



## Probiotic applications in aquaculture for disease prevention and growth enhancement

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### Abstract

Probiotics can serve as a valuable alternative to boost aquaculture productivity sustainably. The appropriate selection of strains and dosage for specific fish species is essential for achieving the intended advantages of probiotic treatment. Probiotics are administered as water additives, feed additives, or via injections, with the latter being extensively utilized in fisheries. The use of probiotics in the aquaculture industry has numerous advantages, including improved growth efficiency, enhanced feed utilization, strengthened immunological defense against pathogens, increased disease resistance, higher-quality water, and greater stress endurance. The utilization of probiotics in farmed animals can be implemented at the farm scale to improve the economic viability of aquaculture animals.

**Keywords:** Probiotic, Aquaculture, Disease prevention, Enhancement, Fish

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## Introduction

Aquaculture refers to cultivating aquatic creatures in marine and inland environments, incorporating treatments in how they are raised to optimize productivity (Verdegem *et al.*, 2023). Global farmed fish manufacturing has significantly increased over the past 74 years, from under one million tons in the first half of 1960 to a staggering 85 million tonnes, valued at USD 350 billion in 2020. Aquaculture's contribution to global fish output increased to 45 per cent in 2020, compared to 26.2 per cent in 2010. From 1960 to 2020, worldwide food consumption for fish rose at a yearly mean of 4.3 percent, which is nearly double the average annual globe population rise of 1.8 percent during the same time frame, and surpassing the growth rate of all other sources of animal protein (flesh, dairy products, milk, etc.), which climbed by 2.3 percent annually (Chatterjee and Sanyal, 2024). In 2018, seafood consumption constituted 18% of the worldwide population's animal-based protein diet and 8% of total protein intake. The data indicates that global aquaculture saw a rapid expansion, with a yearly increase of 5.4 percent from 2000 to 2020 (Çiftçi and Ayas, 2022). The data suggests that worldwide fish consumption has risen at a greater rate than other sources of protein from animals. Aquaculture production methods have been significantly strengthened through technical and functional advancements to satisfy the escalating needs of the expanding worldwide population (Verdegem *et al.*, 2023).

The growth and marketing of the aquaculture industry to satisfy demand presents numerous challenges, including illness and epizootic administration, egg enhancement and cultivation, formulation of suitable feed and feeding strategies, hatcheries and grow-out technological advances, and water quality management (Afewerki *et al.*, 2023; Kumar and Sajjan, 2024). Epidemics are now a significant limitation on cultivating numerous species, adversely impacting aquaculture productivity and hindering economic and social growth in various nations. Infections have resulted in substantial financial losses to the worldwide finfish aquaculture sector, ranging between US\$ 1.4 billion and US\$ 9.6 billion annually. Medicines in aquaculture have been employed as a conventional method for controlling and avoiding the spread of fish illnesses and for enhancing growth and feeding effectiveness (Zhang *et al.*, 2022). The application of antibiotics in aquaculture commenced with recognizing the potential use of antibiotics, specifically sulfonamides, for treating the disease. The World Health Organisation (WHO) expert conference on antimicrobial administration and resistant bacteria in farming concluded that residual and resistant substances are the two primary dangers of antimicrobial usage in agriculture (Farías *et al.*, 2024). The indiscriminate administration of antibiotics leads to the emergence of antibiotic-resistant microbes. It disrupts intestinal microbiota, adversely impacting fish health and causing residual accumulation in fish muscle, posing a possible health risk to consumers. In light of the risks associated

with antibiotic usage in aquaculture, probiotics serve as an alternative strategy for managing fish health in the sector, demonstrating improvements in fish health and, in many cases, the development of fish. Numerous fishermen have identified various non-antibiotic representatives, among which probiotics are crucial in maintaining health within intensive farming. Antibiotics can improve growth, feed efficiency, bolster resistance to illness and immunological reaction, and elevate water quality in agriculture (Yang *et al.*, 2024). This review delineates the choice, kinds, administration techniques, and advantages of probiotic use for aquaculture animals.

## Background

Fish constitutes the most abundant animal protein and represents the most rapidly growing sector in global food production (Boyd *et al.*, 2022). Globally, individuals derive approximately 28% of their protein from animals, such as seafood and fish, and the market for these products continues to increase. Productivity can be optimized by including commercial feeds, growth boosters, antimicrobial agents, and additional additives.

Utilizing these goods undoubtedly results in increased production; a significant concern is that their habitual application causes serious problems, raising questions about sustainability. Probiotics have been used in farming for an extended period; they have become an essential component of farming techniques to enhance growth and resistance to disease (Chamakuri and Janapana, 2024; Saritha and Gunasundari, 2024). This technique

provides numerous benefits to mitigate the restrictions and adverse effects of penicillin and other pharmaceuticals, while also facilitating increased production via improved growth and illness control (Nasrollahi *et al.*, 2022).

The Greek term "probiotic," meaning "for life," existed. Antibiotics are living creatures and chemicals that promote intestinal and bacterial balance. The term beneficial bacteria is defined as a live microbe that positively influences the host by altering the host-associated bacterial community and its interconnected system, increasing the utilization of feed or augmenting its nutritional value, enhancing the host's response to disease, or ameliorating the environment around it. Per the criteria established by the WHO, probiotics are live bacteria that, when provided in enough quantities, provide a health benefit to the host body (Archana Menon and Gunasundari, 2024).

A probiotic living thing must exhibit immunity to acidic gastric conditions, bile, and pancreatic enzymes; possess the ability to adhere to digestive mucosal cells; demonstrate prolonged settlement capacity; remain viable for effective host colonization; produce antimicrobial agents against infectious microbes; and be non-pathogenic, environmentally friendly, and devoid of relocation. Probiotic organisms are at the forefront of current studies in aquaculture due to their remarkable efficacy in enhancing fish development and health while promoting environmental sustainability. Numerous studies have documented growth augmentation after using probiotics in aquaculture (Mohammed *et al.*, 2025). Elevated activities of lysozyme, oxidase

Super Oxide Dismutase (SOD), Catalase (CAT), enzyme protease anti-protease, and Immunoglobulin M (IgM) the amount were also documented in urine and surfaces of the mouth, serving as crucial defense particles toward various infections in mass-cultured aquatic life.

Probiotics are bacteria purported to confer health advantages upon consumption; the phrase gained prominence post-1990 and was frequently utilized from 1995 onward. Probiotics are live microbial feed supplements that positively influence the host mammal by enhancing its intestinal microbial equilibrium. The inception of the idea, albeit not the terminology, is primarily ascribed to Nobel laureate bacteriologist, who hypothesized that Bulgarian farmers who consumed yogurt experienced increased longevity. The reliance of intestinal microbes on dietary intake enabled the implementation of strategies to modify the microbiota in the body, substituting detrimental germs with beneficial microbes (Pereira *et al.*, 2022).

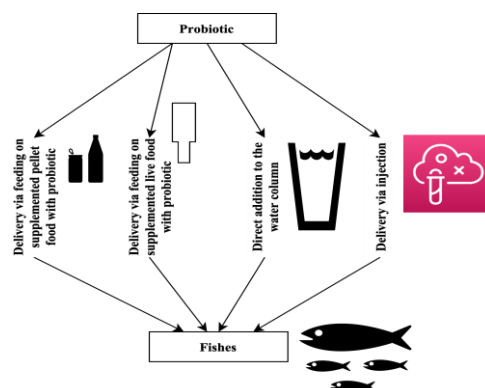
Probiotics are live bacterial feed supplements that positively influence the host mammal by enhancing its gastrointestinal microbial equilibrium. When consumed in adequate amounts, a live microorganism can benefit the person who consumes it by strengthening the gut microbial equilibrium. A comprehensive definition of antibiotics in fish farming is: "live, deceased, or components of microbial cells that, when introduced through feed or raising water, confer advantages to the organism in question by enhancing immunity to diseases, health status, development, feed efficiency, reaction to stress, or overall vitality, primarily through enhancing

their microbial equilibrium or by restoring the microbial equilibrium of the surrounding surroundings." Another description of antibiotics is utilized in farming as "live microbes that are incorporated into feed or surroundings (water) to enhance the vitality (surviving) of the animal."

## Administrative Methods

### *Method of Action of Probiotics*

The selection of probiotics is contingent upon their ability to colonize, inhibit infections, and consequently produce advantageous chemicals such as vitamins, lipids, and gastrointestinal enzymes. The effective use of probiotic varieties as microbial components in fish necessitates additional qualities, such as good viability throughout manufacturing, storage, and post-gastrointestinal transit. A probiotic dose frequently yields beneficial and adverse outcomes in various individuals, whose reactions to differing dietary probiotic concentrations are noted. Significant. The methods of action of probiotics encompass the improvement of the epithelial barrier, enhanced adherence to the intestinal tissue, the competitive elimination of harmful microbes, and the generation of antimicrobial compounds as follows in figure 1:



**Figure 1: Routes for probiotic admission.**

- Augmentation of the epithelial barriers: The intestinal wall is a primary defense system that sustains epithelial integrity and safeguards living things from environmental threats.
- Enhanced adherence to the intestinal epithelium: Adherence to the intestinal wall is essential for colonisation and the relationship between probiotic varieties and their host.
- Competition in eradicating infectious microorganisms: Probiotic organisms adhere to binding locations in the lining of the intestines, establishing a material barrier that obstructs the attachment of pathogens.

The probiotic strain *Vibrio lentus*, administered as a water addition at a  $10^6$  CFU/ml concentration in sea basses, significantly altered gene activity related to immune response, cell growth, apoptosis, cell adhesion, reactive oxygen species digestion, and iron transportation. Application via injection is also feasible. The administration of the probiotics *Enterobacter* sp. strain C6-6 via intravenous and peritoneal routes augmented immunity in rainbow-colored trout.

#### *Individual and Composite*

Probiotics are utilized individually or in conjunction and are available in various forms, including multistrain probiotics, which are combined with plant extracts, and antibiotics with yeast extracts. Most research on probiotics in farming has been on individual probiotics; however, a mixture of probiotics proves more advantageous. Multi-strain probiotics benefit from heightened sensitivity to

infections and efficacy against various aquaculture species. The beneficial effects of multi-strain antibiotics on the development and longevity of rohu were observed during the newborn and fry phases, but not in subsequent stages. The combined use of both *Bacillus megaterium* and the bacterium *Pentosaceus* administered in feed demonstrated superior results compared to the individual application of each antibiotic in zebrafish *Clarias water* sp. The concurrent treatment of *Lactobacillus plantarum* strain and *Bacteria velezensis* resulted in a superior survival rate of 59.4%, compared to the individual applications of *Lactobacillus plantarum* at 55.2% and *Bacteria velezensis* at 42.8%. The synergistic use of probiotics, *Bacillus coagulans*, and extracts of plants, *Mentha piperita*, in conjunction demonstrated superior development, retention of nutrients, and resistance in *Catla catla* compared to their applications.

#### *Dosage*

The correct dosage must be administered to achieve optimal efficacy of the probiotics for the specific species. Optimal probiotic levels are contingent upon the bacterial organisms, types of fish, their physical circumstance, the raising environment, and the particular usage objectives. Growth rates increased when brook trout were fed probiotics, the bacterium B variants at  $1 \times 10^6$  CFU/g, but not  $3 \times 10^6$  CFU/g. This indicates that a rise in bacteria inside feed does not inherently lead to a proportional enhancement in development. The dosage is a critical factor when adding functional elements in aquafeeds.

Weight increase was diminished when dietary probiotics from commercial sources exceeded 0.2% in pabda fish, such as catfish. The findings indicated that elevated concentrations disrupt the overall physiological balance of fish, perhaps causing problems in glucose and fat absorption.

#### *Resistance to Illness*

Studies on probiotics and prebiotics in fish have demonstrated encouraging outcomes in combating numerous diseases, particularly viral, bacterial, and parasite infections. The impact of probiotics on preventing prevalent bacterial illnesses in aquaculture was elucidated. When incorporated into the fish feed, specific probiotic varieties have been shown to diminish the occurrence of bacteria-related illnesses by fostering a healthy microbiota in the gut, hence augmenting the fish's defenses. Another study offered significant insights into using probiotics to combat bacterial diseases across several fish species. The results indicated that specific probiotic supplements significantly inhibited the proliferation of harmful microbes in the animal's gut, consequently reducing infection rates. Parasitic infections, despite the scarcity of information available to investigators, have inflicted significant harm on the fishery. A study on the effects of bacteria on parasitic infections in fish indicated that specific probiotic supplements modified the intestinal environment, rendering it less conducive to parasite growth and survival, reducing parasite populations and enhancing overall fish health. Probiotics significantly influence the prevention and management of parasite illnesses in numerous fish species by

modifying their immune system and gut microbiome.

Countless viral diseases have been documented in fishing, impeding the industry's expansion and growth. Studies have shown that probiotics with particular strains improve immunity against viruses in fish, decreasing vulnerability to pathogenic viruses and mitigating the severity of illnesses. The research elucidated probiotics' effectiveness in managing virus outbreaks in aquaculture facilities.

Their research has shown that consistent probiotic treatment diminished virus levels and strengthened fish immune defenses, considerably reducing viral infection frequencies. These research investigations collectively highlight the possible use of probiotics as a prophylactic strategy against common illnesses in fish, illustrating their efficacy in diminishing the frequency and extent of viral, bacterial, and parasitic diseases by enhancing the fish's defenses and gut health.

#### *Reproductive Promoter*

Probiotics have become a significant focus in aquaculture studies, especially regarding their influence on fish reproduction. These advantageous bacteria have been shown to affect multiple facets of fish reproduction, including gamete quality and larval longevity. The application of microorganisms in fish growth signifies a broader movement towards ethical and ecologically conscious fish farming.

Probiotics primarily enhance fish reproduction by improving livestock's general wellness and immunological function. Probiotic addition in the diet of

zebrafish enhanced fertility and the hatching process. The investigators noted elevated implantation and incubation rates for eggs from females fed probiotics compared to the untreated group. They ascribed these enhancements to the microbes' capacity to regulate the activity of genes associated with reproductive and metabolic functions.

Probiotics have demonstrated the ability to improve sperm quantity in male fish. Research on typical carp has shown that dietary treatment with lactic acid bacteria markedly enhanced sperm movement, survival, and content. The investigators posited that these enhancements were probably attributable to the microorganisms' antioxidant characteristics and capacity to augment nutritional absorption.

In addition to gamete excellence, antibiotics have enhanced larval growth and survival. Investigations on European bass indicated that probiotics administration to larvae led to increased survival rates, potentially correlating with improved reproduction in subsequent stages. The research observed enhanced immune system growth in larvae fed with probiotics, potentially benefiting fish health and reproduction in the long term. The processes by which probiotics affect fertility in fish are complex. A primary mechanism is the regulation of the gut-brain nexus. The injection of bacteria in zebrafish modified the transcription of genes related to hunger regulation and fertility in the brain, indicating the potential of probiotics to affect the reproduction cycle via neuroendocrine systems.

Probiotics improve fish's dietary requirements, facilitating reproduction

activities. Research on the rainbow trout indicated that a probiotic regimen enhanced the digestibility of proteins and the uptake of amino acids. This improved diet promotes the creation of gametes and overall reproductive efficacy. The influence of microorganisms on the development of fish includes stress alleviation, which is essential for good reproduction efficacy. Stress can adversely impact reproductive procedures in fish, resulting in diminished gamete quality and hatching success. Probiotic administration reduced cortisol levels and inflammatory indicators, potentially fostering a more advantageous health state for reproductive.

### **Advantages of Probiotics in Fish Farming**

#### *Enhance Feed Consumption*

Numerous studies have established that incorporating probiotics modifies enzyme activity, enhancing feed consumption. Adding heat-killed *plantarum* bacteria at concentrations of 60, 120, or 1500 mg/kg for twelve weeks dramatically increased the activity of all three enzymes in Nile tilapia. An increase in the levels of protease, amylase, and alkaline phosphatase was noted in narrow-clawed clams, *Astacus leptodactylus*, administered *Lactobacillus plantarum* at concentrations of  $10^7$ ,  $10^8$ , and  $10^9$  CFU/gm.

#### *Growth Promoters*

A primary anticipated outcome of employing bacterial antibiotics is their direct influence on the growth efficiency of fish, either through enhanced nutrient absorption or by supplying essential

nutrients. The notable rise in development outcomes from probiotic treatment results from the increased secretion of digestive enzymes, hunger stimulation, vitamin generation, degradation of indigestible substances, and modification of gut shape. Antibiotics have been extensively utilized in shellfish aquaculture as growth enhancers. The addition of *Bacillus subtilis* at  $10^5$  CFU/kg food for 5 weeks significantly enhanced the development of Pacific white prawns. The dietary addition of *E. faecalis* and the bacterium *acidilacti* markedly enhanced the weight gain and the specific growth rate of the mud lobster, *Scylla paramamosain*. Antibiotics are similarly utilized in decorative fish—the intake of a type of *acidophilus* enhanced development performance in zebra mussels without compromising appetite.

#### *Enhance Illness Opposition*

Microbes can secrete chemical compounds with bactericide or bacteriostatic effects against pathogenic microbes residing in the host's intestine, such as a substance called polymyxin generated by *Bacillus* species, thereby improving disease resistance, through the addition of the probiotics *Enterobacter* species. Augment disease resistance to flavor bacteria *psychrophilum* in rainbow brook trout.

Adding *Lactococcus Garvieae* (LG), obtained from raw cow milk at  $10^6$  cells/gm for 10 days, enhanced immunity to *Staphylococcus aureus* in tilapia from the Nile. The addition of *lactis*, a microbe extracted from the gut of *Cromileptes altivelis* (CA), at doses of 10%,  $10^8$ , and  $10^{10}$  CFU for four weeks improved immunity to *Vibrio harveyi* infection in

CA. The injection of *Enterococcus faecalis*, drawn from a packaged probiotic at  $10^8$  CFU/g for thirty days, enhanced resistance to infection over LG in rainbow trout.

#### *Augmentation of the Immune Response*

Antibiotics can improve several immunological markers in aquaculture animals. Probiotics can impede pathogen infections by augmenting the host's defenses by activating nonspecific and immune cells. The ingestion of viable lactic acid bacteria augmented both cell-based and hormonal resistant responses in the trout species by elevating the number of phagocytically dividing cells from the upper kidneys and stimulating the expression of match receptors. Probiotics linked with the host, like LG *plantarum* and *Bacillus velezensis*, improved innate immune parameters in Nile tilapia, including skin mucus, this enzyme and oxidase action, blood lysozyme, serum peroxide, alternative complements, phagocytosis, and pulmonary burst capabilities. Adding the gut-derived probiotics *Bacillus pumilus* in juvenile yellow pompano, *Trachinotus ovatus*, enhanced lysozyme function and total protein content in the fish. The consumption of lactic acid bacteria at  $10^8$  and  $10^{10}$  CFU/g markedly elevated complement activities after 15 days and considerably improved pulmonary burst activities and lysozyme functioning in black eared cats, *Pangasius larnaudii*, after 30 days and 40 days of nutrition, respectively. Adding feed with *Bacillus subtilis* at  $10^7$  and  $10^8$  CFU/kg in white squid improved the immune system by enhancing phagocytic function and clearing effectiveness. The intake of supplements of the bacterium



Pentosaceus in typical carp leads to a rise in red blood cells, white blood cells, serum total antibodies levels, alternative complement activity, proteolytic and lysozyme activity, and antimicrobial activities.

### *Enhance the Quality of Water*

Good bacteria can immediately assimilate or break down organic materials or harmful substances in the water, enhancing its quality. Probiotics can break down fish or shrimp urine, residual food, phytoplankton remnants, and other organic matter into CO<sub>2</sub>, nitrate, and phosphates, enhancing the nutrition process that sustains a favorable aquatic atmosphere for cultured organisms. The ingestion of combined *Bacillus* bacteria considerably improved the water's quality concerning pH, ammonium N, and nitrite concentrations in the cultivation water throughout the growing of white prawns.

Incorporating probiotic *Bacillus subtilis* at concentrations of 10<sup>3</sup>-10<sup>5</sup> CFU/ml significantly diminished overall ammonia levels and improved the water's condition in shrimp cultivation. Infusing probiotics known as *Bacillus cereus* and the bacterium *acidilactici* at 10<sup>6</sup> CFU/ml into the pond water significantly reduced nitrate and nitrogen dioxide levels and the biological requirements for oxygen.

### **Conclusion**

Aquaculture is now recognized as the fastest-growing business, providing high-quality protein from animals that enhances nutritional and dietary safety. The expanding aquaculture industry has several limits, including disease outbreaks, elevated stress illnesses, and a scarcity of fish meal for nutritional

supplies. These issues are conventionally addressed with antibiotics and chemical-based disinfectants, which present concerns regarding food safety for humans and aquatic organisms due to their lingering effects, ultimately resulting in pollution of the environment. In this sense, probiotics signify a transformative advancement in contemporary aquaculture as a feasible alternative for sustainable practices. Antibiotics can be utilized to create optimal conditions for developing species used in aquaculture, hence improving their well-being. Choosing a suitable strain is crucial for optimizing the efficacy of probiotics before their application. Only the efficacious probiotic at the optimal dosage exerts the most favorable effect on specific species.

Among the several methods of probiotic management, supplementing as food additives is the most prevalent in fishing, while distribution as water additives is the most practical approach. The giving of feed presents challenges during the initial stages of larval development due to the underdeveloped digestive systems of fish, while injection induces significant stress in larval fish. Therefore, incorporating probiotics straight into the raising water is feasible at all developmental phases.

Probiotics augment digestive enzyme activity, promote feed effectiveness, and increase feed consumption. In intense fishing, feeding accounts for 65-85% of operational costs; introducing probiotics might mitigate these costs by improving feed consumption and thus improving the growth rate. Antibiotics can improve farm animals' health by augmenting

beneficial microbes and inhibiting harmful ones.

Antibiotics can also augment the body's natural defense system against fish infections. Using probiotics mitigates ecological and physiological strain in farmed animals by lowering blood sugar and cortisol levels. Probiotics can improve water quality by diminishing the concentration of organic debris and harmful substances. Despite the advantageous effects of probiotics on fishery management, their application in studies has been restricted and not extensively utilized at the farm level. The numerous limitations of probiotics in farming include the inability to develop sufficient strains from aquatic organisms and the failure of enterprises to manufacture probiotic products specifically for the aquaculture sector, resulting in the predominant usage of terrestrial antibiotics in this sector.

Considering the numerous advantages of bacteria in aquaculture, fishermen and other participants in aquaculture administration should integrate probiotics into feed formulas and utilize them as a preventive measure against diseases instead of antibiotic treatment. Farmers should be informed about the administration procedures and the advantages they provide to expand the utilization of probiotics at the farm level. To mitigate the limitations of probiotics in fishing, a collaborative network of fishery professionals, fish nutritionists, and microbiologists is essential.

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