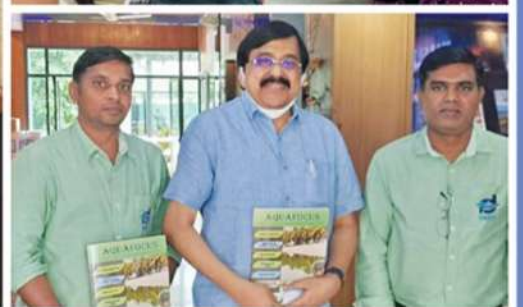


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“Fishing in the right pond is better than fishing in the wrong river.”

**- Canadian Philosopher
Matshona Dhlwayo**

(Dr. Jesu Arockiaraj, highly acclaimed Eminent Scientist with Stanford University World Ranking in Top 2%. Has also received many Awards including Young Scientist Award by Government of India and Government of Tamilnadu)

Greetings!

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In this issue, FAITT brings you well-curated, discerning and insightful articles in the Aquaculture trade and operations by eminent academics and research scientists. With a vibrant and growing Aquaculture industry, India is well poised to build a strong aquaculture industry for many decades to come.

Rainbow Trout is a well adaptable species that grows fast and can easily be cultivated in captive environments. Rainbow Trout Fisheries is gaining a lot of traction in tropical countries and the US and we present you with an insight on the current trends on Rainbow Trout Fisheries in South India.

In Tamilnadu, Aquaculture insurance products launched on World Fisheries Day, November 2, for freshwater fish farming and shrimp farming by the Tamil Nadu Dr. J. Jayalalithaa Fisheries University, Nagapattinam. In August, Honourble Tamilnadu CM, Shri M K Stalin inaugurated various facilities constructed at a cost of Rs 43 crore on behalf of the State Fisheries and Fishermen Welfare Department

Finally, it is great to look forward for a robust year 2023 ahead, as the scintillating year 2022 - a UN designated “International Year of Artisanal Fisheries and Aquaculture” come to a close.

Time to begin to change the future of Aquaculture now!

Regards,
Editors



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Introduction

The aquaculture sector has shown the fastest growth amongst all the various global food production industries, with a record high global production of 114.5 tonnes in live weight in 2018 valued at USD 263.6 billion (2018) (FAO, 2020). It is one of the main driving sectors for sustainable development goals 14 (SDG14) that emphasizes conservation and sustainable use of aquatic resources for sustainable development.

However, the aquaculture industry is still faced with serious problems related to nutritional health such as poor growth and survival in hatchery and grow-out ponds, vulnerability to diseases, and poor production. Nutrition is one of the major determining factors in the success of the aquaculture industry, as it is closely related to the growth, survival, and health of cultured organisms. Feedform the highest portion of the production cost and the use of fish meal in aquaculture is one of the unsustainable practices with a high carbon footprint. Poor quality feeds do not only result in low fish production but are associated with a high waste generation that could cause deteriorating environmental and fish health. Thus, it is very important to improve aquaculture nutrition using environmentally friendly, and highly nutritious feed that could increase precision feeding resulting in maximum production with minimal wastes. One of the approaches to enhance nutrition quality in aquaculture is to use microalgae as feed supplements. Microalgal cells in general are highly nutritious containing biomolecules associated with fast growth and high immunity such as vitamins, antioxidants, and anti-inflammatory, in addition to basic food components such as proteins, carbohydrates, lipids, and trace elements.

The accumulation of carotenoids in fish can also benefit human health which can satisfy the emerging markets of healthy functional foods. In addition, a microalgae culture that consumes CO₂ and nutrients improves the environment by decreasing wastes, thus preventing eutrophication in water bodies, and helping to decrease atmospheric carbon content.

The demand for natural and healthier food is increasing worldwide. Commercially, industries related to food and beverages, nutraceuticals, pharmaceuticals, cosmeceuticals, and aquaculture use algal pigments such as beta-carotene, astaxanthin, fucoxanthin, lutein, chlorophylls, phycocyanin, and phycoerythrin. Due to these valuable health-related properties, the global algae products market accounted for USD3.4 billion in 2017 and is expected to reach USD6.09 billion by 2026, growing at a CAGR (compound annual growth rate) of 6.7% (Globe Newswire, 2019). The natural food colour business in 2016 was USD 1.3 billion with a 6.8% annual growth rate and is expected to reach USD1.77 billion by 2021 (Kannaujiya et al. 2017).

Microalgae

Microalgae are valuable sources basic food components such as proteins, carbohydrates, lipids, vitamins, trace elements, and a variety of biomolecules including pigments such as chlorophylls, carotenoids, phycobiliproteins, and phenolic compounds that are associated with fast growth and high immunity (Begum et al 2016).

With an estimated species in the range of 200,000 to several million species, microalgae are ubiquitous microscopic and macroscopic plants in both freshwater and marine ecosystems (Figure 1). Most species of microalgae have high protein contents that can be valuable sources of protein concentrates, hydrolysates, and bioactive peptides, and together with highly nutritive fatty acids and pigments, make microalgae valuable sources for the development of functional, nutritional, therapeutic, and cosmeceutical commodities (Foo et al 2015, 2017).



Fig.1. Different types of microalgae that can be cultured, freshwater species (F1 -F24) and marine species (M1-M11) (after Khaw et al. 2020)

The presence of polyunsaturated fatty acids (PUFA) such as docosahexaenoic (DHA), and eicosapentaenoic (EPA), and other omega-3 fatty acids which are high in antioxidants renders microalgae as suitable products for immune enhancement in humans and animals (Yusoff et al., 2020). In fact, microalgae can be considered as a potent source for functional foods due to their ability to synthesize compounds with high antioxidative properties. Microalgae have valuable pigments such as astaxanthin, lutein, beta carotene, chlorophylls, and phycobiliproteins.

As human dietary supplements, microalgae pigments such as carotenoids and phycobiliproteins function as anticancer, anti-bacteria, immunity booster, and cardiovascular disease deterrence. Lycopene has been reported to have the capacity to prevent prostate cancer. Microalgae are not only used as food and health products but also as feedstocks for biofuels (Medipally et al. 2015). In addition, microalgae pigments are also used in poultry, livestock, and aquaculture industries. Their high contents of lipids and fatty acids make them valuable alternatives for the replacement of fish meals in livestock and fish feeds. The much-needed polyunsaturated fatty acids (PUFAs) and beta-1,3-glucan, and other biomolecules associated with fish/shrimp health are also found in many microalgae species (Foo et al. 2019).

Applications in Aquaculture

Nutritious and balanced diets can influence the immunity and disease resistance in cultured animals and reduce disease-related economic loss. Dietary supplementation with key amino acids, polyunsaturated fatty acids, vitamins and carotenoids are critical in modulating immune response and productivity of the aquaculture industry.

Most microalgae species have characteristics that are required to be natural supplements in animal feeds including aquaculture. In aquaculture, microalgae can be used as live feeds, formulated feed supplements, health enhancer, water quality bioremediation, growth promoter, and animal color enhancer. Due to their antioxidant and free radical scavenging properties, the best-known function of pigments in aquaculture besides increased growth and enhanced immunity is pigmentation. In fact, there are some pigments, such as fucoxanthin, which are only found in certain microalgae.

Colors play a major role in improving perceived quality, acceptability and willingness to pay for aquaculture products. Colors in organisms are mainly determined by genetic inheritance, but the pigmentation can be greatly influenced by environmental factors and occurrence of diseases. The enhancement of coloration in fish can be due to the proliferation of chromatophores induced by the presence of carotenoids

Carotenoids, which are dependent on diets and body conditions, are responsible for many yellow, orange and red hues in animals including fish and aquatic invertebrates. The color of microalgae is influenced by the pigments which are a part of the cell's photosynthetic system and naturally contain high-value compounds such as polyunsaturated acids, fatty acids, chlorophylls, phycobiliproteins and carotenoids. Carotenoids showed better effects on skin pigmentation compared to xanthophylls. In aquaculture, carotenoids function in pigment development, antioxidants and vitamin enrichment, growth and reproduction improvement, cellular protection from photo-damage and health enhancement. In ornamental fish industry, colouration is one of the product main attributes, as demonstrated by the effects of astaxanthin on the fish pigmentation. However, the effects on pigmentation could vary with the pigment sources. Diet supplementation with a photosynthetic bacterium did not significantly enhance the colouration of the Japanese ornamental koi, *Cyprinus carpio*, compared to a blue-green alga, *Spirulina platensis*. Astaxanthin and canthaxanthin are responsible for the typical red colour in most fish and thus these two pigments are commonly used in the diet of farmed salmonids for pigmentation enhancement

Microalgae are frequently used in hatchery to enrich zooplankton such as copepods and *Artemia* as live-feeds, which could be used to increase growth and survival of fish and shrimp larvae. There were significant increases in protein, carbohydrate, ascorbic acid, and β -carotene in *Artemia* enriched with a green alga, *Nannochloropsis oculata*. Live feeds such as rotifers and microcrustaceans, like other aquatic consumers do not contain endogenous carotenoids, but can be enriched through their diets. Carotenoid astaxanthin and lutein was used to enrich live-feeds such rotifers and the population was more productive and had higher nutritional contents for healthy development of larval fish compared to rotifers without the carotenoid supplements.

Microalgal pigments are also effective in improving growth and health of many cultured organisms. Lim et al (2019a, b) demonstrated that dietary administration of astaxanthin improves feed utilization, growth performance, survival and health of Asian seabass, *Lateolabrax calcarifer*. In addition, supplementary feeding with astaxanthin was also effective in reinforcing fish immunocompetence and disease resistance against *Vibrio alginolyticus* infection in sea bass (Lim et al. 2021).

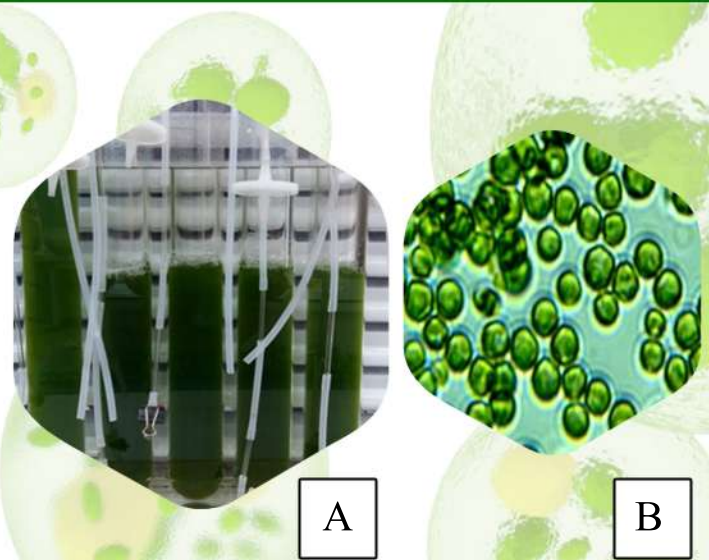


Fig.2. *Chlorella vulgaris* (green algae) culture (A) and cells (B)

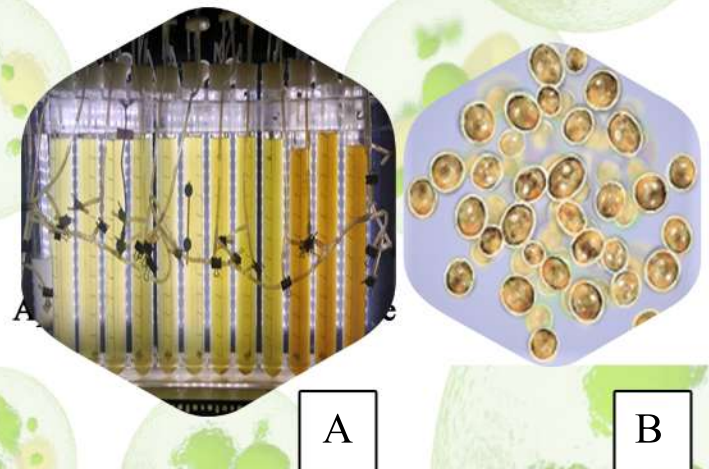


Fig.3. *Isochrysis galbana* culture in column photobioreactors (A) and *I. galbana* cells (B)

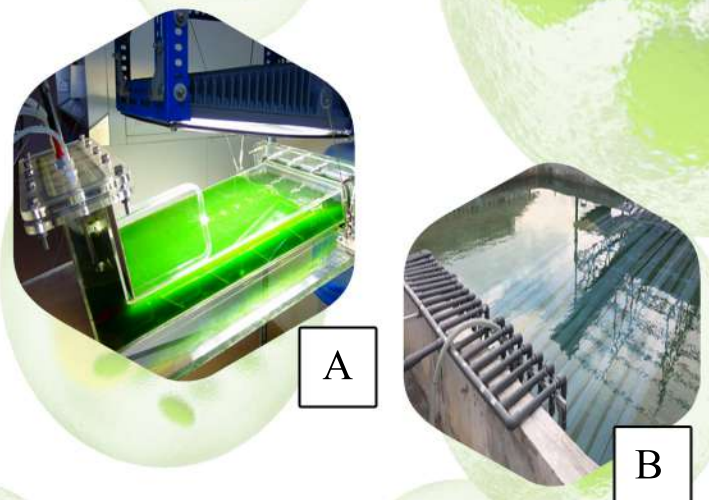


Fig.4. A prototype of the Cradle photobioreactor of the SATREPS-COSMOS project (A) and Outdoor bag photobioreactor in a pond, which can be integrated with fish culture. Placement of culture bags in the water keeps the bags cool and avoid damage from the sun (B)

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Introduction

Rainbow trout (*Oncorhynchus mykiss*, Walbaum 1792) are coldwater species that belong to the family of Salmonidae. They are native to North America and have been introduced worldwide, except Antarctica [1]. Rainbow trout have the capability to adapt to different aquatic habitats (rearing systems, reservoirs and natural waters) and have been introduced widely in other continents of the world such as Eastern Asia, Western North America, Central and Western Europe for aquaculture purposes [1]. Rainbow Trout was first introduced in the Ooty region of South India in the year 1863 [2]; this was followed by other coldwater regions of the Indian subcontinent.

Rainbow trout fisheries have flourished and expanded greatly in the Northern Himalayan regions and are available for commercial aquaculture. However they are neglected in the South Indian regions of Munnar, Ooty and Kodaikanal due to the absence of fisheries specialists and because of the lack of knowledge on trout fisheries and are currently in an endangered state [3].

Rainbow trout fisheries in the Indian scenario and its importance

Rainbow Trout fisheries in the North India first began in the year of 1900 in the Jammu and Kashmir State by Mr. F. J. Mitchell [4] and had arrived from Howeiton in Scotland. Currently, Trout fisheries in North India have become economically important, especially in the Himalayan regions and are in high demand. The fisheries department of the Jammu and Kashmir State government has achieved remarkable success in trout fisheries and has 59 trout rearing units in various districts (<http://jkfisheries.in>) and with one farm in the Kokernag region of the Anantnag district serves as the mother unit and is known as Asia's largest trout fish farm (Fig.1).



Fig. 1. A view of the trout fish farm in Kokernag of Kashmir, also known as “Asia’s largest trout fish farm”

Further in Himachal Pradesh, trout fisheries have significantly contributed to the economic development of this state [5] and quality trout seeds are supplied to other north-eastern states such as Sikkim [6]. Trout farming has also flourished in the State of Arunachal Pradesh with hatcheries being established in this state. Adaptation of trout fisheries in the northern states of India has provided an excellent opportunity for game fishing and trout culture [3, 7]. Thus, trout fisheries have flourished in the North Indian regions and are economically viable. Although rainbow trout stocks are available in the high ranges of Munnar, Ooty and Kodaikanal [8, 9], the status of trout fisheries in South India is not fully developed [3]. Rainbow trout was introduced in India by the British mainly for sport and recreational fishing and it is enjoyed by anglers who do game fishing in India. Trout are reared in Indian uplands, and are contributing significantly to the revenue and economy of the people. Trout are a rich source of polyunsaturated fatty acids, accounting for 25 % of the total fatty acids [10] as such polyunsaturated fatty acids (linoleic acid (C18: 2n-6), docosahexaenoic acid (DHA; C22: 6n-3), arachidonic acid (AA; C20: 4n-6), and eicosapentaenoic acid (EPA; C20: 5n-3)) can be acquired only from the diet, therefore they play an important part of a healthy diet. Trout fisheries in India have become economically significant in the Himalayan regions and are known for producing table-sized trout [3].

Rainbow trout fisheries in Munnar

Trout Fisheries in Munnar was commenced by four Englishmen Koechlin, John Charles, Daisy Bell, and George Howlett of the Kanan Devan Hills Plantations (KDHP) Company in 1909 by introducing Brown Trout (*Salmo trutta fario*). This was initially successful, but further stocking operations with Brown Trout were not possible due to the First World War. In 1932, rainbow trout (*Oncorhynchus mykiss*) was introduced by A.W. John and a hatchery was established in 1941.

Though fisheries record are available in the 1930s and 1940s [11] and 1960s, but no data on trout fisheries are available until the 1970s [8]. However, the data after 1970 showed that rainbow trout have disappeared from a majority of the water bodies namely Kaniyamallay, Lakkam and Chokanadstreams; Devikulam and Letchmi lakes; and Madupatty and Kundale reservoirs (Figs. 2a - f) due to illegal fishing, pollution, siltation, and animal intrusion. Currently, only one water body named as the Rajamallay Stream (Fig. 3a) is considered safe for the rainbow trout (Fig. 3b) for stocking and angling [3]. However, the rainbow trout in the Rajamallay Stream has been gradually decreasing after 1970, and are under continuous fishing pressure due to regular fishing. Moreover, local villagers and tribes are involved in illegal fishing. Furthermore, animal intrusion is also the reason for the decrease in stocks as wild otter packs often visit the stream for their meal because fish is their staple food [12]. All these criteria led to the reduction of stocks in the Rajamallay Stream. The rainbow trout stock in Munnar is in an endangered state [3].

Conservative measures for future management of Rainbow trout fisheries in Munnar

Conservative measures are currently underway to help improve the trout fisheries in Munnar and some of them are as follows, fisheries experts from the Indian Council of Agricultural Research–Directorate of Coldwater Fisheries Research, India are frequently visiting the hatchery to help in the conservation of the trout stock. From 2020 onward, steps towards improvement of feed and water quality are being done to increase production. During 2018-2019, food pellets with improved nutritious value were formulated by ICAR–DCFR, Uttarakhand, India and fed and this gave excellent length and weight results to the fish. The strategy was adopted by the authorities of KDHP Company.



Fig. 2. Water bodies in Munnar once inhabited by Rainbow trout (*Oncorhynchus mykiss*); a) Kundale reservoir, b) Madupatty reservoir, c) Kaniyamallay Stream, d) Letchmi Lake, e) Devikulam Lake, f) Lakkam Stream

Further training was given by ICAR - DCFR scientist's for hatchery workers on breeding, Hatchery management and feeding practices in February 2019 [13]. Efforts are currently being taken to increase the fingerling production for stocking. Construction of a new hatchery will help in the improvement of seed stocking operations. The commercialisation of the trout fish happens on a small scale and table-sized trout are sold at INR800 kg-1 to hotels and restaurants by the Munnar Supplies Association which is an authorized sales outlet of the KDHP Company [3]. On February 2021, nearly 8000 juveniles of rainbow trout were purchased from ICAR-DCFR Experimental Field Centre in Champawatand were released in the Rajamallay Stream [14], this will help in the conservation of the genetic diversity of the trout stock. Some of the other conservative measures that can be done towards conservation are, implementation of the recirculating aquaculture system needs consideration because low water levels are a persistent problem and the streams receive water mainly from the southwest monsoon [8]. Angling rules and regulations need revision, with stringent action against destructive fishing. The KDHP Company should also take steps to restore all water bodies in which rainbow trout was previously found. The hill communities should also be educated on the conservation of the trout fish. Strict implementation of all the above factors can help flourish trout aquaculture in the High Ranges of Munnar in the future [3].

Rainbow trout stocks in Ooty

Trout introduction began in the Ooty region in the year 1863 by Francis Day who commenced by introducing of browntrout (*Salmo trutta fario*) and Loch Leven trout (*Salmo levisis*) in the 19th century, but it was a failure for four decades. In 1909, Henry. C. Wilson introduced rainbow trout from New Zealand and his attempts were successful and a hatchery was constructed in the Avalanche forest zone in 1910. Size and number decrease was noticed in 1913, indicating high fishing pressure. After this, In order to upgrade the stock, nearly five attempts were done and in 1920 rainbow trout were introduced from Kashmir for stock upgradation and this was followed by an introduction of four salmonid species from Japan in 1968 which consisted of golden rainbowtrout (*Oncorhynchus mykiss aquabonita*), brown trout, tiger trout (hybrid between brown trout and brook trout) and Sockeye salmon (*Oncorhynchus nerka*), out of all these releases, the golden rainbow trout established itself as the dominant strain in every anglers catch [2], while rest of the introduced species failed to establish itself.

Another consignment of 10,000 eyed ova of albino rainbow trout was introduced from Japan in 1974, but had perished due to fungal infection, after this yellow bellied Shasta strain of rainbow trout was introduced from Munnar and hybrids were formed.



Fig. 3. a) Rajamallay Stream, current stocking habitation of the Rainbow trout (*Oncorhynchus mykiss*); b) Rainbow trout (*Oncorhynchus mykiss*) caught from Rajamallay Stream.

In 1997, the National Bureau of Fish Genetic Resources initiated a cross-breeding programme, by fertilizing the eggs of the wild spawners in Ooty with cryopreserved milt from Barot hatchery in Himachal Pradesh, but it failed due to untimely weather conditions with a poor success rate of 0.5% [2]. Though, trout fisheries got established in the Ooty region, but no reports are available after 1998. In 2015, fish biologists from Madurai Kamaraj University assessed the present status of trout fisheries in Ooty. It was found that after 2000, trout fishing activities have been banned to avoid fishing exploitation. The fisheries department of Tamil Nadu, Government of Tamil Nadu annually raise fingerlings from ripe brooders and has them stocked in the Avalanche Lake (Fig. 4 a) in small numbers but is majorly stocked in the Upper Bhavani reservoir (Fig. 4 b) of Ooty. Rainbow trout are now designated as 'wildlife fish' (importance of protection of rainbow trout is equivalent to that of the tiger) to save them from being poached. Commercialization of trout is not practised in Ooty. Production records from 2011 to 2019 showed that a total of 633,340 eyed ova were produced and the total number of fingerlings stocked was 462,350. The produced fingerlings have been stocked along with the wild brooders in the Upper Bhavani reservoir. In 2019, torrential rainfall and landslides caused massive damage to the hatchery and the Upper Bhavani reservoir which is the only stocking site on the reservoir wall, the repairing of the wall required the water and the entire trout stock to be flushed out. Thus, only a little stock is now available in the Upper Bhavani reservoir and similar situations persists in the Avalanche Lake and Mukurthi reservoir. The rainbow trout stock in Ooty (Figs. 4c - d) is in an endangered state [15].

Conservative measures for future management of Rainbow trout fisheries in Ooty

Trout fisheries in Ooty hold ambient potential for growth, cultivation and farming, but are bred on a small scale. However, certain conservative measures can help in the sustainable development of this existing rainbow trout stock. There is a rising need for introduction of new rainbow trout strains which can make the population viable in the Upper Bhavani reservoir. Stripping and hatching sheds need construction along the banks of the Upper Bhavani reservoir, Avalanche Lake and Mukurthi reservoir, this can help in catching the brood fish and for quick fertilization of the eggs, thereby reducing the mortality.

Implementation of stringent action against poaching of trout fish must be done. The rainbow trout population in the Ooty region has high development potential [15]. Currently in 2022, the fisheries department of Ooty are taking proper steps towards the improvement of hatchery and are improving breeding procedures in the Upper Bhavani reservoir, Avalanche Lake and Mukurthi reservoir. Some of the other conservative measures that can be done are, introduction of new trout strains for genetic improvement of the stocks, Catch and release angling with supervision of the fisheries officials can be done for monitoring the trout population. Further maintenance of the hatchery premises using sanitary measures along with protection from animal intrusion (mainly by otters) is highly necessary. Cage culture of trout can also be implemented which can help in the employment and benefit of rural communities in Ooty [15].

Culture conditions for Rainbow trout

Rainbow trout can be bred artificially in specialized rectangular tanks called as "raceways" (tanks with free flow of water with an inlet and outlet) and can withstand temperatures upto 30°C. Adult brooders are first checked whether they are ready for spawning, i.e. in female the belly is gently for release of eggs, in male for release of milt. Adults are anaesthetized in clove extract (so that they don't move around while handling). The female fish is then stripped for eggs completely, and the eggs are placed in a bowl, and the male is stripped for milt and both the eggs and milt is mixed together and water is added to increase the sperm motility for fertilization. The fertilized eggs are then placed on flow through trays (trays with wooden sides and hollow glass rods like capillary tubes) and these trays are placed in hatching troughs which allows clear, cool and oxygenated water to pass through at 3-4 litres/min. Meanwhile before hatching, the dead eggs are removed in order to restrict fungal infection. Fungal infections can be controlled by using 37% formalin in the inflow water at a dilution of 1:600 for 15 minutes on an everyday basis, but should be stopped 24 hours before hatching time. The usual time taken for hatching is around 20 – 25 days.

After hatching, the trays are removed and are moved to shallow water with a depth of about 8-10 cm with a reduced water flow until they reach 'swim-up' stage, after which they reach the advanced fry stage and these and advanced fries are reared in fibreglass or concrete tanks and is fed with specially prepared starter feeds. As growth continues, dissolved oxygen is monitored, and the fish are moved to larger tanks and well-developed fingerlings are stocked in wild streams or lakes.

Need for conservation of rainbow trout

Rainbow trout play a vital role in the living of mankind and they need to be conserved for the reasons:

Nutritional Importance

Rainbow trout are a rich source of polyunsaturated fatty acids which makes up to 25 % of the total fatty acids of this fish [10] as they can be acquired only from diet and cannot be synthesised by humans [16], and are also good in protein content and these trout fish can be considered as an important part of a healthy diet.

Medicinal Importance

This fish has medicinal value especially for patients with cardiac diseases, rheumatoid arthritis and nutritional deficiencies.

Economic Importance

In 2002, the trout industry generated about \$9 million in economic output and created 201 jobs, and generated \$3 and \$0.9 million in North Carolina, USA [17].

Ecological Importance

Rainbow trout is sometimes used as a biological indicator for water quality in water purification facilities [18].

Adaptability

Rainbow trout can easily adapt and can be cultured in any cold and pollution free water environment around the world.

Rainbow trout in south india for future directions

Rainbow Trout have not received the importance and status as other aquaculture species in India have, this mainly because of the limited knowledge on trout fisheries to the general public. The availability and knowledge about Rainbow trout is limited only to the people residing in the Coldwater regions or hilly areas in North and South India, but it is also known to people who show great interest in sport and recreational fishery. Rainbow trout fisheries has reached phenomenal heights of growth and has greatly played a vital role in the economy of the fisheries departments of the northern states and in the lives of the public who have their living on trout farming. This is not the scenario in the South Indian regions and needs great efforts towards growth and sustenance, currently rainbow trout stocks have disappeared from most of the water bodies at Munnar and it is currently limited to the Rajamallay Stream of Munnar with the breeding operations in the Rajamallay Hatchery on a small scale. The scenario in Ooty is worrisome as the trout stock in the Upper Bhavani reservoir have been completely washed away and the hatchery too has been devastated due to the torrential rains and floods, thereby requiring reconstruction. Though remnant stocks are available in the Avalanche Lake and Mukurthi reservoir, breeding operations are not carried out here. The trout stocks in Munnar and Ooty are in an endangered state and suitable conservative measures are currently underway. Rainbow trout fisheries and cultivation can be an economic boon to the people of South India if fisheries specialists, scientists and authorities who can implement strong conservative actions towards protecting its habitats are available. If the other viable options that can help in the propagation and promotion of this salmonid species are considered, the fish will increase and flourish as it is seen in the Northern regions of India. The presence of clear water streams and ambient weather conditions have proved that they can sustain Rainbow trout life in the locations of Munnar and Ooty and it will continue to do so in the coming years also with efforts taken as proposed.

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CRESCO and MICROALGAE Production

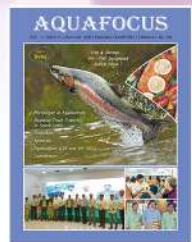
Cresco aims to produce large quantities of Algae and Microalgae. This would encompass production of economically important algal species like *Spirulina sp*, *Chlorella sp*, *Dunaliella sp*, *Hematococcus sp*, *Nanochloropsis sp*, *Diatoms*, etc. These are highly useful in the Aquaculture industry as Fish Nutrition.

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AQUACULTURE, G20 and UN SDGs



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Foundation for Aquaculture Innovations and Technology Transfer (FAITT)

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Introduction

Aquaculture operations are a booming industry worldwide in recent times. It is hailed as the Blue Revolution. The demand for fish, shrimp and algae farming is high in both developed and developing nations and is thus of significant interest to the aquaculture operations in the G20 nations

Among the fish producing countries globally, India ranks third and accounts for over 8% of the total fish production.

About G20

G20 (Group of 20 nations) is a prestigious international collective forum for mutually beneficial economic cooperation. The **G20** membership comprises of a total of **19 sovereign independent nations** which are Argentina, Australia, Brazil, Canada, China, France, Germany, India, Indonesia, Italy, Japan, Republic of Korea, Mexico, Russia, Saudi Arabia, South Africa, Turkey, United Kingdom and United States and also includes the **European Union**, which is a international political and economic union of **27 member states** that are mainly located in Europe.

India has assumed G20 Presidency for 2023 by taking over from its predecessor Indonesia, and will hand it over to Brazil for 2024, followed by South Africa in 2025, as part of the annual presidency rotation system.

India's G20 Leadership Priorities

Under India's G20 Leadership, its Cardinal Priorities pertain to the following aspects :

1. Accelerated, **Inclusive & Resilient Growth**
2. Accelerating progress on **United Nations Sustainable Development Goals (SDGs)**
3. **Green Development**, Climate Finance & Lifestyle for Environment (LiFE)
4. Multilateral Institutions for the 21st century
5. Technological Transformation & Digital Public Infrastructure
6. Women-led development



SUSTAINABLE DEVELOPMENT GOALS



Aquaculture fulfils UN SDGs in G20

In this context, **aquaculture operations** in most G20 Nations, especially in developing nations like Argentina, Australia, Brazil, China, India, Indonesia, Italy, Republic of Korea, Mexico, Russia, Saudi Arabia, and South Africa, form a cardinal aspect of G20 Priority policy initiatives for socio-economic benefits.

In juxtaposition with this, **aquaculture operations** also fulfils multifarious aspects relating to UN SDGs.

As a paradigm, **Aquaculture** food production process fulfils 5 IMPORTANT UN SDGs:

UN SDG 1 - by generating and providing artisanal employment thus leading to alleviation of poverty

UN SDG 2 - by reduce hunger and

UN SDG 3 - by ameliorating nutrition, generate economic growth

UN SDG 8 - Promote Sustained, Inclusive and Sustainable Economic growth

UN SDG 14 - Life below Water

Sustainable aquaculture would require sustainable economic practices. This would entail the use of human and material resources to create long-term sustainable values by optimal use, reduce wastage and adopt measures for re-use and recycling of resources.



Under India's G-20 Presidency, Marine Products Exports Development Authority (MPEDA) is organizing a special buyer-seller meet between Indian seafood exporters and importers of G20 countries. at the 23rd edition of India International Seafood Show to be held at Kolkata in February 2023. This will uniquely help to **revitalise the seafood sector in India and G-20 Nations to a great extent.**

As the **top ten buyers of Indian seafood are from G20 countries**, contributing 73 per cent of total exports from the country, this unique buyer-seller meet will help to improve the cooperation with G20 countries in marine product exports, It can also help in establishing collaborations and exports to other G20 nations where India has less presence and thus open up more new markets for exports.

India, according to published reports, has been exporting around \$100 million worth marine products till this year to the UK market. The EU market has also witnessed a 30 per cent growth this year clocking \$777 million till October.

Aquaculture would thus be a **robust and viable business proposition** for various stakeholders including small and marginal farmers even in the rural segments in **India as well as in other G20 Nations,**

With good long term prospects via efficient aquaculture health management and disease control and improving the net yield and profits, the Indian and other G-20 nations farmers can ensure sustainable aquaculture economics in the long term and contribute to a robust Blue Revolution and contribute to a **sustainable Blue economy.**

PhytoGrow

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COMPOSITION PER 1 KG

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GUARANTEED ANALYSIS PER KG

Cinnamaldehyde \geq 5.00g/Kg, calcium propionate \geq 35.0g/Kg, thymol \geq 35.0g/Kg

INDICATIONS

Natural growth promoter and gut health enhancer for all species and ages.

DIRECTIONS FOR USE

Mix homogeneously with feed at 0.6-5 kg/ton of complete feed. Recommended doses: Broilers: 0.6-1 kg/ton of complete feed. Layers and breeders: 1-2 kg/ton of complete feed. Ruminants: 1-3 kg/ton of complete feed. Shrimp and fish: 2-5 kg/ton. PhytoGrow does not contain antibiotics.

STORAGE AND HANDLING

Store well closed in a cool and dry place. Do not eat, drink or smoke while handling. Avoid any contact with the product.

Net weight: 1 Kg Batch and manufacturing date: See the top of the bag.

Expiry date: Two years after the date of manufacturing

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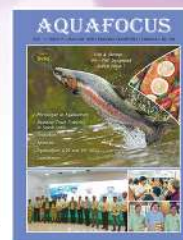
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PHYTOTHERAPY APPROACH IN AQUACULTURE



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Status of Phytotherapy

Phytotherapy is referred as Western Herbal Medicine. It involves use of plant-based therapies or extracts of natural origin as food and medicines typically native to Europe and North America for the improvement of health and avoidance of diseases. Of late, it is getting a lot of positive attention from both holistic and traditional practitioners over the past few years, and is often linked to traditional knowledge.

Phytotherapy includes some unique approaches to manipulating inflammatory and immunological mechanisms. Plants contain rich sources of natural bioactive compounds having various biological activities that provide medicinal value, which are known to traditional healers over several centuries. More than 80% of the world's population still follows the traditional medicines approach for their primary healthcare. About 250,000 higher level and 215,000 lower level plant species have been identified, among only about 6% of these species have been screened for their biological properties, while only about 15% were subjected to phyto-constituents analysis. Some synthetic drugs are developed by pharmaceutical companies which originate from natural resources such as fungi, bacteria, animals, protists, and plants. Plant extracts and their derivatives have received considerable attention as therapeutic agents for preventing and treating of many human health problems.

Application of Phytotherapy

Since the 1940s, among most of the bio-molecules involved in cancer treatment, almost half are either natural products or their transformed products. The promising biological activities of these molecules warrant more research and continued exploration of these natural products to find much needed novel medicines. Understanding how herbal medicines are sourced, processed, and standardized can help providers guide patients who are trying to choose the most clinically effective and affordable treatments.

Multiple herbs are often combined and sold as proprietary blends, which can increase the risk of allergies, adverse reactions, or cross-reactivity with other pharmaceuticals and supplements. Phytotherapeutic strategies for humans include acute inflammations of muscles, joints, connective tissues and glandular and gut tissue; chronic inflammatory diseases of the digestive tract, including gastritis, Crohn's disease and ulcerative colitis; chronic inflammatory diseases of joints, and other connective tissues, including rheumatoid arthritis (RA) and ankylosing spondylitis; psoriasis, scleroderma, other chronic inflammatory skin diseases (dermatitis), including complex and autoimmune conditions such as psoriasis; long-term inflammatory processes underlying chronic conditions such as diabetes and atherosclerosis.

History of Phytotherapy for Aquaculture

Aquaculture is an important revenue sector that contributes significantly to the economy and create various jobs opportunities in almost all countries of the world. Aquaculture activities fulfill more than 30% of nutritional food demands for humans in the world. For the increasing populations, strategies for increasing aquaculture production through intensive and semi-intensive systems are often associated with higher stocking density and massive use of artificial feed, leading to the incidence of diseases including bacteria, viruses, protozoan, parasite, and fungal infections, and resulting in significant economic losses.

In India, losses due to these aspects have been estimated at several million dollars per year. To prevent and manage the infectious diseases, large quantity of antibiotics and other chemotherapeutics are used, resulting in criticism for their negative impacts, residues in environment, high man power requirements, financial support, etc. In addition, the use of antibiotics or chemotherapeutics in aquaculture practice usually cause changes in water quality, resulting in chances of widespread infection, through increasing stress levels which directly affects their immune system and reduces the ability of fish to fight pathogens. Further, the antibiotics or chemotherapeutics may limit the larval growth and affect immune defense of the fish larvae. In this connection, there was demand for alternative prophylactic measures of commercial synthetic drugs.

Effect of individual or mixed herbals for Aquaculture

Medicinal plant extracts are recognized as a great potential for preparing clinically useful drugs to effectively prevent and control many bacteria, viruses, protozoans, parasite, and fungal diseases in aquaculture. These are gaining importance, because herbal treatment is cost-effective, eco-friendly and has limited side effects. Traditional herbal medicines seem to have the potential immune-stimulation that contains rich sources of bioactive compounds and thus serve as important raw materials for the production of drugs that are non-biodegradable and bio-compatible. The application of phyto-therapy in aquaculture started in earlier 1990's as a preliminary study on the effect of herbal extract on hematological and biochemical changes and in vitro antimicrobial activity against infectious pathogens. A number of studies proved that herbal additives enhanced the growth of fishes and also protected from infectious diseases. Herbal extracts have important properties like controlling of infectious pathogens due to their antioxidant and antimicrobial activity.

Natural herbal products have been reported to promote various activities like anti-stress, growth promotion, appetite stimulation, tonic and immune stimulation in fish and shrimps larvae culture. The herbal products are widely accepted as immunostimulants, conferring the non-specific defense mechanisms of fish and elevating the specific immune response. Several studies confirmed that the combinations of herbal extract is better than individual herbal extract and are well known to have many properties like anti-stress, growth promoters, appetizers, tonic and immuno-stimulants. However, till date there was no herbal-resistance immunity reported by any pathogen. Mixed herbal extracts also reported enhanced innate immune response and disease resistance in goldfish against bacterial disease.

Impact of plant active compound for Aquaculture

Several plant-derived compounds had been reported that enhanced non-specific immune, and led to better performance of haematological, biochemical in fish and shrimps. Plant active compounds are also reported to enhance immune response and disease resistance in *Cirrhinus mrigala* against fungal disease. After supplementation of the individual or mixed herbal products in diets, it enhances hematology and biochemical parameters and innate immune response in fish against pathogens.

The herbal active compounds may inhibit or block the transcription of the virus to reduce the replication in the host cells and enhance the non-specific immunity. Although, administration of these immune-stimulants can be done by feeding, immersion, and injection, it requires more studies. However, an overdose of herbals and their active principles could lead to negative effects on aquatic animals.

Conclusion

In conclusion, existing studies have clearly defined that herbal products positively enhanced growth, immunity, and disease resistance in aquaculture species. However, the information on functional and mechanisms of action of herbal products in aquaculture is still scanty. Further details on immunological and molecular studies are required to find out optimum dose, mode of administration, duration of treatment, mode of action in various fish against different pathogens before inclusion of the natural products in aquaculture practices.



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Introduction

Probiotics are now offering a promising alternative approach to controlling fish or shrimp diseases and improving the animal health through their ability to control pathogens and enhance nutrition through the development of digestive enzymes. Several routes are used for probiotic administration in aquaculture systems. Probiotics may be administered directly or added to the water as a dietary supplement. Also, the incorporation of probiotics into animal feed pellets is more effective than the direct application of probiotics in rearing systems. This report analyses the viability of the probiotics incorporated in the aqua diet which passes through the extruder or pelleting temperature.

Probiotic organisms such as live bacteria and yeast are likely to have a positive effect on the gut health of animals only if they reach the gut alive. Feed processing conditions, namely moisture addition and exposure to high temperature, shear stress and high pressure have the potential to lower the viability of probiotics. This report addresses a few techniques that enhance the probiotic survival in the extruded pellet diets. However, more research needs to be focused to improve the probiotics viability in pellet diets for the well-being of the aquaculture organisms.

Probiotics

Probiotics are currently gaining scientific and commercial interest, and are now quite common in health promoting therapeutic, prophylactic and growth supplements in functional foods. Studies are focused on microorganism's characteristic of intestinal microbiota for many years, and the term 'probiotic' was primarily restricted to Gram-positive lactic-acid bacteria, especially representative of the genera Bifidobacterium, Lactobacillus and Streptococcus. Unlike terrestrial animals, due to the flow of water passing through the digestive tract, gastrointestinal microbiota of aquatic species is particularly dependent on the outer environment.

Benefits of probiotics in aqua diet

- ❖ The probiotics of Bacillus in aqua diet have shown a positive approach to increasing the size and weight.
- ❖ Immune response stimulation is performed for host bio-security by probiotics applying different biological functions to kill pathogens.
- ❖ The probiotic results of aqua diet demonstrated a high possibility of prevention of stress in aquatic animals.
- ❖ Reduce the level of ammonia in water to ensure healthier output and increased profits.

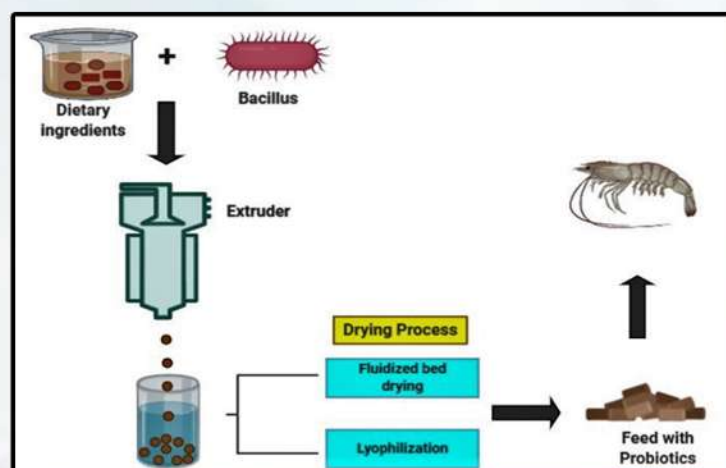


Fig.1. Process of efficient probiotics inoculation in extruder diet for shrimp feeds in aqua diet

Probiotics incorporation into pellet diets

Probiotics viability and stability have been a technological challenge in the production of feed, since probiotics including *Lactobacillus*, *Bacillus* and yeast are susceptible to high temperatures in the pelleting and drying process. *Bacillus* spores in the diet were found to have a loss of more than 99% after the processes of extrusion, extension and drying. Also, the viable yeast count in shrimp feed pellets decreased by 105 fold after being extruded by a meat grinder at 72°C for 31 s, followed by drying at 65°C for 6 h. Some of the methods that help to restore the probiotics in aqua diets (Fig. 1 & Table 1) which are discussed below.

Preparation of shrimp feed with probiotics using fluidized bed dryer method

Fluid bed drying is widely used for the drying of wet particles and granular materials. In a fluidized bed dryer, the probiotic cell suspension is mixed with a vibrating absorber bed or a matrix molecule that helps to form capsules by adhesion. *Lactobacillus lactis* was isolated and cultured overnight in 250 mL Erlenmeyer flasks containing 120 mL of sterile soymilk. For 24 hours, the sample was incubated at 37°C. The formulated feed for shrimps consisted of 40% fishmeal, 8% shrimp head meal, 20% rice bran, 10% wheat flour, 5% sago flour, 10% horse tamarind leaves powder, 5% soybean oil and 2% premix by weight. The mixture was sterilized at 121°C for 30 min and dried overnight in a hot air oven. The culture of the overnight *L.lactis* with and without pH adjustment of 5 M NaOH to 7.0 was added to the mixture. Soybean oil was then applied after 3 minutes of mixing and blended again for 3 minutes. Pressing the feed mixture into pellets at ambient temperature by using a 2 mm diameter pellet mill.

The wet pellets (250 g) were dried at an air inlet temperature of 80°C in a fluidized bed dryer with a 5 L stainless chamber. Milk powder and monosodium glutamate were applied to the overnight culture of *L. lactis*. It had a protective effect on probiotic viability at a high temperature of 80°C. Dried pellets (5 g) of shrimp feed were packed in plastic zip bags and kept at 4°C for 6 months. The viable cell counts in the feed can be measured on monthly using pour plate technique.

Probiotics in low-fishmeal extruded pellet aquafeed

Bacillus licheniformis received attention as probiotic supplements in aquafeed due to the production of heat-stable and low pH resistant spores. The spores also survived the extrusion process during low-fishmeal aquafeed production. Soybean meal (40 to 51% crude protein) with a balanced amino acid profile as well as low cost and market availability is considered to be one of the most promising fish meal substitutes in the aquafeed industry. To achieve a final concentration of 106 CFU/g, *B. licheniformis* was thoroughly mixed with low-fishmeal aquafeed. The mixed feed was then subjected to the extruder with twin-screw. Low-fishmeal EP feed was then air-dried for 3 hours at 60°C followed by vacuum fish oil coating and processed at -20°C before use. The high survival rate of *B. licheniformis* spores at high temperatures at 90°C indicates that it can be used as a food additive.

Conclusion

Research on modern technology that can protect and retain the feasibility of probiotics in aqua diet is needed. Technology should be further developed for the monitoring and maintenance of probiotics survival in the feed. For example, a brief understanding of the chemical composition and activity of the native microorganism and its microbial cultures and molecular knowledge is required to overcome the concept.

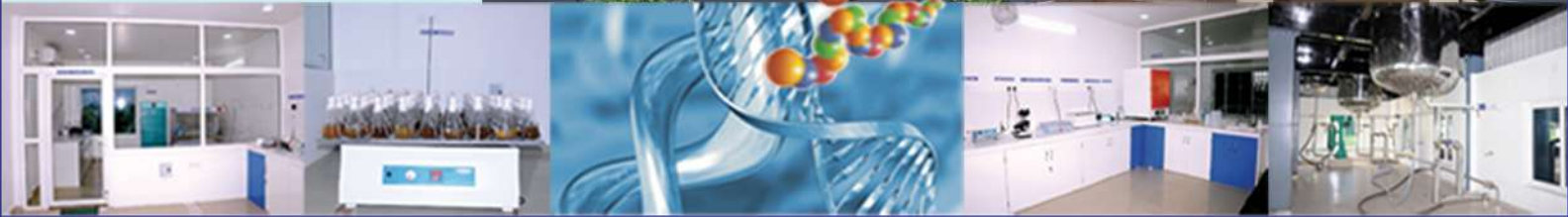
PROBIOTICS	EXTRUDER PROCESS	DRYING PROCESS
<i>Bacillus megaterium</i>	An extruder was used for pellet production at temperatures between 70 and 75°C	Pellet was air-dried in a vent hood at room temperature overnight
<i>Bacillus S11</i>	Probiotics were mixed thoroughly with low-fishmeal aquafeed and then subjected to the twin-screw extruder with the following conditions: feeder supply speed, 70 kg/h; conditioner temperature, 80°C	A low-fishmeal feed with probiotics was air-dried at 60°C for 3 h followed by vacuum fish oil coating and stored at -20°C until use
<i>Lactobacillus acidophilus</i>	Probiotics were added to the feed mix after cooling and passed through an extruder, barrel temperature of 60°C and cutter speed of 1100 rpm	Pellets thus obtained were dried at 60°C in an oven and put in airtight polyethylene until their use
<i>Saccharomyces cerevisiae</i>	Diet with yeast was added and mixed until a stiff dough was obtained. This was then extruded through a meat grinder. The pellet temperature just after pelleting was 82°C	After drying in a vertical cooler, the pellets (4mm diameter) were sampled and stored in plastic bags at 4°C
<i>Bacillus subtilis</i>	Extruder at 95°C was applied to the pellets to test the probiotic temperature resistance. Feeds were broken up, sieved to convenient pellet size	Later the fishmeal with probiotic was stored at -20°C

Table 1. Probiotics survival at different temperature during extruder and drying process

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Introduction

Aquaculture is one of the most significant and fast growing food industry, that provides the nutritional needs to consumers all over the globe (Ansari et al., 2019). Fish protein meets a significant percentage of the human daily requirement of protein to maintain normal physiology. As a result, aquaculture production is expanding to meet the global demand for human nutrition. The productivity rate on the other hand is influenced by many factors such as feed, seed, environment, water quality, etc. However, the major effect on production and profitability is contributed by feed (accounts for >65% of the production costs). Due to this, finding an affordable substitute and a feed component with a balanced nutritional profile is crucial for ensuring the global sustainability of both aquatic feed and production.

Spirulina, a microalga has gained popularity in recent times due to its rich nutritional profile (Zhang et al., 2020). Owing to the well-balanced nutritional profile such as macronutrients, amino acids, proteins, and fatty acids garners, spirulina can be utilised as an as an effective alternative feed in aquaculture and the commercial production of spirulina has thus gained popularity. Thus, this microalgae represents a promising possibility for boosting aquaculture production, particularly in developing nations such as India, where spirulina can be grown on a modest scale in rural regions (Mo et al., 2018).

Spirulina

Spirulina, a multicellular blue-green cyanobacteria of phylum Oscillatoriaceae has, has been utilized as food for generations by various tribes. Spirulina is easier to digest than other microalgae and grabbed the attention of the food and pharmaceutical industry as a unique supplement containing high protein level, mineral composition, vitamins, carotenoids, and essential fatty acids (Table 1) (Fig.1). The two most common species, *Arthrospira plantensis* and *Spirulina maxima* are more important due to their high nutritional content.

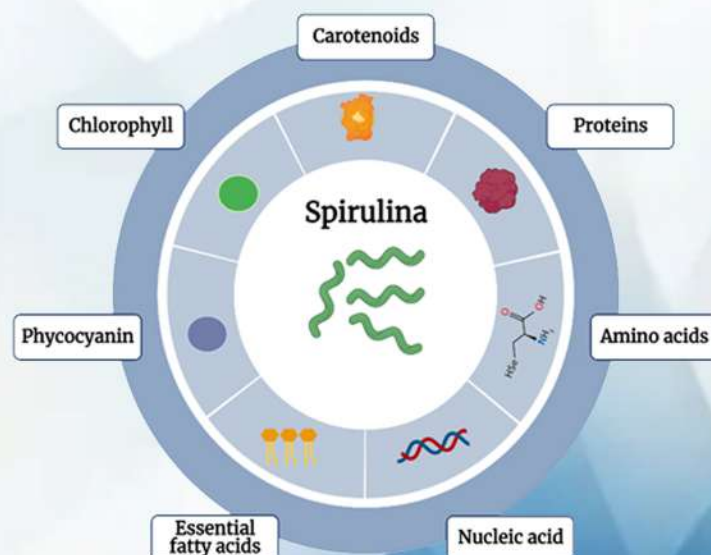


Fig.1. The biochemical composition of spirulina

Table 1: Biochemical composition of Spirulina
(Rosas et al., 2019)

Composition	Spirulina (%)
Crude protein	57.5
Amino acids	
Isoleucine	0.81
Leucine	1.28
Lysine	3.03
Phenylalanine	0.80
Tyrosine	0.54
Methionine	1.15
Histidine	3.21
Valine	3.51
Arginine	1.09
Tryptophan	0.93
Carbohydrates	15-21
Nucleic acid	2.2-3.5
Essential fatty acids	1.5-2.0
Natural pigment enhancers	
Phycocyanin	14
Chlorophyll	1
Carotenoids	47

Data from National Research Council (2011)

Spirulina as an effective functional feed in aquaculture

Spirulina has been deemed a beneficial aquafeed supplement because of its nutritional content and inclusion of bioactive substances that improve disease resistance and stress tolerance in the organism. Consequently, over the last decades, there have been several researches that involve spirulina as an effective feed supplement in fish and shrimp aquaculture (Altmann and Rosenau, 2022). Spirulina also improves immune response and increases resistance to pathogenic microbial infections (Macias-Sancho et al., 2014). As a result, extensive multi-nutritional and therapeutic potential has made spirulina as an excellent functional feed in aquaculture (Figure 2).

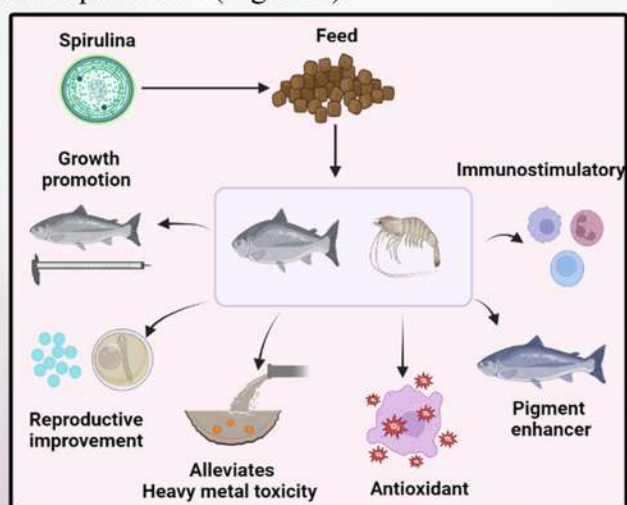


Fig.2. Functions of spirulina as an effective feed in aquaculture

Spirulina as a growth promoter

Various research has demonstrated that spirulina improves the growth and performance of various fishes and shrimp. The improvement in growth and feed utilisation after feeding with live spirulina has been recorded in many aquatic species. In context, *A. platensis* has a good effect on Nile tilapia fish growth and performance (Velasquez et al., 2016). Additionally, fish feed combined with *A. platensis* increased feed efficiency and disease resistance in aquatic animals (Macias-Sancho et al., 2014). Hence, spirulina could be considered a superior replacement for a protein source of feed as a growth booster in the aquaculture industry.

Spirulina as an Immunostimulant and an antimicrobial agent

Immunostimulants could defend infection and stress in aquatic animals by improvement of non-specific immune response (Talpur et al., 2013). Various immuno-stimulants, including lipopolysaccharides, chitin, and glucan, have been proven to increase fish immune defences when administered orally (Sakai, 1999). Spirulina has been recognized to stimulate the immune system in aquatic organisms such as fishes and shrimp by enhancing phagocytic activity, lysozyme activity, bactericidal activity, and white blood cell functions. Additionally, spirulina regulated the expression of gene signalling molecules such as cytokines, etc. Spirulina has also shown significant antibacterial characteristics that can effectively combat dangerous pathogens. Spirulina excretes a variety of metabolic by-products, including organic acid, vitamins, and phytohormones. Spirulina cell extract has also demonstrated antibacterial activity against pathogenic microorganisms such as *Saccharomyces* sp., *Bacillus* sp., *Streptococcus* sp., *Escherichia coli* and *Staphylococcus aureus* (Vo et al., 2015). Thus, spirulina could function as an efficient modulator of the immune system.

Spirulina boosts reproductive performance in aquatic organisms

Spirulina supplements boost reproductive performance such as improved spawning, fecundity, fertility and hatching rate in fish and shrimp due to their compositions like essential fatty acids, ascorbic acid, and carotenoids. In this context, carotenoids in the feed have been reported to improve broodstock performance in yellowtail fish (Watanabe and Vassallo-Agius, 2003). Additionally, spirulina is high in linolenic and linoleic acid, both of which are precursors of arachidonic acid. Arachidonic acid is required for the synthesis of prostaglandin, which is required for steroidogenesis, oocyte maturation and ovulation. Hence, spirulina is rich in the nutrients which are required for the reproduction of aquatic animals.

Spirulina as a colour enhancer in ornamental fishes

Ornamental fish farming generates significant revenue for exporting countries (Leal et al., 2016). Pigmentation is an important feature in ornamental fish pricing and carotenoids are pigments that are responsible for their colour (Storebakken et al., 1987). Thus, the healthy growth and pigmentation of ornamental fishes is reliant on diet that contains an appropriate amount of carotenoids. On the other hand, spirulina is rich in carotenoids, beta carotene and zeaxanthin and incorporating it into the fish diet can enhance fish pigmentation. Thus, spirulina can be utilized as a colour enhancer in the ornamental fish farming industry.

Spirulina alleviates heavy metal poisoning and its role in nanotherapeutics

Heavy metal poisoning exhibits severe ill effects in aquatic animals, causing oxidative stress resulting in the production of reactive oxygen and nitrogen species, which are implicated in cellular damage (Lushchak, 2011). Spirulina contains a variety of pigments, including beta carotene and phycocyanin, which are both antioxidants and anti-inflammatory. Spirulina's chlorophyll content also serves as a cleaning agent and a phytonutrient detoxifier (Wu et al., 2016). As a result, spirulina-rich diets reduce the harmful effects of heavy metals in aquatic animals and could be considered as an effective supplement that could alleviate heavy metal poisoning. In recent times spirulina is also employed in nanoscience (photosynthesized nanomaterial system).

Conclusion

Spirulina could serve as an effective dietary supplement with positive effects on growth performance, immuno-stimulation, antioxidant, antimicrobial, anti-inflammatory, reproductive improvements, pigments synthesis and alleviation of heavy metal toxicity. Hence integrating spirulina into traditional feed systems in aquaculture appears to be a great alternative feed.

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Introduction

Over 1 trillion species of microbes are found extensively throughout the world. Of these, bacteria are the predominant microbes. To survive in a competitive environment and to fight with competitors, bacteria employs various strategies. One of these defence strategies is the production of an antimicrobial peptide called bacteriocins. There are various classes of bacteriocins based on their size, structure and mode of action. Class I bacteriocins are classified by the presence of unusual lanthionine containing amino acids with unique mode of action. These are called lantibiotics.

Mode of action of Lantibiotics

Lantibiotics are antimicrobial peptides, produced mostly by Gram-positive bacteria, that binds to the lipid membrane of the target pathogen. Lantibiotics have two modes of action. Some varieties of lantibiotics interrupt the outer membrane during its synthesis and inhibit the formation of the cell membrane. The second type of lantibiotics bind to the bacterial membrane and disrupt the cells by forming pores. Third type of lantibiotics function in both ways. They inhibit cell wall synthesis and also forms pores in the cell membrane of bacteria (Fig. 1).

Different methods for the production of lantibiotics

Lantibiotics are produced by different methods. Producer bacteria should be given competitive stress or challenge with other microorganisms to make it produce lantibiotics. The microorganism used to challenge lantibiotic producers must be less powerful.

Lantibiotics are produced on a laboratory scale using multilayers of agar, in which one layer contains weak bacteria and the other layer contains lantibiotic producing bacteria. On industrial-scale, lantibiotics can be produced by co-culturing both the bacterias.

Advantages of using lantibiotics

Lantibiotics are bacterial killing peptides produced naturally by Gram-positive bacteria. They can tolerate salt and heat stresses. They are non-toxic and easily digestible. They have strong antibacterial activity hence very less amount will be required to use in aquaculture.

Lantibiotics can be sometimes combined with antibiotics to produce antibacterial activity against a wide range of bacterial pathogens. Some of the lantibiotics can be used against viral and fungal diseases.

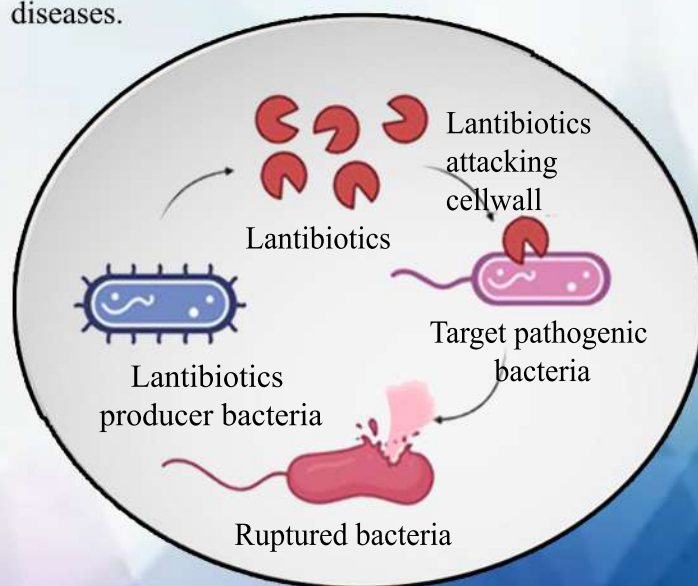


Fig. 1. Mechanism of lantibiotics in killing target pathogen

Commercially available lantibiotics

Nisin is a well-established FDA approved lantibiotic that is in commercial use for a long time. It is produced by lactococcus bacterium. Lantibiotics are commercially used in the food industry as a food preservative because it is tasteless, colourless, odourless and inhibit the growth of food spoilage microorganisms. Consumption of lantibiotics will not create harmful side effects like commercial antibiotics. Due to these advantages usage of lantibiotics are successful in the food industry.

Epidermin is another lantibiotic used in dairy products. It is used as a preservative to inhibit cheese spoiling bacteria.

Lantibiotics for the aquaculture industry

Lantibiotics such as Nisin, and Epiderminare have been studied extensively in inhibiting aquatic pathogens and these lantibiotics showed strong antibacterial activity against aquatic pathogens. Application of these lantibiotics can also increase the water quality of aquaculture farms and can prevent disease-causing pathogens. Since lantibiotics are used to store food products, they can also be used as a preservative in aquaculture products. The implementation of lantibiotics in aquaculture could be a better replacement for commercial antibiotics. Lantibiotics can be used for aquaculture farms either in powdered or solution form.

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APPLICATIONS OF POLYSACCHARIDE, POLYPHENOLS & PHAGES (3Ps) FORTIFIED FISH FEED IN AQUACULTURE



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Introduction

The demand for food is constantly increasing worldwide due to population growth. In this context, aquaculture food industry is a sustainable and fast-growing food-producing sector in the world.

India's fishery sector has been growing steadily and production is increasing every year. Due to the fast depletion of resources of wild fish species in marine ecosystems and to meet the growing demand, Government of India seeks to promote aquaculture through the "Blue Revolution" program, to cultivate the targeted commercial fishes in mainland ponds and water bodies. There is a high demand for such farmed fish feeds.

Recent studies suggested that the majority of fish feeds are supplemented with fishes caught from natural sources and these marine captures again leads to further exploitation of marine natural resources. The balanced feed that contains carbohydrate, protein, lipids, minerals and vitamins are very vital for growth and well-being of fish. There are several types of fish feed available commercially. Due to advancement of feed technology, tailor-made feeds for both aquariums and aquacultures are available. At present, selection of right feed at an affordable price is a challenge for commercial farmers. Medicated fish food is often used by the farmer to combat infection and bacterial outbreak in aquaculture. However, use of antibiotics in fish feed are not much encouraged due to emergence of antimicrobial resistance and also due to regulatory restrictions.

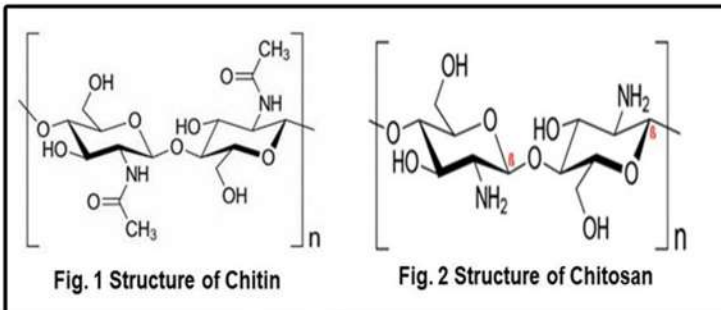
Chitosan is a natural polysaccharide polymer, which is endowed with potential biological benefits in fish and animals.

It is eco-friendly and recently got lots of attention to use in fish feed because it meets environmental requirements and provides sustainability to aquaculture. Chitosan plays a significant role in feed additives due to its low side effects, improves growth, provides immunity and also acts as antimicrobial agents (Abdel-Ghanyand Salem 2020). Polyphenols obtained from natural sources are endowed with potential with antioxidant and anti-inflammatory activities. Polyphenols such as flavonoids, phenolic acids, lignans and stilbenes are widely used as food additives in fish feed to improve the health status and production of fish in terrestrial fish farms (Ahmadifar et al., 2021). Polyphenolic compounds like caftaric acid, chlorogenic acid, cynarin, echinacoside and cichoric acid are present in plant parts. For example, purple coneflower that have been found as immune-modulator and antioxidant agent of fishes.

Vibriosis causes endemic bacterial infections and outbreaks in marine aquaculture farms, which may enter into fish through water and contaminated feed. To combat the microbial pressure in aquaculture, the bacteriophages are widely used as natural antimicrobial agents. Due to increase in antimicrobial resistance among the fish pathogens, there is an increasing trend in use of bacteriophages for multiple applications in aquaculture. Polysaccharide like chitosan, polyphenols and bacteriophages has shown individually as a promising candidate for providing health benefits and growth of fish. There is a vast potential scope for biological applications and health benefits using a combination of these 3Ps such as Polysaccharide, Polyphenols and Phages in aquatic organisms.

Source of chitosan and its applications

Chitin is a large, structural polysaccharide made from chains of modified glucose. Chitin (Fig. 1) is abundantly present in the exoskeletons of insects, cell walls of fungi, invertebrates and fish.



Chitosan (Fig. 2) is obtained by deacetylation of chitin (Fig. 3), which is composed of β-(1-4)-linked d-glucosamine and N-acetyl-d-glucosamine randomly distributed within the polymer. The cationic nature of chitosan is rather special, as the majority of polysaccharides are usually either neutral or negatively charged in an acidic environment. Chitosan has been recognized as versatile biomaterials for their unique properties such as biocompatibility, biodegradability, non-toxicity and low allergenicity (Kumar et al., 2004). However, the application of chitosan depends on the degree of deacetylation, molecular weight and characteristics like appearance and turbidity of polymer in aqueous solution (Nwe et al., 2014). Several studies have been carried out for biomedical applications of chitosan including pharmaceuticals industry, tissue engineering, drug delivery, antitumor and antimicrobial effects (Cheung et al., 2015).

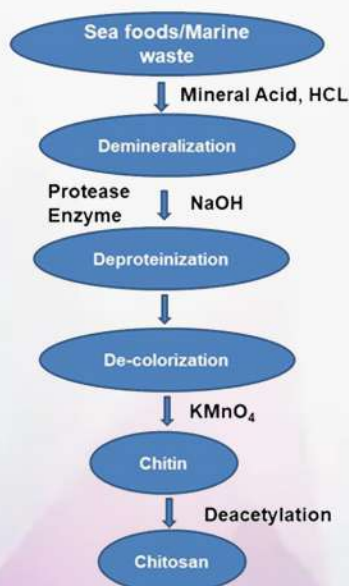


Fig. 3. Purification of chitosan from biological sources

Water Treatment

Chitosan is an excellent biomaterial for adsorption of organic and inorganic pollutant in the water treatment systems. Presence of multiple functional groups enables the molecule to interact with chemically active substance such as dyes, metals and organic micropollutants. To increase the adsorption properties of chitosan, functional groups may be modified by cross linking and grafting with active molecules or with elements. Chitosan cross-linked with epichlorohydrin, ethylene glycol diglycidyl ether, glutaraldehyde and tripolyphosphate have shown to improve the physical properties and also dye adsorption properties (Kyzas et al., 2015). Recently, chitosan-oxalic acid-biochar composite proved for adsorption of synthetic azo-dye (Doondani et al., 2022). Chitosan structure modified with carboxylic acid functional (-COOH) group improved the solubility and acquire tendency to chelate the heavy metals in addition to other functional groups such as amino and hydroxyl (-NH₂ and -OH) functional groups (Boamah et al., 2015).

Aquaculture

In aquaculture, chitosan renders several functions like coagulant, adsorbent, or bactericide. It has been used to improve the water quality of aquaculture wastewater by adsorption of suspended solids, organic compounds, NH₃, PO₄³⁻, and pathogens. The treatment efficiency improved based on the degree of deacetylation and acidic pH (Chung et al., 2006). The nanocomposite forms of chitosan are used for food preservation purposes and diagnosis of fish diseases (Ahmed et al., 2019). In fish models, chitosan has shown to have many physiological functions such as growth promoting, antioxidant, antimicrobial effect and immunostimulant effect. The antioxidant activity of chitosan was attributed due to radical scavenge ability, presence of hydroxyl moiety and metal chelation properties. This activity was directly proportional to the concentration of chitosan. The dietary supplementation of chitosan alone or with vitamin C increased the efficiency of antioxidant properties of fish exposed to heavy metals like cadmium, chloride and also involved in the regulation of enzymes like alanine aminotransferase, creatine phosphokinase and catalase (Banaee et al., 2015). The optimal concentration of chitosan in the fish feed was estimated between 1 and 2 g /kg for fish growth.

However, the concentration above 4 g/kg feed attributed to heavy development of intestinal microvilli that leads to blockage food mobility and fish growth depression and death (Zaki et al., 2015). In contrast to this observation, a basal diet supplemented with chitosan upto 20g/kg diet was fed to Asian seabass for 60 days challenged with *Vibrio anguillarum* and showed highest haematological and innate immune parameters compared to control group. In addition, 10 g/kg diet was found effective concentration for prophylactic purpose against marine vibrio infections (Ranjan et al., 2012). Chitosan has been used as a candidate for delivery of nucleic acid, vitamin C and hormones. Delivery of these molecules into several organism such as *Litopenaeus vannamei*, *Labeo rohita*, *Solea senegalensis* and *Lates calcarifer* have been reported (Bhoopathy et al., 2021). There are several research works on successful delivery of chitosan-based therapeutics and nucleic acid into fish and aquaculture. Dietary RNA has been successfully prepared and used in rohu (Ferosekhan et al., 2014) and nanoparticles of chitosan encapsulated with inactive particles of the viral haemorrhagic septicaemia virus (VHSV) tested against Olive flounder (*Paralichthys olivaceus*) (Kole et al., 2019). Chitosan coated membrane vesicles from intracellular fish pathogen *Piscirickettsia salmonis* was injected into zebrafish that provided immunogenic and increased survival against pathogen challenge (Tandberg et al., 2018). Chitosan is a versatile biomaterial and widely used many fields such as food industry, photography, wastewater treatment, aquaculture and chemical industry. However, low solubility in neutral and alkaline solution poses to limit the use of this compound abundantly. Chemical modification or functionalization of chitosan with other biologically active materials enhanced the physico-chemical and functional properties of this compound for different applications.

Applications of Polyphenols in aquaculture

Polyphenols are plant-derived compounds endowed with biological and potential health benefits. Polyphenols are considered a viable alternative to synthetic chemicals like antimicrobial compounds to improve fish health status and to enhance the fish quality, productivity and food safety. Polyphenols are extensively investigated in aquaculture as functional feed additives in the form of polyphenols and polyphenols rich additives. Polyphenolic compounds such as flavonoids, phenolic acids, lignans and stilbenes are

known to provide health benefits on the overall performances and immunity of fish; thereby improving the health status and production of fish farms. These compounds are expected to act on the pathways of antioxidant, pro-oxidant activities, regulation of gene expression and different immune parameters (Ahmadifar et al., 2021). The purple coneflower [*Echinacea purpurea* (L.) Moench.] were added to fish feed as adjuvant therapy for the prevention of fish diseases. The most active compounds of *E. purpurea* have been reported to contain polyphenols such as caftaric acid, chlorogenic acid, cynarin, echinacoside and cichoric acid (Oniszczuk et al., 2019). Plant extracts of polyphenols are known as tea phenols. It has been used to enhance and preserve the yellow croaker (*Pseudosciaena crocea*), which is known for important commerce and aquaculture particularly in Chinese cuisine.

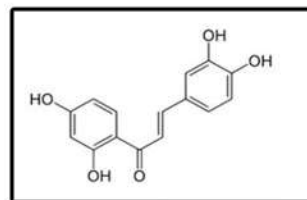


Fig. 4. Structure of Butein

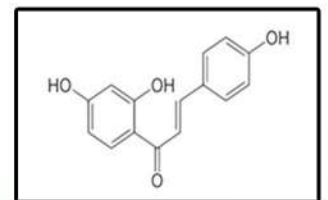


Fig. 5. Structure of Isoliquiritigenin

Phenolic plant extract has shown to interact with lipidperoxy or lipidoxo free radicals obtained from lipid oxidation and further stop breakdown of lipids. Hence inclusion of polyphenolic compounds in fish meat products would enhance the shelf life of fish food products (Ali et al., 2019). Myofibrillar proteins (MP) are most abundantly present in fish, which is likely to hydrolyse during storage. The polyphenols like chlorogenic acid and quercetin were investigated for the binding affinity with this protein and shown that chlorogenic acid bind to MP by Van der Waals forces and hydrogen bonds; quercetin binds to MPs by electrostatic interactions with tryptophan and tyrosine residues there by prevent degradation of protein (Xie et al., 2020). The polyphenolic compounds like butein (Fig. 4) and Isoliquiritigenin (ILT) (Fig. 5) are endowed with potential biological and pharmacological activities but very poorly soluble in water. Polyphenolic compounds are expected to promote the growth of symbiotic commensal bacteria and also ease the stress of the host as an antioxidant thereby increasing the health benefits of the host when consumed.

Among the polyphenolic compounds, Resveratrol has been extensively investigated in in-vitro and in animal models and found to increase lifespan of yeast, roundworms and fish (Cherniack et al., 2010). Butein and ILT have structural similarity with resveratrol but yet not investigated its health benefits in fish. These polyphenolic compounds containing plant extracts were tested against fish pathogens and proven effective. However, many polyphenolic compounds have low oral bioavailability, which limits the application of polyphenols in nutraceuticals. Food-grade delivery carriers encapsulation; and delivery of polyphenolic compounds could resolve poor water solubility and low bioavailability of polyphenols for various practical applications.

Interaction of polyphenols and polysaccharides

Naturally, polysaccharides and polyphenols coexist in many plant-based food products. Interactions of this polyphenol-polysaccharide affect physicochemical, functional and physiological properties, such as digestibility, bioavailability and stability of foods (Guo et al., 2022). The presence of multiple functional groups in polysaccharide makes them ideal for conjugation. The conjugations of polysaccharide with polyphenols are primarily to increase the solubilisation and control release of hydrophobic moieties.

Several approaches are employed for conjugation of polysaccharides with polyphenols such as esterification, free radical grafting, etc., these methods are selected based on the structural nature of conjugants. However, the integrity of the compound must not be affected. A polysaccharide like chitosan was conjugated with small molecule drugs, natural compounds, proteins/peptides, nucleic acids, etc., Introduction of these active molecule impart better biological activities like antimicrobial and antioxidant and also improve solubility. The accepted mechanism action of antimicrobial action of chitosan conjugated polyphenols is electrostatic interactions between the protonated amino group of the chitosan molecule and the anionic surface of the pathogen under acidic conditions (Qin et al., 2020).

Bacteriophages

Bacteriophages or phages are bacterial viruses that can invade bacterial cells and cause cell death by disruption of bacterial metabolism. Phages are isolated from environmental water samples or from host pathogens (Fig. 4). Before the discovery of bacteria, bacteriophages were widely used as antimicrobial agents. In order to combat the microbial pressure in aquaculture, the bacteriophages are used as natural antimicrobial agents. Due to increase in antimicrobial resistance among the fish pathogens, there is an increasing trend in use of bacteriophages for multiple applications in aquaculture.

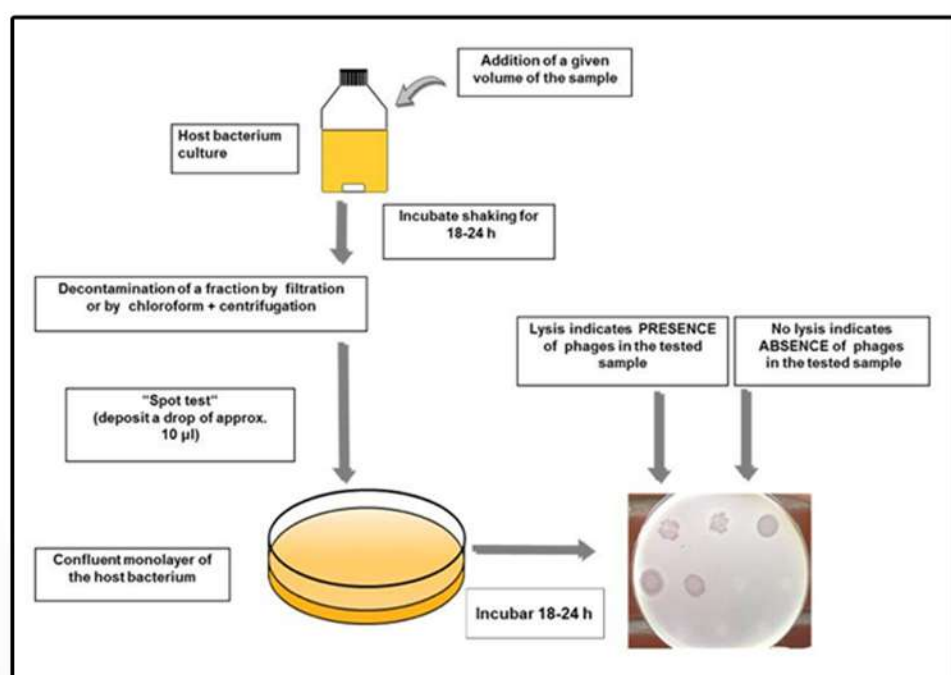


Fig. 6. Isolation of phage from marine water or sewage sample (Courtesy: Jebri et al., 2017)

The bacteriophages may be used as natural, genetically engineered phages and also phages derived from lethal genes and with other cargoes are used as antimicrobial agents (Nair et al., 2022). Phage based approaches have gained importance for antimicrobial treatment or as prophylactic measures due to its sustainable alternative to chemical antimicrobials. Recently, live artemia-mediated phage delivery methods were found promising tools for phage-based therapy against pathogenic bacteria to prevent aquatic diseases (Nikapitiya et al., 2020).

Phages can be administered through oral feed pellets sprayed with concentration range between $1-2 \times 10^8$ PFU/g and other routes of administration such bath or injections. The concentration and route of administration needs to be selected based on phage titer value against specific bacteria killing ability (Donati et al.,2021). The successful phage therapy treatment in aquaculture depends on the optimization of phage delivery methods and use of appropriate phage to control bacterial infections (Kunttu et al.,2021). The better bio-control was achieved by mixing therapeutic phages into cocktails of different virus types. In clinical and aquaculture, infrequently detected with appearance of MDR (multidrug-resistant), XDR (extensively drug-resistant) and PDR (pan drug-resistant) bacteria that leads to failure in the infecting the control. In these cases, the bacteriophages have become one the best hope for the future treatment of resistant bacteria that do not respond to available antimicrobial agents. However, for the commercial application of phage based therapies in aquaculture require more experimental evidence and field trials.

Application of 3Ps (Polysaccharide, Polyphenols and Phages) in Aquaculture

Polysaccharide like chitosan, polyphenols and bacteriophages has shown individually a promising candidate for providing health benefits and growth to fish. Chitosan itself acts as a carrier polymer molecule for delivery of a variety of compounds into the target of the host including polyphenols and bacteriophages. Chitosan functionalized with polyphenols are expected to improve the solubility of these compounds and expected to provide antioxidant, improve health benefits by overall wellbeing and when these are combined with phage are expected to provide better infection control. Chitosan is a biocompatible and mucoadhesive polymer. Due to the mucoadhesive property of chitosan, polyphenol and phages enter into the fish through gill, skin and into the intestine through the feeds. All these three agents are antimicrobial by themselves and when combined together, these may have synergistic effects, thereby providing better infection control of fish. Polyphenols are expected to provide stress relief and also modify the gut microbiome.

Conclusion

Based on the available data, it is obvious that chitosan (polysaccharide), polyphenols and bacteriophages have great potential for health benefits in fish. These three components were tested individually or in combination of two against the health benefits of fish. It was found that Chitosan conjugated polyphenol particular, butein and ILT with phage cocktail will have most desirable properties for healthy growth of fish, supports free from infection, stress and also expected to modify the gut microbiota. More investigations are required on these 3Ps with reference to the health benefits for fish when administered together through the food, particularly relating to infection control and analysis of gut microbiome.

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**Nov 17, 2022 -
Nov 18, 2022**

**4th International Conference on Aquaculture
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**Dec 13, 2022 -
Dec 15, 2022**

**AlgaEurope 2022 - Conferences about
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Jan 10, 2023

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**Feb 15, 2023 -
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**Feb 19, 2023 -
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**Aquaculture America
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**Feb 23, 2023 -
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**13th Global Summit on Aquaculture &
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March 19, 2023

**International Conference on Fisheries And
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Mar 25, 2023**

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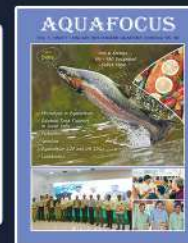
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FAITT SHOWCASES SPIRULINA in AQUA EXPO



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ABOUT FAITT

Foundation for Aquaculture Innovations and Technology Transfer (FAITT), is a nascent and highly unique Non-profit Private Research and Development (R&D) Foundation in the field of Aquaculture, It was established by eminent scientists, in Eluru, Andhra Pradesh, India in 2020 to uniquely focus on “Aquaculture Research And Development” – both nation-wide and globally. With its primary focus on **Research, Publishing, Skill Development, Training and Social Empowerment** activities.,it aims to benefit both the Aquaculture community as well as society – both nationally and internationally, with **quality, innovative and Sustainable Aquaculture operations and practices.**

FAITT, as a **Govt of India recognized Start Up and Triple-ISO Certified firm**, believes in the maxim: “Human knowledge belongs to the entire world for the benefit of all, and thus collaboration is better than competition”. Also, **Aquaculture** has been declared to have immense potential and **Spirulina** has been hailed as the **next global Super-food** by the UN (United Nations) due to its very high nutrient potential. Considering the necessity and importance of such a potentially vast food resource sector, FAITT has been established to initiate and fulfil the R&D as well as Training requirements in Aquaculture, to **innovatively and scientifically support the farmers and allied stakeholders.**



AQUAEX INDIA EXPO - 2022

FAITT participated in the “AQUAEX INDIA” expo held on 4,5 & 6 of November at Bhimavaram. Its associate company CRESCO ALGAE FORT, is involved in Spirulina Production and successfully showcased the same. They supplied energy drinks, which is made up of Spirulina, in juxtaposition with traditional ingredients, to the expo visitors and participants which numbered to more than 5000 people. All of them were really happy to taste the energy drink. Their highly positive feedbacks are a real boost to the CRESCO ALGAE FORT team.

Andhra Pradesh Undi constituency MLA, Shri Mantena Rama Raju Garu visited CRESCO stall at expo and had a glass of Spirulina Juice and expressed great delight. In juxtaposition with this, and many other visitors, farmers, students and other delegates liked it very much.

FAITT and CRESCO are very grateful to Mr. Ramchandra Raju Garu (President, AquaEx) and the expo organising committee who have supported and help achieve a great success to promote FAITT awareness and Spirulina Juice. Dr. Dhanaraj, from FAITT, who is an achiever of Young Scientist award, took a presentation on “Role of Micro Algae in Aquaculture”, which is quintessential innovative paradigm for current aquaculture growth and development. FAITT also released its nascent issue of AQUAFOCUS magazine on the eve of Aquaex India Expo – 2022.

Many farmers and entrepreneurs cognized the importance and benefits of algae, especially Spirulina, and expressed their interest to help in sustainable aquaculture by growing and maintaining Microalgae in their aqua farms.

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