

# THE USE OF PROBIOTICS IN AQUACULTURE - A REVIEW

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**Abstract** - Aquaculture is emerging as one of the most promising sources for providing animal protein for the rapidly growing population. Over the years, the aquaculture industry has been affected by environmental damage, disease outbreak leading to low productivity of the aquatic species. Over the decade, different methods such as antibiotics, chemicals have been employed to sustain the aquaculture production. These methods infer a lot of disadvantages to the aquatic species. Alternative to these methods will be the use of probiotics. In this review, it highlights the use of probiotics in the aquaculture field, its mechanism, and also its application.

**Key Words:** Aquaculture, Probiotics, sources, Mode of action, Application of probiotics.

## 1. INTRODUCTION

Aquaculture provides aquatic foods like shell fish and fish. Fishes provides high-quality protein, and they are majorly consumed in areas where livestock is short in supply [1]. The total animal protein consumption to human diet, fishes contribute about 17.4% of protein in Africa, 9.2% protein in Europe, 26.2% protein in Asia, 7.4% in Central America [2]. Aquaculture has evolved as the fastest growing food producing sector which employs many people in various different countries, thereby increasing the economy of the country. It has been developed as an important component in the global food security [3]. During the large-scale production, the aquatic species undergo a lot of stressful conditions, environmental issues, diseases which ultimately leads to economic loss.

Among the production limiting challenges, the infectious disease takes the lion share by causing multibillion-dollar loss annually [4]. Fish disease impedes both economic and social developments in many ways: directly, through production losses and increased operational costs, and indirectly, through cost of society, adjustment in market shares and increase in price due to lower supply [5]. It is essential to act upon the constraints based on

scientifically proven, as well as locally applicable ways to overcome the economy loss.

To control and prevent the spread of disease, application of chemicals like organophosphates [6], formalin [7], furazolidane [8], maintaining hygiene conditions have been employed. In the recent decades, it has led to a sizeable increase in the application of antibiotics which are used in the veterinary therapy [9]. The amount of antibiotics used by different countries have exerted a huge compression towards resistance among bacteria by the promiscuous flow of resistance gene [9]. The resistance is passed through 2 ways, acquisition of plasmids (mainly *Vibrio* species) or through chromosomal mutations [10]. The best-known transmission of resistance gene between aquaculture ecosystem and humans, is *floR* (florofenicol resistance gene) in *Salmonella typhimurium* DT104, confers resistance to chloramphenicol which almost has sequence identical to *floR* described in bacteria found in fish, *Photobacterium damsella* [11]. Use of antibiotics also has adverse effects on the aquatic species, as its residues are harmful to the aquatic environment [12].

Of late, the use of probiotics in aquaculture has gained an imaging recognition for controlling the pathogenic spread among the aquatic species. The application of probiotics will be an eco-friendly measure for sustainable aquaculture practices [1].

## 2. WHAT IS A PROBIOTIC?

The word "Probiotics" comes from a Greek word – *pro* and *bios* which means "prolife" [13]. Dr. Elie Metchnikoff in 1905 [14] was the first person to explain the beneficial role played by the bacteria among farmers who consumed milk containing pathogens. In 1987, Fuller expanded it as live microbial feed supplement, which affects the host in a beneficial way by improving its microbial intestinal balance [15].

According to WHO/FAO [16], probiotics are Live microorganisms when administered in adequate amounts will confer a health benefit on the host. It has been proposed that a potential probiotic organism must fulfil the following criteria.

1. Should have a beneficial activity on the host.
2. Should be non-pathogenic, non-toxic, and free of significant adverse side effects.
3. Should be able to survive through the gastrointestinal tract.
4. Should be in an adequate number of viable cells to confer the health benefit in the product.
5. Should be compatible with product matrix, processing, and storage conditions to maintain the desired properties and accurately labelled [17].

The probiotics can be sourced from a host or from a non-host origin. It can be a Bacteria, Algae, Fungi [18]. The microorganism associated with the host confers better results with diverse biochemical features [19]. It provides leverage in some biotechnical concerns like salinity, temperature, familiarity to the environment features [20]. Mostly in aquaculture, the microorganism isolated from the intestine of aquatic as well as terrestrial animals are used extensively as probiotic [21].

*Bacillus cereus var toyoi*, *Bacillus licheniformis*, *Bacillus subtilis*, *Enterococcus faecium*, *Lactobacillus casei*, *Lactobacillus plantarum*, *Lactobacillus rhamnosus*, *Pediococcus acidilactici*, *Saccharomyces cerevisiae*, *Streptococcus infantarius* are the list of microorganisms which are authorized by European Union as probiotics in feeding stuffs in aquaculture sector [22].

For animal nutrition, mostly lactic acid bacteria are used as a probiotic to improve the feed efficiency, to increase the growth, to prevent intestinal disorders, rate of survival, also microbial monitoring [23][24][25].

Over the decade, probiotics has gained focus among researchers to implement in their study. Thus, probiotics mostly promote the health of other organisms, when consumed or applied in the environment.

### 3. COMMERCIAL PREPARATION OF PROBIOTICS

1. Probiotics can be applied in combination or as a single organism in aquaculture field [27]. Single strain cultures are less effective than those based on mixed strains [28]. Multispecies and Multi-strain probiotic culture shows increased protection against infection [29]. Combined application of both probiotic and prebiotic is called synbiotics, which provides a competitive advantage over competing endogenous populations. It therefore improves the survival rate and implantation of the live microbial dietary supplement in the GI tract of the host [30].

2. Another fascinating idea is encapsulation of probiotic with the live feed, where the probiotic can remain viable and proliferate to increase the effectiveness [27].

3. Application of probiotics in the water is the method mostly followed in aquaculture. The direct oral administration of probiotic bacteria is mostly followed for prawn culture [32]. Probiotic can be directly added into culture water, as water additives, immersion method of application, bathed in bacterial suspension [27].

4. The probiotics are applied for different time duration, and the frequency of administration plays an important role in maintaining the function of probiotic bacteria. Prolonged exposure would enhance immune suppression of continuous responses of non-specific immune system [31].

5. Probiotic dosage may bring different results to different aquatic species, and they depend upon physiological status, probiont species, application, and rearing conditions [34] High dosage of probiotic bacteria will not result in a high level of protection [35]. Dosage of  $10^8$  CFU  $g^{-1}$  of *L. lactis* improved the growth rate, protein efficiency ratio, improved lysozyme, antiprotease levels of rainbow trout fry [33].

### 4. MODE OF ACTION OF PROBIOTIC

The potential disease-causing pathogens belongs to the genera, *Aeromonas*, *Citrobacter*, *Chromobacterium*, *Flavobacterium* *Pseudomonas*, *Micrococcus*, *staphylococcus*, *Streptococcus* [26]. To eliminate the pathogens from the host, probiotic organisms act in different ways.

1. Competitive exclusion - The pathogenic microorganism adheres to the mucosal membrane of the gastrointestinal tract of the host to develop the disease. The probiotic bacteria adhere to the receptors, colonize and interfere the action of the pathogenic microorganism [36][19]. This is known as competitive exclusion as they directly fight for space, oxygen, energy and nutrients [27]. Attachment of probiotics can be specific or non-specific [19][38]. Lactic acid bacteria, which is a known probiotic group consume nutrients which are essential for the pathogen's growth [37].

2. Immunomodulation, probiotics act by stimulating the immune system of the hosts. They help in increasing the phagocytic activity of leucocytes, phosphatase, antibodies, lysozymes, anti-microbial peptides, cytokines, transforming growth factor [20]. Thus, they improve the growth of the aquatic species.

3. Production of inhibitory substances: - Probiotic bacteria secrete substances which have bactericidal, or bacteriostatic effects on other organisms. The substances like hydrogen peroxide, siderophores, lysozymes, bacteriocins, proteases, lactic acid, butyric acid, propionic acid, acetic acid and indole(S,3-benzopyrrole) are produced by the probiotic bacteria. As a result, they reduce the pH in the lumen of the host, thus preventing the proliferation of the pathogenic organism [20].

Several probiotics have been known and documented to have antibacterial, antiviral, and antifungal activity against pathogens [20].

4. Quorum sensing: - "Quorum sensing is defined as the regulation of gene expression in response to fluctuations in the density of the cell-population" [20]. Halogenated furanones, autoinducer antagonists has the ability to activate or suppress the gene expression in the susceptible pathogenic bacteria. This is considered as a potential anti-infective strategy [39].

Thus, probiotics can be considered for the prophylactic use of antibiotics and chemicals [20].

## 5. APPLICATION OF PROBIOTICS IN AQUACULTURE

Initially, probiotics have been used as a growth promoter but recently they are applied in many areas to check their effect on reproduction and stress

tolerance [40]. Probiotics is also applied in aquaculture for different purposes, such as, to inhibit the pathogens, to improve nutrient digestibility, to improve water quality [40].

Probiotics have been used as a growth promoter to improve the growth of the cultivated aquatic species. In reality, whether these probiotics increase the appetite of the aquatic species or improve the digestibility of the species is unknown [41]. As already discussed, probiotics has the ability to adhere and colonize to the mucosal membrane of the intestine [36] and can exert multiple benefits when administered over a long period as their multiplication rate is higher than the rate of expulsion. *Bacillus* sp. S11 [42], *Bacillus* sp. [43], *Lactobacillus helveticus* [44], *Lactobacillus casei* [45] have been used as a growth promoter for *Penaeus monodon*, Catfish, *Scophthalmus maximus*, *Poeciliopsis gracilis* aquatic species respectively.

The probiotics produce extracellular enzymes and growth factors like amylases, proteases, lipases, vitamins, fatty acids [36] and amino acids which improves the digestibility of the aquatic animals. These enzymes and growth factors are absorbed efficiently when probiotic is supplemented with the feed [46]. Results show that *Lactobacillus helveticus* [44], *Lactobacillus acidophilus* [47] improves the nutrient digestibility of the *Scophthalmus maximus* and *Clarias gariepinus* under appropriate conditions respectively.

As the demand for aquatic species increases, it triggers pressure on the aquaculture industry to increase the production in a shorter time interval, ultimately it exerts pressure on the aquatic species [40]. The Lactate level and plasma glucose level are the appropriate indicators for stress condition. These levels increase as a secondary response to stress, to fulfill the energy requirements [40]. Results show that *Lactobacillus casei* [45], *Lactobacillus delbrueckii* [48] enhances the tolerance level in *Poeciliopsis gracilis*, *Dicentrarchus labrax* respectively.

Antagonistic property of the probiotics helps in controlling the diseases in the aquatic species as these probiotics has the ability to release chemical substances as discussed before. *Bacillus* sp. [50], *Pseudomonas* sp. [51], *Lactococcus lactis* [52] has been applied to *Penaeids*, *Oncorhynchus mykiss*, *Epinephelus coioides* aquatic species respectively.

Application of gram-positive probiotic strains (especially genus *Bacillus*) is preferable, as it is more efficient in transforming organic matter into CO<sub>2</sub> when compared to gram-negative bacteria. Increased concentration of nitrogen compounds like ammonia, in the water affects the water quality. Thereby, water quality can be improved by the application of probiotics into the water [40].

## 6. SELECTION OF PROBIOTIC BACTERIA

Method of selection of Probiotic bacteria include, Collection of information, Acquisition of probiotics, Evaluating and assessing the ability of the potential probiotic bacteria to antagonize pathogenic microorganism, Evaluation of the effect of the probiotics in host, Cost analysis [53].

## 7. CONCLUSION

Probiotics make it a feasible alternative to the use of other chemicals and antibiotics. The effectiveness of probiotics depends on lot of factors. They are, proper management of strains, provision of correct dosage and also its stability during the production procedure.

There are some limitations to the use of probiotics, like, during the application of probiotics, other drugs and chemicals should not be administered for treating any disease, as it interferes the establishment of beneficial microorganisms [49]. Other limitations such as, the safety of the microbial strain, virulent plasmid, the transmission of antibiotic resistance has to be considered. The limitations can be appeased by reducing its inclusion in fish diets by combining it with prebiotics, method called synbiotics. In favor of a more sustainable aquaculture industry, it can be a viable option.

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