

Application of Some Essential Bacterial Probiotics in Aquaculture: A Review

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Abstract

Aquaculture plays an essential role in maintaining food security and sustainable lives for the world's ever-growing population. Due to the expansion, intensification, and variety of aquaculture methods, aquatic animals confront significant challenges, including disease outbreaks, decreasing output, and risks to the sustainability of the sector. Poor soil conditions in aquaculture cause higher mortality, a higher prevalence of diseases, and lower yields of aquatic organisms. Overfeeding, dead animals, and feces from cultured organisms like fish and shrimp are the main causes of soil deterioration in aquaculture ponds. The two main compounds that cause issues are hydrogen sulfide (H₂S) and ammonia (NH₃). To prevent diseases and promote growth, antimicrobial medications, insecticides, and disinfectants have been used in aquaculture. This has resulted in the evolution of bacterial strains that are resistant to these chemicals. Probiotic usage has been suggested as a potential remedy for various problems. Probiotics are a live beneficial bacterium that are ingested through food or water which helps to maintain a healthy balance of microorganisms inside the body. The growth of dangerous pathogens is inhibited by bacteriocins, siderophores, lysozymes, proteases, and hydrogen peroxides produced by probiotic bacteria. This review contains various ways in which probiotics can be used in aquaculture systems. Thereby, probiotics assist the host animals by improving development and nutritional digestibility, boosting disease resistance, and improving the quality of the culture water.

Keywords: Probiotics, aquaculture, application, growth promoter, improved water quality

INTRODUCTION

Aqua feed and health management are extremely important for a sustainable aquaculture economy.

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Aquaculture produced over half of the world's fish in 2019 and is still the industry with the highest growth rate [1]. The Food and Agricultural Organization (FAO) of the United Nations [2] reported that the contribution of aquaculture to global fish production increased from 25.7% in 2000 to 46% in 2018. The average yearly increase in fish consumption from 1961 to 2017 was 3.1%, exceeding the 1.6% global population growth rate, as well as increases in the consumption of all other foods high in animal protein (meat, dairy, milk, etc.). Fish consumption accounted for 7% of all protein consumption and 17% of the global population's animal protein intake in 2017 [2]. In the short time of the antibiotic era, roughly 60 years, tons of antibiotics have been globally dispersed across the biosphere. Twelve thousand tons of eighteen thousand antibiotics are generated annually in the United States for medicinal and agricultural purposes, which contributes to the non-

therapeutic treatment of cattle for growth promotion [3]. Global aquaculture production has generally increased faster than global population expansion and is regarded as one of the most important food production industries. Fish health is now a major concern for aquaculturists because of the increasing commercialization and intensity of aquaculture practices [4]. Degradation of the rearing environment, particularly water quality, is a major problem because it is crucial to the health of aquatic organisms [5, 6]. Poor feed conversion ratios and high mortality rates can lead to a decline in fish output [7]. In the past few decades, antibiotics have been utilized as a conventional approach to manage fish illnesses and enhance development and feed conversion efficiency. Nonetheless, there is ample evidence for the growth and proliferation of bacteria that are resistant to antimicrobial agents [8, 9]. However, antibiotics also cause antibiotic residues to accumulate in fish products, making them unsafe for human consumption. Antibiotics hinder or kill beneficial microbiota in gastrointestinal (GI) ecosystems. Excess ammonia in pond water and sediment, which is brought about by an overabundance of feed, feces, and dead algae deposited on the bottom of the pond, is another significant issue that Indian shrimp culture farmers must deal with. As a result, shrimp are exposed to harmful gases such as NH_3 , NO_2 , and H_2S , which further contribute to eutrophication in the culture system and cause stress in animals. This has led to microbiological illnesses and high mortality rates [10]. In addition to the impacts of drug residues, antimicrobial-resistant microorganisms in aquaculture pose a public health issue. For instance, conjugation between the human-originating bacteria *Escherichia coli* and the bacteria causing fish diseases *Aeromonas salmonicida* has resulted in the transfer of plasmid-borne resistance genes, some of which are harmful to humans [11]. There are two methods by which antibiotic resistance develops: chromosomal mutation and plasmid acquisition. While resistance plasmids can spread quickly to other bacteria, chromosomal changes cannot be passed laterally, leading to a large proportion of pathogenic bacteria that quickly acquire mediated plasmid resistance [12]. Utilizing biotechnological approaches, concerns regarding antibiotic resistance, disease, and seafood demand have been addressed. Probiotic utilization is one of the most successful biotechnological applications in aquaculture. Probiotics are safe live microorganisms that improve water quality, enhance nutrition and immune response in host species, and aid in the competitive exclusion of pathogenic bacteria by producing compounds that inhibit them [13–16].

PROBIOTICS

The word probiotic is a Greek term in which “pro” and “bios,” in the name “probiotic” signifies “for life” [17]. Parker was the first to coin the term “probiotic” (1974). Probiotics are defined as “organisms and substances which contribute to intestinal microbial balance” in his original definition [18]. According to a recent proposal [19], probiotics in the context of aquaculture can be defined as “live or dead, or even a component of the microorganisms that act under different modes of action in conferring beneficial effects to the host or its environment.” The term “live microbial food product which benefits its host (human or animals) by enhancing the microbial balance in the body” was added by Fuller [20], who also stated that it would work under a variety of extreme salinity and temperature fluctuations.

Probiotics are defined by the World Health Organization [21] as living organisms that boost the health of the host when administered in appropriate amounts. The notion of probiotics in aquaculture was suggested to include microbial “water additives” [22]. According to Anjana and Tiwari [23], probiotics can be administered orally, intravenously, or immediately submerged in water. They can be used either separately or in combination [24]. Probiotics have been the subject of an increasing number of studies, and it is now possible to review the status of the science surrounding them, from their empirical application to their scientific methodology [25, 26].

SELECTION OF PROBIOTICS

The safety and production of the desired effects should be the main criteria for choosing probiotics. Prior to reaching the consumer, they must also maintain their abilities during production, manufacture, distribution, and storage [27]. Probiotics must be carefully chosen because incorrect strains may negatively affect their hosts [28, 29]. The following sequence is involved in the selection of probiotics [30]. The sequence of probiotic selection is shown in Figure 1.

- It is necessary to choose a microorganism source.
- By applying selective culture, the microbes that will be used for the task can be isolated and identified.
- A new culture with just the colony of interest is made to perform an in vitro analysis (pathogen inhibition, pathogenicity-targeting species, host resistance conditions, etc.).
- Research on a small- and large-scale exploration of in vivo supplementation is conducted to determine whether the host species benefits from it, provided that there are no restrictions on the application of the target species. Probiotics with favorable results may eventually be produced and marketed.

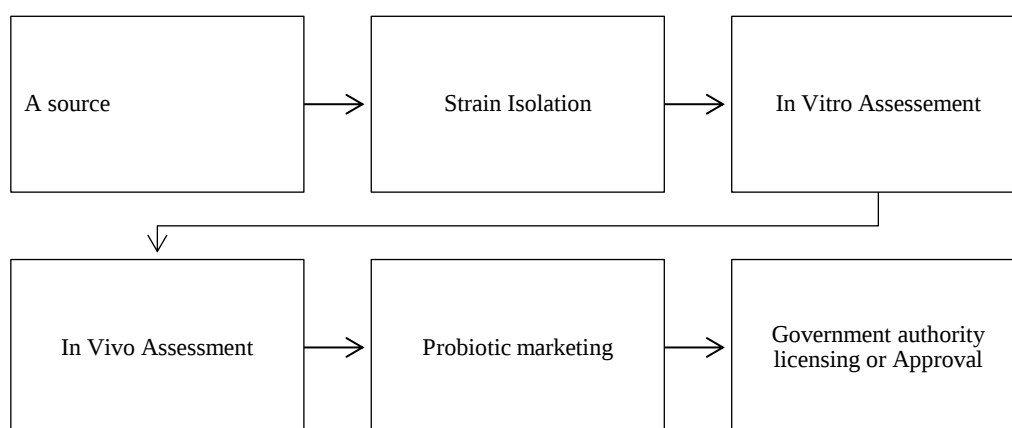


Figure 1. Sequence of probiotics selection.

Source: Adapted from modified [31].

ROLE OF PROBIOTICS IN AQUACULTURE

Aquaculture uses probiotics as part of a practice known as bioprophylaxis, which is the use of living microscopic organisms to prevent illnesses in aquatic animals. The production of bacteriocins by beneficial microorganisms prevents pathogenic organisms from infecting the host or causing diseases is how bioprophylaxis is accomplished [32] (Figure 2).

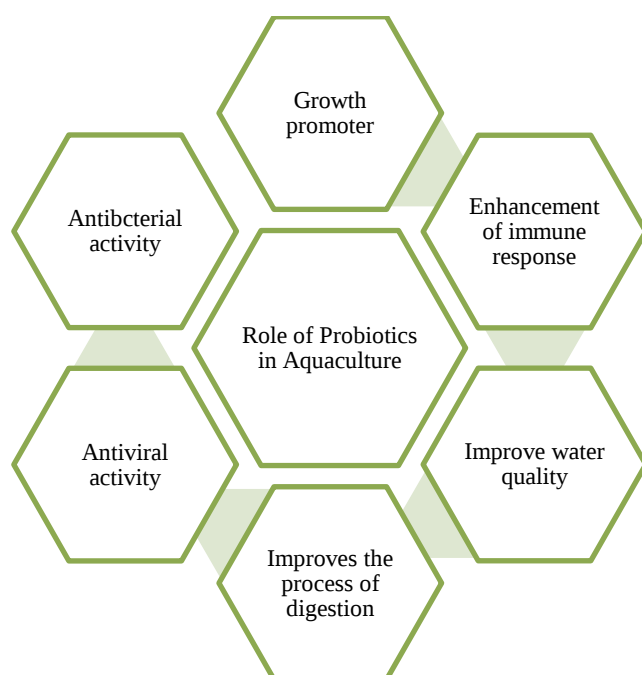


Figure 2. Schematic representation of the role of probiotics in aquaculture.

Growth promoter: The direct impact of probiotics on fish development performance, whether through increased nutrition intake or by supplying nutrients, is one of the most anticipated effects of utilizing bacterial probiotics [33]. Unlike the risk factors linked to commonly used commercially available growth enhancers [34, 35], the growth-promoting action of probiotics on fish is not associated with detrimental health effects [36, 37]. As probiotics are continually added to fish cultures, they adhere to the intestinal mucosa of the fish and develop and exercise their multiple benefits. The author [31] observed that probiotic microorganisms have a more substantial multiplication rate than the rate of expulsion, which allows them to colonize the intestinal tract when administered over an extended period of time. Additionally, probiotics are considered an eco-friendly form of treatment. Probiotics can be added to food in the form of living microbes in the gastrointestinal system to establish a balanced native microfloral population [38]. The probiotic *Streptococcus* strain added to the diet of Nile tilapia (*Oreochromis niloticus*) resulted in a considerable increase in the fish's crude protein and crude fat content, and just nine weeks of culture, the weight of the fish increased from 0.154 g to 6.164 g [39]. To employ probiotics as a tilapia (*Oreochromis niloticus*) growth promoter, Yassir et al. [40] found that *Micrococcus luteus* produced the greatest growth output when used as a probiotic, and the best feed conversion ratio when the same organism was used. The application of *Bacillus* sp. as a probiotic has been shown to increase fish survival by allowing bacteria to colonize the fish's digestive system and culture water [41–43]. Probiotics exhibit varying degrees of efficacy. For instance, *Saccharomyces cerevisiae* and *Bacillus subtilis* showed higher final weights (255.31 g) and specific rates of growth (SGR) (0.77%) at various concentrations [44]. The effectiveness of probiotic applications varies depending on a number of characteristics, including species composition, treatment level, frequency of administration, and environmental circumstances [45, 43], making it challenging to directly evaluate different probiotic studies.

Enhancement of immune response: Probiotics can improve the immunological characteristics of aquaculture species probiotics can prevent the spread of pathogens by boosting the host immune system and stimulating both cellular and nonspecific immunity [46]. According to Taoka et al. [47], tilapia (*Oreochromis niloticus*) treated with viable probiotics exhibited enhanced nonspecific immune responses, as measured by lysozyme activity, neutrophil migration, and bactericidal activity. Lysozyme, a protein present in many vertebrates, including fish, serves as the primary defense against the invasion of Gram-positive and some gram-negative bacteria; it cleaves the β -1, 4 glycosidic bonds between N-acetyl glucosamine and N-acetylmuramic acid in the peptidoglycan layer of bacterial cell walls [48, 49]. Lysozyme levels can rise as a result of feeding diets supplemented with probiotics. It has been shown that giving *Clostridium butyricum* bacteria to rainbow trout increases their ability to fend off vibriosis by boosting leukocyte phagocytic activity [50]. Studies have also shown that providing high doses of probiotics through diet for an extended period of time influences the respiratory burst activity of *O. niloticus* [51], *R. canadum* [52], and *L. rohita* [53, 54]. Probiotic bacteria also compete with one another for resources under aquatic conditions and locations for attachment to the gut. They alter the enzymatic activity in infections. They have demonstrated immunostimulatory properties [55]. Probiotics have a major positive effect on the development of the immune system in juvenile aquaculture species. Because the immune systems of fish larvae, shrimp, and other invertebrates are not as developed as those of adults, their immunity to infection is mostly derived from nonspecific immunological responses [56].

Improved water quality: According to Zhou et al. [57], probiotics have a lot of potential, and it is crucial to provide fish with a healthy habitat. Water quality was monitored in several experiments when probiotic strains, particularly those belonging to the gram-positive species *Bacillus*, were added. This is most likely because this type of bacterium converts organic substances into CO₂ more effectively than gram-negative bacteria do. It has been proposed that fish farmers can reduce the build-up of soluble and insoluble carbon from organic matter during the growing season by maintaining high probiotic levels in their production ponds. Furthermore, this may maintain a balance in phytoplankton production [58]. Ammonia compounds, which are extremely harmful to fish, quickly accumulate when fish excrete nitrogen waste in the form of NH₃ or NH₂⁺ [59]. Nimrat et al. [60] examined the water quality in

treatment and control ponds and demonstrated how the introduction of blended probiotics (*Bacillus sp.*) impacts the development of advantageous bacteria. Because the levels of ammonia, pH, and nitrite in the water were notably lower, they serve as enhancers to improve the quality of the water. Through the nitrification process, ammonia-oxidizing bacteria convert ammonia to nitrite NO_2^- whereas nitrite-oxidizing bacteria convert nitrite to nitrate NO_3^- . Thus, hazardous ammonia build-up in fish-rearing facilities is avoided. In aquaculture plants, aerobic denitrifiers are thought to be ideal choices for reducing nitrate to nitrogen gas (N_2) [61]. Probiotics derived from plant sources, such as yucca extract, potassium ricinoleate, tannic acid, and citrus seed extract, as well as beneficial bacterial species from the genera *Nitrobacter*, *Pseudomonas*, *Enterobacter*, *Cellulomonas*, and *Rhodopseudomonas*, have also been reported to have been used in culture systems noted to have a significant improvement in water quality [62, 63]. Other factors, including pH, temperature, dissolved oxygen, ammonia, and hydrogen sulfide in culturing water, were shown to be of better quality after the application of probiotics. Thus, probiotics maintain the shrimp and prawn larval culture of aquatic systems in a good and healthy environment [64, 65].

Improve the process of Digestion: It is said that through the processes of both aerobic and anaerobic decomposition, microbial metabolism contributes significantly to biogeochemical processes and cycles in the aquatic environment. Probiotics are crucial for recycling organic materials. They also aid in the regeneration of nutrients and the dissolution of insoluble inorganic salts [66]. According to El-Haroun et al. [67] and Mohapatra et al. [68], the majority of probiotics colonize the host and influence digestive processes by increasing the number and generation of microbial enzymes. This improves the gastrointestinal microbial balance and, as a result, the digestibility, absorption, and utilization of feed. Exogenous enzymes are defined as enzymes released by probiotic bacteria. According to Mohapatra et al. [69], these exogenous enzymes can aid in the induction of endogenous enzymes that can withstand a wider pH range than indigenous enzymes, thereby delaying the digesting phase [70]. Proteases are enzymes that secrete and break peptide bonds to release free amino acids that the host can absorb [71]. Because *Bacillus* secretes a variety of exoenzymes, including lipases, carbohydrases, and proteases, which enhance enzymatic digestion and complement the actions of fish, they can help with digestion [72]. Thus, adding probiotics to a meal improves the efficiency of nutrient absorption [73].

LIMITATIONS OF USING PROBIOTICS

Probiotics in aquaculture are limited by the inability of strains to be produced commercially and subsequently demonstrated on a large scale, the challenge of demonstrating efficacy at the farm level, and the inability of companies to undertake comprehensive research on how to make products specifically for aquaculture [74]. Probiotics can be developed in either static or low water exchange environments (circulatory system); they can be used as proactive preventive measures in advance, preventing the disease rather than treating it. They work best when applied as soon as the water medium is sterilized before being contaminated with other microbes [75]. The risk that pathogenic bacteria used as probiotics will mutate and contaminate aquaculture products, probiotic overuse, and the high production costs that will be a burden for small-scale aquaculture farms are all mentioned in an article published in the International Magazine for Animal Feed and Additives Industry (2023). These factors could lead to the introduction of non-native microbes into the aquatic environment, which could alter ecology and impact native species [76].

CONCLUSIONS

Probiotics have potential applications in aquaculture; however, some studies are yet essential as to how we can use some selected probiotics in aquaculture that do not have any negative effects on the health of an organism. It is a viable substitute for chemotherapy and antibiotics, which are harmful and have unfavorable long-term effects. It is well known that 60–80% of operating costs in intensive aquaculture operations are related to feeding; therefore, probiotic usage can reduce feeding costs by improving feed consumption and, consequently, improving growth performance. Probiotics assist host animals by improving development and nutritional digestibility, boosting disease resistance, and improving the quality of culture water. Selecting the appropriate strain based on favorable traits is the

most important stage for boosting the effectiveness of probiotics. Nonetheless, further investigation should be conducted to estimate the right amounts and doses added to meals, in a group of bacteria, and in a variety of physical and chemical elements that influence probiotics need to be researched. Probiotics have potential applications in aquaculture; however, further studies are required. They may have beneficial or detrimental effects on the surrounding environment as well as aquaculture animals. Although there are metal removal methods that use physiochemical approaches, they are costly and have a negative environmental impact. Understanding these mechanisms is necessary to ensure long-term commercial use, despite a slightly less rigorous approach. This is because it helps to determine any potential environmental side effects, such as whether adding probiotics permanently alters the community of microbial species and how this will affect the turnover of inorganic and organic molecules in a specific environment. Numerous studies have demonstrated that the addition of organisms more frequently, and not relying solely on a single probiotic culture treatment, is necessary. However, the strength of the systems, such as the necessary concentration of probiotics, frequency of additions, and impact of temperature changes, have not been measured; therefore, some aspects in detail are yet to be studied for further development.

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