Institute of Biology, Sri Lanka

MARCHING TOWARDS A BIOECONOMY

Marching Towards a Bioeconomy

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Preface

The year 2021 marks the 41st Anniversary of the Institute of Biology of Sri Lanka (IOBSL). It is the main forum which represents the biology professionals from academia, research, and industry in Sri Lanka. Incorporated by the Act of Parliament No. 22 in 1984, the IOBSL is mandated to promote and advance the science of biology and its applications. By upholding the mandate vested upon IOBSL, it contributes to the advancement of the biology professions and welfare of the nation.

The global grand challenges have directed the world a technology shift from synthetic material base to biomaterials and processes. The biologists are at the core of this endeavour as they have to find out how the biological sectors can contribute to solve the economic, social and environmental problems. It is the biology professionals who have the firsthand knowledge on plants, animals, microorganisms, their biological processes, and the systems to use them sustainably to develop competitive bio-based industries through research and innovations. It is also the biologists who can support the development of educated and competent human resources required for an emerging bioeconomy.

The Institute of Biology has performed and will continue to perform its role even better in the future in promoting research, innovations, education, training, and public awareness in the quest to utilize the biological resources sustainably. The publication of this book on the theme of: Marching towards a bioeconomy, intends to inspire especially the young biologists on the concept of the bioeconomy and research and innovations required for sustainable use of biological resources and processes. This publication brings together research contributions by some of the experts on biological sciences which will lead to a knowledge-based bioeconomy, focusing on work done on herbal medicine, microbiota of mosquito vectors, biological control of the dengue vectors, potential crops for sustainable development and ecological concerns on hydro power generation. For all of us a stronger bioeconomy is still far from being a reality. We believe that this publication though not exhaustive would create a discussion and provide an encouragement for all those who work in the biological sectors in the country.

Our thanks go to the contributors of the chapters in this book, the reviewers and the members of the council for their great support in producing this book.

Dr S.A.C.N. Perera
Professor L.D. Amarasinghe
September 2021

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Chapter

01

Bioeconomy:
A Response to Global Grand Challenges

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Abstract

Bioeconomy has evolved as a solution to economic, social and environmental problems within and across all the sectors of world economies. It has brought about the materials technology shift from synthetics to bio-based raw materials to introduce sustainable technologies. Activities in the bioeconomy primarily involve the production of biomass from plants, animals, microorganisms and its conversion into bio-based products. Agriculture is a major component in a bioeconomy. Countries with a larger land mass have a competitive advantage in biomass production through agriculture hence for a larger bioeconomy. Sri Lanka, with its limited land availability cannot expand the land further for biomass production through traditional agriculture. The bioeconomy of Sri Lanka therefore have to move beyond traditional agriculture pursuing more scientific approaches for biomass production and conversion. The use of crops as renewable industrial feedstock and the application of biotechnology are therefore, indispensable for implementation of sustainable development strategies in the country. The rich biodiversity on land and the sea of the country is still underutilized and has a great potential to be used in pharmaceutical and biotechnology industries. There is a pressing need in the country to create a motivated society to pursue the promising prospects offered by the bioeconomy in development.

Keywords: Bioeconomy, Biomass, Biotechnology, Sustainability

Introduction

Figuring out, how to address the urgent challenges posed by population growth and environment pollution is important. There are fundamental problems in different facets of our lives such as health, environment, food, water and energy: and there is still uncertainty about how these challenges can best be addressed. Despite this uncertainty, the concept of bioeconomy has evolved across the globe in response to these urgent challenges and forms an integral part of the solution. It is the solution that nations have put forward to the Global Grand Challenges which underpins sustainable development. Bioeconomy focuses on the development strategies which minimize environmental damage while promoting recycling of resources. It is the shift of economies from petroleum-based synthetics to biomaterial-based products and processes and holds a great promise in securing human needs with minimum environmental damage. This sector of the economy therefore, encompasses all measures to expedite the global green transition by promoting the use of clean energy and greener industries, eliminating pollution by maximizing the use of residual streams from agriculture and food processing, emission savings and enhancing biodiversity through improving the ecosystem services. However, transition towards a bioeconomy is a challenging task especially for developing nations as it constitutes mainly an innovation driven economy. The policy environment and the technological capabilities of many developing countries are not adequate to facilitate the sustainable and eco-efficient use of the biological resources. The recent trend however, to embed the bioeconomy concept into a broader concept on sustainable development and green economy is a promising sign for future expansion of the bioeconomy globally. This way the bioeconomy concept will contribute to achieve the sustainable development goals (SDGs) so that we leave a healthy and joyful life for our future generations.

Bioeconomy

Bioeconomy relies on the renewable resources and technological advancements. It involves the production and sustainable utilization of biological resources, to provide goods and services across all economic sectors. The European Commission defines the bioeconomy as "the production of renewable biological resources and the conversion of these resources and waste streams into value added products, such as food, feed, bio-based products and bioenergy (European Commission, 2017). When developing the concept of the bioeconomy in the European Union, the label “knowledge-based” was added so that it became the “knowledge-based bioeconomy”. The knowledge-based bioeconomy emphasizes the scientific findings in life sciences, biotechnology and other related technologies to be the major driving force for sustainable management, production and use of biological resources. Thus, the knowledge-based bioeconomy was defined as “Transforming life sciences knowledge into new, sustainable, eco-efficient and competitive products” (Janez POTOČNIK, 2005). Further deliberations on this topic in EU emphasised two dimensions of the bioeconomy. They are the:

1. biotechnology innovation perspective which recognized the role of biotechnology as “an important pillar of Europe's economy by 2030, indispensable to sustainable economic growth, employment, energy supply and to maintaining the standard of living”

2. resource substitution perspective which stressed the use of crops as “renewable industrial feedstock to produce biofuels, biopolymers and chemicals”

National Bioeconomy Blueprint in the United States described the bioeconomy as one based on the use of research and innovation in the biological sciences to create economic activity and public benefit. The U.S. bioeconomy was already in place at the time of giving this definition: new drugs and diagnostics for improved human health, high yielding food crops, emerging biofuels to reduce dependency on oil, and biobased chemical intermediates, to name just a few. This definition also reflects the two perspectives of the bioeconomy discussed above: the biotechnology innovation perspective and the resource substitution perspective. When considering these two perspectives the resource substitution perspective predominated in the first decade of the 21st century where increase in prices of fossil energy was expected to continue. In the subsequent decade the emphasis has shifted to biotechnology innovation perspective in order to make the economic use of the advancements of the biotechnology and life sciences. A study carried out by Bugge *et al.* (2016) to enhance the understanding of what the notion of bioeconomy means by exploring the origins, uptake, and contents of the term “bioeconomy” in the academic literature have uncovered an additional dimension in bioeconomy, i.e. the bioecology perspective. In academic literature therefore it identifies three visions of bioeconomy albeit that they are not completely distinct from each other. These three visions of bioeconomy are:

1) The bio-technology vision that emphasizes the importance of bio-technology research and application and commercialization of bio-technology in different sectors. This vision relies mainly on the potential applicability of science.

2) The bio-resource vision that focuses on the role of research, development, and demonstration (RD & D) related to biological raw materials in sectors such as agriculture, marine, forestry, and bioenergy, as well as on the establishment of new value chains. The bio-resource vision emphasizes the potentials in upgrading and conversion of the biological raw materials.

3) The bio-ecology vision that highlights the importance of ecological processes that optimize the use of energy and nutrients, promote biodiversity, and avoid monocultures and soil degradation. While the previous two visions are technology-focused and give a central role to RD & D in globalized systems, this vision emphasizes the potential for regionally concentrated circular and integrated processes and systems (Bugge *et al.,* 2016).

Components of the Bioeconomy

Bioeconomy is the part of the economy which uses renewable biological resources from terrestrial and aquatic origin such as crops, forests, algae, fish, animals and microorganisms to produce food, feed, materials and energy. It primarily involves the production of biomass on soil, water or in culture conditions such as microbial or tissue cultures and their conversion supplemented by the recycling of waste generated in the process. The activities of bioeconomy may extend beyond the primary production and conversion of the biomass. For example, bioeconomy related activities may extend to other sectors in the society such as education, skill development, policy making and to all processes in pre- and post- commercialization steps of bio-based products. Hence bioeconomy value chain constitutes with a multitude of components which comprises not only in terms of value added and employment contribution but also encompassing the socio-economic and environmental aspects of the economy. The European Commission (2017) classified the bioeconomy sectors as core bioeconomy, partial bioeconomy and indirect sectors. Agriculture, fishing and forestry, food industry, bioenergy and biofuel are among the core bioeconomic activities. Some activities included in the partial bioeconomy are chemical and plastic industry, construction, pharmaceutical, textile, waste management and biotechnology. The indirect sectors include technologies, machinery, retail trade and related services etc. which have an indirect impact. A generic pathway to monitoring and assessing of bioeconomy to a nation's economy as identified by the Food and Agricultural Organization of the United Nations is given in Figure 1 (FAO, 2018).

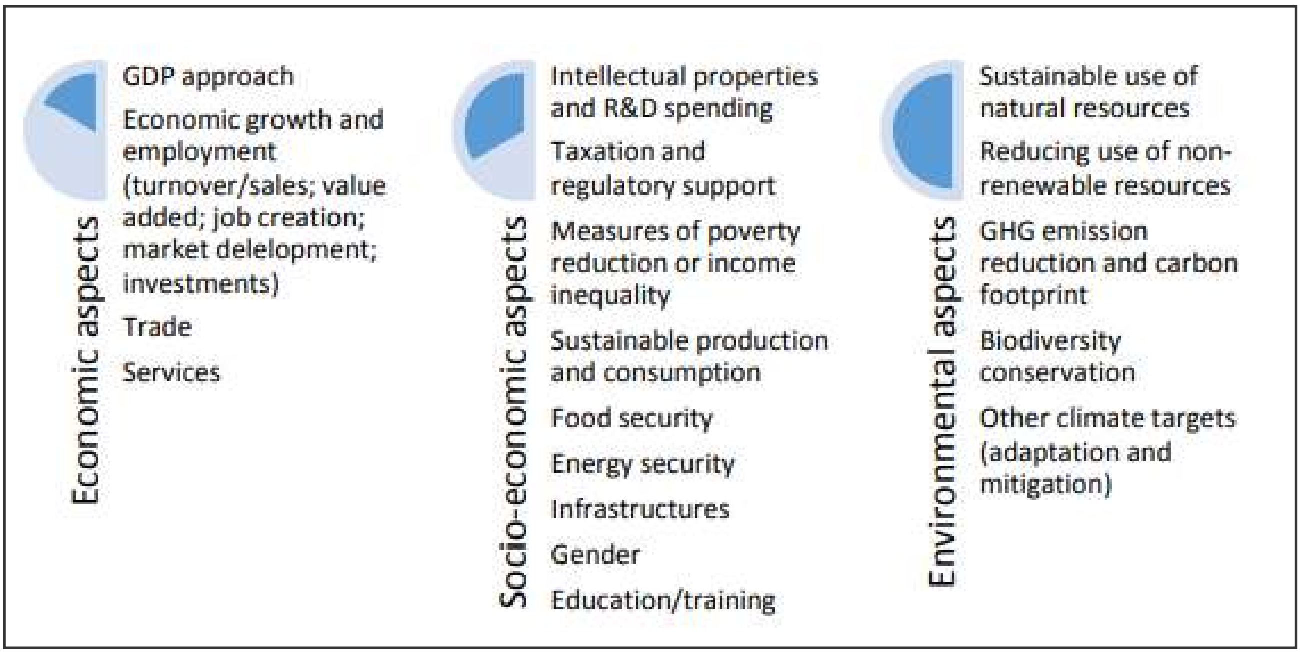

Figure 1: Identified pathway towards a sustainable bioeconomy monitoring (source: FAO, 2018).

The major component in bioeconomy common to all countries is the agricultural sector where it produces the largest part of the biomass used as a raw material for the bioeconomy through crop and animal husbandry, forestry and aquaculture. Countries having a large land mass, favourable agro-ecological conditions and a low population density, have the competitive advantage in biomass production by agricultural activities and to go for resource substitution perspective of the bioeconomy. Similarly, countries having access to marine resources utilize these resources as a main component in the fisheries sector of their bioeconomy. Biomass production in cultures such as cell and tissue cultures, microbial cultures also make a significant contribution to produce bio-based materials and these activities are mainly based on the progress of the scientific findings related to the biotechnology perspective of bioeconomy. The biotechnology perspective of bioeconomy requires comparatively a low land area and can be carried out independently of the external environmental conditions hence provide better opportunities for countries with limited land resources and high population densities. The bioeconomy of small countries like Sri Lanka therefore have to move beyond traditional agriculture pursuing more scientific approaches for biomass production and conversion.

Biowaste such as crop and animal residues, municipal waste usually referred to as second generation biomass are beginning to be recognized as a resource for the production of bioenergy and variety of other products. Agricultural wastes for example contain high level of cellulose, hemicellulose, starch, proteins as well as lipids. Biowaste therefore is a low-cost substrate for biotechnological industries such as for bio-fuel, fertilizer and microbial enzyme production.

The end products obtained directly and by conditioning and conversion of the biomass, are ultimately added to the bioeconomy as food, feed, fibre, fuel, chemicals and other industrial biotechnological products and some key sectors that can be identified in a bioeconomy are agriculture, forestry, fisheries, aquaculture, food and beverages, biobased electricity, biofuel, biobased chemicals, biopolymers and plastics, pharmaceuticals and health care products, wood and wood furniture, biobased textiles, leather, paper and paper products. However, the definition and the classification of bioeconomy vary among nations. Countries in the EU tended to use relatively a broad definition of bioeconomy landscape and include, in their entirety, economic sectors that produce or fundamentally rely on biologically produced materials. For example, the primary sectors such as agriculture, fisheries, forestry are included. Food, beverage, tobacco and wood product manufacturing also included. North American countries tended to adapt a narrower definition of bioeconomy landscape concentrating more on the economic activities driven by research and innovation in the life sciences and biotechnology. For example in the US only the agricultural production which has some form of interaction with the biotechnology or research in life science such as crops derived by genetic engineering and marker-assisted breeding, use of large informatics databases either for breeding applications or enhanced land use capabilities and also the plant biomass used in a downstream processing and/or fermentation utilizing recombinant or synthetic DNA technologies is accounted in the agriculture sector of the bioeconomy (Frisvold *et al.,* 2021). Table 1 shows the various sectors included in the bioeconomy definition identified in a review on “national frameworks of some selected countries including EU on assessing bioeconomy contributions to the economy” (FAO, 2018).

Table 1: Sectors included into bioeconomy strategy and monitoring in selected countries and EU (source FAO 2018).

	Argentina	Australia	Germany	Malaysia	Netherlands*	South Africa	USA*	EU
Agriculture	**XX**	**X**	**XX**	**XX**		**X**	**XX**	**XX**
Automotive and Mechanical engineering			**XX**					
Chemistry (incl. bioplastics)	**XX**	**X**	**XX**	**XX**	**XX**	**X**	**XX**	**XX**
Biofuel/bioenergy	**XX**	**X**	**XX**	**XX**	**XX**	**X**		**XX**
Biorefining		**X**	**XX**	**XX**		**X**	**XX**	
Construction/building industry			**XX**					
Consumer goods such as cosmetics and cleaning products	**XX**		**XX**			**X**		
Feed	**XX**		**XX**	**XX**		**X**		**XX**
Fisheries	**XX**	**X**	**XX**	**XX**		**X**		**XX**
Food and Beverage industry	**XX**	**X**	**XX**	**XX**		**X**		**XX**
Forestry	**XX**	**X**	**XX**	**XX**		**X**	**XX**	**XX**
Health				**XX**		**X**		
Knowledge/Innovation		**X**	**XX**	**XX**	**XX**	**X**		**X**
Mining						**X**		
Pharmaceuticals industry	**XX**	**X**	**XX**	**XX**	**XX**	**X**		**XX**
Pulp and Paper	**XX**		**XX**		**XX**	**X**		**XX**
Textile	**XX**		**XX**		**XX**	**X**	**XX**	**XX**

X- sector included in bioeconomy strategy XX- included in bioeconomy strategy and monitored or measured * results for the Netherland monitor bio-based economy and results for the USA refer to bio-based products industries.

Contribution of Bioeconomy

Over the last two decades there has been an increasing trend in the contribution of bioeconomy for national economies of countries. Apart from the direct primary products from farming, forestry and fisheries, the bioenergy and the new generation of biobased products have shown an increasingly important role in the economic growth of countries. For example, in the energy sector of U.S. bioeconomy the bioethanol production and biodiesel production have reached to 13.8 billion gallons (Statista, 2021) and 1.8 billion gallons respectively in 2020 accounting for 17% of the consumption of renewable energy in the US (Voegele, 2021). The estimated value only from the biotechnology component of the bioeconomy in U.S. in 2017 was USD 388 billion which comprises the revenues from GM crops, biobased products and high value molecular biological reagents and drugs derived from biotechnology (biologics). This has accounted for 2% contribution to the U.S. GDP Bbioeconomy Capital, 2018). The estimate of the size and scope of the U.S. bioeconomy as given in the safeguarding the bioeconomy report of National Academies of Sciences, Engineering and

Medicine (NASEM), for 2016 was USD 959 billion which includes direct contributions, indirect and induced effects representing 5.1 percent of the GDP. In European Union too bioeconomy makes up an important impact. Since early 2000s, the European Commission has been very much promoting the bioeconomy in its member states. In 2017 the value added bioeconomy component of the EU economy was EUR 614 billion which represented 4.7 % of GDP. The indicators for the period 2008-2017 show a steep rise in value added in the bio-based pharmaceutical sector (18%) and in agriculture (12%) (Renewable Carbon News, 2020). In Latin American and the Caribbean countries where more than one fourth of the world's arable land and one third of its fresh water resources are available makes this region to be one of the primary producers of sustainable biomass. For these countries too, the bioeconomy represents a new and powerful opportunity. In addition to the advances made in the pharmaceutical industry, plant biotechnology that started with the use of genetically modified plants has become an important contribution for the bioeconomy of many countries in Latin America. A good example in Latin America is the outstanding efforts made by Chile in applying bioleaching to extract copper. There the bioleaching process was upgraded and there are currently many states and private owned mines that use bacterial bioleaching. Brazil, the world's second largest ethanol producer next to the USA produces its fuel mainly from the fermentation of sugarcane. This process is relatively efficient and environment friendly than ethanol production from maize starch as practiced in the USA (Sasson and Malpica, 2018). Many African countries are endowed with abundant resources of natural resources. Continued improvements in biomass productivity and an optimization of biomass use, combined with a viable bio-business sector that can add value to primary production, has the potential to drive a broader African bio-based economic growth. However, despite its potential to help solve sustainable development challenges, bioeconomy has not been yet adopted in Africa. Recent study by Oguntuase (2020) identified nine countries with bioeconomy related activities in Africa. South Africa stood out as the only country with a dedicated bioeconomy strategy and the other eight countries with bioeconomy related activities were Ghana, Kenya, Mali, Mauritius, Mozambique, Nigeria, Senagal and Uganda.

The potential for Asian countries to be the leaders in biobased production is enormous. The robust and continued economic growth, rich biodiversity and the lack of fossil resources except in the Middle East will provide the impetus for a biobased revolution in this region. Currently, China, India and Thailand have become the fourth, fifth and seventh largest countries in the world respectively, for production and utilization of bio-ethanol mostly using sugarcane molasses. The oil palm biomass in Malaysia and Indonesia provides the base for a variety of high value products including biodiesel. However, the palm oil based biofuel industry has now been affected by the recent decision of the European Commission to phase out palm oil based biofuel from transport, an issue that has to be solved through negotiations. Thailand has already taken steps to promote its bioeconomy mainly based on cassava and sugarcane. These two crops are selected since they are already being produced in large quantities and can be developed into many value-added products including bioplastics derived from cassava starch and biofuels from both sugarcane and cassava. The industries based on advanced scientific technologies such as biopharmaceutical industry in Asia shows a promising growth. China, Japan, South Korea, India, Taiwan, Singapore, Malaysia, Thailand and Indonesia are in the forefront in the biopharma industry. The continuous increase in the innovation capacity of these countries in the fields of life sciences and biotechnology as a result of implementation of national policy and strategies promoting investments in research, training and education has open up new opportunities for bioindustry based economies. The concept of bioeconomy thus remains a viable option for every country to achieve major global challenges both economic and environmental.

Bioeconomy in Sri Lanka

The island of Sri Lanka has a land area covering 65,610 km^2 out of which 33,400 km^2 are under agricultural land. It possesses a territorial sea of 21,500 km^2 and the Exclusive Economic Zone (EEZ) extends outward 370 km from the shore and covers an area about 510,000 km^2 of the Indian Ocean (Arachchige *et al*., 2017). Sri Lanka has the right to resources in the water column, seabed and subsurface in the EEZ. The country is endowed with rich flora and fauna diversity and it along with the Western Ghats is one of the 34 biodiversity hotspots of the world (iucn.org/asia/, 2020). Though further extension of land is not possible for biomass production the sea and biodiversity endowments still remain as underutilized resources with a great potential to contribute for a stronger bioeconomy. The country should adopt the bioeconomy strategies to foster the policy support and investment which will lead to pioneering research, development of new and advanced technologies, expand education, capacity building, create public awareness, drive forward the industrialization processes and stimulate consumer demand on bio-based products.

Currently, the agriculture sector is the major component of the Sri Lankan bioeconomy. It comprises growing crops in plantation as well as non-plantation segments, rearing animals, fisheries and forestry sectors and primarily it provides the biomass for food, feed, fiber, and energy needs. The direct contribution of the agricultural sector for the GDP is currently at 6.9%. The country is almost self-sufficient in rice, vegetables and fruits. However, per capita consumption of fruits and vegetables in Sri Lanka is lower than the quantities recommended by nutritionists. The available statistics do not reflect on actual fruit and vegetable production and consumption, as Sri Lankans consume a significant amount of many different types of seasonal fruits and vegetables grown in their own home gardens, such as jak, bread fruit, *thumba karawila, thibbatu*, leafy vegetables, and many underutilized fruits and vegetables. Details of these do not show under available national statistical information. The country is dependent on the imports of some essential food and feed items such as maize, sugar, condiments and pulses. This shows the need of enhancing production of such crops in order to strengthen food and nutrient security and self-sufficiency. It also reveals that at present Sri Lanka does not have the abundancy of the agricultural biomass to be used as a feedstock for the industries from the dedicated crops such as rice, maize and cassava. Any attempt to use these food crops to produce biofuels or other related biobased industrial products at this moment would invariably create a strong public backlash. Sri Lanka is standing far behind in converting these main annual crops to value added products. However, the part of the biomass which does not directly impair the food and feed supply such as industrial crops, spices, agricultural residues and fuel wood can play an important role in establishing new value chains in the Sri Lankan bioeconomy. For example, currently biomass is the second largest energy supply source which provides 36% share of the total energy requirement of the country (Sri Lanka Sustainable Energy Authority, 2017).

Even though the contribution of the agriculture sector to the gross domestic product has eclined sharply from 20% to less than 7% during the past two decades, the volume of agricultural produce in this period has not been declined. It has steadily secured the food needs of the nation. However, Sri Lanka is now gradually moving towards an economy oriented around manufacturing and services. More and more agricultural produce is used as the raw materials in the value added industrial sector and new opportunities are being created in the service sector expanding the GDP contribution of these two sectors at the expense of the primary agricultural sector. For example, a major share of around 48% of the industrial GDP sector comes from the agri-based manufacturing industries such as food products, beverages, tobacco, leather, paper and pulp, bio-based health products, pharmaceuticals and manufactured natural rubber products. Bio-based activities can also penetrate to the service sector of the GDP such as trade, insurance, disease diagnostics services, education and training. Thus, the bioeconomy cuts across all the GDP sectors of the economy. The main subsectors of the bioeconomy of Sri Lanka and their approximate contribution are presented in Figure 2.

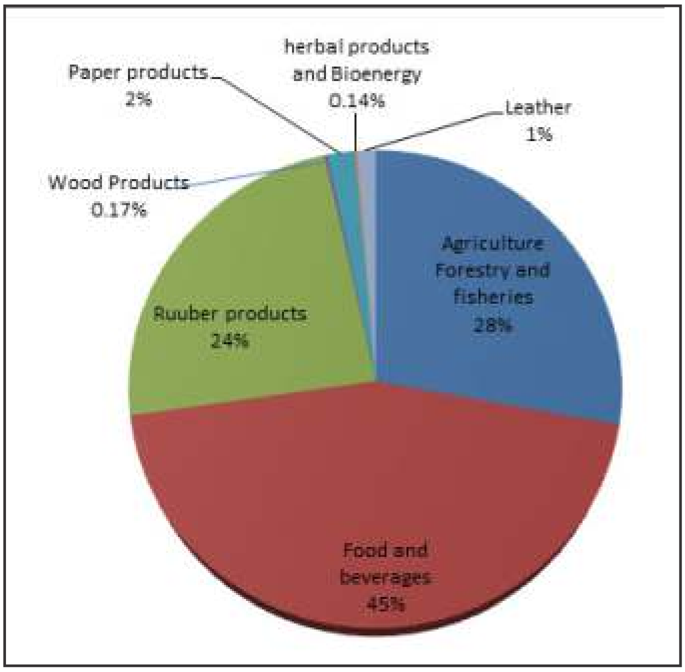

Figure 2: Contribution of subsectors of bioeconomy of Sri Lanka.

The Biotechnology Perspective in Sri Lankan Bioeconomy

In Sri Lanka the biotechnology perspective of bioeconomy is still in its infancy. The main biotechnological applications are seen in the food and beverage sector which uses the traditional fermentation technologies for the production of alcoholic beverages, fermented food and dairy products. The fermented food and dairy product industries are now even successfully adopted in the small and medium enterprise sectors. Among the few biotechnology based industries and services plant tissue culture, mushroom culture, manufacturing of herbal based health promoting agents, cosmetics and pharmaceuticals, bioenergy, organic and biofertilizers, biopesticides, animal vaccine production, services related to disease diagnostic applications and supply and marketing of biotechnological products have now begun to emerge in the Sri Lankan bioeconomy. However, the prospects for a rapid development of a knowledge base bioeconomy in Sri Lanka are still scarce. The progress that is being made in scientific advancements in life sciences to introduce technologies and innovations to use the renewable biological resources is slow as many of the research projects undertaken are short term and do not possess a critical mass of scientists for continuation. However, there are several exceptions to this statement. A big contribution has been made in the seed and planting material production sector. Many new high yielding and disease resistance varieties developed by the breeders have made a noteworthy improvement in agricultural productivity. The breeders and scientists have begun to use new biotechnological tools such as Marker Assisted Selection, protein and nucleic acid based virus indexing methods and tissue culture techniques to develop disease free, quality planting materials for local and export markets. The new technologies are well established at commercial levels in animal vaccine production, fishery and prawn hatchery operations. The expanding herbal product development sector uses many aspects of indigenous knowledge and biodiversity with advanced extraction and formulation technologies. However, still a large opportunity exists in value addition in the herbal sector, predominantly in the spice crop sector where most of the harvest is exported in its primary form. Sri Lankan spices are well recognized to contain a higher concentration of bioactive compounds and fetch a premium price in export market due to their intrinsic value. The real wealth contained in these biomasses at present is exploited only in foreign soils by producing high value end products such as pharmaceuticals and cosmetics. Sri Lanka having a limited land resource cannot continuously expand and depend on the volume of export of herbal biomass in its primary form but has to be resorted to technology based industries producing high value end products for export. In Sri Lanka the sea and microbial bioresources are still untapped. There is a great potential for utilization of renewable aquatic and marine biological resources to make products such as novel food, food additives, animal feed, nutraceuticals, pharmaceuticals and cosmetics. It is also a need to develop the human resources and the infrastructure for bioprocess and biorefinery industries, the prominent applications of biotechnology, to produce biofuels, biopolymers, biochemicals and industrial enzymes to foster the economic growth of the country. In Sri Lanka therefore, biotechnology applications have a great potential to contribute for a stronger bioeconomy. Hence there is a pressing need in the country to create a motivated society to pursue the promising prospects offered by the bioeconomy in development.

Note: The values presented in Figure-2 are estimates calculated by the author based on data from various sources, predominantly from Central Bank reports and serves as a rough guideline.

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Chapter

02

Stepping Towards Green-Economy:
Potential Use of Naturally Occurring Microbiota for Sustainable Development of Mosquito Vector Controlling Programmes in Sri Lanka

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Abstract

Distribution, abundance, and individual fitness of mosquitoes in a particular breeding habitat are known to be dependent on biotic factors. Naturally occurring microbiota associated with mosquito breeding habitats are among biotic factors which serve in the form of parasites, pathogens, predators, competitors, and toxic food items for developing larvae in these habitats. This fraction of microbiota species could serve as potential controlling agents against mosquito larvae, causing lethal effects on them. Further, microbiota composition and their interactions with mosquito larvae influence survivorship and developmental rate of immatures of mosquitoes which can ultimately alter the adult traits and population dynamics and thereby, more importantly, vector competence and their ability of disease transmission. Therefore, studies on microbiota associated with mosquito breeding habitats in terms of interactions with larval development have become important. Several species/taxa of unicellular organisms such as ciliates, bacteria, and some fungi and algae are known to cause negative or lethal effects on developing larvae. Identification of such naturally occurring microbiota and their interactions on mosquito larvae, in terms of parasitic, pathogenic, competitive, or predatory organisms would be beneficial for potential larval controlling approaches in an environmental-friendly manner. On the other hand, this approach would be of beneficial solution to overcome limitation of trained staff and funding through rational use of finances and human resources to achieve better productivity and fruitfulness of mosquito controlling programmes. Hence, in an emerging bioeconomy, the potential of the microbiota in mosquito breeding habitats to control vector mosquitoes makes such microbiota a valuable biological resource for scientists and vector mosquito control organizations to introduce bio-based products and strategies for controlling of vector mosquitoes.

Keywords: Insecticides, Microbiota, Mosquito, Parasites

Introduction

Mosquito-borne diseases are significant public health concerns in Sri Lanka and many other tropical and subtropical countries. Sri Lanka has been suffering from mosquito-borne diseases since ancient times with high prevalence of malaria, filariasis, and Japanese Encephalitis (JE). As a result of successive efforts in control programmes, Sri Lanka has attained some remarkable achievements by receiving the malaria-free certification in 2016, and filariasis as no longer a public health concern (WHO, 2016). However, despite of all the successful efforts of mosquito vector controlling measures that have been implemented throughout the past decades, some arbovirus infections particularly, dengue, have increased rapidly over the last few decades on par with global situation. Not surprisingly, arboviruses have caused vast epidemics with massive numbers of patients and deaths in Sri Lanka. Insecticide-based intervention is the most common method of controlling mosquito populations, since recent past in Sri Lanka. As a result of the reliance on a few active ingredients registered and used in public health, resistance has now evolved in most of the regions to all four classes of insecticides, *ie*. organochlorides, organophosphates, carbamates and pyrethroids. Apart from that the high cost which should be borne by the government, training of health staff for insecticide application, cost for protective equipment used during insecticide applications, impacts on other non-target organisms, bioaccumulation of chemicals along food chains and being non-environmentally friendly have strictly pinpointing challenges of using synthetic chemical insecticides for controlling mosquito populations. Therefore, nowadays scientists are targeting the use of other alterative measures, particularly usage of biological control strategies for vector control interventions.

Mosquitoes in Sri Lanka and their Breeding Habitat Requirements

Immature stages of mosquitoes being aquatic, most of their life stages are spent in water habitats. Therefore, selection of a suitable site for laying eggs is critical for both survival of immature stages and population dynamics of mosquitoes. Mosquito habitat ecology plays an important role in determining the larval densities, proliferation, species assemblage, and species succession. The quality of larval habitat is an important determinant of mosquito abundance as well as temporal and spatial distribution. Therefore, studying the ecological and environmental factors associated with the mosquito life cycle is a vital necessity.

Many mosquito species tend to select both natural and artificial containers as their breeding places. They exploit almost all types of lentic aquatic habitats for their breeding. On the other hand, abandoned rice fields and marshy land habitats in Sri Lanka have significantly influenced the distribution of mosquito populations including vector mosquitoes, thereby facilitating disease transmission (Udayanga *et al.,* 2018). Approximately, 159 mosquito species have been recorded in Sri Lanka under 19 genera (Gunathilaka, 2018). Mainly the larvae of *Culex* and *Anopheles* species are found in rice fields, nursery paddy beds, and large stagnant water bodies in Sri Lanka (Amarasinghe and Weerakkodi, 2014). Ecological factors and physicochemical properties of water significantly affect the mosquito density and abundance where habitats are concerned. The size, type, and nature of waterbody are important in selecting the oviposition sites by mosquitoes (Eitam *et al*., 2002).

Distribution, abundance, and individual fitness of mosquitoes in a particular breeding habitat are known to be dependent on mainly three factors; biotic factors (Blaustein and Chase 2007), abiotic factors (Alfonzo *et al*., 2005), and their interaction between each other as well as with other associated taxa (Chase and Knight, 2003; Beketov and Liess, 2007).

Biotic factors associated with mosquito breeding habitats include several species of macro-fauna and flora, micro-fauna and flora including bacteria, fungi, and protists that can be categorized into autotrophs, heterotrophs, and detritivores. To provide an ideal environment for the development of the mosquito immatures, each mosquito species owes its optimum biotic characteristics that act as oviposition cues. Microbiota associated with mosquito breeding habitats are among biotic factors and serve as parasites, pathogens, predators, competitors, non-competitors, and food items for developing larvae in these habitats. Therefore, there are naturally occurring microbiota species that serve as potential controlling agents against mosquito larvae, some causing lethal effects on them. However, the composition and abundance of such species in particular mosquito breeding habitats vary with the geographical location. Accordingly, the degree of parasitic, pathogenic, or predatory effects may also vary with the geographical location.

Interspecific Resource Competition of Mosquito Larvae

Within the mosquito breeding habitats, multiple mosquito species may occur. Thus, they may experience interspecific resource competition under food-limiting environments. Besides, there are different types of interactions between mosquito larvae and associated macrobiota and microbiota taxa. Such interactions may be of, parasitic, pathogenic, competing, or predatory effects occurred by different species of both macrobiota and microbiota species against mosquito larvae. When there is coexistence or mutualism of different mosquito species along with other biotic organisms, they form a community sharing habitat requirements (Devi and Jahuri, 2007).

Species of many algae, protists, and cyanobacteria, that represent an array of macrobiota in an aquatic habitat serve as food items for developing larvae. Apart from that, there are many associated macrobiota and microbiota, which share the same habitat with mosquito larvae, but causing no considerable negative or positive effect on each other. Preference of mosquito larvae for microbiota as food items may also vary with the species composition of microbiota, availability of other macro-food items, and non-living organic matter such as detritus within the habitat. Further, microbiota composition and their interactions with mosquito larvae influence immature survivorship and their developmental rate which can ultimately alter the adult traits and population dynamics and thereby, more importantly, vector competence and disease transmission ability. Therefore, studies on microbiota associated with breeding habitats in terms of interactions with larval development are important.

Several species/taxa of unicellular organisms such as ciliates, bacteria, fungi, algae are known to cause negative or lethal effects on developing larvae. Identification of such naturally occurring microbiota and their interactions on mosquito larvae, in terms of parasitic, pathogenic, competitive, or predatory organisms against mosquito larvae as controlling agents would be beneficial for potential larval controlling approaches in an environmental-friendly manner. On the other hand, this biological approach would be the solution to overcome lack of trained staff and funding through rational use of finances and human resources to achieve better productivity and fruitfulness of mosquito controlling programs. Hence, in an emerging bioeconomy, the potential of the microbiota in mosquito breeding habitats to control vector mosquitoes makes such microbiota a valuable biological resource for scientists and vector mosquito control organizations to introduce bio-based products and strategies for controlling vector mosquitoes.

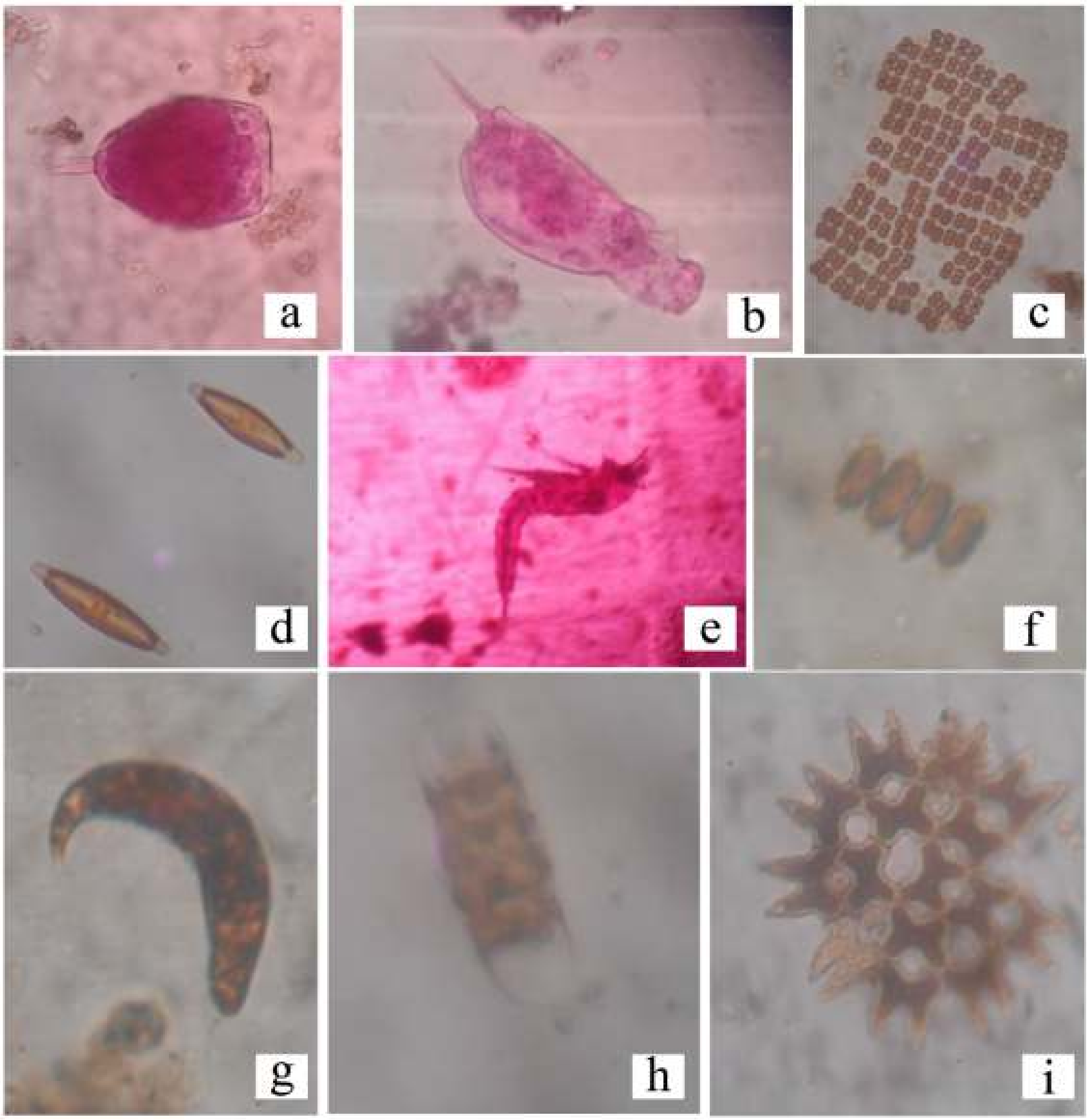
Research Conducted on Associated Microbiota in Mosquito Breeding Habitats

Only a few studies on associated microbiota in mosquito breeding habitats have been carried out locally and internationally. Comprehensive studies or checklists on associated microbiota, specifically targeting a variety of mosquito breeding habitats have been developed in Sri Lanka recently (Amarasinghe and Ranasinghe, 2019; Ranasinghe and Amarasinghe, 2020a; Ranasinghe and Amarasinghe, 2020b). During the above studies a total number of 83 naturally occurring microbiota species has been identified (Table 1) from variety of mosquito breeding habitats that are associated with mosquito larvae in Gampaha, Kandy, Kegalle, and Kurunegala districts of Sri Lanka (Amarasinghe and Ranasinghe, 2019; Ranasinghe and Amarasinghe, 2020a; Ranasinghe and Amarasinghe, 2020b; Ranasinghe and Amarasinghe, 2021). Microbiota species identified were belonged to 11 phyla namely Cilliophora, Rotifera, Ochrophyta, Charophyta, Amoebozoa, Bacillariophyta, Cyanophyta, Euglenozoa, Sarcodina, Chlorophyta and Arthropoda. Figure 1 shows some examples of microbiota species showing morphological characteristics. Majority of microbiota species that were associated with mosquito larvae belonged to Phylum Rotifera. Identification of naturally occurring microbiota and their interactions on mosquito larvae, would be beneficial for developing novel larval controlling approaches in an environmental-friendly manner.

Table 1: Naturally occurring microbiota associated with variety of mosquito breeding habitats in Sri Lanka.

Microbiota species recorded	
Phylum- Ciliophora	**Phylum- Rotifera**
Vorticella microstoma	*Cephalodella forficula*
Paramecium caudatum	*Monostyla hamata*
Strombilidium viride	*Lecane luna*
Zoothamnium sp.	*Philodina roseola*
Colpoda sp.	*Lepadella ovalis*
Coleps hirtus	*Asphlachna priodonta*
Paramecium bursaria	*Asphlachna brightwelii*
	Keratella tropica
Phylum-Cyanobacteria/ Cyanophyta	*Keratella valga*
Spirilina major	*Brachionus clavciflorus*
Microcystis sp.	*Lecane unguitata*
Difflugia corona	*Brachionus falcatus*
Oscillatoria sp.	*Lecane lunaris*
Mycrocystis sp.	*Brachionus forficula*
Anabaena affinis	*Brachionus quadridentatus*
	Brachionus urceus
Phylum-Amoebozoa	*Lecane papuana*
Arcella arenaria	*Monostyla bulla*
	Monostyla hamata
Phylum-Sarcodina	*Notholca acuminate*
Acanthocystis aculeate	*Philodina citrina*
	Diurella stylata
Phylum- Charophyta	*Brachionus* sp.
Cosmarium margaritiferum	*Euchlanis dilatate*
Cosmarium dimorphus	

Cosmarium antilopeum	**Phylum-Chlorophyta**
Cosmarium quadrum	*Crucigenia rectangularis*
Chlosterium sp.	*Crucigenia quadrata*
	Gloeocystis gigas
Phylum-Euglenozoa	*Pediastrum biradiatum*
Phylum-Ochrophyta	**Phylum- Arthropoda**
Pinnularia sp.	*Alona* sp.
Gloeobotrys limneticus	*Cyclops strenuous*
	Canthocamptus staphylinus
Phylum-Bacillariophyta	*Latonopsis australis*
Siurella sp.	*Daphnia* sp.
Gomphonema angustatum	*Parastenocaris brevipes*
Cymbella affinis	*Metacyclops minutus*
Arthrodesmum incus	*Sidacrystallina*
Synedra sp.	*Diaphanosoma brachyurum*



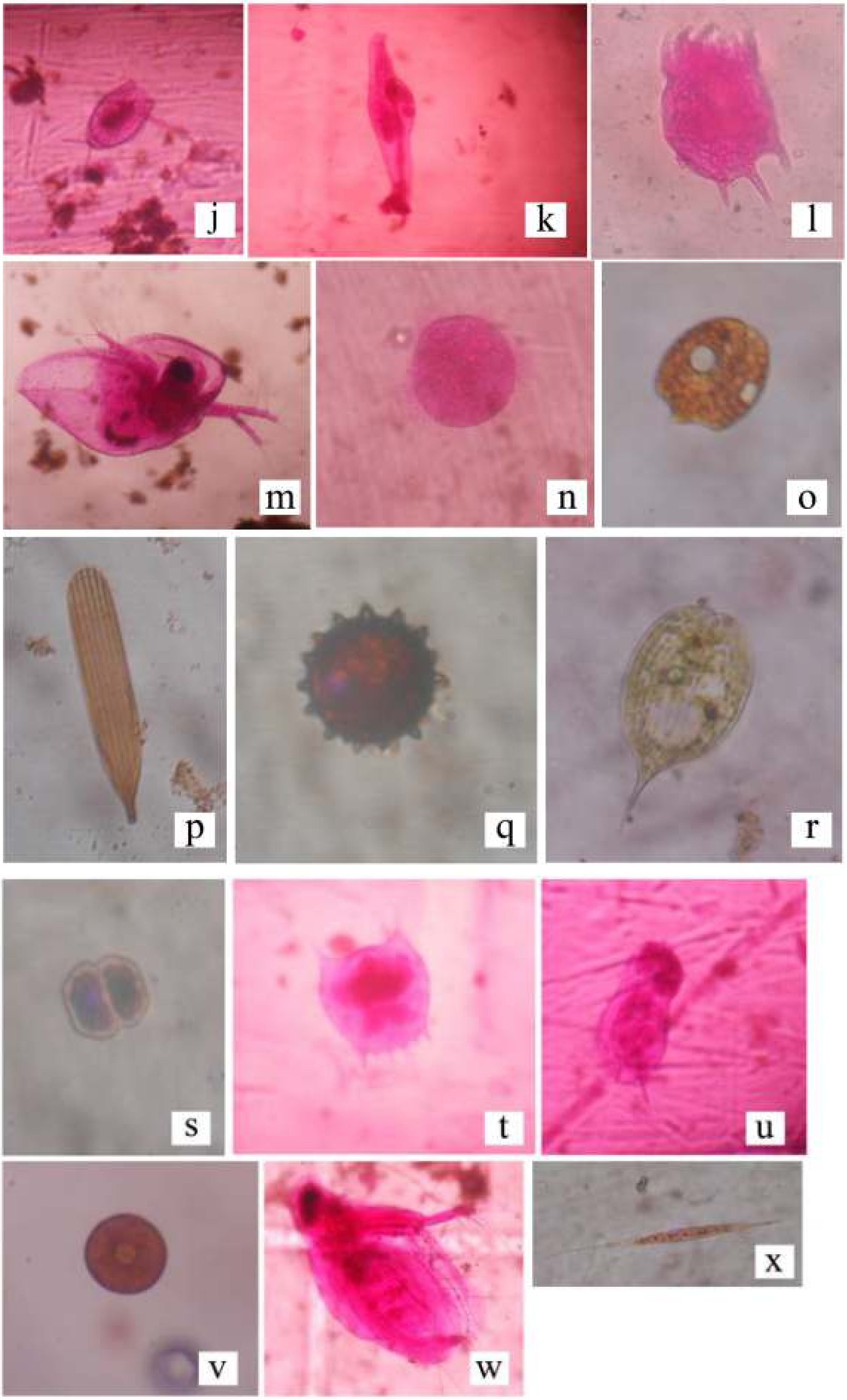

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Figure 1: Microbiota species identified from mosquito breeding habitats in Kurunegala, Gampaha, Kegalle and Kandy districts, Sri Lanka, x400 magnification; **a-***Monostyla hamata,* **b-***Diurella stylata,* **c-***Crucigenia rectangularis,* **d-***Gomphonema* sp., **e-***Canthocamptus staphylinus*, **f-** *Scenedesmus bijuga*, **g-** *Euglena acus,* **h-***Scenedesmus quadricauda*, **i-** *Pediastrum biradiatum,***j-***Lecaneluna,* **k-** *Philodina citrina,***l-***Keratellavalga,* **m-***Sidacrysallina*, **n-***Acanthocystis aculeata,* **o-***Phacus pleuronectes*, **p-***Notholca acuminata,* **q-** *Volvox aureus*, **r-***Phacus pleuronectes*, **s-***Cosmarium quadrum*, **t-***Brachionus quadridentatus*, **u-***Euchlanis dilatata*, **v-***Arcella arenaria*, **w-***Latonopsis australis,***x-***Chlosterim* sp.

Efficacy of Microbiota Activity on Vector Mosquito Larvae

Three species of naturally occurring microbiota namely *Vorticella microstoma*, *Zoothamnium* sp., and *Chilodinella* sp. as mosquito larval epibiont species were recorded from paddy fields in Sri Lanka. They are found to be effective epibiont on mosquitoes of the genus *Culex* which were also found most prominently and abundantly in paddy fields. When natural population of *Culex tritaeniorhynchus* mosquito larvae transferred to the laboratory and kept under observation, an unusual high larval mortalities reaching to 100% were observed and prompted to detect the causative organism (Ranasinghe and Amarasinghe 2020 a,b); a peritrich ciliate, *Vorticella microstoma.* Figure 2 shows that *V. microstoma* attached to the body of such dead larvae.

Percentage natural infection of immature stages of mosquitoes by epibiont parasitic ciliate, *V. microstoma* was different from mosquito species to species and the degree of mortalities were found to be different. The mortality rate of *Cx. tritaeniorhynchus* larvae collected from the paddy fields in Gampaha district was found to be higher compared to that of *Cx. quinquefasciatus* collected from the same fields. Similarly, the highest mortality rate was observed from the same mosquito species collected from paddy fields and irrigation canals in Kurunegala district due to *Vorticella microstoma* parasitism. Though *Cx. quinquefasciatus* larvae have been reported abundantly from associated abandoned paddy fields in Kurunegala districts, *Vorticella* species was not usually found associated with them. Laboratory experiments proved that only 26.78% mortality of *Cx. quenquefasciatus* larvae due to *V. microstoma* infection. This indicates a relatively low susceptibility of *Cx. quenquefasciatus* larvae to ciliate infection. Antagonist potential of *V. microstoma* on different mosquito species has also been determined in bioassays. When the mosquito larvae infested with *V. microstoma* were observed under a microscope (40× magnification) live sessile stalked trophont stage of the organism was found in high densities attached to the head, saddle, and abdominal regions of the body of dead mosquito larvae. *Vorticella microstoma* usually was not found attached to the siphon region of live mosquito larvae. However, *V. microstoma* was seen attached to the siphon and head regions of dead larvae left in culture bottles in extended period. *Culex tritaeniorhynchus* was the most preferred host of the trophont stage of *V. microstoma*, causing the death of 100% of the mosquito larvae, followed by *Cx. gelidus* with mean percentage mortality of 69.6 ± 7.41. *Anopheles subpictus* also showed the mean mortality percentage lower than to that of *Cx. gelidus* whereas, *Aedes albopictus* and *Ae aegypti* larvae were not preferred host species for *V. microstoma*. However, the precise reason for the death of the host is still unknown. It is possible that the biopolymer glue used for the surface attachment may damage the sensory systems, pore formation of infected larvae, and thereby interfering respiration (Mick, 1955).

Natural Occurrence of Effective Microbiota on Vector Mosquito Larvae in Sri Lanka

Vorticella microstoma was mainly abundant in flooded paddy fields in the early stages of the crop growth. During the harvesting time of paddy, or during dry season the fields usually get dried off hence, vector breeding habitats become unavailable. Coincidently, it affects parasitic or pathogenic ciliates that usually thrive in water habitat. Thus, survival of the parasitic agent should overcome the dry periods of the year until the next season of paddy cultivation coupled with monsoon rains during which high vector density situation returns. The encystation of these ciliates seems a possible way to escape the drought. When the environmental conditions are optimum, the organism excysts to release the trophont or free-swimming stage of this ciliate. Cysts and the processes of encystation and excystation have been described for *V. microstoma*. In addition, the cysts and desiccation resistance of *Chilodinella uncinata* (Das, 2003) have also been described.

Zoothamnium sp. has been also recorded as an epibiont in *Cx. gelidus* and *Cx. tritaeniorhynchus* mosquito larvae (Figure 3) in Gampaha, Kegalle, Kandy, and Kurunegala districts, Sri Lanka. However, no lethal effect on mosquito larvae was observed. *Zoothamnium* sp. had one main stalk with many branches ending in zooids which is a major difference from that of *Vorticella*. Upon stimulation, *Zoothamnium* contracts the entire colony into one large globule and then folds the main stalk. A previous study conducted by Amarasinghe and Rathnayaka (2014) has identified *Zoothamnium* sp. as the most prevalent microbiota in marshy lands in Sri Lanka. Further, *Zoothamnium* sp. has been reported as a microbiota species associated with the rice fields in the Gampaha district, Sri Lanka which was also found from the rice fields in the study carried out by Ranasinghe and Amarasinghe (2020a). They reported that *Zoothamnium* was acting as an epibiont species on *Cx. tritaeniorhynchus* mosquito larvae with no lethal effect. The possible reason for this finding may be due to the absence of *Zoothamnium* colonies in sufficient numbers to cause a lethal effect on mosquito larvae. *Zoothamnium* sp. also serves as external parasites for many freshwater shrimps and some fish species, thus the field applications must be carried out only after a risk assessment of the usage of these agents in higher densities. *Chilodinella* sp. was identified as another ciliate causing pathogenic effect (Figure 4) under natural environmental conditions on *Cx. tritaeniorhynchus* mosquito larvae collected from a paddy field in Gampaha district (6°57.959' N, 79°59.492'E). However, considerably high mortality was not observed (4.58% mortality of larvae compared to controls) due to the infestation of the pathogen to mosquito larvae. Endoparasitic ciliates were observed in the host larval body under microscopic observations. Anopheline larvae found less susceptible to *Chilodonella* infection than culicine larvae revealing that this ciliate also has a different degree for pathogenicity and host selection over different species of mosquito larvae. The body cavities of dead and transparent larvae were found packed with thousands of motile endoparasitic stage of *Chilodinella* sp. They attack mosquito larvae and invade the host hemocoel by dissolving the host cuticle while forming cuticular cysts. After the death of the host larvae, ciliate continues to reproduce for some more time and fill almost the entire body cavity of the susceptible host. At this stage, the infected larvae turn transparent, with thousands of motile microscopic endoparasitic stages visible inside the host body cavity (Das, 2003). Moreover, this ciliate was found highly virulent, desiccation resistant, and has a high reproductive potential with the capability to disperse in the environment by transovarial transmission through the mosquito host (Das, 2003).

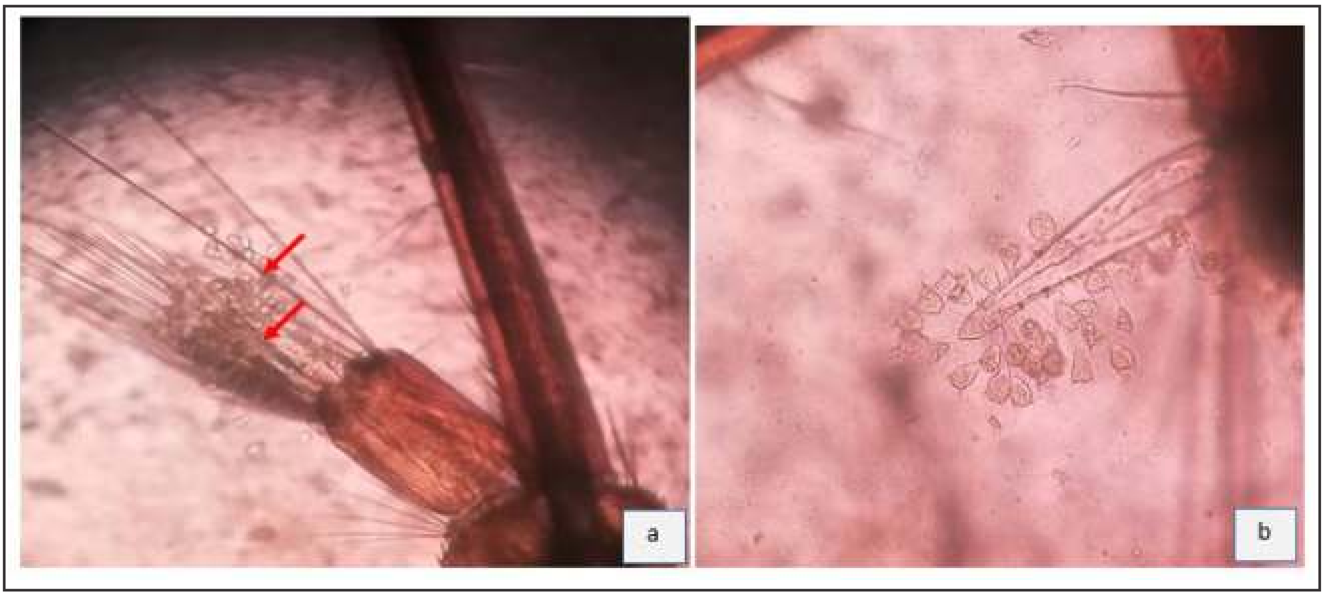

Figure 2: (a) Infection of the parasite, *V. microstoma*, to 3rd larval instars of *Cx. tritaeniorhynchus* anal papillae region (X 40 magnification); (b) attached trophonts of *V. microstoma* (X 100 magnification).

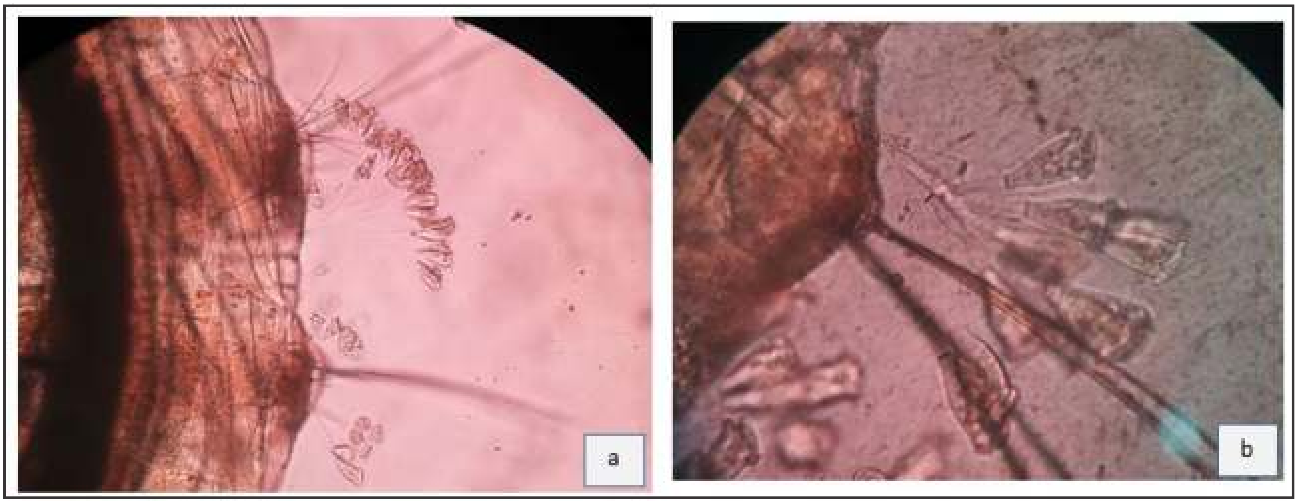

Figure 3: (a) Infection of parasite (*Zoothamnium* sp.) to 3rd larval instars of *Cx. tritaeniorhynchus* abdominal region (X 100 magnification); (b) attached colonies (X 400).

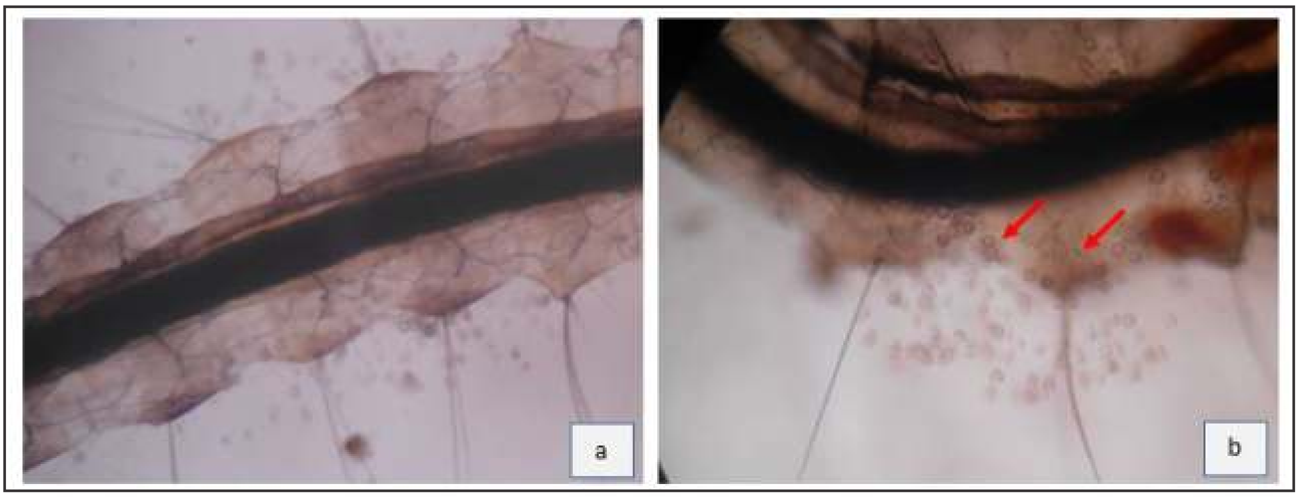

Figure 4. (a) Transparent *Cx. tritaeniorhynchus* larva (magnification x 40) showing endoparasitic ciliates, *Chilodinella,* in the host body; (b) cuticular invasive cysts of the pathogen (magnification x 100).

Discussion and Way Forward

It is evident that there is a naturally occurring microbiota with a potential for vector mosquito control interventions in an environmentally friendly manner, after proper evaluation of scenarios happening to non-target populations. However, with the container type mosquito breeding habitats, there is little effect of these ciliates on non-target organisms.

Cyclopoid copepods may act as effective biocontrol agents of mosquito larvae (Marten, 1984). Only one such species, *Metacyclops minutus* with low occurrence frequency was recorded from paddy fields from Gampaha district in Sri Lanka. Studies with copepods have been carried out using *Mesocyclops aspericornis* against the larvae of *Ae. polynesiensis* and *Ae. aegypti* (Riviere *et al*., 1987). These cyclopoid copepods reduce the survival of mosquito larvae by feeding on young first and second instars. In Sri Lanka, there are only limited investigations available on the predatory effect of copepods on mosquito larvae. The only published study recently has indicated that *Mesocyclops leuckarti* manifests the highest predatory effect on *Ae*. *aegypti* and *Ae*. *albopictus* larvae within 24 hours of exposure followed by *Mesocyclops scrassus* (Udayanga *et al*., 2019).

By knowing the composition of food that makes habitat particularly favorable for mosquito survival, it might be possible to manipulate those habitat types to eliminate mosquito breeding. This could be applicable as an additional tool for integrated vector control approaches. In line with this, Ranasinghe and Amarasinghe (2020a,b) have shown that food items represented in the larval gut are fairly in proportion to their abundance in the larval habitats. The larval gut of *Ae. aegypti* found from Kandy district mainly contained with food items from Phylum Bacillariophyta (38.98%) while it was from Phylum Chlorophyta (40.63%) for *Ae. Albopictus.* Similarly, food items in *Cx. tritaeniorhynchus* larval gut contained 37.5% Chlorophyta while from the identified gut content of *Cx. gelidus* larvae, 63.64% of the gut composition was from Cyanobacteria/Cyanophyta. Therefore, the preference for food items by mosquito larval species is different. Marten (2007) found that algae are generally represented in the gut of mosquito larvae in proportion to their abundance among the micro-flora and micro-fauna in larval habitats. This was true for *Ae. Aegypti, Ae. albopictus*, *Cx. tritaeniorhynchus* and *Cx. gelidus* larvae found in Kandy district, Sri Lanka, as they were found strongly associated with naturally occurring algae in their habitats. Kaufman et al. (2006) indicated the importance of algal biomass to the growth and development of *An. gambiae* larvae. Although many algal species were identified as a source of food for larvae in higher proportions for *Ae. Aegypti, Ae. albopictus*, *Cx. tritaeniorhynchus* and *Cx. gelidus* larvae, some studies have shown that while most of the algal species are nutritious food for many species of mosquito larvae, a few species were able to kill the larvae if ingested in large quantities. For example, *Cyanobacteria,* the blue-green algae were able to cause larval mortality by their toxicity and some species of green algae in the order of *Chlorococcales* was able to kill larvae by being indigestible (Marten, 1986; Marten, 1984). However, no species belonged to the Order *Chlorococcales* were identified in association with mosquito larvae (Ranasinghe and Amarasinghe, 2020 a,b). *Kirchneriella*, *Scenedesmus*, *Coelastrum*, *Selenastrum*, *Dactylococcus* and *Tetrallantos* species were found virtually indigestible by *Culex, Aedes,* and *Anopheles* species mosquito larvae, hence reduces the survival (Marten, 1984).

Krishnamurthy *et al*. (1989) reported that there are strains of *Mycrocystis aeruginosa* which are producing microcystin, a group of substances known to be toxic to various organisms. *Mycrocystis* sp. showed a significant negative effect on developing mosquito larvae where they are grown in the presence of *Microcystic* sp. were significantly smaller. *Mycrocystis* sp. was recorded from the study carried out by Amarasinghe and Ranasinghe (2019) in lower densities from the paddy fields and reservoirs associated with *Cx. quinquefasciatus* and *Cx. tritaeniorhynchus* larvae in Kurunegala district, and from abandoned wells where *Ae. albopictus* larvae are present in Kegalle district (Ranasinghe and Amarasinghe 2021), Sri Lanka. However, ~~a~~ no mortality effect was observed. Cyanobacteria seemed to have an important role in the diet of mosquito larvae. Apart from *Mycrocystis* sp. other species such as *Spiriluna major*, *Anabaena affinis, Gloeocapsa minima,* and *Oscillatoria* sp. were recorded as food sources from study sites in their investigation.

Marten (2007) has reported that many species of *Scenedesmus* were found to kill the larvae; although a recent study (Garcia-Sánchez *et al*., 2017) stated that, *Scenedesmus* species were encountered from both larval gut and in larval habitats, but its larvicidal property is yet to be confirmed. During a study conducted by Amarasinghe and Ranasinghe (2019), *Scenedesmus* sp. was only encountered from larval gut of *Cx. gelidus*, but no considerable lethal effect was observed from larvae. Further, Howland (1930) has reported that *Scenedesmus quadricauda* showed no signs of digestion in the mosquito gut. The same species was recorded from ponds from Gampaha and Kandy districts with no lethal effect on *Ae. albopictus* mosquito larvae (Ranasinghe and Amarasinghe 2020b). Probably the density of them was lower in the habitat to show mosquito larval mortality. Meanwhile, Duguma *et al*. (2017) has reported that increased abundance and diversity of micro-eukaryotes in the larval habitat significantly reduced the abundance of adult *Culex* mosquitoes. The reason the authors claim was that these organisms competed with mosquito larvae for small size class microbial biomass. However, ciliates, protists, and rotifers singly or in combination, found to inhibit larval growth. They altered other microbial populations in mosquito breeding habitats ~~at~~ with their presence and thereby inhibited larval growth as they compete with early instar mosquito larvae for food items (Duguma *et al*., 2017).

The abundance of *Aedes* spp. was found to be affected by the presence of micro-crustaceans, *Ceriodaphnia* spp, *Chydorus* spp, *Daphnia* spp, *Simocephalus* spp, Calanoida and larvae of Chiranomidae as they compete efficiently with mosquito larvae for food resources (Elono *et al*., 2010). Competition created by such crustacean species for larval food, could be exploited as a potential tool for integrated vector control approaches.

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Chapter

03

The Potential Strength of Eco-friendly Non-modified Biological Control Approaches as Additional Tools in Integrated Management of Dengue Vectors in Sri Lanka

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Abstract

Dengue has been recognized as one of the major public health concerns in Sri Lanka nowadays. Suppression of vector population and limiting the vector-human contact has become the main strategies in the disease control programmes. In control of dengue vectors, chemical-based methods are widely used to reduce vector densities. However, downsides allied with the chemical-based control approaches such as the development of resistance against insecticides, adverse impact to the non-target organisms and environment have widened the attention toward eco-friendly methods in the integrated vector control concept for vector management. In Sri Lanka, several biological-based control approaches have been evaluated and used in control progammes with different magnitudes. However, many of these approaches have not been involved in the vector control strategic plan. The present work reviews the biological vector control approaches which have been evaluated in Sri Lanka at some scale and reiterate the potential strengths in integrated vector management.

Keywords: *Aedes*, Agents, Biological vector control, Management

Dengue as a Major Public Health Problem

Dengue Fever (DF) is a disease caused by an arbovirus that is transmitted by *Aedes* mosquitoes, particularly *Aedes aegypti* (Linnaeus). *Ae. albopictus* (Skuse), a secondary vector has drastically expanded the geographic range in recent years (Lambrechts *et al*., 2010). Today, the geographic distribution of dengue mosquitoes includes more than 100 countries worldwide. The global burden of dengue is large, an estimated 50 million infections per year occur across approximately 100 countries, with potential for further spread (Figure 1).

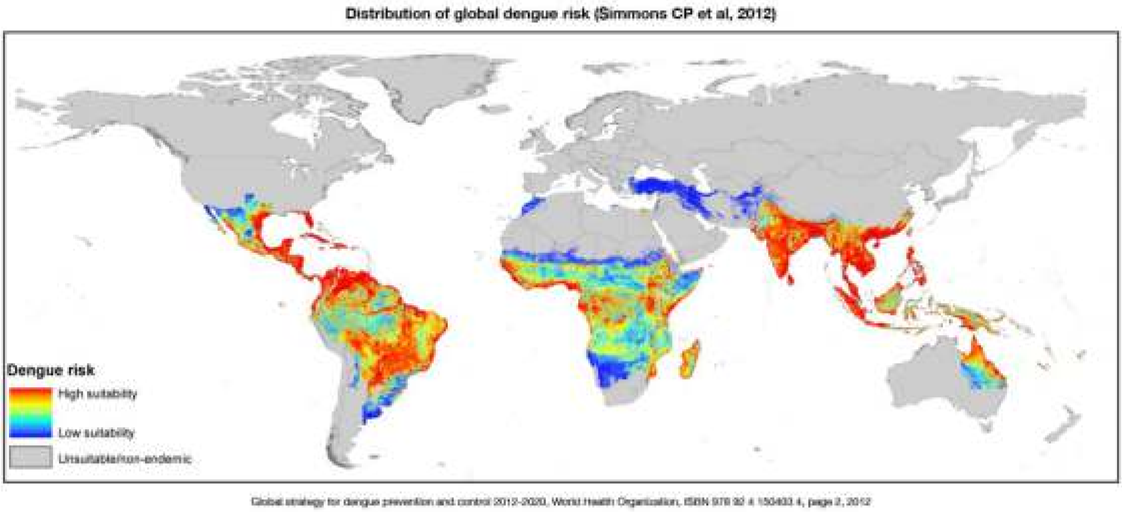

Figure 1. Distribution of global dengue risk (Source: Simmons *et al*., 2012)

In Sri Lanka, the history of dengue infection way back to 1960 which is the first dengue case recorded from the country as an imported case followed by the first local case in 1962. However, the dengue infection was not a major public health concern until it is declared as a notifiable disease in 1996 (National Control Unit, Sri Lanka) Since early 2000, progressively larger epidemics of dengue with more cases of Dengue Hemorrhagic Fever (DHF) occurred at regular intervals. Major epidemics of dengue infection were reported since 2009 in Sri Lanka. Most of the cases (47-55%) have been recorded from the western parts of the country, mainly in three districts namely; Colombo, Gampaha and Kaluthara (Dalpadado *et al*., 2021).

Vector control and minimizing the vector-human contact are widely used principles in the control of dengue infection (Morrison *et al*., 2008; Hamid *et al*., 2018). Currently, vector control approaches are mainly based on source reduction, application of insecticides, public health education, and legislation (WHO, 2009; Karunaratne *et al*., 2013; Dalpadado *et al*., 2021).

The use of insecticides is considered the most efficient application in vector control programmes. However, insecticide-based control is not the only intervention practice by the modern integrated vector management approach practices in the world. This is mainly due to limitations allied with the insecticide-based control methods such as developing resistance, adverse effects to the non-target organisms and detrimental effects cause to the environment.

In Sri Lanka, the government has taken a policy decision to ban the use of agrochemicals and agro-fertilizers in the country due to health concerns and adverse impacts on the environment.

Therefore, it is important to explore the possibilities of using environmentally friendly all possible biological control methods as vector control approaches in disease control programmes. Therefore, this chapter reviews biological vector control approaches (non-modified organisms) that have been evaluated in Sri Lanka at some scale.

Biological Approaches to Control Disease Vectors

Biological control of dengue vectors is based on the concept of introducing organisms that prey upon, compete with, or otherwise reduce the density of vectors. The immature stages of the vectors in household water containers provide a suitable target for the introduction of biological control agents. Their introduction must be safe, inexpensive, easy to produce on a large scale and be culturally and socially acceptable to the target population.

Larvivorous fish

For dengue control, larvivorous fish have been used successfully as biological control agents in water jars and other large containers in many countries (Chang *et al.,* 2008). In Sri Lanka, water storage cement tanks, barrels, ornamental ponds and wells have been identified as important breeding sites of *Ae. aegypti* and *Ae. albopictus* (Kusumawathie and Fernando, 2003; Kusumawathie and Siyambalagoda, 2005; Kusumawathie *et al*., 2010). Therefore, introducing a self-propagating predator into these breeding habitats such as larvivorous fish could be a good alternative to control vector population (Kusumawathie *et al*., 2015).

In Sri Lanka, *Oreochromis niloticus, O. mossambicus, Poecilia reticulata, Aplocheilus dayi, Puntius bimaculatus, Rasbora daniconius* and *Rasbora caveri* (Figure 2) have been tested for larvivorous potential and indicated predatory activity against dengue vectors (Surendran *et al*., 2008; Wijesinghe *et al.,* 2009; Kusumawathie *et al.,* 2015; Ranathunge *et al.,* 2021). It has been recommended that since *Ae. aegypti* and *Ae. albopictus* breed in water storage containers such as cement tanks, barrels and ornamental ponds, the fish species that showed high larvivorous potential to be considered for dengue vector larval control in such containers. Further, a field and laboratory-based study conducted by Surendran *et al*. (2008) using *O. mossambicus* has also confirmed that the introduction of this species into water storage tanks is effective in eliminating *Aedes* larval breeding within 3 days (Surendran *et al.,* 2008).

Figure 2: Larvivorous fish commonly used in mosquito control (a): *Poecilia reticulata* (Source: Deacon *et al.,* 2015), (b): *Aplocheilus dayi* (Source: National Dengue Control Unit, Sri Lanka, 2019), (c): *Puntius bimaculatus* (Source: Dignall, 2021; © Beta Mahatvaraj), (d): *Rasbora daniconius* (Source: Tropical Fish Keeping), (e): *Oreochromis spp*. (Source: National Dengue Control Unit, Sri Lanka, 2019).

Copepods

Although cyclopoid copepods have long been known to prey on mosquito larvae (Hurlburt, 1938; Bonnet and Mukaida, 1957; Fryer, 1957), the unique potential of these tiny crustaceans for mosquito control was first esteemed in the 1980s (Marten, 1984). Most large species of cyclopoids are voracious predators of first instar mosquito larvae (Figure 3), more effective for biological control than other predatory invertebrates because it is so common for cyclopoids to be numerically abundant. Cyclopoids are practical for large-scale use because most species are easy and inexpensive to mass-produce. However, the application of copepods as control agents are associated with some limitations such as the predacious efficacy may depend on the stage/size of the mosquito larvae, species-specific predatory potential and they are appropriate as a control tool only for some breeding habitats.

According to the published information, predation against the first instar of *Aedes aegypti* and *Aedes albopictus* by five copepods species namely; *Cyclops languides*, *C. varicans*, *C. vernalis*, *Mesocyclop leuckarti* and *Mesocyclop scrassus* have been evaluated in Sri Lanka under laboratory condition. It has been stated that among these five species only *M. leuckarti* and *M. scrassus* have been suggested as suitable candidates to be considered as potential candidates (Udayanga *et al.,* 2019). However, there could be many potential candidates with higher capacities in the country. Therefore, investigations should be driven to explore such candidates to be used in integrated vector management. In addition, some species records from different geographical locations and zone may not be the ideal candidates for other regions, because the predatory efficacy can be different with the species and strain records from different regions.

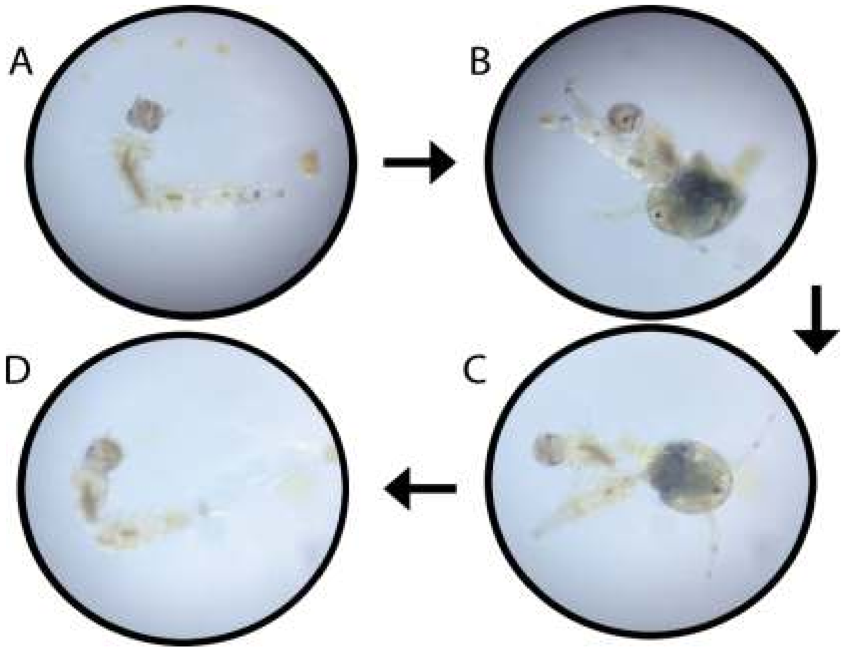

Figure 3: Predatory activity by Copepods on the first instar of *Aedes* larvae. (A); First instar *Aedes* larvae, (B) & (C); Praying on *Aedes* larvae by the Copepod, (D); Dead *Aedes* larvae after the Copepod attack (Source: Udayanga *et al.,* 2019).

Use of dragonfly/ damselfly nymphs

Dragonflies belong to the order Odonata, which includes dragonflies (Suborder Anisoptera), and damselflies (Suborders Zygoptera). Both, the nymph and the adult stages of dragonflies and damselflies predate on mosquito larvae (Shad and Andrew, 2013). Prey-capturing by nymph involves visually locking onto the prey to be captured (Figure 4). In certain cases, pressure build-up in the abdomen with water helps to generate quick forward movement using propulsion (Kundanat *et al.,* 2021). Although dragonflies are often associated with ponds, streams and swamps, they have rarely been studied as potential biocontrol agents against medically important mosquito vectors (Chatterjee *et al.,* 2007).

This approach is associated with several limitations. The main drawback is since the life cycle of dragonflies are long which the nymph stages consist of 10-15 instars. Therefore, it takes nearly one year or more (Chatterjee *et al.,* 2007). Hence, maintaining these cultures artificially under laboratory conditions may be challenging. Further, these candidates are suitable only to some selected breeding habitats due to the bioecology of dragonflies and damselflies.

In Sri Lanka, only one published information available thus far. In that study, predation by nymphs of five dragonfly species; *Anax indicus, Gynacantha dravida, Orthetrum sabina sabina, Pantala flavescens,* and *Tholymis tillarga* have been tested against *Ae. aegypti* larvae under laboratory conditions (Samanmali *et al.,* 2018). Predation rate has been observed highest in *A. indicus* followed by *P. flavescens*, *G. dravida* and *T. tillarga* (Samanmali *et al.,* 2018). Further, this investigation has recommended *A. indicus* and *P. flavescens* as potential candidates to be used for filed applications.

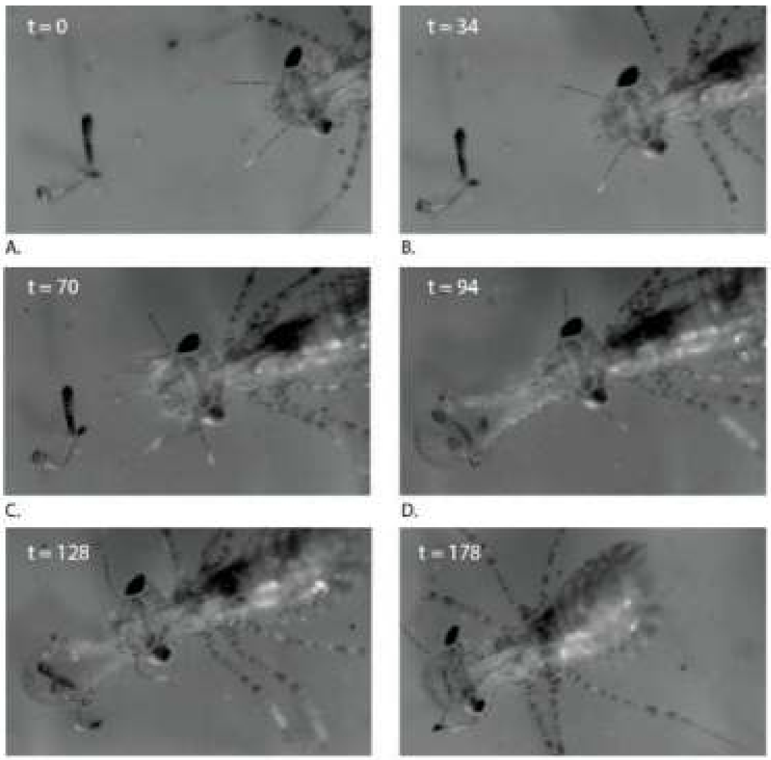

Figure 4: The sequence of dragonfly nymph capturing a mosquito larva at different time points in milliseconds (Source: Kundanat *et al.,* 2021).

Use of carnivorous mosquito larvae

Mosquitoes are well known to transmit diseases through bites of female misquotes when taking a blood meal. However, not all species feed on blood and they obtain their required nutrient from other sources such as pant nectar. Species in genus Toxorhynchites is such an example. These mosquitoes are entirely non-blood feeding and not considered medically important. Interestingly, Toxorhynchites larvae prey on other mosquito larvae (Alonso-Palomares *et al.,* 2018).

As a result of this behaviour, these species have received wider attention since they may be useful as biological control agents against medically important vector mosquitoes (Figure 5). However, the appropriate application of Toxorhynchites as a biological control method is not common due to several restrictions (Donald *et al.,* 2020). The aggressive feeding behavior and cannibalistic tendency depend on species to species which enable them to consider as potential candidates in vector control (Donald *et al.,* 2020). This method has been recommended mostly to control vector breeding in domestic water storage containers (Donald *et al.,* 2020).

Figure 5: Toxorhynchites larvae consuming larvae of a medically important *Culex* mosquito (Source: Schiller *et al.,* 2019).

There are two species namely; *Tx. amboinensis Tx. splendens* and *Tx. minimus* have been recorded in Sri Lanka (Gunathilaka, 2018). However, only a few studies have been conducted in the country and of them, a limited number of research findings are available as published information. The application and effectiveness of the use of potential candidates may not readily be available since many of these studies are laboratory-based investigations. Therefore, it is essential to check the feasibility of these control agents to be used as effective measures in the natural environment.

A laboratory-based study was conducted with *Tx. splendens* against *Ae. albopictus* larvae have consumed 10 larvae within 330.0 minutes (Wijesinghe *et al.,* 2009). Further, some field-based investigations have been observed that, *Tx. splendens* and *Tx. minimus* breed in water-filled barrels (Wijesinghe *et al.,* 2009). Therefore, it has been emphasized the possibility of establishing *Tx. splendens* and *Tx. minimus* as a part of an integrated vector management programme targeting the breeding sites of *Aedes* mosquitoes in peridomestic cemented water tanks and water collecting/holding containers (Wijesinghe *et al.,* 2009).

Use of carnivorous aquatic plants

Carnivorous plants are adapted to grow in low-nutrient habitats. Pitfall traps in *Nepenthes*, flypaper traps in *Drosera*, snap traps in *Dionaea* and bladder traps in *Utricularia* are the major prey trapping devices among carnivorous plants. The ability of prey trapping is focused on mosquito control methods.

Genus *Utricularia* is a carnivorous angiosperm belonging to the family Lentibulariaceae, comprised of approximately 235 species (Silva *et al.,* 2016). They usually grow in nutrient-poor shallow habitats with standing waters like small lakes, ponds, oligotrophic marshes, and their distribution is highly fragmented (Beretta *et al.,* 2014). These habitats are usually poor in nitrogen and phosphorus. All the nutrients required for the plant, are directly taken up from the water by shoots or from animal prey by traps (Adamec, 2011). These traps capture small aquatic animals such as mosquito larvae (Figure 6).

Thus far there is only one study conducted in Sri Lanka on this aspect using *Utricularia vulgaris* and *U. reticulata* species. These plant species tested against early (I & II instars) and late (III & IV) stages of *Aedes aegypti* larvae have been indicated more than 70% of cumulative predatory efficacy within 72 hours of exposure (Perera, 2021). Further, these biological control agents have been recommended to use for slow-moving small water bodies, fish tanks, ornamental or man-made recreational ponds (Perera, 2021).

These carnivorous plant species may attract dragonflies for oviposition which enhances further biological control by nymph sages of dragonflies (Sawchyn and Gillott, 1975). Since *U. reticulata* is an endemic species to Sri Lanka, introducing this species in man-made ponds and tanks indoor or outdoor would enhance biodiversity and species conservation. Further, since this is a flowering plant having an attractive bright blue flower, which ultimately enriches the recreational value and contributes to ecological services such as the attraction of various pollinators like bees and butterflies.

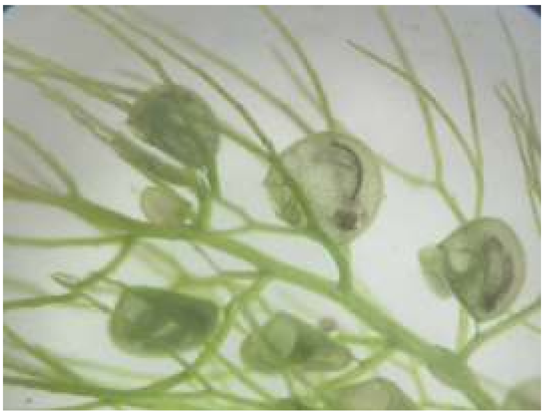

Figure 6: *Aedes aegypti* larvae trapped in the *Utricularia vulgaris* bladders (Source; Perera, 2021).

Endoparasitic ciliates causing an antagonistic effect on mosquito larvae

Endoparasitic ciliates (Protista: Ciliophora) have been known to infect mosquito larvae about a century ago with the report of *Lambornella stegomyiae* infection in the larvae of *Ae. albopictus* (Lamborn, 1921). Ciliates infect mosquito larval hosts within 48 to 72 hr after exposure (Figure 7).

However, some larvae can escape from parasitization by molting and reaching the next stage in their life cycle (Washburn *et al.,* 1991). Ciliates are also dispersed by infected adults that can infect larvae and form desiccation-resistant cysts. Invasion of ovaries in female mosquitoes induces oviposition and actively disperse ciliates through deposition into water. Adults of both sexes also passively disperse ciliates by dying on water surfaces (Egerter *et al.,* 1986).

Parasitic effects of ciliates such as *L. clarki* (Corliss and Coats, 1979; Egerter *et al.,* 1986; Washburn and Mercer, 1991) and *Tetrahymena pyriformis* (Corliss, 1961) have been reported in *Aedes* and *Anopheles* mosquitoes, respectively. Arshad and Sulaiman (1995) reported the virulence of *L. stegomyiae* to *Ae. albopictus* (mortality rate: 99.53%) and *Ae. aegypti* (mortality rate: 90.83%) under laboratory conditions.

Figure 7: Microscopic observation (x100) of *Vorticella microstoma* infected in the larvae of; (a): *Cx. tritaeniorhynchus* head region, (b): *An. subpictus* thorax region, (c): *Cx. gelidus* abdominal region, (d): *Cx. gelidus* anal papillae region (Source: Ranasinghe, 2020).

In Sri Lanka, a recent study has reported the parasitic and antagonistic effect of *Zoothamnium sp*., *Chilodinella sp*., *Vorticella microstoma,* and *V. microstoma* against some medically important mosquito larvae tested (Ranasinghe, 2020). Laboratory bioassays confirmed that there is a potential use of *V. microstoma* as a potential biological larviciding agent for *Cx. tritaeniorhynchus, Cx. gelidus* and *Anopheles subpictus* mosquitoes (Ranasinghe, 2020). Therefore, it is essential to test the effectiveness of these ciliates to control *Aedes* mosquito larvae and identify suitable ciliates that can be parasitized in dengue vectors.

Use of Bacillus Strains for Larval Control

Bacillus thuringiensis var. israelensis (Bti) is a gram-positive, spore-forming entomopathogenic bacterium first isolated in 1976 (Goldberg and Margalit, 1977). As a biological control agent, Bti has demonstrated high efficacy against target organisms, primarily mosquito and black fly larvae (Mittal, 2003; Lacey, 2007). *Bacillus thuringiensis israelensis* exerts its lethal effects through producing a variety of toxic proteins that are ingested by the larvae of susceptible organisms. These toxins are then activated in the gut of the larvae where they disrupt the cell membranes and death of the organism within 24-48 hours.

The specificity of this mechanism has been demonstrated in multiple studies with no adverse effects on non-target invertebrates and vertebrates (Lee and Scott, 1989; Merritt *et al.,* 1989; Lacey and Mulla, 1990; Saik *et al.,* 1990). Because of the complex mechanism of action involving many proteins, the potential for resistance development is greatly reduced. *Bacillus thuringiensis israelens* is available in several formulations that can be applied by hand or with conventional spray equipment (Lacey, 2007), allowing Bti to be utilized in a variety of breeding habitats. In comparison with synthetic insecticides.

In Sri Lanka, a local strain of *B. thuringiensis* strain has been identified from soil displaying insecticidal activity (Medical Research Institute, Sri Lanka; Baragamaarachchi *et al.,* 2019). The formulation of this bacteria culture has been tested against the dengue vector mosquito larvae (MRI, 2021) and a commercial product has been formulated with technical support from the Industrial Technology Institute, Sri Lanka (Bacto Bti).

Conclusion and Future Directions

Successful control of disease transmission, in particular by diurnal species, such as *Ae. aegypti* and *Ae. albopictus*, is challenging. Limitations of insecticides such as; resistance, high cost, damage to the environment, hazards on non-target organisms and human health, non-biodegradable nature have led to biological tools as a measure of vector mosquito control. However, the insecticide-based control measure cannot be disregarded since it is still contributed to reducing the vector densities drastically in control programmes. Therefore, to minimize the use of insecticide-based control, there should be more alternatives from other approaches to support the integrated vector management strategy.

Biological control approaches through non-modified organisms have received wider attention due to their eco-friendliness and promising outcomes. Larvivorous fish, copepods, dragonfly/ damselfly nymphs, carnivorous mosquito larvae (*Toxorhynchites spp*.), carnivorous aquatic plants, endoparasitic ciliates, use of Bacillus strains have been evaluated in Sri Lanka and used in the control programmes.

Biological-based control techniques have not been popularized and used in control programmes, since many of the evaluations are restricted to laboratory investigations which ended up only with a scientific publication. Therefore, the value to control programmes has become minimal. Hence, it is essential to disseminate findings to the vector control entities/decision-makers and evaluate the feasibility with field-based trials in coordination with the National Control Programmes.

Another limitation is that the applications of biological approaches are breeding site-specific and need rearing of organisms which is laborious, time-consuming and requires optimization of rearing protocols. Therefore, breeding sites that are suitable to use different biological control should be defined and described. In addition, effective organisms having larvivorous activity should be evaluated through field trials. Hence, it is vital to include recommendations and guidelines in the vector control strategic plans specifying the suitable breeding habits that can be targeted by different biological control methods. Further, sustainability of such control efforts should be ensured by establishing rearing facilities or recognizing the laboratories at the national level to maintain and continuous supply of organisms to the national control programme.

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Chapter

04

The Role of Microalgae in Sustainable Bioeconomy

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Abstract

Microalgae are a broad group of ubiquitous, microscopic and photosynthetic organisms which are widely present in aquatic habitats across different climatic zones in the world. Microalgae provide foundation for the life on earth due to their significant contributions for oxygenation of the atmosphere and primary production through photosynthesis. They have received new attention with global trends for bioeconomy which aims to meet the growing demand for food, products and energy using renewable biological resources and sustainable methods. In addition, marine microalgae play a major role in mitigating atmospheric CO_2, level by sequestration.

Applications of microalgae have become promising technology for producing renewable biofuels, food, animal and aquaculture feed, other value-added products such as cosmetics, nutraceuticals, pharmaceuticals, biofertilizers, bioactive compounds and waste management. Most commercialized strains as super food are green algae *Chlorella vulgaris* and blue green algae *Spirulina (Arthrospira)*. In pharmaceutical industry, *Dunaliella salina* and *Haematococcus pluvialis* are used to extract antioxidants such as beta-carotene and astaxanthin respectively.

Number of researches have shown the potential for effective applications of different microalgae species in Sri Lanka e.g. biofertilizer, biofuel, pharmaceuticals and waste management. Continuation of these projects at next level with pilot scale experiments focusing on optimization of culture conditions and product quality is highly important to expand the bio-based economy of the country.

Although microalgae are abundant in both marine and freshwater ecosystems and terrestrial environments of Sri Lanka, in most of the environments they are not identified or fully studied. Systematic studies for sample collection, isolation and identification of the species are very important as the foundation for marching towards microalgae-based bioeconomy in Sri Lanka. The isolated pure cultures need to be preserved in standard culture collections and a national algae culture collection for the future applications.

Keywords: Bioeconomy, Cyanobacteria, Microalgae, Phycoremediation, Sri Lanka

Introduction to Microalgae

The word 'algae' originates from the Latin word "*Alga alligata*" which means seaweed. Algae thrive in both saline water environments as oceans, seas and lagoons and freshwater environments such as lakes, ponds and rivers. Based on the cellular morphology and size algae are classified as macroalgae and microalgae. Multicellular, larger algae which are visible with the naked eye are termed "macro algae", while microscopic, smaller, single cell organisms are named "microalgae". The latter generally includes prokaryotic cyanobacteria (Blue green algae) and eukaryotic microalgae including green algae, red algae, and diatoms.

Microalgae are more popular as nuisance organisms for causing problems such as "dense algal blooms" in lakes and "red tides" in oceans. However, these organisms deserve to be recognized and appreciated for one reason that is around 3.5 billion years ago, the beginning of eukaryotic life paving path for evolution of land plants and animals on earth was facilitated by a special group of microalgae, the blue green algae (cyanobacteria). The latter performed chlorophyll a mediated photosynthesis releasing oxygen as a biproduct and filling the earth's atmosphere. At present, microalgae in marine and aquatic environments produce half of the oxygen on the earth, while the total biomass of algae is only tenth of the biomass of other plants (Chapman, 2013).

Primary Production in Aquatic Environments

Microalgae as primary producers use carbon dioxide or dissolved CO_2 in aquatic environments for photosynthesis using light energy and become the key source of food and energy for other organisms in aquatic ecosystems. In the ocean environment too, the unicellular microalgae including cyanobacteria, generally known as "phytoplankton" are the major primary producers in the food web. These are usually invisible but contribute for half of the annual global primary production (50×10^{15} g C), an equal amount produced by all the vascular plants in lands (Marnon, 2009).

Carbon Sequestration

Phytoplankton play a major role in the regulation of atmospheric CO_2 levels by sequestration. One fifth of the net primary production by planktons is moved to the ocean interior as sinking particles or dissolved material. Large fraction (99%) of the organic matter mineralizes to form inorganic carbon and enters deep ocean circulation in span of time 100 – 1000 years. Thus, the phytoplankton act as a biological pump to transport atmospheric CO_2 to the deep ocean, where it is sequestered for longer times (Marnon, 2009).

Ubiquitous in the Environment

Microalgae are a broad group of ubiquitous photosynthetic organisms which are widely present in aquatic habitats across different climatic zones in the world. However, they can with stand dry periods and winter season with resistant spores (e.g. Akinetes by blue-green algae *Anabaena* and cysts by green algae *Haematococcus*). Some algae types can grow in terrestrial environments as well. For example, cyanobacteria are the main photosynthetic organisms in biological soil crusts

(Moore, 2010). In paddy fields, planktonic and benthic algae are common during the period of submergence, while during the dry period they form resting spores and found as surface crusts (Darsha *et al.*, 2019). Some types can grow even in extreme environments as deserts, on rocks, wastewater and in symbiosis with other organisms.

Many freshwater algal species have planktonic (free-floating) and benthic (substrate-associated) stages in the life cycle. Planktonic algae drift freely within the water body, while some species are able to regulate their movement. Benthic algae grow usually by attaching to the underwater sediments such as rocks, mud and organic matter and form biofilms with bacteria, fungi and micro invertebrates. Some can grow as epiphytes on the higher plants, macro algae and colonies of planktonic algae.

Diverse Morphological Forms

Microalgae are diverse in shape, size and motility of the cells or colonies. There are four main types of algal cells or colonies based on the shape; i.e. unicellular, colonial, unbranched filamentous and filamentous forms. Unicellular non-motile algae are the simplest forms, which are generally sphere shaped e.g. *Chlorella* or with different cell shapes (e.g. *Selenastrum*). Unicellular motile forms have differentiated appendages such as flagella (e.g. *Chlamydomonas*) which enable them to move around. Colonies of microalgae are formed by small groups of cells that may come together (e.g. *Scenedesmus*) or large number of cells in aggregates with no definitive shape (e.g. *Microcystis*) or globular colonies with characteristic shape (e.g. *Volvox*). Filaments are colonies formed by cells arranged in linear forms. The filamentous microalgae can be unbranched (e.g. *Anabaena*) or branched (e.g. *Stigonema*). Some species show pseudo branching (e.g. *Johannesbaptistia*) (Figure 1). These growth forms have been described in relation to the growth of vegetative cells. During the winter months and dry periods, the vegetative cells are converted to thick-walled resting spores (e.g. Akinetes by Blue green algae, cysts by green algae species).

Other than flagellated species, microalgae gain motility with the aid of the surface mucilage. For example, blue-green alga *Oscillatoria* and some diatoms as *Navicula* can "glide" by secretion of mucilage. Mucilaginous sheath or cover in many algae is an important factor deciding the cell or colony morphology.

Based on the ability to form large colonies and motility, microalgae greatly vary in the size from micrometer size unicells to large colonies which are visible to the naked eye. Planktonic algae are classified based on the size of cells or colonies such as picoplankton (<2 μm, e.g. Blue-green algae *Synechococcus* and *Synechocystis*), nanoplankton (2–20 μm, Blue-green algae and Cryptophytes – *Cryptomonas*, *Rhodomonas*), microplankton (20–200 μm, Dinoflagellates *Ceratium*, *Peridinium*, colonial Diatoms *Asterionella*) and macroplankton (>200 μm, colonial Blue-green algae, *Anabaena* and *Microcystis*) (Bellinger and Sigee, 2015). The macro colonies of blue-green algae (diameter around 2000 μm) can be easily seen in the water column. In the benthic environment too, microalgae ranging from small unicells to large colonies of extended filaments to several centimeters (e.g. *Cladophora*) are found. They can grow as small free colonies or attached epiphytes on the substrate. Microalgae are the major component of "periphyton community" which is a complex biofilm of algae, bacteria, fungi, and meiofauna bound together with a polysaccharide matrix.

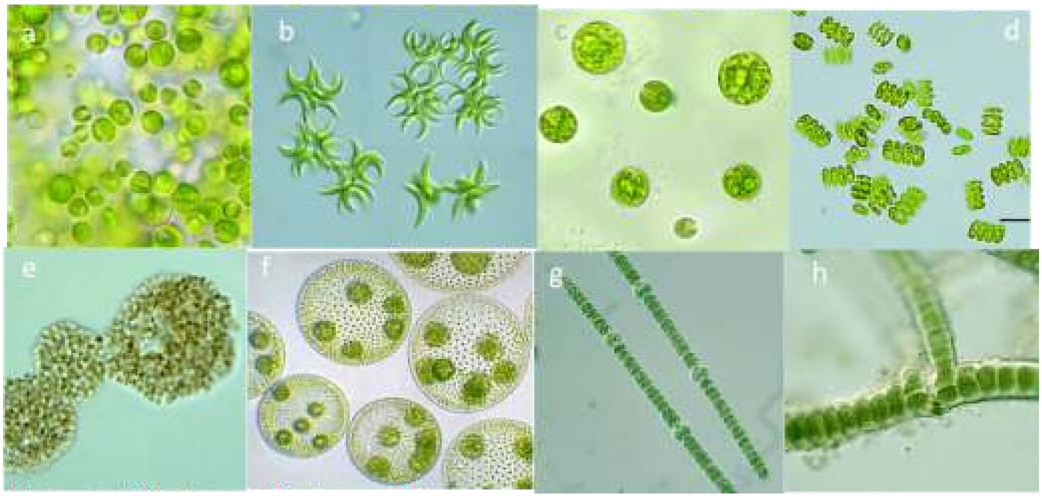

Figure 1. Main morphological forms of microalgae. Unicellular non-motile (a) *Chlorella* and (b) *Selenastrum*; Unicellular motile (c) *Chlamydomonas*; Small colonies (d) *Scenedesmus*; Large irregular shape colonies (e) *Microcystis*; Globular shape colonies (f) *Volvox*; Unbranched filaments (g) *Anabaena* and branched filaments (h) *Stigonema*. Images Source: (Baker *et al*., 2012)

Major Groups of Microalgae

Algae are not identified as a consistent taxonomic group, but constitute of several divisions or phyla which share common characteristics such as aquatic, photosynthetic, no vascular tissues in simple vegetative structures and simple reproductive bodies (Wehr and Sheath, 2002). "Phycology", the study about algae dates back to late 18th century by identifying and naming a kelp species *Fucus maximus* (currently *Ecklonia maxima*) by Swedish explorer Pehr Osbeck, an associate of the Farther of taxonomy, Carolus Linnaeus. There are agreements and disagreements among phycologists on the classification of different taxonomic groups as "Algae" (Kim, 2015). The commonly recognized algal groups are Rhodophyta (red algae), Phaeophyta (brown algae), Chlorophyta (green algae), Diatoms (Bacillariophyta), Haptophyte algae (Haptophyta), Dinoflagellates (Dinophyta), Euglenoids (Euglenophyta), Yellow-green algae (Xanthophyta), Cryptomonads (Cryptophyta), Chrysophytes (Chrysophyta) and Blue–green algae (Cyanobacteria) (Table 1) (Wehr and Sheath, 2002, Bellinger and Sigee, 2015). These groups vary in their photosynthetic pigments hence the color and several other cytological characteristics. Some groups have entirely microscopic unicellular species (e.g. Euglenoids) while some groups have entirely macroscopic species except microscopic growth stages (e.g. brown algae). Most of the algal groups are ubiquitous (e.g. blue green algae), and found in marine, freshwater and terrestrial environments, but some groups are predominant either in marine (red algae) or freshwater (e.g. green algae) environments.

There are an estimated several hundred thousand of alga species and about ten thousand have been classified so far. Various applications of microalgae have been revealed and currently about 20 microalgae are used for economic purposes (Posten and Chen, 2015).

Table 1: Major Groups of Microalgae and Commercial Species

Algal group	Photosynthetic Pigments	Typical color	Habitats and morphology	Examples of commercial species
1. Green Algae (Chlorophyta)	chl	Grass Green	Predominantly freshwater algae and some species are found in marine and terrestrial environments Planktonic, benthic and epiphytic species Range of growth forms; unicellular, flagellated, globular colonial, filamentous, large complex algae with branches	*Chlorella vulgaris*, *Dunaliella salina*, *Haematococcus pluvialis*
2. Diatoms (Bacillariophyta)	chl, fucoxanthin	Golden Brown	Major primary producers in both marine and freshwater environments, Planktonic, benthic and epiphytic species, Non-flagellated single cells, simple colonies or chains of cells Cell wall is distinct as a siliceous frustule	*Chaetoceros muelleri*, *Skeletonema* spp., *Thalassiosira pseudonana*
3. Haptophyte algae (Haptophyta)	chl, fucoxanthin	Golden Brown	Mainly marine and some freshwater species Unicellular or colonial, Distinctive haptonema filament which, is inserted between two flagella Some groups have "coccoliths", calcium carbonate platelets borne on the cell surface	*Pavlova salina* *Tisochrysis lutea*
4. Dinoflagellates (Dinophyta)	Chl, peridinin	Red-brown	Predominant in marine surface waters, limited number of species in freshwater environments Mostly biflagellated and unicellular, several plates form the cell wall, some species have projected plates as "horns" Some species are heterotrophic	*Crypthecodinium cohnii*
5. Cryptomonads (Cryptophyta)	Chl, PC or PE	Various colors	Found in both marine and freshwater environments; however, relatively less abundant Unicellular, planktonic and motile with two flagella Some species are heterotrophic	
6. Chrysophytes (Chrysophyta)	chl	Golden brown	Predominantly in freshwater and some species are in brackish and salt waters Planktonic; Unicellular, spherical and branching colonial types Flagellated motile spiciest and non-flagellated species Some species are heterotrophic or phagotrophic	

7. Blue–green algae (Cyanobacteria)	Chl, PE, PC, APC	Blue Green	Ubiquitous; found in marine, fresh water and terrestrial environments Withstand extreme environmental conditions Planktonic, benthic and epiphytic Prokaryotic cells, phycocyanin pigments give the characteristic color Range of growth forms; unicellular, colonial, un branched filamentous and branched filamentous, some species are motile by buoyancy or gliding	*Spirulina plantensis (Arthrospira plantensis)*
8. Euglenoids (Euglenophyta)	chl	Various colors	Predominantly present in freshwater; shallow lakes, wetlands, some species can withstand low pH and salinity Almost entirely unicellular flagellated motile organisms	
9. Yellow-green algae (Xanthophyta)	chl	Yellow-green	Mostly found in freshwater; in small water bodies and damp soils, Range of morphologies; from unicellular to colonial and filamentous	
10.Red Algae (Rhodophyta)	chl, PE, PC, APC	Red Blue – olive green	Predominantly marine species (97%) Freshwater members are mostly found in streams and rivers, most are macroscopic, some unicellular species	*Porphyridium cruentum*
11.Brown Algae (Phaeophyta)	chl, fucoxanthin	Brown	Almost entirely marine species, less than 1% of species, present in freshwater habitats, benthic growth, All are multicellular, some species have microscopic growth stages	
12.Eustigmatophytes (Eustigmatophyceae)	chl		all unicellular, found in marine, freshwater, and terrestrial environments.	*Nannochloropsis* spp.

chl = chlorophyll (green); PE = phycoerythrin (red); PC = phycocyanin (blue); APC = allophycocyanin (blue)

Applications of Microalgae

Growing population exerts a great pressure on the finite natural resources such as land and fossil energy. On the other hand, climate change exerts an impact on conventional production systems, such as agriculture. We need to find alternative ways for safe and secure supply of food, feed, energy and other demands by the humans, animals and the environment.

The bio-based economy – encompassing the sustainable production of renewable resources from land, fisheries and aquaculture environments and their conversion into food, feed, fiber, bio-based products and bio-energy as well as the related public goods (Khan *et al*., 2018), provides a way we must move forward for the sustainability of the earth.

During the transition towards an optimal use of renewable biological resources, we face challenges to select sustainable primary production and processing systems that can produce more food, fiber and other bio-based products with fewer inputs, less environmental impact and reduced greenhouse gas emissions. Currently, microalgae-based applications are being sought as promising methods for producing renewable biofuels, food, animal and aquaculture feed products and other value-added products such as cosmetics, nutraceuticals, pharmaceuticals, bio-fertilizers and bioactive substances simultaneously with sequestration of atmospheric CO_2 (Ryan, 2009; Harun *et al*., 2010). They also have applications in wastewater treatment which can be coupled with biofuel production which has inspired a new focus in biorefinery (Khan *et al*., 2018).

Exploration of microalgae as renewable and sustainable sources of biofuel has been initiated in 1970s. The first large-scale culture of the microalga *Chlorella* for commercial purposes was reported in Japan, in the 1960s. Over the last few decades, algae culturing expanded to new fields, such as food and feed, biofuels, and biopharmaceuticals. Number of high-value products have already been identified and marketed. Some of the most commonly produced valuable products obtained from microalgae are Lutein, β-carotene, α-carotene, α-tocopherol, Carotenoids, Astaxanthin, Docosahexaenoic acid (DHA), Eicosapentaneoic acid (EPA) and Glycerol. The poly unsaturated fatty acids (PUFA), DHA and EPA have a global market value of over 700 million US$/yr. followed by β-carotene (261 million US$/yr.), astaxanthin (240 million US$/yr.), lutein (233 million US$/yr.) and phycobiliproteins (60 million US$/yr.) (Bhalamurugan *et al.*, 2018)

Commercially Produced Microalgae

Many species of algae have been harvested as food from ancient times in many countries. Commercial cultivation of microalgae started only a few decades ago. The freshwater microalgae *Chlorella vulgaris* and blue green algae *Spirulina* (*Arthrospira*) are the main microalgae produced as dietary supplements for humans and animals in many countries as Japan, China, Australia and USA. Green microalgae *Dunaliella salina* and *Haematococcus pluvialis* are used to extract antioxidants such as beta-carotene and astaxanthin respectively. Astaxanthin, a strong antioxidant, is used as a nutritional supplement and anticancer agent, and exhibits preventive properties for diabetes, cardiovascular diseases, and neurodegenerative disorders, and also stimulates immunization (Kim, 2015).

The unicellular red algae genus *Porphyridium* has high potential in the production of polysaccharides (Arad and Levy-Ontman, 2010), arachidonic acid (Ahern *et al*., 1983), and phycobilin (Kathiresan *et al*., 2007). These microalgal polysaccharides and arachidonic acid have a variety of uses in medicine, nutrition, and cosmetics. Phycobilins are water soluble pigments found in the chloroplasts of Rhodophyta, which can be used for food and cosmetic colouring (Borowitzka, 2013), biotechnological uses like fluorescent tags (Glazer, 1994) and medical uses as photosensitizers in cancer treatment (Kim, 2015). The main commercially used genera of Haptophytes are *Isochrysis* and *Pavlova*, which are important aquaculture feeds due to their high content of DHA and generally high nutritional value (Guedes and Malcata, 2012). *Isochrysis* aff. *galbana* has also been identified for its high content of fucoxanthin (Kim *et al*., 2012), a pigment with medical properties in treating cancer, obesity, inflammation, and diabetes (Muthuirulappan and Francis, 2013). The marine cocolithophore *Emiliana huxleyi* has fast growth rate forming vast blooms. Thus, they play an important role in earth's carbon cycle and have great potential for production of renewable energy (Wu *et al*., 1999; Kim, 2015).

The main genus with commercial application for production of aquaculture feed is *Nannochloropsis*. They have high fatty acid content, mainly in the form of triacylglycerides and large proportion of PUFAs, including the essential eicosapentaenoic acid (EPA) (Huerlimann *et al*., 2010). Therefore, they are also considered for the production of EPA for humans (Winwood, 2013) and biofuel (Passell *et al*., 2013). Similarly, the freshwater Eustigmatophyceae *Monodus subterraneus* can be used for the commercial production of EPA (Lu *et al*., 2001).

Bacillariophyceae genera *Chaetoceros*, *Nitzschia*, *Phaeodactylum*, *Skeletonema*, and *Thalassiosira* are known to have high fatty acids, especially PUFAs thus commercially produced as feedstock for larval cultures in aquaculture. These are also under consideration for the production of biofuels and essential fatty acids for human consumption. In addition, *Phaeodactylum tricornutum*, and *Odontella aurita* are considered for the production of fucoxanthin (Xia *et al*., 2013), aquaculture feed (Guedes and Malcata, 2012) and EPA (Haimeur *et al*., 2012).

The genera *Aurantiochytrium*, *Schizotrichium*, *Thraustrochytrium*, and *Ulkenia* have commercial potential with their high lipid content in general and high DHA content (Lee Chang *et al*., 2014). This leads to potential applications as fish feed, the production of DHA for human consumption (Gupta *et al*., 2012), or the production of biofuels (Lee Chang *et al*., 2013). Even though Dinophyta are mainly associated with harmful algal blooms, they are also known for their generally high fatty acid content, especially the valuable omega-3 PUFAs (Kim, 2015). *Crypthecodinium cohnii*, a marine, heterotrophic dinoflagellate is commercially grown for the production of starter feeds for fish and known for high DHA content (30%) (Atalah *et al*., 2007).

Abundance of Marine and Fresh Water Microalgae in Sri Lanka

Phycological research in Sri Lanka dates back to the early studies on seaweeds carried out by the Dutch scientist Paul Hermann (1646 – 1695). List of “Ceylon algae” was compiled by G. Murray in 1887 and later new algal species were reported by Durairatnam in early 1960s and other studies including De Silva and Mallikarachchi in 2004. In the recent past, Prof. E. Coppejans from Belgium has compiled 'Sri Lankan seaweeds' based on extensive field research carried out along the coastline (Coppejans *et al*., 2009). The book provides an excellent reference for diverse green algae, brown algae and red algae species in the country. More recent studies on diversity of algae in south coastal water have been conducted by Premarathne et al, 2020. However, almost all of these studies represent only part of the algal biodiversity, as they have omitted microalgae including cyanobacteria, which dwell as smaller epiphytes, turf algae and crusts on coral reef.

Studies on freshwater algae in Sri Lanka can be traced back to 1902, where West and West have described 30 species of algae identified in samples collected at several locations across the country (Kulasooriya, 2017). Early studies have reported the presence of diverse microalgae species i.e. cyanobacteria, diatoms and green algae in the reservoirs of Sri Lanka. However, later studies have indicated the dominance of toxigenic genera such as Microcystis and *Cylindrospermopsis* in reservoirs influenced by the human habitation (Jayatissa *et al.*, 2006; Kulasooriya, 2017; Yatigammana and Perera, 2017). Recent studies in forty-five (45) reservoirs from the three major climatic regions of Sri Lanka (Wet, Intermediate and Dry Zone) have identified thirteen species of Cyanobacteria (*Microcystis aeruginosa*, *Nostoc* sp., *Cylindrospermopsis raciborskii*, *Chroococcus* sp., *Merismopoedia* sp., *Oscillatoria* sp., *Pseudoanabaena limnetica*, *Anabaena* sp., *Lyngbya* sp., *Coelaspharium* sp., *Aphanocapsa* sp., *Spirulina* sp., *Aphanothece* sp.,) and one species of dinoflagellate, *Peridinium aciculiferum* (Senanayake and Yatigammana, 2017).

Paddy fields are also considered as a habitat for microalgae growth due to the prevalence of favorable conditions as adequate light, water, high temperature and nutrient availability (Rodger and Kulasooriya, 1980). Previous studies have identified blue green algae, green algae, diatoms and other microalgae in paddy fields of Sri Lanka, with seasonal patterns (Dharsha *et al.*, 2019; Kulasooriya, 2011, Bambaradeniya *et al.*, 2004).

Current studies by the author and research team have identified 18 cyanobacteria species in genera *Aphanothece, Aphanocapsa, Chroococcus, Johannesbaptistia, Microcystis, Synechococcus, Synechocystis, Merismopedia, Cyanotetras, Anabaena, Leptolyngbya, Lyngbya, Nostoc, Oscillatoria, Phormidium, Pseudanabaena, Spirulina* and *Komvophoron*, four green algae species in genera *Chlorella*, *Spirogyra*, *Closterium* and *Scenedesmus* and two diatoms in genera *Navicula* and *Stauroneis* in selected paddy soils from 25 locations in four major paddy growing areas, Anuradhapura, Polonnaruwa. Kurunegala, Matale and Gampaha (Amarawansa *et al.*, 2018, Balasooriya 2019). Therefore, further studies to identify freshwater and terrestrial microalgae from the paddy fields in other locations as well as from other crop fields are important prior to screen them for potential applications.

Potential Applications of Algae and Microalgae in Sri Lanka

Pioneer research studies on the antibiotic potential of seaweed species in Sri Lanka has been initiated in 1975 by Sachithananthan and Sivapalan. However, later studies on bioactive compounds were scattered (Mageswaran and Sivasubramanian, 1984; Lakmal et al., 2014) and no successful bioproducts have been developed. During 1960s, cultivation of seaweeds and extraction of alginic acid have been reported in the country (Arumugam *et al.*, 1981).

Cyanobacteria has been recognized as a major contributor for N fixation in paddy fields (Rodger and Kulasooriya, 1980). Current studies by the author and research team have developed a cyanobacteria based biofertilizer for paddy cultivation in Sri Lanka. Based on field experiments in several seasons and at many locations, this biofertilizer indicated a potential to replace the current recommendation of Urea fertilizers significantly (unpublished data).

Fernando *et al.* (2021) have shown potential of freshwater microalgae, *Haematococcus pluvialis* and *Chromochloris zofingiensis* for extracting astaxanthin which is used in pharmaceutical industry for treatment of Parkinson's and Alzheimer's diseases, and as an antioxidant in cancer treatment. Several researches have revealed the potential of freshwater microalgae species for biodiesel industry based on their high fatty acid content (Kalana *et al.*, 2016; Hossain et al., 2020).

Studies have also revealed the potential of microalgae for waste management. Marine algae Nannochloropsis has shown high potential in comparison with Chlorella and Spirulina sp, for removal of CO_2 in flu gas from a cement factory (Gunasena et al., 2019). Fernando *et al.* (2021) have coupled phycoremediation and Astaxanthin production by growing microalgae in Palm Oil Mil Effluent (POME). The results revealed moderate removal of Chemical Oxygen Demand, total Nitrogen and total Phosphorus in POME.

Our current research on phycoremediation using *Chlorella* species has shown significant potential for removal of Nitrates and Phosphates, whereas *Oscillatoria* species has shown potential for removal of Phosphates from agricultural, domestic and industrial wastewater (Balasooriya *et al.*, 2021).

Conclusions and Future Directions

Microalgae are abundant in both marine and freshwater ecosystems and terrestrial environments of Sri Lanka. However, microalgae species in most of the environments are not identified or fully studied. Systematic studies for sample collection, isolation and identification are very important as the foundation for marching towards microalgae based bioeconomy in Sri Lanka. The isolated pure cultures need to be preserved in a standard or national culture collection for future applications including genetic improvements. Many countries in the world have established national algae culture collections and standard culture collections by universities and research institutes. Microalgae research and development and education should be motivated at institutional level as well as at national level through the relevant polices and strategies.

Number of researches have shown the potential for effective applications of microalgae in Sri Lanka. Continuation of these projects on identification and application of microalgae for bioactive compounds, biofuel, biofertilizer and phycoremediation is highly important to expand the bio-based economy of the country. Further steps in utilization of microalgae may include optimization of culture conditions, control of contaminants, molecular identification and conservation and maintenance of pure cultures, product quality and toxicology analysis, waste management and economic assessment of the microalgae-based technologies in pilot scale experiments.

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Chapter

05

A step towards strengthening bioeconomy of Sri Lanka with two potential candidates:

finger millet; the crop of future and sorghum; the camel of crops

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Abstract

The transition from the conventional economic practices to bioeconomy is identified as a requisite for ensuring food and energy security for the increasing human population in a sustainable manner and maintaining ecological balance. Sri Lanka as a biodiversity hotspot as well as a developing country fits perfectly in the model of bioeconomy. Agriculture plays a major role in the economy of Sri Lanka and also in the bioeconomy. Increasing the productivity of the existing agricultural lands, rather than utilizing more land, would be the strategy to ensure food security. In achieving this goal, there is a need to cultivate the crops having the potential for high yields and the feasibility for crop improvement. The crop species, finger millet and sorghum are recognized as perfect candidates in this context. Finger millet is a highly nutritious crop with enormous health benefits and possessing the capacity for high yields with low input. Further, finger millet displays the potential for bioethanol production and to be used as fodder, qualifying to the title 'Crop of future'. Sorghum is mainly used as a staple food, livestock feed and fodder, biofuels and in the brewery industry. It is the cereal crop recording the highest levels of drought tolerance and is considered as the 'Camel of the crops'. As both crops are highly recommended to be used in the crop improvement programmes, the bioeconomy of Sri Lanka can be strengthened using these two crops. This chapter highlights the benefits of upgrading the two crops from 'underutilized' state to be of crops having great potential in marching towards bioeconomy.

Keywords: Agriculture, Finger millet, Sorghum, Underutilized cereals

Introduction to Bioeconomy

Bio economy has been in debate over the last decade as the major solution to address the issues in environmental changes, food and energy security, health and industries. It is defined as 'the knowledge-based production and utilization of biological resources, biological processes and principles to sustainably provide goods and services across all economic sectors' (Anonymous, 2018). This concept includes three facets namely; the economy, the environment, and the society. It is intended to reduce the dependence on natural resources, transform manufacturing, promote sustainable production of renewable resources and their conversion in to food, feed, and bio-based products which will in return upsurge different industries and employment opportunities while causing no negative impact on the sustainability of the system and the environment (Figure 01) (https://ec.europa.eu/programmes/horizon2020/en/h2020-section/bioeconomy).

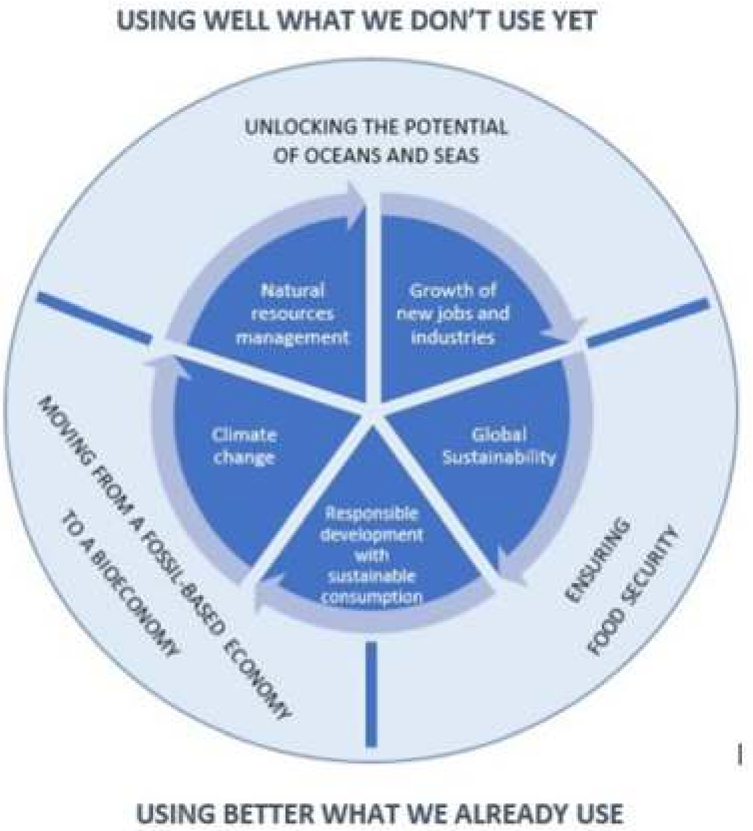

Figure 1: The components of bioeconomy.

As a country with extensive biodiversity yet susceptible of depleting it due to developmental activities, Sri Lanka is in need of a bioeconomy based concept to be followed to meet the developmental goals while maintaining sustainability and biodiversity. Agriculture plays a vital role in the practices of bioeconomy. Although the attention is always grabbed by the major cultivations like rice, coconut, rubber, tea and sugarcane, there are other cereal crops available in Sri Lanka with wide-ranging applications in the attempts to enhance bioeconomic practices in the country. The bioeconomy has already been adopted by a significant number of low and middle income countries as a new vision of development (Anonymous, 2018). As it is identified as a timely requirement under these circumstances, this chapter focuses on bringing up the potential of two underutilized crops finger millet and sorghum as potential candidates worth to be considered in the practices of bioeconomy in Sri Lanka.

1. Finger millet: the Crop of Future

Finger millet [*Eleusine coracana* (L.)] (2n=4x=36) is an annual crop cultivated in arid and semi-arid regions of the world predominantly in Africa and Asia (Figure 2). Origin of finger millet is believed to be around 5000 years ago in Ethiopian highlands followed by dissemination to African lowlands and from there to India approximately 3000 years ago via sea trade between Africa and India (Hilu and de Wet, 1976; Dida *et al.*, 2008). Finger millet cultivation in Sri Lanka dates back to ancient times and was the dominant cereal crop in Sri Lanka during early and middle historic periods between 500 and 300 B.C. (Panabokke, 2010). Finger millet cultivation in Sri Lanka is mainly linked with rainfed '*chena*' cultivation and irrigated cultivation can also be observed in variable extents all over the island. It is estimated that finger millet accounts for approximately 10% of the world's 30 million tons of total millet production (Dida *et al.*, 2008) and it ranks fourth in importance among millets after sorghum, pearl millet and foxtail millet (Upadhyaya *et al.*, 2007). Finger millet grain is processed in different methods and used to prepare a large array of food.

Figure 2: The wide array of ear types in finger millet.

Importance of Finger millet

Finger millet is a highly nutritious functional food with enormous health benefits due to which it is recognized as 'wonder crop, or 'super cereal' by the nutritionists. It contains about 5-8% protein, 65-75% carbohydrates, 15-20% dietary fiber and 2.5-3.5% minerals and finger millet contains the highest content of calcium (344 mg/100 g) among all cereals. Finger millet is enriched in the essential amino acids like lysine and methionine (Kumar *et al.,* 2016a). Further, finger millet contains phytates, polyphenols, tannins, trypsin inhibitory factors, and dietary fiber which are called nutraceuticals (Devi *et al.*, 2014). A nutraceutical is defined as any substance that is a food or part of a food and provides medical or health benefits, including the prevention and treatment of disease (Trottier *et al.*, 2010).

The high calcium content in finger millet is good for bone health; strengthening of bones and prevention the risk of diseases like osteoporosis and bone fracture. Finger millet possess the ability to prevent premature aging and metabolic diseases due to the presence of phytates, polyphenols and tannins that are responsible for antioxidant activity (Bravo, 1998) and offering several other health benefits such as anti-diabetes, hypo-cholesterolaemic, antimicrobial effects and protection from diet-related chronic diseases (Devi *et al.*, 2014). Polyphenols and dietary fiber in finger millet facilitate the slow digestion of carbohydrates (Kavitha and Prema, 1995) controlling the blood glucose levels making finger millet diabetes relief food. The high content of dietary fiber makes a feeling of fullness after consumption of finger millet and therefore it is excellent as a reduced snack (Thilakeratne, 2010). Antioxidants in finger millet assist in wound healing in diabetes patients by partially protecting the insulin producing cells from alloxan-mediated cell damage thereby promoting the healing process (Rajasekaran *et al.*, 2004). Finger millet is currently gaining popularity among Sri Lankan elderly population for the above mentioned qualities of this crop.

Finger millet as a potential candidate in strengthening the bioeconomy in Sri Lanka

Food security plays an important role in bioeconomy. Finger millet, with all the nutrients it contains, possesses a great potential towards strengthening of bioeconomy by ensuring food security. The high nutritional value of finger millet over most of the cereals and millets makes it a food security crop (Dida *et al.*, 2007). Finger millet itself is a balanced diet. Accordingly, the different types of food items prepared with finger millet, will be beneficial for the poor in obtaining a balanced diet, fighting malnutrition and will provide an economical solution for hunger. The remarkable properties of finger millet is highly useful to treat both of the opposite scenarios; undernutrition and obesity, which are comm among the children in Sri Lanka today. Beginning from recent past, pregnant and lactating mothers mostly depend on artificial nutrient supplements to fulfil their high demand of nutrition. This may not be affordable for everyone and also may not be healthy in the long-run. Finger millet can provide healthy and economical solutions for this issue. Currently, more people are changing their dietary habit to be vegetarian but facing health issues aroused by missing essential amino acids. Finger millet contains several essential amino acids hence can nurture the vegetarians. If finger millet is popularized highlighting the potential, the demand for the crop will mark an exponential growth and the rural farmers of finger millet will get benefitted. Developing different types of attractive food items or snacks with finger millet targeting above mentioned diverse groups of consumers rather than conventional '*kurakkan*' diets, would pave way for improving finger millet to an industry scale would create plenty of job opportunities. On the other hand, in a situation where importing finger millet has been restricted from the year 2020, the only way of fulfilling the demand is to improve cultivation of finger millet within the country. Further, finger millet grain can be stored for a long time; for years without getting damaged from insects or microbes due to its hardness which is a promising feature in terms postharvest quality and food security.

The Hippocrates' quote (400 BC) 'Let food be thy medicine and medicine be thy food' implies that the important properties of food can be utilized to stay healthy as well as in prevention and curing of non-communicable diseases. Given their health benefitting values, whole-grain cereals can be acclaimed as major nutraceutical candidates for human consumption (Kumar *et al*., 2016a). In case of finger millet, it is a promising nutraceutical candidate for prevention of cardiovascular diseases, cancer, aging, osteoporosis, anemia, obesity and for promoting blood glucose and cholesterol lowering. If awareness is built among the public on safety of nutraceuticals over conventional drugs it will open an enormous market which will in return develop a healthy nation. Finger millet-based nutraceuticals can be improved as an industry for supporting the bio-economy in a direct manner by raising job opportunities. The above mentioned non-communicable diseases accounts for more than 75% of annual deaths in Sri Lanka. In the long-run, the potential for exploitation of export market and saving of foreign exchange spent on drugs and treatments will mark a positive effect on the country's economy.

Finger millet can be grown in lands which are marginal to most of the other crops. It can withstand significant levels of salinity, resistant to water logging (Dida *et al.*, 2007) and resistant to the pests and for most of the diseases as well. It is beneficial to the farmers that is has a short growing season of three and a half to four months.

Finger millet is considered to be a crop needing less input to obtain maximum yield due to two different reasons. Firstly, Finger millet is less susceptible to diseases and affected by only a few diseases (Upadhyaya *et al.*, 2008) with birds and squirrels being the pests attacking the crop. Therefore, less effort or input are needed for disease and pest control of the crop to obtain the expected yield. Secondly, being an annual rain fed crop the agronomical sustainability of the crop makes it an ideal in dry farming as it requires medium rain-fall (average annual rain fall 500 mm - 1000 mm) and temperature of 18 - 27 °C. It can withstand a soil pH of 5.0 - 8.2 (Duke, 1978) ranging from moderate acidity to moderate alkalinity. Finger millet can withstand drought and significant levels of salinity and resistant to water logging. It can adopt to a wide range of environmental conditions and further can be cultivated at altitudes over 2000 m from the sea level and thrives at higher elevations than most other tropical cereals (Anon, 2012, https://www.doa.gov.lk). This makes finger millet requiring less inputs for growth yet offering optimum yields. Chandra *et al*. (2016) states that, being a hardy crop, it is relatively easy to grow finger millet under stressful regimes, without hampering the net productivity, Also, with increasing world population and decreasing water supplies finger millet represents an important crop for future human use, which reiterating the slogan 'crop of future' given for finger millet.

Finger millet provides sufficient amounts of straw for animal feeding which is of great value for backyard animal husbandry (Sood *et al.*, 2016). Using finger millet as fodder is beneficial in several ways. And also, it has a potential to be used as an alternative source of energy in poultry diets. Since finger millet is containing a high quantity of protein it may allow formulation of fodder without a supplementation of protein reducing the cost of feed for livestock production and consequently reduce the cost of livestock products for people who rely on it as source of protein (Hassan *et al.*, 2021). This can be counted as a benefit of finger millet in assuring food security in an indirect manner. After harvesting grains as the main product, the use of remaining straw for fodder fulfills the zero-waste concept of bioeconomy.

Greenhouse gases (GHG) emissions and fossil fuel depletion are currently viewed as the major challenges of global industrial development and bioethanol is considered at the moment as one of the most promising substitutes for petroleum (Aditiya *et al.*, 2016). Recent reports highlight the utilization of finger millet as a feedstock source for bioethanol production to be considerably effective as its tolerance to drought and the ability to grow under low nutrient input conditions (Yemets *et al.*, 2020).

Kumar *et al.* (2016a) affirm that, in the global scale, exploitation of the high nutritional value of finger millet would be important to provide food security, agricultural development, self-dependence and economic enhancement of developing countries (Figure 3). With agreement to this statement, it can be stated that, the significant attention finger millet has currently gained in Sri Lanka as an excellent nutrient and nutraceutical source and its potential as a crop with a less input, its ability to withstand abiotic and biotic stresses and its potential to be used as fodder as well as to produce bioethanol can lead Sri Lanka's bioeconomy.

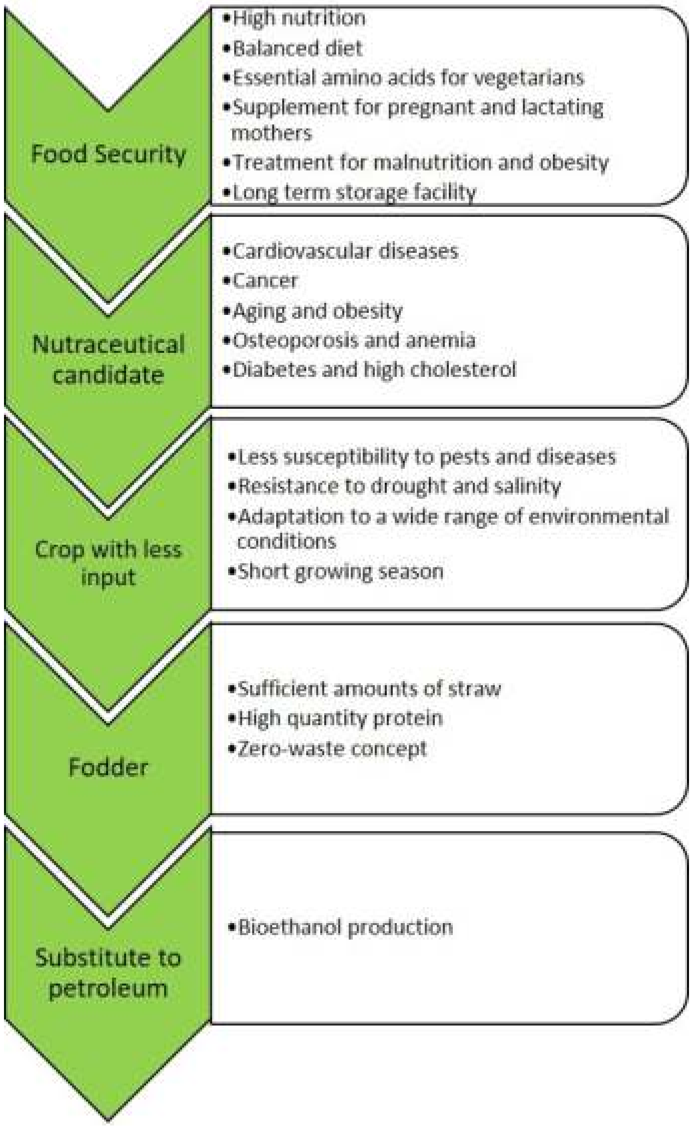

Figure 3: Finger millets for bioeconomy: A summary

2. Sorghum: the Camel of Crops

Sorghum (*Sorghum bicolor* (L.) Moench) which is belonging to the family *Poaceae* is an important cereal crop in many tropical and subtropical areas of the world (Figure 4). Sorghum is a self-pollinated diploid (2n=20) crop, but recording out-crossing in various frequencies. Sorghum had been domesticated in Ethiopia (Doggett, 1965; Etuk *et al*., 2012) and people who migrated into Ethiopia through the Middle East or through Arabia may have domesticated the wild sorghum which occurred as a weed in their wheat fields (Pursglove, 1972). It is considered as the fifth most important cereal crop grown worldwide on the basis of both production and area planted (FAO, 2004; Kumar *et al*., 2011). Sorghum cultivation spreads over 105 countries in the semi-arid areas of the world (Rakshit *et al*., 2014). Africa, Central America and South Asia are leading as per the production area of sorghum. More than one third of the world's sorghum production is fulfilled by the developed countries. In Asia, both India and China produce sorghum in massive-scales. More than 500 million people in more than 30 countries depend on sorghum as their dietary staple. This crop is mainly used as a staple food, livestock feed and fodder, biofuels and in the brewery industry (ICRISAT, 2009; Grains Council, 2010).

Figure 4: Panicle diversity of sorghum

Importance of sorghum

Sorghum has the ability to utilize water efficiently and it is the cereal crop with the best drought tolerance among the currently cultivated cereal crops. Therefore, it is considered as the “camel of the crops” in the world of crops. Among the C4 cereals, *Sorghum bicolor* is the most suited species to be cultivated in drought stressed environments and it is the most preferred cereal to be grown in areas where the rainfall is erratic and insufficient (Kudajie, 2005). Drought tolerance ability of sorghum is a consequence of morphological and anatomical characteristics; thick leaf wax and deep root system and physiological responses; osmotic adjustment, stay green, and quiescence (Dugas *et al*., 2011). Due to the high genetic variability among sorghum genotypes and the relatively small genome size, sorghum serves as an appropriate model for identification of drought related genomic regions and genes that are valuable to unravel the high complexity of drought tolerance related traits (Sanchez *et al*., 2002; Paterson *et al*., 2009).

Sorghum is well known for its nutritive and medicinal values. Polysaccharides, proteins, lipids, minerals and vitamins are the nutritive components of sorghum (Martino *et al*., 2012). Sorghum contains phosphorus, potassium and zinc depending on the place of cultivation (Martino *et al*., 2012; Shegro *et al.,* (2012) and several B-complex vitamins (thiamine, riboflavin and pyridoxine) and fat-soluble vitamins (D, E and K) (Ochanda *et al*., 2010; Martino *et al*., 2012; Cardoso *et al*., 2014).

Sorghum is a gluten free cereal and thus can be recommended for patients suffering from celiac disease (Taylor *et al*., 2006). Sorghum is believed to be useful in controlling diabetes as it is rich in dietary fiber and record low glycemic index (Park *et al*., 2012). Sorghum grain is rich in antioxidants and is highly suitable as an alternative for people with wheat allergy (Grains Council, 2010). A rare class of plant pigments known as 3-Deoxyanthoxyanins (3-DXA) can be found in sorghum (Yang *et al*., 2006) and it is believed to be useful in reducing the incidence of gastrointestinal cancer.

There are four classes of sorghum varieties based on the application including grain, sweet, fodder and high biomass types (Murray *et al*., 2009; Shakoor *et al*., 2014). Sorghum grain is processed into a wide variety of food; such as baked products, tortillas, couscous, gruel, steam-cooked products semi-leavened breads, popped form, fermented or non-fermented porridges and alcoholic or non-alcoholic beverages (Anglani, 1998). Sweet sorghum has a sugary stalk and it is well known for plant-based bioethanol production (Disasa *et al*., 2018). Sweet sorghum is characterized by the synthesis of sugar during flowering stage and translocating the synthesized sugar into seeds during seed filling stage and further, it is be considered as a natural variant of grain sorghum (Rao and Kumar, 2013). Sweet sorghum produces high biomass under adverse conditions and therefore it is highly recommended as a raw material for second generation bioethanol production and lignocellulosic feedstocks in temperate zone (Smith and Buxton, 1993; Windpassinger *et al*., 2015). Renewable fuels can be produced using sorghum as well (Kisgeci *et al*., 1983). Stamenkovic *et al*. (2020) reported that there is a potential to produce different kinds of bio fuels using sorghum biomasses in parallel to utilizing as a food and nutritional security crop. Not only the grain, but also the leaves provide fodder for farm animals and the stalks are used in fencing, roofing, weaving baskets, brooms, mats and also as fuel wood (Bantilan *et al*., 2004).

Sorghum as a potential candidate in strengthening the bioeconomy of Sri Lanka

Sorghum conquers high demand in agriculture-based bio economic trends in Sri Lanka. Sorghum is locally called as '*Idal Iringu*' and it is believed that sorghum was consumed by the ancient Sri Lankans, but it is yet to be proven by sufficient archeological evidences. As per the records, sorghum had been introduced to Sri Lanka some decades ago and the highest sorghum production of 5,586 tons was recorded in 1975 (Factfish, 2016). However, sorghum failed to conquer much demand in Sri Lanka, due to a number of reasons including lack of availability of seed sorghum. Currently, the popularity of sorghum is less in the local market and high in the international market. Kaluthanthri and Dasanayaka (2019a) outlined the production and use of sorghum and its adaptability to Sri

Lanka in a review.

Methodical enhancement of the production of cereal crops offers a number of ways to address the combined societal challenges including food insecurity, natural resource insufficiency, fossil-fuel dependence and climate change acceleration. In this scenario, sorghum displays multipurpose flexibility and becomes promising for bioeconomy. Due to the high drought tolerant ability of sorghum, it is highly suitable for the hot, dry and arid areas of Sri Lanka. Drought is the most important abiotic stress in terms of limiting crop productivity worldwide (Fracasso *et al*., 2016). Drought tolerance is the ability of a plant to produce its components of economic value with minimum loss in drought prone environments. The C_4 photosynthetic pathway of sorghum makes it even more suitable for use as a component in bioeconomy, due to capability of C_4 plants to absorb ambient atmospheric carbon dioxide and transform into biomass. Furthermore, greenhouse gas emission can be reduced by the replacement of fossil energy with the biomass derived renewable energy. Production of different kinds of bio-fuels using sorghum biomasses in parallel to utilizing as a food facilitates opportunities for rural smallholder farmers to have an additional income as it is adapted to low agronomic inputs which are characteristic of smallholder farming areas (Mengistu *et al*., 2016). Sorghum can be considered as a suitable candidate for fuel production due to high yield of biomass, low input requirements, adaptation to constraints characteristic in marginal lands, and rich genomic resources (Regassa and Wortmann, 2014). Also, sorghum biomass, grains and sugary juice derived from the stem can be used in bioethanol production (Wang *et al*., 2008).

The three major scope of sorghum in Sri Lanka; elevated agro-ecological value, low input and drought tolerance and farmers expertise and large genetic diversity, it can be used to enhance the food production under climate change and to develop novel green economy outputs (Figure 5). The genetic diversity of currently available sorghum germplasm in Sri Lanka, is a valuable measure in utilizing sorghum as a crop in the bioeconomy approaches. In the present day, there are number of sorghum varieties cultivated by local farmers, namely: *Idal iringu*, *Karal Iringu*, *Poth Iringu*, *Sudu Thiringu*, *Rathu Thiringu*, *Rata Kurakkan*, *Karalliya*, *Waguru*, etc. Currently, sweet sorghum varieties are less available for both small-scale and large-scale cultivations in Sri Lanka.

Evidently, sorghum varietal development is a crucial need for bioeconomy of the country. This could be an opportunity and a threat as well. As an opportunity, varietal development facilitates a path for yield enhancement, nutrient improvement, additional income generation and diversification. On the other hand, erosion of agrobiodiversity can occur due to preferences of limited number of varieties with specific qualities leading to the neglect of traditional and non-patented varieties. The genetic diversity can be considered as a reservoir for both numerous cultivation practices and bioeconomy. Therefore, the plant breeders need to carefully select local varieties based on the specific populations, preferences and uses as well. Bioeconomy practices in Sri Lanka should not be totally market driven. It should be strengthened towards agrobiodiversity preservation.

Agricultural practices utilizing inorganic fertilizers and pesticides cause negative environmental effects. Therefore, promoting the organic farming trends is vitally important especially in developing countries. However, when cultivations using inorganic agricultural inputs over a long period of time is to be converted organic farming practices, use of cereal crops that can be grown in adverse conditions is highly recommended. In Sri Lanka, sorghum is highly feasible for organic farming practices as the camel of crops. This can be cultivated during both '*Yala*' and '*Maha*' seasons in Sri Lanka with low levels of fertilizer input a requirement of supplementary irrigation depending on the rainfall.

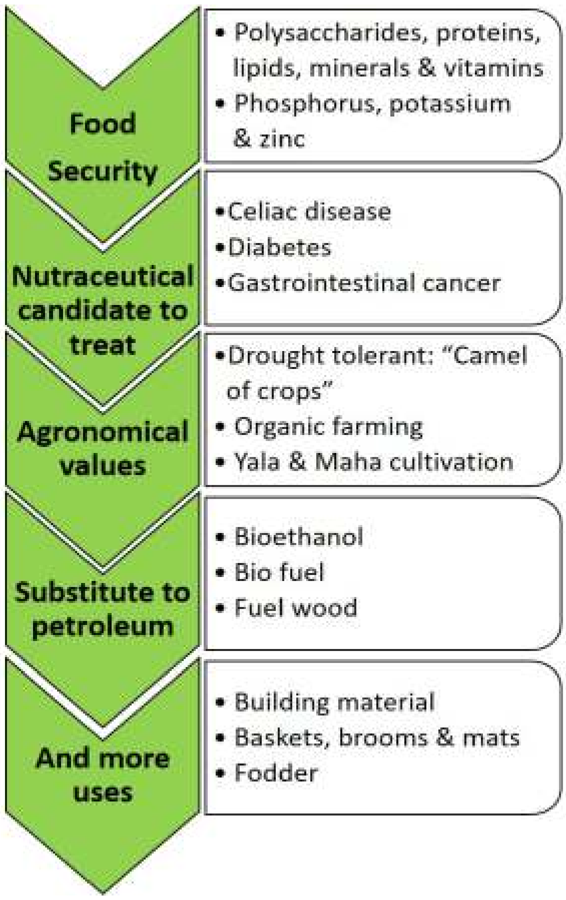

Figure 5: A summary: sorghum towards bioeconomy.

Conclusions and Future Directions

Research and development of agricultural components of bioeconomy is fundamental in achieving the goals of bioeconomy. Increasing the area of cultivation is no longer a feasibility and thus the efforts should be concentrated on maximizing the productivity of crops. Development of novel improved crop varieties pave the way for achieving maximum yields amidst other desirable characteristics. Accordingly, crop improvement is a compulsion for finger millet and sorghum for them to be better valued in the bioeconomy of Sri Lanka. Crop improvement involves the identification of desirable traits, most importantly yield related characteristics in crop plants and this is achieved by germplasm characterization and evaluation. Making the germplasm available, identification and selection of elite germplasm involves five main steps; germplasm collection, conservation, characterization, evaluation and utilization. There is a rich collection of finger millet and sorghum germplasm accessions including both local and exotic germplasm collections conserved at Plant Genetic Resource Center, Gannoruwa and several important studies have been undertaken to reveal a gamut of important information that can be utilized in genetic improvement (Wakista, 2018; Kaluthathanthri, 2021). Such studies have shed light on information related to selection of elite germplasm for cultivation and core accessions for conservation and utilization, identification of potential candidates to be used as parents in hybridization, and the research efforts need to be strengthened for them to fit better in the bioeconomy in Sri Lanka.

In addition to crop improvement research, it is vital to generate knowledge, processes and practices to promote the two crops for to realize their true potential. Such knowledge should include agronomy, processing and value addition and market promotion etc. which will enhance the value of the two important crops, finger millet and sorghum, which in turn will enrich the bioeconomy of Sri Lanka.

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Chapter

06

Is Sesame a Potential Crop that Contributes to the Bioeconomy of Sri Lanka?

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Abstract

Sesame (*Sesamum indicum* L.), often regarded as the "Queen of oil seed crops", is the oldest oil seed crop mainly grown in the tropical and subtropical countries. Sesame seed oil (SSO) is of high-grade quality and has been cultivated for its multiple benefits since ancient times. Increased awareness on the medicinal, nutritional, beauty-cultural, and industrial applications of sesame has grown the international demand which in turn has increased the production in the sesame growing countries and expanded the international trading. Despite the continuous expansion in the global market, currently, Sri Lankan sesame remains as an orphan crop. Lack of high-yielding varieties, limited accessibility to developed varieties, resources, guidance, and facilities of transport and storage, seed shattering nature of the available varieties, changing weather patterns, unsteady market prices, and less awareness of the farmers on the market opportunities are the key factors limiting sesame cultivation in Sri Lanka. Limited product range and awareness on the numerous benefits of SSO, occasional unavailability of seeds, allergies reactions, taste of SSO have downturn the consumption of sesame. Sri Lanka where more than two third of the land experience dry climate, naturally has the ideal environment for cultivation of sesame, the only oil seed crop being exported from the country, yet with sporadic export volumes. Along with raised awareness among farmers and consumers, among many other factors, opportunities to generate numerous economic activities (expanded market opportunities, increased product range and customer base, introduced value-added products, modern economic uses such as harnessing biodiesel) may encourage both cultivation and trading while contributing to bioeconomy of the country. Overall, Sri Lanka inherits an excellent prospective for expanding sesame cultivation and improve the level of production.

Keywords: Agricultural benefits, Biodiesel from SSO, Food values, Medicinal properties, Sri Lankan sesame

Introduction

Sesame (*Sesamum indicum* L., Family Pedaliaceae), is an ancient oil seed crop that constitutes high-grade oil and hence, is more often regarded as the "Queen of oil seed crops". Compared to other oil seed crops, sesame seeds contain a greater oil content (Eskandari *et al.*, 2015). Oil and protein contents in the sesame seeds are reported to vary from 37% to 63% and 17% to 32%, respectively. Unsaturated fatty acids make up to 80% of sesame oil (Hegde, 2012) and its quality index varies from 83% to 87% (Wei et al., 2015). For edible oils, the quality index is calculated as the ratio of unsaturated fatty acids to saturated fatty acids. The oleic acid (30% to 53%) and linoleic acid (33% to 52%) are the key unsaturated fatty acids present in the sesame oil (Wei et al., 2015). Nutritionally, sesame seeds are enriched with minerals (i.e. iron, calcium, phosphorus) and vitamins (i.e. thiamine, vitamin A, riboflavin) (Weiss, 2000). Further, sesame seeds contain various antioxidants such as sesamolin, sesamol and sesamin (Anilakumar *et al*., 2010) providing stability and adding significant medicinal value to sesame seed oil (SSO).

Sesame seeds, treasured as the currency in the ancient world (Weiss, 2000), are presently used as a popular ingredient in the confectionary industry. Seeds, born in dehiscent capsules, are small and ovate in shape with smooth or rough seed coat. The pigment composition in the seed coat is responsible for creating white, yellow, brown, red and black colour seeds. Raw seeds are pressed to obtain cooking oil recognized as a good source of unsaturated fatty acids, antioxidants and proteins. Both seeds and oil serve multiple purposes including culinary, medicinal and beauty cultural uses (Adu-Gyamfi *et al*., 2019; Anastasi *et al*., 2017; Elleuch *et al*., 2007; Hegde, 2012; Pathak *et al*., 2014; Ross, 2007; Weiss, 2000; Were *et al*., 2006).

Owing to its high antioxidant content and the ability to improve the skin fairness and healthiness, sesame oil is an essential ingredient in cosmetics (Table 1). Sesame seeds are immensely beneficial even after they have been pressed to obtain oil and hence, oil cakes, the residue seed hulls, are used to feed livestock. The crude protein content in the sesame meal ranges from 47.1% to 52.9% and in terms of the amino acid composition of the protein, it is similar to that of soybean meal (El-Saidy, Mahmoud and Tonsey, 2009).

Table 1: Uses of sesame and its bioactive components.

Uses	Bioactive components
Nutraceutical	
Antioxidant and providing hepatoprotection	Lecithin Myristic acid
Cancer preventive	Fiber, Sesame oil
Tumor prevention, cardio protective	Sesamin, Sesamolin
Antioxidant property	Lecithin, Lignans
Inhibition of cholesterol production	Sesame oil
Skin softener	
Pharmaceutical	
Drug vehicle and laxative	Sesame oil
Hypoglycemic activity	Flavonoids
Inhibition of malignant melanoma	Linoleate in triglyceride form
Industrial	
Antibacterial mouthwash	Sesame oil
Antifungal properties	Chlorosesamone
Bactericidal and insecticidal properties	Sesamin, Sesamolin Myristic acid
Cosmetics	Sesame oil
Biodiesel	
Traditional	
Intestine lubrication	Sesame oil
Constipation	Sesamin
Intestinal worms	Sesamin, Sesamolin

Source: Anilakumar *et al.*, (2010)

Apart from its food condiment values, sesame oil brings about diverse health benefits and generates wide pharmaceutical applications. The oil has been applied for healing wounds for many thousand years. The seeds and fresh leaves are used as a poultice. The SSO maintains good cholesterol [High-Density Lipoprotein (HDL)] and lowers bad cholesterol [Low-Density Lipoprotein (LDL)] (Sirato-Yasumoto *et al*., 2001). The antioxidant sesamin reduces LDL levels while HDL levels are increased. Sesamin is anti-hypertensive and anti-inflammatory (Ide *et al*., 2003). It enhances the recovery of vitamin E, functioning of liver and provides protection against alcohol induced oxidative stress. The SSO is slightly laxative and holds moisturising and soothing characteristics. It is naturally antibacterial for common skin pathogens as well as common skin fungi such as athlete's foot fungus (Kaliyamoorthy *et al*., 2015). The SSO, extracted from high-grade seeds and more purified than normal edible oil, is used in the preparation of a number of pharmaceuticals and cosmetics, perfumes, soaps, paints and insecticides. The important antimicrobial properties of sesame have been explored in the production of antiseptics, bactericides, viricides, disinfectants, in repellants and anti-tubercular agents (Nyongesa *et al*., 2014).

Since recently, sesame has been explored to extract biodiesel from its seed oil through transesterification with methanol (Ahmad *et al*., 2010; Mujtaba *et al*., 2020). Compared to mineral diesel, combustion of sesame-based biodiesel is shown to be environmentally friendly and hence, the extraction of biodiesel from sesame seeds is a sustainable alternative to the traditional mineral diesel (Khan *et al*., 2019).

Sesame, the oldest of the oil seed crops, is mainly grown in the tropics and subtropics of the world. It has been cultivating in the Asian region for more than 5,000 years (Bisht *et al.*, 1998). Even though it was originally believed that sesame has a polytopic origin (Nayar and Mehra, 1970), with the latest scientific data it is evident that sesame is originated in Indian region (Bedigian, 2003; 2004; & 2015). Sesame cultivation has then been expanded to many African countries including Sudan, Uganda, Nigeria and Ethiopia, and Asian countries including India, China, Myanmar, Pakistan and Bangladesh. Among them, China, India, Myanmar, Sudan, Ethiopia, Tanzania, and Nigeria are the biggest producers in the world (Anastasi et al., 2017). According to Food and Agriculture Organization (FAO) (2019), the global sesame production in 2017 was over 5.5 million tonnes, of which more than half (57 %) was produced in Africa while Asia accounts for 40%. Nearly 5% of world's sesame production is produced by South American countries such as Mexico, Guatemala and Venezuela. The world's largest exporters of sesame seeds, oil and oil cakes are India, Ethiopia, Sudan, China, Myanmar and Mexico (FAO, 2019) while the US, China, UK, Japan, Australia, Turkey, Korea, Netherlands, Belgium, Indonesia, Malaysia and Singapore are the main export destinations of sesame and sesame products in the world (FAO, 2019). Analysis of market data shows that global annual demand for sesame is following a positive trend due to the increased awareness on medicinal values among consumers. The price of sesame is also continuously rising despite some recent fluctuations. As per the future projections, the global consumption of sesame is forecasted to reach 9.3 million tonnes by 2040 (Rahman, c. 2019). Accordingly, US$ 17.8 worth of growth is expected in the international market by the year 2025 compared to the market value of US$ 6.5 billion estimated in 2018.

Current Status of Cultivation and Consumption of Sesame in Sri Lanka

Sesame, commonly known as gingelly in Sri Lanka, is the main oil crop cultivated in Sri Lanka (Weeraratna and Weerasinghe, 2009). Among Sri Lankan flora, *Sesamum* is reported to be one of the two genera recorded in Sri Lanka belonging to the family Pedaliaceae. *S. indicum, S. radiatum* and *S. prostrate* are three different species of the genus *Sesamum* (Theobalt and Grupe, 1981)*. S. radiatum* and *S. prostrate* are the wild sesame varieties. The most economically valuable species, *S. indicum,* was introduced into the dry areas of Sri Lanka where it is being currently cultivated. MI, *Uma, Malee* and ANKSE3 are the varieties developed by the Department of Agriculture (DOA) in Sri Lanka regardless of which the landraces (*Idal, Pokuru* etc) are famous among sesame farmers. Further, Sri Lanka has a collection of over 70 accessions of sesame in the gene bank at the Plant Genetic Resources Centre (PGRC), Gannoruwa.

According to the cultivation data, sesame is grown at varying degrees in 16 (during *Maha* season) - 19 (during *Yala* season) number of districts in the country (Department of Census and Statistics, 2021) out of which nearly14 are categorized under the dry or arid zones according to the climatic conditions. It has been widely grown in the districts of Anuradhapura, Monaragala, Kurunegala, Hambantota, Mullaitivu, Vavunia, Puttalam, Jaffna, and Mannar where the temperature settings are best suited for the growth of the plant. Sesame cultivation requires an annual rainfall of about 625-1100 mm and a temperature varying between 25°C and 37°C during the cultivation to obtain maximum harvest (Langman, 2008). In addition, medium texture fertile soil is the most suitable for the cultivation. Sesame is cultivated in about 11,872 ha of which 8,886 ha is cultivated during *Yala* season under rain-fed condition, whilst the balance is cultivated during *Maha* season in 2018 (Socio Economic and Planning Centre, 2019).

The national total production and the extent of cultivation sesame in Sri Lanka for the period from 2001 to 2020 are shown in Figure 1. From 2001 to 2008, the values remain nearly constant and after 2009 when the values dropped drastically, a downward trend in the values are observed.

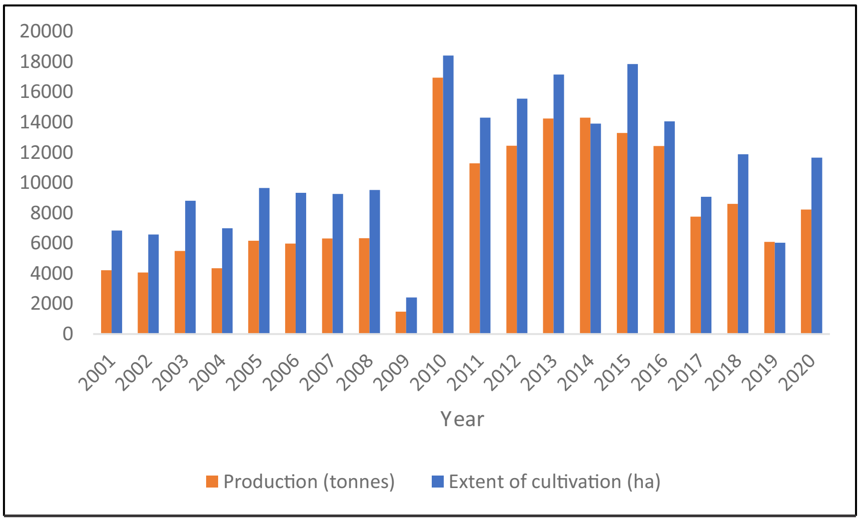

Figure 1: Total cultivated area (ha) and annual production (tonnes) of sesame in Sri Lanka (Source: Department of Census and Statistics, 2021).

A farmer survey (n=251) carried out in selected sesame growing districts (Anuradhapura, Hambanthota, Ampara, Mannar, Puttalam) in Sri Lanka revealed that sesame is cultivated mainly as a secondary crop in the marginal lands with minimum agricultural inputs (Dissanayake *et al*., 2017). It was widely grown in *Yala* season as a secondary crop to maintain the agricultural lands for primary crops grown during *Maha* season. Many farmers (nearly 80%) who have entered sesame cultivation since recently (<5 years) while few farmers have been with sesame cultivation for many decades. In general, sesame was cultivated by smallholders (<5 ha) and the production reported to be less than 1 tonnes/ ha in all study areas. A recent questionnaire-based farmer survey (n=44) carried out in Anuradhapura district where the highest annual sesame production is reported, showed that the number of farmers cultivating sesame as a commercial crop was 37% (Jaythilaka and Dissanayake, 2020).

Sesame is cultivated in Sri Lanka mainly to obtain seeds and to extract edible oil. Baked seeds, confectionaries made from seeds including gingelly rolls, sweetened gingelly balls, crackers, *halva* and *dodol* and bakery foods containing sprinkled seeds are quite popular among Sri Lankans. A survey to collect the information on consumer preferences, awareness about important properties and consumption of sesame, was carried out based on a questionnaire using convenience sampling method (Jaythilaka and Dissanayake, 2020). The consumer survey (n=126) revealed that 2.4% of respondents have never tasted food products of sesame. Among the respondents who have consumed sesame foods, the higher consumer preference (45.6%) was observed for sesame-based bakery products. Consumption of sesame oil was very rare (3.2%) among the respondents. Less awareness of medicinal and beauty cultural applications of sesame was also observed among the sampled individuals and the majority (55.6%) seemed to use these products very rarely. Even though various degrees of use of sesame oil from traditional massages to the treatments in the modern day has been published in the literature, limited modern applications of sesame oil i.e. in the preparation of cosmetics and bath oils, were reported during the survey. Use of stalks, seeds, leaves and oils of sesame for treating the diseases and conditions including dysentery, diarrhoea, catarrh, haemorrhoids, gonorrhoea, coughs, burns, constipation, head lice, general pains, nervous system disorders, ulcers, cystitis, urinary bladder disorders, wounds, otorrhoea and strangury are reported in the indigenous and ayurvedic medicinal practices in Sri Lanka (Jayaweera, 1982; Kodikara, 1994; Rajapaksha, 1998).

Sesame is the only oilseed crop exported from Sri Lanka (Socio Economic and Planning Centre, 2020). Analysis of export data shows that Sri Lanka has been exporting sesame 20 years before it started depending on imported sesame (Figure 02 & 03). Export of sesame seeds has started in 1970 after which it had been increasingly exported during that decade to reach a peak around 1982. Since then, the amounts dropped and started to fluctuate notably after 2004 in a positive direction in general [Figure 2 (a)]. Commencement of export of oil is marked in mid-eighties since when an upward trend was observed till 2003 after which the amounts dropped slightly across the last two decades [Figure 2 (b)].

Sri Lanka started to import sesame seeds and oil continuously since 1995 (Figure 3) and import of seeds fluctuated till 2010 since when the country tends to increasingly depend on imports [Figure 3 (a)]. Currently, imports of oil show a decreasing trend [Figure 3 (b)] while the annual imports of seeds and export volumes move in a positive direction.

In the light of the present significant rise in the international market for sesame seeds as well as projected increasing demand in future, these data suggest that there is an opportunity for Sri Lankan sesame to expand its cultivation to meet the local needs as well as to export the production surplus as it had occurred in early years. It was also mentioned in the vision statements of the former and current governments that the country should target zero sesame imports by the year 2020 by improving sesame production in the country.

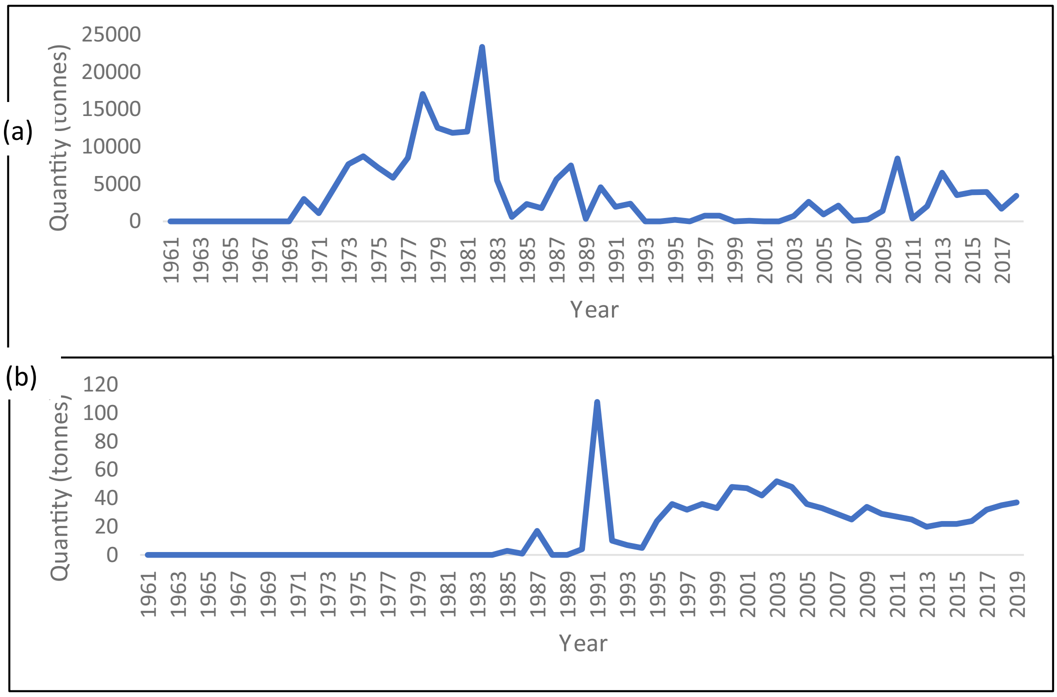

Figure 2: Export volumes (tonnes) of sesame seeds (a) oil (b) from 1961 to 2019 (Source: FAO, 2021),

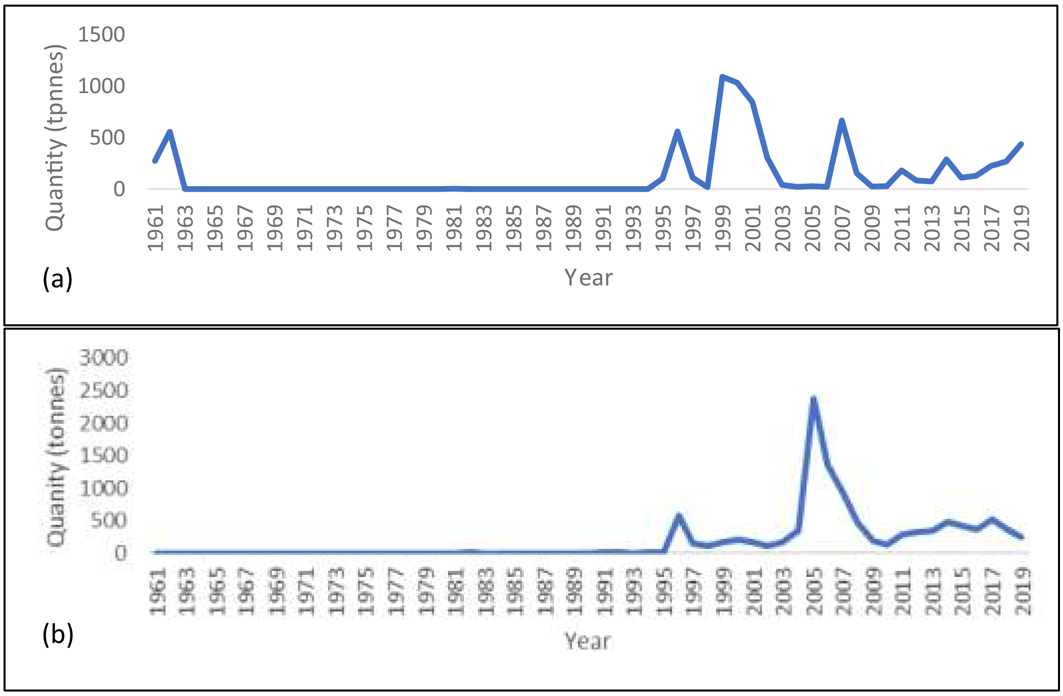

Figure 3: Import volumes (tonnes) of sesame seeds (a) oil (b) from 1961 to 2019 (Source: FAO, 2021).

Issues Related to Cultivation and Consumption of Sesame in Sri Lanka

Farmer surveys have showed that sesame is occasionally grown as a commercial crop in Sri Lanka. Many farmers were not aware of the sesame cultivars released by the DOA (Dissanayake *et al*., 2017). Little over 80% of the respondents of the farmer survey conducted in Anuradhapura have not even heard about those varieties (Jaythilaka and Dissanayake, 2020). None of them had grown improved varieties. Usually, white seeded sesame cultivation was prominent in Hambanthota district while black seeded sesame cultivation was popular among the farmers in Puttalam and

Mannar districts (Dissanayake *et al*., 2017). Easy accessibility to the seeds has apparently been the cultivar selection criteria by these farmers. The farmers had received limited resources (seeds and fertilizers) and advice on sesame cultivation practices. Application of agrochemicals (fertilizers, insecticides, weedicides) during cultivation was merely recorded or often used only during land preparation. Inaccessibly to high yielding cultivars, early seed shattering of the available varieties, environmental effects including prolong drought conditions, change of rainfall patterns and animal attacks limit the expansion of sesame cultivation in Sri Lanka. The low yield of the cultivation ultimately leads to a low income and hence the growers are not much attracted to expand the cultivation to a larger scale. The involvement of the middlemen, low farmgate prices, unsteadiness of the local market, and lack of storage and transport facilities also generate low income from sesame cultivation. The surveys indicated that sesame remains an orphan crop in the country as it has been continued to cultivate in marginal lands utilizing minimum human resources, and attention.

The consumer survey revealed a very few consumers (1.6%) with allergies to sesame. From the consumer perspective, some (15%) of the consumers have not preferred the taste of sesame. As far as the awareness of respondents about the important properties of sesame is concerned, many (45%) were unaware of the importance of sesame. Findings from the consumer survey suggested limited consumption of sesame among the consumers.

Sesame Related Scientific Research in Sri Lanka

As identified by Dossa *et al*. in 2017, the history of sesame related research in the world can be comprehended under three eras: "germplasm collection and gene bank constitution", "classical breeding and genetics", and "Omics" era (Figure 4).

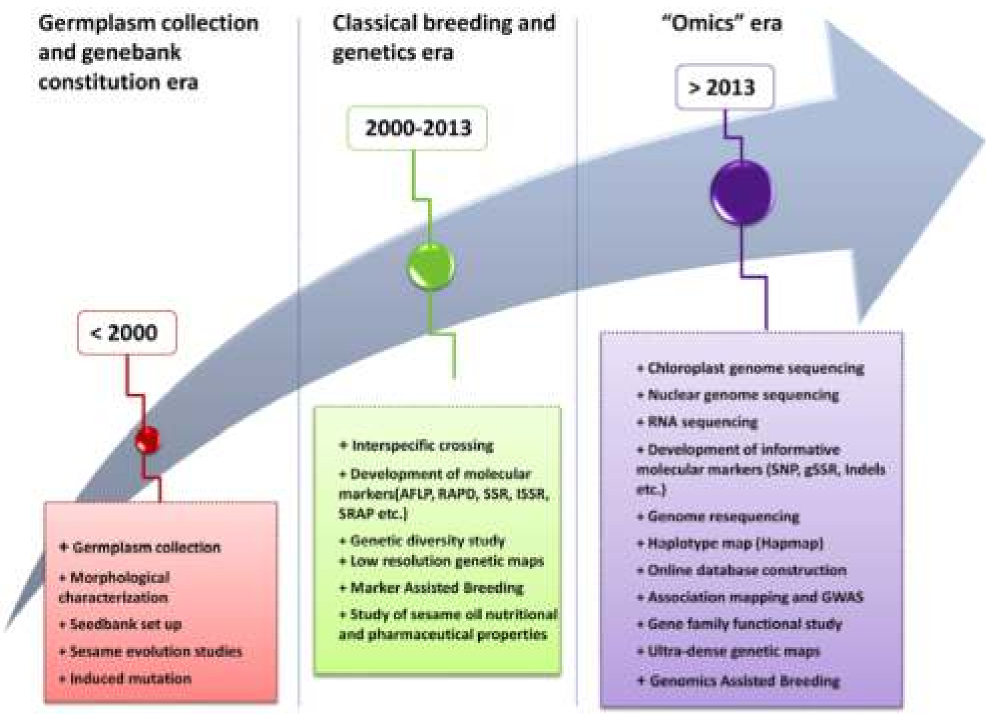

Figure 4: Evolutionary history of the scientific research on sesame

(Source: Dossa *et al*., 2017).

Morphological characterization of cultivated and wild relatives of sesame, installation of seedbanks, confirmation of the origin of centres and domestication process are considered as the key events in first period of the research history. During the second era, application of classical and molecular marker assisted breeding and analysis of diversity and oil quality were attempted. Many of the sesame growing countries are now following the era of "Omics" when much more focus was drawn to further improve the crop with the availability of transcriptomic data and genomic resources (Dossa *et al*., 2017).

Compared to other sesame growing countries, scientific research on Sri Lankan sesame is scanty. However, numerous researches on sesame are available during 1990s, the period considered as the golden era in relation to sesame cultivation in Sri Lanka. For instance, optimization of callus induction from sesame anthers by Ranaweera & Pathirana (1992) and generation of gamma ray-induced mutants by Pathirana (1992) and Pathirana *et al*., (2000). Field trials are regularly carried out at Grain Legumes & Oil Crops Research & Development Centre (GLOCRD), Angunakolapelessa in order to determine the best sowing dates of the selected cultivars. In determining the extent of genetic diversity with respect to geographical origins, some international studies have included some accessions of Sri Lankan sesame (Ali *et al*., 2007). Few attempts are reported on diversity analysis: on morphological diversity among ecotypes by Dissanayake *et al*. (2015) and genetic diversity to select parental lines for breeding programmes by Perera & Pushpakumara (2014). In addition, drought responses of selected sesame cultivars have been revealed by Dissanayake *et al*. (2016) and Dissanayake *et al*. (2019a). Further attempts were resulted with preliminary yield and growth models of sesame by Dissanayake *et al*. (2019b) and Dissanayake *et al*. (2019c).

Still there is room for further research in pacing with 'Omics' era such as developing tolerant varieties against abiotic and biotic stresses, non-shattering varieties to allow mechanical harvesting, improving taste, innovating non-allergy products, deriving biodiesel, exploring applications of crop residues, harnessing soil conditioning ability and developing forecasting models for yield.

Future Opportunities for Sesame Cultivation

An upward trend is noted in the world sesame production over the last 50 years. Despite the growth in supply, the price trend has followed a similar positive growth. Demand for sesame in top importing countries also show a gradually rising trend. Among the sesame importing countries, China holds the largest share of international trade. For instance, China has imported around 1 million tonnes from various countries in 2016, accounting for nearly 64% of the global sesame imports (Rahman, 2019c). Similarly, Japan remains as an export destination of sesame from several exporting countries. The import market of Korea is dominated by the sesame producing Asian countries. These largest sesame importing countries could be prime destinations for Sri Lankan grown sesame. There are vast opportunities for high quality raw seeds in the global market. The growing inclination in global demand, production, and unit price, will generate opportunities to increase local production, manufacture sesame-based products and develop novel value-added products. Compared to raw sesame seeds, value-added products are economically more profitable owing to its high price in the global market. Development of multiple industries (e.g. confectionary, condiment, cosmetic, and livestock feed) based on sesame seeds and oil could directly foster expansion of sesame cultivation. This attempt will generate subsistence for farmers, create opportunities for employments and alleviate poverty in rural areas (Myint *et al*., 2020; Wacal *et al*., 2021).

In the international market, black sesame attracts higher price compared to that of white seeds with a price variation ranging from 40% to 45% (Rahman, 2019c). Similarly, hulled seeds also have a greater price by about 10% to 15% than the unhulled seeds (Rahman, c. 2019). These facts suggest the need for the improvements of local sesame production towards the popularization of black seeded sesame cultivation and the use of appropriate processing methods for hulling raw seeds in creating a niche in the international market.

Organic farming is currently trending in the world. A greater potential for extension of sesame cultivation as 'organic by default' on degraded dry lands in Sri Lanka was revealed through the farmer surveys as less or no agro-chemical usage was observed through the farmer interviews. The international organic market has grown by an annual rate of 50 % (Olowe et al., 2009). African and Asian countries such as Nicaragua, Peru, Turkey, Uganda, Mexico, China and El Salvador have already entered into the global organic sesame market (Augstburger et al., 2002). Main sources of organic fertilizers applied by these countries included plant extracts, bone meal, compost manure, and green manure. In addition, bee-keeping in the vicinity of cultivation increases sesame yield by encouraging cross-pollination (Augstburger et al., 2002). Further, crop rotation alternatively with legumes promotes growth of soil mycorrhiza and biological methods including the use of plant extracts are recommended to deter disease causing agents and pests in sesame cultivations (Augstburger et al., 2002). Good agronomic practices such as dressing seeds with hot water, timely seed sowing, diseased plants disposal, and selection of disease resistant varieties are also recommended to prevent diseases. Similarly, farmers in Sri Lanka have the ability to transit from 'organic by default' practices to 'certified organic farming' and enter the international organic market. Use of recommended varieties may further increase the yield ensuring a continuous supply.

Sesame is highly responsive to many abiotic factors and as per the research evidence, drought effects and responses of sesame are genotype specific (Bahrami et al., 2012; Boureima et al., 2011; Hassanzadeh et al., 2009; Kadkhodaie et al., 2014). Similarly, studies done by Dissanayake *et al*. (2016, 2019) in Sri Lanka on drought responses also suggest that sesame becomes tolerant to drought stress and develops adaptations to thrive dry weather. Many districts in Sri Lanka belong to intermediate and dry zones and hence, create appropriate climatic conditions required for sesame cultivation. Although mature sesame plants are drought-tolerant, yield penalties may result under severe water deficit conditions, hence, supplementary irrigation is required at seed setting and flowering to maximize yield and optimize oil quality. Subject to the sesame cultivar and developmental stages exposed to water deficit conditions, sesame expresses various morphological adaptations to thrive drought period and include excessive root growth, high root to shoot ratio, reduced leaf area and plant stature, early maturity and high plasticity (Dissanayake *et al*., 2016, 2019). These traits will be important in screening for tolerant cultivars or developing tolerant cultivars for the subsequent breeding programmes. Drought tolerant mechanisms such as morphological and physiological adaptations are also well established for the cultivars grown in many other sesame producing countries (Bahrami et al., 2012; Boureima et al., 2011; Hassanzadeh et al., 2009; Kadkhodaie et al., 2014). Further research on similar aspects is needed for understanding such responses of the Sri Lankan sesame. For instance, establishing the exact drought tolerant mechanisms (osmoregulation, increased photosynthesis etc) expressed by Sri Lankan sesame is important. For instance, qRT-PCR to examine if genes for specific osmolytes (e.g. proline) are highly expressed in leaves under osmoregulation. Similarly, use of effective molecular markers (e.g. random amplified polymorphic DNA) will be valuable for characterizing sesame cultivars in addition to the morphological analysis.

According to the farmer surveys, major causes for the limited cultivation in Sri Lanka were the low yield and pod dehiscence. The level of shattering is a key characteristic of sesame varieties as it decreases harvest and causes yield losses during harvest limiting receipt of maximum value from the available seeds (Dissanayake *et al*., 2017). Achieving delayed shattering will require breeding of new non-dehiscent varieties and allows shifting to mechanical harvesting. Further, it requires optimizing of harvesting equipment, and tactical agronomic techniques such as the use of pod sealants to control seed loss from capsules.

During the surveys, the farmers suggested that the introduction of new varieties and subsequent proper distribution of seed material will encourage farmers to continue with sesame cultivation and attract new farmers. Availability of high yielding tolerant varieties with determinate growth habit and delayed shattering will make sesame a more attractive crop. Enhanced sesame production will financially benefit the sesame farming rural communities and elevate the standards of living. Increased production may lessen the need to import and reduce the cost of annual importing which follows an increasing trend (FAO, 2021).

The government has a major role to play in expanding sesame cultivation in the country. The government program of cultivating 15 additional field crops including sesame under '*Saubahgya*' program in 2020 was implemented to assure a promising price and a purchase mechanism for sesame produce. Under this program, the government expects to purchase a kilo of seeds for a price of 200 rupees. The Agricultural and Agrarian Insurance Board (AAIB) is also prepared to provide an insurance scheme for cultivating these additional crops. The release of water from the Maduru Oya and Pimburattewa reservoirs has encouraged the cultivation in *Yala* season in the Polonnaruwa district. Similarly, in other areas under sesame cultivation can also be supplied with sufficient amount of irrigated water when required. Famers should be trained to access agronomic information and instructions on sesame cultivation from the DOA, Sri Lanka. Parallelly with the promotion programmes for developing other crops such as rice, promotion programmes can be implemented to encourage sesame farmers and provide seeds, instructions, and incentives. Further, lessons learnt from other sesame growing countries can be adopted for better production within Sri Lanka. For instance, a government initiative to facilitate transition towards technological interventions including improved agronomical practices and post-harvest techniques such as cleaning, grading, and packaging, has led to boost in sesame production in Tanzania. The farmers have been trained to use smartphone applications in accessing information on agronomy and business support in Tanzania (African farming and food processing, 2020). In response, the country developed into the largest sesame producer in the world in 2017. Similarly, raising the awareness of sesame growers with respect to proper agronomic, post-harvest, and processing will be an encouragement for the Sri Lankan sesame growers. In addition, an introduction of technology such as sensors to detect soil nutrient availability, physiological state of the plants, disease occurrence in cultivation, and proper harvesting time, development of weather forecasting systems, and the supplement of agronomy and business support information through smartphone technologies, will attract young farmers and be influential in expanding the cultivation in Sri Lanka.

Farmers demanded for a steady local market for the sesame produce. At present, farmers are unable to fetch a reasonable price corresponding with the cost of sesame production. Therefore, direct intervention of the government in manipulating the market is essential to deter the influence of the middlemen and in turn, ensure a guaranteed farmgate price for unit of production. Segregated transport along with proper storage is needed by the sesame industry. If industry opt for widening the product range and pursue value-added products, new facilities for processing, packaging, and marketing will also be required. Manual harvesting which is the only practice in Sri Lanka, is inefficient and sesame production is generally labour-intensive. Therefore, the introduction of mechanized harvesting could minimize this cost and the risk allowing the producers to fetch a competitive price in the export markets and assure a longer return on the investment.

Sesame is a great soil constructor, retains and improves moisture levels sufficient for the next crop and reduces soil blow in eroded lands (Langham et al., 2008). This shows the opportunity for exploration of the possibility of transforming into a zero-waste cultivation by converting agricultural wastes of sesame into a soil conditioner that can be promoted through a organic farming. After harvesting, the stumps of the plants are retained in the cultivated lands with no use.

In addition, opting for biofuel production with low grade sesame oil can be seen as a new production line supporting the country in moving towards ecofriendly transportation.

Variability in rainfall pattern, land degradation, excessive water loss due to evapotranspiration and low water holding capacities limit the suitability of land use for cultivation of many other crops in the dry areas of Sri Lanka. Therefore, sesame is suggested as a suitable crop for cultivating in the marginal lands in the dry areas, as it has proven that sesame thrive well without much of agricultural inputs and survive water limited conditions up to some extent. Overall, Sri Lanka inherits an excellent prospective for expanding sesame cultivation and improve the level of production.

Consumption of sesame oil is popular in Asia. However, as far as the status of consumption of sesame is concerned, there was less or no frequent consumption of sesame-based products in Sri Lanka. Not much awareness on the important values of sesame was reflected by the consumers. In addition, there is less variability in the range of sesame-based products in the market. The consumer survey revealed that the social awareness about the beneficial aspects of sesame, introduction of novel and value-added products, opening of the product accessibility and improving the sesame taste, could greatly enhance the consumption. Increased awareness on assorted values of sesame in the society may widen the consumer base and stabilize the local sesame market.

According to the Food Allergy Research and Education Institute (FARE) in the US, sesame is the ninth most common cause of food allergy reported among children and adults (Food & Nutrition, 2021). An anaphylaxis reaction occurs during consumption leading to a series of symptoms such as rashes and low level of blood pressure. Fortunately, only low number of cases was evident in terms of sesame allergies through the consumer survey in Sri Lanka and so does the other Asian countries (Jaythilaka and Dissanayake, 2020).

Food innovations such as sesame milk and instant *Tahini,* a dip prepared from toasted sesame, have led to the growing demand for sesame in the world. In addition, the high-grade sesame meal has multiple uses including making sesame flour for baking purposes and preparing tahini and sesame cake. The meal with lower grade is excellent as feed for poultry and livestock. Ground sesame seed is used in soup in the African cuisine, sweets in the Middle East cuisine and in various curries in the Indian cuisine. Use of roasted black sesame as meal dressing including salad, rice, or meat, is popular in countries such as China, Japan, Taiwan and Korea. Black Sesame ice cream is a novel product now available in many countries (Bhat *et al*., 2014). Such value-added foods can be promoted in Sri Lanka and specially at the local Food Stalls established at the district level by the DOA.

Conclusion

This chapter reviews the uses, cultivation and future potential of sesame as an oil seed crop in the Sri Lankan context. Sesame possesses many important nutritional, medicinal, antioxidant and antimicrobial properties and offers many agricultural benefits. In spite of its high international demand and climate suitability to expand sesame cultivation in Sri Lanka, sesame remains an underutilized crop. Sri Lanka exhibits great potential to develop sesame cultivation, be self-sufficient in sesame production and popularize the consumption of sesame. Proper awareness of both growers and consumers would be an ideal action to improve the sesame cultivation and consumption in Sri Lanka. Despite the raising awareness on market opportunities and global demand growth, the introduction of improved varieties, proper distribution of seed material for growing, shifting to eco-friendly and organic farming, establishment of a stable market, loan facilities, insurance schemes, transport and storage facilities, use of smart technology and creating new business ventures would promote sesame cultivation in Sri Lanka. Along with raising awareness on the values of sesame, innovation of value-added food products will promote sesame consumption. In addition, exploration for avenues for producing biodiesel and soil builder will also encourage sesame cultivation. Further, Scientific research in improving sesame cultivars and discovering new applications from sesame needs to be continued.

As future direction, it is suggested conducting a thorough cost benefit analysis of sesame farming, systematic market survey along with SWOT analysis and quantify the real potential of sesame to be part of the bioeconomy of the country.

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Chapter

07

Hydroelectricity as a Sustainable Energy Source:
Are We Maintaining the Ecological Regimes?

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Abstract

There are growing concerns whether the world has adequate energy resources to support economic development and the increased global climate change due to fossil fuel consumption. The concept of 'Bio-economy' was introduced to address these concerns. Bio-economy bridges economic development and sustainability. It facilitates economic growth while promoting sustainable development. Hydro-power can be considered one of the most promising long-term sustainable energy sources in the world. Similarly in Sri Lanka, 40% of the energy is generated from hydro-power. Thus, major Sri Lankan rivers are heavily exploited and the natural flows are altered by dams threatening riverine ecosystems. Powerful stakeholders justify water needs for human consumption while neglecting ecosystem-water-needs in the decision-making process. Demand for water for human needs and ecosystems often overlap with each other. This not only threatens the quality, but also the quantity of the water in the rivers. Safeguarding ecosystems when managing waters to meet human demands is a must. Humans benefited from hydroelectricity as a sustainable energy source towards the economic development of the country, but this is not the case for ecosystems – the silent water user. Ecosystems are frequently omitted from water allocation decision-making. It is accepted that maintaining a set of flows which termed as ecological regimes or environmental flows and are beneficial in social-economical and environmental aspects. Therefore, this chapter discusses the harvesting hydro-power while maintaining ecological regimes, by giving especial focus to Sri Lanka.

Keywords: Ecological Regimes, Environmental Flows, Environmental Policy, Hydro-power, Sustainable Energy

Global Economy and Renewable Energy

In the context of the global economy, energy plays a major role as it is a basic need of society to ensure the quality of life. As a source of energy, fossil fuel is dominant in many parts of the world. In the total primary energy supply, fossil fuels account for 81 %, nuclear energy represents 5%, and the renewable energy sources 14 % (Popp *et al*., 2020). On the other hand, the type of energy that is used directly affects the quality of the environment, especially the degradation of the atmosphere (Sibuea *et al*., 2012). Further, over the last number of years, there has been an increasing fear among the scientific community and decision-makers around the world about the adverse impacts of copious consumption of energy from fossil fuels (Alvarado *et al*., 2021). For example, it is estimated that energy-associated levels of carbon dioxide emissions are projected to rise by 61% from 2014 to 2025 in the ASEAN (Association of Southeast Asian Nations) region. Thus, the significance of the transition towards renewable energy was highlighted by the UN under the 7th sustainable development goal "affordable and clean energy". As per the UN SDG (United Nations Sustainable Development Goals), there are encouraging signs of the world moving towards sustainable energy sources (UN SDG, 2021)

Renewable Energy

Progression towards renewable energy is imperative for de-carbonising the world economy and alleviating global climate change. Compared to conventional energy sources such as fossil fuel, renewable energy technologies provide considerable advantages to future energy systems and environmental sustainability (Levenda *et al.*, 2021). Solar, wind, hydro-power, nuclear power, biomass, and geothermal energy are renewable energy sources. Unlike fossil fuels, renewable energy resources are also unevenly distributed throughout the globe. Fortunately, almost every continent in the world is blessed with at least one type of renewable resource. Thus, almost all countries have energy security at a certain point, without having to depend on a foreign energy source. Depending entirely on foreign energy resources may create political instability or manipulation, which can be seen in many developing countries (Bull, 2001). At the global level, 15 % of primary energy consumption is renewable energy and the bulk is hydro-power and wood fuel in developing countries. Solar and wind share a diminutive percentage compared to total (Lund, 2007) consumption. However, moving forward, renewable energy has many challenges. Demand for the energy to be used as electricity is one major challenge while the other is its utility-driven demand in the transportation sector (Lund, 2007). With progressive anxiety on the adverse effects of global warming, most of the countries have endorsed the vision of sustainable development goals to use renewable sources of energy and economic growth (Tiwari *et al*., 2020). Therefore more focus and attention is essential for strengthening the usage and popularization of renewable energy sources and to expand the use of renewable energy beyond the electricity sector (UN SDG).

Hydroelectricity - A Sustainable Energy Source

The "fuel" of electricity generated from fossil fuels can vary from coal, oil, and to natural gas. Similarly, water is the fuel of hydroelectricity. It is clean, renewable, and cheap, and identified as the world's most rapidly expanding major fuel (Bermann, 2007; SLSEA, 2021). Initially, the power of moving water was found in ancient Greece 2,000 years ago; they used water power to run wheels for grinding grain. Hydro-power is technically mature, economically combative and accounts for 18% of the world's total electricity supply . World statistics show that there is a steady increase (around 3% annually) in hydroelectric power generation (Edenhofer, 2011). Hydroelectricity plants reduce the necessity and the demand for thermal power plants. There is substantial spatial heterogeneity among hydroelectricity in the world. For example, the capacities range from 76.6% in Brazil, to 18% in China, and 62 % in Canada. China, Brazil, Canada, and the United States are the world's largest hydro-power producers accounting for more than half of the world's hydroelectricity (EPI, 2021). As per the statistics, developing nations have reached about 23% of their financially sustainable hydro-power projects compared to developed nations (developed nations exploited 70% of their capacities) (Zhou, 2015).

Hydroelectricity generation is supported by the hydrological cycle which provides endless and continuous recharging of fuel (water). To generate hydro-power, rivers or other waterways were altered by dams or diversions. The simple physics behind hydroelectricity is the potential energy of stored water that is converted to the kinetic energy of flowing water as it flows downstream. The amount of energy generated depends on both the volume of the water flow and the elevation change. There are three types of hydroelectricity plants, namely impoundment (the most comment one, stores water using a dam), a diversion or a “run-of-river” (uses the kinetic energy of natural down-flow water), and pumped storage (generates electricity while sending water through turbines from a upper reservoir to lower reservoir and pumps water back to upper reservoir).

Sri Lankan Status

Sri Lanka is blessed with freshwater resources and there are around 103 major and minor rivers radially draining into the sea. Therefore, generating electricity from hydro-power is considered the most fitting method for Sri Lanka. Currently, around 40% of Sri Lanka's energy requirement is fulfilled by hydro-power. History of hydro-power in Sri Lanka dates back to 1912. The first-ever hydro-power system was a small-scale hydroelectric power plant at Black Pool which was inaugurated at the Nuwara Eliya Electricity Scheme. Further, in 1918, Engineer D. J. Wimalasurendra identified the potential of hydro-power in Sri Lanka. In 1950, the Laxapana Power Station started to generate electricity and was accompanied by around 20 hydro-power plants. Until 1990, Sri Lanka solely depended on hydro-power. Currently, Sri Lanka has a 1719 MW electricity generation capacity and is envisioned for 247 MW to be added to the electricity grid in the coming years (CEB, 2021; ADB, 2019).

One major drawback of hydro-power is the difficulty in developing hydro-power plants in all possible locations due to demographic and geographical reasons. Thus, the Sri Lankan government started to promote mini hydro-power projects (Producing hydro-power at a small scale for eg. for industry plant or small community) to get the maximum out of water resources. In addition, it is considered that mini hydro-power plants are more environmentally friendly than large-scale plants (Rupasinghe & De Silva, 2007). British planters were the first to build mini-hydro-power plants from small streams. As per the records, until the 20th century, there had been around 500 mini-hydro-power plants (Fernando, 2002).

The government encourages private sector entrepreneurs to develop mini hydro-power plants, but there is no proper mechanism to track efficiencies of these plants, especially with the growing impacts of climate change. Climate change is heavily affecting hydro-power generation as the temperature and rainfall variations in most parts of Sri Lanka have become prominent in the recent past. Even though climate change and related repercussions are acknowledged by the authorities, there is an apparent niche in research on forecasting power generation with changing rare climate factors such as floods and droughts. It is essential to identify the patterns and consequences of hydro-power generation related to climate change. Most Sri Lankan hydro-power plants were built in the early fifties and even though the life of a hydro-power plant is around 50 years, in case one major plant stops functioning due to reduced water capacity (due to climate change) or mechanical failure, the country may undergo a dearth in power and present the government with an economic crisis by forcing the country to import fossil fuel.

Advantages and Disadvantages of Hydro-power

Hydro-power is produced through the hydrological cycle that is driven by the sun. It has no greenhouse gas emissions, and thus, makes it a clean source of energy. The development of reservoirs promotes inland fishing, irrigation water especially for seasonal crops, recreational activities, flood control, etc. Hydro-power is considered a domestic source of energy. (i.e. generates energy locally). Especially on a regional scale, mini-hydro-power projects provides electricity on a small scale for villages; even for individual houses. Hydro-power not only provides electricity but also provides clean drinking water. In many regions around the world, there are water purification plans along with reservoirs. Hydroelectricity is considered a flexible power source as many plants can rabidly move from zero to ceiling capacity. This makes hydro-power a backup power system, where it can Provide power directly to the main grid. Hydro-power can be coupled with other renewable resources as well. For instance, in pumped and storage hydro-power, the energy for pumping is received from solar or wind power. Lastly, it is affordable, compared to other conventional energy sources (Bartle, 2002).

Despite of ample advantages of hydro-power, there are disadvantages as well. One main constrain is, due to demographic and geographical impediments most of the waterways and the locations are not suitable and safe to create dams. Besides, hydro-power generation is carbon-free, in the damming process water stagnation and decaying of vegetation and other biological components emit huge quantities of methane and carbon dioxide. Also, despite the relatively cheap power generation and low operation cost, construction of the dam is quite expensive, and therefore, the initial cost of hydroelectricity plant generation is similar to the construction cost of equal-sized fossil fuel plants. Additionally, although without a doubt, hydro-power has shown great potential as a renewable energy source, the production of energy also greatly relies on climatic and weather conditions such as droughts. There are many examples around the world including Sri Lanka of long-lasting blackouts due to the low capacities of reservoirs stemming from droughts. Finally, even though the dams are constructed to be strongholds, there is a risk of the failure due reasons such as earthquakes, extreme rainfall, and failures to open the gates. The focus of this chapter is, however, the disruption of the riverine ecosystem, especially the non-maintenance of sufficient water in the river channel (environmental flows) and the consequences on not only the in-channel ecosystems, but also faunal and floral health in the river floodplains due to diversion of water as a result of construction of dams.

Hydro-power and Maintaining Ecological Regimes

Rivers and Their Flow Patterns

Rivers can be considered as one of the most extensively used natural resources. They facilitated great civilizations for hundreds of years, and in addition to fulfilling the thirst, rivers played a significant role in agriculture, transportation, recreation, flood control, and most importantly sustaining the ecosystem (Aheeyar *et al*., 2008).

Natural freshwater ecosystems are replenished by precipitation and lost via evaporation, sub-surface seepage, percolation and discharge to the ocean. Rivers are not only just waterways that carry water and support human needs and development activities, but they also can be considered as a very complex network that serves the ecosystem and support biodiversity. Specifically, rivers interact with their catchments by aggregating runoff that is comprised of sediments (nutrients and organic matter) and propagating them downstream (Arthington and Pusey, 2003). However, along with the economic development of countries, the rivers have been heavily affected both in quality and quantity.

The health of a riverine ecosystem depends on the flow, physical structures and riparian zone, quality of the water, exploitation level, and other physical barriers to the connectivity. The quality of the water, basically the level of pollution, riparian zones, sediments, food supply, and biotic interactions have been vehemently discussed in the literature. However, in general, the quantity of the water and physical barriers have been neglected (Poff, & Matthews, 2013). River flow determines the habitat availability for aquatic organisms. Organisms evolve life-history strategies in accordance with the natural flows. Longitudinal and lateral connectivity of waterways is critical for the viability of these populations. The distribution and success of invasive/exotic species also depend on river flows.

Ecological Regimes

Ecological regimes or the environmental flows are defined as 'the quantity, timing, and quality of water flows required to sustain freshwater and estuarine ecosystems and the human livelihoods and well-being that depend on these ecosystems (Arthington *et al*., 2018). Ecological regimes maintain a balance between economic development and ecological integrity of waterways. Even the World Conservation Union (IUCN) endorses the fact that natural flows are crucial to supporting the riverine ecosystems. Maintaining low flow levels reduces groundwater recharge. Further, if there are no high pulses or floods then downstream regions will be lacking sediments and debris which are essential components of agriculture (Dyson *et al.,* 2003). Maintaining ecological regimes is not only important for the reverie ecosystems and aquatic organisms, but also for human communities who utilise the water.

Initial environmental flow assessments were based on the idea that maintaining at least the minimum flow throughout the year may fulfill the required condition. The hypothesis researchers had at that time was the health of the river is based on the low flow levels. Thus, until you maintain the flow levels at a minimum or just above average, the riverine ecosystems were thought to be preserved. Incremental development of science later on has proven that entire flow regimes including high pulses, medium, and low pulses are vital for ecosystem sustainability (Suwal, 2020).

Hydrological Alterations

Any artificiality-built obstacle that interrupts the magnitude or timing of natural flows can be considered as hydrological alteration. Rivers are continuously altered due to dams, weirs, and other impediments. The most common hydrological alteration is the construction of dams to build hydroelectricity plants. It is widely accepted that dam construction, related diversion, and/or stocking of water are some of the biggest threats to riverine health. Thus, ecological regimes play a major role in catchment management and initial environmental impact assessments related to construction of waterways. In general, all related stakeholders accept there is a need to preserve the ecosystems while disturbing the natural waterways for development projects. However, when it comes to economic development and political agendas stakeholders with power justify their needs and get projects approved . This makes the ecosystems, the silent water users, frequently omitted from water allocation decision-making (Korsgaard, 2006).

Environmental Flow Assessment in Sri Lanka

Sri Lanka's power sector is heavily dependent on hydro-power. Therefore, waterways in Sri Lanka are increasingly exploited, particularly for dams built for hydro-power plants. Most rivers in Sri Lanka are blocked to construct dams and the flows are significantly altered. Even though estimating ecological regimes are considered an integral component of environmental impact assessments of river basin development projects, regrettably, the assessment of ecological regimes is not yet fully incorporated. There is a shortage of studies related to ecological regimes in Sri Lanka. In most cases, the annual flow or the minimum flow was considered as the environmental flow (Smakhtin and Weragala, 2005; Bellanthudawa, 2021).

In the literature, there are very few studies carried out in Sri Lanka related to environmental flows. Dissanayake *et al*. (2010) can be considered as the first-ever study related to environmental flows in Sri Lanka. The study was conducted based on Walawe river. Halwatura and Najim (2014) researched the Attanagalu Oya ('Oya' is akin to a mid-order stream) catchment to determine the environmental flows and revealed most of the flow regimes are not maintained due to water extractions. Munasinghe *et al*. (2021) assessed the environmental flows for Gurugoda Oya by combining the study with benthic macroinvertebrate communities, where the results highlighted asignificant variation among the zoobenthic organisms above and below the weir.

Hydro-power and Maintaining Ecological Regimes

Hydro-power generation affects the ecological regimes in four different ways. 1. Total loss of flow, 2. Altered flow regime, 3. Changes to connectivity and 4. Inter-basin transfer. These factors can be seen all together or individually, in each case. It might depend on the geographical location, design of the dam, the catchment characteristics and the operating system of the plant. Thus, it is advisable to design the projects by minimising the possible impacts on the ecosystem while optimising the productivity of the plant (World Bank Group, 2018).

Sri Lanka experienced widespread development after the 1950s and hydro-power was the focal point. Evaluation of the hydrorological regimes while constructing the plants was the least aspect considered during this era. Thus, most of the Sri Lankan rivers that are altered due to the hydro-power plants do not meet ecological regimes. For example, Mahaweli, Menik, Deduru Oya, and Malwatu Oya are extensively affected by hydro-power and irrigation-related projects (Eriyagama *et al*., 2016). Implementing conservation policies for affected rivers would be more complicated as the rivers are affected in different ways such as quality of water/quantity of water. Therefore, it is more realistic to apply ecological regimes for proposed projects as it would be more effective and straightforward. The concept of ecological regimes should be introduced in to or an Initial Environmental Examination (IEE) and Environmental Impact Assessments (EIA), where hydrologists and ecologists can work together to maintain a healthy ecosystem while optimally using the hydrological potential of flowing water. As per the National Environmental Act of Sri Lanka, it is mandatory to conduct an IEE/EIA to get approval for major irrigation or river basin development projects. However, national guidelines on implementing environmental flow requirements have not yet been stipulated (Eriyagama *et al.*, 2016), and instead, project proponents use a set of discharges such as minimum flow or mean annual flow. Therefore, a properly defined set of ecological regimes should be incorporated into the IEE and EIA guidelines and should be integrated into the national policies in Sri Lanka. Eriyagama *et al*. (2016) also emphasize the importance of incorporating ecological regimes to the national policies as developed countries have done since 1994. Further, they have presented some examples of how Sri Lanka manages environmental flows in major river basin projects. According to the findings, it is clear that maintaining ecological regimes in those projects lack the scientific aspects of ecological flows. In the Mahaweli Development Program, the condition for the flows was that 'the minimum discharge of the Mahaweli River should not drop below the historical minimum. Further, in the Weheragala diversion, nearly 50% of its flow was diverted to Kirindi Oya with the condition to maintain a minimum flow (of 1.5 m3 /s) river through the Yala National Park.

Assessing Ecological Regimes

There are two different approaches (bottom-up and top-down) that researchers used in the past decade to assess environmental flows. In the bottom-up approach, only the most essential components of environmental flows will be identified and assessed. In the top-down, it is the entire ecosystem and the flows will be identified as important, and the flow regimes that are not critical can be considered to be modified for ancillary usages (e.g. irrigation, power generation). As evident from above, the significance of identifying or measuring the ecological flow regimes has also been initiated as management actions. There are two management responses based on ecological flows. The first one, proactive responses try to maintain flows as much as possible natural status or provide some sort of conservation to natural flows. The reactive response is for the already modified rivers. In reactive management, it tries to restore the possible altered flows (Arthington and Pusey, 2003).

One main constraint in assessing ecological regimes is that assessments require daily flow data of rivers which are not available in most of the cases. Similar to other developing countries in the world, daility flow data is scarce in Sri Lanka too. Lack of data availability prompts hydrologists to simulate the flows in selected rivers. The simulation process requires diverse modeling approaches and the complexity of the hydrological regimes makes the analysis more intricate. Therefore, it is of timely importance to include ecological regimes in national policies.

Recommendation

The project proponents always prefer to follow simplified guidelines for scientific approaches to assess the impact on the environment. Therefore, it is recommended to develop guidelines and methods that are easy to follow by the project proponents. However, these guidelines also have to be incorporated into National Environmental Policies, making them legislatively secure for authoritaties conducting such assessments and viability of projects. Further, the integration of an entire group of stakeholders is vital in planning basin management projects. For example, in the process of assessing ecological regimes, not only the Ceylon Electricity Board must be consulted, but also the Department of Irrigation, Department of Wildlife Conservation, Department of Forestry and the Ministry of the Environment should be drawn in. Lastly, it is important to create awareness about existing environmental flow assessments methods. When it comes to environmental flow assessment in Sri Lanka, there are a number of methods. One widely used method in Sri Lanka is the Range of Variability approach (developed by Richter *et al*., 1996), of which the application to the Sri Lankan context is explained in detail by Halwatura and Najim (2014). Supplementarily, Eriyagama *et al*., (2016) developed a tool (Sri Lanka Environmental Flow Calculator (SLEFC)) to provide a rapid estimation of environmental flows at known locations on major rivers of the country. This would be a great step forward in ecological analysis in Sri Lanka. In many cases, there is a lack of scientists with ground-level experience and skills to guide the assessments. In some instances, science expertise may not get the necessary legislative support which consequently results in lack of keenness of such scientists to do conduct assessments. Therefore, it is timely to be aware and advocate for the public and relevant stakeholders about the latest concepts of ecological regimes. Further, it is recommended to have the Avoid, Minimize, Restore, offset approach concerning hydro-power generation and maintaining ecological flows. Avoid the threats to the ecosystem by careful planning design and operation. Minimize the effect on the basin ecosystem and organisms. Restore where necessary by using conventional methods. Offset residual impacts by suitable actions World Bank Group (2018).

Conclusion

Bio-economy not only applies to diverse sectors of primary production but also ecosystems services. Further, it seeks the pathways to replace fossil fuels in energy production by encouraging sustainable energy sources. Hydro-power is the dominant source of sustainable energy worldwide.

However, some negligence in power plant construction may threaten the ecosystems by altering the flow regimes. The balance that has to be mainlined in ecological regimes is that it has to comfort human demands while sustaining the ecosystem needs. The more the ecological regimes are closer to the natural flows, the healthier the riverine ecosystem is. Sri Lanka, a country heavily dependent on hydro-power also faces a similar situation of not maintaining ecological regimes. Thus, it is recommended to have the Avoid, Minimize, Restore, offset approach. By adhering to these practices Sri Lanka can get the optimum use of hydro-power for the betterment of the country while conserving the biodiversity.

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