VOL.
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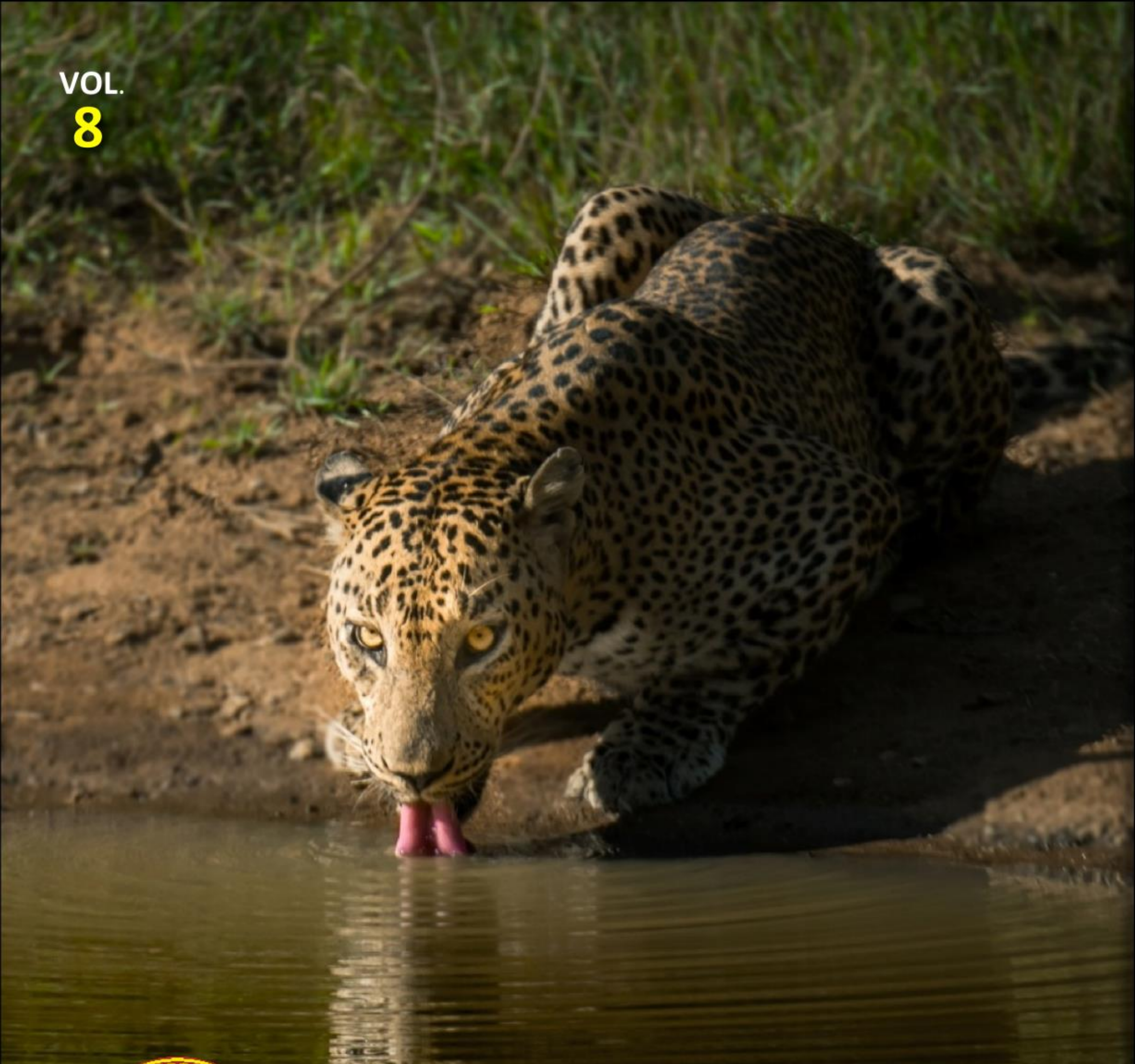
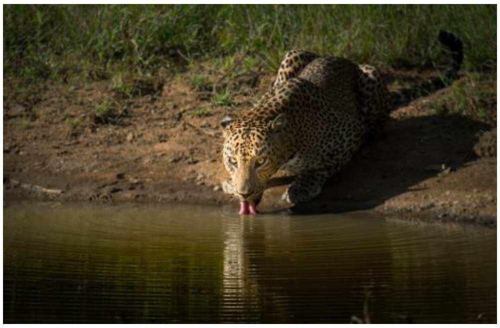
POSTGRADUATE RESEARCH HIGHLIGHTS



Postgraduate Institute of Science
University of Peradeniya
Sri Lanka

RESEARCH:
Ph.D., M.Phil., & M.Sc. (SLQF 10)

2023



Cover Image: *"Scientists, like animals searching for water, have an unending thirst for knowledge."*

The cover page displays the Sri Lankan leopard, *Panthera pardus kotiya*, (also recognized as the Ceylon leopard), drinking from a puddle of water. This endemic subspecies in Sri Lanka is the only remaining native wild cat on the island. These leopards are known for their adaptability to various habitats, including dry evergreen forests, scrublands, and rainforests, and serve as one of the apex predators in Sri Lankan terrestrial ecosystems. *Panthera pardus kotiya* is considered a flagship species that not only contributes to biodiversity conservation but also fosters the growth of ecotourism. It is classified as an endangered species, primarily due to habitat loss and human-leopard conflicts. Conservation efforts have been undertaken to protect their natural habitats and reduce conflicts with humans.

Cover Design: Dr. Kapila Wijayaratne and Dr. Ashwini Amarasighe, Faculty of Science, University of Peradeniya.

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PGIS Research Highlights 2023
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POSTGRADUATE INSTITUTE OF SCIENCE
UNIVERSITY OF PERADENIYA
SRI LANKA

POSTGRADUATE
RESEARCH HIGHLIGHTS 2023

3rd and 4th November 2023

PGIS POSTGRADUATE RESEARCH HIGHLIGHTS 2023

ISBN 978-955-8787-09-0

Published by

Postgraduate Institute of Science (PGIS)
University of Peradeniya
Peradeniya, Sri Lanka

MESSAGE FROM THE DIRECTOR

The Postgraduate Institute of Science (PGIS) at the University of Peradeniya has been a hub of excellence, providing a wide range of postgraduate programmes leading to diplomas, M.Sc., M. Phil and Ph.D. degrees. Our commitment lies in fostering a global reputation for innovation through exceptional scientific research across various disciplines. At PGIS, we facilitate advanced scientific research facilities and environment conducive to achieve the goals of both Institute and society.

Our students' research endeavors occur across many well-equipped laboratories within PGIS, various faculties at the University of Peradeniya, and other prominent national universities and research institutes. PGIS has recently implemented a research grant program aimed at providing additional support to our students. Furthermore, PGIS actively engages in collaborative research with eminent scholars from various overseas universities and research institutes. PGIS is pleased to present the publication "PGIS Postgraduate Research Highlights - 2023" as part of our mission to enhance visibility and industry outreach. This publication of outstanding discoveries made by Ph.D. and M.Phil. graduates. Many of these findings have already been published in reputable journals, and several of our researchers have received prestigious awards for their exceptional contributions to their respective fields.

I express sincere gratitude to Prof. W.A. Priyanka P. De Silva, the Editor-in-Chief of this publication, and the review group members for their invaluable contributions. I am grateful to all M. Phil and Ph.D. holders from PGIS and their supervisors for providing summaries of their research work and consenting to their publication.

Prof. H.M.T.G.A. Pitawala
Director, Postgraduate Institute of Science
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MESSAGE FROM THE EDITOR IN CHIEF

It gives me immense pleasure to introduce the compilation of the PGIS Research Highlights 2023. The accomplishment of this compilation is a direct result of the invaluable contributions from each member of the Committee. I would like to express my deepest gratitude to all the M.Phil and Ph.D. graduates, along with their dedicated supervisors, whose exceptional research has transformed this vision into a tangible reality.

During the 2021 convocation, we proudly celebrated the achievements of 33 students who received their Ph.D. and M.Phil. degrees., showcasing outstanding research outputs. This publication serves as an additional channel for disseminating the wealth of knowledge produced by our postgraduate researchers during the 2020/21 academic period. Those interested in exploring the topics covered here are encouraged to contact the respective authors or their supervisors. I am confident that this publication will serve as a valuable resource for fostering collaborative research endeavors, both locally and internationally.

I wish to extend my heartfelt appreciation to the Postgraduate Institute of Science for their steadfast support of these research initiatives. A special note of gratitude goes to the Director of the PGIS, Prof. H.M.T.G.A. Pitawala, the Chairperson of the RESCON 2023, Dr. T. C. Weeraratne, and the Secretary of the RESCON 2023, Dr. Kapila Wijeyaratne, alongside all the dedicated members of the Editorial Committee of RESCON Research Highlights - 2023, for their indispensable assistance in bringing this publication to fruition. Finally, I extend my best wishes to the RESCON 2023 team for the success of the conference.

Prof. W. A. Priyanka P.de Silva
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Contents

MOLECULAR PHYLOGEYLOGENY AND TAXONOMIC REVISION OF SELECTED JUMPNG SPIDER GENERA (ARANEAE: SALTICIDAE) OF SRI LANKA

A. Satkunanathan *graduated in 2017 with a B.Sc. Special in Agricultural Resource Management and Technology with Second Class Upper Division Honors from the University of Ruhuna, Sri Lanka. Soon after her graduation, she worked as a demonstrator at the Department of Animal Science, Faculty of Agriculture at the University of Ruhuna, Sri Lanka. Later, in 2019, she joined the National Institute of Fundamental Studies, Kandy, Sri Lanka, as a Research Assistant, initiating her research career. In 2022, she completed her M.Phil. by research from the Postgraduate Institute of Science (PGIS), University of Peradeniya, Sri Lanka.*

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Salticidae (jumping spiders) is the most diverse spider family and includes species most encountered in many habitats (WSC, 2022). Sri Lanka together with the Western Ghats is one of the 34 biodiversity hotspots with a highly diverse endemic fauna arising from most probably from autochthonous diversification (Myers. *et al*., 2000). Sri Lanka is known for several unique lineages of jumping spiders and Sri Lankan salticid diversity is represented by 149 species placed in 66 genera, including 58 endemic species (Benjamin *et al*., 2012; WSC, 2022).

The majority of salticid genera lack proper descriptions and revisions. Comprehensive molecular phylogenies, evolutionary relationships, and distribution patterns of Sri Lankan Salticidae have rarely been studied. Therefore, it is essential to incorporate the island's biodiversity as representatives in a global Salticidae phylogeny.

The main objectives of the current study were to investigate the systematics of selected jumping spider genera in Sri Lanka through the use of molecular phylogenetic analysis and morphology.

To achieve the aims, study was conducted with the following Specific objectives:

(I) Taxonomic revision of the Sri Lankan endemic jumping spider genus *Epidelaxia*, including the phylogenetic placement of the genus in the salticid tree of life.

(II) Taxonomic revision and phylogenetic placement of Sri Lankan species of the jumping spider genera, *Carrhotus*, and *Telamonia*, including the phylogenetic placement of the two genera in the salticid tree of life.

(III) Provision of distributional records of Sri Lankan species of the Jumping spider genera *Epidelaxia*, *Carrhotus*, and *Telamonia*.

Methodology

Specimens were collected from all major climatic-physiographic zones of Sri Lanka, primarily through methods such as beating vegetation, hand collection and sifting litter. Spiders from the sieved litter were extracted using a Winkler extractor. The collected spiders were preserved in either 70% or 100% ethanol, depending on taxonomic and molecular purposes. Collected specimens were examined using an Olympus SZX stereo microscope and identified using recently published works and available databases.

Male palps and female epigastric regions were observed and illustrated with the aid of an Olympus BX51 compound microscope attached with a drawing tube. Digital images of the specimens were taken with a Leica MC170 HD camera mounted on a Leica M205C stereomicroscope using the software package Leica Application Suite, LAS version 4.6.2. Acquired image stacks of different depths were assembled using Helicon Focus (version 6, Helicon soft Ltd).

For molecular analysis, genomic DNA was extracted from 100% ethanol-preserved leg tissues using the DNeasy® Blood and Tissue Kit (Qiagen). The polymerase chain reactions (PCR) were carried out using 15 primer pairs. PCR products were purified using the Gene Clean TM Turbo Kit. All purified PCR products were Sanger sequenced in both direction by MACROGEN (Seoul, South Korea). Sequences were edited and assembled using Geneious 11.0.2 software package. Sequence alignment was done by Geneious 11.0.2 using default parameters. Mesquite (version 3.51) software was used to edit the sequence manually. The appropriate evolutionary model for each partition for individual and concatenated datasets was determined by the program PartitionFinder2. Maximum likelihood and Bayesian analysis were performed using RAXML and MrBayes v 3.2.6 respectively. Parsimony analysis of the morphological data matrix was done using TNT 1.1

Molecular phylogeny of tribe Nannenini was based on a combined molecular data set of H3+CO1+28S+18S gene fragments, and for Salticini and Plexippini, COI+28S gene fragments. Morphological phylogeny of tribe Nannenini was based on 61 morphological characters coded for 17 taxa.

Results and Discussion

The total length of the final matrix of Nannenini was 2492 bp. Out-groups consisted of distantly and closely related taxa to *Epidelaxia* and *Genus X.* The out-group taxa are from the 20 taxa belonging to 8 tribes.

The monophyly of tribe Nannenini was robustly supported by *28S*-single gene and concatenated all genes (*COI+28S+18S+H3*) ML analysis. All individual gene trees (*COI*, *28S*, *18S*, *H3*) and concatenated all gene analysis corroborated the monophyletic origin of genus *Epidelaxia*. The individual gene trees (*COI*, *28S*, *18S*) and concatenated gene analysis corroborated the monophyletic origin of *Genus X.*

In the Tribe Salticini likelihood analysis, the sequence length was 1336 bp, involving 34 taxa (16 ingroup and 18 outgroup). The phylogenetic tree, obtained from Maximum Likelihood (ML) analysis of the combined data matrix, supports the monophyly of the jumping spider genus Carrhotus. Our phylogenetic hypothesis confidently places Carrhotus within the tribe Salticini, belonging to the subfamily Salticinae. With three known species and three new species, Carrhotus exhibits significant diversity within Sri Lanka

Tribe Plexippini likelihood analysis, the sequence length was 860bp and included 23 taxa (17 ingroups and 06 outgroups). The phylogenetic tree obtained from ML analysis of the combined data matrix recovers a well-supported *Telamonia* clade. This clade includes the monophyletic *Telamonia* and resolves as a sister clade to the other taxa in tribe Plexippini.

We revised all the members of the genera *Epidelaxia*, *Carrhotus*, *Telamonia* of Sri Lanka and re-described *Epidelaxia albocruciata*, *Epidelaxia albostellata*, *Epidelaxia obscura*, *Carrhotus silanthi*, *Carrhotus taprobanicus*, *Carrhotus viduus* and *Telamonia sponsa.* We further reported that, *Epidelaxia* sp. C, *Epidelaxia* sp. G, *Genus X.* sp. A, *Genus X.* sp. B, *Carrhotus atratus*, *Carrhotus albosetosus*, *Carrhotus lobatus* as endemic species in Sri Lanka.

The species introduced through this study bear immense importance as they are mostly endemic and also for their localized distribution in the native forests of Sri Lanka. The conservation of the remaining patches of montane, sub-montane, and lowland wet forests is, therefore, critical in this regard. These high-elevation cloud forests are referred to as hotspots within hotspots. Human-caused climate changes may also have high impact on the abundance and distribution of these endemic species. Thus, due to the plethora of environmental destructions, it’s high time to implement necessary measures to protect these habitats to protect the spider fauna of Sri Lanka. This study emphasizes the importance of conducting, investigating, and documenting the spider diversity in Sri Lanka as well as the collaborations with national-level institutes such as the Forest Department and the Department of Agriculture, the Department of Wildlife Conservation, IUCN, and the National Museum of Sri Lanka.

Conclusion

Based on multi-locus molecular dataset (*H3+COI+18S+28S*) and 61 morphological characters this study provides the first internal phylogenetic structure of the *Epidelaxia* and its placement within the tribe Nannenini. The existence of a new genus, *Genus. X* which shows close affinity to *Epidelaxia* was revealed. Further, this study also adds four new species and highlights the evolutionary history of *Epidelaxia* and *Genus X*.

The monophyly of the genus *Carrhotus* was resolved as a sister clade to the other taxa with the tribe Salticini. The outcomes add three new species to the existing list while demonstrating the evolutionary history of *Carrhotus*, confirming its phylogenetic placement within the tribe Salticini through molecular analysis. *Telamonia sponsa* was also identified as a monophyletic group, and for the first time, the molecular analysis revealed the phylogenetic placement of *Telamonia* under the tribe Plexippini.

Source of Funding

This research work was financially supported by the National Institute of Fundamental Studies, Sri Lanka.

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THE CURRENT STATUS OF SECONDARY LEVEL SCIENCE EDUCATION IN SCHOOLS WITH INDIGENOUS STUDENTS IN UVA PROVINCE OF SRI LANKA

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Science plays a crucial role in propelling social, economic, and environmental development in the modern world. Science education is the process that shares the science culture among the public. However, contemporary literature depicts standard science that is commonly taught in schools has a Western perspective. Therefore, students from non-Western cultures, especially Indigenous students whose home culture is very different from school science culture, are alienated from science learning. Sri Lanka is also a non-Western multicultural country with a distinct group of Indigenous people who inhabit the island for about 30,000 years. These Indigenous people are called Vedda people. Vedda people are also considered under compulsory education laws in Sri Lanka. Hence, Vedda children learn alongside mainstream students making those classes unique and multicultural. However, research on the formal education of Vedda students is scarce and literature on science education of Vedda students is almost non-existent. Therefore, this study aimed to explore the status of science education in three selected secondary level schools with Vedda students in Uva Province, Sri Lanka.

The objectives of the present study were,

(I) To explore the status of secondary level science teaching and learning process in the three selected schools with Vedda students in Uva Province, Sri Lanka

(II) To identify constraints for secondary level science education for students in the three selected schools with Vedda students in Uva Province, Sri Lanka

(III) To collect suggestions from parents, students, science teachers, and Vedda leaders/ senior community members to enhance secondary level science education of students in the three selected schools with Vedda students in Uva Province, Sri Lanka

(IV) To identify best practices in Australia regarding science education of Aboriginal and Torres Strait Islander peoples to enhance science education of Vedda people.

Methodology

This study was carried out in three schools associated with two Vedda settlements in Uva Province, Sri Lanka, from 2018 to 2021. A mixed methods research approach with an inductive convergent mixed methods research design was employed predominantly in data collection and analysis. Qualitative data were collected through science lesson observations, semi-structured interviews (conducted with Vedda parents, Vedda students, mainstream parents, mainstream students, science teachers, and Vedda leaders/ senior community members), and selected documents. Questionnaire surveys were used to collect quantitative data from Vedda parents, Vedda students, mainstream parents, mainstream students, and science teachers associated with the three selected schools. Qualitative data were analyzed with the thematic analysis method (documents were analyzed independently from science lesson observations and semi-structured interviews). Descriptive statistics were used to analyze quantitative data. The methodology paid attention to research ethics. Further, necessary measures were taken to ensure the reliability and validity of quantitative (Cronbach's Alpha for reliability) data and the trustworthiness of qualitative data.

Results and Discussion

Convergent analysis of quantitative and qualitative data generated five shared themes namely, view towards general education and science learning, support towards general education and science learning, challenges for science teaching and learning, suggestions to enhance science learning of Vedda students and mainstream students, and socioeconomic factors of parents.

Apart from the shared themes, two unique themes were identified. Those two themes were good interaction between Vedda students and mainstream students and poor concern of authority on education of Vedda people.

In addition, document analysis resulted in two themes, namely incorporation of Indigenous histories and cultures into the science curriculum and development of participation and achievement of Indigenous students in science through a variety of programs. The above themes were used to address the objectives of this study.

Status of secondary level science teaching and learning process in the three selected schools

Results revealed that both Vedda and mainstream parents expressed positive attitudes towards general education and keen on providing formal education to their children. They identified general education to secure a better future for their children. Vedda parents also considered, general education is crucial for their engagement with the broader society. However, they tended to less involved with general education of their children compared to mainstream parents.

Both Vedda parents and mainstream parents had a limited understanding of school science, with Vedda parents being particularly unable to provide academic support for their children's science education. Furthermore, science education followed a traditional teacher-centred approach.

Although there was no evidence of discrimination against Vedda students, they did not receive specialized attention and tailored teaching methods that accounted for their cultural diversity. While both Vedda and mainstream students showed interest in learning science, a higher percentage of Vedda students found science as a challenging subject to learn.

Constraints for secondary level science education for students in the three selected schools

The low education and economic status of Vedda parents impede their ability to provide sufficient academic support and additional learning opportunities in science to their children. This results in many Vedda students relying solely on their school science teachers for learning. The schools also face challenges in terms of inadequate scientific equipment and chemicals, lack of water and electricity in science laboratories and shortages of science teachers. The heavy science syllabus limits the time available for student-centred teaching. Similar to many indigenous groups globally, Vedda parents, experience financial constraints that hinder their capacity to offer necessary physical resources and learning opportunities for their children. Furthermore, issues related to distance and transportation make accessing educational facilities difficult.

Suggestions to enhance secondary level science education of students.

Suggestions made by the Vedda parents were not specifically on developing science education of the respective schools. However, the students at school "A" showed their interest in science learning by requesting a permanent science teacher to their school. The lack of suggestions emphasizing science education could be due to multiple reasons such as poor understanding of Vedda parents and students on school science. Most of the suggestions made by the Vedda parents were on developing physical resources. In contrast, mainstream parents suggested to improv more learning opportunities. These suggestions depict that the mainstream parents are more concerned about the education of their children compared to Vedda parents.

The suggestions made by the mainstream students on developing physical and human resources to enhance science education confirmed the limited resources available in their schools. Suggestions made by the Vedda leaders/ senior community members highlight the importance of teachers being familiar with their culture. Science teachers also emphasize the value of incorporation of culturally relevant practices in teaching science. Hence, it would be worthy to consider incorporation of cultural practices of Vaddha community to science teaching and learning process. Furthermore, science teachers stated that activities such as science camps and science seminars could be beneficial to motivate Vedda and mainstream students towards learning science.

Best practices in Australia regarding science education of Aboriginal and Torres Strait Islander peoples

Incorporation of Indigenous cultures and histories into the school science curriculum and delivery of various targeted programs to promote STEM education of Indigenous students are two best practices that could be identified from the Australian context to enhance the science education of Vedda people. Australia has paid much attention to closing the achievement gap between Indigenous and mainstream students in science through the above actions. Further, the incorporation of Indigenous histories and cultures provides opportunities to learn science in a more culturally relevant manner. To support culturally relevant science lessons, science teachers in Australia are provided with elaborations and background information on histories and cultures of Aboriginal and Torres Strait Islander peoples. In contrast, Sri Lanka has not paid enough attention to providing culturally relevant science education to the Vedda people. Similar to Australia, Sri Lanka also could make necessary arrangements to incorporate Vedda history and culture in the secondary level science curriculum and support science teachers by providing guidance and supplementary materials to conduct culturally relevant lessons.

Conclusion

It can be concluded that Vedda students at the above schools were interested in learning science. Still, no special methods were used to teach science to "Vedda" students and science education of "Vedda" students hindered by the low educational attainment and poor economy of "Vedda" parents. Incorporation of Indigenous content in the science curriculum and delivery of different science programs could be adopted to enhance the science education of "Vedda" students.

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MORPHOMETRY-BASED RADIOMICS FOR PREDICTING THERAPEUTIC RESPONSE FOR GLIOMAS AND PREDICTING PROGNOSIS FOR SOFT TISSUE SARCOMAS OF EXTREMITIES IN RADIOTHERAPY

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Cancer is a leading cause of death worldwide, and globally, about every sixth death is due to cancer. Despite its high incidence and high mortality, the challenges in accurate detection, characterization, treatment and monitoring of cancer make it difficult to achieve the desired effectiveness to reduce morbidity and mortality. Especially cancers like gliomas and soft tissue sarcomas of extremities (STSE) require better management approaches to improve survival rates while reducing morbidity and mortality.

Approximately half of all cancer patients require radiography upon diagnosis. Imaging plays an essential role in radiotherapy treatment, including treatment planning, evaluating therapeutic response, and assessing patient prognosis. The preferred imaging modality for assessing gliomas is magnetic resonance imaging (MRI), while for evaluating soft tissue sarcomas of extremities (STSE), a combination of MRI, computer tomography (CT), and positron emission tomography (PET) is use.

The high-throughput extraction and analysis of image features from radiological images, known as radiomics, is a promising approach in precision medicine that has emerged in recent years. This approach has a significant impact on the delivery of individually tailored radiotherapy treatments to improve clinical outcomes. However, only a limited number of radiomics studies have been conducted to assess the therapeutic response and prognosis in patients after receiving radiotherapy.

The objectives of the present study were,

(I) To develop a morphometry-based radiomics signature (Rad-score) for patients with glioma after radiotherapy
(II) To determine the potential of morphometry-based radiomics signature (Rad-score) as a potential biomarker for the prediction of treatment response in patients with gliomas after radiotherapy

(III) To determine the potential use of morphometry-based radiomics features to predict the prognosis of patients with soft tissue sarcomas of extremities after radiotherapy.

Methodology

Predicting therapeutic response in patients with glioma following radiotherapy.

Initially, 110 patients with pathologically confirmed gliomas following radiotherapy were retrospectively evaluated. MRI scans were done prior to treatment and had received radiotherapy treatment for the primary tumor site. This study used the segmented glioma regions of interest (ROIs), deidentified images and clinical data obtained from Brain Tumor Segmentation datasets, The Cancer Imaging Archive (TCIA) and Genomic Data Commons Data Portal in compliance with their data usage policies and restrictions.

The image processing and feature extraction were performed using MATLAB 2014a. The morphometry-based radiomics features identified by the image biomarker standardization initiative (IBSI) were extracted. After feature extraction, a supervised learning method was applied to build a model to predict treatment response after radiotherapy. The feature analysis including the model construction was done using Python 3.7.

After preprocessing data, the dataset was divided into two sets as training (70%) and test (30%) datasets. Age, gender, tumor type and grade of the tumor were also included to the data set. The ANOVA test was done to determine the most predictive twenty features from the training data set to forecast the treatment response. Hence, a radiomics signature was developed based on f-test scores of the selected features. The most appropriate machine learning algorithm was used to predict the treatment response which was selected using ten-fold cross validation. Using the latter algorithm, the selected normalized features were used to train the prediction model for the developed radiomics signature. Once the model was constructed its performance was evaluated. Then random search and grid search were used to optimize the hyperparameters and the model performance.

Predicting prognosis in patients with soft tissue sarcomas of extremities following radiotherapy

Thirty patients with histologically proven soft tissue sarcomas of extremities following radiotherapy were retrospectively evaluated. FDG-PET/CT and MRI scans were done for each individual prior to treatment. Patients who received chemotherapy were excluded from this study. This study used deidentified fused images, contours which were available as radiotherapy structure set (*RTstruct*), objects and clinical data from TCIA in compliance with its data usage policies and restrictions.

The morphometric features were calculated, and mean values were computed for each feature. The two-sample t- test (one-tailed) with 95% confidence level was performed for each morphometric feature to determine the significant difference between the patients who developed recurrence or metastasis (RM) and those who remain recurrence or metastasis free (RMF) after radiotherapy.

Results and Discussion

Predicting therapeutic response in patients with glioma following radiotherapy

Among the selected patients, 54% were males, and 46% were females. The mean age of the sample ranged from 51.5 ± 14.5 years. The chosen machine learning algorithm was the random forest, which exhibited the highest classification accuracy at 81.59%. Initially, the prediction model achieved an accuracy of 82%, which improved to 86% after optimizing hyperparameters. The performance metrics for predicting the absence of a tumor included precision (91%), recall (81%), and F1-score (86%). For predicting the presence of a tumor, the precision, recall, and F1-score were 82%, 92%, and 87%, respectively. The constructed model demonstrated its ability to predict therapeutic response with an area under the curve-receiver operating characteristic (AUC-ROC) of 0.92 according to ROC curves. After grid search evaluation, the AUC-ROC value remained high at 0.91.

Predicting prognosis in patients with soft tissue sarcomas of extremities following radiotherapy

Among the selected sample 33% were males while 67% were females. The mean age of the sample was 56.6 ± 18.84.

The area density based on the oriented minimum bounding box was uniform across all patients, and the center of mass shift was also uniform for all patients except one. Excluding those two features, the results of the. two-sample t- test (one-tailed) showed a significant difference (*p*-value of 0.029) between RM and RMF categories only for surface to volume ratio.

Conclusion

This study developed a morphometry-based radiomics signature that could predict the therapeutic response in patients with gliomas after receiving radiotherapy. This radiomics signature served as the foundation for the prediction model, which utilized the random forest algorithm to determine outcomes. The morphometric feature, surface-to-volume ratio, demonstrated a significant difference between the patients who developed recurrence or metastasis and those who remained free of recurrence or metastasis after receiving radiotherapy for soft tissue sarcomas of extremities. Therefore, the surface-to-volume ratio could be employed as a predictor for assessing the prognosis of patients with soft tissue sarcomas of extremities following radiotherap.

Source of Funding

Self-funding.

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DEVELOPMENT OF NOVEL POLY (ETHYLENE OXIDE)/CONDUCTING POLYMER-BASED ELECTROLYTES AND GRAPHITE-BASED COUNTER ELECTRODES FOR DYE/Q-DOT SENSITIZED SOLAR CELLS

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A solar cell or photovoltaic (PV) cell is a device which absorbs sunlight and converts it directly into electricity. A solar panel is made from a large number of interconnected small solar cells. Most solar cells and solar panels commercially manufactured today and widely used worldwide are made of the semiconductor material silicon. A silicon solar cell is formed as a junction between a n-type silicon layer and a p-type silicon layer. When a p-n junction solar cell is exposed to sunlight, an electric current is produced. Today's silicon solar cells convert about 20-25% of the light energy into electrical energy. Also, at present, solar electricity is relatively more expensive than the conventional electricity produced by burning fossil fuels such as coal and diesel. Scientists are focusing their research on ways to find more efficient and low-cost solar cell materials and solar cells to make solar electricity more competitive with electricity generated by fossil fuel burning.

To make solar cells and panels more affordable for widespread use by common people, a few different types of solar cell technologies, other than silicon solar cells, have emerged during the past three decades. Among these, dye-sensitized solar cells and quantum dot-sensitized solar cells have emerged as a competitor to silicon solar cells due to their low fabrication cost and flexible design capabilities. These two types of solar cells offer an efficient and low-cost technology categorized under the third-generation PV technology. The efficiency of these devices primarily depends on the properties of the metal oxide semiconductor, the dye or the quantum dot, the electrolyte, and the counter electrode.
This overall objective of this study was to develop a low-cost, nanostructured materials for applications in dye/q-dot-sensitized solar cells with enhanced device performance. In this work, several dye and quantum dot-based solar cell systems were studied and can be categorized under three main contributions development of electrolyte, counter electrode, and photoanode.

Methodology

Electrolyte modifications for dye/q-dot sensitized solar cells.

Solar cells were fabricated using four different types of gel and solid electrolytes based on poly (ethylene oxide) (PEO) polymer together with PANI conducting polymer, PVdF-HFP polymer, ionic liquids, different cations (LiI, Pr_4NI, and KI)) and plasticizers (EC, PC). The effect of blending host polymer for gel polymer electrolytes was tested by blending PEO and PVdF-HFP polymers. In order to improve the performance of the device, BMII ionic liquid-based nanofiber gel electrolyte was prepared through an electrospinning technique. Prepared electrolytes with optimized compositions have been characterized by ionic conductivity measurements, DC polarization test, Fourier Transform Infrared Spectroscopy (FTIR), Scanning Electron Microscopy (SEM), and Differential Scanning Calorimetry (DSC).

Novel counter electrodes for dye/q-dot sensitized solar cells.

Under this section, different types of counter electrode (CE) materials were prepared and tested in solar cells, starting from Sri Lankan vein graphite. Initially, attempts were made to find an excellent binder to fabricate graphite-based CEs with good adhesion to the FTO substrate and solar cell performance. For this purpose, graphite, reduced graphene oxide (RGO), conducting polymers, and activated carbon-based CEs were prepared in different ways, and to improve the electrical conductivity and electrocatalytic properties, composites of the above materials were used to prepare CEs. Different materials such as SnO2, PANI, Pt, and PbS nanoparticles were incorporated while preparing the graphite and RGO-based CEs to improve the adhesion to the substrate and enhance the electrocatalytic activity. For activated carbon (AC) based CEs, stainless steel was used as the substrate to replace the widely used but expensive FTO glass, while sugar was used as the binder. Efficiencies comparable to Pt-based CEs were achieved after optimizing the procedure of making carbon-based CEs.

Photoanode modifications for efficiency enhancement in dye/q-dot sensitized solar cells to maximize the light-harvesting and improve the performance of solar cells, several techniques were used to modify the titanium dioxide (TiO_2) based photoanodes such as surface treatment, co-sensitization with inorganic semiconductor quantum dots (QDs), and carbon QDs. Therefore, this study tested the possibility of co-sensitization with quantum dots towards achieving higher efficiencies in DSSCs. TiO_2-based photoanodes were modified by $TiCl_4$ surface treatment and co-sensitization. TiO_2 photoanodes were co-sensitized with PbS/CdS QDs formed by the SILAR technique and the Ru N719 dye. In a different solar cell system, photoanodes were co-sensitized with carbon quantum dots and Ru N719 dye.

Results and Discussion

The major findings in this research project are as follows. Gel and solid polymer electrolytes consisting of PEO and PANI conducting polymer showed that the enhanced iodide ion conductivity is very likely due to the "cation trapping" effect caused by the strong cation-PANI and cation-PEO polymer interactions. Structural studies on the blended polymer gel electrolyte showed that the blending of PEO and PVdF-HFP polymers in a suitable ratio (20:80 wt% ratio) enhances the amorphous fraction of the polymer matrix and facilitates fast ion conduction. Performance of the DSSC with PEO/PVdF-HFP based blend gel polymer electrolyte enhanced by the "mixed cation effect" which increases the ionic conductivity of iodide ions of Pr_4NI and KI salts mixed in a proper ratio.

From the results of preliminary studies of carbon-based CEs, including Sri Lankan vein graphite, it is concluded that the spray technique was the best technique to prepare carbon-based CEs while the ionic liquid containing liquid electrolyte gives the best performance of the carbon CE-based devices. Making carbon-based composite materials provided the formation of a nanoporous structure with improved interconnections and pathways for improved electrocatalytic effect for triiodide ion reduction at the CE and efficient ionic mobility for iodide/triiodide ions in the electrolyte medium.

The overall performance of the device has increased significantly after introducing the optimized amounts of PbS and CdS quantum dots in TiO_2 photoanode, largely by increasing the current density. The results of co-sensitized solar cells demonstrate the efficient charge transfer dynamics between the two sensitizers, thus reducing the recombination in the solar cell. Green emissive CQDs were synthesized in the next system using a simple and cost-effective technique with atmospheric pressure air plasma. Two different configurations of DSSC were fabricated in order to explore the potential of synthesized CQDs for solar cell application. The higher number of carboxylic group sites on the CQD surface provides better anchoring towards TiO_2 and enables more active sites. Also, it facilitates faster electron transport by creating sub-energy levels due to bandgap modifications. This has improved the electron transfer and reduced the recombination significantly.

Conclusion

This research provides three significant contributions to the field of dye/quantum dot-sensitized solar cells. The first significant contribution presented here is the synthesis and characterization of gel and solid-state electrolytes for solar cell applications as a replacement for the widely used solvent-based electrolytes. It could provide several polymer-based gel and solid electrolytes with comparable efficiency and high stability to liquid-type electrolytes. The second significant contribution presented here involves the application of low-cost carbon-based materials such as Sri Lankan vein graphite, reduced graphene oxide, activated carbon, and their composites as a low-cost alternative to expensive Pt used as the counter electrode in solar cells. This project has convincingly shown that Sri Lankan natural vein graphite and its derivatives can be suitably modified and adapted to be used as the counter electrode material in dye/Q-dot solar cells. The third contribution in this thesis involves the modifications of TiO_2-based photoanode to improve light harvesting in solar cells. For this purpose, co-sensitization was mainly used with quantum dots and dye to fabricate dye/q-dot co-sensitized, hybrid solar cells and improved device performance. With these modifications, it was possible to achieve dye/q-dot co-sensitized hybrid solar cells with a conversion efficiency of over 10%. It can be further developed through a research and development phase to fabricate practical and prototype solar cells.

Source of Funding

National Science Foundation (Sri Lanka-Pakistan collaborative research) grant no. NSF-PSF/ICRP/2017/EA&ICT/04 is acknowledged for funding this research

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SOFTWARE COMPLEXITY MEASURES: A NOVEL APPROACH BASED ON HUMAN COGNITION FOR ADEQUATE QUANTIFICATION OF COMPLEXITY IN SOFTWARE PROGRAMS

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In his second law of software evolution, Manny Lehman, the father of software evolution, stated, "As a program is evolved, its complexity increases unless work is done to maintain or reduce it" (Lehman, 1996). However, a considerable number of software applications are currently being developed or upgraded without sufficient consideration of maintainability and understandability aspects. Over time, the codes of such software can become vastly intricate, making it almost impossible to enhance them further without undergoing a significant revision, requiring an enormous amount of money. Although the elimination of software complexity is inconceivable, it can certainly be controlled and reduced to an acceptable level. Accurate quantification of complexity is an essential prerequisite for restraining it.

Over the years, there have been many attempts to introduce new software complexity measures. Although existing complexity measures have their merits, they also have numerous limitations in accuracy, suitability, and ease of use. Furthermore, most of the measures proposed in the recent past have revolved around the same set of factors without introducing novel factors. The cognitive approach is regarded as the latest trend in measuring software complexity. However, the studies which used the cognitive approach have either employed the subjective weights proposed by the researchers or the weights of originators, which have no published empirical basis.

Thus, this research undertakes a comprehensive analysis of existing complexity measures and introduces nine novel factors that the existing studies have not considered. Moreover, it introduces a standard set of new cognitive weights for fundamental programming elements. The research also proposes a new complexity measure, the Weighted Program Complexity (WPC) measure, which computes complexity based on the novel factors and cognitive weights proposed through this study. Unlike the existing measures, the WPC measure is the first measure to employ a collection of sixteen factors to decide on program complexity. WPC was theoretically validated via Weyuker's framework and empirically validated via a study involving 167 programmers who ranked the complexity of open-source programs. WPC was the most practically applicable measure among those that compute complexity at the lowest possible level, having the highest correlation (0.964) with expert rankings.

The primary objective of this research is to introduce a new measure that computes complexity based on the novel factors and cognitive weights proposed in this study.

The research also aims to achieve the following specific objectives:

(I) To develop an in-depth understanding of software complexity.
(II) To identify and analyse the existing complexity measures.
(III) To introduce a set of novel complexity factors which have not been considered by the previously defined measures.
(IV) To propose a standard set of novel cognitive weights for fundamental programming elements.
(V) To examine the appropriateness of incorporating the novel factors into the proposed measure.
(VI) To assess the theoretical and practical validity of the newly introduced complexity measure.

Methodology

A multi-stage approach was devised using a mixture of empirical research and theoretical analysis to achieve the research objectives.

Initially, an extensive study was conducted to gain an in-depth understanding of software complexity and identify its relevance in the field of software engineering. From the study, it was noticed that the current literature on this subject does not offer a commonly accepted definition of software complexity. Thus, based on the knowledge obtained from the literature survey, a new definition of complexity was proposed for the purpose of the study.

Subsequently, the authors focused on identifying and analysing the existing complexity measures. Based on the obtained information, in addition to proposing nine novel factors that the existing studies have not considered, the study introduced a standard set of new cognitive weights for fundamental programming elements based on an empirical study that involved one hundred and twenty programming experts who had a working experience of two or more years.

The study then introduced the Weighted Program Complexity (WPC) measure, a new complexity measure that computes complexity based on the novel factors and cognitive weights introduced in this research.

Since the novel factors were proposed solely based on the opinion of the authors, a study was conducted to examine the appropriateness of incorporating those factors into the WPC measure by considering the views of twenty-two established industry personnel with five or more years of experience. The industry personnel were chosen randomly, ensuring that the sample consisted of personnel attached to renowned software companies in Sri Lanka that have been established for more than twelve years.

The theoretical validity of the WPC measure was assessed using Weyuker's framework (Weyuker, 1988), the most extensively used complexity validation framework in the software engineering field (Srinivasan and Devi, 2014).

Finally, an empirical study was conducted to identify the practical applicability of the WPC measure with the other measures that computed complexity at the lowest possible level. Ten open-source Java programs and a hundred and sixty-seven software engineers who had a working experience of two or more years were used for the study. Although the ten programs were different from one another, it was ensured that they were conceptually the same by limiting them to the same programming language.

Results and Discussion

The following are some of the notable findings derived from the literature survey:

- The first complexity measure to be proposed is McCabe's Cyclomatic Complexity (MCC).
- MCC and Halstead's Software Science measures are the two most widely known measures of all time.
- Based on the programming style, the existing measures can be divided into two categories: procedure-oriented (PO) and object-oriented (OO).
- Based on the characteristic that has been considered, PO measures can divided into three groups: structural, size, and hybrid.
- PO measures have revolved mainly around five factors: comments, coupling, decision statements, size, and variables.
- The first cognitive-based OO measure to be introduced is Kushwaha and Misra's Total

Complexity of OO Software Product (KMTCOOSP) measure.

- The validity of complexity measures has been mainly assessed theoretically, can be further categorized into expert opinions and case studies.
- Based on the program component in which the complexity is calculated, the metrics of OO measures can be categorized into four groups: application level, class level, method level, and program statement level.
- Based on the proposed artefact, the OO complexity measures could be categorized into two main groups: design-level and code-level. However, it was discovered that code-level measures are more useful in predicting the maintenance effort (Li and Henry, 1993) and fault-prone modules (Jiang *et al*, 2008) than the design-level measures. Furthermore, it was found that from all the artefacts that are released in each phase, source code is the easiest to measure (Oram and Wilson, 2010). Due to these reasons, most of the OO measures have been suggested based on the program code. In fact, it was observed that 73% of the OO measures introduced within the last fifteen years had been proposed based on the program code. Moreover, it was noticed that most of the code-level measures had been proposed based on the cognitive weight concept. Hence, it is apparent that proposing measures for the program code based on the cognitive weight concept is the way forward in measuring complexity.
- The majority of the code-level measures have only revolved around coupling, inheritance, type of basic control structures, nesting level of basic control structures, and size factors. Hence, it is apparent that the existing OO code-level measures are unable to capture the complexity of the OO-specific factors to a satisfactory level.

From the empirical study that was performed to determine a standard set of new cognitive weights, it was evident that 95% of the programming experts examine the complexity of their programs at least once a month. This confirms the importance of measuring program complexity. It also shows that measuring complexity is becoming a statutory activity in the software development life cycle. Furthermore, the study revealed a lack of awareness among programming experts regarding the code complexity measures that have been proposed in the recent past. Moreover, it was found that Sonar Cube is the most used code complexity measuring tool in the industry.

From the study conducted to assess the practical applicability of the WPC measure, it was found that the WPC is the measure to report the highest correlation (0.964) with expert rankings among the ones that compute complexity at the lowest possible level.

Conclusion

In conclusion, this study introduces nine new factors that have not been considered by any of the existing complexity measures. The novel factors were suggested to direct research on software complexity measures to a new pathway such that those factors will be considered when introducing new complexity measures. In addition, the study introduces a standard set of new cognitive weights for fundamental programming elements.

Central to the work of this research is the introduction of the Weighted Program Complexity (WPC) measure. By leveraging the new factors and cognitive weights established in this study, the WPC measure offers an advanced approach to evaluating software complexity. As software systems evolve and grow intricately, the WPC measure provides a valuable tool for guiding design, development, and maintenance decisions.

In essence, this research marks a significant step forward in the domain of software complexity measurement. The introduced factors, cognitive weights, and the WPC measure collectively contribute to an enhanced understanding of complexity's multifaceted nature. As the software landscape continually evolves, this study's findings hold the potential to shape the future of complexity assessment methodologies.

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IMPLICATIONS ON SELECTED ENVIRONMENTAL AND GEOGENIC FACTORS ON CHRONIC KIDNEY DISEASES OF UNCERTAIN ETIOLOGY (CKDu)

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Chronic kidney disease of undermined etiology (CKDu) is a severe health problem that prevails in certain districts of the dry zone of Sri Lanka (Athuraliya *et al*., 2011; Chandrajith *et al*., 2011). It was first recognized in the mid-1990s in the North Central region of the country, and since then, the disease has been reported elsewhere in dry regions with alarming death rates (Jayatilake *et al*., 2013). Most of the affected individuals in Sri Lanka were found in three provinces, namely North Central, Uva and Northwestern. Regional clustering of the disease is one of the important features of the CKDu.

During the last few years, several causative factors have been proposed for the etiology of CKDu. These include the use of low-quality aluminum utensils in food preparation (Ileperuma *et al*., 2009); nephrotoxic heavy metals such as Cd, As, Pb (Bandara *et al*., 2008; Jayasumana *et al*., 2015; Jayatilake *et al*., 2013); toxins produced by cyanobacteria in water (Jayasekara *et al*., 2015); exposure to agrochemicals (Jayatilake *et al*., 2013); and viral infections (Gamage *et al*., 2017; Yoshimatsu *et al*., 2019). Continuous exposure to direct sunlight and consequent heat stress with recurrent dehydration (Nerbass *et al*., 2017; Senevirathna *et al*., 2012) has also been considered as an etiological factor for CKDu. The aim of this study is to investigate the relationship between agricultural practices and the occurrence of CKDu in the dry zone of Sri Lanka. The CKDu hotspots of Girandurukotte and Wilgamuwa were selected for this study.

The objectives of the present study were,

(I) To study the geographical distribution of CKDu in Wilgamuwa and Girandurukotte.

(II) To investigate the geo-environmental conditions that might influence CKDu in selected regions, correlating CKDu prevalent data, spatial distribution, and geochemical parameters of environmental materials (such as soil and water). fertilizers) in soil-water-plant s

(III) To explore the fate of environmental contaminants (agrochemicals, fertilizers) in soil-water-plant systems in CKDu regions.

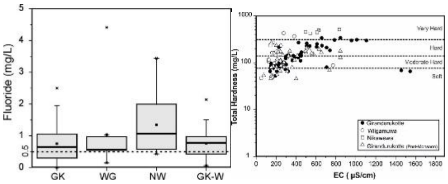
Methodology

Environmental samples were collected in CKDu regions (Girandurukotte-GK, Wilgamuwa-WL, Nikawewa-NK) in the dry zone and control regions in Sri Lanka. Ginnoruwa village, considered one of the important CKDu hotspots in Girandurukotte, was also studied in detail. Water (surface, groundwater) (n=531), soil (paddy, garden) (n=218), rice and rice plants (n=15), fertilizers (n=36), and pesticides (n=20) samples were collected to investigate their trace and major element compositions. Control samples of rice paddy soils and rice samples were also collected from the wet zone regions where CKDu is non-prevalent. All water samples were preserved at 4 °C until analysis. Chloride (Cl^-), fluoride (F^-), nitrate (NO_3^-), phosphate (PO_4^{3-})and sulfate (SO_4^{2-})were determined by ion chromatography (Thermo DionexICS-1100). Repeated analyses of samples yielded analytical precisions better than ± 3%. Total hardness was determined by the EDTA titration method (APHA 2012). Major and trace element contents in the samples were quantified using ICP-MS. The stable isotope abundance in water (18O and 2H) was determined using cavity ring-down spectroscopy methods (Picarro L1102-i).

Results and discussion

Surface and groundwater

The results of this study showed that the geochemistry of groundwater in all regions is controlled primarily by the chemical weathering of underlain rocks, though the impact of precipitations is also significant. In all groundwater samples studied, HCO_3^- and Cl^- are the most dominant anions and the anionic contents varied in the order $HCO_3^- > Cl^- > SO_4^{2-} > F^- > NO_3^- > PO_4^{3-}$. Ca^{2+} and Na^+ are the most predominant cations that varied in the order: $Ca^{2+} > Na^+ > Mg^{2+} > K^+$. The classification of groundwater based on total hardness (TH) showed that most groundwater samples fall in the hard water category. The hardness values ranged from 89.5 to 815 mg L^{-1} with an average value of 212 mg L^{-1}. The lowest hardness of 10 mg L^{-1} was observed in the Girandurukotte region near the stream sample, while the highest value of 516 mg L^{-1} was recorded from the Nikawewa region (Figure 1b). The fluoride content in well water in these regions does not exceed the WHO recommended limit of 1.5 mg L^{-1}, but in most samples, the fluoride contents were above 0.5 mg L^{-1}, the limit recommended for tropical countries by WHO (WHO 1994) (Figure 1a). Groundwater from the CKDu-affected regions showed extremely low levels of arsenic, which are below the WHO recommended limit. The highest arsenic concentration (0.73 μgL^{-1}) was recorded in the GK region. The geochemistry of groundwater in all regions is controlled primarily by the chemical weathering of underlain rocks, though the impact of precipitations is also significant.



(a) (b)

Figure 1. (a) Box and whisker plot showing fluoride distribution in study regions (b) Relationship between hardness and electrical conductivity of groundwater from study regions.

Soil

Rice paddy soils in the wet zone were acidic in nature, with depleted Ca and Mg contents. Most trace elements in rice soils were geochemically incorporated with Fe and Mn oxides/hydroxides, particularly in the dry zone area. Although a slightly higher content of As in CKDu regions compared to the wet zone was noted, the levels in all climatic regions are found to be well below the average levels in soils found elsewhere in the world. The median Cd content in dry zone rice paddy soils was 0.07 mg kg^{-1}, while it was 0.09 mg kg^{-1} in the wet zone. In CKDu regions, the median Cd content was 0.07 mg kg^{-1}. Cadmium contents were not significantly higher in rice soils from both regions, including CKDu hotspots. As indicated in this study, the total metal contents were generally below the threshold values and do not provide direct evidence of possible health risks.

Rice plants

A high content of Mn is observed in all plant parts. Overall, these metal contents in roots were much higher than those in leaves and grains, following the order root>leaves>grains. above 0.5 mg L^{-1}, the limit recommended for tropical countries by WHO (WHO 1994) (Figure 1a). Groundwater from the concentrations of As and Pb showed little difference between leaves and grains. It may suggest that these metals remobilizing abilities from leaves to grains are much higher than those of Cr, Cu, Fe, Mn, Ni, and Zn. The higher accumulation of As in roots (2.60 mg kg^{-1}) of the rice plant was found to be followed by the accumulation in the shoot (0.13 mg kg^{-1}), leaves (0.08 mg kg^{-1}), husk (0.08 mg kg^{-1}), and the lowest was observed in grain (0.03 mg kg-1). However, none of the rice samples exceeded the total As level (300 µg kg^{-1}) recommended for inorganic As in rice (200 µg kg^{-1}) by the Codex Committee on Food Additives and Contaminants (FAO/WHO 2012). The highest content of Cd was found in the root (1.02 mg kg^{-1}) of the rice plant, followed by shoot (0.09 mg kg^{-1}), leaves (0.08 mg kg^{-1}), grains (0.05 mg kg^{-1}), and husk (0.03 mg kg^{-1}). The concentration of Cd in rice grains (0.05 mg kg^{-1}) is comparatively lower compared to other parts. These values were below by the Codex Committee on Food Additives and Contaminants (FAO/WHO 2012).

Fertilizers and pesticides

Samples of triple super phosphate (TSP), Urea, Muriate of Potash (MOP) and Top Dressing Mixture (TDM) were collected from households of farmers. Among the samples, TSP samples contained higher concentrations of trace elements. Trace element content in TSP fertilizer samples of Girandurukotte varies in the order of Fe> Mn> Al> Sr> Zn > Ba> Ni> Cr> Cu> U> As> V> Bi> Pb> Se> Tl> Co> Ga> Cd > Cs> Hg. In general, trace metals such as Zn, Ni, Cu, Fe, Ti, Sr and Mn are available in high concentrations. All fertilizers have a higher Fe content, an element that is found in greater amounts with respect to other heavy metals and trace elements. The mean concentration of As was present only in the five pesticide samples with comparatively low concentrations, and Cd was detected in low concentrations from the three samples.

Case study in Ginnoruwa

Fluoride levels in groundwater from CKDu wells in Ginnoruwa varied from 0.23 to 5.00 mg L^{-1} with a mean value of 1.02 mg L^{-1} (Figure 2). In our sample population, 60% of the wells showed fluoride levels higher than 0.6 mg L^{-1}. However, groundwater showed higher concentrations of both fluoride and hardness, and Mg^{2+} was found to be the most prominent factor for water hardness in CKDu-affected wells. This, in turn, may increase the risk of CKDu. In non-CKDu wells, groundwater is often influenced by nearby surface water bodies, thus leading to the dilution of critical element levels. CKDu wells showed a mean As content of 0.17 µg L^{-1}, which was similar to non-CKDu wells (0.20 µg L^{-1}). With this, both types of wells showed almost 100 times lower As levels compared to the recommended value of 10 µg L^{-1} (WHO 2011). Similar to Cd and As, all other toxic trace elements measured, including Cr and Pb, were also lower than recommended levels by WHO. Results of this study suggest that drinking water fluoride in combination with Mg^{2+} may be a far underestimated for the deterioration of kidney functions. Overall, the antagonistic health effects of fluoride in combination with Mg^{2+} and the molecular mechanism associated with fluoride-Mg toxicity need further investigation.

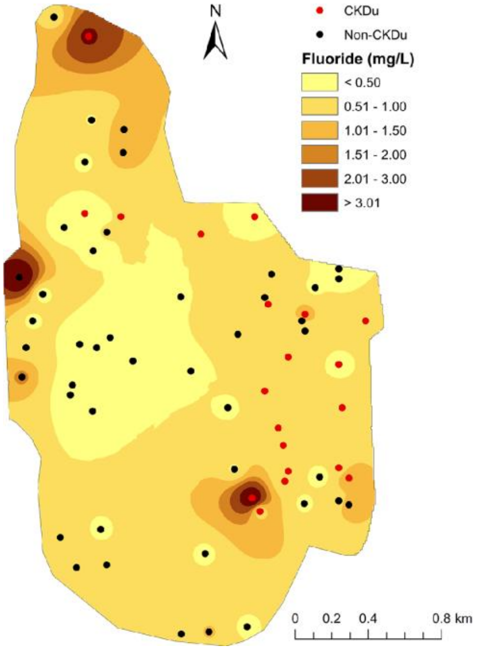


Figure 2. Spatial distribution of groundwater fluoride in the Ginnoruwa region

Conclusion

This study showed that the geochemistry of groundwater in all regions is controlled primarily by the chemical weathering of underlain rocks, though the impact of precipitation is also significant. Since groundwater trace element levels are well below the stipulated WHO standards in both seasons, their contribution towards CKDu can be eliminated. Groundwater is the prime source of drinking water in the Ginnoruwa region, where CKDu is widespread. Among the sampled wells, both CKDu and non-CKDu groundwaters showed low concentrations of known nephrotoxic trace elements such as Cd, As, and Pb. There is no reliable evidence of any unique cause of the disease. Several studies have shown that the disease's occurrence is mainly due to the combined effects of several adverse environmental conditions. Concerning soil and water, they are in safer stages concerning their trace element content. However, the continuous application of fertilizers and agrochemicals without proper management will adversely affect the environment. Also, the results confirmed that the availability of toxic elements (As and Cd) and their impact on CKDu is negligible. In addition to fluoride and hardness (Ca and Mg) in groundwater, no other element contributes to the etiology of CKDu in the studied regions.

Source of Funding
This research work was financially supported by the Nation Research Council (NRC), Sri Lanka (Grant 14-05).

Source of Funding
This research work was financially supported by the Nation Research Council (NRC), Sri Lanka (Grant 14-05).

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MICROBIAL BIOFILMS AND THEIR NETWORK INTERACTIONS

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Biofilms are complex, functional microbial communities comprising multiple species attached to surfaces or physical interfaces in nature. They can also be created in vitro using beneficial microbes for various applications such as biofertilizers, pharmaceuticals, fuels, foods, and electricity. Within biofilms, resident microbes are interconnected through gene regulatory, protein, and signaling networks that govern their functionality and sustainability. Natural biochemicals, including enzymes, RNA, proteins, polysaccharides, and quorum sensing (QS) molecules, play a crucial role in these networks (Seneviratne, 2015). These biochemicals are found within the extracellular polymeric substances (EPS), which are secreted as biofilm exudates (BFEx) by the biofilm itself. The EPS is often referred to as the 'dark matter of biofilms' due to its undefined nature, and the contributions of its components to matrix integrity are not fully understood at the molecular level (Flemming & Wingender, 2010; Seviour et al., 2018).

The identification of this 'dark matter' and a deeper understanding of its role in complex interactive ecosystem networks are crucial for leveraging biofilms in various biotechnological applications, such as biofilm biofertilizers (BFBFs) and biofilm medicines (BFMs). Therefore, it is hypothesized that biofilm formulations can contribute to the restoration of degraded agroecosystems and the human body ecosystem, respectively.

Overall objective:
To understand the holistic ecosystem approach of using microbial biofilms developed in-vitro for remediating two contrasting ecosystems i.e. paddy agroecosystem and human gut environment.

Specific objectives:
(I) To manipulate biofilm microbes to maximize the productivity of EPS biochemicals i.e., lipids, proteins, and polysaccharides.
(II) To find out the most compatible and productive biofilm type in terms of biochemical production
(III) To identify important biochemicals produced by the productive biofilm.

(IV) To apply a productive biofilm as a biofertilizer to paddy agroecosystem as a case study, and to evaluate reinstation of ecosystem networks by constructing networks using data obtained from its soil-plant-microbial parameters.

(V) To investigate the potential of biofilm exudates in reinstating human body ecosystem via growth and dormancy breaking of gut microbes.

Methodology

A cross-disciplinary study of agriculture, ecology, and medicine was conducted to investigate the effect of the composition of microbes on network interactions and the production of biochemicals exuded during growth and maturation of fungal-bacterial, fungal-cyanobacterial, bacterial-cyanobacterial, and fungal-bacterial-cyanobacterial biofilms developed in-vitro. To examine the potential restoration and its effect on consequent crop yield, and gut microbiota, productive biofilms were applied to a degraded paddy agroecosystem and to a simulated human gut environment, respectively. Data were analyzed to identify the outcomes of interactions in diverse biofilms and their effects on the two ecosystems.

Results and Discussion

The biofilms exhibited distinct morphological characteristics during their development. Fungal-cyanobacterial-bacterial biofilms and fungal-bacterial biofilms displayed an enormous potential in generating EPS in highest quantity and biochemical diversity, respectively. Also, the capacity of producing diverse biochemicals of the biofilms could be improved further by manipulating microbial composition and inoculation ratio. Moreover, 214 EPS biochemicals were detected and 9% of them were identified of the fungal-bacterial biofilm, which were ecologically and medicinally important, and indicative of restoration of degraded managed/natural ecosystems including the human gut. The biofilms when applied to paddy agroecosystem as BFBF were found to enhance crop yields by 24% via restoring soil-plant-microbe network interactions. The results also showed that with the BFBF application, the crop yields could be improved further by optimizing several soil physico-chemical parameters i.e., soil total phosphorus, nitrogen, potassium, and organic carbon. However, with the farmers' chemical fertilizer alone application, the crop yields mainly depended on the amount of chemical N fertilizers. In the context of human gut environment, the EPS biochemicals were not cytotoxic, and they increased the growth and possibly dormancy breaking of gut microbes leading to restore gut microbiota.

Conclusion

The EPS biochemicals trigger biochemical cycles in living organisms and in the ecosystems. The present study concluded that the biofilms with EPS biochemicals can be formulated as next generation biofertilizers and medicines/nutraceuticals within the limitations of the study.

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Source of Funding

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SEED BIOLOGICAL TRAITS OF SELECTED RAINFOREST SPECIES FROM SRI LANKA: TOWARDS CONSERVATION OF BIODIVERSITY

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Tropical rainforests which are important as gene banks for future research and development are faced with wide scale degradation leading to loss in biodiversity and ecosystem services, leading to negative social and economic impacts. Restoration of the degraded forests is vital to ensure their existence and safeguard ecosystem services provided by them for the future. Ex-situ conservation and ecological restoration have been identified as important elements in biodiversity conservation at both regional and global scales (Li and Pritchard, 2009; Wyse *et al.,* 2018).

The successful implementation of cost-effective conservation strategies requires knowledge on seed biology since seeds play a critical role in angiosperm regeneration (Kildisheva *et al*., 2020). Techniques of seed collection, determination of germination requirements of seeds, storage behaviour, dormancy and dormancy alleviation are some of the significant seed biological information required to plan successful conservation projects (Miller *et al*., 2017). However, lack of knowledge on seed biology is a significant impediment to the effective conservation of these tropical rainforest species.

This research was conducted to gather information on seed dormancy, seed germination, seed storage behavior and other seed biological traits of 56 plant species in tropical lowland rainforests of Sri Lanka.

Further, information on plant functional groups considering the reproductive functional traits was applied in the identification of variation in seed traits of genus *Mesua* within different climatic zones. Further, the above information was applied to determine the early seedling development of invasive *vs.* native plant species from tropical lowland rainforests in Sri Lanka.

Methodology

Seeds of 56 plant species were collected from the tropical lowland rainforests in Sri Lanka, based on their ecological significance, threatened status, availability and endemicity. Seeds were incubated on moistened tissue papers in Petri dishes at 25°C in light/dark (12hr/12hr) conditions to identify the presence or absence of dormancy. Imbibition experiments, embryo length: seed length ratio calculation, germination experiments after scarification and gibberelic acid treatments were conducted to identify dormancy class.

Hundred seed method was conducted to identify the seed storage behaviour of collected species. Initial moisture content was also calculated using the oven-dry method.

Effect of storage at 25, 8 and -20 °C on seed viability and vigour of some selected species was determined. Change in the anti-oxidant and antifungal activity with storage of some of the study species was documented.

Results and Discussion

Determination of seed dormancy

A dormancy profile for Sri Lankan tropical lowland rainforests was created using seed dormancy and germination behavior data. According to the study, 63.7% of the species produced non-dormant seeds while 23.6, 9, 7.2 and 1.8% were producing morpho physiologically, physiologically, morphologically, and physically dormant seeds, respectively (Figure 1). Among the studied species, 32 species were identified as non-dormant (57% of the total species), while 24 species (43%) were dormant. Dormant species included 1 physically dormant species, 5 morphologically dormant species, 8 physiologically dormant species and 10 morpho physiologically dormant species. Embryos of 13 species were underdeveloped, while 43 species had developed embryos (Figure 2). The seeds with underdeveloped embryos have morphological or morphophysiological dormancy.

Alleviating dormancy is critical in restoration programs to accelerate seedling emergence during seed sowing (Zanetti *et al*., 2020). Here we have identified mechanical scarification as the most effective method in overcoming physical dormancy while the application of Gibberellic acid (GA_3) as the most successful treatment to overcome physiological dormancy in many tropical rainforest plant species.

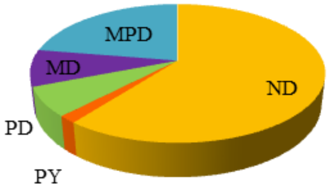

Figure 1. Seed dormancy profile of tropical rainforest species in Sri Lanka. MD, morphological dormancy, MPD, morphophysiological dormancy; ND, nondormancy; PD, physiological dormancy; PY, physical dormancy.

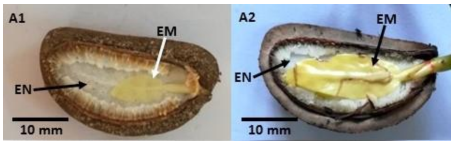

Figure 2. Underdeveloped embryos of fresh (A1) and developed embryo of the germinating (A2) seeds of *Artabotrys hexapetalus* shows Morphologial dormancy.

Determination of seed storage behaviour

Seed storage behaviour profile was constructed for tropical lowland rainforests in Sri Lanka. Fifty-three percent of the species studied produced desiccation-sensitive seeds, while the other 47% produced desiccation-tolerant seeds. Although desiccation-sensitive species slightly dominated the tropical lowland rainforest ecosystem in Sri Lanka, our results revealed that both desiccation-sensitive and desiccation-tolerant behaviours have a significant role in it. In this research, both predictive models and experimentation procedures were used separately to determine the seed storage behavior types. A higher percentage of tree species recorded desiccation-sensitive seed storage behaviour, while most of the shrubs, herbs and lianas recorded desiccation-tolerant behavior.

A. significant difference between the proportions of desiccation sensitive/desiccation tolerant among different strata was observed (χ^2=215.6, P<0.001). Sixty-three, 91 and 60 % of the species in canopy, subcanopy layer and understory trees were dominated by desiccation-sensitive species, respectively. Understory shrubs (68%), herbs

(83%) and lianas (83%) mostly consisted of desiccation-tolerant species

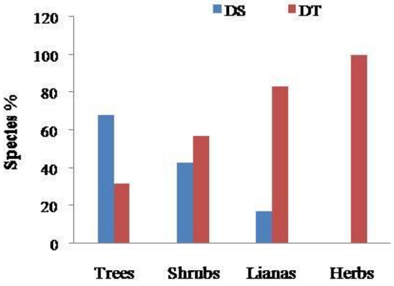

Figure 3. Percentage of species producing DS (Desiccation Sensitive) and DT (Desiccation Tolerant) seeds representing different life forms in tropical lowland rainforests in Sri Lanka.

Desiccation-sensitive behaviour of canopy seeds enables them to germinate fast and produce high vigour seedlings while desiccation-tolerant behaviour of understory species helps them to survive in the soil seed bank until they receive favorable conditions for germination.

Storage treatments

Storage treatments were conducted for ten species. Of the five species which were stored for a period of 12 months, two (40%) showed decreased germination after storage. This result suggests that these species do not tolerate storage for long periods (Godefroid *et al*., 2020), however, they survived during short-term storage (6 months). Our results further described that most of the desiccation-tolerant species retained viability in a higher percentage at 8 °C compared to those stored at 25 and -20°C.

In this study, seeds of desiccation-sensitive species could be stored only for a very short period (~ 2 months). However, the highest viability of recalcitrant seeds was obtained by storing them at 25 °C. Hence, in the absence of reliable information on other methods for storage, seeds of recalcitrant species should be used as soon as possible for planting, after the seed collection to avoid viability loss (Miller *et al*., 2017).

Conclusion

The dormancy profile constructed for the tropical rainforests in Sri Lanka provides important information on the germination ecology of trees, shrubs, herbs and lianas which is important in describing the ecosystem dynamics. Understanding and considering seed dormancy and germination traits in restoration planning can assist in achieving sustainability of restoration. Further, seed storage and germination information could be used to determine the most suitable method for the propagation of the concerned species and to design successful seed-sowing strategies.

Identifying the seed storage behaviour category is an essential step to deciding appropriate conditions to store seeds for conservation projects. Our data suggest that ex-situ conservation is possible for many species with desiccation-tolerant storage behaviour in tropical lowland rainforests.
Further, this study provides information on storage treatments, which can be applied to selected rainforest species for short-term/long-term storage.

This study contributes significantly to our understanding of the seed biology of Sri Lankan tropical lowland rainforest species and supports future conservation efforts of germplasm by providing options for conventional seed storage and future restoration programmes.

Source of Funding

National Research Council Grant No. 17-095 is acknowledged for funding this research.

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PYRETHROID RESISTANCE, EFFECT OF INSECTICIDE FOGGING AND DISTRIBUTION OF *Wolbachia* WITH REFERENCE TO DENGUE VECTOR CONTROL IN SRI LANKA

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Dengue is a major public health concern in Sri Lanka. The disease control heavily relies on controlling the vector mosquitoes *Aedes aegypti* and *Ae. albopictus*. Continuous application and excess use of insecticides have contributed to the development of insecticide resistance among mosquitoes while making an adverse effect on the non-target organisms. This highlights the need of alternative vector control methods. This study was conducted to investigate the insecticide resistance status of dengue mosquitoes, effect of insecticide fogging on non-target insects and the prevalence of *Wolbachia* (a potential candidate for biological control) in mosquito populations in Sri Lanka.

The objectives of the study were,

(I) To identify the underlying mechanisms of pyrethroid resistance and presence of *kdr* mutations in the voltage gated sodium channel in *Ae. aegypti* and *Ae. albopictus* mosquitoes on different geographical regions of Sri Lanka.

(II) To assess the effect of insecticide fogging on pollinators and other non-target insects.

(III) To identify the presence and the distribution of *Wolbachia* in wild mosquito populations of Sri Lanka and to construct their phylogeny using *wsp* genes.

Methodology

Investigation of underlying mechanisms of pyrethroid resistance in dengue vector mosquitoes in Sri Lanka.

Mosquito adults, larvae and eggs were collected from Badulla (Uva province), Jaffna (Northern), Batticaloa (Eastern), Colombo (Western), Kandy (Central) and Kurunegala (Northwestern) using standard methods. Insecticide-impregnated papers were prepared according to WHO guidelines using recommended discriminating dosages of insecticides. Bioassays were conducted by exposing *Ae. aegypti* and *Ae. albopictus* mosquitoes to insecticide impregnated papers in WHO test kits (WHO, 2018). Mortalities were recorded after 24 h of recovery period. Survivals of bioassays were used to detect *kdr* mutations. Biochemical experiments were carried out according to the guidelines provided by WHO (1998). Fragments of domain II, III, and IV of the voltage gated sodium channel (VGSC) were amplified by polymerase chain reaction (PCR) (Kasai *et al*., 2011). Sequences of VGSC were aligned using BioEdit software with house fly (*Musca domestica*) VGSC genes to identify the *kdr* mutations.

Investigation of the effect of insecticide fogging on non-target insects.

This study was carried out in the Batticaloa district, encompassing three urban sites and three village sites, including areas with paddy fields, cashew, coconut, mango, and other garden crops. Six fogging treatments were administered in the mornings. Each fogging treatment involved the spraying of a 2.5 L insecticide solution over an area of 200 m² for 8 minutes using a handheld pulse jet thermal fog generator.

At each site, ten white polythene sheets, each covering an area of 1 m², were randomly laid out on the ground to collect insects. Thirty minutes after the fogging, the knocked-down insects on the polythene sheets were collected and carefully transported to the laboratory. Mortality and recovery rates were recorded following a 24-hour recovery period. The identified recovered and deceased insects were classified at the family level using the taxonomic keys provided by Bland and Jaques (1984).

Identification of the distribution and phylogeny of Wolbachia in wild mosquito populations in Sri Lanka.

Adults, larvae, and eggs of different mosquito species were collected from previously mentioned study sites. The larvae and eggs were reared to adults under laboratory conditions. Adult mosquitoes were identified using the standard taxonomic keys (Amarasinghe, 1995). Extracted DNA from the mosquitoes were tested by amplifying *Wolbachia* surface protein gene using *wsp* primers. Samples positive for the *wsp* were subjected to *Wolbachia* strain and group-typing with group-specific and strain specific primers. *Wsp* sequences obtained from this study, reference *wsp* sequences obtained from EMBL and GenBank were aligned in BioEdit using CustalW alignment algorithm. Evolutionary analyses were conducted in MEGA7 software package.

Results and Discussion

Investigation of underlying mechanisms of pyrethroid resistance in dengue vector mosquitoes in Sri Lanka.

Colombo district *Ae. aegypti* population showed the highest resistance to pyrethroids (51–52% mortality) followed by Kandy district population (88–89% mortality). All the tested populations of both species were highly resistant to DDT.

According to the biochemical assays, the GST activity levels of both the species from Kandy and *Ae. albopictus* from Kurunegala, were more than the discriminating values.

Amplified sodium channel gene sequences revealed kdr type mutation V1016I Kandy (30%) and S989P from Colombo (100%) in *Ae. aegypti* mosquitoes. Domain III mutation F1534C was found from Colombo (25%) *Ae. aegypti* mosquitoes. Previous studies have observed that over-expression of cytochrome P^{450}gene and enhanced GST activities influence development of pyrethroid resistance in *Aedes* mosquitoes (Karunaratne *et al*., 2013). Although, Colombo population did not show any elevation of enzymatic action, *kdr* type mutations S989P and F1534C were found. Kandy *Ae. aegypti* populations also showed resistance to several pyrethroids with high GST levels and V1016I *kdr* type mutation. Therefore, possible association of these three *kdr* type mutations in developing pyrethroid resistance in *Ae. aegypti* populations is evident.

Investigation of the effect of insecticide fogging on non-target insects.

A total of 1,708 knocked-down insects belonging to ten orders and 71 families were collected during the six fogging treatments. Among the knocked-down insects, 7.1% recovered during the 24-hour recovery period. Of these, the family Formicidae (Hymenoptera) accounted for 28.93%, and the family Sminthuridae (Collembola) accounted for 14.05% in urban settings. In village settings, the majority of the recovered insects (12.34%) belonged to Formicidae (Hymenoptera). The highest number of dead insects in the urban setting belonged to the family Sminthuridae (Collembola) at 28.79% and Formicidae (Hymenoptera) at 16.03%. In the village setting, the highest mortality was shown by the family Aleyrodidae (Hemiptera) at 30.78%.

Despite fogging operations targeting mosquitoes, including Ae. aegypti and Ae. albopictus, only 6 mosquito deaths (0.35%) were observed in both urban and village settings. A previous study conducted by our team emphasized the significance of directly collecting knocked-down insects using polythene sheets (Abeyasuriya et al., 2017).

Pollinator service is a vital ecosystem service provided by insects. In addition to their role as pollinators, insects such as Collembola play a crucial part in the plant litter decomposition process, contributing to the formation of soil microstructure. Ants, on the other hand, are involved in creating, modifying, and maintaining various habitats and microhabitats within the ecosystem. The reduction or removal of these insect populations within an ecosystem can disrupt the services they provide, ultimately impacting the entire ecosystem.

The Batticaloa district of Sri Lanka is renowned for its cashew cultivation, with approximately 652.2 hectares of land dedicated to cashew production. While it was previously believed that cashew trees were wind-pollinated, recent studies have demonstrated the heavy reliance of cashew pollination on biotic pollinators. Among these, wild bees, particularly Trigona species, wasps, ants, thrips, and flies, have emerged as the key pollinators of cashew trees.

In the current study, it was found that potential cashew pollinators accounted for 32.8% of the total insect mortality, highlighting a significant decline in the population of cashew pollinators. The research demonstrates that insecticide fogging has a substantial impact on non-target insects, thereby significantly affecting both the environment and crop yield.

Identification of the distribution and phylogeny of Wolbachia in wild mosquito populations in Sri Lanka.

A total of 330 individual mosquitoes, belonging to 22 species and collected from 7 provinces of Sri Lanka, underwent individual screening for the presence of Wolbachia via PCR, utilizing wsp primers. The results indicated that 26.4% of the mosquitoes were positive for Wolbachia infection. Out of the 22 screened species, only 4 species, namely Ae. albopictus, Culex quinquefasciatus, Armigeres subalbatus, and Mansonia uniformis, were found to be infected. However, the primary vector of dengue fever, Ae. aegypti, tested negative for Wolbachia infection. Among the species, the secondary vector of dengue fever, Ae. albopictus, exhibited a 100% infection rate among the mosquitoes collected from the Sabaragamuwa province, followed by 34.6% in the Southern province and 31.0% in the Northwestern provinces.

Results of the PCR performed with *Wolbachia* group specific *wsp* primers showed that *Ae. albopictus* is super-infected with *Wolbachia* strains belonging to A and B supergroups. Further screening using strain-specific *wsp* primers revealed that the strain of *Wolbachia* in supergroup A is *AlbA* and the strain in supergroup B is *Pip*. *C. quinquefasciatus* and *M. uniformis* were infected with *Pip* strain of *Wolbachia* which belongs to the supergroup B and *Ar. subalbatus* was infected with *AlbA* strain. According to the phylogenetic analysis of *Wolbachia* using *wsp* sequences obtained from the present study and other 43 reference sequences clearly showed that *Wolbachia* strains harbored by *Ae. albopictus* separated into A and B supergroups with strong bootstrap values. Of five *Wolbachia* sequences obtained from *Ae. albopictus*, three were in super group A and two were in super group B. *Wolbachia* harbored by *C. quinquefasciatus* and *M. uniformis* were separated into *Wolbachia* supergroup B.

Conclusion

The incidence of dengue is rapidly increasing in Sri Lanka, prompting concerns about the efficacy of space spraying of adulticides, both in terms of its actual impact on mosquitoes and the extent of damage it causes to non-target insects. Urgent measures are required to explore alternative techniques, such as Wolbachia-infected replacement strategies, to effectively combat dengue. To achieve this, understanding the resistance levels of Ae. aegypti and Ae. albopictus mosquitoes to the currently used pyrethroids and DDT, including their underlying resistance mechanisms to insecticides, is crucial. Identifying the three mutations associated with the VGSC, resulting in kdr-type resistance, can strengthen the country's vector control programs.

The continuous exposure to pyrethroids can lead to the easy selection of mutations present in VGSC proteins of Ae. aegypti on a larger scale. In the absence of elevated esterase-based and insensitive AChE mechanisms, organophosphates and carbamates emerge as viable alternatives for controlling Sri Lankan dengue vectors. Findings from fogging studies underscore the detrimental impact of insecticide fogging on non-target insects and the environment, highlighting the urgent need to prioritize alternative vector control methods.

Moreover, the present study represents the first report of Wolbachia presence in four mosquito species in Sri Lanka. This provides important information to develop novel approaches in using *Wolbachia* to mosquito control programmes.

Source of Funding

National Research Council (NRC) of Sri Lanka grant no. NRC 14-94.

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IMPROVED BAYESIAN MODELS FOR HOMOSCEDASTIC AND HETEROSCEDASTIC METHOD COMPARISON DATA

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Method comparison can be considered as an essential and colossal aspect of clinical science. In essence, a method comparison could be effectively explained by the studies that are performed to compare a new method with the existing standard reference method to check whether the prevailing method could be replaced by a new method. If the two distinct methods agree sufficiently well, the cheaper or, the less invasive method will be preferred, and they can be used interchangeably. Interchangeability in method comparison refers to the situation where the measurements obtained from the new method for a subject are very close or similar to those obtained from an existing method, allowing the new method to replace the existing one in clinical interpretation. Statistical method comparison studies are employed to achieve this assessment.
Any measurement method is subjected to errors in measurement that cause the observed values to differ from the true values. Also, the percentage of errors varied according to the method. Moreover, these types of data should be handled extremely carefully and accurately. Because of that reason, identification of the most appropriate and accurate measurement method is essential.

The objectives of the present study were,

(I) To introduce the Bayesian model for methods comparison data when those measurements follow the homoscedasticity and verify the characteristic of the model using a simulation study carried out using MCMC techniques.

(II) To extend the proposed model for homoscedasticity to deal with heteroscedastic measurements by adding heteroscedastic dealing parameters and verify the characteristics of the model using simulation under different settings by the MCMC technique.

Methodology

Proposed Bayesian Linear Regression Model for Homoscedastic Measurements.

The first step of this proposed model is to introduce the Bayesian Linear Regression Model for method comparison studies that satisfy the homoscedastic condition. A simulation study to

verify the model performance is considered as the second step. The simulation study was used to verify the performance of the new model and the accuracy compared with the existing related models. The third step is to evaluate the agreement between the two methods under the proposed model.
Let y_i; i =1, 2…,n be the observed test response, and x_i; i =1,2,...,n be the observed reference response on the i-th measurement. Besides, y_i^'s are assumed to be normally distributed with mean μ_i and variance σ^2, where μ_i has a linear relationship with x_i which is not estimated as a single value. Then, the proposed Bayesian model is defined as,
Y_i=α+β*X_i+ε_i ; i=1,2,...,n,
where μ_i= α+β*X_i and 〖ε〗_i ~N(0, σ^2).
Y_i \α,β,σ^2 ~ N(α+β*X_i,σ^2)
where α and β are constants.

The precision of this model can be expressed as, τ = 1⁄σ^2 . The Cardiac Ejection fraction dataset was used to illustrate the proposed model.

Proposed Bayesian Regression Model for Heteroscedastic Measurements
The primary purpose of this proposed model is to address heteroscedasticity, which is achieved using a non-linear equation. In this study, we selected various prior distributions based on past literature and evaluated their performance. Error values were calculated for each prior, and the one with the lowest error value was identified as the prior for this research work. The error measurements used include Mean Square Error (MSE), Mean Absolute Error (MAE), and Mean Absolute Percentage Error (MAPE). The simulation process of this model was carried out under six different settings based on the strength of the variance and agreement. The model's behavior was evaluated by calculating point estimators for each parameter, credible intervals, and coverage probabilities.

The extended version of the homoscedastic model for the heteroscedastic model,
Y_i=∝+β*X_i+ε_i where i=1,2,…,n
*μ*_i= ∝+β*X_i and 〖ε〗_i ~N(0,σ_i^2)
Y_i \∝,β,σ_i^2 ~ N(∝+β*X_i,σ_i^2)
where ∝ and β , are constants.

The precision of this model can be expressed as, τ = 1⁄(σ_i^2). A gold particles dataset was used to examine the properties of the proposed model and get an insight into the practical view of the model.
Results and Discussion

According to the simulation results of the homoscedastic model, in each sample size, the setting related to the medium agreement obtained the highest coverage probability when compared to the others. Small sample sizes have higher coverage probabilities than others related to the agreements in each setting.

Hecteroscedatic model simulation results show that when the sample size is larger, all parameters obtain higher coverage probabilities than sample sizes in each variance and agreement level. Especially in a sample size of 500, all parameters have acceptable coverage probabilities in each setting. Moreover, when the agreement is high, the coverage probabilities of all parameters are shown higher values than other agreement levels.

According to the results of the practical illustration of the homoscedastic model, it can be identified that when the sample size increases, the coverage probability gets low. When both the sample size and the settings are considered, the highest coverage probability for both α and β was found in the setting (0.5, 0.5) in a sample size of twenty. Furthermore, according to the coverage probability, the moderate agreement gives better results for the proposed model in each different type of sample size. Also, according to the coverage probabilities, the proposed model fits well when the sample is less than 500. When comparing the proposed model with existing models, the lowest values of the error values imply the best-fitted model is the newly proposed one.

A practical illustration of the heteroscedastic model shows that coverage probabilities of ∝,β,η_0 and η_1 are imply that each estimator's probability lies between appropriate credible intervals higher than 0.95. Four error measurements are calculated, MSE, RMSE, MAPE, and SMAPE. Those values are 0.034, 0.1829, 0.4316 and 0.4811, respectively. All the low error values imply that the proposed model has high accuracy.
Conclusion

The Bayesian linear regression model demonstrated excellent performance for homoscedastic measurements in method comparison data, outperforming other existing models in terms of accuracy, model fitting ease, time efficiency, and its assumption-less nature. Additionally, it exhibited strong performance even with sample sizes of up to 500. The second

Bayesian model proposed in this study introduces a new approach to Bayesian inference for heteroscedastic measurements in method comparison. The proposed model showed a good fit with low error values and high coverage probabilities. Simulation results indicated that all parameters had coverage probabilities of 0.95 or higher, signifying the superior fit of the proposed model. Furthermore, the model appeared to be most suitable for sample sizes of around 500, but it did not perform as well with sample sizes below 20. One limitation of the proposed model is its relatively poor fit with small sample sizes.

Source of Funding
Self-Funded.

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Institutions where the research was carried out.
Postgraduate Institute of Science, University of Peradeniya, Sri Lanka.

MODIFIED ANT COLONY OPTIMIZATION ALGORITHM FOR SOLVING A TRANSPORTATION PROBLEM LEADING TO OPTIMAL SOLUTION

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Transportation problem (TP) is another well-known optimization problem which aims to minimize the total cost of distributing resources from sources to destinations. Various techniques have been developed to solve TP, with some focusing on finding an initial basic feasible solution (IBFS) and others aiming for the optimal solution (OS). The Northwest, Least Cost, and Vogel's Approximation methods are used to find an IBFS, while the MODI Method and Steppingstone Methods are used to find an OS. However, these methods often fail to find an optimal or near-optimal solution within a reasonable amount of time, especially for large-scale TPs. In this paper, we propose a heuristic method called the Modified Ant Colony Optimization Algorithm, which is based on the Ant Colony Optimization Algorithm (ACOA) and has been found to provide near-optimal solutions for large-scale TPs, satisfying the requirements to a reasonable extent.

Ant colony optimization (ACO) is a well-known biologically inspired technique that is used in combinatorial optimization problems. Marco Dorigo developed ACO in his Ph.D. thesis to address the optimal path problem in a graph. The artificial ACO algorithm is based on natural foraging behavior, in which an ant colony seeks the shortest path between a food source and the colony. In this novel approach, such as the Modified Ant Colony Optimization Algorithm (MACOA), the degree of satisfaction with the optimal solution was improved by modifying ACOA with the incorporation of the Transition Rule and Pheromone Update Rule.

The objectives of the present study were,

(I) To propose methods for solving the TP to meet demands at destinations within the capacities at origins while minimizing total transportation costs in a reasonable amount of computational time.

(II) To develop new approaches based on the ACO algorithm to solve single and multi-objective TP.

(III) To analyze the computational efficiency of the proposed methods with the different existing methods for solving TPs.

Methodology

Here, we propose a meta-heuristic solution method based on the Ant Colony Optimization Algorithm (ACOA) to solve the transportation problem, which has been successfully applied to solve several combinatorial optimization problems.

The principles of ACO are based on the natural behavior of ants. Ants must search for food about their nest daily. While on such a quest, the ants leave a chemical substance known as a pheromone on the ground with two objectives in mind. On the one hand, it allows ants to return to the nest, but on the other hand, it permits other ants to know the path they have taken so that they can follow it. Because hundreds or even thousands of ants exhibit this behavior, if the pheromone were laid on the ground as a kind of light, the ground would be a large network with some arcs brighter than others. Dorigo et al. (1996) developed the first foraging ant algorithm, the Ant System. Using the notation in Dorigo et al. (1996), the first foraging ant algorithm, Ant System, was developed.

There are two main rules followed by the ants as the pheromone intensity on the path between the cities i and j at time t is the information provided by the algorithm. The heuristic information controlling the move from city i to city j can be determined by a heuristic algorithm according to the problem to be solved.

Generally, it can be given as:

$$\eta_{ij} = \frac{1}{d_{ij}} , \qquad (1)$$

where d_{ij} is the length of the path between the cities *i* and *j* .

By using the two rules, at time *t*, the ant *k* at city *i* selects the next city *j*, which it has not yet visited, according to the following probability:

$$p^k{}_{ij} = \begin{cases} \dfrac{\tau^{\alpha}{}_{ij}\eta_{ij}{}^{\beta}}{\sum_{j\in N_i{}^k}\tau^{\alpha}{}_{ij}\eta_{ij}{}^{\beta}} & if\ j \in N_i{}^k \\ 0 & if\ j \notin N_i{}^k \end{cases} , \qquad (2)$$

where $\alpha \geq 0$ and $\beta \geq 0$ are adjustable parameters describing the weights of the pheromone trail and visibility when choosing the route. When $\alpha = 0$, the nearest city is chosen, which corresponds to a *greedy algorithm* in the *classical optimization theory*. When $\beta = 0$, only the *pheromone* trail is taken into account, which implies that all ants select one *suboptimal* route. To provide good optimization dynamics, it is recommended to set $\beta \geq \alpha$. $N_i{}^k$ is the feasible neighborhood of ant *k* when it is at city *i*, that is, the set of cities that ant *k* has not visited yet. In addition, the ACO algorithm has been successfully applied to solve a wide range of combinatorial optimization problems, including the minimum spanning tree, the traveling salesman problem, the transportation problem, and the quadratic assignment problem with various modifications. (e.g., $\alpha = 1$, and $\beta = 1$).

After the ant completes one cycle (visiting all cities), the pheromone intensity on all paths should be updated. That is, after the paths of the ants have been built, the pheromones for all ants are updated using the following equation:

$$\tau_{ij}(t+1) = \rho\tau_{ij}(t) + \sum_{k=1}^{m} \Delta\tau^k{}_{ij}, \qquad (3)$$

where ρ is the residual ratio of the pheromone. However, to avoid an infinite accumulation of the pheromone, ρ must be less than 1. $\Delta\tau^k{}_{ij}$ is the increase of the trail level on edge (i, j) caused by ant *k*. Depending on the problem, there are three descriptions of $\Delta\tau^k{}_{ij}$, as follows:

$$\Delta\tau^k{}_{ij} = \begin{cases} \frac{Q}{T^k} & ;\text{ if ant } k \text{ travels on edge } (i, j) \\ 0 & ; \qquad \text{Otherwise} \end{cases} \qquad (4)$$

$$\Delta\tau^{k}{}_{ij} = \begin{cases} \frac{Q}{d_{ij}} & ; \text{if ant } k \text{ travels on edge } (i,j) \\ 0\,; & \text{Otherwise} \end{cases} \qquad (5)$$

$$\Delta\tau^{k}{}_{ij} = \begin{cases} Q & ; \text{if ant } k \text{ travels on edge } (i,j) \\ 0 & ; \qquad \text{Otherwise} \end{cases} \qquad (6),$$

where Q (> 0), is an adjustable parameter which is the quantity of pheromone laid by an ant per tour and T^k is the length of the tour that ant k has found. In the three descriptions above, global information is used for the first, while local information is used for the other two descriptions. Generally, the first description is used. A MACOA method is proposed in this study to identify the optimal solution for TP. The algorithm's performance is evaluated in different simulated settings and compared to existing methods. The results demonstrated that the novel method outperforms existing methods. This novel method for addressing TP is simple, easy to understand, and useful for decision making; it provides the minimum solution to TP.

Using the new modified probability function (7) from (3).

$$p_{ij} = \begin{cases} \dfrac{\frac{1}{c_{ij}+\theta}}{\sum_{(i,j)\in N_{(i,j)}} \frac{1}{c_{ij}+\theta}}, & if\ (i,j) \in N_{(i,j)} \\ 0, & if\ \ (i,j) \notin N_{(i,j)} \end{cases} \qquad (7)$$

where , $N_{(i,j)} = (i = 1,2,\dots m; j = 1,2,\dots n)$, $\alpha = 0,\ \beta = 1$, and $\theta =$ minimum c_{ij} .

The TP can be formulated as an LP model and is usually represented in a tabular form. Let us assume that, in general, a particular product is manufactured at m production plants known as supply, denoted by $S_1, S_2, \dots, S_m$ with respective capacities $a_1, a_2, \dots, a_m$, and distributed to n distribution centers known as demand, denoted by $D_1, D_2, \dots, D_m$with respective demands $b_1, b_2, \dots, b_n$. Also, assume that the transportation cost from i^{th} - supply to the j^{th} - demand is c_{ij}(unit transportation cost) and the amount of product shipped is x_{ij}, where i = 1, 2,..., m and$j$ = 1, 2,..., n. The following is known as the transportation cost table (table 1):

Supply / Demand	D_1	D_2	D_3	...	D_n	Supply
S_1	c_{11}	c_{12}	c_{13}		c_{1n}	a_1
S_2	c_{21}	c_{22}	c_{23}		c_{2n}	a_2
⋮	⋮	⋮	⋮		⋮	⋮
S_m	c_{m1}	c_{m2}	c_{m3}		c_{mn}	a_m
Demand	b_1	b_2	b_3	…	b_n	

The novel method transforms transportation cost, demand, and supply (Table .1) into a probability table (Table 2) using formula (7):

Table 2: Probability table for MACOA

Supply / Demand	D_1	D_2	D_3	...	D_n	Supply
S_1	p_{11}	p_{12}	p_{13}	⋯	p_{1n}	a_1
S_2	p_{21}	p_{22}	p_{23}	⋯	p_{2n}	a_2
⋮	⋮	⋮	⋮		⋮	⋮
S_m	p_{m1}	p_{m2}	p_{m3}	⋯	p_{mn}	a_m
Demand	b_1	b_2	b_3	…	b_n	

The new approach involved solving a TP with a MACOA. Use the updated ACO approach to convert any TP to probability values (table 2) and then proceed with the iteration method. The steps involved in achieving the best performance by MACOA are as follows:

Step 1: Construct the initial transportation table $m \times n$ with unit cost c_{ij} , supply a_i and demand b_j for i = 1, 2...,m and j = 1, 2,..., n.

Step 2: Compute the path according to the probability table (Table 2).

Step 3: Initially, start an allocation process from a minimum of supply/demand. Allocate this minimum of supply/ demand in the place of the maximum probability cell in Step 2. If demand in the column (or supply in the row) is satisfied, delete the column (or row) then move to the next maximum probability cell.

Step 4: Repeat this process until all supplies and demands are satisfied, then proceed to Step 5. Otherwise, proceed to Step 3.

Step 5: Stop and calculate the minimum transportation cost.

Results and Discussion

The efficiency of the proposed method was compared with several cost-minimizing TPs, and it showed a comparatively better result. Because the OSs obtained by applying the MACOA to the vast majority of problem instances were based on benchmark instances from the literature, they were used to evaluate the heuristics based on solution excellence. In the context of benchmark instances and randomly generated problems, the proposed heuristic approach produced OSs. Finally, the MACOA is an excellent and effective method for achieving near-optimal performance by acquiring minimal iteration and running time. However, the MODI method is guaranteed to find the OS every time, whereas the case for MACOA is not the same.

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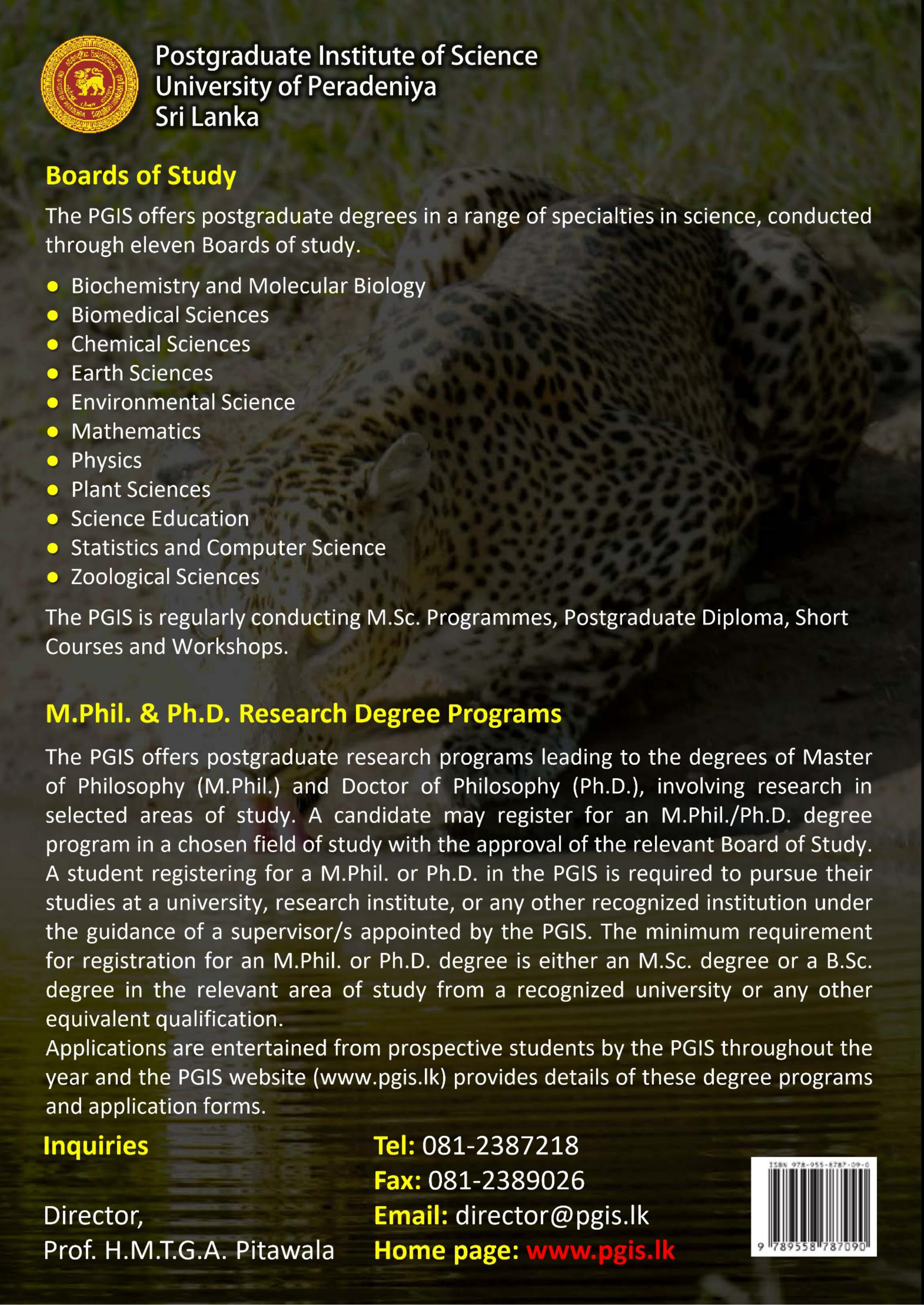

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ISBN 978-955-8787-09-0
9 789558 787090