



## REVIEW PAPER

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## Role of probiotics and their mode of action in aquaculture: A review

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### Abstract

The extensive and discriminate practice of antibiotics has affected severe biological and ecological problems, mainly the development of antibiotic resistance. Probiotics, known to be beneficial, non-pathological microbes, are being suggested as an adequate and eco-friendly alternative to antibiotics. More than three decades ago probiotics are first used in aquaculture farming, but the effective application had been given only in the recent years. Different probiotics have been identified and used in aquaculture and many of them are observed as host origin. Alternatives used for aquaculture (antibiotics) up to now have a unilateral mode of action, whereas an indefinite number of potential probiotics has multiple modes of action conferring various health benefits to the host. Recent studies suggest that a number of probiotic research publications in aquaculture focus on disease resistant and health-promoting alternatives and immune development for cultured fish and shrimps. This paper gives updated information on the use of probiotics in aquaculture farming specially targeted on the mechanism of action. It analyzes the enhancement of immune responses, antibacterial, antifungal, the antiviral activity of probiotic microbes, and water quality management in farming and inhibitory metabolites.

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## Introduction

Aquaculture farming is rapidly growing and nowadays it is considered as a major contributor to the worldwide food production. As per the United Nations Food and Agriculture Organization, the production of aquaculture division is greater than any other types of animal food production systems (FAO/WHO, 2001). To reach the global demand, the growths of aquaculture species have been enhanced to a higher extent in technological and practical measures (Tuan *et al.*, 2013). On the other hand, aquaculture production growth was inhibited by uncertain mortalities, which are affected due to pathogenic microorganisms. Majority of the pathogenic microbes are naturally existing saprophytes, which are adapted for the use of organic and mineral matter in the aquatic environment for their growth and proliferation. Most of the bacterial disease in fishes and shrimps are caused by punctiform/small, Gram-negative rods which belong to the families *Pseudomonadaceae*, *Enterobacteriaceae* and *Vibrionaceae*. These bacterial diseases are majorly observed in aquaculture even though chemicals like drugs and antibiotics are administered to control disease and health management (Newaj-Fyzul and Austin, 2014). Initially, antibiotic application for health management has been an active method, later on, residuals remaining in the rearing climate exert a pressure for long periods of storage and became a problem (Lakshmi *et al.*, 2013).

The main aim of the review is to discuss that the antibiotics and chemicals used previously are harmful and diminish the environmental conditions and we observe the major loss in aquaculture (Bachere, 2000). Accordingly, severe loss due to an immediate spread of diseases has been highly recorded. A different alternative process has been introduced to increase the growth and quality of aquaculture farming (Rekiel *et al.*, 2007; Li *et al.*, 2006), to replace those pathogenic agents we are using probiotics which are rapidly using in aquaculture now a day's. In this review, we discuss updated information on probiotics use, its mode of action and antagonistic activity. Of all those methods, probiotics have been selected as the best application method and also plays a crucial role in aquaculture (Skjermo and Vadstein, 1999).

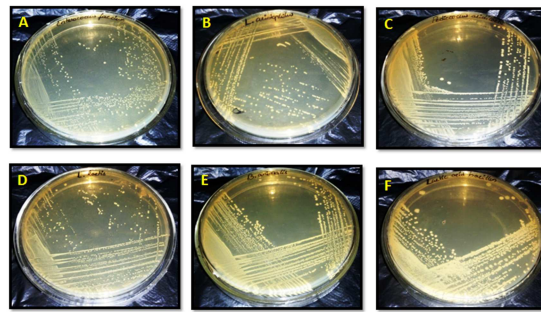
But for this probiotics till now researchers don't know the complete mechanism of beneficial bacteria role in aqua and the human body, for better interpretation of mode of action might be used for effective and appropriate administration of probiotic in aquatic farming. Along with it, the selection of appropriate administrative process provides the great advantage of optimum condition for probiotics which are able to perform the better mode of action. Probiotic supplementations have been widely applied in water and feed additives (Moriarty, 1998; Skjermo and Vadstein, 1999) with either in single or a combination two or more cultures or along with the mixture of prebiotic and vitamins (Hai and Fotedar, 2009).

### Definition of Probiotics

Metchnikoff, noble laureate researcher detected probiotics for the first time in fermented dairy products, who recorded that some acid producing microbes might eradicate fouling in the large intestine and thus aid to a prolongation of the lifespan of the consumer (Metchnikoff E, 1908). Lilly and Stillwell (Lilley DM and Stillwell RH, 1965) reported beneficial bacteria as a substance deposited by one microorganism that can encourage the growth of another. It was evaluated that an agent which has the inverse activity of antibiotics. Later, Sperti (G. S. Sperti, 1971) updated the concept of "tissue extracts that stimulate microbial growth." In 1974 Parker used the term microbial feed/food supplement (R. B. Parker, 1974). Parker described it as "organism and substances that contribute to intestinal microbial balance. Fuller defined the definition to live microbial food supplement that benefits the host by increasing the microbial growth balance of the host and later intimated that it may be effective in a range of higher temperature and salinity diversity. Later, authors suggested that probiotics are "monoculture or mixed culture of microorganisms applied to animals or humans, which benefits the host by elaborating properties of indigenous microflora" (R. Havenaar and I. Huis, 1992). Later many authors recognized probiotics as microbial cell preparation or microbial cell component, which have a beneficial action on health and well being of the host (Fuller R, 1989; Salminen S *et al.*, 1999; FAO/WHO, 2001). Guarner

and Schaafsma in 1998 reported that probiotics are live microorganisms which, when administered in adequate amounts, confer health benefits to the host (F. Guarner and G. J. Schaafsma, 1998). In 1999, Gatesoupe distinguished them as “microbial cells consumed in a certain way, which reaches the gastrointestinal tract and remain alive for improving the health (F. J. Gatesoupe, 1999). At the same time, research was carried out on the pathogenic inhibition by using probiotics; this work suggests that live microbial cell supplement gives health benefits to the host by enhancing its microbial growth (L. Gram *et al.*, 1999).

Usage of probiotics has been increased nowadays as these microbes have an antimicrobial activity through modifying the intestinal microflora, secreting antibacterial substances which fight with the pathogen to prevent their attachment to the intestine, oppose for nutrients required for pathogen survival, and producing an antitoxin effect. Extension of the probiotic concept was applicable when administered microbes survive in the Gastrointestinal tract (GIT) otherwise, general condition were recommended such as biocontrol when the treatment is antagonistic to pathological agents whenever water quality is improved. Despite for the first time probiotics were tested in fish were commercial preparations formulated for terrestrial animals. Then, these probiotics strains have been isolated from both aquatic and non-aquatic animals. On the other hand, those aquatic sources might be endogenous or exogenous microorganisms and the identified strains from endogenous sector might depend on genetic and environmental conditions. Moreover, these isolated probiotic strains have multiple modes of action conferring to health benefits. Based on functional role i.e. immunity development, the growth of the bacteria, disease resistant and as a supporter these probiotics were selected. Generally, in aquaculture the following probiotics are frequently used, *Enterococcus faecium*, *Lactobacillus acidophilus*, *Pediococcus acidilactic*, *Lactobacillus lactis*, *Bifidobacterium animalis*, and *Lactic acid Bacillus*, morphological colonies in agar plates are illustrated in fig. 1.



**Fig. 1.** Illustrates the list of Probiotic cultures used in Aquaculture sector for growth and development A- *Enterococcus faecium*, B- *Lactobacillus acidophilus*, C- *Pediococcus acidilactic*, D- *Lactobacillus lactis*, E- *Bifidobacterium animalis*, and F- *Lactic acid bacillus*.

#### Selection of Probiotics

It is essential to identify and characterize the mechanism of action of potential probiotic and its ability on the pathological agents and viability and safety of the aquatic species. A Probiotic selection criterion is based on Gomez-Gil (Gomez-Gil *et al.*, 2000) which was done through many *in vitro* and *in vivo* experiments.

- 1) Background information collection
- 2) Addition of potential probiotics
- 3) Assessment of the efficacy of potential probiotics to fight against pathogenic strains
- 4) Evaluation of the pathogenicity of potential probiotics
- 5) Estimation of the effect of the potential probiotics on the host and
- 6) Economic benefit analysis

Later on, the United Nations has suggested several numbers of conditions that should follow when the probiotic product is preferred and approved (Lee K and Salminen S, 2009). And the list of the specification includes:

- 1) The Viability of the potential probiotic strain should survive in the GIT which when administered to the host.
- 2) Multiplication and growth of the probiotic should occur when probiotic available in the GIT of the host.
- 3) For adhesion to the intestinal surface probiotic bacteria should fight against the pathogenic bacteria, and it should happen successfully.

- 4) The probiotic should be capable of inhibiting pathogenic bacteria according to *in vitro* tests
- 5) To the other contaminant agents or disinfectant probiotic bacteria should be tolerant.
- 6) For identification of a probiotic product, it should be marked on the label by genus and species name as per the international norms.
- 7) Usage, dosage, and date of expiration should be mentioned on the label and
- 8) Data mentioned on the product will not infect immune compromised animal is essential.

#### Probiotic Benefits

A few of the beneficial effects of probiotic uptake include: the balancing of microflora leads to improvement of intestinal tract health and activation and progression of the immune system, integration, and production of bioavailability of nutrients, suppressing signs of lactose intolerance and reducing the risk of some diseases. Mechanism of probiotics in the host body and its effects are mostly unknown but parts of its modification are understood based on the modification of gut of pH, inhibiting the growth of pathogens by the productions of antimicrobial substances. In this review, we clearly update on some of the benefits of probiotics and its mode of action in the aquaculture sector.

#### Enhancement of Survival and Growth

In aquatic animals the survival and growth performance was enhanced by the following probiotics: In larvae of the crab *Thalassobacter utilis* (*Portunus trituberculatus*) (Nogami *et al.*, 1997), in channel catfish (*Ictalurus punctatus*) *Bacillus sp.* (Queiroz F and Boyd C, 1998), in turbot (*Scophthalmus maximus*) rotifers with food additives are having lactic acid bacteria or *Bacillus spores* (Gatesoupe FJ, 1991).

In juvenile *P. monodon* *Bacillus sp.* (Dalmin G *et al.*, 2001) Bactocell (*P. acidilactic*) and Levucell (*S. cerevisiae*) in Artemia cysts and naupili (Gatesoupe FJ, 2002). In Nile tilapia (*O. niloticus*) *Lactobacillus acidophilus* and *Streptococcus faecium* and the yeast *S. cerevisiae* (Lara-Flores M *et al.*, 2003), In post-larval stage of freshwater prawn (*Macrobrachium*

*rosenbergii*) *Lactobacillus sporogenes* and *Lactobacillus acidophilus* (Venkat HK *et al.*, 2004) *Vibrio P62*, *Vibrio P63* and *Bacillus P64* in shrimp (Gullian M *et al.*, 2004), juvenile tilapia and freshwater prawn *Bacillus subtilis* (Gunther J and Jimenez-Montealegre R, 2004).

In hybrid striped bass yeast (GroBioticR-A) (Li Peng *et al.*, 2005), in gilthead sea bream larvae (*Sparus aurata*) *Cytophaga sp.*, *Roseobacter sp.*, *Ruegeria sp.*, *Paracoccus sp.*, *Aeromonas sp.* and *Shewanella sp.* (Makridis P *et al.*, 2005), *Lactobacillus delbrueckii* in sea bass juveniles (Carnevali O *et al.*, 2006), on juvenile dentex *Bacillus toyoi*, T, and *Bacillus cereus*, E (Hidalgo MCA *et al.*, 2006). In India major crap (Labeorohita) *Bacillus subtilis* (Kumar R *et al.*, 2006), In fingerling diet of Nile tilapia (Biogen®) (El-Haroun ER *et al.*, 2006), whereas in newly hatched larvae of fresh prawn and *L. vannamei*- *Bacillus subtilis* bacterium (Keysami MA *et al.*, 2007) and in adult shrimp *V. alginolyticus* UTM 102, *B. subtilis* UTM 126, *Roseobacter gallaeciensis* SLV03 and *Pseudomonas aestumarina* SLV22 was used for survival and growth of the fresh prawns (Balcazar JL *et al.*, 2007).

In gilthead, sea bream *lactobacillus* species were provided (Suzer C *et al.* 2008). *B. subtilis* in Cirrhinus mrigala (Hamilton) (Ghosh S *et al.*, 2008); *L. acidophilus* and *Saccharomyces cerevisiae* in juvenile common carps (*Cyprinus carpio*) and in fishes (Ramakrishnan CM *et al.*, 2008), *Bacillus pumilus* in tilapia and shrimps (Aly SM *et al.*, 2008); Based on the above evidence it is well known that probiotics have been extensively suggested to improve the growth and survival rate and feed utilization and to improve the production ratio.

Method of administration, strains of microorganisms, species, age, variability in farm practices and its diet were influenced by the effects of probiotics on animals. On confirmation by the recent research, probiotics can improve aqua and animal activity through competitive exclusion with pathogens in the digestive process. Summarised in the table 1.

**Table 1.** List of Aquatic animals and its beneficiary organisms.

Aquatic animals	Beneficiary Organisms	Reference
Nile tilapia	<i>B. subtilis</i> and <i>Lactobacillus acidophilus</i>	Aly <i>et al.</i> , 2008a
	<i>Bacillus pumilus</i>	Aly <i>et al.</i> , 2008a
	<i>Streptococcus faecium</i> and <i>Lactobacillus acidophilus</i> <sub>1</sub> and the yeast <i>Saccharomyces cerevisiae</i>	Lara-Flores <i>et al.</i> , 2003
	(Biogen®)	EL-Haroun <i>et al.</i> , 2006
Nile tilapia and freshwater prawn ( <i>M. Bacillus subtilis rosenbergii</i> )		Gunther <i>et al.</i> , (54)
Shrimp ( <i>Penaeus vannamei</i> )	Photosynthetic bacteria and <i>Bacillus sp.</i>	Keysami <i>et al.</i> , (63)
Adult shrimp	<i>L. vannamei</i> , <i>V. alginolyticus</i> UTM 102, <i>B. subtilis</i> UTM 126, <i>Roseobacter gallaeciensis</i> SLV03 and <i>Pseudomonas aestumarina</i>	Balcazar <i>et al.</i> , (29)
White shrimp ( <i>Litopenaeus vannamei</i> ) post-larvae	<i>Vibrio alginolyticus</i>	Garriques and Arevalo, 1995
Juvenile <i>Penaeus monodon</i>	<i>Bacillus sp.</i>	Dalmin <i>et al.</i> , 2001
Shrimp <i>Macrobrachium rosenbergii</i>	<i>Vibrio P62</i> , <i>Vibrio P63</i> and <i>Bacillus P64</i> , <i>Lactobacillus sporogenes</i> and <i>Lactobacillus acidophilus</i>	Gullian <i>et al.</i> , 2004 Venkat <i>et al.</i> , 2004
Larvae of the carb	<i>Thalassobacter utilis</i>	Nogami <i>et al.</i> , 1997
Indian major carp	<i>Bacillus subtilis</i>	Kumar <i>et al.</i> , 2006
Allogynogenetic crucian carp	<i>Rhodopseudomonas palustris</i> -loaded montmorillonite	Wen <i>et al.</i> , 2008
Juvenile common carp	<i>L. acidophilus</i> and <i>S. cerevisiae</i>	Ramakrishnan <i>et al.</i> , 2008
Channel catfish	<i>Bacillus sp.</i> ,	Queiroz and Boyd, 1998
Hybrid striped bass	Yeast (Grobiotic R-A)	Li <i>et al.</i> , 2005
Gilthead sea bream larvae	<i>Cytophaga sp.</i> , <i>Riseobacter sp.</i> , <i>Ruergeria sp.</i> , <i>Paracoccus sp.</i> , <i>Aeromonas sp.</i> , and <i>Shewanella sp.</i>	Makridis <i>et al.</i> , 2005
Juvenile dentex	<i>Bacillus toyoi</i> , <i>T.</i> and <i>Bacillus cereus</i> , <i>E</i>	Hidalgo <i>et al.</i> , 2006
Seabass juveniles	<i>Lactobacillus delbrueckii delbrueckii</i>	Carnevali <i>et al.</i> , 2006
Gilthead sea bream	<i>Lactobacillus spp.</i>	Suzer <i>et al.</i> , 2008
Cirrhinus mrigala	<i>Bacillus subtilis</i>	Ghosh <i>et al.</i> , 2008
Rainbow trout fry	<i>Bacillus spp.</i>	Bagheri <i>et al.</i> , 2008

*Mode of Action*

During the last decade advanced research studies have been published on probiotics. Even though the methodological and ethical limitation of animal studies is difficult to understand the mechanism of action of probiotics, only the limited explanation is present. Some beneficial applications of probiotic in host have been reported by several authors: (i) Competitive exclusion of pathogenic bacteria (Garriques and Arevalo, 1995; Moriarity, 1997; Gomez-Gil *et al.*, 2000; Balcazar, 2003; Balcazar *et al.*, 2004; Vine *et al.*, 2004a); (ii) upgrade of immune response against pathogenic agents (Andlid *et al.*, 1995; Scholz *et al.*, 1999; Rengpipat *et al.*, 2000; Gullian and Rodriguez 2002; Irianto and Austin 2002; Balcazar *et al.*, 2004); (iii) Production of Inhibitory substance (Servin, 2004; Panigrahi and Azad, 2007); (iv) Source of Vitamins and Nutrients (Dall W and Moriarty DJW, 1983; Sakat T, 1990). (v) Source of Enzymes (Priour G *et al.*, 1990); (vi) Water Quality

Management (Nogami K *et al.*, 1997); (vii) Antibacterial activity (Zhou *et al.*, 2010; Balcazar *et al.*, 2008; Zapata and Lara-Flores, 2013), (viii) Antiviral activity (Kamei Y *et al.*, 1988; Girones R *et al.*, 1989).

*(i) Competition Exclusion*

Most of the pathogenic agents desired to adhere to the mucosal layer of the host gastrointestinal tract to activate the effect of disease (Adams, 2010).

An important mode of action in beneficiary bacteria is competition for adhesion sites, known as competitive exclusion. The Ability of bacteria to colonize the gut and attachment to the epithelial surface and equally prevent the adhesion of pathogens is an essential criterion in the opting of probiotics (Balcazar *et al.*, 2006; Lazado *et al.*, 2011). Lactobacilli which are non-pathogenic intestinal bacteria encounter with pathogenic microbes for attachment to the intestinal

mucosal layer, especially on intestinal villus and enterocytes (Brown, 2011).

Addition of probiotic is well suggested as an initial husbandry practice in larviculture considering the competitive exclusion for adhesion sites might supply supportive rearing conditions (Irianto and Austin, 2002a). On the basis of a physiochemical agent, adhesion of probiotics may be non-specific or specific on the basis of attachment of beneficiary microbe on the surface of the adherent bacteria and receptor molecules on the epithelial cells (Salminen *et al.*, 1996; Lazado *et al.*, 2015).

#### (ii) Role of Probiotic in Immune response

The immune system of an aquaculture species has mainly two basic parts which include innate or nonspecific and the adaptive or specific immune system, unlike higher vertebrates. Cellular, as well as series of humoral factors, are engaged in the activation of both immune systems. Probiotics support defence mechanisms against pathological agents through the stimulation of aqua animal immune responses by damaging the negative impact of antibiotic and chemotherapeutic agents. The substances produced by the beneficiary bacteria are capable of intensifying the immune response of fish and crustacean (Sakai M 1999). Upon these *Lactobacillus acidophilus* and *Bacillus subtilis* can be administrated efficiently to develop the health status, thus improving the disease resistance ability of Nile tilapia and development of colonizing response by increasing the non specific immune response (Aly SM, 2008) By the supplement of compound probiotic diet in *Cobia* results in improved growth performance by increasing feed utilization, immune response and survival against *Vibrio harveyi*, which it might be due to improvement in the non specific immunity of *Cobia* after feeding with probiotic diets (Geng X *et al.*, 2012) Immunoglobulin and lysozyme are the one of the important antimicrobial peptide present in the blood serum of fish, which can play as the act of defence. Additionally, they have the ability to prevent the growth of the pathogenic microbe, which leads to the prevention of disease (Alexander JB and Ingram GA, 1992). Lysozyme function in fish has been recorded to

improve at the time of challenged with pathogenic bacteria or at the update of natural bacterial infections (Balcazar JL, 2007). Similar results have been observed when various experiment conducted where higher lysozyme content was produced in the blood serum of snakehead fish supplemented with LAB diet (Talpur AD *et al.*, 2014). Similarly, another most important humoral immune factor in a fish body is an immunoglobulin, which plays a crucial role in the prevention of pathogenic organisms.

#### (iii) Production of Inhibitory substance

Beneficiary bacteria produce a substance with bactericidal or bacteriostatic effects on other microbial populations such as bacteriocins, hydrogen peroxide, siderophores, lysozymes, proteases and many other ( Panigrahiand Azad, 2007; Tinh *et al.*, 2007). Along with some bacteria produces an organic acid and volatile fatty acids likewise lactic, acetic and butyric and propionic acid this can helps in lowering the acidic condition in the gastrointestinal lumen, thus inhibiting the growth of pathogenic bacteria (Tinh *et al.*, 2007). On recent studies evidence that a compound indole (3-benzopyrole) was identified in some microbes which have antifungal and antibacterial capability to inhibit the growth of pathogens (Gibson *et al.*, 1999; Lategan *et al.*, 2006).

#### (iv) Source of Vitamins and Nutrients

Probiotic might serve as an additional source of food and microbial efficacy in the intestinal tract also can be a source of vitamins or essential amino acids (Dall W and Moriarty DJW, 1983). By supplying fatty acids and vitamins, *Bacteroides* and *Clostridium sp.* contribute nutrition to the host (Sakat T, 1990). *Agrobacterium sp.*, *Pseudomonas sp.*, *Bervibacterium sp.*, *Microbacterium sp.* and *Staphylococcus sp.* supply nutritional development in some fishes Arctic charr (*Salvelinus alpines L.*) (Ringo *et al.*, 1995).

#### (v) Source of Enzymes

A few probiotic strains may involve in the digestion mechanism of bivalves by yielding extracellular enzymes, such as lipases, proteases along with it these probiotic stimulates essential growth factors (Prieur G *et al.*, 1990). Some researcher's similar observation has

been reported in adult penaeid shrimps by microflora, where few enzymes for digestion and synthesize substances which administrated. (Wang *et al.*, 2000).

#### (vi) Water Quality Management

In water, the amount of *Vibrio sp.* was reduced by using *Thalassobacter utilis* (Nogami K *et al.*, 1997) Use of *Bacillus sp.* in water improves the quality and also helps to maintain the health status of juvenile *Penaeus monodon* (Dalmin G *et al.*, 2001). *Rhodopseudomonas palustris*-loaded montomorillonite indicatively reduces the concentration of ammonia, nitrogen, nitrite, nitrate, and sulphide (Wen JH *et al.*, 2008).

#### (vii) Antagonistic Activity

##### (a) Antibacterial activity

It was documented that some probiotics in aquaculture have been possessing antibacterial influence against identified pathogens. Few examples are listed below, Microbe *L. lactis* RQ516 have been using in tilapia (*Oreochromis niloticus*) which shows inactivation against *Aeromonas hydrophila* (Zhou *et al.*, 2010). Later Balcazar *et al.*, 2008 concluded that this *L. lastis* had inhibitory activity against fish pathogens *Aeromonas salmonicida* and *Yersinia ruckeri*. The growth of the pathogenic agents in fish Nile tilapia (*O. niloticus*) was inhibited by the probiotic strain *Leuconostoc mesenteroides* (Zapata and Lara-Flores, 2013). *Bacillus subtilis* automatically lessen the number of motile *Aeromonads*, *Pseudomonas* and total Coliforms in fishes (Newaj-Fyzul and Austin, 2014). Lactic acid bacteria (*Lactobacillus buchneri*, *Lactococcus lactis*, *Lactobacillus acidophilus*, *Lactobacillus fermentum* and *Sterptococcus salivarius*) which are isolated from the intestinal part of Spanish mackerel (*Scomberomorus commerson*) has the antibacterial

activity against the *Listeria innocua* (Moosavi-Nasab *et al.*, 2014). Dhanasekarann suggested that Lactobacilli identified from the intestine of catfish (*Claris orientalis*), Hari fish (*Anguilla sp.*), Rohu fish (*Labeo rohita*), Jillabe fish (*Oreochromis sp.*) and Gende fish (*Punitus carnaticus*) exhibited noticeable inhibiting activity against *Aeromonas* and *Vibrio sp* (Dhanasekaran *et al.*, 2008). Other applications are summarised in Table 3.

##### (b) Antiviral

Some promising probiotics have antiviral effects even though the exact mechanism of probiotics are not known, the inactivation of viruses activity can be acquired by chemical and biological contents likewise extracts of marine algae and extracellular agents of bacteria (Kamei Y *et al.*, 1988). Antiviral activity against the pathological disease hematopoietic necrosis virus (IHNV) can be inactivated by the strains of *Pseudomonas sp.*, *Vibrio sp.*, *Aeromonas sp.*, and *Coryneform* groups (Kamei Y *et al.*, 1988).

For poliovirus, high specificity and more antiviral capacity were showed by Moraxella (Girones R *et al.*, 1989). Antiviral activity against IHNV and *Oncorhynchus masou* virus (OMV) by two strains (NICA 1030 and NICA1031) *Bacillus* and *Vibrio sp.* shows positive results against white spot syndrome virus (Balcazar JL, 2003).

##### (c) Antifungal

In a culture of eels (*Anguilla australis Richardson*), antagonistic activity of *Saprolegnia sp.* was inhibited by the *Aeromonas media* (strain A199) which was isolated from freshwater (Lategan MJ and Gibson LF, 2003). *Aeromonas media* reduces the effect of Saprolegniosis in fishes (Lategan MJ *et al.*, 2004).

**Table 3.** Antagonistic activity of Probiotics.

Aquatic Animals	Pathogenic Agents	Probiotic Used	Reference
	<i>Aeromonas hydrophila</i> , <i>Aeromonas salmonicida</i> , <i>Lactobacillus sp.</i> , <i>staphylococcus epidermidis</i> , <i>Serratia liquefacians</i> , <i>Vibrio anguillarum</i> , <i>Vibrio sal</i>	<i>Tetraselmis suecica</i>	Austin <i>et al.</i> , 1992
Fish	IHNV	<i>Pseudomonas sp.</i> , <i>Vibrios sp.</i> , <i>Aeromonas sp.</i> , <i>coryneforms</i>	Kamei <i>et al.</i> , 1988
	<i>A. salmonicida</i> and to a lesser extent after exposures to <i>V. anguillarum</i> and <i>Vibrio ordalii</i>	<i>V. alginolyticus</i>	Austin <i>et al.</i> , 1995
	<i>Saprolegniosis</i>	<i>Aeromonas media</i> (Strain A199)	Lategan <i>et al.</i> , 2003
	<i>A. hydrophila</i> and <i>V. anguillarum</i>	<i>C. butyricum</i> CB2	Pan <i>et al.</i> , 2008a

Aquatic Animals	Pathogenic Agents	Probiotic Used	Reference
Finned-fish	<i>Vibrio</i>	<i>Bacillus sp.</i>	Dalmin <i>et al.</i> , 2001
Catadromous fish	<i>Edwardsiella</i>	<i>Enterococcus faecium</i> SF68 and <i>B. toyio</i>	Chang and Liu 2002
Ornamental Fishes	<i>Aeromonads</i> , <i>Pseudomonads</i> and total coliforms	<i>B. subtilis</i>	Ghosh <i>et al.</i> , 2008
Atlantic cod fish	Antagonistic activity against <i>Saprolegnia sp.</i>	<i>Aeromonas media</i> (strain A199)	Lategan and Gibson (2004); Lategan <i>et al.</i> , (2003)
Catadromous fish	<i>V. anguillarum</i>	<i>Pseudomonas fluorescens</i> AH2	Gram <i>et al.</i> , 1999
	<i>Vibrio harveyi</i>	<i>Paenibacillus spp.</i> and <i>B. cereus</i>	Ravi <i>et al.</i> , 2007
Nile tilapia	<i>Vibrio parahaemolyticus</i>	<i>L. vannamei</i> , <i>V. alginolyticus</i> , <i>B. subtilis</i> , <i>R. gallacensis</i> and <i>P. aestumarina</i>	Balcazar <i>et al.</i> , 2007
	<i>A. hydrophila</i> and <i>P. fluorescens</i>	<i>B. subtilis</i> and <i>Lactobacillus</i>	Aly <i>et al.</i> , 2008a
	<i>A. hydrophila</i>	<i>B. pumilus</i> , <i>Bacillus firmus</i> and <i>Citrobacter freundii</i>	Aly <i>et al.</i> , 2008b
	<i>Vibrio</i>	<i>Clostridium butyricum</i>	Saikai <i>et al.</i> , 1995
	<i>A. salmonicida</i> , <i>A. hydrophila</i> , <i>Streptococcus iniae</i> , <i>V. anguillarum</i> , <i>Listeria monocytogenes</i>	<i>C. maltaromaticum</i> (B26) and <i>C. divergens</i> (B33)	Kim and Austin 2008
	<i>Vibrio anguillarum</i>	<i>C. divergens</i>	Gildberg <i>et al.</i> , 1997
	Mycobacterial infection	Yeast (Grostbiotic R-A)	Li Peng <i>et al.</i> , 2005
Shrimp larva	<i>Aeromonas infection</i>	<i>Bacillus subtilis</i> AB1	Newaj-Fyzul <i>et al.</i> , 2007
	Vibriosis	<i>Arthobacter</i> XE-7	Li <i>et al.</i> , 2006
	<i>A. salmonicida spp. salmonicida</i>	<i>Lactobacillus lactis</i> ssp. <i>lactis</i> , <i>Leuconostoc mesenteroides</i> and <i>Lactobacillus sakei</i>	Balcazar <i>et al.</i> , 2003
	<i>A. hydrophila</i>	Yeast glucan	Selvaraj <i>et al.</i> , 2005
Shrimp	IHNV and OMV	<i>Vibrio spp.</i> (NICA 1030 and NICA 1031)	Direkbusarakom <i>et al.</i> , 1998
	Protection against white spot syndrome virus	<i>Bacillus</i> and <i>Vibriosis sp.</i>	Balcazar 2003
	<i>Vibrio harveyi</i>	<i>Vibrio sp.</i>	
	<i>Vibrio harveyi</i> (S2)	<i>Vibrio</i> P62, <i>Vibrio</i> P63 and <i>Bacillus</i> P64	Gullian <i>et al.</i> , 2004
	<i>V. harveyi</i> , <i>V. vulnificus</i> , <i>Vibrio parahaemolyticus</i> , <i>Vibrio. alginolyticus</i> , <i>Vibrio fluvialis</i> , <i>Aeromonas spp.</i>	<i>Pseudomonas sp.</i> PS-102	Vijayan <i>et al.</i> , 2006
Black Tiger Shrimp	<i>Vibrio spp.</i> , <i>V. harveyi</i> , <i>V. anguillarum</i> , <i>V. vulnificus</i> and <i>Vibrio damsela</i>	<i>Bacillus</i> BT21, <i>Bacillus</i> BT22 and <i>B. subtilis</i> BT23	Vaseeharan and Ramasamy, 2003
Carp	Furunculosis	<i>A. hydrophila</i> , <i>Vibrio fluvialis</i> and <i>carnobacterium sp.</i>	Irianto and Austin, 2002
Fishes, Shrimps, and carp	Poliovirus	<i>Moraxella</i>	Girones <i>et al.</i> , 1989
	<i>Escherichia coli</i> and <i>Pseudomonas aeruginosa</i>	<i>Lactobacillus</i>	Oyetayo 2004

## Conclusion

Present food crisis and increasing production costs required focused on government and the international community to assure an adequate amount of food supply for the growing population. Aquaculture is presented as a way to accommodate the growing demand for fresh water food and to face challenges to the ongoing globalization of trade, intensification, and augmentation of aquaculture farming, improvement in technological innovation for food production, development in ecological system and human behavior. These challenges will give the advantage to improve aquaculture practices and also will become an important alternative for modification of aquatic ecosystems caused by capture fisheries. Now a day's use of probiotics can potentiate the benefits against the pathological agents, improving survival and growth performance, immune responses in the field of aquaculture. It is necessary to understand the mode of action in order to define selection criteria for potential probiotic. Hence, more

information is required for microbe interaction *in vivo* and development of monitoring tools are essential for better fig. out the composition and function of the microbial culture of probiotics.

Probiotics are an important aspect of aquaculture as they are an alternative to the use of antimicrobials and low usage of vaccines. Furthermore, regulation and legislation for the small scale and large scale administration of probiotics in aquaculture should be increased and implemented to keep the biological balance in an environment and to avoid undesirable biological interaction.

## Conflicts of interest

All authors have none to declare.

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