

Application of Probiotics in Aquaculture

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Abstract – Probiotics are micro-organisms or their products with health benefit to the host, have found use in aquaculture as a means of disease control, supplementing or even in some cases replacing the use of antimicrobial compounds. The action of the probiotics includes competitive exclusion, i.e. the probiotics actively inhibit the colonization of potential pathogens in the digestive tract by antibiosis or by competition for nutrients and/or space, alteration of microbial metabolism, and/or by the stimulation of host immunity. Probiotics may stimulate appetite and improve nutrition by the production of vitamins, detoxification of compounds in the diet, and by the breakdown of indigestible components. The application of probiotics in aquaculture shows promise, but needs considerable efforts of research. It is essential to understand the mechanisms of action in order to define selection criteria for potential probiotics. This paper summarizes and evaluates current knowledge of characteristics of good probiotics, the various functions of this intestinal microbial community, modes of application and the potential for further application of and probiotics in aquaculture.

Keywords – Probiotic, Aquaculture, Microorganisms, Pathogens, Feed.

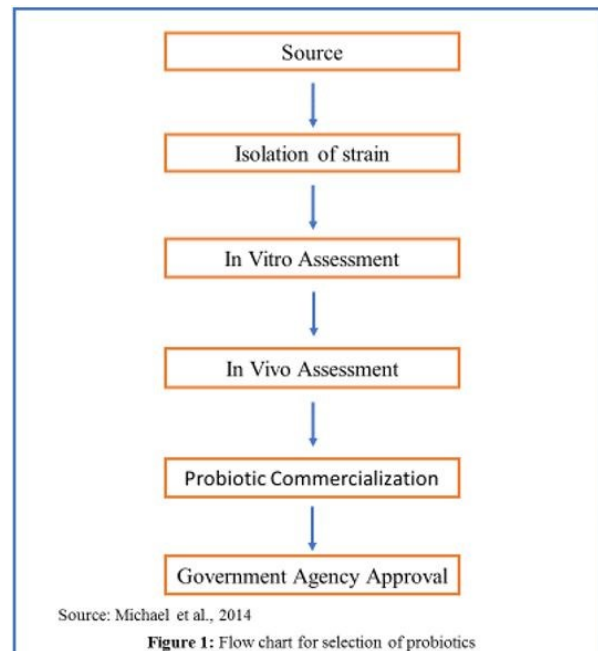
I. INTRODUCTION

The use of probiotics in the culture of aquatic organisms is increasing with the demand for more environment-friendly aquaculture practices (Ziaei-Nejad *et al.*, 2006). Most probiotics are supplied as live supplements in diets, which has the ability to survive passage through the intestinal tract (Saad *et al.*, 2009). Probiotics are beneficial microorganisms that protect the host from diseases (Kumar *et al.*, 2006). Fuller (1989) defined probiotics as ‘live microbial feed supplements which beneficially affect the host animal by improving its intestinal microbial balance’. Gram *et al.* (1999) proposed that a probiotic is any live microbial supplement, which beneficially affects the host animal by improving its microbial balance. The application of probiotics in aquaculture has been widely used as a means of controlling disease, enhancing immune response, providing nutritional and enzymatic contributions to the digestion of the host, and improving water quality (Kumar *et al.*, 2016). Probiotics are also regarded as an environmentally friendly treatment method. The probiotics may be added to feed as live microorganisms to create a balanced indigenous micro floral community in the gastrointestinal tract. Moreover, probiotics are being considered for use as therapeutic agents and some farmers are already using them preferentially over antibiotics (Tuan *et al.*, 2013). Microbes play very important and critical roles in aquaculture systems, both at hatchery and the grow-out level, because water quality and disease control are directly affected by microbial activity (Kumar *et al.*, 2006).

Selection of Probiotics:

Selection of probiotic bacteria has usually been an empi-

rical process based on limited scientific evidence. Many of the failures in probiotic research can be attributed to the selection of inappropriate microorganisms. Selection steps have been defined, but they need to be adapted for different host species and environments. It is essential to understand the mechanisms of probiotic action and to define selection criteria for potential probiotics its efficiency on the pathogen and safety to the aquatic animals and aquaculture environment. This can be done through several in vitro and in vivo experiments (Pandiyana *et al.*, 2013).



Characteristics of Good Probiotics:

A high-quality probiotic should have the following mentioned characteristics (Akter *et al.*, 2016 and Michael *et al.*, 2014):

- ✓ Should be a strain which is capable of exerting a beneficial effect on the host animal, e.g. increased growth or resistance to disease.
- ✓ Should not have any side effect; should neither be pathogenic nor toxic, not only with regard to the host species but also with regard to aquatic animals in general and human consumers.
- ✓ Should be viable under normal storage conditions and able to survive during industrial process.
- ✓ Should be capable of surviving and metabolizing in the gut environment, e.g. resistant to bile and low pH due to organic acids enrichment.
- ✓ Possess high ability to multiply in the intestine.
- ✓ Possess strong adhesive ability with the gut of the fish.
- ✓ Should have strong antagonistic activity against pathogenic microorganisms.

II. MODES OF APPLICATION OF PROBIOTICS IN AQUACULTURE

Probiotics can be applied through feeding, injection or immersion and directly by adding in the water (Irianto and Austin, 2002):

Application in Feed

Usually, probiotics are used by adding directly in the feed ingredients or by spraying in the prepared feed (Fuller, 1989). In aquaculture, commonly used probiotics are *Lactobacillus sp.*, *Bacillus sp.* or *Saccharomyces cerevisiae* (Akteret *et al.*, 2016). According to the guidelines of Food and Agriculture Organization (FAO) and World Health Organization (WHO), probiotics should have the capability of surviving while passing through the gut as well as resisting the gastric juices and bile. Additionally, probiotics should have ability to flourish and settle in the gut, which should be safe and effective for the host species.

Application through Immersion or Injection

Probiotics also can be applied by injecting or immersion. A study also demonstrated the effect of administration of a well-known probiotic *Micrococcus luteus* by intra peritoneal injection to *Oreochromis niloticus* and observed only 25% mortality while 90% mortality with *Pseudomonas* (Yassiret *et al.*, 2002).

Direct Application to Culture Water

Probiotic application directly in ponds and tanks water also exhibited beneficial effect on fish health by modulating microbial composition and water qualities of water and sediments (Venkateswara, 2007). Probiotic bacteria, particularly *Bacillus spp.* and some other species including *Aerobacter sp.*, *Nitrobacter sp.* and *Saccharomyces cerevisiae* (yeast), played a remarkable task in the water quality improvement (Akteret *et al.*, 2016).

III. FUNCTIONS OF PROBIOTICS

Several studies have demonstrated the benefits of probiotics for aquatic animals, such as the stimulation of growth or to improve feed digestion, immune responses and water quality control.

Growth and Digestive Process Promoter

Supplementing the diet fish with probiotics can reduce the use of antibiotics and synthetic chemicals in the feed (Fuller, 1989). Consequently, the addition of probiotics to fish diets has become widespread on aquaculture farms. The application of probiotics results in reduced feed costs, which plays an important role in determining the practices of aquaculture. Interestingly, previous research findings have shown that the beneficial effects of probiotics can manifest as enhanced feed utilization of cultured aquatic animals through the supplementation of digestive enzymes, improved feed efficiency and higher growth, the prevention of intestinal disorders and the pre-digestion of antinutritional factors present in mixed feed (Tuan *et al.*, 2013).

Improvement in Water Qualities

Nitrogenous compounds contamination such as ammonia, nitrite and nitrate in fish culture systems/ponds

has been a serious concern. The susceptibility of cultured aquatic species to high concentration of these compounds is generally species-specific, but in high concentrations, these compounds may be extremely harmful and cause mass mortality in all cases. Ma *et al.*, (2009) reported the ability of *Lactobacillus spp.* JK-8 and JK-11 simultaneously removes nitrogen and pathogens from contaminated shrimp farms. In several other studies, water quality has been improved by the addition of probiotics especially *Bacillus spp.* (Verschuereet *et al.*, 2000).

Effect of Probiotics on Digestive Enzymes

Some researchers have suggested that microorganisms have a beneficial effect in the digestive processes of aquatic animals. In fish, it has been reported that *Bacteroides* and *Clostridium sp.* have contributed to the host's nutrition, especially by supplying fatty acids and vitamins. Some microorganisms such as *Agrobacterium sp.*, *Pseudomonas sp.*, *Brevi-bacterium sp.*, *Microbacterium sp.*, and *Staphylococcus sp.* may contribute to nutritional processes in *Salvelinus alpinus* L (Michael *et al.*, 2014). In bivalves, some probiotic bacteria may participate in the digestion processes by producing extracellular enzymes, such as proteases, lipases, as well as providing necessary growth factors. Also, some enzymes for digestion and synthesize compounds are assimilated by microbial flora of adult penaeid shrimp (*Penaeus chinensis*) (Aly, 2009).

Enhancement of the Immune Response

The non-specific immune system can be stimulated by probiotics. It has been demonstrated that oral administration of *Clostridium butyricum* bacteria to rainbow trout enhanced the resistance of fish to vibriosis, by increasing the phagocytic activity of leucocytes (Sakai *et al.*, 1995). Rengpipat *et al.* (2000) mentioned that the use of *Bacillus sp.* (strain S11) provided disease protection by activating both cellular and humoral immune defences in tiger shrimp (*P. monodon*). Balca'zar, (2003) demonstrated that the administration of a mixture of bacterial strains (*Bacillus* and *Vibrios sp.*) positively influenced the growth and survival of juveniles of white shrimp and presented 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 increasing phagocytosis and antibacterial activity.

Antiviral Effects of Probiotics

Some bacteria used as candidate probiotics have antiviral effects. Although the exact mechanism by which these bacteria exerts its antiviral effects is not known, laboratory tests indicates that the inactivation of viruses can occur by chemical and biological substances, such as extracts from marine algae and extracellular agents of bacteria. It has been reported that strains of *Pseudomonas sp.*, *Vibrio sp.*, *Aeromonas sp.*, and groups of *coryneforms* isolated from salmonid hatcheries, showed antiviral activity against infectious hematopoietic necrosis virus (IHNV) with more than 50% plaque reduction (Pandiyaneet *et al.*, 2013).

Competitive Exclusion of Pathogenic Bacteria

The aim of probiotic products designed under competitive exclusion is to obtain: stable, agreeable and controlled microbiota in cultures based on the following: competition for attachment sites on the mucosa,

competition for nutrients and production of inhibitory substances by the microflora which prevents replication and/destroys the challenging bacteria and hence reduce colonization (Moriarty, 1998). Different strategies are displayed in the adhesion of microorganism to those attachment sites as passive forces, electrostatic interactions, hydrophobic, steric forces, lipoteichoic acids, adhesions and specific structures of adhesion (Salyers and Whitt, 2002). Adhesion and colonization of the mucosal surfaces are possible protective mechanisms against pathogens through competition for binding sites and nutrients (Westerdahl *et al.*, 1991).

IV. DISCUSSION

The application of probiotics in aquaculture shows promise, but needs considerable efforts of research. However, a number of probiotic products have been thoroughly researched, and evidenced their efficacy a possible use on aquaculture. Beneficial bacterial preparations that are species-specific probiotics have become more widely available to the aquaculture community. These preparations show specific beneficial effect as disease prevention and offer a natural element to obtain a stable healthy gut environment and immune system. The establishing of strong disease prevention program, including probiotic and good management practice can be beneficial to raise aquatic organism production.

V. CONCLUSION

The application of probiotics in aquaculture shows promise, but needs considerable efforts of research. It is essential to understand the mechanisms of action in order to define selection criteria for potential probiotics. Therefore, more information on the host/microbe interactions in vivo, and development of monitoring tools (e.g. molecular biology) are still needed for better understanding of the composition and functions of the indigenous microbiota, as well as of microbial cultures of “probiotics”. The use of probiotics is an important management tool, but its efficiency depends on understanding the nature of competition between species or strains.

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