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COVER STORY

Captured in a moment of breathtaking beauty, this image emerged as the most popular entry in the IOBSL e-Photography Competition 2023

Photograph by Mr. Achchuthan Seetharan

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CORNER FOR YOUNG BIOLOGISTS

IOBSL NEWS AND EVENTS

Highlights from the IBO 2024 Training Sessions

International Biology Olympiad 2024 training sessions were successfully held at Allergy, Immunology and Cell Biology Unit at Faculty Medical Sciences, and Ecosphere Resilience Research Center at the University of Sri Jayewardenepura, Sri Lanka, and Zoology and Biochemistry Labs at the University of Ruhuna, Sri Lanka. These sessions provided valuable insights and hands-on experience for the participants.


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Sri Lankan Team Shines at International Biology Olympiad 2024 in Kazakhstan

A team of four exceptionally talented Sri Lankan biology students participated in the International Biology Olympiad (IBO) 2024, held in Kazakhstan. The distinguished team comprised:

- Neth Wijesooriya from Ananda College, Colombo
- Methasa Wijyeyasuriya from Sirimavo Bandaranaike Vidyalaya, Colombo
- Gamitha Senaratna from Richmond College, Galle
- Senulya Samarawickrama from Sujatha Vidyalaya, Matara

The team was accompanied by Prof. Hiran Amarasekera and Emeritus Prof. M. J. S. Wijeyaratne, who provided invaluable support and guidance throughout the competition.

Applications are now open for the Sri Lankan Biology Olympiad 2024

Sri Lankan students have the exciting opportunity to compete and win medals at the Sri Lankan Biology Olympiad 2024. The top four students from this national competition will be selected to represent Sri Lanka at the International Biology Olympiad (IBO) 2025, which will be held in the Philippines.

This competition not only offers a platform for students to showcase their biological knowledge and skills but also provides a chance to advance to the international level. It is an excellent opportunity for aspiring young biologists to make their mark on the global stage.

For more information, please visit https://www.iobsl.org/olympiad/sri-lankan-biology-olympiad-2024

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Sri Lankan Journal of Biology Earns Prestigious Indexation in ASCI-Database: A Milestone in Academic Excellence!

The Sri Lankan Journal of Biology (ISSN 2513-2245) has been accepted for indexation in the prestigious Asian Science Citation Index (ASCI)-Database!

This recognition highlights the journal's academic significance and its valuable contributions to scholarly research.

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Published by The Institute of Biology, Sri Lanka

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The BIO-NEWS editorial team extends its warmest congratulations and best wishes to everyone involved in this milestone

FEATURE ARTICLES

From Serendipity to its Secret Sauce: Connecting the Dots of *Candidatus* (*Ca.*) *Thiomargarita magnifica*, a Giant Macroscopic Bacterium

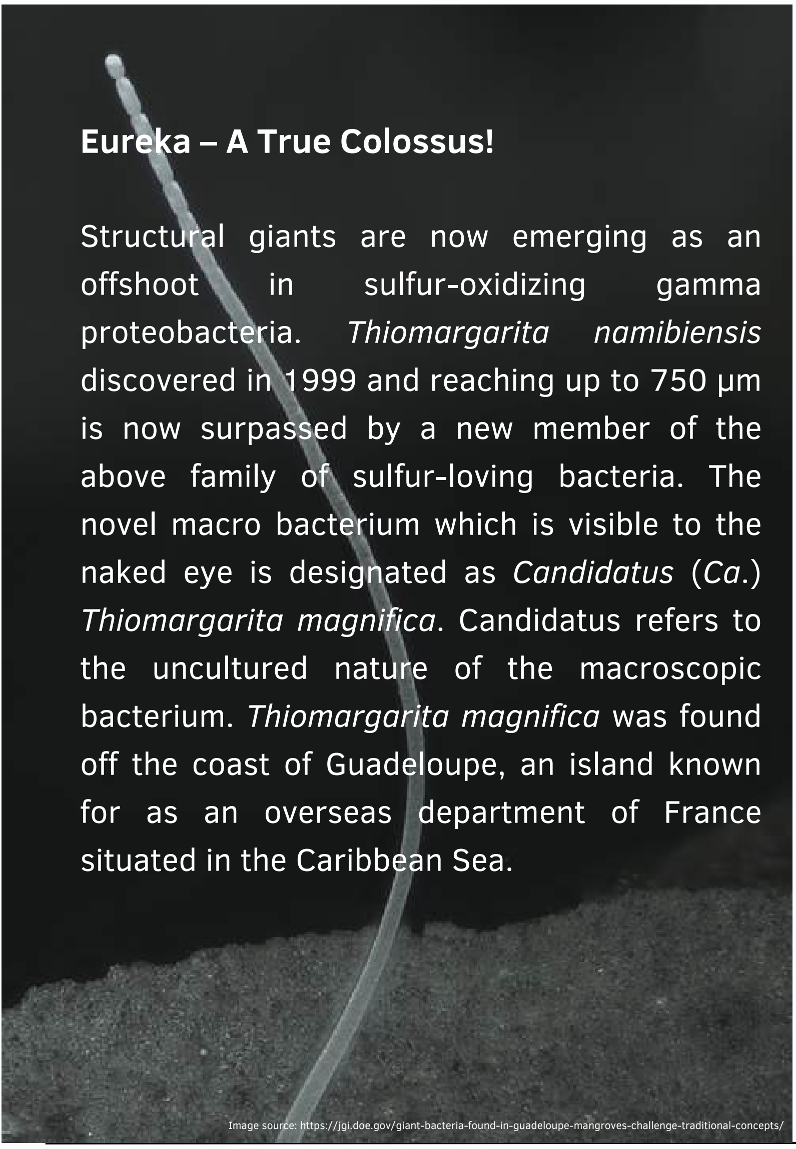

Image source: https://jgi.doe.gov/giant-bacteria-found-in-guadeloupe-mangroves-challenge-traditional-concepts/

Eureka – A True Colossus!

Structural giants are now emerging as an offshoot in sulfur-oxidizing gamma proteobacteria. *Thiomargarita namibiensis* discovered in 1999 and reaching up to 750 µm is now surpassed by a new member of the above family of sulfur-loving bacteria. The novel macro bacterium which is visible to the naked eye is designated as *Candidatus* (*Ca.*) *Thiomargarita magnifica*. Candidatus refers to the uncultured nature of the macroscopic bacterium. *Thiomargarita magnifica* was found off the coast of Guadeloupe, an island known for as an overseas department of France situated in the Caribbean Sea.

Thiomargarita magnifica has been identified as sessile, being able to move while being fixed to a substratum but not having the capacity of free locomotion away from the bedrock. The environment that *T. magnifica* was identified from can be classified as a sulfur rich environment, where sulfur oxidation plays the central role in energy production. The sulfur oxidizing bacterium is known to outsize the next-in-line giant bacterium - *Thiomargarita namibiensis* - by a whopping 50 fold.

Dr. Dilantha Gunawardana, M. I. Biol. (Sri Lanka)

Connecting the dots: a healthy dose of biology

In order to characterize the structural details of *Ca. T. magnifica*, the membranes of this prokaryotic giant were highlighted by using osmium tetraoxide and a fluorescent dye known as FM 1-43X, to observe the 3-D arrangement of the cell, using X-ray tomography and new tricks in confocal microscopy. Electron microscopy allowed the scientists to measure the size of cells spanning the whole length to the apical extremity. There was some degree of tapering towards the distal end with the cytoplasm narrowing till the apical tip. There were no septa or walled structures separating the filamentous cell into smaller subunits. The “budding” of daughter cells took place at the apical end and was instrumental for the generation of new cells.

A large vacuole sits at the center of the cell, which took up 73.2 ± 7.5% (n = 4) of the total volume of the elongated bacterium. The cytoplasm took a narrow path around the vacuole being only 3.34 ± 1.48 μm thick, therefore allowing for efficient metabolic diffusion to be the cornerstone of its biochemical needs. Lucid vesicles made of deposits of sulfur showed a dotted appearance in the cytoplasm.

The unprecedented took the shape of compartmentalized DNA and ribosomes surrounded by membranous structures, showcasing that there was a jump in bio-complexity for this marine bacterium. These “blebs of cytoplasm” either contained ribosomes with DNA fibrils or exclusively enveloped nuclear material or took the shape and identity of “endobionts”. The scientists of the study could not find relevant answers to these membrane-bound organelle-like structures in their initial dive into the biology of this macroscopic wonder. However, it is suspected that compartmentalized DNA bolsters the polyploidy of the identified species, a common signature in many giant bacteria identified this far.

Biological appartments: floor plans and furniture

The compartmentalization of functions of bacteria has been shown to offer anaerobic ammonium oxidation, photosynthesis, and magnetic orientation as a bacterium’s newly acquired skill sets. Therefore, it was not totally surprising that to accommodate such a giant structure, there was the need to compartmentalize both the DNA and protein making machinery. Deep but incomplete invaginations have been shown to occur in many bacteria (such as *Gemmata obscuriglobus*) prior to the discovery of *T. magnifica*. These incomplete prototypes - stepping stones of future membrane-bound compartments -, show the way in which biology evolves in steps to nestle critical molecules (DNA, etc.) and assembly units (ribosomes) in nurtured easy-to-control environments. FISH (Fluorescence In Situ Hybridization) which entails targeting probes to ribosomal RNA, was the method of choice to demonstrate that ribosomes were indeed compartmentalized. The compartments harboring ribosomes and DNA were named as “pepins”, the etymology derived from pips that are encountered in watermelons and kiwi fruits, a term for “smallness” that is symbolic of the miniature size of the compartment next to eukaryote concentrates bordered by enveloping membranes.

ATP synthase, which is the custodian of energy production (to build chemical blocks called ATP) is known to be located on the inner mitochondrial membrane in eukaryotes, while in bacteria, ATP synthases are localized on the exterior cell membrane. Immunohistochemistry narrowed down the ATP synthases to pepins and to the interconnected network of membrane compartments but not to the exterior cell envelope. This heralds new life into the taxonomic unit we call bacteria where the compartmentalized production of ATP will be beneficial to the distribution of ATP synthesizing units, and consequently the logistics and availability of the chemical ATP for bioenergetics.

Lost in translation

Bioorthogonal noncanonical amino acid tagging (BONCAT) was the method of choice for the detection of protein production. BONCAT revealed that protein synthesis was localized to certain pepins but it was not universally distributed to all pepins. The apical areas of the filamentous bacterium possessed stronger protein production activity than the basal parts of the cell and consequently these “active pepins” were called protein synthesizing hotspots. The growth of an axenic culture will be a requirement of the future, to zoom in on the molecular events that are symbolic of the pepins, as well as the growth and proliferation events of this macro-bacterium. It was observed that the *T. magnifica* did not have to double its cell volume to engage in fission, since apical budding was the method of choice to form bacterial offspring.

Diversity in ploidy: genetic stockpiles

Polyploidy leads to higher degrees of gene duplication, reassortment, and divergence that could be the impetus for a rich diversity of unique and rare biochemical signatures. In all, *T. magnifica* contains the highest estimated number of genome copies of any living organism, and this apical level of genome duplication, containment, compartmentalization, and preservation, requires studies that will hone in on the remarkable biology of this macro bacterium. A largely homogenous genome population sets the stage for harmonized biology – all cells appeared to contain the same signature of DNA assemblies. *Thiomargarita magnifica* appears to contain up to 11788 genes with half of unknown pedigree, which points to a high degree of neofunctionalization (finding new roles in biology). The gene families for sulfur oxidation and carbon fixation appear to be highly enriched, suggesting chemoautotrophy, and specifically thioautotrophy.

The lower number of epibiotic bacteria is indicative of the opulence of secondary metabolites produced by the assortment of metabolic pathways that may discourage the growth of mutually beneficial bacteria on external surfaces. It was also evident that cell division genes were found lacking in the genome of this macroscopic bacterium but cell elongation genes were found in their entirety, which may shape the filamentous and elongated nature of the macroscopic cell. The budding of apical daughter cells brings into perspective that there will only be a subset of the pepin genomes in the bacterial offspring, and this points to a dimorphic cycle where parent and daughter possess a distinct genomic signature.

This sort of budding phenomena draws parallels to dispersion by fruiting bodies of the social myxobacteria or aerial hyphae of *Streptomyces* spp.

Alice in Ponderland

In a synopsis, this is a wondrous discovery that will make us look through the rabbit holes of under sampled niches that may harbor a richness of macro contenders that may defy the standards of biology but will not defy logic or a gradual switch towards complexity. The most enthralling story in this uncharted new base of biology is the compartmentalization of DNA fibrils and protein production units, together and in seclusion. Harboring two types of molecular contenders in fusion, points to the easy logistics of coupled transcription and translation as a possible phenomenon that will benefit the biological pathways. Energy derivation from internal ATP synthases also points to a function comparable to mitochondria that may offer less labor costs for the maintenance of cells.

Long, narrow and oozing with confidence due to its polyploidy genome architecture containing a rich collection of unique genes, make this specimen a true novelty.

Culturing this giant – or the dispersion prone daughters – will be the next logical step. An axenic culture will help in the further detection of biology, and it is time that scientists believe that Alice in Wonderland is not the only place for freaks of biology, the waters of Guadeloupe are right up there.

From holler to ruler

In Alice's adventures, she encounters a 3 cm long hookah smoking caterpillar that can be compared to this centimeter-long bacterium that smokes sulfur. Just like a caterpillar turns into a butterfly, we do not know the catalog of wonderful findings that await the team of scientists of this study. So let's empower science to define the next paradigm of findings of this megalo bacterium, to culture it on a petri dish, and dissect the secret sauce that makes it what it is in the contemporary – *a freak so far, but only until the next serendipitous discovery*. An incremental trend spiraling upwards is associated with the size of mimiviruses and dinosaurs, and now by extension, centimeters-long filamentous bacteria.

Here's a mighty "Eureka" to the next exception to the rule(r) !!!

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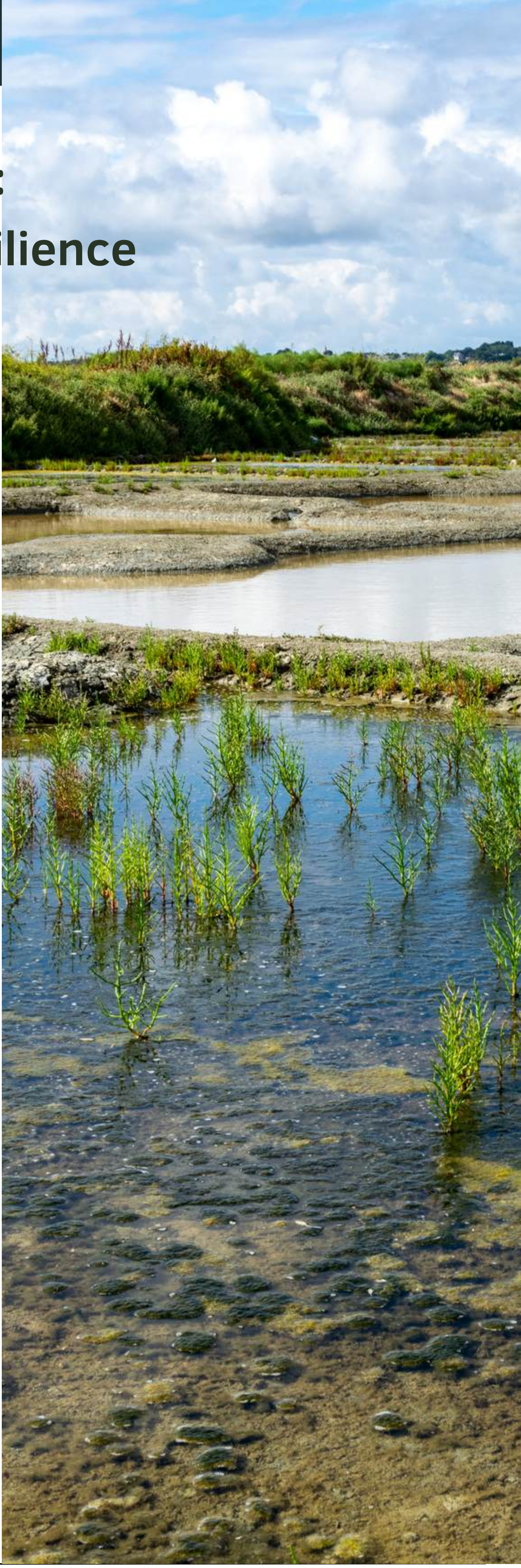
Salt Marshes in Sri Lanka: Guardians of Coastal Resilience



The salt marsh habitat is a hidden gem of nature, representing one of the most productive and valuable ecosystems between the ocean and land. This remarkable ecosystem, as part of the coast, provides essential services to marine flora, fauna, and other animals that rely on it. Every day, the ebb and flow of tides define the rhythm of life in a salt marsh. Often submerged by the tides, the fauna that thrive here face extreme conditions, from fluctuating salinity levels to drought, high temperatures, direct exposure to UV radiation, and waterlogging. Interestingly, these organisms have shown special adaptations to cope with these stresses.

Worldwide, salt marshes offer an array of essential services. They supply food and medicine, provide habitat for coastal biota, maintain biological diversity, and serve as resting areas for migratory birds. These habitats are crucial nursery grounds, feeding and nesting areas, and breeding grounds for a diverse range of organisms, including terrestrial species like angiosperms, mammals, insects, and birds, as well as marine organisms such as algae, mollusks, crustaceans, and fish. Moreover, salt marshes protect shorelines from erosion by acting as a buffer against wave action and by trapping soils. In flood-prone areas, they reduce the flow of floodwaters and absorb rainwater, mitigating the impacts of inundation.

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By filtering runoff and excess nutrients, salt marshes also contribute to maintaining water quality in coastal bays, sounds, and estuaries. Beyond that, salt marshes serve as "carbon sinks," sequestering carbon that would otherwise be released into the atmosphere and contribute to climate change. Figure 1 depicts the associated ecosystem services of the salt marsh ecosystem.

More than 500 species of salt marsh plants have been identified worldwide, with 18 species recently reported in South Asia alone. Of these, only seven species have been recorded in Sri Lanka. Spread over a total area of 23,819 hectares, the salt marshes of Sri Lanka are mainly found in the districts of Puttalam, Gampaha, Mannar, Jaffna, Kilinochchi, Mullaitivu, Trincomalee, Batticaloa, Ampara, Hambantota and Galle.

The seven species of salt marsh plants identified in Sri Lanka namely *Sesuvium portulacastrum*, *Salicornia brachiata*, *Suaeda maritima*, *Suaeda monoica*, *Suaeda vermiculata*, *Tecticornia indica* (*Halosarcia indica*) and *Cressa cretica* have been well evaluated for their unique physical characteristics.

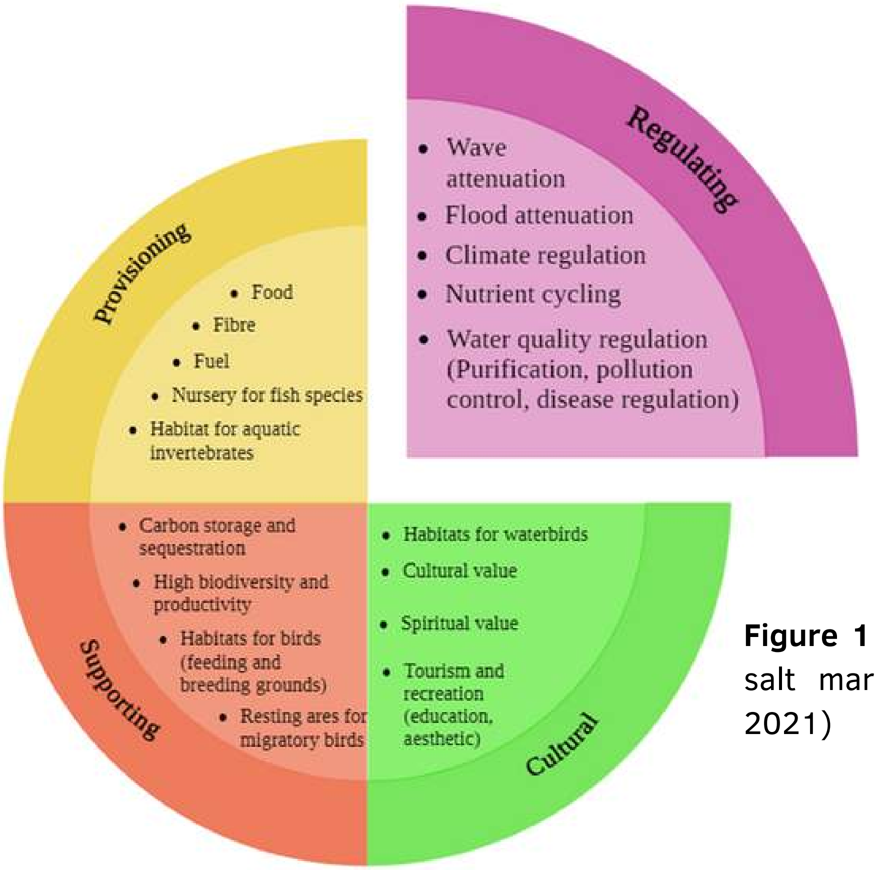

Figure 1 Ecosystem services provided by the salt marshes (adopted from Adams et al., 2021)

Regulating services of salt marshes as a coastal guardian

Salt marshes provide a variety of benefits, known as 'ecosystem services,' and one of the most important is their role as buffers in protecting coastlines, acting as coastal guardians of nature. Serving as natural buffers, they defend against coastal hazards such as erosion, flooding, and storm surges. Their resistance against erosion and subsequent change in marsh elevation, such as morphological stability, is crucial in withstanding wave forcing. Additionally, the strength of the vegetation layer plays a crucial role in benefiting from wave attenuation and bottom friction, further enhancing their protective capabilities.

Wave attenuation

As waves pass through marsh vegetation, there is a reduction in wave strength and height. Frictional drag caused by vegetation and bottom friction in shallow water areas maintained by marshes accounts for this reduction. Characteristics of the vegetation, such as density, biomass production, and marsh size, play key roles in positively influencing both wave attenuation and shoreline stability. The presence of extensive marshes with dense and productive vegetation is crucial for effective wave energy reduction and shoreline stabilization, highlighting their importance in coastal protection.

Floodwater attenuation

Salt marshes not only act as "wave absorbers" but also function as "flood fighters" that lower flood depth by limiting the size of breaches. They serve as natural flood absorbers, effectively reducing the impact of flood peaks in coastal areas. During heavy rainfall or storms, salt marshes absorb excess water, slowing its flow and reducing the volume of water reaching inland areas. The dense vegetation and complex root systems of salt marsh plants play a critical role in this process, providing natural barriers that trap and hold back floodwater.

Storm surge control

Salt marshes mitigate the effects of storm surges and coastal erosion caused by wind waves. These ecosystems are particularly effective at managing surges from fast-moving storms compared to slow-moving ones, which can push water beyond the natural barriers provided by the marshes. The structure of salt marshes reduces water velocity, accounting for about 60% of wave dissipation, with the peat absorbing water and further decreasing wave energy. During small storm events, these marshes can fully absorb and neutralize wave and flood energy. In larger storms, although the marshes may be overtopped, they still help reduce wave energy and protect inland structures from storm surges.

Water filtration

In addition to acting as a sponge for excess quantities of water, marshes also function as filters. As water flows through a salt marsh, marsh grasses and peat (a spongy matrix of live roots, decomposing organic material, and soil) filter out pollutants such as herbicides, pesticides, and heavy metals, as well as excess sediments and nutrients, before it reaches coastal waters.

Carbon sequestration

Apart from the above functions, salt marshes exhibit remarkable carbon sequestration potential, primarily due to their high primary production rates and slow decomposition of soil organic matter. Carbon pools in salt marshes are influenced by the composition and structure of plant communities. The diversity and composition of plant species largely dictate the amount of above- and below-ground biomass, as well as rates of litter production and decomposition. Moreover, the developmental stage of plant communities can impact their capacity for carbon sequestration, with mature stands typically exhibiting slower vegetation growth. Additionally, soils contribute significantly to carbon storage, often containing higher carbon levels compared to vegetation. Figure 2 illustrates a schematic representation of the regulating services provided by salt marsh ecosystems.

Threats to salt marshes in Sri Lanka

Salt marshes, despite their vital ecosystem services, face increasing vulnerability due to anthropogenic threats and climate change. Primarily driven by human activities, these threats pose significant risks to these invaluable ecosystems. Natural hazards such as storm surges, floods, and sea-level rise compound these threats, with projections showing an increase in their intensity and frequency due to climate change. Globally, salt marsh loss is estimated between 25% and 50%. According to observations, human activities have the greatest impact on salt marshes in Sri Lanka. The conversion of marshlands into playgrounds, vehicle parks, tourist hotels, and shrimp farms is rampant, leading to habitat destruction and loss of biodiversity in salt marshes. The expansion of shrimp farms and salt pans, along with the

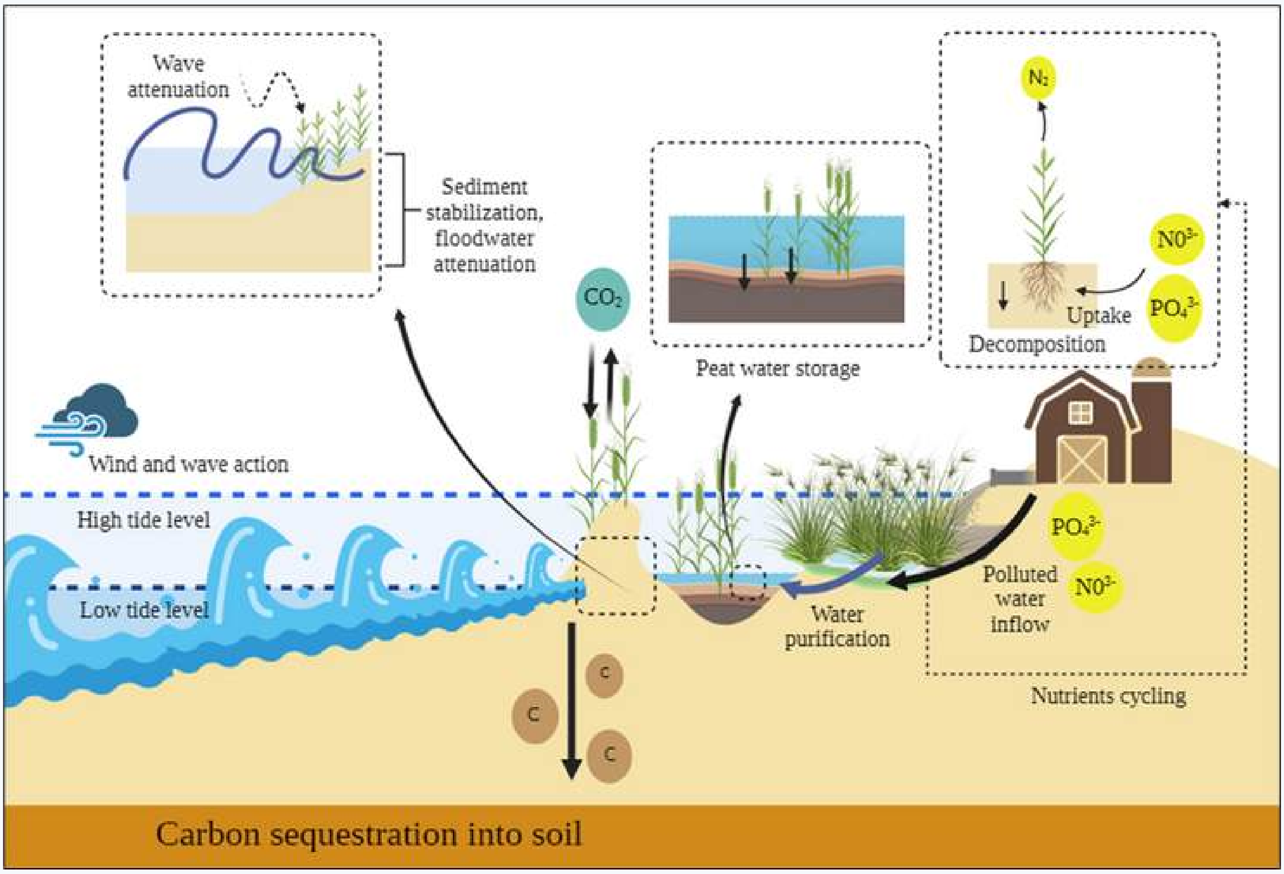

Figure 2 Schematic diagram of regulating services of salt marsh ecosystem

expansion of coconut plantations, for example, has led to a 60% increase in the area of shrimp farms and salt pans, especially reported in the Kalpitiya area within a short period.

Coconut groves have increased by 17%. Many shrimp farms are abandoned after a short period, leaving unproductive landscapes and disrupting carbon sequestration and storage services. Other than that, human activities also introduce pollutants such as fishing debris, plastics, heavy metals, and synthetic chemicals into salt marsh ecosystems, harming microorganisms, invertebrates, and other organisms (Figure 3). Oil spills, construction of landfills, unregulated dumping, and runoff from piggeries further degrade these fragile ecosystems.

The cumulative impact of these pollutants can weaken the resilience of salt marshes, making them more vulnerable to other stressors such as climate change. Natural factors such as crabs' burrowing, overgrazing by animals, tropical storms, changes in sea level, and climate change also disturb salt marsh ecosystems.

Figure 3 Marine debris pollution in salt marshes in Kalpitiya, North Western Province, Sri Lanka. (a) Clothes, (b)-(e) Plastics, (f) Fishing debris.

Sea-level rise, in particular, poses a significant threat, leading to habitat submergence and "coastal squeeze." Rising sea levels are projected to impact 90% of salt marshes by 2100, further highlighting the urgent need for conservation and mitigation efforts to safeguard these invaluable ecosystems.

Salt marshes along coastlines provide a variety of ecosystem services, including provisioning, regulating, cultural, and supporting services. They protect coastlines by attenuating wave energy, shielding against storms and floods, resisting shoreline erosion, and acting as coastal guardians. The benefits of salt marshes are particularly crucial given the increasing destruction caused by storms and flooding, which are expected to worsen with climate change.

However, salt marsh habitats face significant threats from infrastructure development projects, tourism, agriculture, and aquaculture practices. The ongoing destruction of ecosystems will inevitably diminish the products and services that people can derive from the environment in the future. To conserve these precious ecosystems, integrative and inclusive conservation and coastal management practices must be adopted carefully, ensuring that the actions taken protect the natural benefits and ecosystem services provided by salt marshes.

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Low-Carbon Dairy: Do Farms Present Tremendous Mitigation Opportunities?

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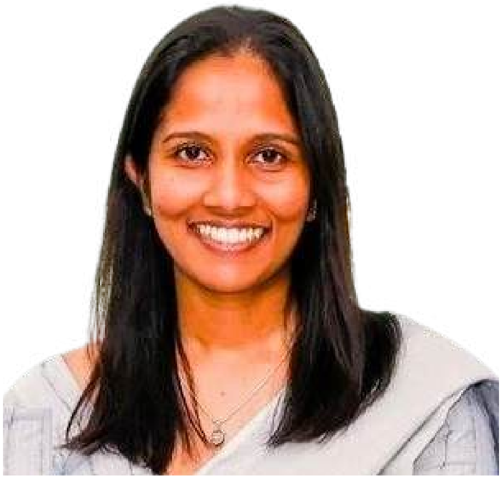

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Climate change along with population growth, poverty alleviation, environmental degradation and global food insecurity is one of the defining challenges of the 21st century. Over the past 50 years, greenhouse gas (GHG) emissions resulting from 'agriculture, forestry and other land use' have nearly doubled, and projections suggest a further increase by 2050. Within agriculture, the livestock sector has come into focus because of its large interface with the environment. Traditionally, livestock was supply-driven, converting waste material and other resources of limited alternative use into edible products and other goods and services. Its size was relatively limited and so were the environmental impacts. However, since the livestock sector has become increasingly demand-driven, growth has been faster and the sector now competes for natural resources with other sectors. Environmental impacts have become greater and the sector is often pointed out as being particularly resource-hungry.

Emergence of 'mega dairy farms'

Dairy farming is considered as a heterogeneous branch of livestock agriculture, as it has largely derived from cattle, buffaloes, goats, sheep and camels. Especially in tropical countries, these ruminant livestock are an important part of agricultural systems. They play crucial and multiple roles in smallholder systems since they produce a wide range of goods in addition to food. They generate manure for use as fuel and fertilizer, as well as direct cash income, making them capital assets.

Milk animals are usually raised in a variety of production systems such as; rural smallholder dairying, pastoral/agro-pastoral dairying, landless peri-urban dairying, large-scale dairy farming, and more (Figure 1).

Figure 1 Intensive livestock farming systems Source: Lebacq at al. (2013)

In most cases, cattle breeds in these farms have been intensively selected for milk production through the importation of breeding heifers. This is certainly occurring in many Asian countries. These stocks can be found in developed countries such as Australia, New Zealand, the United States, and Europe. However, these modern dairy farms with high-performing dairy herds around the world, share many similarities in cattle breeds, use of modern technologies, good veterinary care, dairying equipment, and record systems.

Key activities of a dairy farm

The dairy production technology of any farm which can be considered as a profitable dairy farm, regardless of its size or location, can be broken down into nine key activities as shown in Figure 3. These are also part of their supply chain, and each step must be carefully managed to avoid severe consequences for overall farm performance.

The emergence of modern commercialized large scale dairy farms to meet the global dairy requirements has expanded commercialization of the dairy sector in the last few decades. This is not a new concept in the developed world, but there is an increasing interest in establishing such "mega dairy farms" in developing countries (Figure 2).

Figure 2 Resource intensive large scale dairy farming systems in developing countries Source: Available at https://www.foresightsl.com/projects/importation-of-dairy-animals/ (Accessed: 20 January 2024)

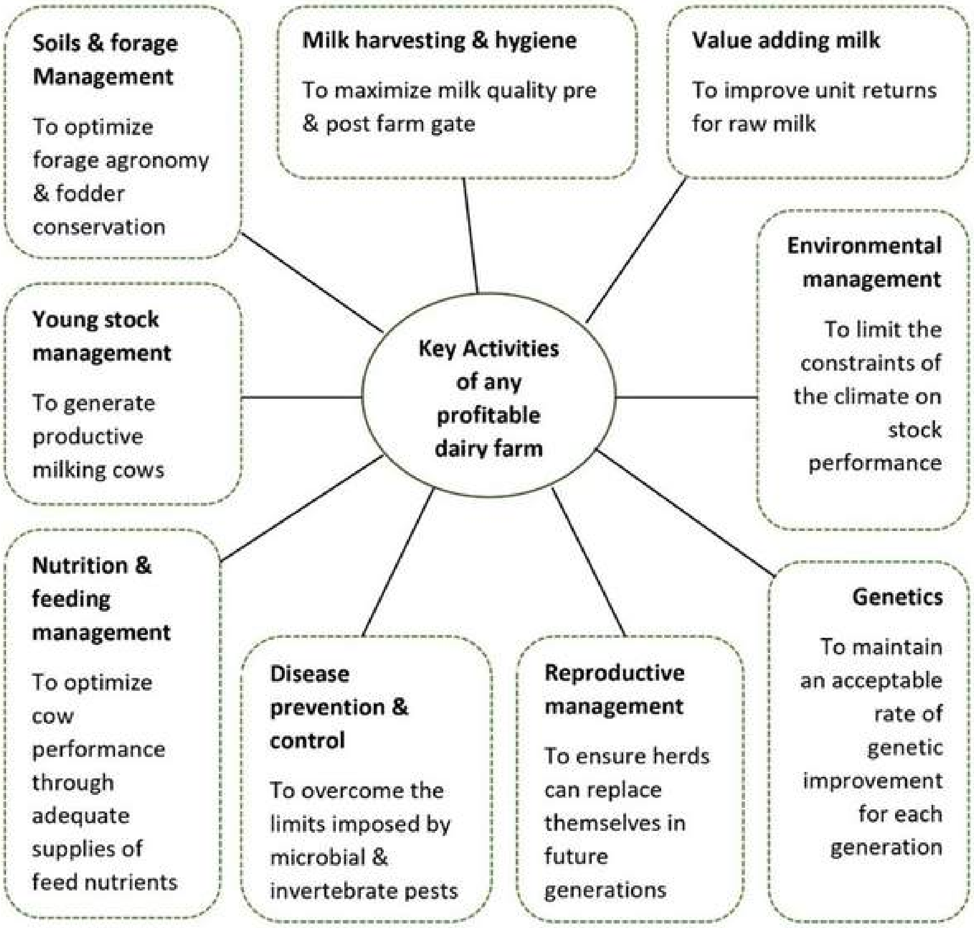


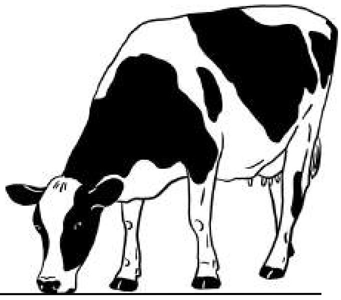
Figure 3 Key activities of a profitable dairy farm (Adapted from: Moran, 2009a; Moran and Chamberlain, 2017)

Where do emissions come from?

In dairy production, emissions result from a variety of complex biological processes and various sources and sinks throughout the farm. Dairy farms contribute significantly to total GHG emissions from milk and other dairy products over their entire life cycle. For example, methane (CH_4), nitrous oxide (N_2O), and carbon dioxide (CO_2) emissions, are the three main GHG emitted by the sector, where methane is produced as a by-product of the digestion process in enteric fermentation. Additional CH_4 and N_2O emissions arise throughout the livestock manure management processes and result from various physical, chemical, and biological processes that change over time and space in response to the ambient conditions (e.g., temperature, wind), the surrounding environment (e.g., soil, building type), characteristics of livestock such as their physiological stage, and farming operations. Other emissions include CO_2, where multiple processes emit CO_2 from dairy farms, primarily associated with energy use, input production and transportation, and land use changes. These emissions result from both upstream and downstream operations of dairy supply chains as well as from on-farm direct operations. Table 1 shows some of the sources of GHG emissions which can be resulted from a typical dairy farm.

Table 1 Sources of GHG emissions of a typical dairy farm (FAO, 2010; Gerber et al., 2013; Rotz, 2017; Guzmán-Luna et al., 2021)

Supply chain	Activity	GHG	Sources
Upstream	Feed production	N_2O	Direct and indirect N_2O from: • Application of synthetic N • Application of manure • Direct deposition of manure by grazing and scavenging animals • Crop residue management • Biomass burning • Emissions of non-N fertilizers
		CO_2 N_2O CH_4	• Energy use in field operations such as land preparation, cultivation, fertilizer application, and harvesting • Production and transport of seeds, fertilizer, and pesticides into the farm • Energy use in feed transport and processing • Feed blending • Changes in C stocks from land use under constant management practices
	Non-feed production	CO_2	• Energy use in manufacturing farm buildings and infrastructure, and equipment • Production of cleaning agents, antibiotics, and pharmaceuticals • Transport of livestock into the farm • Transport of cleaning agents, antibiotics, pharmaceuticals, concentrates all the other feed inputs from outside suppliers into the farm • Use of farm vehicles for inspections
Animal Production Unit	Livestock production	CH_4	• Enteric fermentation • Manure management
		N_2O	• Direct and indirect emissions from manure management
		CO_2	Direct energy use on-farm for livestock: • Feeding, milking, cooling, ventilation, lighting, and heating
Downstream	Post farm gate	CO_2 CH_4 N_2O HFCs	• Transport of livestock to slaughter • Transport of milk to processing plant • Transport of processed dairy products to retail points • Refrigeration during transport and processing • Manufacture of packaging materials • On-site wastewater treatment • Emissions from animal waste or energy generation from waste • Emissions related to slaughter by-products • Energy use in retailing and post retailing • Waste disposal at retail and post retail stages



Options for reducing GHG emissions from the dairy sector

Researchers have shown that the potential to reduce the sector's emissions is large. Technologies and practices that help to reduce emissions exist within the farm, but are not widely used. The mitigation potential can be achieved within existing systems by improving practices rather than changing production systems, and also can be achieved in all climates, regions, and production systems. Possible interventions to reduce emissions are to a large extent based on the adoption of more efficient technologies and practices that improve productivity at farm level. The mitigation options outlined below are consistent with improving the efficiency and profitability of the dairy farm. It has several dimensions including feeding, reproduction, health, genetics and overall management of the animal operation. In many parts of the world, particularly in low-and-middle income regions, the single most effective GHG mitigation strategy is to increase animal productivity. Adopting these practices and technologies could significantly reduce the emission intensity of milk.

Feed and feeding management: Increase feed efficiency by optimizing the energy and protein content in the feed, use precision feeding techniques to match animal requirements with dietary nutrient supply, use more locally produced feed and source low-emissions feeds such as by-products, store more carbon in the soil by means of better grassland management.

Manure management: Improve manure collection, storage, and utilization, by using cow manure in biogas systems it is possible to reduce emissions of GHGs associated with the storage of manure; improve the quality of fertilizer, and replace fossil energy sources, switch from raw to composted manures can greatly reduce emissions.

Fertilizer management (manure and commercial fertilizer): Optimize consumption relative to need, lower manure application rates, and the incorporation of manure into soils can reduce emissions while maintaining farm productivity, use commercial fertilizer produced in an environmentally friendly way with a low carbon footprint, spread fertilizer at the optimum time and with the best technology.

Energy use at the farm: Reduce fossil fuel energy use (e.g. electricity and diesel), increase the use of sustainable energy, e.g. wind energy and biofuel to replace fossil energy sources, biogas for power generation from dairy cattle.

Properly manage and optimize water use: Improving irrigation efficiency (equipment, timing, and precision), fuel switching (including renewable energy), managing plant demand (e.g. drought-tolerant varieties), conservation of agricultural practices to improve soil water holding capacity, properly managing the use of inputs and release of wastewater in surrounding water sources.

Soil quality and quantity: Maintaining good soil fertility and preventing soil erosion and pollution by growing deep-rooted pastures, maintaining a dense pasture cover, professional design and layout of a drainage system, avoiding overgrazing, avoiding cultivation through or disturbing natural drainage lines, testing the soil regularly, and use of a nutrient budget to determine fertilizer requirements.

Animal health and husbandry: Management of herd structures to reduce the number of non-productive animals through improved animal and herd fertility and reproduction is an effective approach to reduce emissions per unit of milk and increase dairy profitability, reducing the prevalence of diseases and parasites would generally reduce emissions intensity as healthier animals are more productive, and thus produce lower emissions per unit of output, improving the genetic potential of animals through planned crossbreeding or selection within breeds, and achieving this genetic potential through proper nutrition.

More ambitious action needed now

There is a clear case for immediate and more ambitious action. Dairy farmers are already part of the solution to limit climate change, but there is an urgent need to accelerate and intensify the sector's response to avoid climate tipping points. While new research and technologies will continue to be developed, many mitigation options are already available and their adoption can be accelerated. Further delaying adoption will result in a greater amount of emissions overall, given that CO_2 emissions accumulate in the atmosphere for hundreds to thousands of years. Even with implementation of best practice and technologies, it is likely that some residual emissions will remain in the future. The dairy industry will therefore need to also consider how these residual emissions will be offset.

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The Cosmological Contingencies for the Evolution of Human Intelligence

Across the vast expanse of the observable Universe, spanning over 13.8 billion years and containing over 100 billion galaxies, a highly improbable series of events produced the first intelligent life we know: humanity. The contingencies required for the emergence and existence of the human species appear to be a statistical improbability, raising questions about how frequently intelligence may arise elsewhere or whether species with greater intelligence than humans are a statistical possibility.

According to the "Rare Earth" hypothesis proposed by palaeontologist Peter Ward and astronomer Donald Brownlee (2000), multiple pivotal events had to transpire precisely in sequence for human intelligence to emerge. This cosmic lottery spanned billions of years, from the earliest seeding of heavy elements to the environmental triggers that transformed ancient apes into modern humans. This article examines these pivotal cosmic contingencies.

The metallicity of life's raw ingredients

The Big Bang theory describes the birth of our Universe around 13.8 billion years ago as an incredibly hot, dense mathematical singularity that began expanding rapidly in all directions. At the beginning of the Big Bang, all the matter and energy in the observable Universe were condensed into an extremely hot, immensely dense point.

In the first few minutes after the initial expansion, the Universe consisted of high-energy radiation and fundamental particles like quarks and leptons. As it cooled, these particles combined to form the simplest atomic nuclei—hydrogen, helium, and trace amounts of lithium and beryllium.

Over 4.5 billion years ago, the solar nebula—a vast, rotating cloud of gas and dust—condensed to form our Sun and planets. Initially, this solar nebula consisted primarily of hydrogen and helium left over from the Big Bang, along with trace amounts of lithium and beryllium. The other heavier elements critical for planetary bodies and biochemical life—like carbon, nitrogen, oxygen, and iron—could only have been supplied from stellar synthesis, the nucleosynthetic process of element generation in massive stars.

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The emergence of life as we know it critically requires elements like carbon, nitrogen, oxygen, and phosphorus that can only be synthesised in stellar interiors. These elements form the backbone of biological macromolecules such as DNA, RNA, proteins, and lipids, which are essential for cellular structure and function.

The first cosmic contingency was the enrichment of the solar nebula with elements heavier than hydrogen and helium, known as metallicity. Evidence for this enrichment is found in some of the oldest known materials in the solar system—refractory calcium-aluminium-rich inclusions (CAIs) within primordial meteorites like Allende. These CAIs contain trace amounts of the Al-26 radionuclide. This supernova seeding enriched the solar nebula with heavy elements like aluminium, calcium, iron, and others, providing the raw ingredients for terrestrial planet formation and the future biochemical evolution of life. These elements play crucial roles in biological processes: iron in haemoglobin for oxygen transport, calcium in bone formation and cellular signalling, and phosphorus in ATP for energy transfer. The presence of these elements in the early solar system provided the chemical diversity necessary for the evolution of metabolic pathways, the development of cellular structures, and ultimately, the emergence of the vast biodiversity we observe on Earth today, from single cell organisms to complex multicellular organisms.

The trigger for the formation of planets

Evidence suggests this same supernova shock front may have directly instigated the gravitational collapse of the presolar nebula, allowing the accretion of planets like Earth to occur. CAI minerals show evidence of having formed under brief high temperatures, potentially from being cross-cut by the supernova shockwave. Astrophysical models indicate nebular shock waves can trigger dust concentrations that gravitationally collapse into dense planetary embryos on timescales of just 10,000 years. Thus, the seeding of heavy elements from supernovae and the supernova shock front that began planetary accretion appears intricately linked to the very formation of Earth-like rocky planets capable of harbouring life.

The contingency of the KT extinction event

Another pivotal contingency was the specific asteroid or comet impact that triggered the Cretaceous-Paleogene (K-Pg) mass extinction event around 66 million years ago. This cataclysmic impact near the current Yucatan peninsula sparked decades or centuries of global cooling, droughts, and a collapse of photosynthesis due to atmospheric aerosols and dust loading. Mathematical analysis suggests the Chicxulub impactor had to be within a fairly narrow range around 10 km wide to drive the K-Pg extinction without causing an even more severe global catastrophe.

Detailed modelling indicates impactors much larger than ~20 km could have instigated severe global cooling and reduced photosynthesis enough to wipe out even small survivors. Meanwhile, geochemical studies confirm the global distribution of impact materials as well as severe shocks to the climate and ecosystems, extinguishing the non-avian dinosaurs along with about 75% of all species.

This catastrophic event disrupted established ecosystems and food webs, allowing the previously unassuming ancestors of mammals, around 225 million years old to undergo a remarkable evolutionary radiation into vacated ecological niches left by the demise of the ruling dinosaurs and other reptilian megafauna. The dominance of large-brained mammals and primates in the evolutionary landscape in the next 30 million years only became possible after this contingency removed the long-entrenched competition of their diminutive ancestors of Mammalia.

The contingency of the upheaval of ancestral environments

But even after mammals gained a foothold, the direct path to human-like intelligence required a final pivotal contingency - the geological forces ripping apart the African continent and transforming the habitats harbouring humanity's hominid primate ancestors. This geological event would prove to be a powerful driver of natural selection and adaptive radiation in the primate lineage.

Around 30-35 million years ago, the Earth's tectonic motions sparked the gradual spreading of the East African Rift Valley system, slowly splitting apart the ancient landmass and creating highlands, volcanic landscapes, and ultimately drying out the lush forest environments where ape-like hominids lived. This dramatic shift in habitat acted as a strong selective pressure, favouring adaptations for a more terrestrial lifestyle. This environmental transformation led to significant changes in flora and fauna, altering food webs and creating new ecological niches.

Fossil evidence indicates this grassland transformation placed intense selective pressures on a branch of hominids around 6-8 million years ago, favouring bipedal locomotion and larger brain sizes. The shift to bipedalism brought about numerous anatomical changes. These adaptations allowed for more efficient locomotion in open environments and freed the hands for tool use. Concurrently, the expansion of brain size increased cognitive capabilities, social complexity, and eventually, the development of language.

The changing environment also influenced dietary adaptations. As forest fruits became scarcer, hominids evolved more versatile digestive systems capable of processing a wider range of foods, including tough grasses and meat. This dietary shift is evidenced by changes in tooth morphology. The ability to exploit diverse food sources, coupled with the development of stone tools, allowed early hominins to access new energy sources, potentially fueling further brain growth. The unique combination of geological and climatic factors in East Africa created a 'natural laboratory' for human evolution, driving the development of traits that would ultimately lead to *Homo sapiens*.

An intertwined cosmic lottery

Taken together, this concatenation of pivotal events—the seeding of a stellar chemical enrichment, the triggering of runaway accretion, an asteroid impact recalibrating biodiversity, and a linear geographic restructuring of a key species' environment—highlights the sheer improbability of the evolution of human intelligence.

As Ward and Brownlee argue, this interlinked chain seems exponentially implausible from a cosmic perspective, leading them to posit that intelligent life may be extremely rare in the Universe. They propose that the specific orchestration of contingencies like those on Earth required for complex intelligent observers appears staggeringly unlikely to be replicated elsewhere.

A persisting mystery

Some scientists contend Ward and Brownlee overstate the degree of improbability and underestimate the potential cosmic prevalence of Earth-like contingencies. Astrobiologists like David Grinspoon argue the supposed contingencies may be more frequent across billions of planetary opportunities than assumed. Others argue the "rare Earth" reasoning succumbs to observational selection effects - we cannot account for the myriad unseen pathways intelligence may have taken elsewhere, limiting our probability calculations.

While captivating, the degree to which this convergence of chemical, astrophysical, geological, and biological contingencies on Earth represents a truly improbable cosmic outlier remains an open question actively debated by scientists. Did humanity arise through an exponentially unlikely chain of events? Or are the steps along Earth's pathway to intelligence more likely to be replicated across the cosmos than currently assumed?

Whether our cosmic solitariness implies a genuinely improbable contingency or just the patience required for intelligence to emerge across the full spacetime immensity remains one of the greatest unanswered questions confronting humanity. For now, what is clear is that the path travelled toward observers like ourselves was neither straightforward nor obligatory, but an intricate cosmic choreography of contingent events whose varying cadences and sequences may be the key to revelations awaiting across the Universe.

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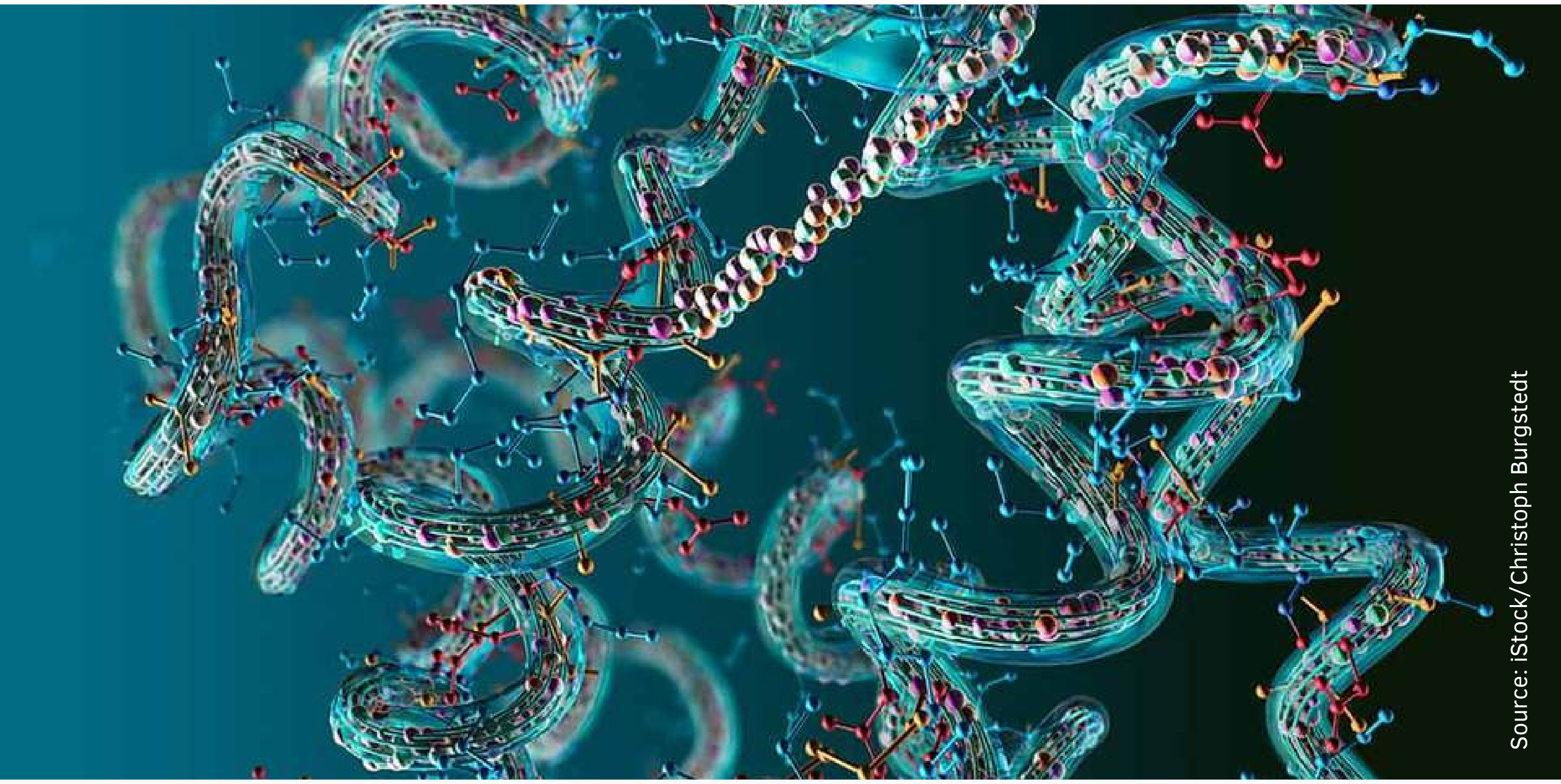

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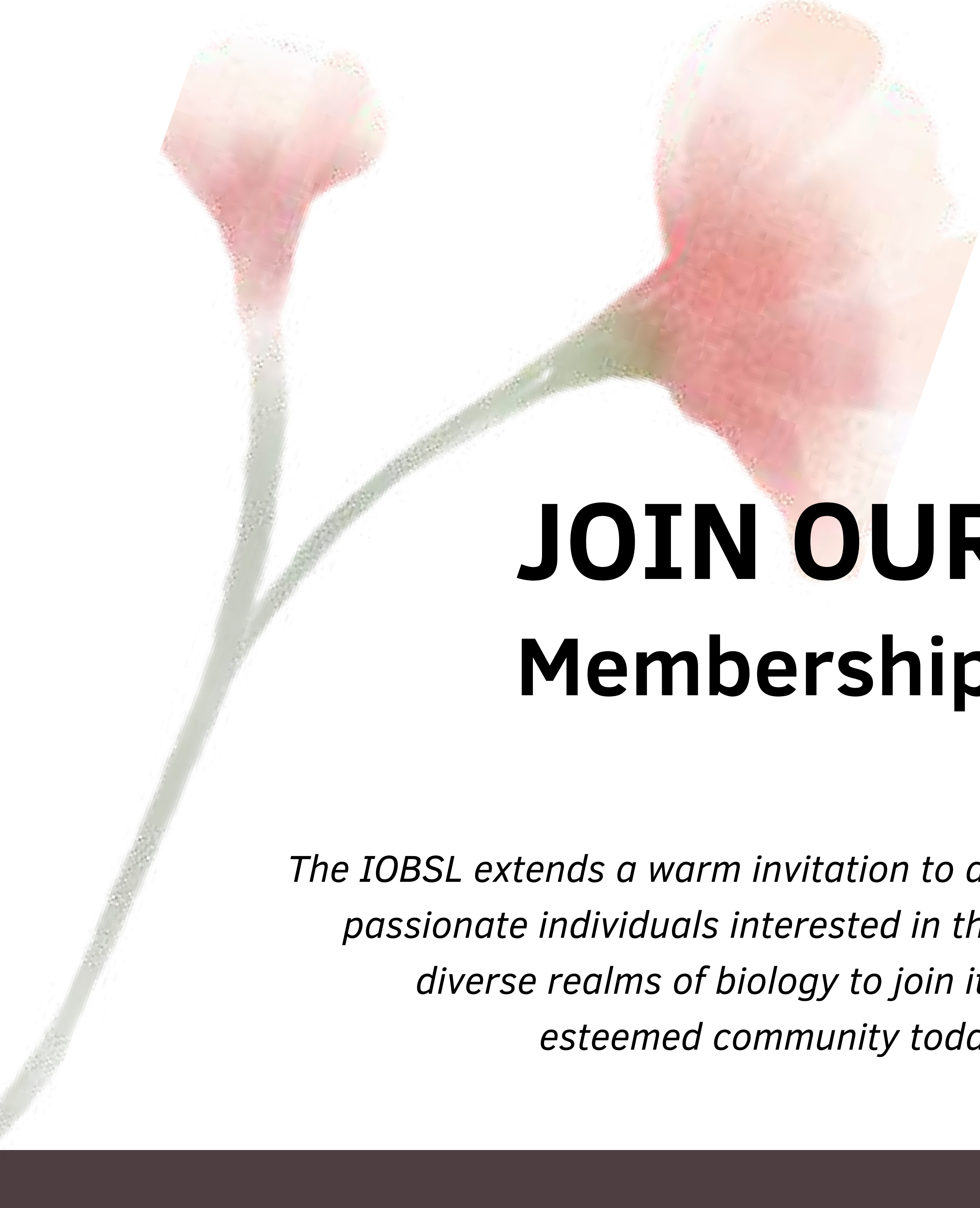

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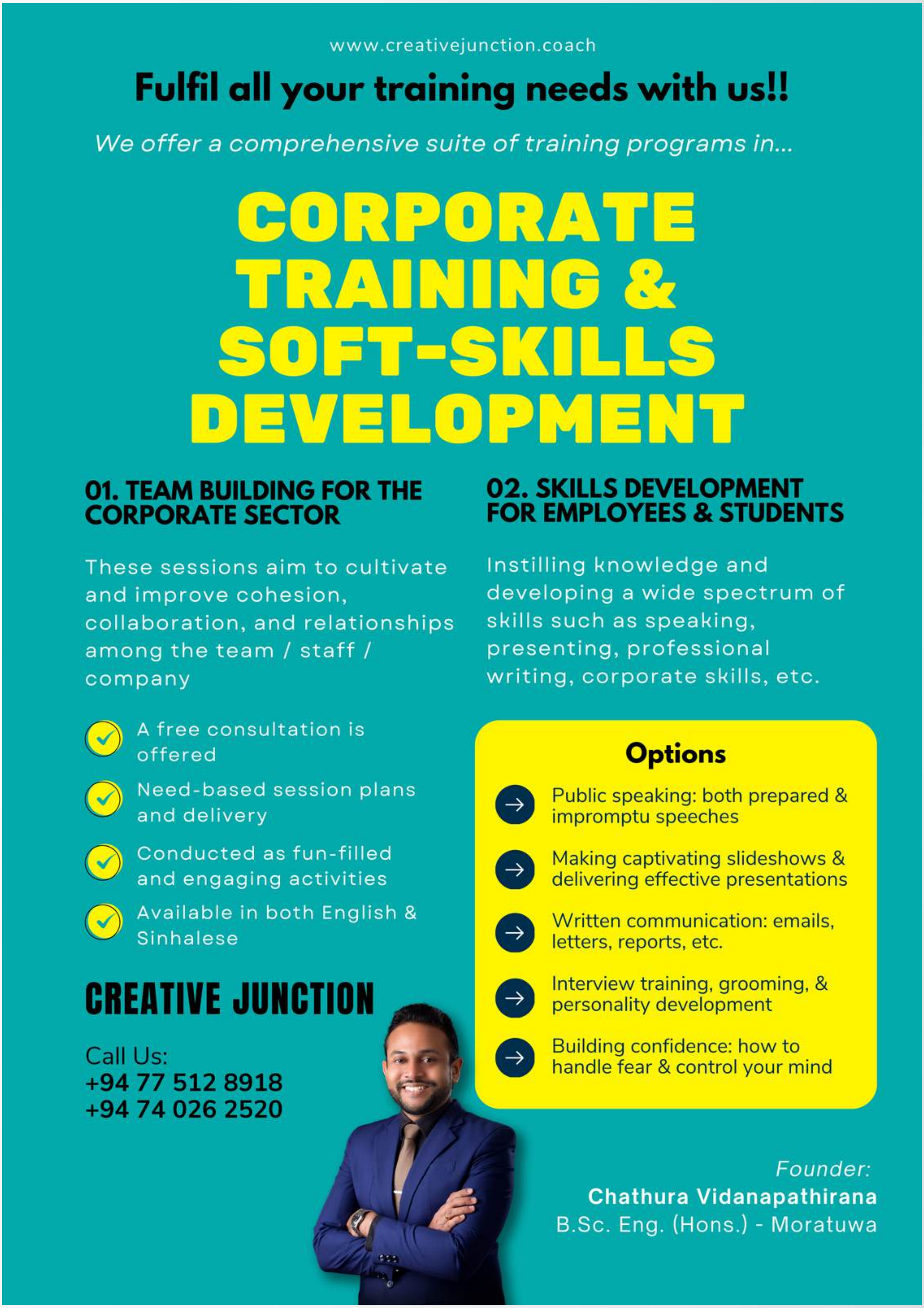

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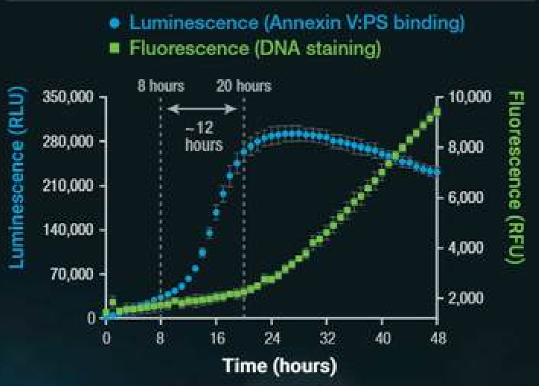

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UPDATES ON THE LATEST RESEARCH

Hornets of Sri Lanka: Beyond the Stings

A colonial past

Hornets are among the most widely distributed medically important insects in Sri Lanka. Wasps belonging to the genus *Vespa* of the family Vespidae (Order Hymenoptera) are known as hornets. The first-ever record of hornets in the scientific literature from Sri Lanka is by Walker in Tennent's Ceylon: An account of the island[2] in 1859. The description appears to be on lesser-banded hornets, though it is mentioned as *Sphex ferruginea*. In 1896, Bingham mentions two species, *V. cincta* Fabricius (synonym of *V. tropica*) and *V. obliterata* Smith (synonym of *V. basalis* Smith, 1852), in his publication "A contribution to the knowledge of the Hymenoptera fauna of Ceylon," based on the collections made by Col. Yerbury, R.A., and Mr. E.E. Green. In 1897, again, he mentions two hornet species, *V. basalis* Smith and *V. cincta* Fabricius (synonym of *V. tropica*), in the publication Fauna of British India Ceylon and Burma Vol, I. Wasps and Bees.

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However, in regional publications, there are four recorded hornet species from Sri Lanka: *V. basalis* Smith, 1852, *V. mandarinia* Smith, 1852, *V. affinis* (Linnaeus, 1764), and *V. tropica* (Linnaeus, 1758). Even though these four species are also included in the specimen registers of the Hymenopteran collection of the National Museum of Sri Lanka, the specimens of *V. basalis* and *V. mandarinia* are missing. Hence the validity of these identifications cannot be established. Further, Das and Gupta, 1989 commented that the literature records of the occurrence of *V. mandarinia* in Sri Lanka appear to be incorrect. A recent investigation carried out by the authors from July 2022 to November 2023, covering 30 locations in all the provinces in Sri Lanka and the citizen science (iNaturalist) and social media (Facebook) platforms resulted in finding the occurrence of only two species, *V. affinis* and *V. tropica*. Considering all these facts, the occurrence of the other two species, *V. basalis* and *V. mandarinia*, in Sri Lanka is doubtful.

Figure 1 Lesser-banded hornet (*Vespa affinis*)
Source: Hettiarachchi, T. (2020) [online]. Available at: https://www.inaturalist.org/photos/95406324 (Accessed: 25 July 2024)

The two valid species

Vespa affinis is commonly known as the Lesser-banded Hornet in English and "Heen Debara/Gu Debara/Debara" in Sinhala (Figure 1). The name Gu Debara, which is translated as "Dung hornet," comes from the belief that they use animal dung/fecal matter to build their nests. *Vespa affinis* is indigenous to eastern, southern, and southeastern Asia and was introduced to Australia, New Zealand, and the USA. In Sri Lanka, this wasp makes large open arboreal nests, which are hanging or fixed onto the surface (Figure 2). It is a very common species and prefers to nest in forest edges and human-altered habitats, due to the abundance of prey items. Looking at the distribution, *V. affinis* has been recorded from all the climatic zones of the country and more abundantly in higher elevations.

Figure 2 Nest of Lesser-banded hornet (*Vespa affinis*)
Source: Kennerley, P. (2008) [online]. Available at: https://www.inaturalist.org/photos/186733534 (Accessed: 25 July 2024)

Vespa tropica is known as the Greater-banded Hornet in English, “Bin Debara/Debara” (Figure 3). It is known to make nests underground or in secluded locations which provides its vernacular name, translating into “ground hornet”. This wasp is also indigenous to the same region as the lesser-banded hornet. It is a common species distributed mainly in the low country and appears to be more frequent in the wet zone. They tend to visit inside houses and buildings looking for nesting sites. *Vespa tropica* has been recorded from all the climatic zones of Sri Lanka.

These two species can be distinguished from each other mainly using size and color patterns. The Lesser-banded hornet is usually smaller in size, and its mesosoma (thorax) is entirely reddish-brown dorsally with a few black markings. The second tergum (dorsal plate) of the metasoma (abdomen) is largely or entirely yellow. The color form recorded from Sri Lanka does not match with any of the subspecies from India, as the thorax is entirely reddish-brown dorsally.

In comparison, the Greater-banded hornet is larger, and the mesosoma is prominently black, except for the mesoscutellum which is reddish-brown. The first tergum of the metasoma is also entirely black, sometimes with a narrow posterior yellow band. The second tergum of the metasoma is orange-yellow with a dark brown basal band.

More than just a sting

Hornets are eusocial wasps, and they occur in colonies where fending off predators and foraging is more successful. This contributes to their survival in large numbers when compared with solitary species. They show progressive provisioning of the young and parental care.

The public perception of hornets is generally placed negatively mainly due to their ability to sting. A hornet can inflict multiple stings because, unlike bees, its stinger has no barbs and does not get detached from the body after stinging. Very often wasp stings are ignored or treated with home remedies such as the application of red onion bulbs (*Allium cepa*) and ripe areca nut (*Areca catechu*) husks in sting sites. Hymenopteran stings are a common public health hazard in Sri Lanka. The hornet stings have been reported to cause ailments ranging from localized painful swellings, skin lesions to acute pulmonary edema, acute renal failure, severe multi-organ, severe anaphylaxis, and death. Therefore, if a nest is noticed, people tend to either drive away the colony using natural repellents such as Kaffir Lime (Kudalu dehi, *Citrus hystrix*) and smoke or destroy the nests using kerosene, petrol, and fire at nighttime or sometimes using commercial insecticides such as “Mortein” spray.

Figure 3 Greater-banded hornet (*Vespa tropica*)
Source: Gallege A. (2020) [online]. Available at: https://www.inaturalist.org/photos/92822494 (Accessed: 25 July 2024)

Despite their fearsome reputation, the hornets play an essential ecological role in the environment, contributing to many primary ecosystem services. They act as critical predatory insects in regulating arthropod populations in natural ecosystems. They often prey on other hymenopteran species such as bees. Hornets scavenge over proteins on dead animals such as birds, rodents, and lizards, suggesting a role as scavengers. They are known to be generalist pollinators of plants. While they are not as efficient as bees or butterflies, they can act as backup pollinators when the primary pollinators of the plants are not present.

As ongoing research refines our understanding of them, the public's awareness of their ecological importance should be improved. Balancing the need for human safety with an appreciation for these insects' ecological contributions will foster coexistence and informed conservation efforts, ensuring the sustainability of hornet populations and the ecosystems they inhabit.

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Grain Level Identification of Widely Cultivated Sri Lankan Rice Varieties

Rice is a principal cereal crop worldwide, distributed on all continents except Antarctica. It serves as the staple food for over half of the world’s population including Sri Lanka and is reported to provide significant quantities of essential nutrients and bio-active phytochemicals for rice consumers. Rice belongs to the family Gramineae/Poaceae and is considered a semiaquatic annual grass plant. It comprises around 22 species under the genus *Oryza* of which only two species are mainly cultivated throughout the world and the rest are wild rice. The two mainly cultivated species of rice are *Oryza sativa* L., which grows in most parts of the world and *Oryza glaberrima* Steud., which is grown in Africa. Generally, rice can be grown in a broad range of soil-water systems and is highly adaptable to its growing environment. Due to the long history of cultivation along with both natural and artificial selection, rice has evolved into a highly morphologically, physiologically and genetically diverse crop with about 120,000 distinct rice varieties existing throughout the globe. Between the two main cultivated rice species, the *O. sativa* which is also known as Asian rice, exhibits exceptional varietal diversity compared to the *O. glaberrima* (African rice). Further, high-yielding, nutritionally and functionally superior new hybrid rice varieties are continuously being introduced for cultivation to feed the growing population. Rice varieties also differ in endosperm traits, categorized as glutinous and non-glutinous, with the majority being non-glutinous. Additionally, rice varieties could be categorized based on grain size as short, medium or long grain, and based on hull/pericarp colour as pigmented or non-pigmented.

Rice varietal identification

Consumer preference and the commercial value of rice are increasingly diversifying on grain quality characteristics such as physicochemical, nutritional, functional and sensory properties, which are also varietal dependent. Further, the identification of rice varieties at the grain level is highly important to maintain their genetic purity. Therefore, identification of the exact rice variety at the grain level has become a key interest among plant breeders, seed certifying authorities and rice growers.

Rice varietal identification techniques broadly fall into morphological, molecular, chemical and physical methods. Conventionally, rice varieties have been identified at the plant level using plant morphological characters which may be altered by environmental factors and do not facilitate identification at the grain level. Moreover, the morphological methods are tedious, time-consuming and require more area to carry out field grow-out tests. On the other hand, molecular methods are more accurate and specific but need substantial investment and thorough technical knowledge and skills. In contrast, physical and chemical test methods are relatively fast, easy to conduct, reproducible and inexpensive. Internationally, physical and chemical test methods have been used by many researchers for varietal identification of various crops, including rice to a certain extent, at the grain level.

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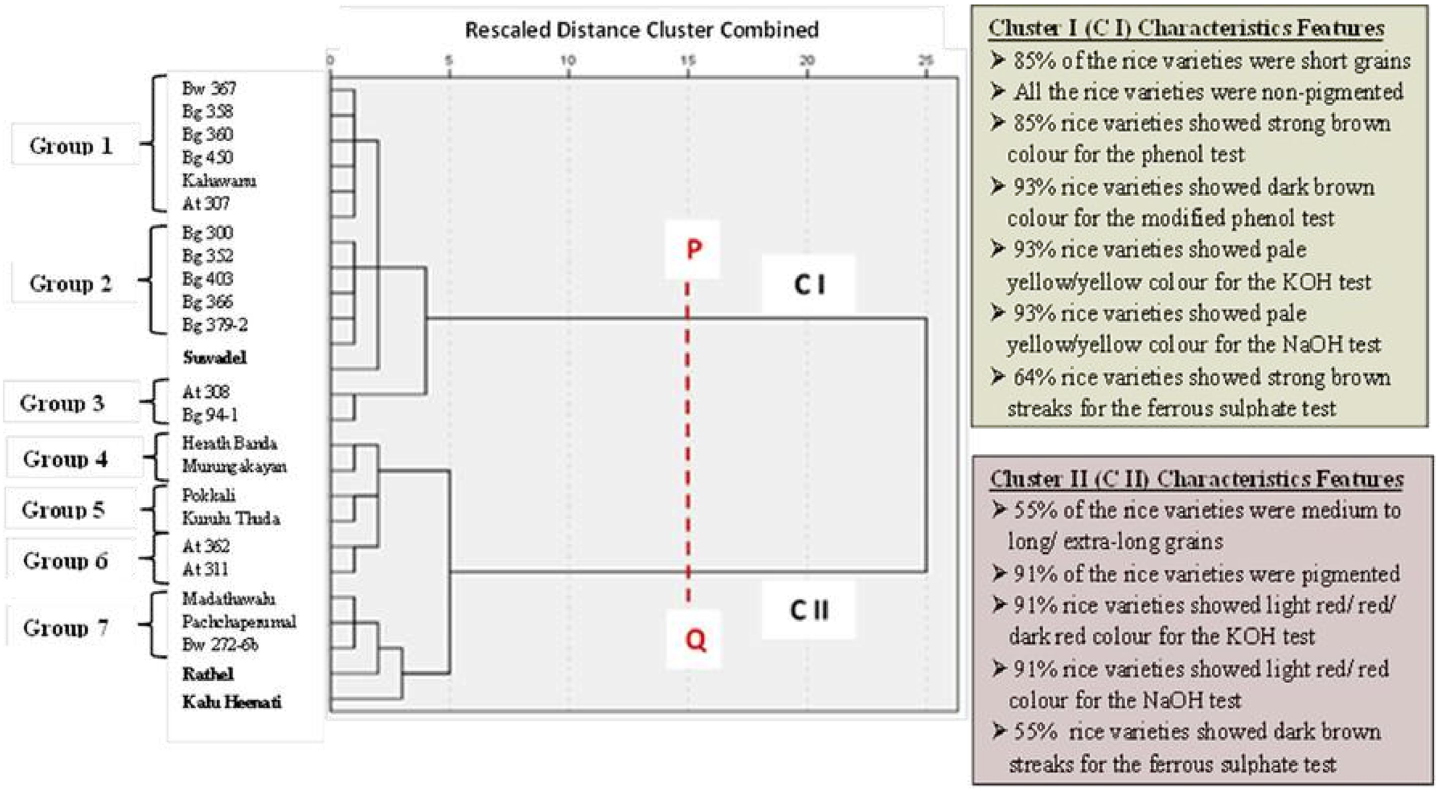
Diversity of rice in Sri Lanka

In Sri Lanka, rice is the dietary staple and two of the three daily meals of the majority of Sri Lanka's population depend on rice and curry. The annual per capita consumption of rice in the country is approximately 107 kg. Rice cultivation in Sri Lanka is believed to date back to 540 BC. The country holds more than 1000 traditional rice varieties and new improved high-yielding rice varieties are also being introduced by the Department of Agriculture to fulfill the increasing demand of rice consumers. Additionally, the research carried out during the last 10-12 years in the country has been able to prove that nutritional, physicochemical and functional properties of rice varieties vary among the rice varieties and some of the Sri Lankan rice varieties have greater nutrient densities, desirable physicochemical and functional properties. However, the identification and differentiation of traditional and new improved rice varieties of Sri Lanka, especially at the grain level is a great challenge due to their high diversity. This article highlights the research findings of an attempt on the identification of commercially important and widely cultivated traditional and new improved rice varieties (*O. sativa* L.) in Sri Lanka at the grain level.

An attempt to identify Sri Lankan rice varieties at grain level

Ten traditional rice varieties (namely Rathel, Suwadel, Madathawalu, Pachchaperumal, Herath Banda, Kalu Heenati, Kahawanu, Pokkali, Kurulu Thuda, and Murungakayan) and fifteen new improved rice varieties (namely At 307, At 308, At 311, At 362, Bg 300, Bg 352, Bg 358, Bg 360, Bg 366, Bg 379-2, Bg 403, Bg 450, Bg 94-1, Bw 272-6b and Bw 367) were grown and harvested under experimental field conditions at the Rice Research and Development Institute (RRDI), Batalagoda, Sri Lanka. They were characterized using a range of internationally accepted physical and chemical test methods. Physical tests included the study of grain length and size, while chemical tests involved ferrous sulphate, phenol, modified phenol, NaOH and KOH tests on the paddy of selected rice varieties.

According to the results, the studied rice varieties were grouped into four categories based on grain length and size: extra-long (≥ 9.9 mm), long (9.8-8.8 mm), medium (8.7-8.0 mm) and short (< 8.0 mm) grain rice varieties. Additionally, based on the colour responses from ferrous sulfate, phenol, modified phenol, NaOH and KOH tests, rice varieties grouped into four (no colour change, brown, strong brown and dark brown streaks), five (no colour change, strong brown, dark brown, reddish yellow and black), five (no colour change, strong brown, dark brown, dark reddish brown and black), five (pale yellow, yellow, olive yellow, light red and red) and six (pale yellow, yellow, olive yellow, light red, red and dark red) groups, respectively. None of the chemical or physical test methods alone used in the study could be used to identify the studied Sri Lankan rice varieties at the grain level. However, combining both methods enabled the clustering of selected rice varieties into two main clusters (CI & CII) and identified three rice varieties (Rathel, Suwadel and Kalu Heenati) at the grain level using hierarchical cluster analysis (Figure 1). Further, the other selected rice varieties were categorized into seven groups based on similar characteristics (Figure 1). **Overall, the selected physical and chemical test methods need to be coupled with other varietal identification methods to characterize the selected Sri Lankan rice varieties at the grain level.**



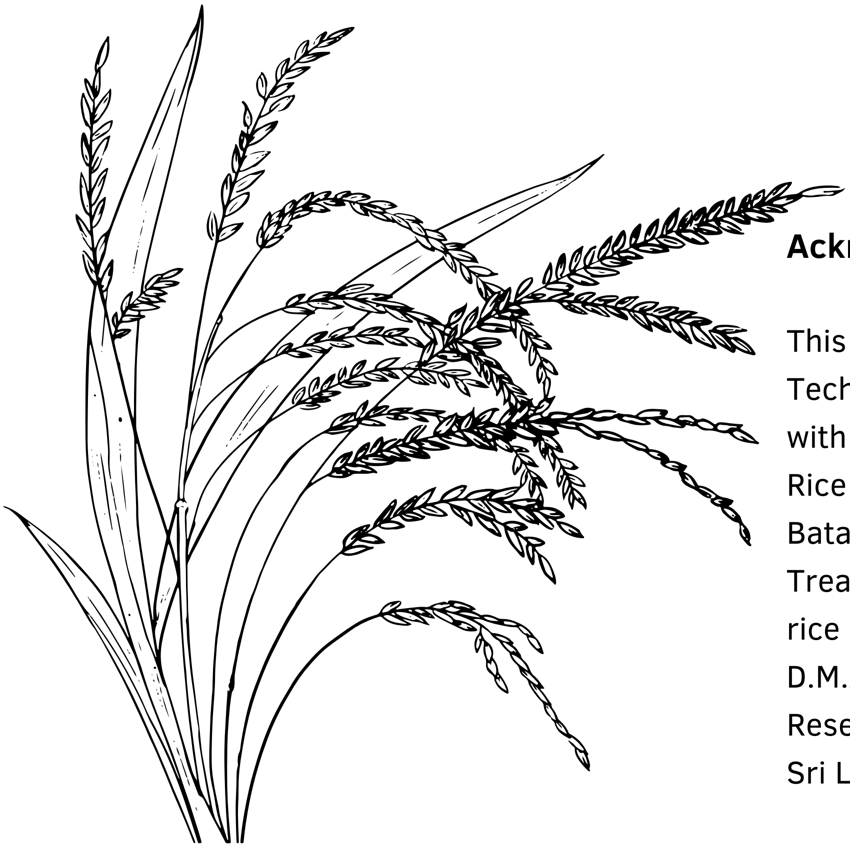
Cluster I (C I) Characteristics Features

- 85% of the rice varieties were short grains
- All the rice varieties were non-pigmented
- 85% rice varieties showed strong brown colour for the phenol test
- 93% rice varieties showed dark brown colour for the modified phenol test
- 93% rice varieties showed pale yellow/yellow colour for the KOH test
- 93% rice varieties showed pale yellow/yellow colour for the NaOH test
- 64% rice varieties showed strong brown streaks for the ferrous sulphate test

Cluster II (C II) Characteristics Features

- 55% of the rice varieties were medium to long/ extra-long grains
- 91% of the rice varieties were pigmented
- 91% rice varieties showed light red/ red/ dark red colour for the KOH test
- 91% rice varieties showed light red/ red colour for the NaOH test
- 55% rice varieties showed dark brown streaks for the ferrous sulphate test

Figure 1 Dendrogram for tested physical and chemical properties of selected rice varieties of Sri Lanka using hierarchical cluster analysis



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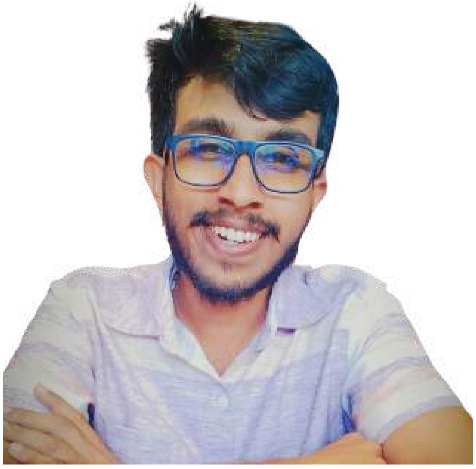
CORNER FOR YOUNG BIOLOGISTS

Light of Neuroscience: Optogenetics

The human brain is an outstanding component of the human body, comprising approximately 100 billion neurons that function both chemically and electrically. While these neurons are nearly similar to one another, they can be grouped into thousands of types based on their size, and the length of their axons and dendrites. Consequently, these neurons form networks by connecting with each other, creating a complex neuronal network. These networks controlhumans' thoughts, emotions, memories, and desires. Disorders such as Parkinson's and depression can be caused by dysfunction within these networks. Optogenetics is a newly emerged field that provides a novel approach for understanding and controlling the animal brain, while also offering potential treatments for certain brain diseases. It is a science that involves collaboration between neuroscience, genetic engineering, and optics. The term ‘optogenetics’ was coined by Karl Deisseroth and his team at Stanford University when they published a groundbreaking paper in 2005. They used a mouse model to control the activity of specific brain regions, making a significant advancement in neuroscience research.

What is optogenetics?

"*Optogenetics is an elegant approach of precisely controlling and monitoring the biological functions of a cell, group of cells, tissues, or organs with high temporaland spatial resolution by using opticalsystem and genetic engineering technologies.*" In other words, protein molecules that can convert light into electricity are introduced into cells, tissues, or organs. In neuroscience, specific proteins are introduced into brain neuron cells. This process can be divided into three main steps: finding a light-sensitive protein, introducing the opsins to the cell, and lighting.



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Finding a light-sensitive protein

Normal neurons are not sensitive to light. Therefore, the first step of this process is introducing a photosensitive protein or a biomolecule to neurons. These proteins can sense light and open specific protein channels (Na^+ channels, Cl^- channels), causing changes in the cell membrane's polarization. This change in polarization, known as the membrane potential, occurs between the outside and inside of the membrane (Figure 1). Once a certain amount of voltage difference, known as the threshold value, is reached, the neuron is stimulated and can transport this electrical charge. There are two types of photosensitive proteins: those that stimulate membrane potential and those that block the stimulation of the membrane potential.

Light-activated proteins known as opsins are used in optogenetics. These proteins can be divided into two types based on their primary sequence and mode of action: type I opsins and type II opsins. Type I opsins are found in archaea, eubacteria, fungi, and algae, whereas type II opsins are found in higher animals. Type I opsins typically encode ion channels, facilitating the flow of ions across the cell membrane upon exposure to light, while type II opsins act via G protein-coupled receptor pathways. Type II opsins are slower and mostly used in dim light vision and circadian clock regulation, compared to type I opsins.

Some of the commonly used microbial opsins in optogenetic experiments include channelrhodopsin (ChR), rhodopsin, panopsin, and melanopsin. ChR isolated from the algae *Chlamydomonas* sp., serves as both a protein and a light-gated channel, primarily localized in the eye spot of *Chlamydomonas*. Responsive to blue light, ChR possesses the ability to depolarize the neuronal membrane. This family of proteins includes various types, such as ChR1 and ChR2. ChR1 is stimulated by a wavelength of λ_{max} = 500 nm, while ChR2 responds to a wavelength of λ_{max} = 470 nm. Both these ChRs primarily function as H^+ ion channels. Additionally, there may be channels that respond to other ions such as Na^+ and

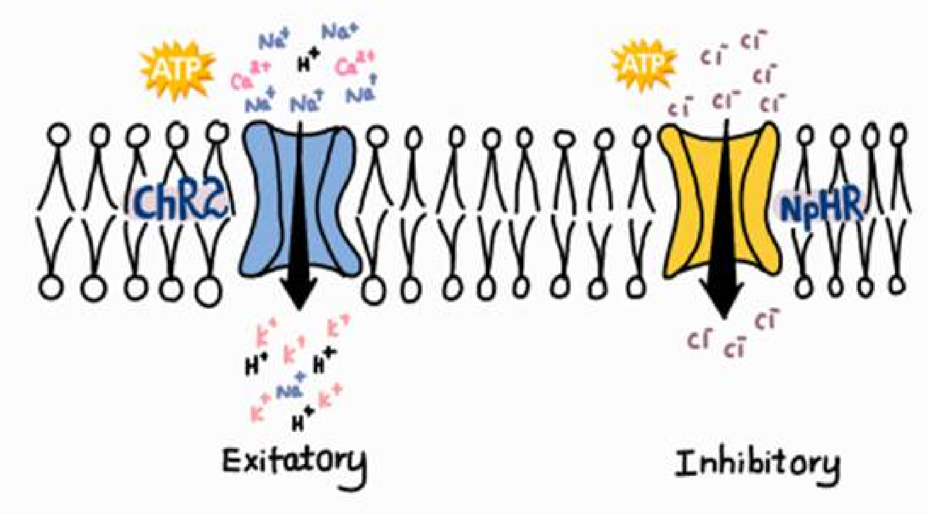


Figure 1 Function of NpHR and ChR2 pump. ChR2: channelrhodopsin2, NpHR: halorhodopsin.

Ca^{2+}. To block membrane potential, there is a channel protein known as halorhodopsin (NpHR), which is isolated from a chloridepump derived from the halobacterium *Natronomonas pharaonis*. NpHR actively pumps Cl^- ions into cells in response to yellow light. Additionally, archaerhodopsin-3 (Arch), derived from *Halorubrum sodomense*, functions by actively pumping hydrogen ions outside of the cell in response to yellow light. This yellow light typically falls within the range of nearly 580 nm wavelength. Furthermore, there are multiple optogenetics sensors to assist in monitoring changes in electrical and biochemical parameters and increase the opsins activity. These sensors can generate action potentials in response to light stimuli.

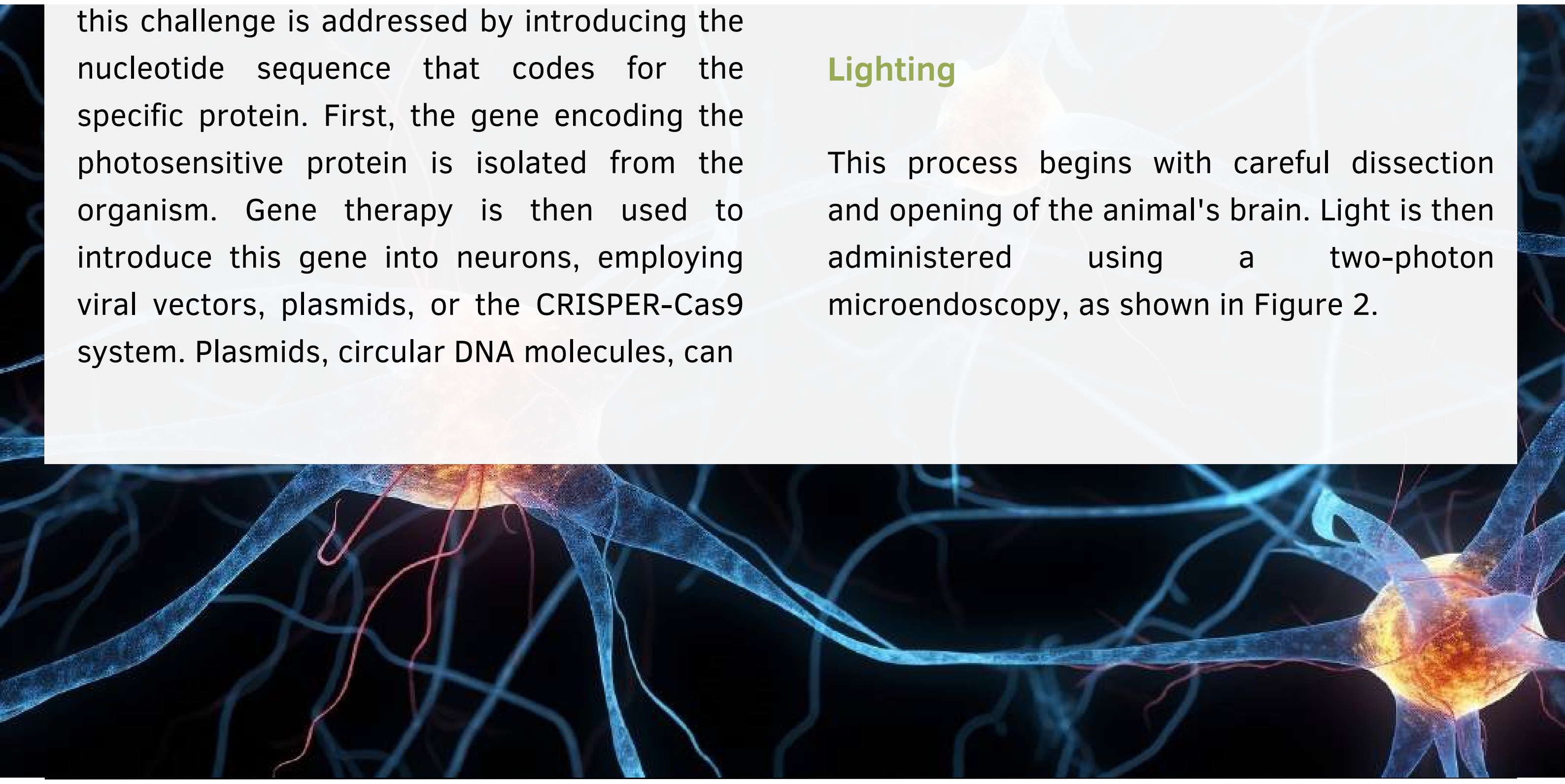
Introducing the opsins to the cell

This involves introducing the protein into neurons, which is where genetic engineering comes into play. Directly introducing the protein into neurons is not practical, therefore, this challenge is addressed by introducing the nucleotide sequence that codes for the specific protein. First, the gene encoding the photosensitive protein is isolated from the organism. Gene therapy is then used to introduce this gene into neurons, employing viral vectors, plasmids, or the CRISPER-Cas9 system. Plasmids, circular DNA molecules, can be introduced into targeted cells using techniques such as electroporation. The CRISPER-Cas9 technique involves cleaving palindromic sequences using the Cas9 restriction enzyme. Viral vectors, such as adeno-associated viruses (AAVs), are commonly used in optogenetic applications due to their efficiency in delivering genetic material into neurons.

Once the gene is introduced into the cell, its expression is regulated by promoters and transcription factors. Moreover, there are genetically targeted reporters which can confirm this expression and transformation. However, this process presents challenges, particularly because the genes being introduced originate from organisms such as algae, bacteria, and archaea. This introduces a high level of immunogenicity for mammals due to the phylogenetic distance between mammals and these organisms. Given the risks involved, such experiments are typically limited to animal models.

Lighting

This process begins with careful dissection and opening of the animal's brain. Light is then administered using a two-photon microendoscopy, as shown in Figure 2.



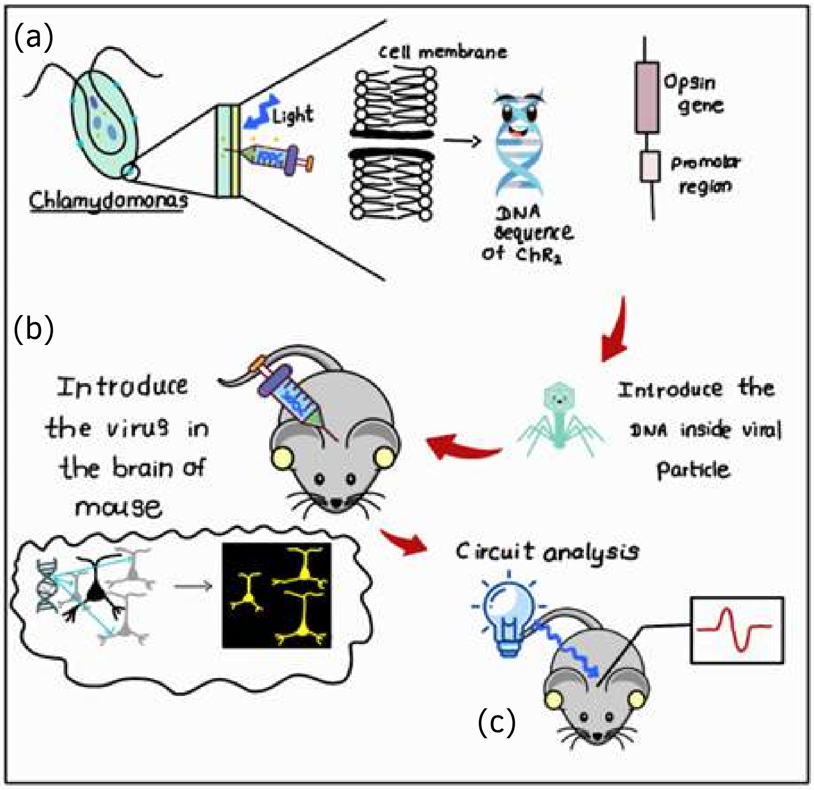


Figure 2 An optogenetic experiment. (a) Isolating specific gene from *Chlamydomonas* sp., (b) Gene engineered into Rat's specific neuron, (c) Lighting the rat's neuron and observed the changes by electric pulses.

Uses of optogenetics

As mentioned earlier, optogenetics integrates optics and genetics to control and monitor the activity of individual neurons in living tissues. This method involves genetically modifying neurons to express opsins, which are then activated or inhibited using light. This emerging field of science holds promise in uncovering neuroscience-based phenomena such as memory, addiction, and sensory perception. Currently, there are only a few examples of experiments in this field, with the majority being ongoing research.

- Memory research: Researchers have used optogenetics to investigate how neurons are involved in the formation and retrieval of memories. By selectively activating or inhibiting neurons, they have sought to understand the underlying mechanisms of learning and memory.

- Addiction studies: Optogenetics has been applied to understand addiction-related behaviors. Through the manipulation of activity within specific brain regions or neurons associated with reward and motivation, researchers can study the neural basis of addiction and potentially develop treatments for addiction.

- Pain sensitivity: Optogenetics has been used to elucidate how specific neurons in pain sensation pathways influence the sensitivity of pain. Insights gained through such studies can help in developing new treatments for chronic pain, especially in surgeries.

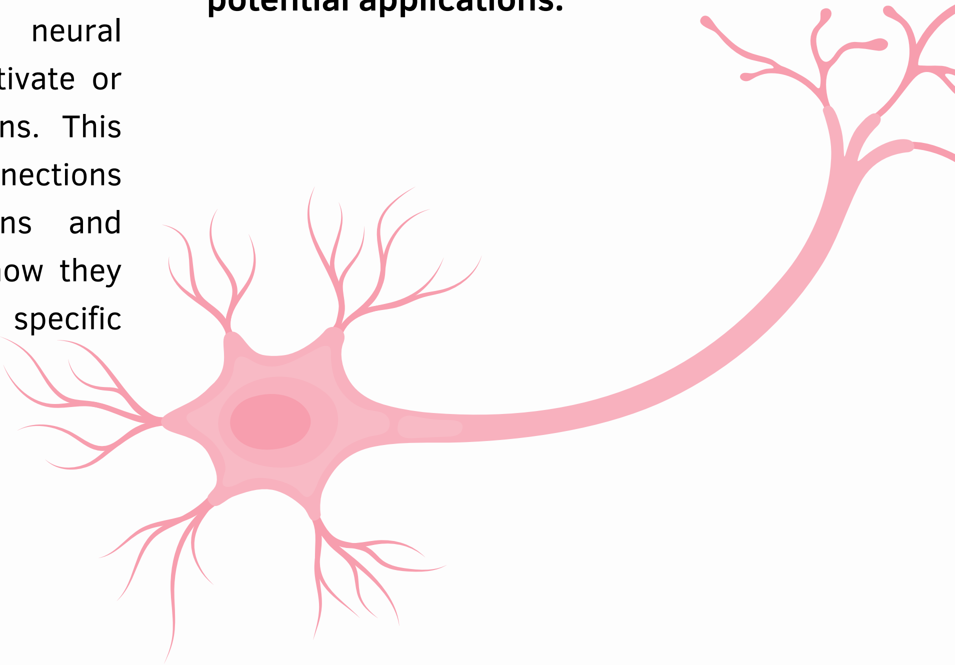
- Motor control: Investigations using optogenetics have paved the way for a deeper understanding of motor control and movement disorders. By controlling the activity of motor neurons, researchers study how the brain regulates movements and explore potential therapeutic applications. Moreover, these experiments offer insights into controlling artificial body parts such as legs and hands via brain signals, thereby aiding individuals with disabilities.

- Sleeping cycle research: Optogenetics has also been used to investigate the neural mechanisms underlying sleep-wake cycles and circadian rhythms. Researchers utilize this technique to manipulate specific neurons.

- Depression and anxiety: Studies have used optogenetics to examine the role of specific brain regions and neurons in mood disorders, such as depression and anxiety. Such investigations not only deepen our understanding of the underlying neurobiology but also identify potential targets for therapeutic interventions aimed at alleviating symptoms and improving the lives of individuals affected by these conditions.

- Neural circuit mapping: Optogenetics proves valuable for mapping neural circuits, allowing scientists to activate or inhibit specific groups of neurons. This enables the tracing of connections between different brain regions and facilitates the understanding of how they work together to produce specific behaviors.

These experiments have significantly advanced our understanding of the brain and hold promise for developing new treatments for various neurological disorders. However, all these experiments are conducted as either *in vitro* or *in vivo* studies; no experiments have been conducted on humans due to ethical and safety concerns surrounding gene engineering and therapy integral to optogenetics. Furthermore, the use of type I opsins can potentially cause immune responses in the host organism. Despite these limitations, the field of optogenetics presents a promising future. As technology and knowledge progress, researchers continue to improve techniques and address challenges, opening new avenues for discoveries and potential applications.



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NATURE CAPTURES

The Versatile Blue Beauty (Plumbago auriculata)

Plumbago auriculata, native to Mozambique and South Africa, is a captivating scrambling shrub that thrives in subtropical biomes. Known for its enchanting sky-blue flowers, this plant is more than just a visual delight. This is an introduced species to Sri Lanka, it serves various roles, including as animal food, a natural poison, and a traditional medicine. Additionally, it has significant environmental and social uses, contributing to biodiversity and cultural practices. *Plumbago auriculata*'s versatility and beauty make it a cherished addition to gardens and natural landscapes worldwide.

The Wishbone Flower (Torenia fournieri)

Torenia fournieri, known as the Wishbone flower, is a vibrant annual native to regions from India to Southern China, Indo-China, and Taiwan. Thriving in wet tropical biomes, this enchanting plant is celebrated for its diverse, colorful blooms. Introduced to Sri Lanka, it adds a splash of color to gardens and landscapes. Beyond its ornamental appeal, the wishbone flower holds environmental and medicinal significance, making it a cherished addition to tropical flora. Its resilience and adaptability symbolize the beauty and utility of nature's botanical treasures.

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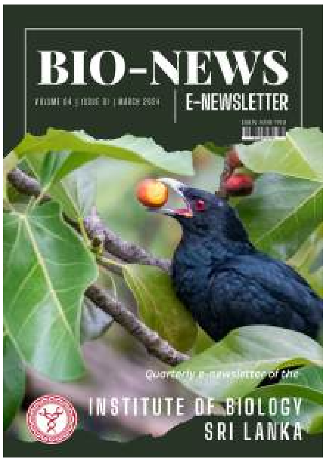

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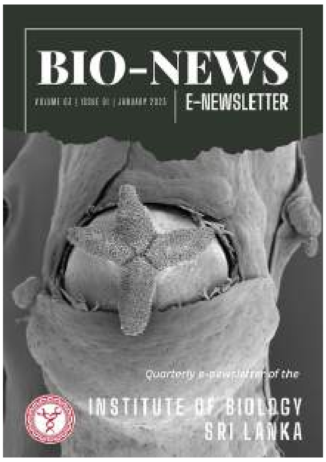

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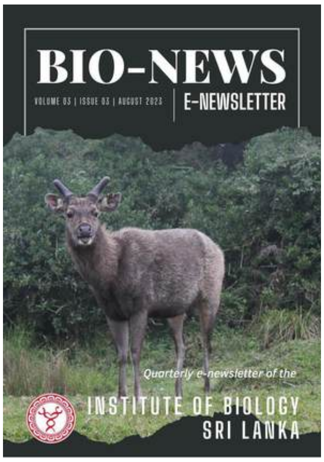

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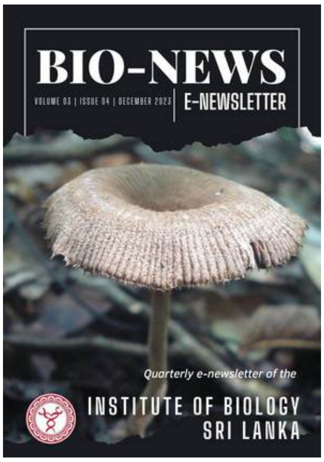

Issue 4

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