



## Research Progress on the Application of Probiotics in Aquaculture

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### Abstract

With the global aquaculture industry transforming towards green intensification, microecological preparations, as the core carrier of antibiotic alternatives, have achieved leapfrog development in their research and application. This article systematically reviews the classification system and innovation in the mechanism of action of microecological preparations, with a focus on analyzing the application effects of probiotics, prebiotics, and synbiotics in water quality regulation, disease prevention and control, and growth enhancement. Studies have shown that the screening system combining genomics with multi-dimensional experiments, AI-driven intelligent application technologies, and the synergistic effects of compound preparations have become research hotspots. However, issues such as stability control and standardized production still need to be addressed. In the future, it is necessary to promote the large-scale application of microecological preparations in aquaculture through interdisciplinary technology integration and standardization construction.

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### 1. Introduction

Aquaculture, as a core pillar of global animal protein supply, is facing the dual crises of antibiotic resistance and ecological pollution caused by the abuse of antibiotics under the dual pressures of population growth and resource constraints. Data shows that the resistance rate of pathogenic bacteria such as *Vibrio parahaemolyticus* to commonly used antibiotics like enrofloxacin has exceeded 85%, and a single disease outbreak can cause a 30%-50% reduction in production for aquaculture enterprises. Microecological preparations, with their characteristics of "environmental friendliness, targeted regulation, safety and high efficiency", have become a key means to solve the industry's predicament.

### 2. Classification of Probiotics

Probiotics are biological agents developed based on microbiological principles. They contain active microorganisms and their metabolites, and are composite products that maintain and improve the health of the host through microorganisms and their related components.

#### 2.1. Classification by Composition

Microecological preparations can be divided into probiotics, prebiotics, and synbiotics according to their composition. Probiotics can increase the number of beneficial bacteria in the intestines, regulate the balance of intestinal microbial communities, and enhance the host's immune system function and overall health. Prebiotics refer to substances that are not easily completely digested by the animal's digestive tract or are mostly undigested. They can be absorbed and fermented by beneficial bacteria in the intestines, promote the growth of certain beneficial bacteria in the host's intestines, and improve the body's non-specific immune function. In animal breeding, common prebiotics include mannan oligosaccharides,  $\beta$ -glucans,

transgalactooligosaccharides, fructooligosaccharides, and lactulose. Synbiotics, also known as symbiotics, are composite preparations that mix probiotics and prebiotics in a specific ratio and can achieve the combined effect of the two. After animals ingest synbiotics, the probiotics in them can better colonize and reproduce in the intestines with the help of prebiotics, making beneficial bacteria dominant in the intestinal microbiota and enhancing the host's immunity.

## 2.2. Classification by Bacterial Species

Microecological preparations can be classified into Bacillus, lactic acid bacteria, yeasts, photosynthetic bacteria, and compound microecologies based on their bacterial species. Bacillus are Gram-positive bacteria, which have the advantages of high stability and ensuring the quality of feed processing. Bacillus are stored in the form of spores, with low loss, resistance to gastric acid, rapid reproduction, and a short growth cycle. The digestive enzymes they secrete can decompose complex carbohydrates. The commonly used Bacillus preparations in aquaculture are Bacillus subtilis and Bacillus licheniformis. Lactic acid bacteria refer to a general term for Gram-positive bacteria that produce lactic acid as the main metabolic product. The main functions of such preparations include: reducing pH value by producing lactic acid, secreting bacteriostatic substances to inhibit harmful bacteria; providing nutrients such as amino acids and vitamins to promote growth; activating digestive enzymes to improve feed efficiency; enhancing non-specific immunity; and improving water quality. Yeast is a facultative anaerobic microorganism, which can provide high-quality protein, promote the growth of beneficial bacteria, inhibit harmful bacteria, regulate intestinal flora, and enhance immunity. The commonly used yeasts in aquaculture are mainly yeast extract, yeast cell wall, and yeast culture. Photosynthetic bacteria are a type of prokaryote that can use light energy for metabolism. Photosynthetic bacteria can improve feed utilization, significantly increase the growth rate of fish, and also play a role in purifying water quality. In addition, photosynthetic bacteria can promote the growth of beneficial algae (such as diatoms and Chlorella) and inhibit the reproduction of harmful algae (such as cyanobacteria). Compound microecological preparations are microbial preparations composed of two or more probiotics mixed in a certain proportion, and the application effects of different compound microecological preparations vary. When Bacillus subtilis and Bacillus licheniformis are added, the feed efficiency and specific growth rate of rainbow trout are significantly higher than those of the single bacterial species group. However, the test results of adding photosynthetic bacteria + Bacillus to the feed of juvenile sea cucumbers show that the growth performance of the single bacterial species group is better.

## 3. The role of probiotics in aquaculture

### 3.1. Regulating immunity

Probiotics can enhance the body's immune response by activating the activity of B cells and macrophages, thereby stimulating the body's own immune defense system. The antibacterial substances produced by lactic acid bacteria can inhibit the growth and reproduction of harmful bacteria, thereby reducing the damage of pathogenic microorganisms to aquatic animals; the active ingredients in probiotics can stimulate the mucosal lymphoid tissue of the intestinal tract of aquatic animals to secrete related digestive enzymes,

improve the activity of digestive enzymes, improve the digestive and absorption functions of the body, and thus enhance the disease resistance of the body. Studies have shown that there are Toll-like receptors, formylated peptide receptors on the intestinal mucosa of animals, as well as intracellular NOD-like receptors (NLRs). These receptors can specifically recognize and bind to the ligands of microbe-associated molecular patterns (MAMPs) (lipopolysaccharides, flagellins, peptidoglycans, and formylated peptides). Probiotics can interact with the mucosal immune system of animals, and alleviate the body's inflammatory response by regulating the growth and metabolism of small intestinal epithelial cells and mucosal tissues.

### 3.2. Maintaining Intestinal Health

In the intestines of healthy aquatic animals, the microbial flora is mainly composed of anaerobic bacteria, with aerobic bacteria accounting for only about 1%. The types and quantities of these microbial flora maintain a dynamic balance. Aerobic probiotics in microecological preparations help anaerobic bacteria regain their dominant position by consuming oxygen in the intestines. Acetic acid, hydrogen peroxide, and other active substances produced by probiotics during their growth and metabolism can not only inhibit the growth and reproduction of pathogenic bacteria but also help maintain the balanced state of the intestinal flora in aquatic animals.

### 3.3. Improving Feed Digestibility

After probiotics successfully colonize and reproduce in the intestinal epithelium, they release various compounds beneficial to the host's growth (such as vitamin B, coenzyme Q, growth-stimulating factors, and multiple digestive enzymes). These compounds can work synergistically with the host's own digestive enzymes to improve the digestive utilization rate of feed, thereby promoting the healthy growth of the organism. Most probiotics and their metabolites contain rich nutrients. For example, Saccharomyces cerevisiae contains high-quality protein, oligosaccharides, vitamins, selenium, and other trace elements. Bacillus, after colonizing the intestinal tract, can secrete digestive enzymes such as amylase and compound protease. These enzymes can break down plant cell walls, thereby improving the absorption efficiency of nutrients by intestinal epithelial cells.

### 3.4. Improving Water Quality

Adding microecological preparations to aquaculture water can effectively decompose harmful substances in the water, inhibit the growth of harmful bacteria and algae, thereby purifying the water quality and reducing the risk of diseases in aquatic animals. Studies have found that Bacillus and EM bacteria can decompose ammonia nitrogen and nitrite in water. Microecological preparations can convert harmful substances in water (ammonia nitrogen, nitrite nitrogen and sulfides) into relatively harmless nitrates. These nitrates can be absorbed and utilized by beneficial algae, which produce oxygen through photosynthesis for aquatic animals to breathe, thereby maintaining a healthy water cycle. Some studies have pointed out that the use of EM bacteria in seawater shrimp culture ponds can increase the dissolved oxygen level in the water.

#### 4. Conclusion

Probiotics have upgraded from auxiliary aquaculture products to core technical support for green aquatic products. Their application has realized the transformation from single water quality improvement to dual regulation of "water body - intestine" and from empirical use to precise application. Breakthroughs such as genomics-driven strain screening, improved stability through embedding technology, and AI-empowered intelligent applications have provided technical guarantees for industrial transformation. Although there are still challenges in standardized production and adaptation to extreme environments, with the integration of interdisciplinary technologies and strengthened policy support, probiotics will surely play an irreplaceable role in the sustainable development of aquaculture and contribute core strength to global food security and ecological security.

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