



# Probiotics as alternative control measures in shrimp aquaculture: A review

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## ABSTRACT

Shrimp aquaculture industry is facing diseases problems, viral and bacterial pathogens that take advantage of the weak immunity of shrimp and an altered gut microbial community. Furthermore, probiotic practices in aquaculture started to be of importance for increased disease resistant, aquatic organisms growth, and feed efficiency. Later they are used to improve the water quality of bacterial infections. Today, there is an evidence that probiotics can improve the digestibility of nutrients, increase tolerance to stress, and encourage reproduction. They are substances containing live microorganisms secreted by microorganisms that stimulate the growth of other organisms. At presently, many commercial probiotic products made by bacterial species such as *Lactobacillus* sp., *Bacillus* sp., *Carnobacterium* sp., *Enterococcus* sp., and also from yeast, i.e., *Saccharomyces cerevisiae* are available. This review article gives a brief explanation on probiotics, their types, origin, mode of action, types of diseases they can control, ability to improve nutrients digestion, increase stress tolerance, and increase of reproduction.

## 1. INTRODUCTION

Aquaculture is the culture of organisms in controlled aquatic environments [1]. Fish aquaculture has remain an important financial source and has boost the economy of many developing countries and served as an important source of food, nutrition, income, and livelihoods for many people around the globe [1]. Due to rapid growth and advancement in aquaculture, the report by indicates that aquaculture is responsible for the provision of about 50% of all fish consumed by humans and the world per capita fish supply has reached a new record high of 20 kg in 2014 [1]. This is a clear demonstration of the contribution of marine environment and inland waters to food security as the world population is expected to rise to over 9 billion people in 2050.

Marine shrimp aquaculture has been the mainstay of aquaculture industries due to shrimp's richness in protein supply [1]. Shrimp aquaculture is a common practice in developing nations in Asia, Latin America and throughout the tropical world supporting non-urban communities with means of survival, and consequently reduced poverty [1]. It contributes to over 50% of global shrimp production as cached shrimp cannot serve the export demand for shrimp worldwide and is regarded as the most valued aquaculture business.

Black tiger shrimp (*Penaeus monodon*) is the most widely cultured species in many countries, but due to the emergence of viral pathogens, many farmers have switched to the imported Pacific whiteleg shrimp, *Litopenaeus vannamei* which has been genetically selected and has since 2003 become a domesticated stock [2,3]. Most developing countries have embraced shrimp farming and are serving as a major export commodity (in international trade) to the developing world. The industry is rapidly growing with 7 million metric tons annual production, generating billions of dollars every year in trade (currently worth around US\$10 billion or 16% of all fishery exports), employs millions of people globally and set for a period of strongly growing demand [4]. Considerable achievements have been made in the aquaculture sector to improve production; and disease resistance; however, information on the use of probiotics as control measures for the disease are scanty. Therefore, this review is an effort taken to understand the role of probiotics as an alternate source for disease control in shrimp aquaculture.

### 1.1. Shrimp Aquaculture Diseases

Despite the accelerated development in global aquaculture of shrimp, the farming industry in the past decades, faced different pathogenic diseases, which reported to cause substantial economic losses [5]. The emergence of this major challenge of shrimp diseases has threatened the success and prospect recorded in the business of shrimp farming [2,4]. This has dwarfed the aquaculture of shrimp and relegated it to the second position in terms of value added product after being the most traded product for decades [5]. Outbreak of diseases has led to a significant reduction in shrimp production and had taken many farmers out of business by converting shrimp ponds to catfish ponds, especially in India [6]. The infections devastated the economic

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and societal benefits of shrimp farming with an estimated total loss exceeding 40% of the global production capacity [7].

Infectious diseases in shrimp are caused by viral, bacterial, and eukaryote pathogens as well as some abiotic factors. Infectious diseases have become a stumbling block for expansion of shrimp aquaculture due to losses recorded [8]. Most of these losses have been attributed to viruses, which account for 60%, an estimated 20% has been attributed to bacteria while eukaryotes such as fungi and parasites' losses are comparably low [3]. Shrimp diseases caused by opportunistic bacteria such as *Vibrio* sp. and viruses are the major problems that can lead to huge losses in farmed shrimp industry [2].

Shrimp viral diseases that account for almost 60% of all shrimp losses in shrimp aquaculture are caused mainly by some viruses, namely infectious hypodermal and hematopoietic necrosis virus, Taura syndrome virus, white spot syndrome virus (WSSV), yellow head virus (YHV), and infectious myonecrosis virus [5]. WSSV and YHV are the most lethal and affect both the wild (*P. monodon*) and the domesticated (*P. vannamei*) species [3]. Others are exclusively diseases of *P. monodon* which occurs when captured ones are used for larvae production such as monodon slow growth syndrome, hepatopancreatic parvovirus, and monodon baculovirus [3]. Most viral diseases proliferate and make substantive impact when wild-caught broodstock are used in hatchery production to supply millions of larvae to stock the ponds and importation of live shrimp for aquaculture [6]. Although to date, there have been no effective drugs for the control and treatment for viral infections, the farming industry can be transformed in a safe manner through adequate preventive measures such as rigorous sanitary practices, introduction of tolerant stocks, adoption of new technologies, implementation of good biosecurity practices, and rapid diagnosis of domesticated stocks [1,5]. Biosecurity measures will ensure the exclusion of specific pathogens from cultured aquatic stocks in broodstock facilities, hatcheries, and farms, and secured environment [9]. This can be promising in global shrimp farming in reducing the impact of losses in shrimp production due to viral diseases. The market has since opened a new chapter of rapid growth due to its recovery from major viral pandemics [1].

Unlike viruses, bacterial diseases of shrimp are mostly caused by various species of *Vibrio* [10]. In most diseased shrimp studies, *Vibrio* species are the frequent bacterial flora that causes vibriosis [2]. The pathogens have led to the collapse of many farms due to its severe effect on hatcheries and high rate of morbidity and mortality. In the world of shrimp aquaculture, vibriosis has been a major problem and capable of crippling shrimp culture ponds with consequent huge economic losses [11,12]. The strength of infection depends on the organisms and strain of *Vibrio* involved the stage of development and age of shrimp, and the ambient environmental conditions [13]. The disease manifests when shrimps are weakened or when pond conditions favor the survival, growth, and spread of bacterial pathogens [12]. These conditions include water availability, large number of shrimps in hatchery, regular use of feed that is rich in protein and when environmental conditions are favorable. The several diseases caused by *Vibrio* species include loose shell syndrome (LSS), white gut disease (WGD), tail necrosis, shell disease and red disease of which LSS, WGD, and red disease caused mass mortalities in shrimp culture ponds [13].

## 1.2. Control of Shrimp Disease

The special concern today is focused on how to effectively control and prevent outbreak of diseases in shrimps. In practice, control of shrimp bacterial infections has relied solely on the use of antibiotics and antimicrobial disinfectants or chemical additives [6]. Moriarty

reported that many farmers apply antibiotics in large quantity as a preventive measure even when infections are not apparent [2]. This has consequently resulted in an increase in the multiple antibiotic resistance strains of virulent pathogenic vibriosis through mutations which has been linked to the ability of marine vibriosis to use plasmids to transfer antibiotic resistance genes between the large bacterial pond population density [14]. The marine pathogen *Vibrio parahaemolyticus* becomes severe using a plasmid that expresses a deadly toxin [2]. Biofilm formation allows *Vibrio* species to thrive on surfaces even in the presence of antibiotics which also pose a challenge in the management of shrimp diseases by protecting the bacteria against antibiotics [6]. The use of chemical agents has also been challenged due to the resultant adverse environmental effects [15]. It can accumulate in the shrimp making it unattractive for export.

To effectively manage shrimp diseases, alternative control and management measures are required [10]. This has necessitated the intense search for an environmentally friendly alternative treatment control of shrimp diseases in aquaculture that will be used to overcome the threat of antimicrobial resistance and ensure sustainable, safe and healthy food production for the growing world population. One of the effective alternatives is the use of antibacterial probiotics as an environmentally friendly approach. This approach is capable of disease control in aquaculture, especially against bacterial diseases; however, its use against viruses is still a subject of research. It is newer and safer to use with its capability to improve the animal's nonspecific immune system.

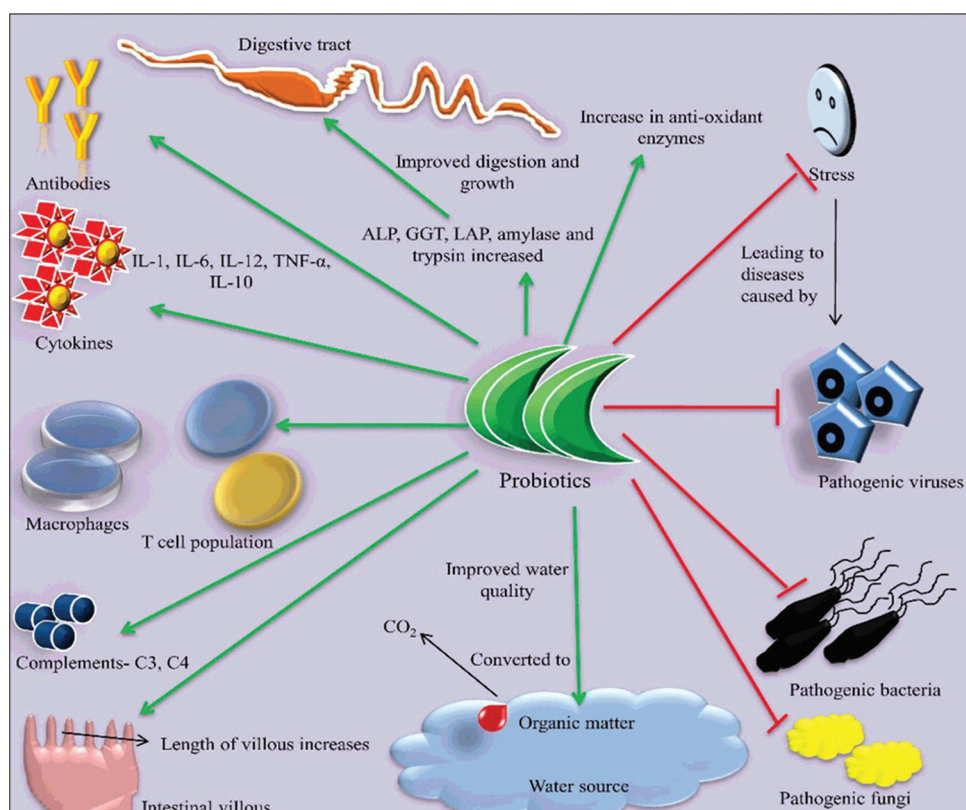
## 1.3. Definition, Source, and Potential Needs

Probiotic was best defined by Fuller as "a live microbial feed supplement which beneficially affects the host animal by improving its intestinal microbial balance" [16]. The probiotics also called beneficial bacteria are capable of improving the host's health status when consumed [17]. They cause inhibition of the pathogens growth in the host, improve the host's innate immunity and hence regarded as a replacement for antibiotic treatments. Probiotics can be commercialized; administered orally and obtained from the animal's host. It can be a single or culture of mixed nonpathogenic bacteria. Its work is based on the introduction into the culture system of harmless and beneficial bacteria that will have a competitive advantage over potential pathogens and also to colonize the ecological niche [17]. The probiotics can overcome the effect of pathogens and increase the survival of the host [10].

Verschuere *et al.* proposed a broader definition of probiotics in aquaculture "as a live microbial audit which has a beneficial effect on the host by modifying the host-associated, ambient microbial community through improvement of its feed or enhancing its nutritional value and also by enhancing the host response toward disease, or by improving the quality of its ambient environment" [18]. Probiotics improve the health status of the shrimp by resisting colonization of pathogens through competition, releasing metabolites that prevent the growth of pathogens and thus increasing the shrimp resistance to diseases [Table 1 and Figure 1].

**Table 1:** Application of probiotic for nutrient digestibility

Probiotic Identified	Applied to aquatic species	Reference
<i>Lactobacillus helveticus</i>	<i>Scophthalmus maximus</i>	[19]
<i>Bacillus</i> NL 110, <i>Vibrio</i> NE 17	<i>Macrobrachium rosenbergii</i>	[20]
<i>Carnobacterium</i> sp. Hg4-03	<i>Hepialus gonggaensis</i> larvae	[21]
<i>Lactobacillus acidophilus</i>	<i>Clarias gariepinus</i>	[22]
<i>Shewanella putrefaciens</i> Pdp11	<i>Solea senegalensis</i>	[23]



**Figure 1:** Overall beneficial effects of probiotic in Aquaculture. Green arrow indicates additive effects. Red lines indicate inhibitory effect

The use of probiotics in the control of diseases in aquaculture of shrimp is being accepted with several researchers showing promising result due to demands for the eco-friendly approach in aquaculture, but it still requires many full scale trials [24]. The use of probiotic bacteria has become increasingly popular for improved nutrition, healthy digestion, and disease prevention. It has been successfully applied in the control of shrimp and other aquatic animal diseases in many developing countries [25]. Probiotics improve the health status of the shrimp by resisting colonization of pathogens through competition, releasing metabolites that prevent the growth of pathogens and thus increasing the shrimp resistance to diseases [10] [Figure 1]. Wang *et al.* have said this qualified probiotic organism needs to be applied for the safe management of shrimp diseases in aquaculture [26].

## 2. ORGANISMS INVOLVED IN AQUATIC PROBIOTICS

In aquaculture practice, probiotics are used as live food supplement and applied in pure or mixed form to increase water quality and enhance immune responses. Many organisms are tested for use as probiotics in aquaculture, including Gram-negative and Gram-positive bacteria, bacteriophages, yeasts, and unicellular algae [27]. The probiotic strains isolated from both indigenous and exogenous microbiota of aquatic animals [28]. Probiotics could be common obtained from various sources that include the gastrointestinal (GI) tracts of aquatic animals and fish mucus [29,30]. In addition, the sources can also be from aquatic environment water or sediment or also could be isolated from microbial bioflakes [28,31]. Some of the potential probiotics are associated with the host, and they have an edge over others due to their biochemical factors [32]. Isolated bacteria from the intestine of aquatic and terrestrial animals are usually and commonly used as probiotics in aquaculture [33].

**Table 2:** Probiotic bacteria identified as growth promoters

Probiotic identified	Aquatic species	Reference
<i>Bacillus</i> sp. S11	<i>Penaeus monodon</i>	[35]
<i>Bacillus</i> sp.	Catfish	[36]
<i>Carnobacterium divergens</i>	<i>Gadus morhua</i>	[37]
<i>Alteromonas</i> CA2	<i>Crassostrea gigas</i>	[38]
<i>Lactobacillus helveticus</i>	<i>Scophthalmus Maximus</i>	[19]
<i>Lactobacillus lactis</i> AR21	<i>Brachionus plicatilis</i>	[39]
<i>Streptococcus thermophilus</i>	<i>Scophthalmus maximus</i>	[39]
<i>Streptomyces</i>	<i>Xiphophorus helleri</i>	[40]
<i>Lactobacillus casei</i>	<i>Poeciliopsis gracilis</i>	[41]
<i>Bacillus</i> NL 110, <i>Vibrio</i> NE 17	<i>Macrobrachium rosenbergii</i>	[20]
<i>Bacillus coagulans</i>	<i>Cyprinus carpio koi</i>	[42]

Many bacteria such as *Vibrio* and *Pseudomonas* sp. from marine fishes are being used as probiotics to target bacterial shrimp diseases mostly vibriosis caused by species of *Vibrio* bacteria. To date, several organisms isolated from different sources such as marine sediments, shrimp ponds, shrimp gut with species ranging from *Bacillus*, *Streptomyces*, *Pseudomonas*, as well as others including vibriosis have been reported to be sources of probiotics to promote an immune response in marine ecosystems [34,35]. Several species reported to be proper probiotic to improve the physiological function and prevent infectious diseases (vibriosis) of cultured Pacific white shrimp *L. vannamei* or the black tiger wild shrimp *P. monodon* as presented in Table 2.

The organisms are dominated by species of *Bacillus* and *Streptomyces*. *Lactobacillus*, which is commonly used as human probiotic, also had effect



on shrimp vibriosis. Others are purple non-sulfur bacteria *Rhodobacter sphaeroides* strains and *Aerifella marina* as well as commercial probiotics in mixed or pure form. The formulated probiotic product composed primarily of nutrients, vitamins as well as a high concentration of the bacteria involved [19]. In consideration of the characteristics of probiotics applied in aquaculture, it should be remembered that unlike terrestrial probiotics applied in aquatic environment, which is influenced by certain factors due to the close interaction of marine animals with their environment; which give way for potential pathogens to establish themselves in the animal's external environment making their take up by the animals a normal occurrence through feeding or osmoregulation [27].

### 3. MODE OF ACTION

The mode of action of probiotics in the aquatic system is different to that of terrestrial animal [43]. In aquatic system, the mode of

action of probiotics is either directly on the host or indirectly on the environment and sometimes involves both. In shrimp, just like other fishes, probiotics do not act just as disease control agents [44]. In aquatic animals, probiotics have several modes of action that confer benefits to their potential host [Figure 2].

The protection from pathogens after probiotic treatment could be attributed to the direct inhibition of pathogens or to the capability of probiotics in modulating the immune system of shrimp [45]. The beneficial effects of probiotics are products of several interrelated or dependent mechanisms. A pictorial representation of the different mechanisms highlighting the interrelationship of the probiotic actions is given in Figure 3. The putative probiotic can be added to the host or its ambient environment in several ways: (i) Addition to the artificial diet, (ii) addition to the culture water, (iii) bathing, and (iv) addition through live food [37].

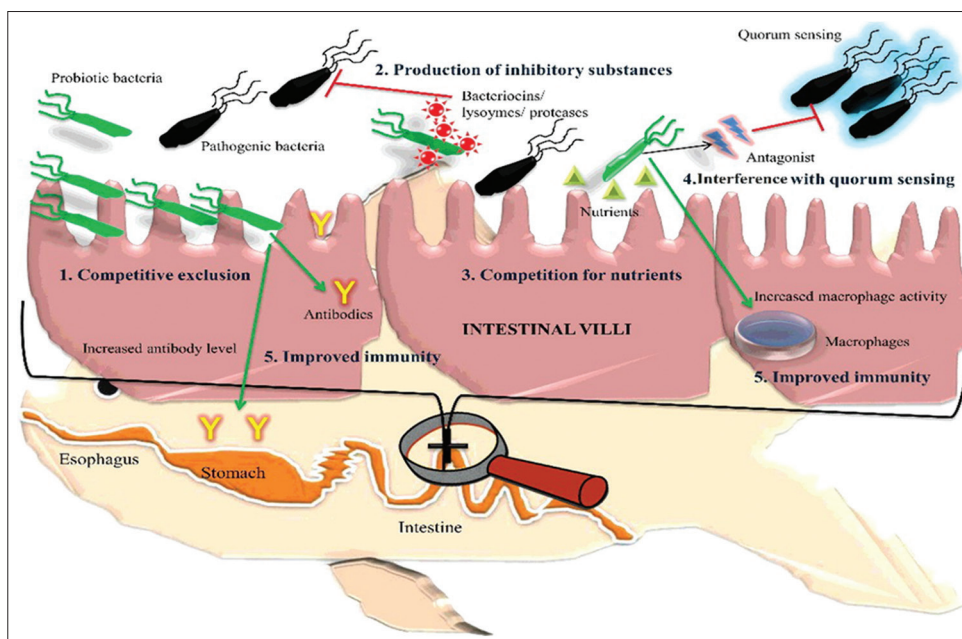


Figure 2: Mode of action of probiotics in the aquatic system

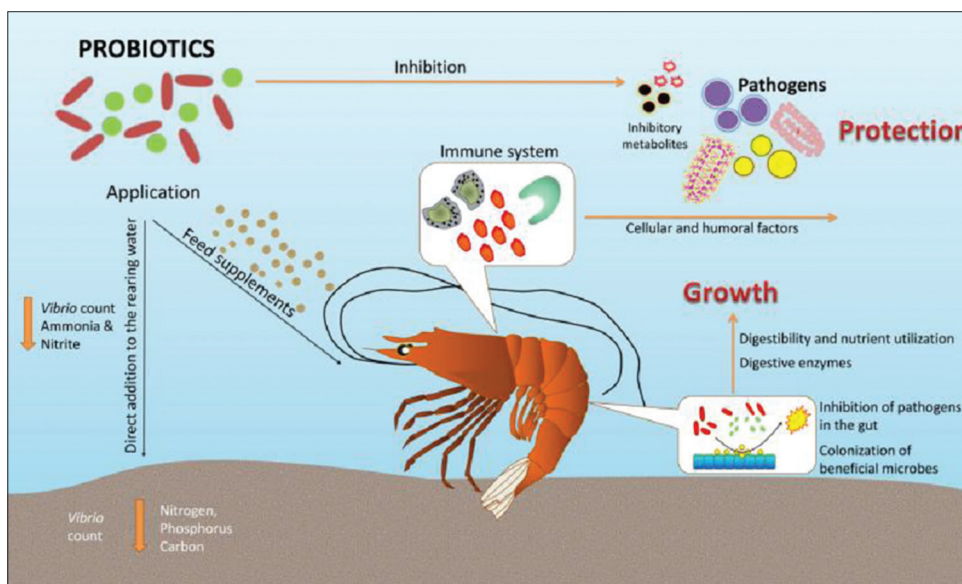


Figure 3: Interrelationship of the mechanisms of probiotic actions in shrimp

### 3.1. Stimulation of the Host Immunity

Many studies showed that some bacteria produce compounds with an immune effect in fish and shrimp. In general, this effect of immunity could be improved by the probiotic in three general ways; first, activate the macrophage, that by the increasing ability to phagocytose microorganisms; second, by increasing systematic antibodies production; and third, by increasing local antibodies at mucus surfaces [46].

Irianto and Austin reported that feeding with 107 cells/g of feed of probiotics both Gram-negative and Gram-positive bacteria led to the stimulation of cellular rather than humeral immunity [47]. Even though the increase in erythrocytes number, lymphocytes and macrophages also enhanced the activity of lysozyme in 2 weeks, and the probiotics behaving like vaccines. Another study confirm an inhibitory effect against vibriosis in turbot by lactic acid bacteria, and also the study proposed mechanisms in this field that was by inhibition of the unlikely microbes by metabolites typical of lactic acid bacteria; the microbial competition for essential nutrients [48].

### 3.2. Competition for Space

One of the major probiotics mechanism is the competition for space in shrimp aquaculture, as many of the bacterial pathogens behave attachment structures [6]. Space competition (adhesion sites) in tissues in the digestive tract is an antagonistic mechanism for probiotic to inhibit the action of pathogenic bacteria [18]. Different reports view the presence of lactic acid bacteria in the intestinal acid bacteria that constitute nonpathogenic members of the indigenous intestinal microbiota of healthy aquatic organism [48]. Furthermore, some probiotics like lactic acid bacteria strain served to fish through the food from GI tract (GIT) [49]. The attached pathogens such as Coliform and Clostridia could be removed by the adhesive probiotic bacteria, such as, and motivate their removal from the infected intestinal tract [50].

To cause diseases to a host, it will need to attach on the layer of mucosin the host GIT [51]. One mode of action of probiotic bacteria is to compete for sites of adhesion with the pathogens and hence the name “competitive exclusion.” This ability of probiotic bacteria to out-compete pathogens in colonizing the gut and in adherence to the epithelial surface and consequently interfere with the pathogens adhesion is a desirable criterion in the probiotics selection [14,32]. *Bacillus* S11, for example, which isolated from *P. monodon* broodstock GIT, the results of a 100-day feeding with probiotic-supplemented and non-supplemented (control) feeds showed effective probiotic protection with *P. monodon*. *P. monodon* showed a significant development in growth, survival, and external appearance between the trialed two groups [35]. In addition, after challenging with adding a shrimp pathogen, *Vibrio harveyi* by immersion for 10 days with the aquaculture shrimp, all probiotic treated groups survived 100%, and 26% of the control group survived, which suggested that probiotic *Bacillus* S11 is a competitive exclusion. On the other hand, some organic acid and volatile fatty acids (e.g. lactic, acetic, butyric, and propionic acids) produce by bacteria, which results in a reduction of GI lumen pH that can preventing opportunistic pathogenic microorganisms growth [52].

### 3.3. Nutrients and Competition

An important role can be played in the composition of the microbiota of the intestinal tract or ambient environment of cultured aquatic species by nutrients competition [18]. Increasing some bacteria such as *Bacillus* and *Lactobacillus* as a probiotic may cause decrease in the

available substrate for some populations of bacteria [53]. Many studies indicate the presence of bacteria in the intestinal tract, which forms nonpathogenic members of the indigenous intestinal microbiota of healthy aquatic organism. Furthermore, such strains could be active in metabolism and grow more than pathogenic bacteria [49].

Usually probiotic bacteria are colonized in the intestine, permanently or temporarily by using the intestinal mucosa as a habitat; and successful bacteria are thriving well in the gut environment, where iron is available for their growth [18]. Smith and Davey (1993) showed that fluorescent strain *Pseudomonas* bacteria during challenge test can beat the growth of *Aeromonas salmonicida* the fish pathogen, these results appear how the fluorescence is probably due to free iron competition [54,55]. The low-molecular-weight siderophores and ferric iron-specific chelating agents can dissolve and precipitated iron in the gut and make it available for microbial growth [18].

Probiotics stimulate the growth and improve the nutrient digestion in aquatic organisms, and the aquaculture is an important option in the production of animal protein, it requires high-quality feeding by proper protein content to keep organisms healthy and favor growth [47,56]. Lara-Flores *et al.* found during studying the effects of strains of some bacteria that all the probiotic-supplemented diets resulted in growth higher than those with the control diets, they also found that probiotics could lower the effects of stress factors [56]. The detoxification of potentially harmful compounds in the diet by hydrolytic enzymes could improve the nutrients in organisms, including both of amylases and proteases, and production of different vitamins such as biotin and Vitamin B12 [47]. Venkat *et al.* observed significant growth for larvae of *Macrobrachium rosenbergii* fed diets with probiotics added and found that the protein gain (>55%) and the protein efficiency ratio were significantly higher during treatments fed by probiotic [57].

### 3.4. Production of Inhibitory Substances that Work on Pathogens

Probiotic bacteria produce substances can beat other microbial populations with bactericidal or bacteriostatic effects those substances such as bacteriocins, hydrogen peroxide, siderophores, lysozymes, proteases, and many others [58]. The effects of *Bacillus* sp., for example, have been linked to the production of different antibiotics, bacteriocins, lysozymes, proteases, and hydrogen peroxide, and the alteration of pH values by the production of organic acids [18]. In aquaculture, several probiotics recorded antibacterial activity against many known pathogens including viruses [Table 3]. *Bacillus megaterium* strain increased the resistance by the shrimp, *L. vannamei* against WSSV, it was observed that probiotics such as *Bacillus* and *Vibrio* sp. could protect shrimp, *L. vannamei* against WSSV [59].

The antibacterial effect of bacteria results from factors such as the production of antibiotics, bacteriocins, siderophores, lysozyme, protease, hydrogen peroxide, the alteration of pH values, and the production of organic acids and ammonia [18]. Several compounds that may inhibit the growth of competing bacteria produces by Lactic acid bacteria and *Bacillus*, and the bacteriocins are one of the most important of those compounds [37,69]. Usually, these are proteins, or protein complexes, and produced by bacteria can have an antagonistic action against species that are closely related to the producer bacterium. Bacteriocins found in four classes: Antibiotics, small hydrophobic, heat-stable peptides, large heat-stable peptides, and complex bacteriocins: Probiotics with lipid and or carbohydrate [53].

**Table 3:** Applications of probiotics as pathogen inhibitor in aquaculture

Probiotic bacteria	Applied to aquatic species	Reference
<i>Bacillus</i> sp.	Penaeids	[2]
<i>Enterococcus faecium</i> SF 68	<i>Anguilla anguilla</i>	[60]
<i>Lactobacillus rhamnosus</i> ATCC53103	<i>Oncorhynchus mykiss</i>	[61]
<i>Micrococcus luteus</i> A1-6	<i>Oncorhynchus mykiss</i>	[47]
<i>Pseudomonas fluorescens</i>	<i>Oncorhynchus mykiss</i>	[55]
<i>Pseudomonas fluorescens</i> AH2	<i>Oncorhynchus mykiss</i>	[61]
<i>Pseudomonas</i> sp.	<i>Oncorhynchus mykiss</i>	[62]
<i>Roseobacter</i> sp. BS. 107	Scallop larvae	[63]
<i>Saccharomyces cerevisiae</i> , <i>Saccharomyces exiguous</i> , <i>Phaffia rhodozyma</i>	<i>Litopenaeus vannamei</i>	[64]
<i>Vibrio alginolyticus</i>	Salmonids	[65]
<i>Vibrio fluvialis</i>	<i>Oncorhynchus mykiss</i>	[42]
<i>Tetraselmis suecica</i>	<i>Salmo salar</i>	[65]
<i>Carnobacterium</i> sp. Hg4-03	<i>Hepialus gonggaensis</i> larvae	[21]
<i>Lactobacillus acidophilus</i>	<i>Clarias gariepinus</i>	[66]
<i>Bacillus</i> spp., <i>Enterococcus</i>	<i>Farfantepenaeus brasiliensis</i>	[67]
<i>Lactococcus lactis</i>	<i>Epinephelus coioides</i>	[68]

**Table 4:** Applications of probiotics for water quality improvement

Probiotic bacteria	Applied to aquatic species	Reference
<i>Bacillus</i> sp. NE 48	<i>Penaeus monodon</i>	[75]
<i>Bacillus</i> NL 110, <i>Vibrio</i> sp. NE 17	<i>Macrobrachium rosenbergii</i>	[20]
<i>Lactobacillus acidophilus</i>	<i>Clarias gariepinus</i>	[22]
<i>Bacillus coagulans</i> - SC8168	<i>Penaeus vannamei</i>	[76]
<i>Bacillus</i> sp. <i>Saccharomyces</i> sp	<i>Penaeus monodon</i>	[77]

### 3.5. Competition for Chemicals or Available Energy

Microorganisms, such as known probiotic group lactic acid bacteria, use the essential nutrients; which used in the growth of a number of pathogens [70]. For example, the low-molecular-weight ferric iron-chelating agents called siderophores have an ability to dissolve precipitated iron or extract it from iron complexes, then it will be ready to use in bacterial growth [71]. Furthermore, the bacteria produce siderophore could be used as probiotics due to its ability to sequester ferric iron in an iron-low environment and make it unavailable for the growth of pathogenic bacteria [52].

### 3.6. Improving Water Quality

By adding probiotic strains of the Gram-positive genus *Bacillus* water quality improves. From several studies, it became proven that application of Gram-positive bacteria, *Bacillus* spp., is beneficial for improving the quality of the water system. In comparison to the Gram-negative bacteria, *Bacillus* sp. has ability to convert organic matter into carbon dioxide, that by converting the proportion of organic matter into a slime [14]. Since this bacterial group is more useful than Gram-negative in transforming the organic matter, regarding the suggestion that high levels of probiotics in production ponds maintainance, it became easy that during the growing season, fish aquacultures can lower the accumulation of dissolved and particulate organic carbon. This also can balance phytoplankton production. Bacterium *Streptomyces* used as a probiotic in the lab culture of *P. monodon* that results a good water quality parameter than the control trial also that was effective in the growth of cultured shrimp [Table 4]. Probiotics

are also proven the capability of pond ecosystem manipulating such as microflora and the physicochemical conditions [72-74].

### 3.7. Disease Control

*Lactobacillus rhamnosus* ATCC (American Type Culture Collection, Rockville, MD, USA) was used in rainbow trout as probiotic for 51 days to reduce mortality by *A. salmonicida*. From the result, it was reduced from 52.6% to 18.9% when 109 cells/g were administered with feed when probiotic dose was increased to 1012 cells/g of feed the mortality reached 46.3% and increasing dosage does not necessarily improve protection against diseases [61]. Farmed shrimp survival increased, and feed conversion occurred when mixed cultures of probiotics were applied [26]. Meanwhile, in the study of mixed cultures of bacteria (*Bacillus subtilis*, *Lactobacillus acidophilus*, and *Clostridium butyricum*) and yeast (*Saccharomyces cerevisiae*), it registered enhancement of the non-specific immune parameters of tilapia *Oreochromis niloticus* such as lysozyme activity, migration and neutrophils, and plasma bactericidal activity, the result showed improvement of resistance to *Edwardsiella tarda* infection [78] [Table 5].

## 4. CONCLUSION

The business of the aquaculture of shrimp is growing and developing at a very fast rate throughout the world and contributes to high world production. The continuous use of antibiotics against infectious diseases in the aquaculture of shrimp and fish lead to a serious problem, while the overuse of antibiotics, which ultimately leads to diseases resistant to the antibiotic treatment such as *Vibrio* species. The successes are using probiotics, which is effective, and sustainable source against viral and bacterial pathogens encountered in shrimp aquaculture. However, many efforts have been served in the field of discovering probiotic products, and it became a fact and efficient to be used in aquaculture. Many application of probiotics in aquaculture clearly used, but there are needs for more efforts to understand the mechanisms of action to define selection for proper probiotics. Meanwhile, there is a lack in the studies of microbial aquaculture ecology and the relation with animal growth and water quality.



**Table 5:** Probiotic bacteria used in shrimp aquaculture

Probiotic bacteria	Source	Shrimp involved	Reference
<i>Streptomyces</i> spp.	Marine sediment	<i>Litopenaeus vannamei</i>	[79]
<i>Bacillus megaterium</i>	Mangrove sediments	<i>Penaeus monodon</i>	[80]
<i>Streptomyces fradiae</i>	Mangrove sediments	<i>Penaeus monodon</i>	[80]
<i>Proteobacteria firmicutes</i>	Commercial	<i>Litopenaeus vannamei</i>	[81]
<i>Streptomyces</i> Spp.	Marine sediment	<i>Penaeus monodon</i>	[82]
<i>Lactobacillus plantarum</i>	Cabbage prickles	<i>Litopenaeus vannamei</i>	[83]
<i>Bacillus subtilis</i>	Shrimp culture ponds	<i>Litopenaeus vannamei</i>	[84]
Probiotic mixture	Commercial	<i>Litopenaeus vannamei</i>	[85]
<i>Bacillus species</i>	Shrimp pond	<i>Penaeus monodon</i>	[86]
<i>Bacillus licheniformis</i>	Shrimp gut	<i>Litopenaeus vannamei</i>	[87]
Purple non-sulfur bacteria	Shrimp pond	<i>Litopenaeus vannamei</i>	[88]

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