



REVIEW ARTICLE

## Probiotics in aquaculture: Importance, influence and future perspectives

Janardana Reddy S.\*

Department of Fishery Science and Aquaculture, Sri Venkateswara University, Tirupati-517 502, Andhra Pradesh, India.

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**Abstract:** Probiotics have been widely used in livestock rearing, have also recently been applied to aquaculture to elevate the production. Probiotics are defined as live cells or a substrate that provides benefits through stimulation of growth, improved digestion, improved immune response and ingested with the aim of promoting good health. Probiotics can also improve water quality and pond management. This review summarizes the current understanding the use of probiotics in aquaculture, including the mechanism of probiotics, and describes their application, and prospects and difficulties associated with their use in aquaculture. This review includes general knowledge of probiotics from previous studies and evaluates the efficacy of probiotics in aquaculture. Research in probiotics for aquaculture such as finfish and shell fish culture is at an early stage of development and much work is still needed.

**Key Words:** Probiotics; Immune system; Antibacterial activity; Growth Promoters.

### INTRODUCTION

It is obvious that the aquaculture is now being the fastest growing protein food-producing industrial sector in the world, is moving in to new directions, intensifying and diversifying [1]. In India fisheries is an important economic and commercial activity and a flourishing sector with varied resources and potentials. Only after the Independence, fisheries sector together with agriculture sector has been recognized as a commercially important sector. The vibrancy of the sector can be visualized by the 11-fold increase that India achieved in fish production in just six decades, i.e. from 0.75 million tons in 1950-51 to 9.6 million tons during 2012-13. This brought about by in an unparalleled average annual growth rate of over 4.5 percent over the years which have placed the country on the forefront of global fish production, only after China. Besides meeting the domestic needs, the dependence of over 15.5 million people on fisheries culture activities for their livelihood and foreign exchange earnings to the tune of US\$ 3.51 billion from fish and fisheries products, properly justifies the importance of the sector on the country's economy and in livelihood security. India is also an important country that produces fish through aquaculture in the world [2]. With the increase in the commercialization of aquaculture production come many challenges, such as combating diseases and epizootics, brood stock improvement and domestication, development of appropriate feedstuffs and feeding mechanisms, hatchery and grow-out technology, as well as water-quality management [3]. Of these, disease ebullitions are one of the important problems that affect aquaculture production, suppressing both economic and social development in many countries. Moreover, the availability of feed for aquaculture is another significant challenge in the intensifying aquaculture industry, as feed accounts

for up to 70% of operating costs for most aquaculture species [4]. Feed quality and feeding methods therefore requirement to be thoroughly considered in order to improve growth performance and feed efficiency of the cultured animals. Several previous reports have suggested that probiotic supplementation can be reduced disease outbreaks by enhancing the immune system of fish and shrimp [5] and can reduce culture costs by improving the growth and feed efficiency of fish. In addition, by improving animal physiology, the application of probiotics can lead to an improvement in water quality, as better feed efficiency may result in fish producing less waste [6-8].

### Probiotics-Origin, development and definition

Elie Metchnikoff has worked at the beginning of this century on probiotics [9], described them as "microbes ingested with the aim of promoting good health". This definition was modified to "organisms and substances which contribute to intestinal microbial balance" [10] and later by Fuller [11] to "a live microbial feed supplement which beneficially affects the host animal by improving its intestinal microbial balance". These definitions are basically applied to farm animals such as ruminants, poultry and pigs and also to humans.

Probiotics, recently, have also being used in aquaculture and therefore, the definition may also be redesigned. In aquatic animals, not only the digestive tract is important but also the surrounding water. Gatesoupe [12] defined probiotics as "microbial cells that are administered in such a way as to enter the gastrointestinal tract and to be kept alive, with the aim of improving health. Gram *et al.*, [13] have given a broadened definition by removing the restriction to the improvement to the intestine: "a live microbial supplement which beneficially

### \*Corresponding Author:

**Dr. S. Janardana Reddy,**  
Head & Assistant Professor,  
Department of Fishery Science and Aquaculture,  
Sri Venkateswara University,  
Tirupati-517 502, Andhra Pradesh, India.



affects the host animal by improving its microbial balance”.

The term ‘Probiotics’ is generally used to indicate bacteria that promote the health of other organisms. Lilley and Stillwell [14] described bacteria as substances secreted by one microorganism, which stimulated the growth of another one. An expert with the Joint Food and Agriculture Organization of the United Nations/World Health Organization (FAO/WHO), stated that probiotics are live microorganisms, which when consumed in adequate amounts, confer a health benefit for the host [15].

In general, probiotic bacterial strains have been isolated from indigenous and exogenous microbiota of aquatic animals. Gram-negative facultative anaerobic bacteria such as *Vibrio* and *Pseudomonas* constitute the predominant indigenous microbiota of a variety of species of marine fish [16], whereas saltwater fish, the indigenous microbiota of freshwater fish species tends to be dominated by members of the genera *Aeromonas*, *Plesiomonas*, representatives of the family Enterobacteriaceae, and obligate anaerobic bacteria of the genera *Bacteroides*, *Fusobacterium*, and *Eubacterium* [17]. Lactic acid producing bacteria, prevalent in the mammal or bird gut such as *Bifido bacterium* in human, *Lactobacillus* in swine, rodent and bird, *Enterococcus* in carnivore, are generally sub-dominant in fishes and represented essentially by the genus *Carnobacterium* [18].

The term *Probiotic* means “for life” and originated from two Greek words “Pro” and “Bios” [19]. It was originally used by Lilley and Stillwell [20] and described as “one of the substances produced by protozoans that stimulates other microorganisms, and it was later used to describe animal feed supplements that benefit the host animal” Fuller [11] revised the definition to “a live microbial feed supplement which beneficially affects the host animal by improving its intestinal microbial balance”. This definition highlighted the essential component of probiotics as being live cells and not only “substances”. Similar definitions have also used in aquaculture indicate that a probiotic is a live microbial food supplement that confers health benefits or disease resistance to the host [21].

The concept of aquatic Probiotics is a relatively new one, and methods for evaluating the efficacy of probiotics are needed. Fuller [11] proposed that a good probiotic should be:

- 1) Effectiveness in application
- 2) Non-pathogenic and non-toxic
- 3) Existing as viable cells, preferably in large numbers surviving and being actively involved in the metabolism of the gut environment being stabilized and remaining viable during long periods of storage and under field conditions.

FAO/WHO defined the Probiotics as “live microorganisms which when administered in adequate amounts confer a health benefit on the host” [22]. The idea of using fermented foods for some health benefits is not a new method, being mentioned in the Persian version of the Old Testament (Genesis 18:8) that “Abraham attributed his longevity to the consumption of sour milk”. Later, in 76 BC, a Roman historian, Pliny, recommended the use of fermented milk products for the treatment of gastroenteritis cases [23]. However, a scientific approach, recognizing the beneficial role of certain microorganisms was applied only in the first decades of the 20th century, with the suggestion of using *Lactobacillus*. Elie Metchnikoff [24] has attributed the longevity of Bulgarian populations to yoghurt consumption; *Bifidobacterium*. Henri Tissie in 1906, observed a greater presence of *Bifidobacteria* in the faeces of breastfed healthy children; and *Saccharomyces boulardii*. Henri Boulard emphasized to the use of this yeast to treat diarrhea of local populations in the East [25]. Several clinical studies have shown the benefits of probiotics to human health such as in diarrhea treatment [26]; in cancer treatment [27]; in lactose intolerance [28]; in allergies treatment [29] and like others.

The use of growth promoters improving zoo technical performance of animals. Initially a large variety of substances with antibiotic function was used to improve performance of poultry, pigs and cattle, especially penicillin and tetracycline. The antibiotics as supplemented to feeds showed great benefits to animal husbandry, expressed primarily in improved weight gain and feed conversion. Antibiotics were used for decades, but are being dissipated from the zoo technical activity, mainly due to the risks posed by antibiotic-resistant bacteria, which can result in problems for animal and human health.

In according to probiotics be good enough for attention from researchers seeking alternatives to the use of normal growth promoters in the field of animal nutrition. Probiotics have also received special attention from researchers seeking animal nutrition alternatives to the use of growth promoters. Therefore, the use of probiotics is being increasingly seen as an alternative to the use of antibiotics in animal production.

Many scientific workers reported that the probiotics showing beneficial effects on supplementation with probiotic strains in diets for poultry, pigs, cattle, fish, crustaceans, mollusks and amphibians [30-32]. Probiotics have been supplemented through diet in order to appraise the balance of the intestinal flora of animals, control digestive tract diseases, provoking the digestibility of feed, resulting to increased use of nutrients and inspiring better zoo technical performance of animals [33, 34].

**Probiotic Organisms-Characteristics**

The essentials that a probiotic organism must meet are [35]:

- I. Resistance to the acid stomach environment, bile and pancreatic enzymes
- II. to the cells of the intestinal mucosa;
- III. Capacity for colonization
- IV. Staying alive for a long period of time, during the transport, storage, so that they can colonize the host efficiently;
- V. Production of antimicrobial substances against the pathogenic bacteria and
- VI. Absence of translocation

The species normally used as probiotics in animal nutrition are usually non-pathogenic normal microflora, such as lactic-acid bacteria (Bifidobacterium, Lactobacillus, Lactococcus, Streptococcus and Enterococcus) and yeasts as Saccharomyces spp.

**Table 1:** Microorganisms used as Probiotics in Aquaculture.

Saccharomyces	<i>S. cremoris</i> , <i>S. faecium</i> , <i>S. lactis</i> , <i>S. intermedius</i> , <i>S. cerevisiae</i> , <i>S. boulardii</i>
Streptococcus	<i>S. diacetylatis</i> , <i>Thermophyllus</i> ,
Bifidobacterium	<i>B. animalis</i> , <i>B. bifidum</i> , <i>B. longum</i> , <i>B. thermophilum</i>
Aspergillus	<i>A.niger</i> , <i>A. orizae</i>
Pediococcus	<i>P. acidilacticii</i> , <i>P. cerevisiae</i> , <i>P. pentosaceus</i> ,
Bacillus	<i>B.coagulans</i> , <i>B. lentus</i> , <i>B. licheniformis</i> , <i>B. subtilis</i> <i>L. acidophilus</i> , <i>L. brevis</i> , <i>L. bulgaricus</i> , <i>L. casei</i> , <i>L. fermentarum</i> , <i>L. curvatus</i> , <i>L. lactis</i> , <i>Plantarum</i> , <i>L. reuterii</i> , <i>L. delbruekii</i> ,
Lactobacillus	

**Probiotics - Mechanisms of Action**

The mechanisms of action of bacteria used as probiotics, although not yet fully elucidated, described as:

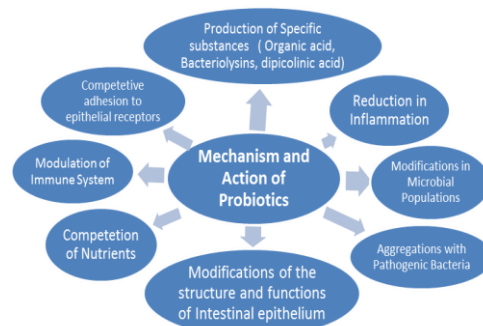
- a) **Competition for binding sites:** also known as "competitive exclusion", where probiotics bacteria bind with the binding sites in the intestinal mucosa, forming a physical barrier, preventing the connection by pathogenic bacteria
- b) **Production of antibacterial substances:** probiotic bacteria synthesize compounds like hydrogen peroxide and bacteriocins, having antibacterial action, mainly in relation to pathogenic bacteria. They also produce organic acids that lower the environment's pH of the gastrointestinal tract, preventing the growth of various pathogens and development of certain species of Lactobacillus,
- c) **Competition for nutrients:** the lack of nutrients available that may be used by pathogenic

bacteria is a limiting factor for their maintenance,

- d) **Stimulation of immune system:** some probiotics bacteria are directly linked to the stimulation of the immune response, by increasing the production of antibodies, activation of macrophages, T-cell proliferation and production of interferon.

**Probiotics - Modes of Actions**

- 1) Probiotics might be able to harmonize the host's gut defences including the innate as well as the acquired immune system and this mode of action is highly important for the prevention and therapy of infectious diseases but also for the treatment of inflammation of the digestive tract.
- 2) Probiotics can also influence on other microorganisms, commensals and pathogenic organisms. This principle is more important for the prevention and therapy of infections and restoration of the microbial equilibrium in the gut of organisms.
- 3) Probiotics effect may also be based on actions affecting microbial products, host products and food ingredients and these actions may result in inactivation of toxins and detoxification of host and food components in the gut.

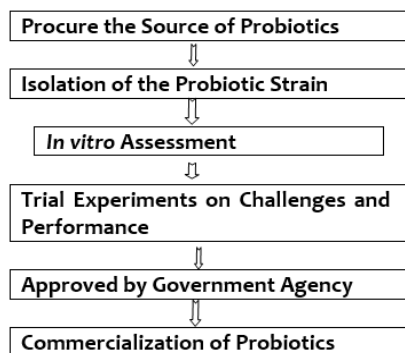


**Figure 1:** Mechanisms of Action of Probiotics

**Probiotics- Selection**

Concisely, for the use of a microorganism as probiotic, it is necessary its isolation, characterization and testing authenticating probiotic efficiency (Figure -2). A customary way to select probiotics is to perform *in vitro* antagonism tests, in which pathogens are exposed to the candidate probiotics or their extracellular products in a liquid [37] or solid [38] medium. However, Gram *et al.*, [39] suggested that *in vitro* activity in well-diffusion assays and broth cultures cannot be used to predict a possible *in vivo* effect. For example, *in vitro* antagonism of *Pseudomonas fluorescens* (AH2 strain) against *Aeromonas salmonicida* does not confer Atlantic salmon protection against furunculosis, but is an effective probiotic in rainbow trout, conferring protection against vibriosis [39].

Therefore, it is highly needed to know the origin of strains isolated from the host, safety (non-pathogenic) and ability of strains to survive to the transit through the gastrointestinal tract of the host for example: resistance to low pH, proteases etc. The potentiality of microorganisms to colonize is contemplated as one of the main selection criteria for potential probiotics that means, the efficient adherence to intestinal epithelial cells is to reduce or prevent colonization of pathogens [37]. Therefore the, potential probiotics must deploy its beneficial effects such as increased immune response in the host. Finally, the probiotic must be tenable under normal storage conditions and also technologically suitable for industrial processes. The results of probiotics which are presented significantly and satisfactory can be manufactured commercially for human consumption and used in aquaculture.



**Figure 2:** The methods to selection of Probiotic bacteria [36]

**The methods to select probiotic bacteria for use in aquaculture: (Fig. 2).**

- (i) Collect the background source information
- (ii) Acquisition of potential probiotics
- (iii) Evaluate the ability of potential probiotics to out-compete pathogenic strains
- (iv) Assessment of the pathogenicity of the potential probiotics
- (v) assessment of the effect of the potential probiotics in the host economic cost-benefit analysis [40]

**The methods to provide host or supplemented to aquatic environment**

- (i) dietary supplemented through live food
- (ii) bathing treatment
- (iii) addition to culture water
- (iv) supplemented to artificial diet [41, 42]

**Uses of Supplemented Probiotics to Aquaculture Pond**

The microorganisms which are in the aquaculture pond are in direct contact with the animals, with the gills and with the food supplied, having easy access to the digestive tract of the animal. The microorganisms are potentially pathogenic, which are opportunistic, cause infections, worsening in zoo technical performance and even death. For this reason, the supplementation of probiotics for

aquatic organisms targeted to direct benefit to the animal, but also their effect on the farming environment. Bergh *et al.*, [43] reported that, when starting its first feeding, the intestinal flora of the Atlantic halibut changed from a prevalence of *Flavobacterium* species to *Aeromonas* species and *Vibrio* species manufacturing the influence of the external environment and food on the microbial community of fish.

It is identified that *Vibrio* spp., *Plesiomonas shigelloides*, and *Aeromonas* species are the main causative agents of diseases in aquaculture, and may even cause food infections in humans. The microorganisms in the water influence the biota of host's intestine and vice versa. Makridis *et al.*, [44] demonstrated that the supplementation of two strains of bacteria through the food directly into the farming water of the incubators of turbot larvae (*Scophthalmus maximus*) promoted the maintenance of probiotic bacteria, as well as the colonization of the digestive tract of the larvae.

Probiotics most used in aquaculture are those belonging to the genus *Bifidobacterium* species (*B. thermophilum*, *B. lactis* and *B. bifidum*), *Bacillus* species (*B. subtilis*, *B. licheniformis* and *B. circulans*), lactic-acid bacteria and yeast [45].

The benefits of supplementation of Probiotics to the in-aquaculture pond [46, 47]:

1. Improvement of the nutritional value of food
2. Enzymatic contribution to digestion
3. Inhibition of pathogens
4. Growth promoting factors
5. Improvement in immune response and
6. Farming water quality

The most recent studies manifested the use of probiotics is very high in the culture of various aquatic organisms such as mollusks, frogs, shrimps fish and animal husbandry and also denote that their working performance is highly potential [12, 33, 48, 49].

**Probiotic Strains Used In Aquaculture**

Most of the probiotics using as biological control agents in aquaculture belong to the lactic acid bacteria (*Lactobacillus* and *Carnobacterium*), to the genus *Vibrio* (*V. alginolyticus*), to the genus *Bacillus*, or to the genus *Pseudomonas*, and the genera or species (*Aeromonas* and *Flavobacterium*) (Table 2).

It is highly requisite to understand the mechanisms of action in order to define selection criteria for perspective potential probiotics. Therefore, more information on the host - microbe interactions *in vivo*, and development of monitoring molecular tools are highly mandatory for better understanding of the composition and functions of the primordial microbiota, and also microbial cultures of probiotics. The choice of using probiotics in aquaculture is a part of result of



historical and pragmatic use and not based on technical criteria. The use of probiotics is an essential management tool, but its efficacy depends on understanding the nature of competition between species or strains.

**Table 2:** Some of the Probiotics considered as biological control agents in aquaculture

Probiotic strain	Source of Probiotic	Used on host
<i>Carnobacterium divergens</i>	Intestines of Atlantic salmon	Atlantic cod fry
<i>Bacillus megaterium</i> , <i>B. subtilis</i> , <i>B. polymyxa</i> , <i>B. licheniformis</i>	Commercial product (Biostart)	Channel catfish
<i>Vibrio pelagius</i>	Turbot larvae Intestines of	Turbot
<i>Carnobacterium</i> sp.	Atlantic salmon	Atlantic salmon
<i>Lactobacillus rhamnosus</i> ATCC 53103	Culture collection	Rainbow trout
<i>Bacillus circulans</i>	Intestines of <i>Labeo rohita</i>	<i>L. rohita</i>
<i>Pseudomonas fluorescens</i>	Iced freshwater fish ( <i>Lates niloticus</i> )	Rainbow trout ( <i>Oncorhynchus mykiss</i> )
<i>Aeromonas hydrophila</i> , <i>Vibrio fluvialis</i> , <i>Carnobacterium</i> sp., <i>Micrococcus luteus</i>	Digestive tract of rainbow trout	Rainbow trout
<i>Vibrio hepatarius</i> , <i>Vibrio</i> sp., <i>Bacillus</i> sp.	<i>P. vannamei</i>	<i>P. vannamei</i>
<i>Vibrio</i> P62, <i>Vibrio</i> P63, <i>Bacillus</i> P64	<i>P. vannamei</i>	<i>P. vannamei</i>
<i>Saccharomyces cerevisiae</i> , <i>S. exiguus</i> , <i>Phaffia rhodozyma</i>	Commercial product	<i>Penaeus vannamei</i>
<i>Roseobacter</i> sp. BS107	Scallop larval cultures	<i>Pecten maximus</i>
<i>Lactobacillus casei</i> , <i>L. brevis</i> , <i>L. helveticus</i> , <i>Lactococcus lactis</i> spp., <i>Leuconostoc</i> , <i>Mesenteroides</i> spp. <i>mesenteroides</i> , <i>Pediococcus acidilactici</i>	Culture collection	<i>Artemia nauplii</i>
<i>Alteromonas haloplanktis</i>	Microalgal cultures	<i>Argopecten purpuratus</i>

### Probiotics - Growth Promoting Effects

Probiotics harmonize the growth of intestinal microbiota, vanquish potentially harmful bacteria and booster up the body's natural defense mechanisms, thus ameliorate resistance against infectious diseases. Bacterial probiotics do not have a mode of action but act on species specific or even strain-specific and immune responses of the animal, and their interaction with intestinal bacterial communities plays a key role. Probiotics bring out

inhibitory substances which might be antagonistic to the growth of pathogens in the intestine [50]. The potentiality of some probiotics is to glued to the intestinal mucous may blump the intestinal infection route common to many pathogens [12, 18]. They can also provoke the appetite and boost up nutrition by the production of vitamins, detoxification of compounds in the diet and disintegration of indigestible components [39, 41].

The primeaval microbiota are being conceded to have a prominent effect on the structure, function and metabolism of the digestive tract of aquatic animals, which are required to sustain the normal physiological functions of an organism, and serve as a source of nutrients, vitamins, enzymes, microbial disintegration of chitin, p-nitrophenyl-N-acetyl-beta-D-glucosamine cellulose and collagen [51]. In fact, it is not yet clear whether feed probiotics increased the appetite or nature itself boosted the digestibility, causing the improved appetite, the aqua scientists are inclined to think that it could be both factors, moreover, it would be more important to determine whether the supplemented probiotics actually flavor good to aquaculture species or blandness to them [51].

Numerous studies have indicated that the application of probiotics can improve the feed conversion, growth rate and weight gain of various fish species. Prevalent applications of the probiotics have also shown a promising improvement in the growth of shellfish. Probiotics are used to reduce the prevalence and grievousness of various diseases caused by *Vibrio alginolyticus* in fish and prawns [41] and that led to a significant improvement in the FCR, FER and PER of shrimp larvae fed with *L. plantarum* bio-encapsulated Artemia [53].

### Probiotics-Influence on Quality of Water in Aquaculture

Another vital aspect of the use of probiotics in aquaculture is improvement of the quality of water in the farming nurseries. Increases in organic load, levels of phosphorous and nitrogen compounds are growing concerns in aquaculture. Boyd and Gross [51] demonstrated that the beneficial effect of probiotics on organic matter decomposition and reduction of the levels of phosphate and nitrogen compounds. Aerobic denitrifying bacteria are considered good candidates to reduce nitrate or nitrite to N<sub>2</sub> in aquaculture pond waters.

Probiotic bacteria are also known to upgrade the water quality in many ways. Heterotrophic bacteria necessitating some organic sources of carbon besides the growth inorganic forms have a significant role in the decomposition of organic matter and production of particulate food materials from dissolved organics. The probiotics entertained a major role in maintaining optimum water quality indices especially dissolved oxygen, ammonia, nitrite, nitrate and phosphates throughout the

culture period. It is obvious from the bacterial load data that the Nitrosomonas and Nitrobacter species were hegemonized and suppressed the Pseudomonas species in the probiotic treated ponds when compared to the control pond [54]. Ameliorated water quality has especially been associated with Bacillus sp. The hypothesis is that gram positive bacteria are better converters of organic matter back to CO<sub>2</sub> that gram-negative bacteria. During the production cycle, high levels of gram-positive bacteria may minimize the buildup of dissolved and particulate organic carbon. It has been reported that use of Bacillus species boosted the water quality, survival and growth rates and increased the health status of juvenile *Penaeus monodon* and bring down pathogenic vibrios [55].

Some of the denitrifying bacteria, identified in aquaculture practices, are Arthrobacter, Acinetobacter, Bacillus, Cellulose microbium, Halomonas, Microbacterium, Paracoccus, Pseudomonas, Sphingobacterium and Stenotrophomas, influencing on crustaceans and finfishes and causing hazardous diseases. Reduction in levels of phosphorous and nitrogen compounds in the farming water of shrimp *Litopenaeus vannamei* was also perceived when commercial probiotics were put in to the water. Similarly, for the shrimp *Penaeus monodon*, an amelioration in the quality of farming water was observed with the supplementation of Bacillus spp. as probiotic [55, 56].

#### **Probiotics - Effect on Immune System of Fishes**

The non-specific immune system is also to be stimulated by probiotics. It has been demonstrated that oral administration of *Clostridium butyricum* bacteria to rainbow trout magnified the resistance of fish to vibriosis, by increasing the phagocytic activity of leucocytes [57]. Rengpipat et al., [58] alluded that the use of Bacillus sp. (strain S11) allowed the disease protection by activating both cellular and humoral immune defenses in tiger shrimp. The administration of mixture of bacterial strains (Bacillus and Vibrios sp.) positively influenced the growth and survival of juveniles of white shrimp and entrusted a protective effect against the pathogens *Vibrio harveyi* and white spot syndrome virus. This protection was due to a stimulation of the immune system, by intensifying phagocytosis and antibacterial activity. In addition, showed that administration of a lactic acid bacterium, *Lactobacillus rhamnosus* at a level of ~105 cfu g<sup>-1</sup> feed, invigorating the respiratory burst in rainbow trout [59]. The larvae of *Scophthalmus maximus* fed rotifera enriched with lactic-acid bacteria increased resistance against infection by *Vibrio* spp. The joint administration of *Lactobacillus fructivorans* and *Lactobacillus plantarum* via dry or live feed promoted the colonization of the intestine of sea bream larvae (*Sparus aurata*) and the decrease in mortality of animals during larviculture and nursery [12]. Gram and his associates [13]

showed that the use of *Pseudomonas fluorescens* AH2 as probiotics dwindled the temporality of juveniles of rainbow trout, *Oncorhynchus mykiss*, exposed to *Vibrio anguillarum*.

#### **Probiotics - Antibacterial Activity**

Probiotics ameliorate the intestinal microflora, which have antagonistic properties, because of the formation of organic acids and bacteriocins they transform the metabolism of microbiota to produce short-chain fatty acids, intensified sodium and water absorption, diminish colonic motility and support the host's good health, extending protection against infections by provoking the immune system, alleviating lactose intolerance, reducing blood cholesterol levels, and boosting weight gain and the feed conversion ratio. The proliferation site of fish pathogens and the mechanisms of antagonism by a probiotic culture influence the choice of a probiotic bacterium [60].

A Probiotic bacterium isolated from the gut is propitious if the targeted pathogenic bacteria infected through the gastrointestinal tract. However, some fish pathogens may escalate on the skin surface and probiotic bacteria attuned and habituated to the outer surfaces could limit pathogen proliferation. The probiotic cultures could also originate from the rearing fields, since Bacillus species usually dwell in the sediment from which shrimps feed, a Bacillus product available in the market was added to it and successfully impeded infection by pathogenic vibrios. It is obviously manifested the levels of *Vibrio vulnificus* in water were conquered by the presence of other bacteria. Therefore, it is conceivable that health-beneficial organisms in fish-rearing systems may be appeared in several other niches than the fish itself [61]. Lactic acid bacteria are normal flora in the GI tract of healthy mammals and aquatic animals; they have probiotic properties with no harmful effects, except in some reports from maricultures in Japan and North America. However, a novel Weissella species has been exemplified as an opportunistic pathogen for rainbow trout [62]. LABs are widely familiar for their ability to inhibit bacterial pathogens by the production of antimicrobial compounds: organic acids, hydrogen peroxide and ribosomally synthesized peptides referred to as bacteriocins [12]. Moreover, Lactobacilli may ferment lactose to lactic acid, thereby decreasing the pH to a level that harmful bacteria cannot be authorized. Hydrogen peroxide is also produced, that inhibits the growth of gram negative bacteria. It has also been proclaimed that lactic acid-producing bacteria of Streptococcus and Lactobacillus species produced antibiotics [21]. Antimicrobial activity against fish pathogens and in vitro safety of 99 LAB formerly isolated from fish, seafood and fish products have also been tested recently [63]. It is also found that *Leuconostoc mesenteroides* manifested more ability to inhibit the growth of fish pathogenic bacteria compared to others and determined that probiotic

bacteria can possibly be used in aquaculture [64]. Overall, the studies have shown that supplemented probiotics are good alternative to protect finfish and shellfish against pathogenic bacteria, namely against *Vibrio* sp. pathogens, the most important in the culture of finfish and shellfish. However, further studies are necessary to broad-ranging the probiotic candidates and the finfish and shellfish species prior they are applied to aquaculture from a practical point of view [65].

### CONCLUSIONS

Probiotics are habitually live microorganisms that administered at adequate doses confer health benefits to the host. In this review, we have fascinated only in those probiotics extending protection to shellfish and fish species important for the aquaculture against viral and bacterial diseases. The results manifested so far with the use of probiotics for aquatic organisms are promising. However, many works have not obtained satisfactory results. Sporadically, the in experiments aquatic organisms are challenged by some pathogenic agent, the probiotic organism does not show inhibiting action against the pathogen, resulting in mortality. Analogously, the conditions to which the animals are subjected during farming may directly influence the effectiveness of probiotics. Thus, when not submitted to stressful situations, the results often do not reveal any significant effect of probiotics on the achievement of animals. Usually, the effects of adding probiotics tend to be most striking in unsuitable operating conditions or in conditions of stress, when the microflora is unhinged, primarily in young animals.

Furthermore, studies have to be registered to increase knowledge on properly use of probiotics to control infections in shellfish and finfish but much more efforts are needed to control viral diseases for the aquaculture industry as it is interminable growing due to the fish and shellfish demand for human consume. Apart from the preparing of better probiotic formulations, improvement of their properties may be more required. So it is better to develop cheaper production methods, in various methods and combination with other therapeutic measures have to be encouraged.

It is essential to manifest the mechanisms of action in order to define selection criteria for potential probiotics. Therefore, more needful information on the host-microbe interactions in vivo, and development of monitoring molecular tools are highly required for better understanding of the composition and functions of the indigenous microbiota, as well as of microbial cultures of probiotics. The application of probiotics in aquaculture has been in large part a result of historical and empirical use but not on scientific criteria. The use of probiotics is an important management tool, but its working potentiality depends on manifesting the nature of competition

between and among species and strains. There is an increasing interest within the industry in the control or elimination of antimicrobial use. Therefore, an alternative method need to be developed to maintain a healthy microbial environment in the aquaculture field. One such method that is gaining adoptees within the industry is the use of probiotic bacteria to control potential pathogenic organisms.

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