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Review

Application of probiotics in Nigeria aquaculture: current status, challenges and prospects

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Aquaculture is the fastest growing food-producing sector in the world. Worldwide, people obtain 25% of their animal protein from fish and shellfish. A significant issue affecting aquaculture production in Nigeria is loss of stock through disease. When faced with disease problem, immediate response has been to use antimicrobial drugs. However, as a result of the growing awareness of the adverse effects of antibiotics and the increasing demand for environmental friendly aquaculture, the use of probiotics in aquaculture is now widely accepted. This review provides summary of benefits of probiotics, selection criteria, safety and evaluation of probiotics using molecular techniques. Current status, challenges and prospects in Nigeria aquaculture industry were also discussed.

Keywords: Probiotics, disease control, aquaculture, quality control, Nigeria.

INTRODUCTION

Aquaculture provided nearly 50 percent of the annual world fisheries production with 110 million tonnes of food fish in 2006. Half of all aquaculture production is fin fish, a quarter is aquatic plants and the remaining quarter is made up of crustacean (such as shrimp, prawn, crabs) and clam, oyster and mussels (FAO, 2007). Although aquaculture activity in Nigeria started about 50 years ago (Olagunju et al., 2007), aquaculture production in Nigeria is currently about 40,000 metric tonnes contributing only 6% of domestic fish production (Adeogun et al., 2007). Nigerians are high fish consumers and offer the largest market for fisheries production in Africa. Thus, Nigeria has become one of the largest fish importers in the developing world, importing about 600,000 metric tonnes annually (Olagunju et al., 2007). Fish production from captured fisheries in spite of its being expensive and risk due to the militancy activities in the coastal line regions of Nigeria has been erratic and on the decline in recent years. To solve the high demand for fish, aquaculture production remains the best option to bridge the wide gap between fish demand and domestic production. Moreover, aquaculture expansion has been a slow process; private sector fish farmers have faced major constraints. Outside the problem of high cost of fish feeds and quality seed, disease outbreak is a major challenge in fish farming in Nigeria.

The prevalence of disease under natural condition is usually low incidence because of the expanse of water and reduced risk of contact between parasite and fish (Hoffman and Bauer, 1971). However, under intensive management the crowded condition of large population of fish would result in heavy parasitic infection, disease and loss of fish (Qi et al., 2009). In recent decades, disease prevention and control have led to a substantial increase in the use of antibiotics. The wide and frequent use of antibiotics in aquaculture has resulted in the development and spread of antibiotic resistance. Because of the health risks associated with the use of antibiotics in animal production, there is a growing awareness that antibiotics should be used with more care (Defoirdt et al., 2011). The resistant bacteria can not only proliferate after an antibiotic has killed off the other bacterial, but also they can transfer their resistance genes to other bacterial that have never been exposed to the antibiotic (Verschuere et al., 2000). Resistance mechanisms can arise one of two

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ways: chromosomal mutation or acquisition of plasmids. Chromosomal mutations cannot be transferred to other bacteria but plasmids can transfer resistance rapidly (Lewin, 1992). Several bacterial pathogens can develop plasmid-mediated resistance. At the high population densities of bacteria found in aquaculture ponds, transfer via viruses and even direct transformation from DNA absorbed to the particles in the water or on the sediment surfaces could all be likely mechanism for genetic exchange (Balcázar et al., 2006).

In view of the above, the development of non-antibiotic agents is one of the key factors for health management in aquaculture. According to Browdy (1998), one of the most significant technologies that evolved in response to disease control problems is the use of probiotics. The application of probiotics in aquaculture is prevalent in United State of America, China, Japan, Indonesia, Thailand and European countries with commendable achievements. However, although the Nigerian aquaculture industry is expanding, the application and development of probiotics is very meager when compared to other countries. The purpose of this review is to describe the principles of benefits, safety and summarize recent applications of probiotics in Nigeria aquaculture. Thereby the increasing of the awareness that application of probiotics in Nigerian aquaculture has tremendous scope and a glorious future.

Probiotics; the concept

"Probiotics", "Probiont", "Probiotic bacteria" or "Beneficial bacteria" are the terms synonymously used for probiotic bacteria. The term, probiotic, simply means "for life", originating from the Greek words "Pro and "bios" (Gismondo et al., 1999). Probiotics were originally defined as "organisms and substances which contribute to intestinal microbial balance" (Parker, 1974). However, as new findings emerged, several definitions of probiotics have been proposed. According to Fuller (1989), Probiotics are "Live microbial feed supplement which beneficially affects the host animal by improving its intestinal balance". In 2001, a Joint Food and Agriculture Organization/World Health Organization Working Group on drafting "Guidelines for the evaluation of Probiotics in food" recommended that probiotics should be defined as " live micro organisms which, when administered in adequate amounts, confer a health benefit on the host" (FAO/WHO, 2001). The application of the above definitions in aquaculture, however, requires some revision. Verschuere et al. (2000) proposed a modified definition of a probiotic as "a live microbial adjunct which has a beneficial effect on the host by modifying the host-associated or ambient microbial community, by ensuring improved use of the feed or enhancing its nutritional value, by enhancing the host response towards disease, or by improving the quality of its ambient environment" This broad definition of probiotics in aquaculture system was able to address the influence of immediate environment on the health of aquatic animals. Based on the above modified definition, probiotics are recognized as microbial adjuncts that (a) prevent pathogens from proliferation in the intestinal tract on the surface structures and in the culture system of cultured species. (b) aid maximum feed utilization. (c) improve water quality, and (d) enhance the immune system of the host.

Presently, probiotics are common place in health promoting food for humans and also used as therapeutic, prophylactic and growth supplements in animal production and human health (Rinkinen et al., 2003; Senok et al., 2005; Anukam et al., 2005; Ng et al., 2009). The lactic acid bacteria (LAB) have been extensively used and researched for human and terrestrial animal purposes (Savadogo et al., 2006; Ukeyima et al., 2010). The most widely researched and used LAB are the lactobacilli and bifidobacteria (Ross et al., 2005; Senok et al., 2005). Other commonly studied probiotics include the spore forming *Bacillus* spp. and yeast Cutting (2011) have given a critical review of *Bacillus* probiotics and products for human use.

The use of probiotics is now prevalent in the aquaculture industry as a means of controlling disease, improving water quality and enhancing the immune system of cultured species (Wang, 2007; Wang et al., 2008; Ma et al., 2009). Nowadays, a number of probiotic products are commercially available in aquaculture. However, very few of these products are available in Nigerian market.

Benefits of probiotics in aquaculture

Potential benefits of probiotics in aquaculture ponds includes: enhanced decomposition of organic matter, reduction in nitrogen and phosphorus concentrations, better algal growth, greater availability of dissolved oxygen, less cyanobacteria (blue-green algae), control of ammonia, nitrite and hydrogen sulfide, lower incidence of disease and greater survival of shrimp and fish production. Although, some of the effects of probiotics have been documented clearly, research is still going on in the area with many questions about some of their benefits remaining unanswered. However, it is crucial to remember that different probiotic strains are associated with different health benefits (Senok et al., 2005).

1. Enhancement of the Immune Response

Among the numerous beneficial effects of probiotics, modulation of immune system is one of the most commonly purported benefits of probiotics. Fish larvae, shrimps and other invertebrates have immune systems that are less well developed than adult stage and are dependent primarily on nonspecific immune responses for their resistance to infection (Söderhall and Cerenius, 1998; Verschuere et al., 2000). Previous studies on fish dealt with growth promoting and disease preventive ability of probiotics. However, recent attention has been shifted towards immunomodulating effects of probiotics in fish immune system. A lot of immunological studies have been performed in several fish using different probiotics and their potency to stimulate the teleost immunity both under in vivo and in vitro conditions is noteworthy (Nayak, 2010). Ogunshe and Olabode (2009) evaluated the ability of *Lactobacillus fermentum* LbFF4 isolated from Nigerian fermented food (fufu) and *L. plantarum* LbOGI from a beverage (Ogi) to induce immunity in *Clarias gariepinus* (Burchell) against some selected fish bacterial pathogens.

Research has shown that several probitoics either individually or in combination with other probiotics can stimulate or enhance both systemic and local immunity in fish. Nayak (2010) critically reviewed the probiotics immunomodulatory activity and the factors that regulate the maximal induction of immune responses in cultured aquatic species.

2. Improvement of water quality

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 remove nitrogen and pathogens from contaminated shrimp farms. In several other studies, water quality has been improved by the addition of probiotics especially *Bacillus spp*. (Verschuere et al., 2000; Kolndadacha et al., 2009). The reason is that gram – positive *Bacillus spp.* according to Stanier et al. (1963) are generally more efficient in converting organic matter back to CO_2 than gram – negative bacteria, which would convert a greater percentage of organic carbon to bacterial biomass or slime. Dalmin et al. (2001) reported that use of *Bacillus spp.* improved water quality, survival and growth rates and the health status of juvenile *Penaeus monodon* and reduced the pathogenic vibrios.

Nitrification has been recognized to help in preventing build up of toxic ammonia. In water recirculatory system (WRS), the start –up of biofilters by transferring medium from an existing filter is a common practice. Nitrifers are responsible for the oxidation of ammonia to nitrite and subsequently to nitrate. Aerobic denitrifiers are considered to be good candidates to reduce nitrate and/or nitrite to N_2 under aerobic conditions in aquaculture systems. In China, Liao et al. (2006) isolated a new aerobic denitrifying strain X0412 from shrimp ponds which was found to contain the nitrite reducdase gene *nirs*. FAO has now designated the use of probiotics as a major means for the improvement of aquatic environmental quality (Subasinghe et al., 2003; Qi et al., 2009).

3. Source of nutrients and enzymatic contribution to digestion

Probiotics have been suggested to have beneficial effects in the digestive processes of aquatic animals (Yanbo and Zirong, 2006; Balcázar et al., 2007). Sakata (1990) reported that *Bacteroides* and *Clostridium spp*. have contributed to host fish nutrition, especially by supply fatty acids and vitamins. Prieur et al. (1990) also reported that some bacteria may participate in the digestion processes of bivalves by producing extracellular enzymes, such as proteases, lipases, as well as providing necessary growth factors. Similar observation have been reported for microbial flora of adult penaeid shrimp (*Penaeus chinensis*), where a complement of enzymes for digestion and synthesized compounds that are assimilated by the animal (Wang et al., 2000).

Probiotics selection criteria

Previous reviews have proposed favourable characteristics for the selection for cultured aquatic species (Fuller, 1989; Verschuere et al., 2000; Vine et al., 2006; Watson-Kesarcodi et al., 2008; Gómez and Balcázar, 2008; Kolndadacha et al., 2009), Merrifield et al. (2010) proposed an extended list of criteria for potential probionts which includes the following:-

- Must not be pathogenic, not only with regards to the host species but also with regards to aquatic animal in general and human consumers.
- Must be resistant to bile salts.
- Should be able to adhere to and /or grow well within intestinal mucosa.
- Should display advantageous growth characteristic.
- Should exhibit antagonistic properties towards one or more key pathogens.
- Should remain viable under normal storage conditions and robust enough to survive industrial processes.

Based on the outlined favourable characteristics of potential probiont, it is not easy to find a candidate that will satisfy all these characteristics, however, through the combined application of multiple favourable probiontic candidates it may be possible to produce greater benefits in aquaculture than the application of single probionts.

Safety and evaluation of probiotics

It is important to note that the safety profile of a potential probiotic strain is of critical importance in the selection process. Therefore, safety considerations of the putative probiotic strain should be taken into account as an integral part of process for the development and marketing of probiotics (Courvalian, 2006). Safety is the state of being certain that adverse effects will not be caused by an agent under defined conditions (Wang et al., 2008). As the search for probiotic bacteria continues, novel species and more specific strains of probiotic bacteria are constantly identified. It cannot be assumed that these new probiotic organisms share the historical safety of tested or traditional strains. Prior to incorporating them into products, new strains should be carefully assessed and evaluated for both safety and efficacy. Evaluation should include consideration for the end product formulation since this can induce adverse effects in some subjects or negate the positive effects altogether. A better understanding of the mechanism by which probiotic organisms might cause adverse effects could help to develop effective assays that predict which strains might not be suitable for use in probiotic products. Furthermore, modern molecular techniques should be applied to ensure that the species of probiotics used in aquaculture are correctly identified for quality assurance as well as safety. Conventional methods relying on phenotypic characterization, growth requirements and characteristics, fermentation profiles, and serology studies have been proven useful but carry inherit deficiencies (Qi et al., 2009). Presently, various molecular finger printing techniques using different genetic markers have been proven useful in strain differentiation. These techniques have been successfully applied in evaluating commercial probiotics in human foods (Fasoli et al., 2003; Temmerman et al., 2003, Huys et al., 2006). In Nigeria, reports on the identification and evaluation of probiotic bacteria and products using molecular techniques are rare. However, recent report by Hu and Yang (2006) using enterobacterial repetitive intergenic consensus – polymerase chain reaction (ERIC-PCR) and polymerase chain reaction – denaturing gradient gel electrophoresis/temperature gradient gel electrophoresis (PCR-DGGE/TGGE) to analyze microbial fertilizers showed that these methods are fast and accurate. Thus, there is urgent necessity for researchers in Nigeria to incorporate these sensitive and reliable molecular methods to identify and characterize the microbial content of probiotic products. In addition to PCR-DGGE/TGGE techniques, improved FISH techniques (CARD-FISH, FISH-MAR, RING-FISH), terminal restriction fragment length polymorphism (T-RFLP), multilocus sequence typing (MLST) and fluorescence amplified fragment length polymorphism (F-AFLP) are available and can be applied in probiotic studies.

CONCLUSION AND RECOMMENDATION

Probiotics are important natural ingredients in finfish aquaculture and they have numerous beneficial effects: improved water quality, improved activity of gastro-intestinal microbiota, and enhanced immune status, growth performance and feed utilization. Thus, probiotics have much potential to increase the efficiency and sustainability of agricultural production.

Nigerian aquaculture industry is expanding, however, the development and application of probiotics is very meager when compared to other countries. There is need for scientist in Nigeria to continue screening for novel probiotic strains from local aquaculture rearing systems to suit the specific requirement in Nigeria. In addition, screening and identification should not only be based on conventional methods, molecular techniques which have been shown to be fast and accurate should be applied. Since limited commercial probiotic products for aquaculture are in the Nigerian market, there is need for commercial production of putative probiotics. Also, issue of safety should not be over looked.

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