

7 Case Study: Evaluation of the Application Effects of Compound Probiotic Preparations in Grass Carp Aquaculture

7.1 Effects of compound probiotic supplementation on growth performance and feed efficiency of grass carp

Compound probiotic supplementation generally improves growth performance and feed utilization in grass carp. In a 6-week feeding trial, a duo-strain preparation of *Bacillus subtilis* and *Lactobacillus plantarum* significantly increased percent weight gain, specific growth rate, and survival rate, while decreasing feed conversion ratio, showing that mixed-strain delivery can enhance both production and feed efficiency. A separate grass carp study using *Lactobacillus buchneri* L3-9 together with its extracellular products also increased weight gain rate and reduced feed conversion ratio within only 21 days, indicating that compound microecological preparations can act rapidly under culture conditions (Gao et al., 2024).

These responses likely reflect complementary functions among strains and bioactive metabolites rather than a simple additive effect. Probiotics are understood to promote growth through improved digestion, enhanced nutrient absorption, and better feed conversion, which is especially relevant in carp farming where feed remains the major production cost. Supporting evidence from other carp models shows that mixed probiotic preparations can reduce feed conversion ratio by 5.04-6.47% and generate measurable feed savings at farm scale, suggesting that the performance gains observed in grass carp are economically meaningful when translated to commercial production (Prazdnova et al., 2025).

7.2 Intestinal microbiota and immune regulation

Compound probiotics regulate the grass carp intestinal ecosystem by enriching beneficial taxa and suppressing potential pathogens. In the duo-strain grass carp study, probiotic feeding increased the relative abundance of Bacteroidetes and Firmicutes while decreasing Proteobacteria and Cyanobacteria, indicating a shift toward a more favorable microbial structure (Luo et al., 2022). Earlier work in grass carp also showed that combined administration of *Shewanella xiamenensis*, *Aeromonas veronii*, and *Bacillus subtilis* increased *Cetobacterium* and reduced potential pathogens such as *Pseudomonas* and *Flavobacterium*, linking microbiota remodeling to lower disease risk.

Immune regulation appears to follow these microbial shifts. Dietary supplementation with *L. buchneri* L3-9 and its extracellular products increased Lactobacillaceae, reduced Aeromonadaceae and Enterobacteriaceae, downregulated the pro-inflammatory gene TNF- α , and upregulated anti-inflammatory markers including TGF- β 1 and IL-10 (Gao et al., 2024). More broadly in juvenile grass carp, probiotic combinations or multiple candidate strains elevate complement components and antioxidant defenses while improving intestinal barrier-related signaling, suggesting that compound preparations work through coordinated effects on microbiota, mucosal integrity, and innate immunity rather than through one pathway alone.

7.3 Water quality and economic value

The broader value of compound probiotics in grass carp farming lies in coupling host benefits with environmental regulation. Probiotics can improve freshwater pond conditions by lowering ammonia, nitrite, and organic loading, and average effects across freshwater fish studies include a 50.7% reduction in ammonia and a 10.7% decrease in feed conversion ratio, both of which directly support more sustainable production. Mechanistically, water-applied probiotics can promote nitrification and denitrification by increasing functional genes such as *amoC_B*, *hao*, *nirS*, and *nosZ*, which explains why improvements in water quality often persist beyond the immediate feeding response (Mang et al., 2024).

Although direct pond-scale case studies in grass carp remain fewer than feed-based trials, related high-density carp systems show the same integrated pattern of benefit. In crucian carp, long-term use of compound probiotics under micro-water-exchange conditions kept total ammonia nitrogen and nitrite in dynamic equilibrium, improved body weight and length, and lowered mortality after bacterial challenge. Likewise, probiotic use in biofloc-based carp culture reduced NH₃-N and NO₂⁻, improved growth and welfare, and supports the conclusion that compound