Single and Multi-Strain Probiotics Supplementation in Commercially Prominent Finfish Aquaculture: Review of the Current Knowledge

Md Afsar Ahmed Sumon1†, Tofael Ahmed Sumon2†, Md. Ashraf Hussain3†, Su-Jeong Lee4, Won Je Jang4,5, S. M. Sharifuzzaman6, Christopher L. Brown7, Eun-Woo Lee4,8, and Md. Tawheed Hasan8,9†*

1Department of Marine Biology, King Abdulaziz University, Jeddah- 21589, Saudi Arabia
2Department of Fish Health Management, Sylhet Agricultural University, Sylhet-3100, Bangladesh
3Department of Fisheries Technology and Quality Control, Sylhet Agricultural University, Sylhet-3100, Bangladesh
4Biopharmaceutical Engineering Major, Division of Applied Bioengineering, Dong-Eui University, Busan 47340, Republic of Korea
5Department of Biotechnology, Pukyong National University, Busan 48513, Republic of Korea
6Institute of Marine Sciences, University of Chittagong, Chittagong 4331, Bangladesh
7FAO World Fisheries University Pilot Programme, Pukyong National University, Busan 48513, Republic of Korea
8Core-Facility Center for Tissue Regeneration, Dong-Eui University, Busan 47340, Republic of Korea
9Department of Aquaculture, Sylhet Agricultural University, Sylhet-3100, Bangladesh

Introduction

Aquaculture, the farming of aquatic organisms under controlled environments, is a diverse food producing activity growing annually by 4.5%, accounting for a value of USD 243.26 billion [1]. Data suggest that fish, macroalgae, molluscs and crustaceans are meeting the protein demand of an increasing global population, contributing 49.1, 27.3, 15.6, and 7.9%, respectively, to total aquaculture production, equalling ~20% of total global animal protein supplies [2]. Aquaculture practices have shifted from extensive to super-intensive to elevate production at the expense of huge amounts of artificial feeds and degradation of aquatic environments. Poor culture management causes immunosuppression, stress and creates suitable conditions for the proliferation of opportunistic microbial pathogens (bacteria, viruses, and parasites) leading to infectious disease outbreaks. Hemorrhage, exophthalmia, meningoencephalitis, anorexia, and disruption of the nervous system are common impacts of infectious diseases responsible for high mortality of in cultured fishes. To control diseases, antibiotics, drugs and vaccines are routinely used in aquaculture [3].

Vaccination is a well-established strategy for long-term disease protection, but it has been associated with
temporary loss of appetite and growth, intra-abdominal lesions and impaired antibody production in fish [4]. On the other hand, excessive use of antibiotics results in the development of antibiotic resistant pathogens [5, 6], immunosuppression in host and reduction or mass killing of normal microbiota in the culture environment and gut [7], including residual effects in human [8]. The magnitude of this problem can be severe; an estimated 1.7 million deaths were attributed to antibiotic resistant bacteria in 2019 [9]. Antibiotic-resistant pathogens have been isolated from fish and humans, and their applications in aquaculture are under strict regulation [2]. To address this concern, one productive line of research has focused on biologically benign approaches such as probiotics, prebiotics, immunostimulants and functional feeds that enhance host immunity to pathogens [10, 11].

The word “probiotics” means “for life” and is based on the Greek word’s “pro” and “bios”. Probiotics were first defined by Parker [12] as “organisms or substances engagement for the maintenance of the microbial balance in the intestine”. These definitions were revised by Merrifield et al. [13] for aquatic organisms as, “a probiotic organism can be regarded as a live, dead or component of a microbial cell, which can be administered via feed or into rearing water, benefiting the host by improving growth performance, feed utilization, immune health status, infectious disease resistance, and stress responses which is achieved at least in part via improving the microbial balance in hosts or ambient environment”. Probiotic research in the last decades has revealed positive effects on growth [14, 15], digestive enzymes activities and feed utilization [16], immunity with upregulation of immune related genes [17, 18], improvement of beneficial microbes in the gut and positive modification of intestinal structure [19], and disease protection [20, 21], leading to an eco-friendly aquatic environmental management [22] in fish and shellfish farming.

Previous reviews of probiotics in aquaculture have considered sustainability issues [3], carp culture [7], disease control [2], mitigation of challenges in aquaculture [23], uses in Chinese aquaculture [5] and so on. There is not a single review that has comprehensively addressed the effects of probiotics on prominent cultured finfishes (e.g., tilapia Oreochromis niloticus, Atlantic salmon Salmo salar, rainbow trout Oncorhynchus mykiss, olive flounder Paralichthys olivaceus, common carp Cyprinus carpio, grass carp Ctenopharyngodon idella and rohu carp Labeo rohita) as relating to growth, haemato-immunological and disease responses. The novelty of the present review is to present up-to-date insights on the use of probiotics in commercially important finfish culture, highlighting their applications in field or open pond system. Probiotics effects on the host transcriptome profiles and aquatic environmental parameters are also discussed.

**Probiotics Functionality Linked to Growth, and Cellular and Humoral Immunity**

Probiotics not only reduce intestinal pathogens but also detoxify harmful compounds in feed through hydrolytic enzymes and the production of vitamins, biotin, and vitamin B12 [24, 25]. Growth elevation with improved feed utilization can ensure improved yields at lower production costs which are of value to aquaculture. Several authors have characterized responses to probiotics including alterations of digestive enzyme synthesis and release [26] and of intestinal structure to increase nutrient digestion and absorption [27], influencing the final body weight gain and production scale.

In aquaculture, probiotics can be supplemented through feed or water to manipulate the microbial balance in host and culture environment. Different nutrient specific digestive enzymes (e.g., amylase, protease, chitinase and lipase) can be increased by probiotics in the intestine [28], improving digestion and absorption while removing toxicity. Jang et al. [29] mentioned that probiotics improved digestive process by enhancing beneficial bacterial population, microbial enzymatic activity, and microbial balance, ultimately improving digestibility and absorption of nutrients toward increased growth rates and diet digestibility. It is unknown whether digestive enzymes are produced directly by probiotics or by modulating synthesis and secretion of enzymes by intestinal cells, or possibly by both mechanisms. However, probiotics reportedly demonstrate in vitro extracellular amylase, cellulase, lipase and protease production and could potentially modulate those enzymatic functions in vivo [30].

Normal light microscopic observation on fish gut histology depicts an intact epithelium barrier, goblet cells, and well-organised villi and microvilli. Supplemented probiotics metabolize the carbohydrates (oligosaccharides/prebiotics) of diet for their growth and survival in the intestine. A combined application of probiotics and prebiotics, termed symbiotics, produced significantly higher effects compared to each individual component [11, 31]. Fermentation of prebiotics by probiotics is reported to produce short chain fatty acids, which can be utilized as an energy source by intestinal epithelial cells [32], and cell proliferation ultimately increases goblet cell intensity, villus and micro villus height, width and density [33]. Alteration of these morphometric structures also can be found in intestinal thickness, fold of change, and mucus layer, contributing to the elevation of nutrient absorption. Higher nutrient absorption through increased absorption area in the intestine in response to probiotics supplementation showed positive changes in apparent digestibility coefficient [19] that correlated with fish growth and utilization of diets.

Probiotics mainly upregulate the fish innate immune parameters through antigen presenting dendritic cells (DCs) to maintain the linkage between innate and adaptive immunity [34]. Mucus on body surface, scale, skin, and gills serve as physical or epithelial barriers as a first line of defence in fish innate immune system. The role of probiotics to improve epithelial barrier is not well defined, but immunoglobulins (Ig), antibacterial peptides and complement proteins containing skin mucus lysozyme activity were enhanced after probiotics supplementation [35, 36]. Macrophage and phagocytic cells (natural killer cell, neutrophil, monocyte, lymphocyte, and cytotoxic cells) provide cellular immune defence in fish [37]. Interaction between microbial-associated molecular patterns (MAMPs) and pattern recognition receptors (PRRs) on DCs can stimulate and activate the phagocytic cells or macrophage and other immune cells to release cytokines and chemokines that recruit and activate immune cells, enhance the body's immune response, and provide protection against pathogens [38]. Lactic acid bacteria (LAB), for examples Bacillus and bifidobacteria are commonly used gram-positive probiotics, and their cell wall components like peptidoglycans, lipopolysaccharides,
flagella, and microbial nucleic acids are collectively known as MAMPs. MAMPs are attracted by PRRs on DCs and after interaction/binding of MAMPs with PRRs, DCs stimulate and activate phagocytic cells [39] to locate and engulf invading pathogens. Stimulation of plasma cells by DCs produces antibodies which can pass through the enterocyte and neutralize the pathogens [8]. Moreover, binding of MAMPs with Toll-like receptors (TLR) and stimulation of T cells by DCs produce mostly pro-inflammatory cytokines [31]. Among different pro-inflammatory cytokines, tumour necrosis factor (TNF)-α stimulates neutrophilic production and stimulates lymphocyte and macrophage [41], interferon (IFN)-γ stimulate resting macrophages to secrete IL-6 and IL-1β, and multi-functional IL-6 are responsible for B-lymphocyte differentiation and maturation [42] to elevate cellular innate immunity.

Humoral immunity refers to activities of bacteriolytic or haemolytic enzymes like myeloperoxidase or peroxidase, serum/skin mucus lysozyme, serum antiprotease, superoxide dismutase, serum bactericidal activity, alternative complement activity, transferrin and Ig in body or tissue fluids [10, 39, 43], which contribute to the elimination of pathogens. Serum proteomic study after *Bacillus* sp. JB-1 supplementation increased transferrin protein quantity in rainbow trout [44]. In human, oral administration of probiotics interacts with intestinal epithelial cells to enhance the production of macrophage chemotactic protein-1, to send signals to other immune cells characterised by an increment of IgA+ cells in the intestine, bronchus, and mammary glands [45]. *Lactobacillus acidophilus* La1 can persist in the gastrointestinal tract and act as an adjuvant to increase serum IgA titre to *Salmonella typhi* Ty21 in human [46]. *Lactococcus lactis* increased the expression of complement receptors and adjuvant potential of *Lactobacillus* GG to elevate the humoral immunity [47]. Lysozyme, defensins, cathelicidins, and phospholipase are antimicrobial peptides secreted by Paneth cells located in the bottom of intestinal crypts [48]. A transmission electron microscopic study depicted *Lactobacillus casei* CRL 431 and *Lactobacillus paracasei* CNCM I-1518 increased Paneth cells activity, apparently increasing the secretion of antimicrobial peptides in intestines [49].

**Probiotics Effects on Cultured Finfish**

**Tilapia**

Tilapia alone contributed 10% (5.377 million tonnes) of global aquaculture production in 2016 [1]. Nevertheless, its intensive production is more susceptible to stress-related disease and mass mortalities [50], and probiotics are viewed a promising potential solution [51].

The effects of single probiotic *L. plantarum* and its strains CGFM639, CCFM8661, and CR1T5 on tilapia were studied [52-55] (Table 1). Supplementation with 107 to 108 CFU g–1 of *L. plantarum* resulted in improvement of growth, feed intake and haemato-biochemical characters, similarly to its other strains CGFM639, CCFM8661 and CR1T5. Among these *L. plantarum* and CR1T5 increased protection against infection, and CGFM639 and CCFM8661 reduced aluminium toxicity and lead accumulation in liver, kidney, spleen, gill, and gonad. A combination of *L. plantarum* N11 + *Bacillus velezensis* H3.1 positively modulated tilapia cellular and humoral immunity [36].

Dietary *Clostridium butyricum* improved final body weight and feed digestion at 1.5 × 108 CFU g–1 [56], but not at 107 CFU g–1 [57] and protection against *A. hydrophila* and *S. agalactiae*, respectively. This probiotic decreased serum malondialdehyde and diamine oxidase but upregulated immune genes expression levels. Similar parameters were modulated along with increment of immunity, and digestive enzymes were also induced by *B. subtilis* HAINUP40 [58] and *B. licheniformis* DAHB1 [59]. Protection against *S. agalactiae* and *A. hydrophila* was also reported after feeding with a combination of *B. subtilis* and *B. licheniformis* [60] and various strains of *Bacillus* spp. [61], respectively.

Immunity modulation was also confirmed by *B. cereus* as a water supplement at 108 CFU ml–1 [62]. Combination of *Bacillus* with *Pediococcus* sp., *Enterococcus* sp. and *Lactobacillus* sp. did not produce any notable enhancement of digestive enzymes or other serum biochemistry parameters [63]. Expression of inflammatory, antimicrobial and T cell response genes after administration of *B. subtilis* ABP1 is an indication of its oral vaccine property [64]. *Pseudomonas chumenssis* NPUS1 [65] and *Aspergillus oryzae* [66] improved growth and immunological competence, but lowered the levels of glucose and cortisol in blood. Similarly, gut probiotic *Rummeliibacillus stabekeensis* modulated the aforementioned parameters with enriched gut microbiota and inhibit *A. hydrophila* and *S. iniae* [67]. In addition, supplementation with *Psychrobacter maritimus* S and *P. nannhaensis* S089 showed similar modulation except lower HSP70 [16, 68] and *B. amyloliquifaciens* increased fat digestibility [69]. *B. velezensis* TPS1N, *B. subtilis* TPS4 and *B. amyloliquifaciens* TPS17 [70]; *Bacillus* sp. KUAQ1 and KUAQ2 [71] improved tilapia innate immunity. In addition, intestinal lipase activity and morphometric alteration, viz. micro-villus height and width, goblet cells count, and muscle thickness were reported by the former *Bacillus* probiotics. Administration of *B. subtilis* WB60 with *L. lactis* for 8 weeks resulted in better growth, immunity and immune genes expression, villus height, without changing the haematology [72]. Similar modulations were observed after supplementation with *L. plantarum* L-137 [73] and only gene transcription by *L. plantarum* [74]. The *Bacillus* and *Lactobacillus* probiotics specified above conferred elevated protection against different infectious diseases caused by *E. faecalis*, *A. hydrophila*, and *S. agalactiae*.

Applications of three commercial probiotics, Proxin (multi-strain, 6 × 108 CFU g–1), *Biogen-s* (*B. subtilis*, 1011 CFU g–1) and Diamond V (*Saccharomyces cerevisiae*, 2.6 × 1010 CFU g–1) increased growth and feed utilizations, immunity and immune genes transcription at 4th and 8th week [75]. Moreover, commercial multi-probiotics AquaStar Pond and EM resulted in higher immune gene transcription relative to single commercial preparation, MicroPan [76]. Available studies indicated that the commercial probiotics containing single bacterial species,
| Probiotics                | Mode of administration and dosage | Duration | Effects on O. nilotica References |
|--------------------------|----------------------------------|----------|-----------------------------------|
| *Lactobacillus plantarum* | Dietary supplementation at 10^8 CFU g⁻¹ | 60 days  | GP; FUP; FCR; IP; TAC, R, B, AL, HBP, T, TSH, M, B, GL, Ch, H₂O⁻¹; IDR↑; GIR↑↑ [52] |
| *L. plantarum* CGFM639   | Dietary supplementation at 10^8 CFU g⁻¹ | 4 weeks  | GP; FUP; FCR; IP; TAC, R, B, AL, HBP, T, TSH, M, B, GL, Ch, H₂O⁻¹; IDR↑; GIR↑↑ [53] |
| *L. plantarum* CCFM8661  | Dietary supplementation at 10^8 CFU g⁻¹ | 4 weeks  | GP; FUP; FCR; IP; TAC, R, B, AL, HBP, T, TSH, M, B, GL, Ch, H₂O⁻¹; IDR↑; GIR↑↑ [54] |
| *B. subtilis* HAINUP40    | Dietary supplementation at 10^8 CFU g⁻¹ | 8 weeks  | GP; FUP; FCR; IP; TAC, R, B, AL, HBP, T, TSH, M, B, GL, Ch, H₂O⁻¹; IDR↑; GIR↑↑ [57] |
| *B. licheniformis* DAHB1  | Dietary supplementation at 10^8 & 10^9 CFU g⁻¹ | 4 weeks  | GP; FUP; FCR; IP; TAC, R, B, AL, HBP, T, TSH, M, B, GL, Ch, H₂O⁻¹; IDR↑; GIR↑↑ [58] |
| *C. butyricum*           | Dietary supplementation at 10^8 CFU g⁻¹ | 30 days  | GP; FUP; FCR; IP; TAC, R, B, AL, HBP, T, TSH, M, B, GL, Ch, H₂O⁻¹; IDR↑; GIR↑↑ [59] |
| *B. subtilis + B. licheniformis* | Dietary supplementation at 3 to 7 g Kg⁻¹ | 4 weeks  | GP; FUP; FCR; IP; TAC, R, B, AL, HBP, T, TSH, M, B, GL, Ch, H₂O⁻¹; IDR↑; GIR↑↑ [60] |
| *B. subtilis*             | Dietary supplementation at 60 mg/kg feed | 42 days  | GP; FUP; FCR; IP; TAC, R, B, AL, HBP, T, TSH, M, B, GL, Ch, H₂O⁻¹; IDR↑; GIR↑↑ [61] |
| *Aspergillus oryzae*     | Dietary supplementation at 10^8 CFU g⁻¹ | 30 days  | GP; FUP; FCR; IP; TAC, R, B, AL, HBP, T, TSH, M, B, GL, Ch, H₂O⁻¹; IDR↑; GIR↑↑ [62] |
| *Rummeliobacillus stabekisii* | Dietary supplementation at 10^8 CFU g⁻¹ | 2 weeks  | GP; FUP; FCR; IP; TAC, R, B, AL, HBP, T, TSH, M, B, GL, Ch, H₂O⁻¹; IDR↑; GIR↑↑ [63] |
| *Psycobacter nanhaensis*  | Dietary supplementation at 10^8 CFU g⁻¹ | 2 months | GP; FUP; FCR; IP; TAC, R, B, AL, HBP, T, TSH, M, B, GL, Ch, H₂O⁻¹; IDR↑; GIR↑↑ [64] |
| *Aspergillus oryzae*     | Dietary supplementation at 10^8 CFU g⁻¹ | 60 days  | GP; FUP; FCR; IP; TAC, R, B, AL, HBP, T, TSH, M, B, GL, Ch, H₂O⁻¹; IDR↑; GIR↑↑ [65] |
| *Rummeliobacillus stabekisii* | Dietary supplementation at 10^8 CFU g⁻¹ | 8 weeks  | GP; FUP; FCR; IP; TAC, R, B, AL, HBP, T, TSH, M, B, GL, Ch, H₂O⁻¹; IDR↑; GIR↑↑ [66] |
| *Psycobacter maritimus* S | Dietary supplementation at (2.8 & 5.6) × 10^8 CFU g⁻¹ | 50 days  | GP; FUP; FCR; IP; TAC, R, B, AL, HBP, T, TSH, M, B, GL, Ch, H₂O⁻¹; IDR↑; GIR↑↑ [67] |
| *Psycobacter maritimus* S | Dietary supplementation at (3.3 & 6.6) × 10^8 CFU g⁻¹ | 50 days  | GP; FUP; FCR; IP; TAC, R, B, AL, HBP, T, TSH, M, B, GL, Ch, H₂O⁻¹; IDR↑; GIR↑↑ [68] |
| *B. subtilis*             | Dietary supplementation at 10^8 CFU g⁻¹ | 4 weeks  | GP; FUP; FCR; IP; TAC, R, B, AL, HBP, T, TSH, M, B, GL, Ch, H₂O⁻¹; IDR↑; GIR↑↑ [69] |
| *B. subtilis*             | Dietary supplementation at (1, 3, & 5) × 10^8 CFU g⁻¹ | 8 weeks  | GP; FUP; FCR; IP; TAC, R, B, AL, HBP, T, TSH, M, B, GL, Ch, H₂O⁻¹; IDR↑; GIR↑↑ [70] |
| *B. subtilis*             | Dietary supplementation at 10^8 CFU g⁻¹ | 8 weeks  | GP; FUP; FCR; IP; TAC, R, B, AL, HBP, T, TSH, M, B, GL, Ch, H₂O⁻¹; IDR↑; GIR↑↑ [71] |
| *B. subtilis*             | Dietary supplementation at 10^8 CFU g⁻¹ | 8 weeks  | GP; FUP; FCR; IP; TAC, R, B, AL, HBP, T, TSH, M, B, GL, Ch, H₂O⁻¹; IDR↑; GIR↑↑ [72] |
| *B. subtilis*             | Dietary supplementation at 10^8 CFU g⁻¹ | 30 days  | GP; FUP; FCR; IP; TAC, R, B, AL, HBP, T, TSH, M, B, GL, Ch, H₂O⁻¹; IDR↑; GIR↑↑ [73] |
| *B. subtilis*             | Dietary supplementation at 10^8 CFU g⁻¹ | 30 days  | GP; FUP; FCR; IP; TAC, R, B, AL, HBP, T, TSH, M, B, GL, Ch, H₂O⁻¹; IDR↑; GIR↑↑ [74] |

Table 1. Effects of probiotics supplementation on growth, feed utilizations, immunological and haematological-biochemical parameters, immune related gene expression and disease resistance in tilapia (*Oreochromis niloticus*).
Table 1. Continued.

| Probiotics | Mode of administration and dosage | Duration | Effects on O. niloticus | References |
|------------|-----------------------------------|----------|------------------------|------------|
| Protexin (L. plantarum, L. rhamnosus, Bifidobacterium bifidum, E. faecium, Candida pintoledvi, & Aspergillus oryzae), Biogen-5 (B. subtilis), Diamond V (Saccharomyces cerevisiae) | Dietary supplementation at 6 x 107 & 2.6 x 109 CFU g−1 | 8 weeks | GP, FUP, PC, IP, HBP, RBP, RIL, TSP, Alb, Glb, Glu, WBC, & RBC | [75] |
| AlCare (B. licheniformis) | Dietary supplementation at 2 x 107 to 4.4 x 108 CFU g−1 | 10 weeks | GP, FUP, PC, IP, SL, SO, SR, IRGE, TSP, SR | [77] |
| DVAQUA (S. cerevisiae) | Dietary supplementation at 2.0 g Kg−1 feed | 8 weeks | GP, FUP, PC, IP, SL, SR, IRGE, TSP, SR | [78] |
| Organic Green & P. pumilus | Water supplementation at 107 CFU ml−1 | 1, 2, & 8 months | GP, FUP, PC, IP, SL, SR, IRGE, TSP, SR | [79] |
| AquaStar Pond (Bacillus sp., Pediococcus spp., Enterococcus Sp.), EM (Rhodopsedomonas spp., Lactobacillus spp., Saccharomyces spp., MicroPan (Bacillus spp.) | Dietary supplementation at 0.0015 g m−2 day−1; 10 ml m−2 day−1; 0.002 g m−2 day−1 | 60 days | GP, FUP, PC, IP, SL, SO, SR, IRGE, TSP, SR | [76] |

Symbol ↔, no change; ↑, increase; ↓, decrease versus controls.

A/G: Albumin/Globulin; ACA: Apparent complement activity; ADPC: Apparent digestibility coefficient; Alb: Albumin; ALP: Alkaline phosphatase; ALT: Alanine amino transferase; AmA: Amylase activity; AST: Aspartate amino transferase; CASP3: Caspase 3; CAT: Catalase; DAO: Diamine oxidase; DEA: Digestive enzyme activities; FBW: Final body weight; FCR: Feed conversion ratio; FER: Feeding efficiency ratio; FUP: Feed utilization parameters; Glb: Globulin; Glu: Glucose; GP: Growth parameters; Glx: Glutathione peroxidase; GutM: Gut modified bacteria; Hb: Haemoglobin; HBP: Haemato-biochemical parameters; HSP: Heat shock protein; Ht: Hematocrit; IDR: Infectious disease resistance; IgM: Immunoglobulin M; IL: Interleukin; INF: Interferon; IRGE: Immune related gene expression; LBP: Lactate dehydrogenase; MDA: Malondialdehyde; MPO: Myeloperoxidase; mVH: micro-Villus height; MyD: Myeloid differentiation factor; NO: Nitric oxide; PhAT: Pb accumulation in tissues; PCa: Phagocytic activity; PER: Protein efficiency ratio; PPV: Protein productive value; PRA: Protease activity; RGR: Relative growth rate; RB: Respiratory burst (NBT assay); RLC: Red blood cell; RLP: Relative level of protection; SBA: Serum bacterial activity; SGR: Specific growth rate (%); SL: Serum lysozyme; SMLA: Skin mucus lysozyme activity; SOD: Superoxide dismutase; SR: Survival rate; ST: Salinity tolerance; TAG: Total anti-oxidant capacity; TGF: Transforming growth factor; TLC: Total leucocytic count; TLR: Toll-like receptor; TNF: Tumor necrosis factor; TSP: Total serum protein; WBC: White blood cells; WG: Weight gain (%).

namely AlCare (B. licheniformis), Biogen (B. subtilis), and BioSaf (S. cerevisiae) resulted in improved growth [77, 78] without affecting measured dietary digestion parameters [77, 78] (Table 1). Moreover, the use of commercial probiotics as water supplements enhanced protection against A. hydrophila [79].

Atlantic Salmon

Comparatively less information is available on the effects of probiotics in Atlantic salmon. Available information can be categorized into in vivo and in vitro studies as summarized in Table 2.

Wang et al. [80] assessed the effect of B. velezensis V4 CGMCC 10149 and Rhodotorula mucilaginosa CGMCC 1013 on juvenile salmon raised in a recirculating aquaculture system. After 62 days feeding with different doses of these strains, fish weight gain, immunity as well as disease resistance against A. salmonicida improved significantly. In the same species, administration with these strains, fish weight gain, immunity as well as disease resistance against A. salmonicida- and Y. ruckeri-prepared damage in the foregut of Atlantic salmon to some extent [87].

Probiotics Effects on Commercial Finfish 685
### Table 2. Effects of probiotic supplementation on growth, feed utilizations, immunological and haemato-biochemical parameters and disease resistance in Atlantic salmon (Salmo salar) aquaculture.

| Probiotics | Mode of administration and dosage | Duration | Effects on S. salar | References |
|------------|---------------------------------|----------|---------------------|------------|
| Bacillus velezensis V4 CGMCC 10149 (BV4) & Rhodotorula mucilaginosa CGMCC 1013 (RM) | Dietary supplementation at BV4 (5 × 10⁷) + RM (5 × 10⁷), BV4 (1.5 × 10⁷) + RM (1.5 × 10⁷), & BV4 (2.5 × 10⁷) + RM (2.5 × 10⁷) CFU g⁻¹ | 62 days | GP, EBP, W, & S + FUP, PCL, & IP, sod; | [80] |
| Bacillcol (Pedococcus acidilactici MA18/5M) | Dietary supplementation at 1.19 × 10⁷ CFU g⁻¹ | 12 weeks | GP, EBP, & S + IRIE, XS; TLR, TNF-α, & IL-1β; HSP70 & PCNA; | [81] |
| Bacillcol (P. acidilactici MA18/5M) | Dietary supplementation at 10⁹ CFU Kg⁻¹ | 6 weeks | IRGE, ME-β, TNF-α, & INF-γ; | [82] |
| Lactobacillus RII & RIH | Dietary supplementation at −10⁶ CFU g⁻¹ | 20 days | GutM; | [83] |
| Lab. fermentum and Lab. plantarum | Dietary supplementation at 10⁶ CFU g⁻¹ | 38 days | GME & GNET; VH & LPW ↑ | [84] |
| Lab. delbruecki subsp. lactis | In vitro incubation at 1.6 × 10⁶ CFU ml⁻¹ | 30 min | PCIT; IDR ↑; TNL ↓; sALβ, IL-1β, T NF-α, & INF-γ ↓ | [85] |
| Carnobacterium divergens | In vitro incubation at 10⁶ CFU ml⁻¹ | 1 h | PCIT; Gut Morphology ↑; Gut cells; Pathogenic antagonism ↑; | [86] |
| C. divergens | In vitro incubation at 6 × 10⁶ & 6 × 10⁶ CFU ml⁻¹ | 1 h | IDR ↑; sALβ, IL-1β, T NF-α, & INF-γ ↑ | [87] |

Symbol: +, no change; ↑; increase; ↓, decrease versus controls.

ACP: Acid phosphatase; ALT: Alanine aminotransferase; AST: Aspartate aminotransferase; CAT: Catalase; FCR: Final body weight; FUP: Feed utilization parameters; GME: GNP; GNEM: The area and number of mucus cells/μm² gill epithelium; GPx: Glutathione peroxidase; GutM: Gut microbiota; MDA: Malondialdehyde; MUL: Mitochondrial ubiquitin ligase activator of NFκB1; MX-1: Interferon-induced GTP-binding protein; NO: Nitric oxide; PCI: Probiotic count in the intestine; PCNA: Proliferating cell nuclear antigen; SGR: Specific growth rate (%); SL: Serum lysozyme; SOD: Superoxide dismutase; SR: Survival rate; TAC: Total antimicrobial activity; TLR: Toll-like receptor; TNF: Tumor necrosis factor; VH: vili height; WG: Weight gain (%).

### Rainbow Trout

Rainbow trout contributed 2% of total global finfish production in 2016 [1]. Numerous benefits of probiotics have been reported in rainbow trout aquaculture, for example, both *L. plantarum* and its strain KC426951 promoted growth and immunity with some exceptions in haemato-biochemical parameters [88, 89] (Table 3). The former probiotic increased alkaline phosphatase (ALP), IgM, and hemoglobin to a significant level, whereas the latter one only increased white blood cells with no alteration of hemoglobin, mean corpuscular volume, and red blood cells concentration. In contrast to the results reported by Soliani et al. [89], some haematological parameters like ALP, triglycerides and total cholesterol depicted improved modulation after *L. rhamnosus* treatment [90].

Dietary supplementation with LABs *L. buchneri*, *L. fermentum*, and the yeast *S. cerevisiae* together for 130 days improved innate immunity and immune genes transcription without alteration of haemato-biochemical parameters [91]. Modulation of these parameters was also observed after feeding with various strains of LAB [92, 93]. Administration with *L. plantarum*, *Lac. lactis*, and *Leuconostoc mesenteroides* upregulated IL-8 & 10, TNF-α, and IgT and decreased IL-1β, TNF-α and TLR5 transcription before and after *Lac. garvieae* infection, respectively directly in contrast to results reported by Pérez- Sánchez et al. [94] (Table 3).

Combination of *Bacillus* sp. with *Lactobacillus* sp. reportedly elevated humoral immunity, intestinal structure and decreased plasma phenoloxidase [95]. The dietary administration of *Bacillus* sp. and *B. licheniformis* individually or in combination showed improved [96], unchanged [63], and decreased [97] results in weight gain and specific growth rate. Humoral immunity was elevated with *B. subtilis* + *B. licheniformis*, but this combination had no influence on serum biochemical parameters apart from total leukocytes.

Trout gut probiotics *Kocuria* SM1 and *Rhodococcus* SM2 failed to induce changes in growth, digestive enzymes, and haematology, with the exceptions of lowered trypsin and increased hemoglobin levels [30]. In another study, single administration of *Kocuria* SM1 improved immunity and serum biochemistry [98]. Besides these, *Kocuria* SM1-fed rainbow trout fingerlings decreased mortality by 10-28% from 73-92% in response to V. anguillarum challenge. Dietary administration of *Enterobacter cloacae* and *B. mojavensis* showed improved protection from *Y. ruckeri*, where survival rate increased to 99.2% compared to 35% in the control group [99]. Probiotics like *E. casseliflavus* positively modulated serum biochemistry by decreasing malondialdehyde and lymphocytes without changing alanine and aspartate aminotransferase [100]. Supplementation of *Bifidobacterium animalis* PTCC-1631 and *Bif. lactis* PTCC-1736 at the lowest dose (10⁷ CFU g⁻¹) improved fish body weight and
Table 3. Effects of probiotic supplementation on growth, feed utilizations, immunological and haemato-biochemical parameters, immune related gene expression and disease resistance in rainbow trout (*Oncorhynchus mykiss*).  

| Probiotics | Mode of administration and dosage | Duration | Effects on O. mykiss | References |
|------------|----------------------------------|----------|----------------------|------------|
| Lactobacillus plantarum | Dietary supplementation at 2 × 10^7 CFU g⁻¹ | 72 days | GP, FCR, PER, FER, TSP | [88] |
| L. plantarum | Dietary supplementation at 10^8 CFU g⁻¹ | 60 days | GP, FCR, PER, FER, HBP, | [89] |
| L. rhamnosus | Dietary supplementation at 10^9 & 10^11 CFU g⁻¹ | 30 days | HBP, TSP, ALP, MCH, MCHC, MCV | [90] |
| L. buchneri, L. fermentum & Saccharomyces cerevisiae | Dietary supplementation at 10^7 CFU g⁻¹ | 130 days | GP, FCR, PER, MCHC, MCV | [91] |
| L. delbrueki subsp. bulgaricus, L. acidophilus & Citrobacter freundii | Dietary administration at 5 × 10^7 CFU g⁻¹ | 60 days | GP, FCR, PER, FER, IP, IL-1β, IL-12 | [92] |
| L. delbrueki subsp. bulgaricus | Dietary administration at 10^7 CFU g⁻¹ | 36 days | IRGE, IL-1β, IL-12 | [93] |
| Bacillus sp. & Pedicoccus sp., Enterococcus sp. + Lactobacillus sp. + P. acidilactici | Dietary administration at 8.6 × 10^6, 1.6 × 10^6, 2.6 × 10^6 & 7.2 × 10^6 CFU g⁻¹ | 8 weeks | GP, FCR, MCHC, MCV | [94] |
| B. subtilis, B. licheniformis | Dietary administration at (7.79, 8.36, 8.05, & 8.23) × 10^7 CFU g⁻¹ | 10 weeks | GP, FCR, PER, IL-1β, MCHC, MCV | [95] |
| B. subtilis & B. licheniformis, & B. subtilis + B. licheniformis | Dietary administration at ≥ 10^7 CFU g⁻¹ | 8 weeks | GP, FCR, PER, IL-1β, MCHC, MCV | [96] |
| B. subtilis & B. cereus toyoi | Dietary administration at ≥ 10^8 × 1.5 × 10^7 CFU g⁻¹ | 20 weeks | GP, FCR, PER, IL-1β, MCHC, MCV | [97] |
| B. subtilis ABP1 & ABP2 | Dietary administration at ≥ 10^7, & 10^7 CFU g⁻¹ | 1 week | IP, MCHC, MCV | [98] |
| Kocuria SM1 & Rhodococcus SM2 | Dietary administration at ~10^7 (SM1) or ~10^7 (SM2) CFU g⁻¹ | 14 days | GP, FCR, PER, MCHC, MCV | [99] |
| Kocuria SM1 | Dietary administration at ~10^6 CFU g⁻¹ | 2 weeks | GP, FCR, PER, MCHC, MCV | [100] |
| Enterobacter cloacae & B. mojaevensis | Dietary administration at ≥ 10^8 CFU g⁻¹ | 60 days | GP, FCR, PER, MCHC, MCV | [101] |
| Enterococcus casseliflavus | Dietary administration at ≥ 10^7, & 10^11 CFU g⁻¹ | 8 weeks | GP, FCR, PER, MCHC, MCV | [102] |
| Bifidobacterium animalis PTCC-1631 & Bif. lactis PTCC-1736 | Dietary administration at (1, 2, & 3) × 10^9 CFU g⁻¹ | 8 weeks | GP, FCR, PER, MCHC, MCV | [103] |
| Bio Aqua (P. acidilactici, E. faecium, B. subtilis, L. acidophilus, L. plantarum, L. casei, L. rhamnosus, B. bifidum & S. cerevisiae) | Dietary administration at (1, 2, & 4) × 10^8 CFU g⁻¹ | 8 weeks | GP, FCR, PER, MCHC, MCV | [104] |
| Proviotic (L. bulgaricus) | Dietary administration at 460 mg Kg⁻¹ | 60 days | GP, FCR, PER, MCHC, MCV | [105] |
| Prima Lac (L. acidophilus, L. casei, E. faecium, & Bif. bifidum) | Dietary administration at 1.5 Kg⁻¹ | 8 weeks | GP, FCR, PER, MCHC, MCV | [106] |
| Aquasline (S. cerevisiae & S. ellipsoides) | Dietary administration at ≥ 10^8 CFU g⁻¹ | 8 weeks | GP, FCR, PER, MCHC, MCV | [107] |
| Bacillus sp., Pedicoccus sp., Enterococcus sp. & Lactobacillus sp. | Dietary administration at ≥ 2 × 10^7 CFU g⁻¹ | 9 weeks | GP, FCR, PER, MCHC, MCV | [108] |

Symbol →, no change; ↑, increase; ↓, decrease versus controls.
A/G: Albumin-Globulin; ACA: Alternative complement activity; ACP: Acid phosphatase; ADC: Apparent digestibility coefficient; Alb: Albumin; ALP: Alkaline phosphatase; ALT: Alanine transaminase; AmA: Amylase activity; AS: Aspartate amino transferase; Ca: Calcium; CAT: Catalase; DWG: Daily weight gain; DE: Digestive enzyme activity; ED: Egg diameter; FATP: Fatty acid transport protein; FBW: Final body weight; FCE: Feed conversion efficiency; FCR: Feed conversion ratio; FE: Feed efficiency ratio; FLS: Fingerlings; FR: Fertilization rate; FUP: Feed utilization parameters; GLb: Globulin; GLu: Glucose; GP: Growth parameters; GSH: Glutathione; GTP: Glutathione peroxidase; TGP: Gamma glutamyl transpeptidase; GutM: Gut microflora; HBP: Haemato-biochemical parameters; HR: Hatching rate; Ht: Hematocrit; IDR: Infectious disease resistance; Ig: Immunoglobulin; IGF: Insulin-like growth factor; IL: Interleukin; IP: Immunological parameters; IRGE: Immune related gene expression; LDH: Lactate dehydrogenase; LPV: Lipid productive value; MCH: Mean corpuscular haemoglobin; MCHC: Mean corpuscular haemoglobin concentration; MCV: Mean corpuscular volume; Mg: Magnesium; MPO: Myeloperoxidase; mVH: Micro-Volhosse height; P: Potassium; PCa: Phagocytic activities; PER: Protein efficiency ratio; PPO: Plasma peroxidase; PPV: Protein productive value; PRA: Protease activity; RB: Respiratory burst (NBT assay); RBC: Red blood cells; RF: Relative fecundity; RP: Reproductive parameters; SAP: Serum antiprotease activity; SBA: Serum alpha-amylase activity; SBP: Serum beta-glucuronidase activity; SBR: Respiratory burst; SGR: Specific growth rate (%); SL: Serum lysozyme; SOD: Superoxide dismutase; SR: Survival rate; TC: Total cholesterol; TG: Triglycerides; TLR: Toll like receptor; TNF: Tumor necrosis factor; TVC: Total viable counts; VCCS: Vertical column compression syndrome; WBC: White blood cell; WG: Weight gain. 

June 2022 | Vol. 32 | No. 6
digestibility compared to higher doses and to the control group [101] (Table 3).

Treatment with a commercial probiotic, Provistic containing L. bulgaricus and Aqualase (mixture of S. cerevisiae and S. ellipsoidea) demonstrated better growth and diet digestion [102, 103]. Similarly, Primalac composed of L. acidophilus, L. casei, E. faecium and Bif. bifidum elevated these two parameters along with protease, amylase and lipase activity in the intestine [15].

Common Carp

Common carp is among the most important commercially cultured fish in the world contributing 8% of total global finfish aquaculture production in 2016 [1]. Probiotics reportedly affect growth, immunological parameters, and disease protection in common carp [7]. Administration of L. delbrueckii for 8 weeks showed improved production, feed intake and various digestive enzymes including protease, amylase, lipase, Na+/K+-ATPase, creatine kinase and γGT activity [104, 105]. Moreover, similar to L. delbrueckii, Lactobacillus sp. and L. casei, E. faecium, Primalac & Paenibacillus sp. and Bacillus coagulans MTCC 9872, B. licheniformis MTCC 6824 & Paenibacillus polymyxa MTCC 122 improved production, feed intake and various digestive enzymes including protease, amylase and lipase activity in the intestine [15].

Table 4. Effects of probiotic supplementation on growth, feed utilizations, immunological and haemato-biochemical parameters and disease resistance in common carp (Cyprinus carpio).

| Probiotics                  | Mode of administration and dosage | Duration | Effects on Cyprinus carpio                                                                 | References |
|-----------------------------|----------------------------------|----------|------------------------------------------------------------------------------------------|------------|
| Lactobacillus delbrueckii   | Dietary supplementation at 10⁷–10⁸ CFU g⁻¹ | 8 weeks  | GP FBW & WG ↑; FUP ↑; HBP ↑; IP ↑; PB, IL-4 ↑; MPO, ACE, SO, CAT, GP, & TAC ↑; MDA ↑; IRGE ↑; INH ↓; SOD ↑; CAT ↑; PR ↑; MDA ↓; SR ↑ | [105]      |
| L. delbrueckii              | Dietary supplementation at 10⁷–10⁸ CFU g⁻¹ | 8 weeks  | GP HSP ↑; WBG ↓; FUP ↑; IP ↑; PB, IL-4 ↑; MPO, ACE, SO, CAT, GP, & TAC ↑; MDA ↑; IRGE ↑; INH ↓; SOD ↑; CAT ↑; PR ↑; MDA ↓; SR ↑ | [104]      |
| Lactobacillus sp., Nitrosomonas sp. and Bacillus spp. | Water supplementation at 10⁴ CFU ml⁻¹ | 10 days  | IRGE ↑; INH ↑; SOD and CAT ↑ | [106]      |
| L. plantarum 44*            | Dietary supplementation at (1, 3, 5; 10⁵–10⁷ CFU g⁻¹) | 60 days  | GP SGR ↑; FUP ↑; HBP ↑; IP ↑; PB, IL-4 ↑; MPO, ACE, SO, CAT, GP, & TAC ↑; MDA ↑; IRGE ↑; INH ↓; SOD ↑; CAT ↑; PR ↑; MDA ↓; SR ↑ | [107]      |
| L. plantarum                | Dietary supplementation at 10⁴ CFU g⁻¹ | 14 days  | IP ↑; PB, IL-4 ↑; HBP ↑; Amp, & TAC ↑; MPO ↑; ACA ↑; SOD ↑; CAT ↑; PR ↑; MDA ↓; SR ↑ | [108]      |
| Bacillus coagulans MTCC 9872 | Dietary supplementation at 10⁴ CFU g⁻¹ | 80 days  | GPinery ↑; FUP ↑; HBP ↑; IP ↑; PB, IL-4 ↑; MPO, ACA, SOD ↑; INH ↓; SOD ↑; CAT ↑; PR ↑; MDA ↓; SR ↑ | [110]      |
| B. coagulans                | Dietary supplementation at (1, 2, 4) × 10⁴ CFU g⁻¹ | 45 days  | GP FBW & WG ↑; IP ACA, IL-1, MPO & RBP ↑; | [109]      |
| Lactobacillus sp. & Bacillus sp. | Dietary supplementation at 0.75, 1.5, & 2.25 g kg⁻¹ | 60 days  | GP WBG ↑; FUP ↑; IP ↑; PB, IL-4 ↑; WQP ↑; PC & cort ↑; MPO, ACE, SO, CAT, GP, & TAC ↑; MDA ↑; IRGE ↑; INH ↓; SOD ↑; CAT ↑; PR ↑; MDA ↓; SR ↑ | [111]      |
| Pediococcus aciditici       | Dietary supplementation at 6 × 10⁴ CFU g⁻¹ | 60 days  | GP FBW & WG ↑; FUP ↑; IP ↑; PB, IL-4 ↑; WQP ↑; PC & cort ↑; MPO, ACE, SO, CAT, GP, & TAC ↑; MDA ↑; IRGE ↑; INH ↓; SOD ↑; CAT ↑; PR ↑; MDA ↓; SR ↑ | [112]      |
| P. pentosaceus              | Dietary supplementation at 10⁷, 10¹, & 10¹⁰ CFU g⁻¹ | 45 days  | GP FBW & WG ↑; IP ↑; PB, IL-4 ↑; WQP ↑; PC & cort ↑; MPO, ACE, SO, CAT, GP, & TAC ↑; MDA ↑; IRGE ↑; INH ↓; SOD ↑; CAT ↑; PR ↑; MDA ↓; SR ↑ | [113]      |
| Primalac (L. acidophilus, L. casei, E. faecium, & B. bifidum) | Dietary supplementation at 1 & 1.5 g kg⁻¹ | 8 weeks  | GP WBG ↑; SOD ↑; FUP ↑; FCR ↑; HBP ↓; WBG ↑; RBC, Hb, Ht, & MC ↑; MPO, ACE, SO, CAT, GP, & TAC ↑; MDA ↓; SR ↑ | [115]      |
| Primalac                   | Dietary supplementation at 1 & 2 g kg⁻¹ | 60 days  | GP FBW & WG ↑; FUP ↑; IP ↑; PB, IL-4 ↑; WQP ↑; PC & cort ↑; MPO, ACE, SO, CAT, GP, & TAC ↑; MDA ↑; IRGE ↑; INH ↓; SOD ↑; CAT ↑; PR ↑; MDA ↓; SR ↑ | [114]      |
| B. subtilis                | Dietary supplementation at 4 × 10⁷–10¹⁰ CFU g⁻¹ | 180 days | GP WBG ↑; SOD ↑; FBP ↑; FCR ↓; SOD ↑; MPO, ACE, SO, CAT, GP, & TAC ↑; MDA ↓; SR ↑; TAC ↑; IP ↓; TD↑; TSP ↑; DNA ↑ | [151]      |

Symbol: ↑, increase; ↓, decrease versus controls. ACHA: Alternative complement activity; ACP: Acid phosphatase; ACH50: Alternative complement; Alb: Albumin; ALP: Alkaline phosphatase; ALT: Alanine aminotransferase; AST: Aspartate aminotransferase; AmA: Amylase activity; CAT: Catalase; CD4+: Cluster of differentiation 4; CD8+: Cluster of differentiation 8; TC: Total cholesterol; CK: Creatine kinase; CP: Ceruloplasmin; DEA: Digestive enzyme activities; DO: Dissolved oxygen; FBW: Final body weight; FCE: Feed conversion efficiency; FCR: Feed conversion ratio; FUP: Feed utilization parameters; Glb: Globulin; Glu: Glucose; GP: Growth parameters; GPS: Glutathione peroxidase; GT: Glutamyl transpeptidase; GuM: Gut microbiota; Hb: Haemoglobin; HBP: Haematopoietic-biochemical parameters; HSP: Heat shock protein; Ht: Haematocrit; IDR: Infectious disease resistance; Ig: Immunoglobulin; IL: Interleukin; iNOS: Inducible nitric oxide synthase; IP: Immunological parameters; IRGE: Immune gene related expression; MCH: Mean corpuscular haemoglobin; MCHC: Mean corpuscular haemoglobin concentration; MCV: Mean corpuscular volume; MDA: Malondialdehyde; MPO: Myeloperoxidase; NF: Nuclear Factor; PCa: Phagocytic activity; PER: Protein efficiency ratio; PrA: Protease activity; RB: Respiratory burst (NBT assay); RBC: Red blood cell; SBA: Serum bactericidal activity; SGR: Specific growth rate (%); SL: Serum lysozyme; SOD: Superoxide dismutase; SR: Survival rate; TAC: Total anti-oxidant capacity; TC: Total cholesterol; TG: Triglyceride; TGC: Thermal growth coefficient; TGF: Transforming growth factor; TNF: Tumor necrosis factor; TSP: Total serum protein; WBC: White blood cell; WG: Weight gain (%); WQP: Water quality parameters.
Bacillus spp. modulated immune-gene transcription when used as water supplements [106]. Comparable modulation of parameters was noted after 60 days administration of L. plantarum 44ª, although the probiotic had no effects on lipase and haematological parameters [107]. In another study, same probiotic supplementation unveiled better serum biochemistry, phagocytic and λ-globulin activity in head kidney and protection against A. hydrophila, whereas phagocytic activity in spleen remains unchanged [108] (Table 4).

Supplementation with B. coagulans, B. licheniformis and P. polymyxa (MTCC 122) at 10⁴ CFU g⁻¹ influenced growth and feed utilizations, innate immunity [109], survival percentage, protection from A. hydrophila and V. harveyi [110]. Combined applications of Lactobacillus sp. with Bacillus sp. at 0.75, 1.5 and 2.25 kg g⁻¹ improved protease activity in the intestine and modulated water quality parameters like pH, temperature and lowered dissolved oxygen concentration [111]. Administration of P. acidilactici [112] and P. pentosaceus [113] for 60 and 45 days, respectively, exhibited contrasting results. P. acidilactici improved feed conversion ratio, but decreased final body weight and daily weight gain, protease activity and immune gene expression. On the other hand, P. pentosaceus improved growth and digestive enzymes activity. However, both probiotics increased innate immunity and serum biochemistry (Table 4). Commercial probiotic mixture viz. Primalac elicited a similar pattern of results, but only level of white blood cells and serum total protein increased among all haematobiochemical parameters [114, 115].

Japanese/Olive Flounder

Olive flounder is among increasingly commercially important cultured marine species in Northeast Asian countries like China, Japan, Korean peninsula and some other countries [11]. This fish is widely accepted all over the world due to fast growth, taste, palatability, wide range of environmental adaptation and disease resistance. In 2018, its production was 37,239 metric ton equaling ~52% of total fisheries production in the Republic of Korea [116].

Extensive studies have focused on the use of single L. lactis BBFE920 (LLBF) [17, 117], LLBF and L. plantarum FGL0001 (LPFG) [17] as well as single and combination of LLBF and LPFG [35] in olive flounder. LLBF and LPFG were isolated from bean sprouts and the hindgut of olive flounder respectively, and their probiotic properties were confirmed in vitro before dietary supplementation. LLBF was also administered orally as vaccine after cloning with E. tarda membrane protein and S. iniae antigen termed as OmpA, FlagD, and OmpA-FlagD [118] and SIMA [117] respectively (Table 5). These oral vaccines increased transcription levels of cluster of differentiation (CD) 4-1, CD4-2, CD4-α, T-bet, INF-γ, weight (20-21%) and feed digestion. Both vaccines also elevated antigen specific antibodies in fish tissues, as well as protection against E. tarda (5 × 10⁴ CFU ml⁻¹ LD₅₀) and S. iniae (10⁵ CFU ml⁻¹) producing 82% higher rates of survival relative to controls. Individual supplementation of probiotics LLBF and LPFG at 10⁴ CFU g⁻¹ increased expression levels of regulatory and pro-inflammatory genes, respectively [17] (Table 5). In addition, gut permeability was increased by LPFG compared to control and LLBF, and immune tone was stable for 30 days. Similar concentration level with combination of LLBF and LPFG increased levels of immunity and immune genes transcription and conferred higher protection against S. iniae challenge [35].

Except Sporolac, Lactobacillus and mixture of probiotics improved immune parameters and resistance against lymphocystis. In other studies, infectious S. parauberis-injected flounder fed mixed probiotics [119] and Uronema marinum infected group fed individual L. plantarum, L. acidophilus, L. sakei, B. subtilis and S. cerevisiae [120] at 0.1% and 2.42 × 10⁴ CFU g⁻¹ respectively. In both cases, fish production, haematology and immunity levels were higher in disease infected probiotics treated groups as compared to controls. Among individual probiotics only L. plantarum elevated survival in U. marinum infected fish compared to other 4 probiotics and control. Another commercial LAB probiotic, L. fermentum was supplemented at 0.5% for 8 weeks to compare the effects with yacon, ginger and blueberry at 0.1% [121]. Commercial probiotics showed no effects except higher protection against S. iniae infection.

A probiotic Bacillus SI-10 (BSJ-10) was isolated from Korean traditional fermented food [122] and complete genome sequence demonstrated similarity with two well-known olive flounder probiotics, B. subtilis and B. licheniformis [123, 124]. Dietary supplementation with this probiotic both live [21, 116] and heat-killed (HK) [125] forms at 10⁴ CFU g⁻¹ for 8 weeks increased rates of diet digestion and growth. Live and HK BSJ-10 and mixture with L. plantarum KCCM 11322 (3:1) protect olive flounder against streptococcosis [21, 43, 125] and Edwardsiella infection [116]. Two different forms of BSJ-10 demonstrated no effect on heamato-biochemistry and intestinal structure, but both increased immunity and cytokines production in different localised organs like liver, kidney, gill and spleen (Table 5). Live BSJ-10 increased probiotics count in the intestine, but had no effect on IL-6, CD4-1 and CD4-2 transcription, and were able to ferment β-glucogalactosaccharides and barley β-glucan as a symbiotic disease biocontrol model in olive flounder. Treatment with 3:1 Bacillus sp. SI-10 and L. plantarum KCCM 11322 at 0.75 + 0.25 × 10⁴ CFU g⁻¹ feed resulted in improved growth, immunity and disease resistance in olive flounder [43]. Similarly, feeding with B. subtilis, B. pumilus and B. licheniformis demonstrated identical patterns of response in olive flounder [22]. Among these probiotics, B. subtilis and B. licheniformis improved disease protection compared to control. In addition, only B. subtilis increased fish survival and decreased ammonia concentration (5 days experiment) in controlled culture environments. Jang et al. [29] used vegetative cells of BSJ-10 and noted improved growth and feed utilization, and spore-modulated immune genes transcription, leading to improved richness and diversity of intestinal bacterial population.

A study with individual (BSJ-10 and L. plantarum) [28] and multi-strain probiotics [126] as supplemented with 30% reduced fish meal diet was found to maintain intestinal homeostasis and immune competence in olive flounder. Both BSJ-10 and L. plantarum increased lipase and trypsin, but amylose activity was only improved by BSJ-10. Individual and multi-probiotics also improved concentrations of proteobacteria, actinobacteria, and...
| Probiotics                          | Mode of administration and dosage | Duration | Effects on P. olivaceus                                                                 | References |
|-----------------------------------|----------------------------------|----------|----------------------------------------------------------------------------------------|------------|
| Lactococcus lactis BFE920         | Dietary supplementation at 10^7 CFU g⁻¹ as oral vaccination | 4 weeks  | GPₘᵥ, IPₘᵥ, FUPₘᵥ, TG, TGF, INF-γ, IL-10, IL-12, TNF-α, IL-6, CD4-3, CD8-3, INF-γ, S. iniae, T-bet, & INF-γ; Gut permeability (mVH); Gut immunity (mVH); GPₘᵥ, IPₑₘᵥ, FUPₑₘᵥ, TG, TGF; IL-12 & INF-γ; IL-10, IL-12, TNF-α; T-bet, & INF-γ; Gut permeability (mVH); Gut immunity (mVH) | [118]      |
| L. lactis BFE920                  | Dietary supplementation at 10^7 CFU g⁻¹ as oral vaccination | 4 weeks & 3 Months | GPₘᵥ, IPₘᵥ, FUPₘᵥ, TG, TGF, INF-γ, IL-10, IL-12, TNF-α, IL-6, CD4-3, CD8-3, INF-γ, S. iniae, T-bet, & INF-γ; Gut permeability (mVH); Gut immunity (mVH) | [117]      |
| L. lactis BFE920 (LLRF) & Lactobacillus plantarum FGL0001 (LPFG) | Dietary supplementation at 10^7 CFU g⁻¹ | 30 days  | IRGE: IL-6, INF-γ, IL-12, TNF-α; CD4-1, CD4-2, CD8α, CD83, CD18; GPₘᵥ, IPₑₘᵥ, FUPₑₘᵥ, TG, TGF, INF-γ, IL-12, TNF-α, IL-10; T-bet, & INF-γ; Gut permeability (mVH); Gut immunity (mVH); | [17]       |
| L. lactis BFE920, L. plantarum FGL0001, & their combination | Dietary supplementation at 10^7 CFU g⁻¹ | 30 days  | GPₘᵥ; IPₑₘᵥ, FUPₑₘᵥ; IL-12, IL-10, TNF-α, INF-γ, IL-6, CD4-1, CD4-2, CD8α, CD83, CD18, CD11b-2; GPₑₘᵥ, IPₑₘᵥ, FUPₑₘᵥ; IL-12, IL-10, TNF-α, INF-γ, IL-6, CD4-1, CD4-2, CD8α, CD83, CD18, CD11b-2; | [35]       |
| L. plantarum + L. brevis + L. acidophilus + Bacillus subtilis + Saccharomyces cerevisiae | Dietary probiotics mixtures in which each probiotic amount is 0.1% | 12 weeks | GPₘᵥ; IPₑₘᵥ; IL-12, IL-10, TNF-α, INF-γ, IL-6, CD4-1, CD4-2, CD8α, CD83, CD18, CD11b-2; | [119]      |
| L. fermentum                      | Dietary supplementation of each probiotic at 2.42 × 10^7 CFU g⁻¹ | 8 weeks  | GPₘᵥ; IPₑₘᵥ; IL-12, IL-10, TNF-α, INF-γ, IL-6, CD4-1, CD4-2, CD8α, CD83, CD18, CD11b-2; | [120]      |
| Bacillus SJ-10                    | Dietary supplementation of heat-killed probiotic at 10^7 CFU g⁻¹ | 8 weeks  | GPₘᵥ; IPₑₘᵥ; IL-12, IL-10, TNF-α, INF-γ, IL-6, CD4-1, CD4-2, CD8α, CD83, CD18, CD11b-2; | [125]      |
| Bacillus SJ-10                    | Dietary supplementation at 10^7 CFU g⁻¹ | 8 weeks  | GPₘᵥ; IPₑₘᵥ; IL-12, IL-10, TNF-α, INF-γ, IL-6, CD4-1, CD4-2, CD8α, CD83, CD18, CD11b-2; | [116]      |
| Bacillus sp. SJ-10 and Lab. plantarum KCCM 11322 B. subtilis, B. pumilus, B. licheniformis | Dietary supplementation at 10^7 CFU g⁻¹ | 8 weeks  | GPₘᵥ; IPₑₘᵥ; IL-12, IL-10, TNF-α, INF-γ, IL-6, CD4-1, CD4-2, CD8α, CD83, CD18, CD11b-2; | [43]       |
| Bacillus SJ-10, Lab. plantarum    | Dietary supplementation of each probiotic at 0.5% & 5 days | 8 weeks  | GPₘᵥ; IPₑₘᵥ; IL-12, IL-10, TNF-α, INF-γ, IL-6, CD4-1, CD4-2, CD8α, CD83, CD18, CD11b-2; | [22]       |
| Bacillus SJ-10                    | Dietary supplementation at 10^7 CFU g⁻¹ | 8 weeks  | GPₘᵥ; IPₑₘᵥ; IL-12, IL-10, TNF-α, INF-γ, IL-6, CD4-1, CD4-2, CD8α, CD83, CD18, CD11b-2; | [28]       |
| B. licheniformis SK3927 + B. amylobiophaciens SK4079 + B. subtilis SK4082 + Lab. brevis SK1751 + Lab. plantarum SK3494 + S. cerevisiae SK3587 Lact. lactis BFE920 | Dietary supplementation at 10⁸ – 10⁹ CFU Kg⁻¹ | 12 weeks | GPₘᵥ; IPₑₘᵥ; IL-12, IL-10, TNF-α, INF-γ, IL-6, CD4-1, CD4-2, CD8α, CD83, CD18, CD11b-2; | [29]       |
|                                  | Dietary supplementation at 5 × 10⁸ – 5 × 10⁹ CFU g⁻¹ in laboratory & 2.5 × 10⁸ – 2.5 × 10⁹ CFU g⁻¹ in field experiment | 2 weeks & 3 months | GPₘᵥ; IPₑₘᵥ; IL-12, IL-10, TNF-α, INF-γ, IL-6, CD4-1, CD4-2, CD8α, CD83, CD18, CD11b-2; | [152]      |

Symbol →, no change; †, increase; ↓, decrease versus controls.

ACA: Alternative complement activity; ALT: Alanine aminotransferase; AmmA: Amylase activity; AST: Aspartate aminotransferase; CD: Cluster of differentiation; DEEA: Digestive enzyme activities; FBW: Final body weight; FCR: Feed conversion ratio; FER: Feed efficiency ratio; FOX: Forkhead box; FUP: Feed utilization parameters; Glu: Glucose; GP: Growth parameters; Glx: Glutathione peroxidase; GutM: Gut microbiota; Hb: Haemoglobin; HBP: Haemate-biochemical parameters; IDR: Infectious disease resistance; Ig: Immunoglobulin; IL: Interleukin; INF: Interferon; IP: Immunological parameters; IRGE: Immune related gene expression; MPO: Myeloperoxidase; mVH: micro-Villus height; PC: Phagocytic activity; PCI: Probiotic count in the intestine; PER: Protein efficiency ratio; RB: Respiratory burst (NBT assay); SAP: Serum antiprotease activity; SGR: Specific growth rate (%); SL: Serum lysozyme; SMLA: Skin mucus lysozyme activity; SOD: Superoxide dismutase; SR: Survival rate (%); TC: Total cholesterol; TG: Triglyceride; TGF: Transforming growth factor; TNF: Tumor necrosis factor; TLR5M: Membrane anchored toll-like receptor 5; TSP: Total serum protein; WG: Weight gain (%).
acidobacteria, and Lactobacillus, Marinilactibacillus and Globicatella in the intestine, while also elevating cytokines transcription levels. Moreover, multiple and individual probiotics had no effects on production and intestinal structure respectively but improved infectious disease resistance of experimentally-farmed fish.

Grass Carp

Grass carp is predominant among cultured finishes, comprising 11% of total global aquaculture production in 2016 [1] with production increasing to over 5.7 million tons in 2018, FAO ranked grass carp as the 1st aquacultured species worldwide in 2020 [127]. Several studies have examined the effects of probiotics in grass carp. Dietary B. licheniformis FA6 enhanced production, humoral immunity, upregulated immune gene transcription, intestinal morphology and Aeromonas infection protection in grass carp [128]. Identical results [129] along with modulation of intestinal microbiome [130] were noted after dietary supplementation of B. subtilis Ch9, and both B. methylotrophicus WM-1 [131] and Streptomyces amrissarensis N1-32 [132] contributed in the modulation of immunity and aeromonosis protection (Table 6).

Individual and mixture of Shewanella xiamenensis A-1, S. xiamenensis A-2, and A. veronii A-7 at 105 CFU g−1 feed enhanced cellular and humoral immunity, serum biochemistry, gene transcription and offered protection [133]. Administration of S. xiamenensis A-1, A. veronii A-7 and B. subtilis modulate the gut microbiota either singly or in combination, in which proportions of Cetobacterium, Citrobacter, Vibrio, Enterococcus and Streptococcus were increased, although a decreasing number was noted for Pseudomonas and Flavobacterium [134].

Several studies were carried out to examine the effects of probiotics on water quality parameters and microbiota

Table 6. Effects of probiotic supplementation on growth, feed utilizations, immunological and haematobiochemical parameters and disease resistance in grass carp (Ctenopharyngodon idellus).

In Table 6, the effects of probiotics on growth, feed utilizations, immunological and haematobiochemical parameters and disease resistance in grass carp (Ctenopharyngodon idellus) are summarized. The table lists the probiotics, their mode of administration and dosage, duration, effects on growth (WG, FBW, SGR, and HBP), feed conversion ratio (FCR), immunity (GP, skin, and TLR4), gene expression (IRGE), antioxidant capacity (TAC), and clotting factors (ALT, AST, and AP). The references are also provided for further reading.

Symbol: →, no change; ↑, increase; ↓, decrease versus controls.

ACP: Acid phosphatase; ALT: Alanine aminotransferase; AST: Aspartate aminotransferase; Alb: Albumin; AmA: Amylase activity; C3: Complement C3; CAT: Catalase; COD: Chemical oxygen demand; DEA: Digestive enzyme activities; FCR: Feed conversion ratio; FUP: Feed utilization parameters; Glb: Globulin; GP: Growth parameters; HBP: Haemato-biochemical parameters; IDR: Infectious disease resistance; Ig: Immunoglobulin; IL: Interleukin; MDA: Malondialdehyde; MEF2C: Myocyte enhancer factor 2C; MPO: Myeloperoxidase; MyD: Myeloid differentiation factor; MyoG: Myogenin; Nrf: Nuclear factor (erythroid-derived-2) like gene; PCa: Phagocytic activity; PrA: Protease activity; RB: Respiratory burst (NBT assay); SGR: Specific growth rate (%); SL: Serum lysozyme; SOD: Superoxide dismutase; SR: Survival rate; TAC: Total anti-oxidant capacity; TC: Total cholesterol; TG: Triglyceride; TDS: Total dissolved solids; TLR: Toll like receptor; TNF: Tumor necrosis factor; TSP: Total serum protein; VH: Villous height; WG: Weight gain (%); WQ: Water quality parameters.

June 2022 | Vol. 32 | No. 6
in the culture environment of grass carp. The addition of *P. stutzeri* F11 [135] and *B. licheniformis* BSK-4 [136] in water led to reduction of ammonia, nitrite, total nitrogen and altered microbial populations at 9 and 18 days, respectively (Table 6). In another study, application of individual and mixture of *P. stutzeri* SC221-M and *B. cereus* BSC24 in culture water improved the rearing water quality as revealed by lower levels of total dissolved solids, chemical oxygen demand, ammonium, nitrite and total nitrogen compared to the control after 6-days post-treatment [137]. Moreover, these probiotics increased proteobacteria and decreased bacteroidetes and actinobacteria in ambient water.

### Rohu Carp
*Labeo rohita*, commonly known as Indian major carp or rohu, is among the most cultured fish species in South Asia due to its commercial value, fast growth rate, consumer preferences and comparative simplicity in weaning to artificial diets [138, 139]. Global production of rohu was 1.843 million tons in 2016, contributing 3% of total global fish production [1]. Weakened immune system and lack of necessary digestive enzymes in juveniles can lead to higher rates of pathogenic bacterial infection and relatively poor feed utilization [138, 140]. Efforts have been made continuously to subdue these problems, including many involving applications of probiotics. Higher serum IgM levels were recorded after 30 days, and the level decreased after 60 days. Similarly, mixtures of these probiotics with *B. subtilis* VSG1 modulated and generally improved similar immunological parameters,

### Table 7. Effects of probiotic supplementation on growth, feed utilizations, immunological and haematobiochemical parameters and disease resistance in rohu fish (*Labeo rohita*).

| Probiotics                                      | Mode of administration and dosage | Duration | Effects on *L. rohita*                                      | References |
|------------------------------------------------|----------------------------------|----------|------------------------------------------------------------|------------|
| *Bacillus subtilis* VSG1, *L. plantarum* VSG3, & *P. aeruginosa* VSG2 | Dietary supplementation at (0.5 & 1) × 10^7 CFU g^-1 | 60 days  | GP \( \uparrow \) FUP, IP, TSP, AC, UA, RR, TCH, TGL, SoD, & SOD; IDR \( \uparrow \) A. hydrophila; TSP \( \uparrow \) | [141]      |
| *B. subtilis*, *L. lactis*, & *Saccharomyces cerevisiae* | Dietary supplementation at 10^7 CFU Kg^-1 | 30 days  | GP \( \uparrow \) HBP, IP, AC, UA, RR, TCH, TGL, SoD, & SOD; A. hydrophila; TSP \( \uparrow \); Apoptosis \( \uparrow \) | [142]      |
| *B. methylotrophicus*, *B. amyloliquefaciens*, & *B. licheniformis* | Dietary supplementation at 10^7 CFU g^-1 | 60 days  | GP \( \uparrow \) FUP, TGL, & PC; GP \( \downarrow \) HBP, IP, AC, UA, RR, TCH, TGL, SoD, & SOD; TSP \( \uparrow \) | [143]      |
| *B. aerophilus* KADR3 | Dietary supplementation at 10^7 & 10^8 CFU g^-1 | 3 & 6 weeks | HBP \( \uparrow \) IP, AC, UA, RR, SoD, & SOD; IDR \( \uparrow \) A. hydrophila; TSP \( \uparrow \) | [20]       |
| *B. amyloliquefaciens* CCF7 | Dietary supplementation at 10^7 & 10^8 CFU g^-1 | 70 & 28 days | HBP \( \uparrow \) IP, AC, UA, RR, SoD, & SOD; IDR \( \uparrow \) A. hydrophila; TSP \( \uparrow \) | [144]      |
| *G. candidum* QAUCG01 & *B. cereus* | Water supplementation at 10^7 CFU L^-1 | 70 days  | GP \( \uparrow \) HBP, IP, AC, UA, RR, SoD, & SOD; IDR \( \uparrow \) A. hydrophila; TSP \( \uparrow \) | [145]      |
| **Enterococcus faecium** | Dietary supplementation at 10^7 CFU g^-1 | 11 weeks  | GP \( \uparrow \) HBP, IP, AC, UA, RR, SoD, & SOD; IDR \( \uparrow \) A. hydrophila; TSP \( \uparrow \) | [138]      |
| **Lactobacillus spp., Streptococcus faecium, *B. bifidum*, *L. subtilis*, *Saccharomyces spp.*, *Torulopsis & Aspergillus oryzae*** | Dietary supplementation at 0.5, 0.75 & 1% | 60 days  | GP \( \uparrow \) FUP, TGL, & PC; GP \( \downarrow \) HBP, IP, AC, UA, RR, SoD, & SOD; IDR \( \uparrow \) A. hydrophila; TSP \( \uparrow \) | [147]      |
| **G. candidum** QAUCG01 | Dietary supplementation at 10^7 CFU g^-1 | 11 weeks  | GP \( \uparrow \) HBP, IP, AC, UA, RR, SoD, & SOD; IDR \( \uparrow \) A. hydrophila; TSP \( \uparrow \) | [154]      |
| *Lactobacillus* spp. | Dietary supplementation at 1 & 1.5 g Kg^-1 | 30 days  | GP \( \uparrow \) HBP, IP, AC, UA, RR, SoD, & SOD; IDR \( \uparrow \) A. hydrophila; TSP \( \uparrow \) | [155]      |

Symbol \( \uparrow \), no change; \( \downarrow \), increase; \( \uparrow \), decrease versus controls.

ACA: Alternative complement activity; ACP: Acid phosphatase; ALP: Alkaline phosphatase; ALT: Alanine aminotransferase; AmA: Amylase activity; AST: Aspartate aminotransferase; CAT: Catalase; DEA: Digestive enzyme activities; FCE: Feed conversion efficiency; FCR: Feed conversion ratio; FUP: Feed utilization parameters; Gb: Globulin; Glu: Glucose; GP: Growth parameters; GutM: Gut microbiota; Hb: Haemoglobin; HBP: Haemato-biochemical parameters; HSP70: Heat shock protein; IDR: Infectious disease resistance; IgM: Immunoglobulin M; IL: Interleukin; IP: Immunological parameters; IRGE: Immune related gene expression; MDA: Malondialdehyde; MPO: Myeloperoxidase; PCA: Phagocytic activity; PER: Protein efficiency ratio; PrA: Protease activity; RB: Respiratory burst (NBT assay); RBC: Red blood cell; SAP: Serum antiprotease activity; SBA: Serum bacterial activity; SGR: Specific growth rate (%); SL: Serum lysozyme; SOA: Superoxide anion; SOD: Superoxide dismutase; SR: Survival rate; TC: Total cholesterol; TG: Triglyceride; TNF: Tumor necrosis factor; TSP: Total serum protein; WBC: White blood cell; WG: Weight gain (%).
including disease resistance [141] (Table 7). Feeding of *B. subtilis, L. lactis* and *S. cerevisiae* mixture at different culture temperatures (28, 31, 34, and 37°C) reduced the degree of apoptosis, augmented the transcription of HSP70, positively modulated haematobiochemistry and innate immunity compared to the control [142]. Supplementation with *B. methylotrophicas, B. amylophilicae* and *B. licheniformis* either singly or in combination elevated digestive enzymes activity, immunity and offered a significant degree of infection protection [143]. Similarly, disease protection and immunity alteration were observed after administration with *B. aerophilus KAD3* [20], *B. amylophilicae* CCE7 [144] and COFCAL_P1 [145] (Table 7). 

Improved protease, amylase and cellulase resulting on good growth in rohu fingerlings were observed after adding *Geothichum candidum QAUGC01* to rearing water [140]. Similarly, *G. candidum QAUGC01, B. cereus* and *G. candidum QAUGC01 + B. cereus* at 10³ CFU g⁻¹ elevated gut microbiome, digestive enzymes and survivability against *A. hydrophila* infection [138]. Similar parameters modulation with improvements in the growth increment were observed after administration of single *E. faecium QAUEF01* and its combination with *G. candidum QAUGC01* [146] and *S. cerevisiae* [147].

**Probiotics Application under Farming Condition**

Most studies on probiotics were conducted under laboratory conditions, in which water and ambient environmental parameters are subject to rigid control. The role of probiotics in open systems under farming conditions remains minimally studied. While performance of probiotics under uncontrolled field levels is subject to uncertainty, commercial manufacturers of probiotics frequently boast that their products are highly effective for farm level application.

Tilapia cultured in earthen ponds treated with two doses of *B. pumilus* and a commercial probiotic for 8 months showed increase growth, immunity and haematological parameters, and *A. hydrophila* infection protection [79] (Table 1). Artificially cultured fishes are subjected in the hatchery to water quality and other parameters that can compromise the consistent production of healthy juveniles [148], although probiotic applications among broodstock, embryos and larve have been studied inadequately. Female rainbow trout brood stock in raceway ponds supplemented with multi-probiotics before spawning season demonstrated production of eggs with higher diameters, increased fecundity, fertilization and hatching, yolk sac absorption, and eye development [14]. These results are consistent with beneficial responses to multiple-species LAB probiotic applications in the zebrafish hatchery, which led to consistent improvements in every measurable parameter of larval performance [149]. Moreover, multi-probiotics supplementation in rainbow trout during grow out in floating cages led to improved haematobiochemistry and operational taxonomic units in the intestine [150] (Table 3). Common carp (2.75 g) cultured in raceways and fed with *B. subtilis* for 180 days at 4 × 10⁸ CFU g⁻¹ showed elevated production and feed utilizations as contrasted with controls [151]. Moreover, probiotics increased muscle RNA-DNA ratio along with crude lipid, protein, and ash content in the body (Table 4). Single supplementation of *L. lactis* BFE920 at different concentrations increased protection against streptococcosis (68-77%) after 2 weeks. Subsequently, 2.5 × 10⁶ CFU g⁻¹ was chosen for field experiment for 3 months in 12,000 olive flounder [152]. That experiment revealed that probiotics fed group displayed improved immunity and immune related gene expression, resulting in 65.7% survival relative to 5.7% in control group when challenged with *S. iniae* (Table 5).

Cage culture of grass carp (44 g) for 56 days with *B. natto* NT supplementation improved body weight and feed utilization [153]. Myostatin and myocyte enhancer factor C were downregulated, whereas micro-RNA (miR)-1a, miR-181a, miR-23a, and miR-206 transcription were upregulated compared to control group (Table 6). Rohu carp in earthen ponds displayed improved immunity and digestive enzyme activities along with enhanced tissue HSP70 expression after 11 weeks feeding with encapsulated *G. candidum QAUGC01* [154]. Multi-probiotics administration in that species in outdoor tanks resulted in higher survival of hatchlings and fry at 8 and 38 days, respectively but those differences were no longer detectable after 68 days [155] (Table 7).

**Research Gaps and Concluding Remarks**

Some urgency is attached to the transition from the use and at times excessive use of antibiotics to control pathogens in aquaculture. The effectiveness of antibiotics is counteracted by intense selection favouring resistance, leading to the emergence of dangerously capable pathogens. The hazards of routine antibiotic applications are well-recognized now, and alternatives are critically needed. Probiotics are being marketed to gain favour, the mechanisms involved are far from clear. Probiotic organisms decrease the likelihood of the impairment or mortality of culture subjects by pathogens through a poorly understood complex of mechanisms, and more specific understanding of these mechanisms is highly desired. Interactive mechanisms of probiotic species with intestinal cells of host subjects require further investigation and analysis.

Probiotics control the development of an unfavourable microflora, thereby disabling opportunistic microbes including many pathogens, thereby reducing the prevalence of outbreaks of infectious organisms. This can happen by direct and indirect means, for example by generating metabolites that are inhibitory to pathogens, or that promote activation of the immune system, or sometimes simply by competing for nutrients and environmental niches. As discussed above, activation of growth- and
immune-related genes have been observed and may be more symptomatic of beneficial health effects than unknown causative factors. Further study of the mechanisms resulting in beneficial impacts of probiotic organisms may lead to more effective treatments and ultimately to improved environmental management of commercially cultured fishes and invertebrates.

Acknowledgments
This study was supported by Brain Pool Scholarship (Grant no.: 2021H1D3A2A01099381) funded by National Research Foundation of Korea (NRF) and NRF grant funded by Ministry of Education (Grant no.: 2021R1J1A1A01049238).

Conflict of Interest
The authors have no financial conflicts of interest to declare.

References
1. FAO. 2018. Fisheries Department, Fishery Information, Data and Statistics Unit. FishStatJ, a tool for fishery statistics analysis. Global aquaculture production: Quantity 1950–2016; Value 1950–2016; Global capture production: 1950–2016; 2018-03-16.
2. Hoseinifar SH, Sun Y-Z, Wang A, Zhou Z. 2018. Probiotics as means of diseases control in aquaculture, a review of current knowledge and future perspectives. Front. Microbiol. 9:2429.
3. Dawood MA, Koshio S, Abdel-Daim MM, Van Doan H. 2019. Probiotic application for sustainable aquaculture. Rev. Aquac. 11: 907-924.
4. Berg A, Rødsæth OM, Hansen T. 2007. Fish size at vaccination influence the development of side-effects in Atlantic salmon (Salmo salar L.). Aquaculture 265: 9-15.
5. Wang A, Ran C, Wang Y, Zhang Z, Ding Q, Yang Y, et al. 2019. Use of probiotics in aquaculture of China—a review of the past decade. Fish Shellfish Immunol. 86: 734-755.
6. Hasan MT, Jang WJ, Lee S, Kim KW, Lee BJ, Han HS, et al. 2018a. Effect of β-glucooligosaccharides as a new probiotic for dietary supplementation in olive flounder (Paralichthys olivaceus) aquaculture. Aquac. Res. 49: 1310-1319.
7. Dawood MA, Koshio S. 2016. Recent advances in the role of probiotics and prebiotics in carp aquaculture: a review. Aquaculture 454: 243-251.
8. Hasan MT, Je Jang W, Lee JM, Lee B-J, Hur SW, Gu Lim S, et al. 2019a. Effects of immunostimulants, prebiotics, probiotics, synbiotics, and potentially immunoreactive feed additives on olive flounder (Paralichthys olivaceus). Fish Shellfish Immunol. 82: 544-553.
9. Parker R. 1974. Probiotics, the other half of the antibiotic story. Anim. Nutr. Health 29: 4-8.
10. Merrifield DL, Dimitroglou A, Foey A, Davies SJ, Baker RT, Bøgwald J, et al. 2016. Probiotics in aquaculture: challenges and outlook. Aquaculture 365-374.
11. Akbari Nargesi E, Falahatkar B, Sajjadi MM. 2020. Dietary supplementation of probiotics and influence on feed efficiency, growth performances, digestive enzyme activities and gut histomorphology of striped catfish (Labeo rohita). Aquac. Nutr. 26: 316-327.
12. Naderi Farsani M, Bahrami Gorji S, Hoseinifar SH, Rashidian G, Van Doan H. 2020. Combined and singular effects of dietary pimaric acid and potassium diformate (KDF) on growth performance and some physiological parameters of rainbow trout (Oncorhynchus mykiss), Probiotics Antimicrob. Proteins 12: 236-245.
13. Makled SO, Hamdan AM, El-Sayed A-FM. 2020. Growth promotion and immune stimulation in nile tilapia, (Oreochromis niloticus) aquaculture. Fish Shellfish Immunol. 265: 9-15.
14. Angeline Ignacimuthu S, Austin B. 2017. Probiotics for disease control in aquaculture. Published: John Wiley & Sons, Inc.
15. Hasan MT, Jang WJ, Kim H, Lee BJ, Kim KW, Hur SW, et al. 2018b. Synergistic effects of dietary Bacillus sp. Sf-10 plus β-glucooligosaccharides as a synbiotic on growth performance, innate immunity and streptococcosis resistance in olive flounder (Paralichthys olivaceus). Fish Shellfish Immunol. 82: 544-553.
16. Beck BR, Song JH, Park BS, Kim D, Kwak I-H, Do HK, et al. 2016. Distinct immune tones are established by Lactococcus lactis BFE920 and Lactobacillus plantarum FGL0001 in the gut of olive flounder (Paralichthys olivaceus). Fish Shellfish Immunol. 55: 434-443.
17. Park Y, Kim H, Won S, Hamidoghli A, Hasan MT, Kong IS, et al. 2020. Effects of two dietary probiotics (Bacillus subtilis or licheniformis) with two prebiotics (mannan or fructo oligosaccharide) in Japanese eel, Anguilla japonica. Aquac. Nutr. 26: 316-327.
18. Akter MN, Hashim R, Satrana A, Siti Azzahah MN, Asaduzzaman M. 2019. Effect of Lactobacillus acidophilus supplementation on growth performances, digestive enzyme activities and gut histomorphology of striped catfish (Pangasianodon hypophthalmus) juveniles. Aquac. Res. 50: 786-798.
19. Remesh D, Souissi S, Ahamed TS. 2017. Effects of the potential probiotics Bacillus aerophilus KAD3 in inducing immunity and disease resistance in Labeo rohita. Fish Shellfish Immunol. 70: 408-415.
20. Hasan MT, Jang WJ, Kim H, Lee BJ, Kim KW, Hur SW, et al. 2018c. Synergic effects of dietary Bacillus sp. Sf-10 plus β-glucooligosaccharides as a synbiotic on growth performance, innate immunity and streptococcosis resistance in olive flounder (Paralichthys olivaceus). Fish Shellfish Immunol. 82: 544-553.
21. Cha I-H, Rahimnejad S, Yang S-Y, Kim K-W, Lee K-J. 2013. Evaluations of Bacillus spp. as dietary additives on growth performance, innate immunity and disease resistance of olive flounder (Paralichthys olivaceus) against Streptococcus iniae and as water additives. Aquaculture 402: 50-57.
22. Wang Y-B, Li J-R, Lin J. 2008a. Probiotics in aquaculture: challenges and outlook. Aquaculture 281: 1-4.
23. Huang W, Chang I, Wang P, Liu C, Yin Q, Zhu Q, et al. 2018. Effect of the combined compound probiotics with mycotoxin-degradation enzyme on detoxifying aflatoxin B1 and zearalenone. J. Toxicol. Sci. 43: 377-385.
27. Nguafack TT, Jang WJ, Hasan MT, Choi YH, Bai SC, Lee E-W, et al. 2020. Effects of dietary non-viable *Bacillus* sp. SJ-10, *Lactobacillus plantarum*, and their combination on growth, humoral and cellular immunity, and streptococcus resistance in olive flounder (*Paralichthys olivaceus*). *Res. Vet. Sci.* 131:177-185.

28. Jang WJ, Lee JM, Hasan MT, Lee B-J, Lim SG, Kong J-S. 2019a. Effects of probiotic supplementation of a plant-based protein diet on intestinal microbial diversity, digestive enzyme activity, intestinal structure, and immunity in olive flounder (*Paralichthys olivaceus*). *Fish Shellfish Immunol.* 92:719-727.

29. Jang WJ, Hasan MT, Lee GH, Lee B-J, Hur SW, Lee S, et al. 2021b. Comparison of spore or vegetative *Bacillus* sp. supplementation on physiological changes and gut microbiota of the olive flounder (*Paralichthys olivaceus*). *Aquaculture* 535:736555.

30. Sharifuzzaman S, Al-Harbi A, Austin B. 2014. Characteristics of growth, digestive system functionality, and stress factors of rainbow trout fed probiotics *Kocuria* SN1 and *Rhodococcus SM2*. *Aquaculture* 418:55-61.

31. Dawood MA, Eweedah NM, Mourafeti EM, Farhat EM. 2020a. Probiotic effects of *Aspergillus oryzae* on the oxidative status, heat shock protein, and immune related gene expression of Nile tilapia (*Oreochromis niloticus*) under hypoxia challenge. *Aquaculture* 520:734669.

32. Barroso C, Ozório RO, Afonso A, Moraes JR, Costa R. 2016. Immune responses and gut morphology in Senegalese sole (*Solea senegalensis*) fed dietary probiotic supplementation and following exposure to *Photobacterium damselae* subsp. piscicida. *Aquac. Res.* 47:951-960.

33. Kashem MA, Uddin MN, Hossain MM, Hasan MT, Haque SA, Khan MNA. 2020. Effects of dietary probiotic *Lactobacillus plantarum* KCCM 11322 combinations enhance growth and cellular and humoral immunity in olive flounder (*Paralichthys olivaceus*). *Probiotics Antimicrob. Proteins* 13:1277-1291.

34. Kordon AO, Karsi A, Pinchuk L. 2018. Innate immune responses in fish: Antigen presenting cells and professional phagocytes. *Turkish J. Fish. Aquat. Sci.* 18:1123-1138.

35. Beck BR, Kim D, Jeon J, Lee S-M, Kim HK, Kim O-J, et al. 2015. The effects of combined dietary probiotics *Lactococcus lactis* BFE920 and *Lactobacillus plantarum* FGL0001 on innate immunity and disease resistance in olive flounder (*Paralichthys olivaceus*). *Fish Shellfish Immunol.* 42:177-183.

36. Van Doan H, Hoseini AF, Khanvandch M, Kanipangi A, Unban K, Srichaiy M. 2018. Host-associated probiotics boosted mucosal and serum immunity, disease resistance and growth performance of Nile tilapia (*Oreochromis niloticus*). *Aquaculture* 491:94-100.

37. Dobato C, Moncunill G. 2018. Naturally acquired immunity (NAI), pp. 1-15. *Encyclopedia of malaria*. Springer New York, New York, NY.

38. Hoseini AF, Mirvaghefi A, Amoozegar MA, Sharifan M, Esteban MA. 2015. Modulation of innate immune response, mucosal parameters and disease resistance in rainbow trout (*Oncorhynchus mykiss*) upon syngeneic feeding. *Fish Shellfish Immunol.* 45:27-32.

39. Akhher N, Wu B, Memon AM, Mohsin M. 2015. Probiotics and probiotics associated with aquaculture: a review. *Fish Shellfish Immunol.* 45:733-741.

40. Kim Y-R, Kim E-Y, Choi S-Y, Lim SG, Kim KW, Oh R-K, et al. 2013. Effect of a probiotic strain, *Enterococcus faecium*, on the immune responses of olive flounder (*Paralichthys olivaceus*). *J. Microbiol. Biotechnol.* 22:526-529.

41. Kono T, Poppompsiat A, Sakai M. 2004. The analysis of expressed genes in head kidney of common carp (*Cyprinus carpio*) stimulated with peptidoglycan. *Aquaculture* 235:37-52.

42. Schellner J, Chalaris A, Schmidt-Arras D, Rose-John S. 2011. The pro- and anti-inflammatory properties of the cytokine interleukin-6. *Biochim. Biophys. Acta* 1813:878-888.

43. Hasan MT, Jang WJ, Lee B-J, Hur SW, Lim SG, Kim KW, et al. 2021. Dietary supplementation of *Bacillus* sp. SJ-10 and *Lactobacillus plantarum* KCCM 11322 combinations enhance growth and cellular and humoral immunity in olive flounder (*Paralichthys olivaceus*). *Probiotics Antimicrob. Proteins* 13:1277-1291.

44. Brunt J, Hansen R, Jamieson DJ, Austin B. 2008. Proteomic analysis of rainbow trout (*Oncorhynchus mykiss*, *Walbaum*) serum after administration of probiotics in diets. *Vet. Immunol. Immunopathol.* 121:199-205.

45. Galdeano CM, Cazorla SI, Dumit JML, Vélez E, Perdigón G. 2018. Oral administration of probiotics increases paneth cell numbers and mucus production in rainbow trout (*Oncorhynchus mykiss*). *FEMS Immunol. Med. Microbiol.* 29:47-52.

46. Sankaran-Walters S, Hart R, Dills C. 2017. Guardians of the gut: enteric defensins. *Ann. Nutr. Metab.* 74:115-124.

47. Link-Amster H, Rochat F, Saudan K, Mignot O, Aeschlimann J. 1994. Modulation of a specific humoral immune response and changes in intestinal flora mediated through fermented foods. *Fed. Immunol.* 7:65-69.

48. Fang H, Elma T, Heikki A, Seppo S. 2000. Modulation of humoral immune response through probiotic intake. *FEBS Immunol. Med. Microbiol.* 49:25-72.

49. Pringle JD, Klesius PH. 2011. Development and efficacy of a novel bovibiotic-resistant *Streptococcus iniae* as a novel vaccine in Nile tilapia (*Oreochromis niloticus*). *Vaccine* 29:5986-5993.

50. Rindinen M, Jalava K, Westermarck E, Salminen S, Ouwehand AC. 2003. Interaction between probiotic lactic acid bacteria and canine enteric pathogens: a risk factor for intestinal *Enterococcus faecium* colonization? *Vet. Microbiol.* 92:111-119.

51. Abou-El-Atta ME, Abdel-Tawwab M, Abdel-Razek N, Abdelhamik TM. 2019. Effects of dietary probiotic *Lactobacillus plantarum* strain and whey protein concentrate on the productive parameters, immunity response and disease resistance of Nile tilapia, *Oreochromis niloticus* (L.), to *Aeromonas sobria* infection. *Aquac. Nutr.* 25:1367-1377.

52. Yu L, Zhai Q, Zhu J, Zhang C, Li T, Liu X, et al. 2017. Dietary *Lactobacillus plantarum* supplementation enhances growth performance and alleviates aluminum toxicity in tilapia. *Toxicol. Environ. Safety* 143:307-314.

53. Zhai Q, Wang H, Tian F, Zhao J, Zhang H, Chen W. 2017. Dietary *Lactobacillus plantarum* supplementation decreases tissue level accumulation and alleviates lead toxicity in Nile tilapia (*Oreochromis niloticus*). *Aquac. Res.* 48:5094-5103.

54. Van Doan H, Hoseini AF, Tapikev H, Tongrse S, Khamtavee P. 2016. Combined administration of low molecular weight sodium alginate boosted immunomodulatory, disease resistance and growth enhancing effects of *Lactobacillus plantarum* in Nile tilapia (*Oreochromis niloticus*). *Fish Shellfish Immunol.* 56:678-685.

55. Poolsawat L, Li X, He M, J D, Leng X. 2020. *Clostridium butyricum* as probiotic for promoting growth performance, feed utilization, gut health and microbiota community of tilapia (*Oreochromis niloticus* O. aureus). *Aquac. Nutr.* 26:657-670.

56. Li H, Zhou Y, Ling H, Luo L, Qi D, Feng L. 2019. The effect of dietary supplementation with *Clostridium butyricum* on the growth performance, immunity, intestinal microbiota and disease resistance of tilapia (*Oreochromis niloticus*). *PLoS One* 14:e0223428.

57. Liu H, Wang S, Cai Y, Guo X, Cao Z, Zhang Y, et al. 2017. Dietary administration of *Bacillus subtilis* strain KCCM 41590 enhances growth, digestive enzyme activities, innate immune responses and disease resistance of tilapia, *Oreochromis niloticus*. *Fish Shellfish Immunol.* 66:326-333.

58. Gohi N, Vasezhanzhan B, Chen J-C, Rekha R, Vijayakumar S, Anijagam M, et al. 2018. Dietary supplementation of probiotic *Bacillus licheniformis* Dahi1 improves growth performance, mucus and serum immune parameters, antioxidant enzyme activity as well as resistance against *Aeromonas hydrophila* in tilapia *Oreochromis mossambicus*. *Fish Shellfish Immunol.* 74:501-508.

59. Abarike ED, Jian J, Tang J, Cai J, Yu H, Lihua C, et al. 2018. Influence of traditional Chinese medicine and *Bacillus* species (TCMBS) on growth, immune response and disease resistance in Nile tilapia, *Oreochromis niloticus*. *Aquac. Res.* 49:2366-2375.
61. Samson JS, Choresca CH, Quiason KMA. 2022. Probiotic effect of Bacillus spp. isolated from African nightcrawler (Eudrilus eugeniae) on the performance of Nile tilapia (Oreochromis niloticus L.). Arch. Microbiol. 204: 235.

62. Wang M, Liu G, Lu M, Ke X, Liu Z, Gao Y, et al. 2017. Effect of Bacillus cereus as a water or feed additive on the gut microbiota and immunological parameters of Nile tilapia. Aquac. Res. 48: 3163-3173.

63. Ramos MA, Gonçalves JP, Costas B, Batista S, Lochmann R, Pires MA, et al. 2017. Commercial Bacillus probiotic supplementation of rainbow trout (Oncorhynchus mykiss) and brown trout (Salmo trutta): growth, immune responses and intestinal morphology. Aquac. Res. 48: 2538-2549.

64. Docando E, Nuñez-Oritz N, Serra C, Arense P, Enes P, Oliva-Teles A, et al. 2022. Mucosal and systemic immune effects of Bacillus subtilis in rainbow trout (Oncorhynchus mykiss). Fish Shellfish Immunol. 124: 142-155.

65. Chen S-W, Liu C-H, Hu S-Y. 2019. Dietary administration of probiotic Paenibacillus ehimensis NPUST1 with bacteriocin-like activity improves growth performance and immunity against Aeromonas hydrophila and Streptococcus iniae in Nile tilapia (Oreochromis niloticus). Fish Shellfish Immunol. 84: 695-703.

66. Dawood MA, Moustafa EM, Gewaily MS, Abdo SE, AbdEl-Kader MF, SaadAllah MS, et al. 2020. Ameliorative effects of Lactobacillus plantarum L-137 on Nile tilapia (Oreochromis niloticus) exposed to deltamethrin toxicity in rearing water. Aquat. Toxicol. 219: 105377.

67. Tan HY, Chen S-W, Hu S-Y. 2019. Improvements in the growth performance, immunity, disease resistance, and gut microbiota by the probiotic Bummelebacillus stabekeis in Nile tilapia (Oreochromis niloticus). Fish Shellfish Immunol. 92: 265-275.

68. Makded SO, Hamdan AM, El-Sayed A-FM, Hafer EE. 2017. Evaluation of marine psychrophile, Psychrobacter nanhaeensis SO89, as a probiotic in Nile tilapia (Oreochromis niloticus) diets. Fish Shellfish Immunol. 61: 194-200.

69. Maas RM, Verdegem MC, Debnath S, Marchal L, Schrama JW. 2021. Effect of enzymes (phytase and xylanase), probiotics (B. amylycopapiliferus) and their combination on growth performance and nutrient utilisation in Nile tilapia. Aquaculture 533: 736226.

70. Kuebutornye FK, Wang Z, Lu Y, Abarike ED, Sakyi ME, Li Y, et al. 2020. Effects of three host-associated Bacillus species on mucosal immunity and gut health of Nile tilapia, Oreochromis niloticus and its resistance against Aeromonas hydrophila infection. Fish. Shellfish Immunol. 97: 83-95.

71. Sookchayaporn N, Srisapono P, Unajak S, Areechoon N. 2020. Efficacy of Bacillus spp. isolated from Nile tilapia Oreochromis niloticus Linn. on its growth and immunity, and control of pathogenic bacteria. Fish. Sci. 86: 353-365.

72. Won S, Hamandhli A, Cho W, Park Y, Jang WJ, Kong I-S, et al. 2020. Effects of four probiotics, singular or combined, on growth performance and gut health of Nile tilapia, Oreochromis niloticus. Fish Shellfish Immunol. 105: 253-259.

73. Dawood MA, Moustafa EM, Gewaily MS, AbdEl-Kader MF, SaadAllah MS, et al. 2020. Meta-omics technologies reveals beneficiary effects of Lactobacillus plantarum L-137 on Nile tilapia (Oreochromis niloticus) exposed to deltamethrin toxicity in rearing water. Aquat. Toxicol. 219: 105377.

74. Foyyal MF, Alam M, Kawser AR, Hasan F, Rahman MM, Tuy C-Y, et al. 2020. Meta-omics technologies reveals beneficiary effects of Lactobacillus plantarum as dietary supplements on gut microbiota, immune response and disease resistance of Nile tilapia (Oreochromis niloticus). Aquaculture 520: 734074.

75. Dowidar M, Abd ElAzeem S, Khater A, Awad Somayah M, Metwally S. 2018. Improvement of growth performance, immunity and gut health of Nile tilapia, Oreochromis niloticus and its resistance against Aeromonas hydrophila. Fish Shellfish Immunol. 87: 229-233.

76. El-Kady AA, Magouz FI, Mahmoud SA, Abdel-Rahim MM. 2022. The effects of some commercial probiotics as water additive on water quality, fish performance, blood biochemical parameters, expression of growth and immune-related genes, and histology of Nile tilapia (Oreochromis niloticus). Aquaculture 546: 737249.

77. Han B, Long W-Q, He J-Y, Liu Y-J, Si Y-Q, Tian L-X. 2015. Effects of dietary Bacillus licheniformis on growth performance, immunological parameters, intestinal morphology and resistance of juvenile Nile tilapia (Oreochromis niloticus) to challenge infections. Fish Shellfish Immunol. 46: 225-231.

78. He S, Zhou Z, Liu Y, Shi P, Yao B, Ringer E, et al. 2009. Effects of dietary Saccharomycodes cerevisiae fermentation product (DVAQUA®) on growth performance, intestinal autochthonous bacterial community and non-specific immunity of hybrid tilapia (Oreochromis niloticus × O. aureus) cultured in cages. Aquaculture 294: 99-107.

79. Aly SM, Mohamed ME, John G. 2008. Effect of probiotics on the survival, growth and challenge infection in Tilapia nilotica (Oreochromis). Aquac. Res. 39: 647-656.

80. Wang C, Liu Y, Sun G, Li X, Liu Z. 2019. Growth, immune response, antioxidant capability, and disease resistance of juvenile Atlantic salmon (Salmo salar L.) fed Bacillus velezensis V4 and Rodotorula mucilaginosa compound. Aquaculture 500: 65-74.

81. Jaramillo-Torres A, Rawling MD, Roditis A, Mikalsen HE, Johannsen L-H, Tinsley I, et al. 2019. Influence of dietary supplementation of probiotic Pedococcus acidilactici MA18/SM during the transition from freshwater to seawater on intestinal health and microbiota of Atlantic salmon (Salmo salar L.). Front. Microbiol. doi:10.3389/fmicb.2019.02243.

82. Kiron V, Kulkarini A, Dahlé D, Lokehi J, Kitiya Y. 2015. A microbial feed additive abates intestinal inflammation in Atlantic salmon. Front. Immunol. 6: 409.

83. Gupta S, Felkannová A, Lokeš J, Kočířová J, Šorensen M, Fernandes J, et al. 2019. Lactobacillus dominate in the intestine of Atlantic salmon fed dietary probiotics. Front. Microbiol. 9: 3247.

84. Nimalan N, Šorensen SL, Felkannová A, Kočířová J, Mudroňová D, Gancarciková S, et al. 2022. Mucosal barrier status in Atlantic salmon fed marine or plant-based diets supplemented with probiotics. Aquaculture 547: 775716.

85. Salinas I, Myklebust R, Esteban MA, Olsen RE, Mesguer J, Ringer E. 2008. In vivo studies of Lactobacillus delbrueckii subsp. lactis in Atlantic salmon (Salmo salar L.) foregut: tissue responses and evidence of protection against Aeromonas salmonicida subsp. salmonicida epithelial damage. Vet. Microbiol. 128: 167-177.

86. Kristiansen M, Ringer E. 2011. Evaluation of probiotic and probiotic effects on the intestinal gut microbiota and histology of Atlantic salmon (Salmo salar L.). J. Aquac. Res. Develop S1: 009.

87. Ringø E, Salinas I, Olsen R, Nyhaug A, Myklebust R, Mayhew T. 2007. Histological changes in intestine of Atlantic salmon (Salmo salar L.) following in vitro exposure to pathogenic and probiotic bacterial strains. Cell Tissue Res. 328: 109-116.

88. Soltani M, Kane A, Taheri-Mirghaed A, Pakzad K, Hosseini-Shakerabari P. 2019a. Effect of the probiotic, Lactobacillus plantarum on growth performance and haematological indices of rainbow trout (Oncorhynchus mykiss) immunized with bivalent streptococcus/streptococcus vaccine. Iran. J. Fish. Sci. 18: 283-295.

89. Soltani M, Pakzad K, Taheri-Mirghaed A, Mirzargar S, Shekarabi SPH, Yesedi P, et al. 2019b. Dietary application of the probiotic Lactobacillus plantarum 426951 enhances immune status and growth of rainbow trout (Oncorhynchus mykiss) vaccinated against Yersinia ruckeri. Probiotics Antimicrob. Proteins 11: 207-219.

90. Panigrahi A, Kiron V, Satoh S, Watanabe T. 2010. Probiotic bacteria Lactobacillus rhamnosus influences the blood profile in rainbow trout Oncorhynchus mykiss (Walbaum). Fish. Physiol. Biochem. 36: 969-977.
92. Mohammadian T, Ghanie-Motlagh R, Jalal M, Nasipour M, Mohtashamipour H, Osroueh E, et al. 2022. Protective effects of non-encapsulated and microencapsulated Lactobacillus delbrueckii subsp. bulgaricus in rainbow trout (Oncorhynchus mykiss) exposed to lead (Pb) via diet. Ann. Anim. Sci. 22: 325-348.

93. Mohammadian T, Nasipour M, Tabande MR, Heidary AA, Ghanie-Motlagh R, Hosseini SS. 2019. Administrations of autochthonous probiotics altered juvenile rainbow trout Oncorhynchus mykiss health status, growth performance and resistance to Lactococcus garvieae, an experimental infection. Fish Shellfish Immunol. 86: 269-279.

94. Pérez-Sánchez T, Balciar J, Merrifield DL, Carravalho J, Gasicchini D, de Blas I, et al. 2011. Expression of immune-related genes in rainbow trout (Oncorhynchus mykiss) induced by probiotic bacteria during Lactococcus garvieae infection. Fish Shellfish Immunol. 31: 196-201.

95. Ramos M, Gonçalves J, Batista S, Costas B, Pires M, Rema P, et al. 2015. Growth, immune responses and intestinal morphology of rainbow trout (Oncorhynchus mykiss) supplemented with commercial probiotics. Fish Shellfish Immunol. 45: 19-26.

96. Park Y, Lee E, Hong J, Kim D, Moniruzzaman M, Bai SC. 2017. Use of probiotics to enhance growth, stimulate immunity and confer disease resistance to Aeromonas salmonicida in rainbow trout (Oncorhynchus mykiss). Aquac. Res. 48: 2672-2682.

97. Merrifield D, Bradley G, Baker R, Davies S. 2010b. Probiotic applications for rainbow trout (Oncorhynchus mykiss) Balwaum II. Effects on growth performance, feed utilization, intestinal microbiota and related health criteria postantibiotic treatment. Aquac. Nutr. 16: 496-503.

98. Sharifuzzaman S, Austin B. 2010. Development of protection in rainbow trout (Oncorhynchus mykiss, Balwaum) to Vibrio anguillarum following use of the probiotic Kocuria SM1. Fish Shellfish Immunol. 29: 212-216.

99. Capkin E, Altinok I. 2009. Effects of dietary probiotic supplementations on prevention/treatment of yersiniosis disease. J. Appl. Microbiol. 106: 1147-1153.

100. Safari R, Adel M, Lazado CC, Caipang CMA, Dadar M. 2016. Host-derived probiotics Enterococcus casseliflavus improves resistance against Streptococcus iniae infection in rainbow trout (Oncorhynchus mykiss) via immunomodulation. Fish Shellfish Immunol. 52: 198-205.

101. Sahandi J, Jafaryan H, Soltani M, Ebrahim P. 2019. The use of two Bifidobacterium strains enhanced growth performance and nutrient utilization of Rainbow Trout (Oncorhynchus mykiss) fry: Probiotics Antiinicrob. Proteins 11: 966-972.

102. Adel M, Lazado CC, Safari R, Yegani S, Zarriezhbrah M. 2017. Aqualase, a yeast-based in-feed probiotic, modulates intestinal microbiota, immunity and growth of rainbow trout Oncorhynchus mykiss. Aquaculture 478: 329-339.

103. Kurdomanov A, Sirakov I, Sotyanaova S, Yelichkova K, Nedeva I, Staykov Y. 2019. The effect of diet supplemented with Proviton on growth, blood biochemical parameters and meat quality in rainbow trout (Oncorhynchus mykiss) cultivated in recirculation system. Aquaculture, Aquar. Conserv. Legislation 12: 404-412.

104. Zhang C, Zhang J, Fan W, Huang M, Liu M. 2019. Effects of dietary Lactobacillus delbrueckii on growth performance, body composition, digestive and absorptive capacity, and gene expression of common carp (Cyprinus carpio) Huanghe var. Nutr. Aquac. 25: 166-175.

105. Zhang C-N, Zhang J-L, Guan W-C, Zhang X-F, Guan S-H, Zeng Q-H, et al. 2017. Effects of Lactobacillus delbrueckii on immune response, disease resistance against Aeromonas hydrophila, antioxidant capability and growth performance of Cyprinus carpio Huanghe var. Fish Shellfish Immunol. 68: 84-91.

106. Yanhur U, Caesar NR, Junirahma NS, Soelystioldy RN. 2022. Immunomolecular response of CD4+, CD4+, TNF-a and IFN-γ in Myxobolus-infected koi (Cyprinus carpio) treated with probiotics. Aquac. Fish. doi.org/10.1016/j.aaf.2022.01.004.

107. Valiaiihh J, Pourzaabadi M, Jalalzadadeh E, Bucio A. 2018. Use of Lactobacillus for improved growth and enhanced biochemical, hematological, and digestive enzyme activity in common carp at Mazandaran, Iran. North Am. J. Aquac. 80: 206-215.

108. Kaurz B, Malaczrewska J, Kaurz K, Zylisius-Urban J, Siwicki AK. 2018. Immune-enhancing activity of potential probiotic strains of Lactobacillus plantarum in the common carp (Cyprinus carpio) fingerling. J. Vet. Res. 62: 485.

109. Xu Y, Wang Y, Lin J. 2014. Use of Bacillus coagulans as a dietary probiotic for the common carp, Cyprinus carpio. J. World Aquac. Soc. 45: 403-411.

110. Gupta A, Gupta P, Dhawan A. 2014. Dietary supplementation of probiotics affects growth, immune response and disease resistance of Cyprinus carpio fry. Fish Shellfish Immunol. 41: 113-119.

111. Rostika R, Azhina ME, Ilcan YN, Andriani Y, Suryardi IB, Dewanti LP. 2020. The use of solid probiotics in feed to growth and survival rate of montap common carp (Cyprinus carpio). Aquac. Aquar. Conserv. Legis. 13: 199-206.

112. Hoseiniar SH, Hosseini M, Paknejad H, Safari R, Jafar A, Yousefi M, et al. 2019. Enhanced mucosal immune responses, immune related genes and growth performance in common carp (Cyprinus carpio) juveniles fed dietary Lactobacillus acidocaldarius MA185M and raffinose. Dev. Comp. Immunol. 94: 59-65.

113. Ahmadzad A, Sadegh TH, Dawood MA, Dadar M, Sheikhzadeh N. 2020. The effects of dietary pediococcus pentosaceus on growth, hematomo-immunological parameters and digestive enzyme activities of common carp (Cyprinus carpio). Aquaculture 516: 734656.

114. Aadjiri A, Ghafarifarsani H, Hoseiniar SH, Javahery S, Narmanizad E, Ghatpyakh R, et al. 2012. Effects of dietary supplementation of primalac, insulin, and biomin im on growth performance, antioxidant, and innate immunity of common carp (Cyprinus carpio). Aquaculture. Aquar. Nutr. doi.org/10.1111/j.2071-5214.2009.00074.x.

115. Mehrabi F, Khalesi M, Hazaae K. 2018. Effects of pre-and probiotics on growth, survival, body composition, and hematology of common carp (Cyprinus carpio L.) fry from the Caspian Sea. Turkish J. Fish. Aquat. Sci. 18: 597-602.

116. Lee JM, Kang WJ, Hanas MT, Lee J-B, Kim KW, Lim SG, et al. 2019. Characterization of a Bacillus sp. isolated from fermented food and its synbiotic effect with barley β-glucan as a biocontrol agent in the aquaculture industry. Appl. Microbiol. Biotechnol. 103: 1429-1439.

117. Kim D, Beck BR, Lee SM, Jeon J, Lee DW, Lee J, et al. 2016. Pellet feed adsorbed with the recombiant Lactococcus lactis BFE920 expressing SiMA antigen induced strong recall vaccine effects against Streptococcus iniae infection in olive flounder (Paralichthys olivaceus). Fish Shellfish Immunol. 55: 374-383.

118. Beck BR, Lee SM, Kim D, Park JH, Lee JK, Koon S-S, et al. 2017. A Lactococcus lactis BFE920 feed vaccine expressing a fusion protein composed of the OmpA and FlgD antigens from Edwardsiella tarda was significantly better at protecting olive flounder (Paralichthys olivaceus) from edwardsiellosis than single antigen vaccines. Fish Shellfish Immunol. 68: 19-28.

119. Harikrishnan R, Kim M-C, Kim J-S, Balasundaram C, Heo M-S. 2011a. Probiotics and herbal mixtures enhance the growth, blood constituents, and nonspecific immune response in Paralichthys olivaceus against Streptococcus panusaberi. Fish Shellfish Immunol. 31: 310-317.

120. Harikrishnan R, Kim M-C, Kim J-S, Balasundaram C, Heo M-S. 2011b. Immunomodulatory effect of probiotics enriched diets on Uronema marinum infected olive flounder. Fish Shellfish Immunol. 30: 964-971.

121. Kim J, Lee KW, Jeong HS, Ansay-M WR, Kim HS, Kim T, et al. 2019. Oral administration effect of yacon, ginger and blueberry on the growth, body composition and plasma chemistry of juvenile olive flounder (Paralichthys olivaceus) and immunity test against Streptococcus iniae compared to a commercial probiotic, Lactobacillus fermentum. Aquac. Rep. 15: 100122.

122. Jang WJ, Choi S-Y, Lee JM, Lee GH, Hanas MT, Kang J-S. 2019b. Viability of Lactobacillus plantarum encapsulated with poly-γ-glutamic acid produced by Bacillus sp. s10 during freeze-drying and in an in vitro gastrointestinal model. LWT 112: 108222.
123. Jang WJ, Lee JM, Kim Y-R, Hasan MT, Kong I-S. 2018. Complete genome sequence of Bacillus sp. Sf-10 (KCCM 90078) producing 400-kDa poly-γ-glutamic acid. Curr. Microbiol. 75: 1378-1383.

124. Jang WJ, Lee JM, Hasan MT, Kong I-S. 2019c. Fusion of the N-terminal domain of Pseudomonas sp. phytase with Bacillus sp. phytase and its effects on optimal temperature and catalytic efficiency. Enzyme Microb. Technol. 126: 69-76.

125. Hasan MT, Jang WJ, Lee B-J, Kim KW, Hur SW, Lim SG, et al. 2019b. Heat-killed Bacillus sp. Sf-10 probiotic acts as a growth and humoral innate immunity response enhancer in olive flounder (Paralichthys olivaceus). Fish Shellfish Immunol. 88: 424-431.

126. Niu K-M, Khorasvi S, Kothari D, Lee W-D, Lim J-M, Lee B-J, et al. 2019. Effects of dietary multi-strain probiotics supplementation in a low fishmeal diet on growth performance, nutrient utilization, proximate composition, immune parameters, and gut microbiota of juvenile olive flounder (Paralichthys olivaceus). Fish Shellfish Immunol. 93: 258-268.

127. FAO. 2020. Sustainability in action. State of World Fisheries and Aquaculture. Rome 200.

128. Qin L, Xiong F, Wang G, Zou H, Li W, et al. 2020. Effects of Bacillus licheniformis on the growth, antioxidant capacity, intestinal barrier and disease resistance of grass carp (Ctenopharyngodon idella). Fish Shellfish Immunol. 97: 344-350.

129. Tang Y, Han L, Chen X, Xie M, Kong W, Wu Z. 2019. Dietary supplementation of probiotic Bacillus subtilis affects antioxidant defenses and immune response in grass carp under Aeromonas hydrophila challenge. Probiotics Antimicrob. Proteins 11: 545-558.

130. Guo D, Xie M, Xiao H, Xu L, Zhang S, Chen X, et al. 2022. Bacillus subtilis in a high-fat diet modulates the gut microbiota and ameliorates hepatic lipid accumulation in grass carp (Ctenopharyngodon idella). Fishes 7: 94.

131. Li Y, Luo L, Yang Q, Zhang W, Tang X, Yu X, et al. 2022. Bacillus methylophilicus WM-1 enhances the immunity of grass carp against Aeromonas hydrophila. Aquac. Res. 53: 2464-2471.

132. Li Y, Hu S, Gong L, Pan L, Li D, Cao L, et al. 2020. Isolating a new Streptomyces amurensis N1-32 against fish pathogens and determining its effects on disease resistance of grass carp. Fish Shellfish Immunol. 98: 632-640.

133. Wu Z-Q, Jiang C, Wang Y, Peng L, Li W. 2017. Nitrogen removal characteristics of Shewanella xiamenensis A-1, Aeromonas veronii A-7, and Bacillus subtilis, single or combined, on the grass carp (Ctenopharyngodon idella) intestinal microbiota. Probiotics Antimicrob. Proteins 8: 386-396.

134. Fu L, Yang Z, Wang Y, Peng L, Li W. 2017. Nitrogen removal characteristics of Pseudomonas stutzeri F11 and its application in grass carp culture. Fish. Sci. 83: 89-98.

135. Liang Q, Zhang X, Lee KH, Wang Y, Yu K, Shen W, et al. 2015. Nitrogen removal and water microbiota in grass carp culture following supplementation with Bacillus licheniformis BSK-4. World J. Microbiol. Biotechnol. 31: 1711-1718.

136. Deng B, Fu L, Zhang X, Zheng J, Peng L, Sun J, et al. 2014. The denitrification characteristics of Pseudomonas stutzeri SC221-M and its application in water quality control in grass carp aquaculture. PLoS One 9:e114886.

137. Amir I, Zuberi A, Imran M, Ullah S. 2018. Evaluation of yeast and bacterial based probiotics for early rearing of Labeo rohita (Hamilton, 1822). Aquac. Res. 49: 3856-3863.

138. Hussain S, Alfar M, Salim M, Javid A, Khichi T, Hussain M, et al. 2011. Apparent digestibility of fish meal, blood meal and meat meal for Labeo rohita fingerlings. J. Anim. Plant Sci. 21: 807-811.

139. Ibrar M, Zuberi A, Amir I, Imran M, Noor Z. 2017. Effect of probiotic Geotrichum candidum on early rearing of Labeo rohita (Hamilton, 1822). Turkish J. Fish. Aquat. Sci. 17: 1263-1270.

140. Gin S, Sukumaran V, Sen S, Jena P. 2014. Effects of dietary supplementation of potential probiotic Bacillus subtilis VSG 1 singularly or in combination with Lactobacillus plantarum VSG 3 or Pseudomonas aeruginosa VSG 2 on the growth, immunity and disease resistance of Labeo rohita. Aquac. Nutr. 20: 163-171.

141. Mohapatra S, Chakraborty T, Prusty AK, PanirPrasad K, Mohanta KN. 2014. Beneficial effects of dietary probiotics mixture on hema-to-immuno and cellular immunity and cell apoptosis of Labeo rohita fingerlings reared at higher water temperatures. PLoS One 9:e100929.

142. Mukherjee A, Chandra G, Ghosh K. 2019. Single or conjoint application of autolchthonous Bacillus strains as potential probiotics: Effects on growth, feed utilization, immunity and disease resistance in Rohu, Labeo rohita (Hamilton). Aquaculture 512: 734302.

143. Pandey A, Baranerje G, Dan SK, Ghosh K, Ray AK. 2018. Evaluation of in vivo probiotic efficiency of Bacillus amyloliquefaciens in Labeo rohita challenged by pathogenic strain of Aeromonas hydrophila MTCC 1739. Probiotics Antimicrob. Proteins 10: 391-398.

144. Khan MIR, Kamila, Choudhury TG, Rathore G. 2022. Dietary administration of a host-gut derived probiotic Bacillus amyloliquefaciens COFCAU_P1 modulates immune-biochemical response, immune-related gene expression, and resistance of Labeo rohita to Aeromonas hydrophila infection. Aquaculture 546: 737390.

145. Ghori I, Tabassum M, Ahmad T, Zuberi A, Imran M. 2018. Geotrichum candidum enhanced the Enterococcus faecium impact in improving physiology, and health of Labeo rohita (Hamilton, 1822) by modulating gut microbiome under memic aquaculture conditions. Turkish J. Fish. Aquat. Sci. 18: 1255-1267.

146. Bandyopadhyay P, Mishra S, Sarkar B, Swain SK, Pal A, Tripathy PP, et al. 2015. Dietary Saccharomyces cerevisiae boosts growth and immunity of IMC Labeo rohita (Ham.) juveniles. Indian J. Microbiol. 55: 81-87.

147. Brown C, Power D, Nézard J. 2010. Disorders of development in fish. Fish diseases and disorders, Volume 2. pp. 166-181.

148. Padeniya U, Larson ET, Septiani S, Pataueg A, Kafui AR, Hasan E, et al. 2022. Probiotic treatment enhances pre-feeding larval development and early survival in zebrasfish Danio rerio. J. Aquat. Anim. Health 34: 3-11.

149. Ozorio RO, Kopecka-Filarczyk, J, Peixoto MJ, Lochmann R, Santos RJ, Santos G, et al. 2016. Dietary probiotic supplementation in juvenile rainbow trout (Oncorhyncus mykiss) reared under cage culture production: effects on growth, fish welfare, flesh quality and intestinal microbiota. Aquac. Res. 47: 2732-2747.

150. Bishu A, Singh UP, Pandey N. 2012. Bacillus subtilis as a potent probiotic for enhancing growth in fingerlings of common carp (Cyprinus carpio Linnaeus). Ind. J. Fish. 59: 103-107.

151. Bishu A, Beck BR, Heo S-B, Kim J, Kim HD, Lee S-M, et al. 2013. Lactococcus lactis BFE920 activates the innate immune system of olive flounder (Paralichthys olivaceus), resulting in protection against Streptococcus iniae infection and enhancing feed efficiency and weight gain in large-scale field studies. Fish Shellfish Immunol. 35: 1585-1590.

152. Li Z, Chen Y, Zhang J, Zhu X, Zhang J, Chen D, et al. 2017. Effects of dietary Bacillus natto supplementation on growth performance and the growth-related genes/micro RNA expression in the skeletal muscle of grass carp (Ctenopharyngodon idella). Aquac. Nutr. 23: 46-53.

153. Amir I, Zuberi A, Kamran M, Imran M. 2019. Evaluation of commercial application of dietary encapsulated probiotic (Geotrichum candidum QAUC011). Effect on growth and immunological indices of Rohu (Labeo rohita, Hamilton 1822) in semi-intensive culture system. Fish Shellfish Immunol. 95: 464-472.

154. Jha DK, Bhujel RC, Anal AK. 2015. Dietary supplementation of probiotics improves survival and growth of Rohu (Labeo rohita) hatchlings and fry in outdoor tanks. Aquaculture 435: 475-479.