



15TH INTERNATIONAL RESEARCH CONFERENCE

*Economic Revival, National Security, and Sustainability through
Advancement of Science, Technology, and Innovation*

29TH - 30TH SEPTEMBER 2022

BASIC AND APPLIED SCIENCES

ABSTRACTS



GENERAL SIR JOHN KOTELAWALA DEFENCE UNIVERSITY



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ADVANCEMENT OF SCIENCE, TECHNOLOGY, AND INNOVATION**

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**General Sir John Kotelawala Defence University
Ratmalana, Sri Lanka**



This book contains the abstracts of papers presented at the Basic and Applied Sciences Session of the 15th International Research Conference of General Sir John Kotelawala Defence University, Ratmalana, Sri Lanka held on the 29th and 30th of September 2022. No part of this publication may be reproduced, stored in a retrieval system or transmitted in any form, without prior permission of General Sir John Kotelawala Defence University, Ratmalana, Sri Lanka.

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Message from the Secretary, Ministry of Defence



I am indeed delighted to pen this message whilst extending my earnest felicitations to the KDU on this significant occasion of the annual International Research Conference. At this juncture, I would also like to congratulate the Vice-Chancellor and the team for continuing the tradition of organising this conference consecutively for the 15th time, despite the numerous economic and social challenges faced by the country in the post-COVID environment.

Further to that, I am delighted to perceive that this year's conference theme; 'Economic Revival, National Security, and Sustainability through Advancement of Science, Technology, and Innovation' focuses on the National Economic Growth and National Security as core concepts, and suggests that 'economic development' and 'security' of a country should always go hand in hand. Therefore, this conference would undoubtedly become a forum for academia to discuss an area of absolute need in the development interests of our motherland.

Moreover, I am pleased to witness that KDU, under our ministerial guidance, is setting an example for all other universities in Sri Lanka in progressing research in many academic fields. I hope this year's conference will produce a significant research outcome that the policy community of Sri Lanka could utilise to support the present development drive of the country. Further, I would like to urge the conference organisers to explore the possibility of distributing the outcomes of the conference to all the relevant Ministries and Departments of the country so that said entities could link with the researchers and employ their valuable research outcomes for the benefit of the nation.

I wish that KDU IRC 2022 will enhance the wisdom of all the participants to serve Mother Lanka for a better tomorrow.

GENERAL KAMAL GUNARATNE (Retd)

WWV RWP RSP USP ndc psc MPhil
Secretary - Ministry of Defence

Message from the Keynote Speaker



It gives me immense pleasure to send this message on the occasion of the 15th International Research Conference of the General Sir John Kotelawala Defence University (KDU). I would like to congratulate KDU for being able to conduct its International Research Conference in 2022, consecutively for the 15th time. It is not an easy task to organize such a momentous event particularly under many difficulties and challenges posed by the COVID 19 pandemic situation and social and economic crisis. It is gratifying to witness that KDU, the only Defence University in the country, has been able to transform a challenge into an opportunity, as it usually does.

The theme of the conference, namely the “Economic Revival, National Security, and Sustainability through Advancement of Science, Technology, and Innovation,” is very timely and of great significance for deliberation in expert panels of this conference. The nexus between National Growth and National Security is closely interwoven. The ‘economic revival’, ‘sustainability’ advancement’ and ‘security’ of a country cannot be compartmentalized and discussed in isolation of each other. There is no security for a nation without economic and social progress, and likewise, economic and social progress cannot be achieved without stability and a secure environment. I hope various panels of this conference will be able to discuss many facets of economic revival, national growth, sustainability and security and their interconnectedness. These two areas have a direct bearing on the development of Sri Lanka, a country which succeeded in ending a 30-year long separatist war. In the context of the present need for robust development, it is absolutely necessary to engage in serious research which leads to discoveries as well as policy-oriented recommendations. Therefore, all academic establishments must provide a conducive space for their intellectuals to reach new frontiers in research. I am glad that KDU is setting an example for all other universities in Sri Lanka in this regard. I hope this year’s conference will produce significant research outcomes that the policy community in Sri Lanka could utilize for the benefit of the country. I wish this conference all the success.

HON PROF SUBRAMANIAN SWAMY

Former Minister of Commerce, Law & justice, India

Message from the Vice Chancellor



The International Research Conference (IRC) of General Sir John Kotelawala Defence University held for the 15th consecutive year is significant in terms of the continued contribution of the University to the field of research in diverse disciplines much needed for the progression of the nation, especially in the face of unprecedented challenges caused by the COVID-19 pandemic and the current economic crisis in the country.

The conference themes carefully selected by KDU each year have addressed contemporary needs of the country that are linked up with national security perspectives, and they are complementary to the development paradigm of the country. This year's theme "Economic Revival, National Security, and Sustainability through Advancement of Science, Technology, and Innovation" encompasses a wide range of research possibilities for scholars of different disciplines to engage in much useful research relevant to the current issues faced by the nation.

It is heartening to note that the number of papers submitted for the conference has increased despite the challenging circumstances, which is a positive indication of the enthusiasm growing in the country on development and security related multi-disciplinary research. In this respect, I am extremely glad that the KDU's efforts in expanding higher educational opportunities, increasing quality of higher education, enhancing research and innovation, linking up research with the industry and so on have increasingly been acknowledged by many, which is also reflected in the Times Higher Education Impact Ranking, 2022 table, where KDU has been ranked 2nd in Sri Lanka for Quality of Education and 4th in the overall ranking in the country and in the 801-1000 range globally.

KDU IRC also creates a sound platform to initiate collaborative research at both national and global levels, and I invite all participants to use this conference to make lasting and productive connections and networks at the individual, institutional, national, and international levels to envisage and explore mutually beneficial research possibilities and higher education experiences for the future.

While appreciating the commitment of the organizers of this year's conference, I wish you all, the presenters and participants taking part in the conference all the very best, and I hope you will enjoy every moment of this two-day academic endeavour.

MAJOR GENERAL MILINDA PEIRIS

RWP RSP VSV USP ndc psc MPhil (Ind) PGDM
Vice Chancellor
General Sir John Kotelawala Defence University

Message from the Conference Chair



For the 15th consecutive year, General Sir John Kotelawala Defence University (KDU), organises its International Research Conference (KDU IRC 2022) under the theme of “Economic Revival, National Security, and Sustainability through Advancement of Science, Technology, and Innovation”. It is with great pleasure and honour that the organising committee extends its compliments to all of you taking part in KDU IRC 2022. Holding the KDU IRC 2022, under the patronage of the Vice Chancellor, amidst many challenges encountered throughout the year, was a remarkable experience for me. I believe that the organising committee accomplished a very successful mission.

Despite the economic crisis, KDU IRC 2022 is a tremendous opening for many researchers all over the world encompassing various disciplines such as Defence and Strategic Studies; Medicine; Engineering; Management, Social Sciences and Humanities; Law; Built Environment and Spatial Sciences; Allied Health Sciences; Basic and Applied Sciences; Computing; Criminal Justice and Technology to present their research to fellow scholars, professionals, and students.

In this context, we have assembled excellent thought-provoking scientific sessions under the conference theme of this year, and it is remarkable to highlight your participation, at this conference through a highly competitive selection process. In addition, world-renowned invited speakers will deliver keynote and plenary speeches while covering a wide range of important sessions with great networking opportunities and providing solutions using science, technology, and innovation. It is the esteem of the conference to bring together a diverse group of people to disseminate high-quality and novel research results, which will assist to chart our journey forward to reach new heights.

Finally, I would like to extend my best wishes to all the presenters, authors and participants, joining the KDU IRC 2022 on site or online, and I hope that all of you will find this conference informative, enjoyable, and encouraging to feel the experience of KDU hospitality during these two fruitful days.

DR KALPA W SAMARAKOON

PhD, MSc, BSc, MACS (USA), M.I.Biol (SL), C.Biol (SL)
Conference Chair
General Sir John Kotelawala Defence University

Message from the Conference Secretary



Together with the committees and participating academia of this university, I share the immense pleasure and honour of perseverance with the 15th International Research Conference of KDU (KDU IRC 2022), amidst many challenges, under the patronage of our Vice Chancellor and Deputy Vice Chancellor.

The timely congregation for IRC 2022, of all our staff, students and contributors from faculties all over the world, under the theme “Economic Revival, National Security, and Sustainability through Advancement of Science, Technology, and Innovation”, is of paramount importance in this current climate of the global recession.

Whilst thanking all of you, I express my sincere hope that this would be an ideal platform for academia and professionals to discuss economically viable intelligent solutions for diverse problems for the nation to emerge stronger out of the recession, with the ability to provide equitable health, food, and social security, quality education, and enforcement of law and order in our country, for the betterment of our society.

DR PANDULA ATHAUDA-ARACHCHI

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ORAL PRESENTATIONS

Evaluation of the Loading Capacity and Releasing Efficiency of Graphene Oxide Based Nanocomposites Loaded with Natural Bioactive Compounds

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Graphene-based nanocomposites have been recognized as effective drug delivery systems due to their unique properties such as two-dimensional structure, biocompatibility, easy surface modification, and high efficacy in drug loading and releasing. Vanillin, gallic acid, and quercetin are natural bioactive compounds that exhibit a variety of pharmacological properties. The objective of the present study is to evaluate the loading capacity and releasing efficiency of vanillin, gallic acid, and quercetin in PEGylated nano-Graphene Oxide (PEG-nGO). Nano-graphene oxide was synthesized using a modified Hummer's method followed by ultrasonication and PEGylation. Then vanillin, gallic acid and quercetin were separately loaded into PEG-nGO in different ratios (1:1, 1:10, 1:100, 1:1000 of PEG-nGO: bioactive compound). The prepared nanocomposites were studied for loading efficiency, loading capacity, and releasing efficiency. Among different nanocomposites, the 1:1 ratio of quercetin-loaded PEG-nGO showed the highest loading capacity of $37.79 \pm 0.016\%$. Vanillin ($3.80 \pm 0.005\%$) and gallic acid ($2.68 \pm 0.012\%$) exhibited comparatively lower loading capacity in the nanocomposites with a 1:10 ratio. Compared to the other two bioactive compounds (vanillin: $39.54 \pm 0.005\%$ and gallic acid: $27.53 \pm 0.012\%$) the loading efficiency was also considerably high in quercetin ($60.74 \pm 0.016\%$) loaded nanocomposite. Further, quercetin showed the highest releasing ability with an initial rapid release within the first 06 hours ($65.89 \pm 0.001\%$) followed by gradual release within the next 72 hours ($95.22 \pm 0.001\%$), while vanillin ($67.25 \pm 0.001\%$) and gallic acid ($46.79 \pm 0.04\%$) exhibited a comparatively less release within 72hours. The results of the present study revealed that PEG-nGO loaded with quercetin could be used as an efficient nanocomposite for the development of smart pharmaceutical products.

Keywords: *graphene oxide, nanocomposite, quercetin*

Comparative Study of Hexavalent Chromium [Cr(VI)] Removal Efficiency of Natural Adsorbent, Planktonic *Bacillus subtilis* KCB07C10, and their Biofilm

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Chromium is a widely used raw material in textile, tanning, electroplating, and wood preservative industries, which releases significant amounts of hexavalent chromium contaminated wastes into inland waters and aquatic environments, causing hazardous effects on biota, i.e., cancers, organ disorders, morphological changes, functional alterations, etc. Therefore, many pollutant removal strategies including chemical, physical and biological remediation methods are used in the chromium-based industry to prevent such adverse effects. This study mainly compared the effectiveness of physical and biological methods for hexavalent chromium removal. The physical remediation was done using agricultural waste; straws as a natural adsorbent, which is abundant in Sri Lanka. Bioremediation was investigated using previously isolated hexavalent chromium tolerant bacterial strain *Bacillus subtilis* KCB07C10. Bacterial biofilms of *B. subtilis* KCB07C10 were formed on straw and visualized by scanning electron microscopy. Straw, bacterial isolate, and biofilm were exposed to 16.0 mg/L of hexavalent chromium in Tris minimal medium (modified), and hexavalent removal was measured spectrophotometrically by 1,5-diphenylcarbazide analysis during 24 hours of time intervals up to 96 hours. The study describes that the complete removal of hexavalent chromium can be achieved by both *B. subtilis* KCB07C10 immobilized straw and planktonic cell suspension within 48 and 96 hours, respectively, while the adsorbing straw indicated 51.47 % removal during the study period. It can be concluded that the efficiency of hexavalent chromium removal differs significantly among the adsorbents, planktonic bacteria, and biofilms in question ($p=0.022$) and that hexavalent chromium removal can be efficiently performed by *B. subtilis* KCB07C10 biofilm on straw.

Keywords: hexavalent chromium, removal, adsorbent, bacillus subtilis, biofilm

Preparation and Characterization of Starch – Cellulose Biodegradable Polymer Composites for Property Enhancement

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Recently, the production of natural biodegradable polymer composites using renewable sources has become an important research area. These biopolymer composites possess enhanced properties than biopolymers. In this study, a biopolymer composite was prepared using Cassava starch and cellulose that was extracted from *Panicum maximum*. During the study, polymers were prepared using the solution casting technique with a glycerol plasticizer. Starch polymers were prepared by mixing starch and glycerol in 1:1, 2:1, 3:1, 4:1, and 5:1 ratio respectively. Starch-cellulose bio composites were prepared by mixing cellulose and starch in different ratios. During the preparation of biopolymer composites, glycerol and acetic acid amounts were kept constant. The biodegradability, water solubility, water absorption capacity, and mechanical properties of those polymer films were determined. Furthermore, the polymer films were characterized using a scanning electron microscope and infrared spectroscopy in order to investigate the surface morphology and the chemical modifications of polymer films respectively. The tensile strength of the starch polymers increases when increasing the starch content. The water solubility and biodegradability of films were decreased when increasing the starch content. The addition of cellulose increases the tensile strength due to the strong interaction between the starch and cellulose. In starch-cellulose composites, biodegradability has been decreased when increasing the cellulose content. Hence, in starch polymers, better properties can be obtained when increasing the starch content. Whereas in starch-cellulose composites better properties have been obtained with higher cellulose content. Furthermore, properties can be improved by the modification of starch using citric acid.

Keywords: *renewable, biodegradability, biopolymer composites*

Acetylated Starch Nanoparticles for Encapsulation of *Flueggea leucopyrus* Leaves Extract for Anticancer Targeted Drug Delivery

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In cancer treatments, the major problem is the target release of drugs to cancer cells. Due to leaky vasculature and poor lymphatic drainage, molecules in the nanometer range can accumulate near cancerous tissues rather than normal tissues, known as the enhanced permeability and retention effect (EPR). In this study starch obtained from *Manihot esculenta* (cassava) was used to synthesize drug-loaded acetylated cassava starch nanoparticles (ACSNP). Acetylated starch was synthesized by an esterification reaction using acetic anhydride and acetic acid as reactants. The nanoprecipitation method was applied for the preparation of ACSNP by using acetone as the solvent and water as the anti-solvent. The *Flueggea leucopyrus* (FL) has been used in the treatments of cancers in Ayurvedic medicine in Sri Lanka. The major active ingredient found in the FL leaves is bergenin. The leaf extract of FL was encapsulated into ACSNP due to hydrogen bonding interactions between active ingredients and the ACSNP. The mean particle diameter of drug loaded ACSNP was 183.8 nm, determined by dynamic light scattering. Loading efficiency (LE%) and loading capacity (LC%) were determined by UV-Vis spectrum, showing clear absorption peaks at 217 nm and 273 nm specific for bergenin in the leaves extract. The LE% and LC% were 54.09% and 27.76% respectively. The drug release studies showed that ACSNP allowed the immediate release of bergenin at pH 5.6 (acidic pH). These results indicate that ACSNP is a promising vehicle for the loading of natural anticancer drugs and a potential candidate for targeted anticancer drug delivery.

Keywords: *Flueggea leucopyrus*, acetylated starch, nanoprecipitation, targeted drug

Biodegradable Polymer Composites using Starch-based Polymer and Teak Sawdust

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Many useful material properties specifications and mechanical functionalities of biodegradable polymers are comparable to conventional plastics. However, the resistance of synthetic polymers to physical, chemical, or biological degradation has caused serious environmental problems. To overcome these problems, biopolymers are being suggested as safer alternatives. In this study, the synthesis of thermoplastic starch-based biopolymer from starch reinforced with sawdust was investigated. Glycerol was used as a plasticizer and acetic acid resisted the fungus to be formed and spread throughout the surface. Biopolymer composites were prepared with various percentages of sawdust (20%, 40%, & 60%) compared to the dry matter of cassava starch. Starch from both edible sources (cassava & jackfruit seeds) and non-edible sources (mango seeds & avocado seeds) and sawdust (~350 µm) from teak were used. The chemical and physical properties of these composites were analysed using Fourier Transform Infrared Spectroscopy, Scanning Electron Microscope, biodegradability, tensile strength, water solubility, water absorption capacity, and moisture content. Unmodified teak sawdust was soaked in 5% of NaOH (aq) solution and washed until base free. Unmodified starch with chemically modified sawdust (S/CMSD) composite resulted in higher tensile strength values compared to the composites with both chemically modified starch with unmodified sawdust (CMS/USD), and unmodified starch with unmodified sawdust (S/USD) composites. S/CMSD & CMS/USD composites had the lowest values for water solubility and biodegradability than S/USD composites. Water solubility, biodegradability, water absorption capacity, tensile strength, and moisture content were decreased with increasing sawdust percentage. These results indicate that S/CMSD and CMS/USD have great potential in the production of biodegradable packaging material.

Keywords: *biopolymer composites, starch, teak sawdust*

Investigation of Wound Healing Ability and Chemical Constituents of “Wathupalu” (*Mikania cordata*) Leaf Extracts

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Mikania cordata (Burm. f.) Robinson is a herbaceous vine belonging to the family Asteraceae. Traditionally it is mainly used for cuts and wounds. This study aims to evaluate wound healing ability and chemical constituents in *M. cordata* leaves responsible for wound healing. Leaf extracts were prepared by either crushing or sonicating with different solvents. Scratch assay was performed to analyse cell migration *in vitro* using ethanolic extract of *M. cordata* leaves and C6 glioma cells. Cells were grown in culture dishes to create a wound as a scratch. Cells were incubated in ethanolic extracts having 10, 100, 500, and 1000 µg/ml concentrations, and cell movement was observed using a fluorescence microscope. Live cell images were recorded and analysed at different time intervals (0th hour, 6-, 12- and 24-hour) to quantify the cell migration with time. The 1000 µg/ml ethanolic extract of *M. cordata* leaves was found to have a positive effect on cell migration with a 100% wound closure rate over a 24-hour incubation period. Next, thin-layer chromatography (TLC) and column chromatography were carried out to separate the chemical constituents. TLC chromatograms showed that sonicated leaf extracts had fewer chlorophyll traces than crushed extracts. The high amount of plant pigment traces could conceal the bioactive chemical constituents. Hence, the study was continued with leaf extracts prepared by sonication. Leaf powder sonicated in DCM had more chemical compounds than the other extracts. Future studies have to be carried out on DCM extract and scratch assay to build the correlation between leaf extract and wound healing.

Keywords: *Mikania cordata* (Burm. F.) Robinson, external wound healing, scratch assay

Protein-Ligand Docking for Atherosclerosis to Uncover Phytochemicals as Therapeutic Exercise

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Atherosclerosis is one of the primary reasons of the global leading cause of death, cardiovascular disease (CVD), and stroke, the world's second cause of mortality. It is an advancing disease with lipid and fibre accumulation in large arteries. PCSK9 (PDB ID:6U26) has become an attributable target that regulates the pathogenesis of atherosclerosis. Inhibition of PCSK9 significantly diminished the development of atherosclerosis. This study is aimed to discover naturally occurring PCSK9 antagonists as potential drug candidates to treat atherosclerosis. In this study, docking of 102 ligands was performed using Autodock vina 1.2.6 to detect PCSK9 inhibitors from NCBI PubChem database. Through a single config.txt file and *.BAT file, vina docking for multiple ligands was executed. BIOVIA DS was used for the visualization of the receptor-ligand interactions. SwissADME web tool was utilized to categorize and filter potential candidates based on Lipinski's Rule of Five against atherosclerosis and GI absorption for oral administration was evaluated. The docking result of phytochemicals showed Silibinin, Canadin, and Manoalide (-10.7, -10.0, -9.7 kcal/mol) respectively and other 99 compounds (binding affinity ranges from -5.7 to -9.7 kcal/mol). TRP461, VAL 460, PRO 438, and PRO 331 amino acid residues were identified as the most common active sites within the binding pocket of the PCSK9 receptor. The study finds Silibinin, Canadine, Manoalide and most of the compounds respect all drug-likeness rules and can be used as effective molecules for PCSK9 receptor antagonists based on the docking method. *In vitro* study can be conducted in the future based on this research.

Keywords: *.BAT file, PCSK9, silibinin, vina

Green Synthesis of Silver Nanoparticles using Five Varieties of Leaf Extracts from *Annona* Species: Evaluation of Antioxidant, Photocatalytic Properties, and Antibacterial Properties

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The development of eco-friendly and sustainable methods to synthesize silver nanoparticles (AgNPs) is a rapid field in nanotechnology due to its eco-friendly nature. In this study, five varieties of *Annona* leaves (*Annona muricata*, *Annona reticulata*, *Annona squamosa* green, *Annona squamosa* red, and *Annona glabra*) were used to synthesize AgNPs and the photocatalytic, antioxidant, and antimicrobial properties were investigated. One mL of AgNO₃ solution was mixed with 9 mL of the extract. Initially, the formation of AgNPs was observed by a colour change at room temperature after 24 hours. AgNPs were further confirmed by the UV-Vis spectroscopy which displayed a plasmon resonance peak between 420-440 nm. The size and shape of the AgNPs were analyzed by the scanning electron microscope and the analysis showed that AgNPs are spherical with a size of ≈40 nm. The antioxidant properties were analyzed by total flavonoid, total phenolic, and total antioxidant contents. All three antioxidant assays confirmed that the AgNPs have higher antioxidant activity than their respective water extracts. The photocatalytic activity of 267 ppm and 4000 ppm *Annona squamosa* red AgNPs was analyzed using methyl red under sunlight. The degradation of methyl red was achieved within 75 minutes. The antibacterial properties of AgNPs and the water extracts were analyzed against *E. coli* and *S. aureus* using the well diffusion method and showed a significant difference ($p= 0.001$ and 0.03 respectively) in the zone of inhibition. Therefore, the results confirmed that the green synthesis AgNPs from five varieties of *Annona* plants can be used in a variety of medical applications.

Keywords: silver nanoparticles, annona leaves, green synthesis, antioxidant, photocatalyst, antimicrobial

Characterizing Surfactant Effect on Phytoremediation of Carcinogenic Anthracene by *Eichhornia crassipes* Plant

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Anthracene is a mutagenic and carcinogenic polycyclic aromatic hydrocarbon mainly found in crude oil. Anthracene shows bioaccumulation and biomagnification due to its high lipid solubility. Therefore, anthracene has the ability to accumulate in toxic levels at higher trophic level organisms. A high stability makes this organic pollutant harder to remove from the environment. This study aims to characterize alternative plant-based bioremediation techniques to clean up anthracene from crude oil-contaminated aquatic ecosystems. We tested whether *Eichhornia crassipes*, an invasive water plant commonly known as water hyacinth is capable of absorbing anthracene from contaminated water. Considering the low water solubility and mass transfer of anthracene, we further investigated how a non-ionic surfactant Triton-X-100 could affect anthracene absorption into the water hyacinth plant. Plants were treated with anthracene-spiked crude oil-containing water with or without Triton-X-100 and grown for 11 days. Plant roots were cleaned, freeze-dried, and subjected to ultra-sonication. Anthracene was extracted from root tissues using dichloromethane: acetone mixture. Silica column chromatography was performed for further purification. Identification and quantification of anthracene were performed using high-performance liquid chromatography, using a C18 reverse phase column. Control plants were grown in fresh water. Results indicated that the water hyacinth plant absorbed 1.98 ± 0.66 mg/kg of anthracene into the root tissues. The presence of Triton-X-100 significantly increased anthracene uptake to 11.06 ± 0.31 mg/kg. This represents a surfactant-mediated 5.6-fold increase in anthracene uptake in the plant. Using Triton-X-100 can be a viable option to enhance the efficacy of anthracene removal from contaminated waters by water hyacinth plants.

Keywords: *Eichhornia crassipes*, anthracene, phytoremediation, Triton-X-100

Evaluation of *in vitro* Antioxidant Potential of Multifaceted Phytogetic ZnO Nanoparticles Synthesized from Peel Waste Extracts of *Ananas comosus* Varieties

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Green synthesized nanoparticles (NPs) reveal many advantages over conventional physiochemically synthesized NPs and exhibit numerous applications in various fields. The present study focused on the synthesis of zinc oxide (ZnO) NPs from *Ananas comosus* peel waste extracts that are obtained from two varieties: Murusi and Kew. The effect of various parameters on green NPs synthesis was studied, including Zn (CH₃COO)₂ · 2H₂O ion precursor concentration, peel extract to ion precursor ratio, irradiation methods (solar, microwave, UV), pH of the medium, and incubation time. The structural properties of NPs were investigated by UV-Vis spectrophotometer, FTIR spectroscopy, XRD, SEM, and TEM. The antioxidant potentials were determined using 2,2-diphenyl-1-picrylhydrazyl (DPPH) and ferric reducing antioxidant power (FRAP) assays. The optimal conditions for the synthesis of ZnO NPs were 0.1 M Zn (CH₃COO)₂ · 2H₂O concentration, 4:1 metal ion solution to peel extract ratio, microwave irradiation, medium pH of 12, and a 24-hour incubation time for both peel extracts. The absorption of surface plasmon resonance (SPR) peaks appeared in the range of 340 to 350 nm. The presence of O-H and N-H bioactive functional groups involved as capping agents in the synthesis of ZnO NPs was confirmed by FTIR. According to the XRD pattern, the average crystallite size of the Murusi and Kew peel mediated ZnO NPs under optimal conditions was 14.74 nm and 14.47 nm, respectively. SEM analysis revealed that Murusi and Kew mediated NPs were spherical in shape. The presence of rod-shaped ZnO NPs synthesized from Murusi and Kew peel was confirmed by TEM images with sizes of 15.44 nm and 10.23 nm, respectively. The biosynthesized ZnO NPs were found to have a high antioxidant potential. At 100 ppm, Kew peel-mediated ZnO NPs had a higher DPPH radical scavenging capacity (84%) than Murusi peel-mediated ZnO NPs (79%). Similarly, Murusi and Kew peel mediated ZnO NPs showed high FRAP scavenging power when compared to metal ion precursor and peel extracts.

Keywords: *antioxidant activity, agro waste, Kew and Murusi, peel extract, Zinc oxide nanoparticles*

***In vitro* Antioxidant and Anti-inflammatory Activity of Leaf and Stem Bark of Selected *Mangifera* Species in Sri Lanka**

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Mangifera indica (MI), locally known as ‘Amba’ in Sinhalese is the richest species of *Mangifera* found in Sri Lanka with its multiple varieties. This research determined the antioxidant and anti-inflammatory effects by *in vitro* bioassays of ethanol extracts of the leaf (L) and stem/bark (SB) of three native MI varieties; Karthucolomban (K), Willard (W), and Vellaicolomban (V). Ethanol extractions were performed on the collected L and SB samples where, Total Phenolic Content (TPC), Total Flavonoid content (TFC), antioxidant assays such as DPPH, ORAC, and an anti-inflammatory assay known as human red blood cell membrane stabilization assay (HRBC), and also thin layer chromatography (TLC) were conducted. When the sample extracts were compared to gallic acid (GA) the highest TPC was recorded for KSB 3.33 mg/gGAE and KL 3.34 mg/gGAE. When compared against quercetin (Q) by the TFC assay, WL demonstrated the highest flavonoid content (1.15 mg/gQE). DPPH assay was conducted to determine the antioxidant activity, and results were compared against ascorbic acid (AA). WSB showed the highest percentage inhibition (95%), while the second highest inhibition was noted for WL (91%), and while AC only showed 77% inhibition. ORAC assay was also conducted for these two samples where it was found 0.83 mg/g AAE for WSB and 0.64 mg/g AAE for WL. Moreover, when compared to ibuprofen (97% inhibition), the highest inhibition was noted for VSB (97%), and KL and VL showed similar inhibitions (96%) by the HRBC assay. Interestingly, KL showed two prominent spots by TLC. As per the findings, it can be concluded that the tested three varieties of MI, do contain antioxidant and anti-inflammatory properties at varied levels. Nevertheless, WL and WSB showed good antioxidant activity while VSB demonstrated noteworthy anti-inflammatory activity. Thus, more detailed scientific research is warranted to effectively evaluate the medicinal effects of all parts of these varieties of the MI plant.

Keywords: *Mangifera indica*, antioxidant activity, anti-inflammatory activity

A Solvent Polymeric Membrane Ion-Selective Electrode Based on N, N'-bis(Salicylaldehyde)-4-Chloro-O Phenylenediamine Ligand

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Schiff bases are one class of supermolecules that has their applications in biochemical and analytical reagents. They have proven to play an important role as ionophores of potentiometric membrane sensors which are used for selective determination of both the cations and anions in environmental and biological samples. This study developed a solvent polymeric membrane electrode using N, N'-bis(salicylaldehyde)-4-chloro-o-phenylenediamine (a salophen type Schiff-base) as a neutral ionophore. The membrane consists of a poly (vinyl chloride) matrix with 2-nitrophenyl octyl ether (o-NPOE) as plasticizer and Potassium tetrakis(4-chlorophenyl)borate (K-TCPB) as ion-exchanger. The potentiometric response of the electrode towards the metal ions: K⁺, Ni²⁺, Co²⁺, Cu²⁺, Cr³⁺, and Pb²⁺ were investigated in the aqueous medium. Based on the results, the selectivity pattern and the observed slopes of the calibration sensor is as follows: Co²⁺ (16 mV/dec) < Ni²⁺ (23 mV/dec) < Cu²⁺ (33 mV/dec) < K⁺ (49 mV/dec) < Pb²⁺ (39 mV/dec) < Cr³⁺ (73 mV/dec). As observed Cr³⁺ shows a super-Nernstian response and the stability was quite low as well. Therefore, the sensor was characterized using Pb²⁺. The membrane electrode demonstrated a Nernstian response of 33mV/decade over the concentration range of 1×10^{-8} to 1×10^{-1} mol dm⁻³ of Pb²⁺. Therefore, the sensor can be used to determine very low concentrations of Pb²⁺ ions.

Keywords: carrier based ion-selective electrodes, poly(vinylchloride) membranes, schiff-bases as ionophores

Silver Nanoparticles as SERS-Active Probes for Detecting Melamine

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Milk is one of the essential food items throughout the world. Intentional adulteration of milk products with melamine falsely increases the apparent protein content in milk and it is a health concern associated with kidney damage. Therefore, developing an effective method for detecting melamine is important. In this research, a highly sensitive SERS (Surface Enhance Raman Spectroscopy) technique was applied to detect lower concentrations of melamine with silver nanoparticles to indicate the potential of Raman spectroscopy to design an optical biosensor. This preliminary investigation can address shortcomings of high-cost and sophisticated detection techniques and enables the applicability in onsite detection of melamine. In the experiment, the nanoparticles and samples were mounted on tsetupan set up using the hanging drop method and drop casting method. The Raman measurements were obtained using a custom-built research-grade Raman spectrophotometer with a laser wavelength of 532 nm. The most sensitive SERS probe was developed when classical silver nanoparticles were drop cast on the glass slide. Therefore, further quantitative analysis including method validation and application into commercial milk samples was performed using the drop-casted sample. The calibration plot for the intensity of the SERS peak at $\sim 691\text{ cm}^{-1}$ versus melamine concentration shows linearity in the range of 0.2 ppm- 50 ppm for the SERS probe developed via drop casting classical silver nanoparticles. The limit of detection and limit of quantification for this method are 3.05 ppm and 10.16 ppm, respectively. This developed SERS probe allows quick, simple, and cost-effective qualitative and quantitative analysis of melamine.

Keywords: melamine, SERS, adulterant

Identifying the Removal Efficiencies of As^{3+} in Wastewater by Functionalized Nanocellulose

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Nanocellulose (NC) gains significant attention as a promising candidate in water purification and environmental remediation studies due to its superior chemical and physical properties. The present study explored the capacity of removing the hypertoxic As^{3+} in wastewater, with the use of NC after functionalization, namely sulfonation, phosphorylation, and xanthation. Cellulose was extracted through an alkaline treatment followed by bleaching with NaOCl from *Panicum maximum*, which is an invasive plant in Sri Lanka, selected as the cellulose source. Acid hydrolysis on extracted cellulose with 50% sulfuric acid, 85% phosphoric acid, and 21.9 % hydrochloric acid results in sulfonated, phosphorylated, and non-functionalized NC respectively. Xanthation on non-functionalized NC with NaOH and CS_2 results xanthated NC. Functionalized NC was separately fabricated on each filter paper with 50 mg loading and allowed to filter the As^{3+} -containing water through it. The As^{3+} concentrations in the medium were determined by the colour intensity of formed I_2 with the reduction of IO_3^- , which corresponds to the oxidation of As^{3+} into As^{5+} in an acidic medium, with the aid of UV-Vis spectroscopy. Among the functionalized NC, the sulfonated NC has shown the highest removal efficiencies in 200 ppm, 150 ppm, and 100 ppm As^{3+} concentrations with removal percentages of 46.8, 38.4, and 50.1. Further advancement of this work can reach the development of bio-degradable and affordable columns for As^{3+} removal. Dynamic Light Scattering results of sulfonated, phosphorylated, and non-functionalized NC were 295.7 nm, 271.4 nm, and 320.9 nm respectively, indicating that the particle sizes were in the nanoscale range.

Keywords: Arsenite, functionalized nanocellulose, water purification

Graphene Derivatives for Supercapacitor Applications

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Graphene has superior properties which are enormously advantageous in energy applications. Despite graphene's superior properties, it has limitations such as the utilization of complicated synthesis methods and restacking. One of the effective strategies to overcome these limitations is the synthesis of graphene derivatives. This study reports the synthesis of porous graphene (PG) *via* a novel one-step/one-pot electrochemical exfoliation approach. Electrochemical exfoliation was carried out using a graphite rod and a stainless-steel electrode as the anode and cathode. The electrolyte used was 0.1 M $(\text{NH}_4)_2\text{SO}_4$ in 1 M H_2SO_4 and contained 0.03 M Phytic acid (PA). As-synthesized PG was characterized using scanning electron microscopy (SEM), X-ray diffraction (XRD), and Fourier transform infrared (FT-IR) spectroscopy. SEM, FT-IR, and XRD confirmed the formation of a porous structure, the formation of new bonds in PG, and the formation of graphene derivatives respectively. The electrochemical characterization of materials for supercapacitor applications was carried out using electrochemical impedance spectroscopy (EIS), cyclic voltammetry (CV), and galvanostatic charge-discharge (GCD). EIS showed that in PG, the resistance decreased, capacitance increased, and diffusion improved. CV confirmed that PG produced the highest specific capacitance (SC) compared to activated carbon (AC), graphite, and graphene. The percentage increments SC of graphene and PG compared to graphite are 42280.7 % and 93346.5 % respectively. GCD confirmed PG has low potential drop, resistance, and good charging time. These results collectively provide strong evidence of easy synthesis of PG *via* one-step/one-pot electrochemical exfoliation.

Keywords: *graphene derivative, supercapacitors, electrochemical exfoliation, specific capacitance, porous graphene*

Selection of RNA Aptamers to Distinguish the V600E Mutation Status of BRAF Protein: A Potential *in silico* Approach

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The valine to glutamate substitution at the 600th residue of B-type rapidly accelerated fibrosarcoma protein (BRAF V600E) is the most common mutation in the *BRAF* gene. Due to its high prevalence in a number of cancers, the development of efficient diagnostic and prognostic assays and therapeutics is essential for their management. Aptamers have become promising candidates in a variety of biomedical applications due to many favourable properties. However, no aptamers that can distinguish the V600E mutation status of the BRAF protein have been experimentally determined. Therefore, this study was conducted to create an initial knowledge base for *in silico* design of aptamers for wild-type and mutant (V600E) BRAF (mutant BRAF) proteins. It was achieved using molecular docking employing HADDOCK 2.4 web server. In the absence of aptamers for BRAF, five RNA aptamers targeted to the activation loop of ERK 1&2 proteins were selected for docking, considering the similarity of the 3D structure of the kinase domains of the above proteins to BRAF. Docking was done for ten protein-aptamer combinations (five aptamers with wild-type BRAF and mutant BRAF). Three complexes were selected based on the HADDOCK score and their intermolecular hydrogen bonds and salt bridges were determined. Three aptamers obtained negative HADDOCK scores signifying that they presumably target the activation loop of wild-type and mutant BRAF. Considering the total intermolecular hydrogen bonds and salt bridges, Aptamers_1 and 3 (Apta-Index IDs: 481 and 263) would preferably bind with wild-type and mutant BRAF, respectively. They have the potential to be used as starting structures in the *in-silico* aptamer modeling workflow for wild-type and mutant BRAF proteins.

Keywords: aptamers, BRAF V600E, hydrogen bonds, molecular docking, salt bridges

A Comparison of Classical Time Series Models and Machine Learning LSTM Model to Forecast Paddy Production in Sri Lanka

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The Autoregressive Integrated Moving Average (ARIMA) family models and the Exponential Smoothing family models are the most widely used and successful conventional techniques for univariate time series forecasting. As a result of the recent development in more sophisticated machine learning methodologies, such as the Long-Short-Term Memory modelling approach, new algorithms are being developed to evaluate and forecast time series data. The objective of this study was to identify the best time series forecasting model among classical time series models and machine learning LSTM model to forecast the annual paddy production of Sri Lanka. The results showed that the estimated error of ARIMA & Double Exponential Smoothing (DES) models is much higher than the estimated error of the preferred LSTM model based on the RMSE, MAE, and MAPE values. Hence LSTM outperforms traditional-based algorithms like ARIMA and smoothing models for forecasting the paddy production in Sri Lanka. The forecasts for paddy production from 2022 to 2024 were 4.92, 4.89, and 5.34 million Mt respectively. Researchers can use this model to forecast the paddy output of Sri Lanka, and it should be continuously improved by including new data.

Keywords: *time series, forecasting, paddy production, Sri Lanka, ARIMA, double exponential smoothing, LSTM*

A Mixture of Two Endemic Plants as Various Forms of Food Supplements with Heightened Biological Activity

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Evidence-based medicines/supplements have emerged as an important dimension in the modern world. We developed two food supplements with two plants: *Osbeckia octandra* (OO) and *Wrightia antidysenterica* (WA). The two food supplements were a tea bag and a capsule, and they were developed with a 1:1 ratio of OO and WA. The newly developed supplements demonstrated good phenolic content by the Folin-Ciocalteu assay. The tea bag food supplement showed 87.20 mg /Gallic Acid Equivalent/Grams and the capsule food supplement showed 68.63 mg /Gallic Acid Equivalent/Grams. The products also showed good flavonoid content by the aluminum chloride colorimetric assay; tea bag- 44.52 mg /Quercetin Equivalent/Grams and capsule- 34.09 mg /Quercetin Equivalent/Grams. Further, the selected combination mediated very high anti-oxidant activity, which was estimated by the DPPH assay when compared with individual plant material where we found that the tea bag food supplement showed an IC₅₀ of 0.004 mg/ml and the capsule food supplement showed an IC₅₀ of 0.01 mg/ml. Butylated hydroxytoluene only showed an IC₅₀ of 0.02 mg/ml. Anti-oxidants mediate significant protection against oxidative stress and play a protective role in malignancies, and inflammatory and neurological diseases. Furthermore, the products showed anti-inflammatory activity, which was an added benefit, of the egg albumin assay which is a protein denaturation assay. The tea bag showed an IC₅₀ of 0.363 mg/ml and the capsule showed an IC₅₀ of 0.651 mg/ml while Ibuprofen demonstrated an IC₅₀ of 0.169 mg/ml. These supplements/nutraceuticals will benefit in maintaining a healthy lifestyle. It is also a useful invention developed from our very own endemic species which could be developed as a commercial application that will bring revenue to Sri Lanka.

Keywords: food supplements, nutraceuticals, medicinal plants, endemic plants

Overflying the Himalayas; the Northward Migration of Sri Lankan-wintering Brown-headed Gulls

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The Himalayan Mountain range, an average elevation of 6,100 m above mean sea level (AMSL), forms the most formidable geographic barrier to birds migrating along the Central Asian Flyway (CAF). In an attempt to understand the poorly studied migration routes of species occurring in the CAF, we are tracking several migratory waterbird species including Brown-headed Gull *Larus brunnicephalus* from Sri Lanka, which encounters the Himalayas during its northward migration to reach its breeding grounds in south-central Asia. In March-April of 2021 and 2022, two Brown-headed Gulls were caught at their non-breeding sites in Mannar Island of Sri Lanka, and were fitted with GPS-GSM transmitters, just before their northward migration, and their movements were tracked. The birds departed Mannar between 25th April and 14th May and arrived at their breeding grounds on the Tibetan plateau between 4th and 20th May. This northward journey of the gulls lasted 7.5 ± 2.1 (Mean \pm SD) days during which they covered a distance of $3,173.2 \pm 534.3$ km. Both gulls stopped over in the Ganges River, India, for approximately two days. Accordingly, their overall migration speed (including stopover duration) was 451.2 ± 198.9 km/day while the travel speed on travel days was 24.2 ± 10.5 km/h. During migration, the gulls crossed the Himalayas through Nepal at an altitude of $5,744.1 \pm 298.5$ m AMSL. In the course of this crossing, they flew 888.0 ± 116.8 m above ground (where surface elevation is $4,856.1 \pm 181.7$ m AMSL), over the peaks of Panbari Himal & Cho Oyu. This is the first evidence of the Brown-headed Gull encountering such high altitudes during migration. Weighing only 372.5 ± 53.0 g, the Brown-headed Gull is likely to be the lightest gull species to be recorded to cross the Himalayas.

Keywords: brown-headed gull, Himalayan crossing, central asian flyway

Evaluation of Effect of Bacterial Consortia on Growth and Yield of Selected Rice Varieties under Greenhouse Condition

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Overuse of inorganic fertilizer (IF) for high yield of new improved rice (NIR) is a threat to the Sri Lankan economy, human health, and the environment. The objective of the study was to determine the effect of selected bacterial consortia (C) on 2 common NIR. A greenhouse experiment was conducted at the Faculty of Agriculture, the University of Ruhuna during *Maha* season 2021/2022. There were 22 treatment combinations for 2 NIR (Bg 366 and Bg 379/2) and 11 methods with eight C (C1,2,3,4,5,6,7 and 8), one isolate (B1), IF, and negative control (NC) following a complete randomized design with five replicates in pots. Days to flowering (DF), height at flowering in cm (HF), and grains per the first panicle (GPP) were evaluated. ANOVA followed by mean separation was performed by SPSS. In Bg 366, HF of C3 (81.8 ±2.3) and C5 (80.8 ± 3.7) were higher than NC (64 ±4.1). In contrast to the lowest GPP (55.2 ±8.9) of NC, C1(87.2 ±8.5), C8(81.7 ±6.9), and B1(89.75±8.8) produced the highest GPP. In Bg 379/2, DF of C3 (100.2 ±2.7) was significantly early than IF (111 ±2.3). C1 (69.6 ±4.9), C8 (72.4 ±3.2) and C4(68.2 ±3.6) were significantly shorter in HF in contrast to IF (84.4 ±3.8). In comparison to GPP of IF (124 ±3.8), those of C2 (141 ±20.3), C3(137 ±11.4), C4(120.2 ±7.8), and C5(139.5 ±20.6) were not significantly different, while NC produced the lowest GPP (96.4± 8.3). The above results indicate the need for field trials of potential consortia to replace IF.

Keywords: bacterial consortia, agronomic and yield characters, new improved rice

Aspartic Protease Inhibitory Activity of *Pleurotus ostreatus* (Black Oyster) Growing in Sri Lanka

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The literature revealed that the irregular secretion of aspartic proteases is contributed to numerous health problems including hypertension, AIDS, malaria, and Alzheimer's disease. Therefore, the discovery of natural aspartic protease inhibitors provides a novel therapeutic strategy. The aim of the present study is to investigate the pepsin inhibitory activity (PIA) of the mushroom, *Pleurotus ostreatus* (black oyster) and to assess the effect of temperature, pH, metal ions, detergents, oxidizing and reducing agents on PIA. Mushrooms were collected from cultivators who used seeds developed from the mushroom's development and training center in Dehiwala-Mount Lavinia, Sri Lanka. A series of concentrations of aqueous mushroom extract (1.25, 2.5, 5, 10 and 20 %w/v) was screened for PIA using egg albumin as the substrate at 280nm. Then, the effect of different temperatures, pH, metal ions, detergents, and oxidizing and reducing agents on PIA was assessed. The highest PIA (69.11±0.001%) was exhibited by 1.25% concentration and it was used for characterization studies. Among different temperatures and pH conditions, the maximum PIA was exhibited at 60°C (67.65±0.001%) and at pH 2.0 (68.65. ±0.001%) respectively. The presence of Ba²⁺ (34.13±0.01%), Na⁺ (33.21±0.02%), Cu²⁺ (28.42±0.01), Zn²⁺ (28.77±0.01) and Fe³⁺ (21.21±0.01%) ions reduced the PIA activity significantly (p<0.05) compared to the control. Tween-20 (25.78±0.01%), dimethyl sulfoxide (28.14±0.02%), and β-mercaptoethanol (23.07±0.006%) also decreased the PIA significantly (p<0.05) compared to the control. The results of the present study revealed that the black oysters growing in Sri Lanka is a potential source of active aspartic protease inhibitors and the characterization data will be applied in future studies.

Keywords: *Pleurotus ostreatus*, aspartic proteases, pepsin inhibitory activity

Insecticide Susceptibility of *Aedes aegypti* in CMC Area for a Commonly used IRS Agent, Lambda Cyhalothrin

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Being a predominantly abundant area for primary vector of dengue, Colombo Municipal Council (CMC) has often exposed to chemical vector control strategies in successive periods of outbreaks to control *Aedes aegypti* density with space spraying and Indoor Residual Spraying (IRS) using Lambda cyhalothrin as IRS agent. The present study was conducted to assess the susceptibility of *Aedes aegypti* in CMC area towards Lambda cyhalothrin as an aspect of resistance monitoring for chemicals used in mosquito control in Sri Lanka by National Dengue Control Unit (NDCU). WHO standard test kits, procedures and 0.03 % (Discriminative dosage), 0.25 % (8 time higher than discriminative dosage) Lambda cyhalothrin impregnated papers were utilized to conduct susceptibility test on 500 mosquitoes in 2020 and 2022 where each test consisted with five replicates with 20 mosquitoes in each replicate. In 2020 *Aedes aegypti* in CMC area were 5% and 44% susceptible for the discriminative dosage (0.03%) of Lambda cyhalothrin where in 2022 they were 12% and 3% susceptible for repeat tests for the same which indicates very high resistance. With 0.25% the susceptibility has increased steeply up to 88% which still indicates the replaced resistant population. A resistant population of *Aedes aegypti* for Lambda cyhalothrin which even can tolerate higher dosages has almost established, where using Lambda cyhalothrin to control *Aedes aegypti* in CMC area is no longer productive. Irrational use of insecticide with no shifting between chemical classes has led to establishment of highly resistant mosquito population which should be a highly notable fact in vector control programmes in Sri Lanka.

Keywords: *insecticide resistance, susceptibility, Aedes aegypti*

Factors Affecting the Performance Anxiety for Under 19 Male Cricket Player's Performances in Colombo District, Sri Lanka

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One aspect of the complex picture of what makes sportsmen successful is their physical makeup. Successful athletes must develop a mindset that enables them to succeed in the face of potentially adverse environmental or psychological factors in addition to ability and aptitude. The prominent objective of the current study is to identify the factors affecting the performance anxiety of under-19 male cricket players in the Colombo district. The survey covered 47 male cricket players who have participated in the "Singer" under-19 one-day tournament and gathered data for a Sports Competition Anxiety Test (SCAT) questionnaire. This study used Minitab 17 software to analyze the data. The study's findings indicated that anxiety levels are not significantly affected by all-rounders' batting and bowling averages. The moderate anxiety level of all-rounders' batting average is higher and that level of all-rounders balling average is lower than high and low anxiety levels of them. The turkey test reveals that bowlers with moderate anxiety had lower average scores than those with low and high levels. Also, batsmen with intermediate levels of anxiety have greater batting averages than those with high and low levels. The researchers discovered that gamers with intermediate anxiety levels perform better than those with low and high levels of anxiety. Thus, this study has shown enough evidence that players' batting and bowling performances in Sri Lanka are significantly impacted by a moderate level of Anxiety. Repeating this study with additional cricket teams is recommended to maintain a moderate degree of anxiety for these players.

Keywords: *anxiety levels, batsman's performance, bowler's performance, all-rounders performance, (SCAT)*

POSTER PRESENTATIONS

Presence of Collagen X in the Dentine of the Developing Tooth

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In physiological conditions, collagen X is mainly found at the hypertrophic cartilage. Hypertrophic chondrocytes transiently produce collagen X as their major extracellular product. This is shortly followed by the mineralization of the cartilage matrix. Dentine of the tooth is a highly mineralized tissue. Collagen I dominates the dentin matrix. Non-Collagenous proteins (NCPs) represent about 10% of the organic components. There are no previous records of presence of Collagen X in the dentine. We have investigated into the developing tooth of the 18-day old mouse embryo ($n=16$). Immunohistochemistry was conducted using rabbit polyclonal Collagen X antibody to see if there is any presence of Collagen X. Von Kossa method was used to detect the biomineralization. We found immuno-histochemical evidence of Collagen X in the dentine. Bio-mineralization was observed on the locations where the Collagen X was present. Collagen X may be having a negative effect on mineralization and it acts as a barrier to hold up the progressing mineralization front. Animal care and experiments performed were in accordance with the protocols approved by the Committee on the Use of Live Animals in Teaching and Research of the University of Hong Kong.

Keywords: collagen X, Dentine, mineralization

Screening of Phytochemical and Biomedical Effects of some Selected *Psidium sp* Leaf Ethanol Extractions

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Psidium sp leaves have traditionally been used to manipulate several diseases over the last decades. Exploring the potential bioactive metabolites from fruit plant leaves has widely increased. There are many approaches carried out to discover the bioactive compounds present in fruit plants and their parts. In this study, three different *Psidium* species, including *Psidium guineanse*, *Psidium guajava*, and *Psidium cinereum* leaves were subjected to ethanol extraction and the phytochemical analysis by total polyphenolic content (TPC) and total flavonoid content (TFC) assays was evaluated. Anti- α -amylase, and radical scavenging activity by oxygen radical absorbance capacity (ORAC), ferric reducing antioxidant power (FRAP), and DPPH assays were performed to screen the bioactivities. Among the extracts, *P. guajava* showed the highest TFC (7.33 ± 1.92 mg quercetin equivalent/g), and *P. guineanse* showed the highest TPC (240.75 ± 77.58 mg GAE/g of extract) compared to other extracts. In addition, *P. guajava* leaf ethanol extract showed the highest DPPH (93.05 ± 0.85 mg Trolox equivalent/g of extract), ORAC (1.04 ± 0.05 μ g Trolox Equivalent/g of extract), and FRAP (256.85 ± 48.15 mg Trolox equivalent/g of extract) radical scavenging activities, respectively. α -amylase inhibitory activity of *P. guajava* has resulted in the highest (IC_{50} of 0.975 mg/mL) compared to other extracts followed by the highest yield (14.33%). Collectively, the ethanol leaf extracts of *P. guajava* were determined with high phytochemicals such as polyphenolics, radical scavenging, and anti- α -amylase activity compared to the other extracts. However, further research to evaluate *in vitro* toxicity is considered.

Keywords: *Psidium guineanse*, guava leaves, anti- α -amylase, antioxidant activity

A Review on Wastewater Treatment using Constructed Wetland Systems: Implications from Sri Lanka

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The Constructed Wetlands (CWs) are engineering designs that improve wastewater quality in a harmless way to the ecosystem. Even though it is a low-cost method of water quality improvement, it is not widely used in Sri Lanka and not maintained in a proper manner. The current study reviews the utilization of constructed wetlands within the Sri Lankan context and the studies done related to the application were analyzed through a critical desktop search. Presently, CWs are being used in real-time applications in the fields of landfill leachate treatment, slaughterhouses, greywater, and the dairy industry within the country. Studies have proven the potential of using CWs as an optimum wastewater treatment option in the above categories. It is recommended to conduct further studies on the usage of CWs in other industrial fields as well and to determine the best fit for vegetation matching with the type of wastewater plus the climate region of the island.

Keywords: *constructed wetlands, landfill leachate, slaughterhouse wastewater, greywater, dairy wastewater, Sri Lanka*

Comparison of Phytochemicals and Antioxidant Activity of the Polysaccharide and De-Polysaccharide Methanol Extracts of Brown Seaweed *Chnoospora minima*

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Chnoospora minima is a brown algae enriched with unique bioactive compounds which are ideal candidates for pharmaceutical, cosmeceutical, and nutraceutical industries. During this study, *C. minima* were extracted using 80% methanol and de-polysaccharide crude methanol extract was obtained by ethanol precipitation followed by separation of the polysaccharide portion. The crude methanol extract of *C. minima* and its fractions were subjected to analysis of phytochemicals and antioxidant activities. For the assessment of radical scavenging activity, DPPH, FRAP, ABTS⁺, and ORAC assays were conducted. Ethyl acetate fractions of both polysaccharide (IC₅₀:0.67 ± 0.01 mg/ml) and de-polysaccharide (IC₅₀:0.59 ± 0.015 mg/ml) crude methanol extracts exhibited DPPH radical scavenging activity in terms of antioxidant activity. Similarly, the highest level of ORAC, FRAP, and ABTS⁺ activity was observed in the ethyl acetate fractions of de-polysaccharide (ORAC:19.73 ± 5.31 mg TE/g; FRAP: 20.34 ± 1.72 mg TE/g; ABTS⁺: 0.06 ± 0.001 mg/ml) and polysaccharide crude methanol extracts (ORAC:16.22 ± 4.31 mg TE/g; FRAP: 19.23 ± 1.98 mg TE/g; ABTS⁺:0.08 ± 0.002 mg/ml). High TPC was observed in the de-polysaccharide crude methanol extract (298.07 ± 0.003 mg GAE/g) and aqueous fraction (141.2 ± 0.002 mg GAE/g) of the polysaccharide crude methanol extract. Highest TFC was observed in both aqueous fractions of de-polysaccharide (594.23 ± 0.001 mg QE/g) and polysaccharide (113.46 ± 0.001 mg QE/g) crude methanol extracts. Chloroform fractions exhibited the highest TAC for polysaccharide (2.20 ± 0.45 mg PE/g), and de-polysaccharide (2.79 ± 0.31 mg PE/g) samples. Therefore, the de-polysaccharide *C. minima* sample exhibited a high level of antioxidant activity along with a high content of phytochemicals which can further be utilized to determine bioactivities that lead to future drug development.

Keywords: *Chnoospora minima*, de-polysaccharide crude methanol extract, polysaccharide crude methanol extract, antioxidant activity, phytochemicals

***In vitro* Bioactivity of Methanol Extracts of *Elaeocarpus serratus* Leaves and Fruit**

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Plants are getting significant attention globally and the worldwide annual market for herbal products is ~USD 60 billion. *Elaeocarpus serratus* which is an underutilized fruit crop in Sri Lanka was selected for scientific evaluation. Oven-dried mature leaves and fruit (without the seeds) were subjected to methanol extraction. It was found that the leaf extract showed high phenolic content of 100.62±7.28 mg Gallic acid equivalent (GAE)/g and flavonoid content of 14.72±0.85 mg Quercetin equivalent/g. The fruit also showed phenolic content of 157.35±4.82 mg GAE/g. The extracts showed good antioxidant activity by the DPPH assay: leaf- 38.42 ± 6.18 mg Trolox equivalent (TE)/mg and fruit- 185.95±6.07 mg TE/mg. Similarly, the extracts showed notable anti-oxidant activity by the ORAC assay: leaf- 52.45±7.61 mg TE/mg and fruit- 15.35±3.29 mg TE/mg. The leaf extract also demonstrated high anti-inflammatory activity by the human red blood cell membrane stabilization assay where the highest percentage of inhibition was recorded as 85% and an IC₅₀ of 6×10⁻⁵ mg/ml whereas standard ibuprofen showed an IC₅₀ of 5×10⁻³ mg/ml only. The leaf extract also showed a percentage inhibition of 47% by the protein denaturation egg albumin assay whereas the +ve control ibuprofen showed a similar value (51%). Further, inductively coupled plasma-mass spectrometry data revealed the presence of biologically significant both essential minerals and trace elements such as Na-125.73 ppm, Al-53.06 ppm, Fe-44.43 ppm, Mn-16.71 ppm, Zn-7.93 ppm, and Sr-6.11 ppm in the leaf extract. Furthermore, the leaf demonstrated ~40% of cell growth inhibition, while the fruit showed ~35% growth inhibition in Vero cells after 24 hours of treatment by MTT assay. This study demonstrated noteworthy anti-oxidant and anti-inflammatory activity along with essential minerals, especially in the leaf extract. Further studies are underway to elucidate additional health benefits of the leaf and fruit, by *in vitro* bioassay since the research on this underutilized plant has been minimal. This work may help to develop processed products such as new preparations of pickles/jams that will instigate economic benefits.

Keywords: *Elaeocarpus serratus*, underutilized fruit plants, health benefits

Microclimatic Buffering Potential of Forest Types in the Wasgamuwa National Park, Sri Lanka

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The habitat-wise differences in microclimatic conditions have the ability to create heterogeneous landscapes by generating different potential buffering capacities. The objective of this study was to evaluate the microclimatic buffering potential across habitat types in a mosaic dry zone landscape in Sri Lanka. The Vapour Pressure Deficit (VPD) values which have been previously used in studies conducted elsewhere in the world, were used as a suitable indicator. The study was conducted in three forest types including Dry-Mixed Evergreen Forests (DMEF), Riverine Forests (RVF), and Scrub Forests (SCF) in the Wasgamuwa National Park from September 2021 and March 2022. The Grasslands (GR) closest to each sampling station were used as reference sites. Forty-five sampling stations were randomly picked RVF(n=8), DMEF(n=15), SCF(n=11), and GR(n=11) and ambient temperature (T) and relative humidity (RH) were recorded at ground level and at 2 m, using specially constructed sensors and data loggers (NodeMCU sensor node and DHT22 digital temperature and humidity sensor) that recorded data every 30 minutes for 24 hours. The study revealed that the highest VPD was in GR (1.05-1.08) and the lowest in RVF (0.51-0.55), at both height levels. All three forest habitats were moister than the GR. The magnitude of the buffering capacity decreased in the order of RVF>DMEF>SCF. This study generated novel information on the microclimatic buffering capacities of Sri Lankan forests, which highlights the need to protect these ecosystems because local-scale microclimatic conditions in forest understories play a key role in shaping the composition, diversity, and function of the ecosystems.

Keywords: *dry zone forest, microclimatic buffering potential, vapor pressure deficit, Sri Lanka*

Biodegradation of Burnt Engine Oil by Surfactant Producing Marine Isolates of *Pseudomonas* sp.

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The impact of pollution from petroleum hydrocarbons and their derivatives, including burnt engine oil, on the environment, is growing. One of the most environmentally benign approaches for the degradation of such pollutants is bioremediation by microbial degradation. In this study, we aimed to assess the potential of newly isolated marine *Pseudomonas* sp. in deteriorating burnt engine oil. One litre of a fed-batch bioreactor with 450 mL of minimal salt medium (MSM) supplemented with 2% (w/v) burnt engine oil as a sole source of carbon and energy. The fed-batch reactor was started with the addition of bacterial inoculum (2%) which was grown in MSM with 2% burnt oil. The reactor was operated at ambient temperature and a magnetic stirrer was used to mix the culture broth. Viable microbial cells were recorded on the 10th and 20th 30th days of incubation. On the 20th day, the bioreactor was fed 100 mL of fresh MSM supplemented with burnt oil, and an equal volume of culture broth was removed prior to the addition. The presence of biosurfactant in the culture broth was determined using the drop collapse method, oil spreading technique, and emulsification assay. Viable bacterial cells found on the 30th day were 2.55×10^6 . Biosurfactant production was observed and oil displacement was found in the range of 2.8 – 3.2 cm and emulsification index was found to be in the range of 50-58%. Therefore, *Pseudomonas* sp. can be suggested as a good candidate to develop a cost-effective method for the bioremediation of burnt engine oil. Further studies are currently in progress to quantitative and qualitative analysis of biodegradation profiles of burnt oil using gas chromatography-mass spectrometry. Proceeding the research further with the characterization of the biosurfactant will undoubtedly pave the way for biosurfactant-mediated bioremediation of hydrocarbon pollutants.

Keywords: *microbial degradation, biosurfactant, bioremediation, burnt engine oil, marine Pseudomonas sp.*

Formulation and Sensory Evaluation of Malted Drink Powder Incorporated with Selected Legumes, Seeds, and Rhizomes

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The study aims to address the prevailing nutritional deficiencies in society by formulating micronutrients and bioactive compounds rich malted drink while replacing palm oil, the most common fat source of commercially available malted drinks with an underutilized seed. For the formulation of the malted drink, Soybean (*Glycine max*) flour, malted green gram (*Vigna radiata*) flour, Watermelon seed (*Citrullus lanatus*) flour, and Ginger (*Zingiber officinale*) powder were selected as the major ingredients. An appropriate experimental design was selected for the product formulation and the best formulation among the formulated samples was screened out through three sensory evaluation sessions. In each session, the sensorial acceptance of the samples was evaluated by a semi-trained panel with 30 members, in terms of appearance, aroma, taste, mouthfeel, and overall acceptability, under a five-point hedonic scale. For the evaluation of responses, Friedman and Wilcoxon test statistics along with radar diagrams were analyzed by IBM SPSS 25 statistical software. The selected experimental design for the product formulation was Taguchi's L8 Orthogonal array. Eight malted drink powder samples were formulated according to it. There was a significant difference among the eight formulations in terms of all the sensory attributes at a 95% confidence interval. The formulation which was incorporated with 52.17% of Soybean flour, 26.00% of malted green gram flour, 17.39% of Watermelon seed flour, and 4.34% of Ginger powder, obtained the highest mean rank in terms of all the sensory attributes. Thus, it acquired the highest sensorial acceptance, and it could be screened as the best formulation among all eight malted drink samples.

Keywords: malted drink, Taguchi's L8 design, sensory evaluation

Determination of Response of Selected Rice Genotypes to Planting Date During *Maha* Season on Selected Agronomic and Yield Characters under Greenhouse Condition

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Ma wee is a group of traditional rice accessions with abiotic stress tolerance. *Ma wee* is grown only during *Maha* season due to the short-day requirement for flowering. This experiment was conducted to determine the response of selected *Ma wee* accessions to planting date (PD) during *Maha* season on agronomic and yield characters. PDs were September 15, 30, October 15, and 30, 2021 with a photoperiod ranging from 12:09 to 11:53. *Maha ma wee* accessions (8541, 8543), one *Ma wee samba* accession (4561), and an improved variety Bg 300 were planted in pots in a complete randomized design with five replicates under greenhouse conditions. Yield and agronomic characters were evaluated. Days to fifth leaf (DFL), Plant height at the fifth leaf (HFL) and Height at flowering (HF), Grains per the first panicle (GPP), and effective tillers at harvest (ETH) were affected by PD. Days to flowering (DF) at PD1 were lesser (124,, 135 and 136) in *Ma wee* in contrast to other PDs while DF of Bg 300 was not affected. GPP was affected by PD as the plants of PD1 produced the highest GPP in all genotypes (225, 269, and 286). The plants of other PDs produced low GPP (from 178 to 64). The above results indicate the optimum PD requirement of *Ma wee* for incorporation in breeding programmes.

Keywords: *Ma wee* accessions, planting date, agronomic and yield characters

Evaluation of Sri Lankan Wild Fruits Based on Free Radical Scavenging Activity, Polyphenolic Content and Cytotoxic Activity

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The study of free radical chemistry has been of recent interest in the scientific community and represents an emerging paradigm in nutraceuticals and disease management. The objective is to incorporate phytochemicals into nutraceutical preparations as an alternative to natural antioxidants, which are being phased out due to possible health hazards and toxicity. This study examined the free radical structure, phenolic content, and cytotoxic nature of different wild fruits (*Syzygium caryophyllatum*, *Careya arborea*, and *Mangifera zeylanica*) in Sri Lanka. Hexane (Hex) ethyl acetate (EA) and aqueous (AQ) fractions were fractionated from crude methanolic extracts (CR) of fruits and assessed for antioxidant activity by 1-diphenyl-1-picrylhydrazyl (DPPH) and ferric reducing antioxidant power (FRAP) tests. The results revealed that EA and AQ fractions of *Careya arborea* fruit showed 95.51 and 115.74 of DPPH radical scavenging percentage, and CR and EA fractions of *Careya arborea* exhibited 341.34 and 261.84 mg TE/g ferric reducing power respectively. The total phenolic capacity content of the evaluated fruit species ranged from 22.8 to 285.3 mg GAE/g dry weight. The present study revealed a strong correlation between free radical scavenging activity and total phenol activity, representing an $R^2 = 0.9989$ value. Moreover, neither plant extracts nor fractions were toxic to a normal Vero cell line. Thus, it was concluded that *Syzygium caryophyllatum*, *Careya arborea*, and *Mangifera zeylanica* species are positive potent free radical resources

Keywords: free radicals, antioxidants, cytotoxicity

Relationship between Selected Anthropometric Parameters and 50m Freestyle Swimming Time in Teenage Swimmers

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Swimming is an Olympic sport, and it is a very popular endurance development activity around the world. Anthropometry is one of the main factors that influence the swimming performance of teenagers. This study aimed to examine the relationship between selected anthropometric parameters and 50m freestyle swimming time in teenage swimmers. The sample was thirty (n=30) provincial level male swimmers 13-17 years of age. Their body weight, height, and BMI were 35.0-96.5 Kg, 136.0-181.9 cm, and 21.81 Kgm⁻². The dependent variable was 50m freestyle swimming time and the independent variable was anthropometric parameters including; body weight, height, length measurements (upper arm, lower arm, hand, upper leg, lower leg, foot, arm span), circumference (chest, abdomen) and skinfold measurements (bicep girth, tricep girth). Data were collected from a 50m freestyle swimming race and measuring thirteen anthropometric parameter sites of the body. Stadiometer, digital weighing scale, skinfold caliper, measuring tape, and the stopwatch was used as measuring instruments. Pearson correlation coefficient in SPSS 26.0V was applied to determine the relationship between 50m freestyle swimming time and anthropometric parameters of sample swimmers at P<0.05 level of significance. As a result, the mean values of swimming time, body weight, and height were 39.9±7.30 seconds, 57.65±15.91 Kg, and 162.56±9.74 cm. The body weight (r=0.376, P=0.041), height (r=0.375, P=0.041) and hand length (r=0.397, P=0.030) had a significant positive moderate correlation with 50m freestyle swimming time. The study concludes that some anthropometric parameters influence the swimming time of teenage male swimmers. Therefore, they have to manage the anthropometric parameters of the body, to achieve their target apart from the other influencing factors related to swimming.

Keywords: anthropometric, freestyle, swimming, teenage male swimmers

A Comparative Study on Phytochemical Screening, Antioxidant Activity and Photoprotective Property of Ethanolic Extracts of Peel and Pulp of Mango (*Mangifera indica* L. cv. TomEJC)

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Less research has been conducted to investigate the bioactivity of peel and pulp of locally grown TomEJC mango cultivar. Therefore, this research was mainly focused on the exploration of phytochemicals, antioxidant activity, and photoprotective properties of ethanolic extracts of the peel of ripe fruit of mango cultivar TomEJC and compared with pulp. The compounds of mango peels and pulp were extracted into ethanol by Soxhlet extraction. Phytochemical screening revealed that proteins, phenols, cardiac glycosides, coumarins, alkaloids, and saponins were present in both peel and pulp. Total phenolic content (TPC), total flavonoid content (TFC), and antioxidant activity of peel and pulp extracts were determined using Folin-Ciocalteu assay, aluminum chloride colorimetric assay, and DPPH free radical scavenging assay respectively. Mango peel showed a higher TPC, TFC, and antioxidant activity (17.69 ± 0.51 mg of gallic acid equivalent per g of dry weight, 13.39 ± 0.43 mg of quercetin equivalent per g of dry weight, 76.96 ± 5.11 µg/mL IC₅₀ value respectively) than that of pulp (10.57 ± 0.57 mg of gallic acid equivalent per g of dry weight, 1.05 ± 0.94 mg of quercetin equivalent per g of dry weight, 282.21 ± 16.60 µg/mL IC₅₀ value respectively) Crude fiber content of mango peel and pulp was found to be 15.66 ± 0.28 % and 4.66 ± 0.76 % respectively. Photoprotective properties of the extracts were determined using a UV Visible spectrophotometric method and the sun protection factors (SPF) of peel and pulp extracts calculated by the Mansur equation were found to be 31.10 ± 1.18 and 2.00 ± 0.20 at 3.0 mg/mL respectively indicating a higher UV-B absorption property of the extract of mango peel. All experiments were carried out in triplicate. As the peel extract exhibited higher antioxidant and photoprotective properties it was analyzed by GC-MS and the results revealed that mango peel was rich in phytochemicals including 2-Furancarboxaldehyde, Docosane, Hentriacontane, Diethyl Phthalate, 1-Octadecene, Heptadecane, and 1-Heptadecene. Results suggested that among the peel and pulp extracts of TomEJC mango cultivar, the peel is rich in phenolics, flavonoids, and phytochemicals with antioxidant and photoprotective properties and thus will be important in herbal nutraceutical and cosmeceutical industries.

Keywords: *TomEJC, antioxidant activity, phytochemical screening*

Forecasting of Female Labour Force Participation Rate Data with Missing Values Imputation, Sri Lanka

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Female Labour Force Participation Rate (Female LFPR) is defined as the proportion of the female labour force to the total working age population. This study is based on the female LFPR quarterly data published by the Department of Census and Statistics, Sri Lanka during the period 2004 to 2021. However, it was found that data for eight quarters in the above time period are missing. The main objective of this study is to forecast female LFPR using ARIMA models by imputing the missing values. In the first part of the analysis, missing values were imputed using nine imputation algorithms available in “*imputeTS*” package in R software. Missing values were generated under four missing rates and thirty random seeds. By comparing MAPE and RMSE plots the Exponential Weighted Moving Average (EWMA) method was found to be the best imputation method. In the second part of the analysis female LFPR were forecasted using ARIMA models. In this analysis, the data were divided into two parts as training and test data. In the training data set, trend, seasonal and random components were identified using “*decompose ()*” function in R software. Furthermore, functions “*arima ()*” and “*auto.arima()*” in library “*forecast*” in R software were used to fit ARIMA models. It was found that ARIMA(1,1,1) model without drift was the best model to forecast the female LFPR which has the minimum AIC value. Errors for the fitted values were calculated using the test data. Female LFPR for the next ten quarters were forecasted using the ARIMA(1,1,1) model. Results showed a small increment in female LFPR at the end of 2022.

Keywords: female LFPR, ARIMA, missing value imputation

Physico-chemical and Microbial Analysis of *Morinda Citrifolia* (Noni/ Indian Mulberry) Fruit Extract

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Indian Noni or Indian Mulberry is the common name for *Morinda citrifolia*, a tropical fruit that is one of the most important traditional medicinal plants used in folk remedies for its broad range of therapeutic and nutritional value. Noni fruit remains an underutilized plant, and scientific studies about its therapeutic and nutritional properties are essentially lacking in the Sri Lankan context. This research mainly focuses on analysing the biochemical and microbial qualities of noni fruit extract because the bioactive compounds present in noni fruit extract and its medicinal power mainly depend on phytochemical constituents. The qualitative phytochemical analysis in the present study has revealed the presence of a broad spectrum of secondary metabolites including Glycosides, Saponins, Flavonoids, Alkaloids, Terpenoids, Anthraquinones, Phenols, and Tannins. Results of physicochemical analysis revealed that the pH of the sample is 4.14 ± 0.1 and brix acid ratio is 20.3% and the total dissolved solids are 84.03 %, indicating that it has a high degree of simple sugar. The total plate count (CFU/g) of the sample is 8.4×10^3 (SLS 516: part 1 - at 30 °C, 72h and the yeast and mold count of the sample is (CFU/g) is 2.7×10^4 (SLS 516: part 2) at 25 °C - 5 days. Though preliminary studies demonstrated potentially beneficial effects, further in vitro and in vivo studies are essential for further development as a commercial product. However, the present study could ultimately help in the sustainable use of underutilized *Morinda citrifolia* fruit to produce a commercially available drink.

Keywords: *Morinda citrifolia*, fruit extract, noni

A Comparative Study on Growth Performance at Seedling Stage in Response to Different Particle Compositions of Coir-based Nursery Substrates

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Coco pith is widely used as a growing substrate and the particle composition of the substrate can substantially affect plant growth and productivity. This study was conducted to investigate the effects of seven different particle compositions; 2 mm/6 mm mix (50/50), 6 mm/2 mm/fine mix (50/30/20), 2 mm/coarse mix (20/80), 2 mm/coarse mix (40/60), 2 mm/coarse mix (50/50), 6 mm/sieved mix/2nd particles mix (60/20/20) and 6 mm/sieved mix/2nd particles mix (40/30/30) of coir-based substrates as the nursery media together with two standard fine and coarse mixtures on nursery growth performance of Chilli pepper (Scotch bonnet), Green chili (MICH HY 1) and Brinjal (Raveena) varieties. Almost all growth parameters (Seedling height [cm], Root length [cm], and Total dry matter content [g]) had diverse responses to substrate blend where the data was analysed using SPSS. According to the results, seedling height and total dry matter of MICH HY 1 and seedling height of Raveena showed a significant effect of substrate blend treatment. Tested mixtures with minimum deviations from two standard mixtures were selected as the best substrate blends with relevance to growth parameters and the crop variety. T₅ (2 mm/coarse mix [20/80]), T₆ (2 mm/coarse mix [40/60]) and T₇ (2 mm/coarse mix [50/50]) blends had best performances over the standards for MICH HY 1 and Raveena. However, the growth parameters of Scotch bonnet did not show significant effects. In conclusion, new particle compositions with the best performance for Green chili and Brinjal could be used to replace the standard mixtures. These findings will be vital for the horticultural industry and enable growers to better cultivation without quality losses.

Keywords: brinjal, chilli pepper, coir-based substrates, green chili, growth performance

Treatment of Aquaculture Effluent through Combined Phytoremediation and Biosorption Techniques

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Aquaculture has significant effects on the environment, the discharge of aquaculture wastewater without treatment causes several environmental impacts such as destruction of natural habitats, reduction in the abundance and diversity of plants and animals, and changes in the quality of soil, water, landscape, and vegetation. Therefore, the treatment of aquaculture wastewater is emphasized. This study focused on the use of phytoremediation techniques and biosorbents to remove contaminants from aquaculture wastewater and enhance the reusability of the treated water. In this context, a mini aquaculture system was developed, a phytoremediation unit with *Lemna minor* plant and a column structure packed with rice hull as biosorbent to treat the wastewater. The aquaculture wastewater was discharged to the phytoremediation unit and then discharged to a column structure packed with a rice hull of 43.2 g. The flow rate was maintained at 10 ml per minute and the Hydraulic Retention Time (HRT) of the phytoremediation unit was maintained for 3 days. The rice hull packed in the column was replaced with the new one weekly to enhance the removal efficiency. The maximum removal of Nitrate, Phosphate, BOD, and COD by the phytoremediation unit was 76.7%, 57.1%, 96.2%, and 67.6%, whilst the column structure packed with rice hull reduced the same at 48.5%, 53.2%, 36.5%, and 51.8% respectively. The performance of the total experimental setup was found to be 78.8%, 72.3%, 96.9%, and 77.5% respectively. The small sample t-test shows the combined phytoremediation and biosorption column setup was significant (p -value < 0.05) in terms of reducing nitrate, phosphate, BOD, and COD from raw wastewater.

Keywords: aquaculture, phytoremediation, bio-sorbent, column

Antibacterial Activity of Ethanolic and Methanolic Extracts of *Trigonella foenum-graecum* (Fenugreek) and *Mentha piperita* (Peppermint) against *Escherichia coli* and *Staphylococcus aureus*

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The significant increase in antimicrobial resistance exhibited by microorganisms has caused the urgency for novel broad-spectrum antibiotic alternatives. This has led to extensive research on the therapeutic potential of medicinal plants, its beneficial effects typically resulting from the presence of plant secondary products. The current study was sought to detect and determine the effect of the antibacterial activity of the ethanolic and methanolic extracts of the seeds of fenugreek (*Trigonella foenum-graecum*) and the leaves of peppermint (*Mentha piperita*) against bacterial strains of *Escherichia coli* and *Staphylococcus aureus*. The antimicrobial susceptibility test, including minimum inhibitory concentration and minimum bactericidal concentration were determined using the well-diffusion and broth-dilution methods, respectively. The tested bacterial strains showed a concentration-dependent growth inhibition towards the plant extracts. The highest mean growth inhibition zones, being 12.5 ± 1.8 mm and 20.6 ± 2.1 mm, were obtained with the 100 mg/mL methanolic extracts of both fenugreek and peppermint, respectively, against *S. aureus*, with a minimum inhibitory and minimum bactericidal concentration of 25 mg/mL and 50 mg/mL, respectively, for the peppermint extract.

Keywords: antibacterial activity, crude extracts, *Mentha piperita*, *Trigonella foenum-graecum*, growth inhibitory effect, *S. aureus*, *E. coli*

Diversity of Pathogenic Bacteria in Commercially Available Mango Fruits in Western Province, Sri Lanka

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Foodborne illnesses are reported every year throughout the world due to contaminations with pathogenic microorganisms such as *Escherichia coli* and *Salmonella*. Preventing contaminations during any stages of a post or pre-harvesting, transportation, and storage conditions is important in reducing the risk of foodborne illnesses. The present study was aimed to determine the pathogenic bacterial diversity and abundance in the fresh mango samples from Moratuwa, Pettah and Wellawatte areas in Western Province, Sri Lanka. Four varieties of purchased mango (n=200) from every 50 samples from commercial markets were used for analysis. Isolated microorganisms were then identified by conventional standard methods using Biochemical keys. Biochemical tests included Indole, Citrate, MRVP, Triple sugar iron agar, Motility test and microbiological culture methods such as MacConkey, Chromogenic culture and Gram staining. Along with the bacterial characteristics of lactose fermentation and non-lactose fermentation, bacterial chemical reaction to each biochemical test and overall colony characteristics on Chromogenic culture media were used for identifying each pathogenic bacterium in mango samples. Distance-Based Redundancy Analysis (dbRDA) was used to identify the distribution pattern of bacterial isolates from different mango collection sites. Seven pathogenic bacterial species namely; *Escherichia coli* (25%), *Shigella sonnei* (25%), *Klebsiella pneumonia* (30%), *Enterobacter intermedius* (15%), *Proteus vulgaris* (5%) and *Salmonella enterica* (30%) were identified from mangoes. As indicated by Distance-Based Redundancy Analysis (dbRDA) on Bray- Curtis Similarity, the bacteria in mango varieties of study regions were significantly different from each other ($\chi^2=556.167$; $P < 0.001$). As indicated by the loadings of the dbRDA axes, the bacterial diversity in mangoes from Moratuwa and Pettah indicated similarity of 72.46%. The dbRDA 1 axis was significantly influenced by the abundance of *E. coli* and dbRDA 2 axis was significantly subjective with the abundance of *Klebsiella oxytoca*, and *Proteus vulgaris*. Poor hygiene and handling practices of the vendors, storage in unsafe containers and in high-temperature for long periods, and unsafe marketing environments were identified as key facts contributing to the pathogenic contamination of mangoes.

Keywords: contamination, culture, isolate, mango, microbiological

Development of a Lignin-Based Flocculant for Algae Harvesting

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Microalgae are photosynthetic microorganisms, which have both harmful and useful aspects. In the presence of nutrients in the water bodies, harmful algal blooms can rapidly develop, affecting the water quality. Therefore, in this study, a cationic lignin polymer as a flocculant was synthesized to harvest harmful microalgae from their water medium, for productive applications. Even though many organic, inorganic, and biopolymers have previously been used as flocculants, cationic lignin polymers have never been used to harvest microalgae. Extracting from coconut husks, or recycling the waste generated in pulp industries are some common ways that lignin can be obtained. Since lignin is abundant in nature and low in cost, it can be considered an economical raw material. The flocculant was synthesized by first increasing the active sites of lignin polymer via a phenol pre-treatment. Then secondary amine groups were introduced to the phenolated lignin polymer via a Mannich reaction and converted into quaternary amine groups. The formation of the flocculant was confirmed using FTIR analysis. Flocculant was then used in jar test experiments to find out optimum conditions required for maximum flocculation efficiency of algae, using water samples collected from Beira Lake, Sri Lanka. A series of six concentrations (0 mg/L, 5 mg/L, 10 mg/L, 15 mg/L, 20 mg/L, 25 mg/L) of the flocculant at three different pH values (pH 7, pH 8 and pH 9) were tested to obtain the optimum condition. At pH 8 a maximum flocculation efficiency of 84% was obtained using 20 mg/L of the flocculant. These preliminary results suggest that the cationic lignin polymer developed in the current study could be used as an efficient flocculant for harvesting algae from water samples.

Keywords: *microalgae, lignin, cationic polymer*

The Antimicrobial Activity of Crude Extract of Pyocyanin from a Marine Isolate of *Pseudomonas sp.* against Clinical Strain of *Staphylococcus aureus*

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Pyocyanin is a water-soluble blue-green, phenazine pigment synthesized by different strains of *Pseudomonas aeruginosa*. It has a powerful inhibitory effect on the bacterial growth and/or biofilm forming ability of the numerous clinically important pathogenic bacterial isolates. Thus, it is also a valuable addition to the existing antimicrobial drugs which can be potentially used as an antimicrobial compound against bacterial biofilm formation. This study aims to determine the antibacterial activity of crude extract of pyocyanin pigment obtained from a marine isolate of *Pseudomonas sp.* cultures against the clinical strain of *Staphylococcus aureus* NCTC6571. The crude pigment was extracted from four days old cultures of *P. aeruginosa* grown in nutrient broth using liquid-liquid extraction with Chloroform: methanol (2:1 (v/v)). The chloroform layer containing pyocyanin blue pigment was collected and concentrated using a roto-evaporator. The antimicrobial activity of crude pigment against *S. aureus* NCTC6571 was determined using the Kirby-Bauer Disk Diffusion assay in Muller Hinton Agar (MHA) plates. Clinical cultures were grown in the presence of pyocyanin-impregnated filter paper disks (7 mm) in MHA overnight at 37 °C. The assay was performed thrice independently. The sharply marinated circle of bacterial growth was observed with *S. aureus* NCTC6571. The diameter of the inhibition zones against *S. aureus* was in the range of 15.5±1.1-16.3±1.2. Further studies are currently in progress to identify minimum inhibitory concentration (MIC) and susceptibility range using purified pyocyanin extract against *S. aureus* NCTC6571. Also, scaling up of pigment production using a low-cost submerged fermentation technique is currently in progress.

Keywords: *Staphylococcus aureus*, antimicrobial activity of pyocyanin, marine isolate of *Pseudomonas sp.*

Molecular Dynamics Study of Dengue Pre-Fusion Conformation

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Dengue is caused by one of four related viruses. Even though few vaccines are already available and more are in the stage of the clinical trial, all of them have their drawbacks. Therefore, developing a vaccine against the dengue virus is among the top priorities in the field. The main objective of this research is to study the dynamics of the pre-fusion conformation of the dengue virus envelope protein to design a vaccine candidate using computational methods. For that, the changes in the dengue virus pre-fusion envelope protein were investigated using Molecular Dynamics simulations. The present study focused on the dengue virus envelope protein, serotype 2 (PDB ID: 10KE), and serotype 3 (PDB ID: 1UZG). RMSF, RMSD, experimental B-factor, and secondary structure profile analyses were carried out using NAMD and VMD software packages to investigate the dynamics of the pre-fusion conformation. Mean RMSD, mean RMSF, and B-factor were higher in serotype 3 when compared to serotype 2. Domain III displayed higher fluctuations in RMSD when compared to other domains. RMSF of the side chain displayed higher values in the positions where glycans are present. The secondary structure profile revealed that both serotypes contain a higher percentage of β sheets. Our preliminary data indicated that the structural stability of serotype 2 is relatively higher compared to serotype 3. Additionally, the lowest structural deviation was observed within domain II of the dengue virus. The observations obtained from this study lays the foundation for the development of a vaccine against the dengue virus.

Keywords: dengue, molecular dynamic simulations, pre-fusion conformation

Analysis of Heavy Metals in Omega-3 Fish Oil Soft Gels Commercially Available in Sri Lankan Market

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Heavy metals (HM) in commercial fish oil (CFO) are one of the major non-clinical disadvantages which may challenge health benefits arising from the omega-3 fatty acids present in CFO. This study focused on the determination of HM levels (Arsenic, Cadmium, Lead, and Mercury) in twelve omega-3 CFO available in the Sri Lankan market and ensuring their safety for human consumption. The levels of As, Cd, Pb, and Hg in CFO were investigated using Inductively Coupled Plasma-Mass Spectrophotometry (ICP-MS) after microwave digestion. The As levels were ranged from 0.004 -3.029 mg/kg in CFO-9 and CFO-7 while Cd were varied from 0.003 – 0.080 mg/kg in CFO-3 and CFO-1 respectively. CFO-6 reported the lowest Pb (0.043 mg/kg) while CFO-1 reported the highest Pb (0.239 mg/kg). The highest Hg level was found in CFO-1 at 0.003 mg/kg and CFO-12 accounted lowest Hg level (0.001 mg/kg). When compared with Omega-3 Trade Association standards (<0.1 mg/kg for each metal) maximum accepted level (MAL) for As were exceeded in CFO-2, CFO-7, and CFO-12 while CFO-1 and CFO-2 exceeded MAL for Pb. Moreover, in all samples, Cd, Pb, and Hg levels were below the MAL values (1.0, 3.0, and 0.1 mg/kg, respectively) recommended by the European Pharmacopeia. Even though As and Pb values of some samples exceeded the MAL, the calculated daily intakes for As, Cd, Pb, and Hg in all CFOs were below the permissible daily intakes recommended by Food and Agriculture Organization and World Health Organization, thus indicating that the studied CFO brands are safe for human consumption.

Keywords: *commercial fish oil, heavy metals, ICP-MS*

Parasite Profile and Trends in Parasite Composition in Yellow-Billed Babblers (*Turdoides affinis taprobanus*) in Low Land Wet Zone, Sri Lanka

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The study's primary objectives were to characterize the parasitic spectrum in Yellow-billed Babblers and investigate trends in parasitism in individuals from different habitat types, flock sizes, and according to the level of host maturity. Thirty-seven birds were sampled from 18 different locations in Sri Lanka's Low Land Wet Zone from November 2021 to May 2022. Identifying and quantifying gastrointestinal parasites from faecal samples were done using direct smears and concentration techniques. Thin, thick, and buffy-coat blood smears were prepared to conduct microscopic identification of haemo-parasites. Ectoparasites were mounted on glass slides for identification. Data were analyzed using Minitab-21 and SPSS-26 software. Parasitic prevalence in the Yellow-billed Babblers was 68%. Five intestinal parasitic species (two protozoans; *Isospora* spp, *Entamoeba* spp, 3 nematode species; *Trichuris* spp, *Strongyloid* spp, and hookworm type eggs), one haemoparasite species (*Trypanosoma* spp) and two ectoparasite species (Family – Philopteridae and Ixodidae spp) were identified. 51.4 % were infected with gastrointestinal parasitic species, 16.2% were infected with ectoparasites and 2.7% were infected with haemoparasites. Among the gastrointestinal parasites, protozoan infections (37.8%) were about two times higher than nematode infections (18.9%). Sub-adults and individuals from large flocks had a high prevalence of parasites. Overall parasitic prevalence was highest in the sub-urban areas while ectoparasitic infections were most prevalent in urban habitats. The only blood parasite recorded, the *Trypanosoma* spp found from the sub-urban habitat is a notable finding as the trypanosomes have zoonotic potential. The study depicts the importance of investigating the parasitic profiles of wild animals in order to prevent zoonoses.

Keywords: yellow-billed babbler, urban, ectoparasites, gastrointestinal parasites, haemo-parasites

Knowledge and Motives towards the Functional Food and Nutraceutical Consumption by Adults in Sri Lanka

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Functional foods and nutraceuticals (FFN) play an important role in adults' health by preventing and reducing the risk of non-communicable diseases (NCD). This cross-sectional study was designed to evaluate knowledge, attitude, and behaviour (KAB) towards the consumption of FFN by Sri Lankan adults. The study sample comprised of 441 adults above 20y of age living in Sri Lanka. A pre-tested online questionnaire was used for data collection and was analysed using SPSS 23 and AMOS software. Out of the participants, 60% were female and 74% were in 20 – 29y age category. Significant relationships were observed between knowledge and attitude ($p < 0.01$), knowledge and behaviour ($p = 0.004$), and behaviour and FFN consumption ($p = 0.003$). District of residence, education level, income, and use of medications for NCDs, significantly affected to the knowledge of FFN consumption. However, the above socio-demographic factors did not significantly affect towards attitude or behaviour. Gender and age did not significantly affect the KAB towards FFN. Average scores of knowledge, attitude, and behaviour were 72,72, and 71%, respectively. More than 50% of the respondents consumed plant-based functional foods, fruits and vegetables, and functional beverages at moderate levels while their nutraceutical consumption was inadequate. Vegetable curries, black tea, green leaves, dhal, and unpolished rice were identified as the top five foods consumed daily. In conclusion, knowledge of FFN affects the attitude and behaviour towards FFN consumption. Socio-demographic factors are associated with the knowledge of FFN consumption. It is required to make people aware of the health benefits of FFN and encourage FFN consumption.

Keywords: attitude, behaviour, functional food, knowledge, nutraceuticals, Sri Lanka

Evaluation of the Antibacterial Activity of Miswak (*Salvadora persica*) and Persian Lime (*Citrus latifolia*) Extracts against *Escherichia coli* and *Staphylococcus aureus*

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Despite advances in medicine, the phenomenon of emerging drug resistance provokes novel research on active botanical compounds and alternative therapy development. Bioactive compounds present in plants possess potent antibacterial properties. The current study aims to evaluate and compare the bacterial susceptibility between miswak (*Salvadora persica*) and Persian lime (*Citrus latifolia*) extracts; a novel combination. Miswak sources unique phytochemicals, making it a superior tool for oral hygiene, while the volatile oil harbored within lime is traditionally used as a flavouring and cosmetic agent. Crude extraction of phytochemicals was done via cold maceration, employing polar solvents methanol and ethanol. Varying concentrations (150 mg/mL and 200 mg/mL) of extracts were subjected to antibiotic susceptibility testing (ABST) using agar well diffusion, while gentamicin and vancomycin served as positive controls. Both *Escherichia coli* and *Staphylococcus aureus* exhibited susceptibility toward all extracts that were assayed. Triplicate readings were statistically analyzed using a two-way analysis of variance (ANOVA) and student's t-test with a 95% confidence interval ($p \leq 0.05$). Mean zones of inhibition (ZOI) were wide-ranging, from 10.7 ± 0.6 mm to 13.7 ± 0.6 mm for miswak and 16.7 ± 0.6 mm to 19.7 ± 1.2 mm for lime. Methanolic lime of 200 mg/mL (M/L2) demonstrated a pronounced ZOI against *E. coli* (19.7 ± 1.2), proving its supremacy over miswak. Upon further testing, lime extracts displayed a minimum inhibitory concentration (MIC) at 12.5 mg/mL and a minimum bactericidal concentration (MBC) at 25 mg/mL. Nonetheless, based on overall results, both miswak and lime extracts serve as potential candidates that can be developed into therapeutic drugs in the phytopharmaceutical industry.

Keywords: *miswak, persian lime, ABST, MIC, MBC*

Evolutionary and Ecological Characterization of the Laughingthrush of Sri Lanka

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The Ashy-headed Laughingthrush, an endemic bird to Sri Lanka, has been historically considered a laughingthrush in the genus *Garulax*. However, recent phylogenetic studies have suggested a new classification for the species under the genus *Argya* (true babblers). However, as the name suggests, the Ashy-headed laughingthrush shows remarkable morphological similarities to other laughingthrushes, despite the genetic signal showing evolutionary affinity to true babblers. This study attempted to address these seemingly divergent signals in the Ashy-headed Laughingthrush by reconstructing the molecular phylogeny of babblers and laughingthrushes of the Indo-Himalayan region and comparing it with key morphological, plumage features of the two groups. The phylogenetic relationships were reconstructed using the Bayesian inference using four mitochondrial and seven nuclear gene regions. Hierarchical clustering was performed to assess the phenotypic characters. The phylogenetic tree placed the Ashy-headed Laughingthrush with the genus *Argya*, with the *A. malcolmi* (Great Grey Babbler) of India being the sister taxa. Morphometrics and plumage placed Ashy-headed Laughingthrush with laughingthrushes in the phenotypic clustering. Therefore, a discordance is evident between phenotype and phylogeny. This discordance suggests a rapid phenotypic divergence from babblers and convergent patterns of evolution with laughingthrushes of the Indo-Himalayan region. Our findings agree with the recent placement of Ashy-headed Laughingthrush in *Argya* based on phylogenetic evidence despite seemingly divergent phenotypic signals. We think that the convergence of phenotypic evolution has historically confused the taxonomic delimitation of the Ashy-headed Laughingthrush of Sri Lanka.

Keywords: babblers, convergent evolution, morphology, plumage, phylogeny

Survey on How Many People in Sri Lanka are Interested in Fish Oil Dietary Supplements

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Consumption of fish or fish oil containing ω -3 polyunsaturated fatty acids (PUFAs) reduces the risk of coronary heart diseases, decreases mild hypertension, lowers the incidents of diabetics, and is important during pregnancy as a critical building block of foetus brain and retina. Therefore, fish oil dietary supplements have attracted more attention from people around the world recently. The present study attempts to gain an understanding of Sri Lankan people's interest in and awareness of fish oil dietary supplements. A questionnaire was used to collect the most reliable and accurate data from 200 individuals. In order to gather useful and relevant information, the questionnaire contained various components including general information about participants, fish consumption pattern, fish oil dietary supplement usage, diseases, etc. A recent survey indicates that Sri Lankan people have generally less interest in fish oil dietary supplements. According to the data analysis, only 10% of people use fish oil dietary supplements and 85% of them are between 20-30 age limits. Moreover, most of them eat fish usually in their diets and non-fish eaters do not take both fish and fish oil dietary supplements. Omega-3 fatty acids play a vital role in the human body. It has been suggested that non-consumers of fish should take fish oil as a dietary supplement to fulfill their omega-3 requirement. Based on the findings, this research can contribute towards informing the general public about fish oil dietary supplements and their accurate usage.

Keywords: *fish oil, fish consumption, dietary supplements, Omega-3 fatty acids*

A Preliminary Study Conducted on a Traditional Formulation used for the Treatment of Dengue

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Currently, around the globe, there are no effective drugs for the treatment of acute dengue infection. Even though a vaccine was licensed recently, it was found to have limited efficacy. Scientists are currently seeking for a novel treatment for dengue. Bamboo Swarasa is a Sri Lankan traditional medicinal preparation made from shoots of yellow and green common bamboo (*Bambusa vulgaris*) extracts together with Palmyra sugar. This preparation is currently given to patients with dengue fever. Initially, we assessed the phenolic, flavonoid and antioxidant content of this preparation by Total Phenolic Content (TPC), Total Flavonoid Content (TFC), 1,1-diphenyl-2-picrylhydrazyl assay (DPPH). We also analysed the anti-inflammatory activity by the human red blood cell membrane stabilization (HRBC), and the egg albumin assays. Finally, the cytotoxicity of this preparation was assessed by the MTT assay. When 75mg/ml of sample preparation was compared to Gallic acid (GA) the total phenolic content was found to be 5mg/g GA equivalent. The total flavonoid content of 75mg/ml sample was 0.02mg/g Quercetin equivalent. For the DPPH assay, the highest percentage radical scavenging value was reported to be 91.79%, at the maximum concentration (75 mg/ml) tested and the IC₅₀ was found as <3.75mg/ml. The anti-inflammatory activity of the preparation was found to be 46.09% inhibition of protein denaturation at 75mg/ml sample while Ibuprofen showed a similar inhibition: 51.30%. The HRBC assay depicted 52.85% inhibition of red cell lysis at 75mg/ml, while Ibuprofen showed 97.20% inhibition. Finally, a moderate cytotoxic effect was noted in Vero cells by the MTT assay where a concentration of 10µg/ml depicted a ~35% cell growth inhibition at 24h. This commercial preparation of Bamboo Swarasa demonstrated excellent antioxidant activity and moderate anti-inflammatory activity at a concentration of 75mg/ml. Viral infections are usually accompanied by oxidative stress, which plays an important role in their pathogenesis, and antioxidants are able to reduce the reactive oxygen species level in infected cells. Thus, this preparation could be an effective medicine for viral diseases such as dengue. This work provides only preliminary scientific evidence, and further research will be conducted to determine its antiviral activity.

Keywords: *Bambusa vulgaris*, dengue fever treatment, traditional plant extract

Optimization of yeGFP Expression as a Reporter for Prospective RNAi Analysis in *Pichia pastoris*

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Green fluorescent protein from the jellyfish *Aequorea victoria* is a widely used reporter gene in genetic engineering. Yeast codon optimized green fluorescent protein (yeGFP) has been found to have higher expression, and hence it is more reliable as a yeast reporter. *Pichia pastoris* can be used as a model organism to study gene regulation such as RNA interference (RNAi) because it does not have RNAi, by monitoring the yeGFP expression. This study focuses on the optimization of the expression of yeGFP in the *Pichia pastoris* GS115 strain, under the control of AOX1 promoter for prospective RNAi studies. yeGFP gene was cloned into the pPICZ A vector to create pPICZ A-yeGFP expression plasmid. *P. pastoris* GS115 strain was transformed with pPICZ A-yeGFP and positive transformants were selected on Zeocin plates and further confirmed by PCR. Five transgenic colonies were tested for the yeGFP expression by methanol induction in MMH (Minimum Methanol Histidine) media containing 0.5%, 1%, and 2% (v/v) methanol for six days. Fluorescence intensities were measured using SpectraMax M3 spectrophotometer at an excitation wavelength of 395 nm, and emission of 510 nm. According to the Relative fluorescence units of the induced cultures, expression increased with the time till the fifth day and decreased on the sixth day. 0.5% methanol and 2% methanol-induced cultures showed the least and highest fluorescence respectively. Transgenic lines 1 and 4 had the highest yeGFP expression in 2% methanol on the fifth day. For studies on gene regulation by RNAi, optimum yeGFP expression under the AOX1 promoter can be measured in 2% methanol on the fifth day of induction.

Keywords: *Pichia pastoris*, yeGFP, methanol induction

