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Review

NANOPARTICLES IN AQUACULTURE INDUSTRY: CURRENT STATUS AND FUTURE PERSPECTIVES

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Abstract

The fastest-growing aquaculture industry plays a crucial role in fulfilling the increased demand for animal protein. However, the outcomes of the sectors are greatly hindered by unexpected disease prevalence, environmental degradation, organic and inorganic pollutants in aquaculture operations, and ineffective feed exploitation to achieve the global food security goal. There has been little research on the impact of nanotechnology on fish nutrition and improved health, despite the fact that the application of nanotechnology in food production systems is being studied globally. In this respect, scientists are generating and inventing new potential ideas and technology to cope with these challenges in aquaculture. Among them, the use of nanoparticles (NPs) has grown in popularity due to their numerous attractive and distinctive features and also tremendous potential to improve aquaculture production. New ideas regarding the application of NPs in fish medicine, drug delivery techniques through NPs, diet formulation to enhance feed efficacy, and as an antiviral or antimicrobial agent to control disease prevalence in aquaculture systems to augment overall production have been discussed. Commercially cultured fish species, including shrimp, Thai koi, Common carp, Grass carp, and Tilapia, have been shown to respond favorably to a variety of nanoparticles (eg. silver, gold, iron, copper, zinc, selenium, and chitosan). Thus nanoparticles researched-based information in finfish and shellfish aquaculture have short-term or long-term prospects and can be used as a potential candidate for nanomedicines combined with fish feed. These new ideas can be studied further before being utilized commercially in the aquaculture fish feed industries and it has huge merit in the future aquaculture need to be considered.

Keywords: Antimicrobial effect, diet formulation, feed efficacy, nanoparticle, nanotechnology.

Introduction

Nanoparticles (NPs) are atom clusters that range in size from 1 to 100 nanometers. Because of their enormous outer region-to-volume ratio, these metallic NPs have been shown to have exceptional antibacterial properties by increasing the formation of reactive oxygen species like H_2O_2 . Microemulsions, vaccinations, metallic, inorganic, lipid, and polymeric-based nanoparticles are among the many nanotechnological uses for infectious disease treatment that have emerged recently. Nanotechnology is a rapidly emerging new science and technology platform among recent scientific accomplishments toward the future era of agricultural development and modernization.

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Nanotechnology has formerly been used in a variety of sectors, such as chemistry, pharmaceutical research, diagnosis, and therapies, to name a few (Leso *et al*., 2019; Stefan and Monchaud, 2019). NPs, which are at the cutting edge of nanotechnology, are used in a wide range of consumer items because of their enormous potential when opposed to their bulk counterparts (Jeevanandam *et al*., 2018). As a result, the food and agriculture industries have paid close attention to the unique and growing qualities of these new-age materials. With the help of these cutting-edge materials, modern agriculture is evolving into precision agriculture, allowing farmers to get the most out of their resources. Even though, as the industry develops, there is still a question mark over its long-term viability, as the effect of rising aquaculture waste has a detrimental effect on both aquaculture productivity and the aquatic habitats surrounding it. Furthermore, Antibiotics used to treat various diseases, as well as other synthetic substances used as feed additives, have negative impacts on watery habitats (Awad and Awaad, 2017; Bandeira *et al*., 2018). From an environmental standpoint, nanoparticles have gained widespread attention for use in aquaculture and as a potential alternative to managing infectious pathogens. Nanoparticles are presently generating a lot of buzz as a potential new way to combat infectious pathogens in therapeutics because of their significant level of antimicrobial efficacy over viruses, fungi, and bacteria (Seil *et al*., 2012). From that aspect nanomaterials (NMs) are being used for the development of innovative medication administration methods for people, at the same time they could be potentially employed to develop drugs for fish. The prevalence of fish

diseases has a negative economic impact on aquaculture. Epizootic Ulcerative Syndrome (EUS) and vibriosis in fish, as well as White-Spot Syndrome (WSS) in shrimp species, are some of the most frequent diseases caused by fungi, viruses, and bacteria. Microbes, like viruses and disease-infecting agents, function on the same magnitude, controlling these diseases would necessitate early identification and elimination of pathogens using nanomaterials. Nanomaterials can be used to produce microbiome fish seedlings as well as prawn or shrimp PL for commercial farming so that the disease can be avoided (Muruganandam *et al*., 2019).

Studies showed that silver nanoparticles made with Citrus lemon juice as a reducing agent showed antibacterial and anticyanobacterial action against *Edwardesiella tarda* and *Staphylococcus aureus* bacteria, as well as anti-cyanobacterial activity against *Oscillatoria* and *Anabaena* bacteria (Swain *et al*., 2014). Nanoparticle mixed feed greatly impacts overall fish growth and survival rates, especially in the young stages. Some studies have shown that nanoparticles induce rapid growth and have a great impact on the intestinal microbiota community (Merrifield *et al*., 2013). But sometimes they may produce metal toxicity due to excessive amounts or sensitivity of particular species or life stages. Research should be done on their impact on our regular native fish species that have been cultured to a large extent. There is limited information regarding the impact of nanoparticles in the fisheries sector in Bangladesh as fish medicine, and the effect of nanoparticles enriched fish feed. Therefore, our central emphasis will be on the application of some important nanoparticles in the

realm of fish medicine, including the use of nanoparticles in the diets of aquaculture fish species.

Application of nanoparticles in aquaculture

The utilization of nanoparticles in the fisheries sector is a new practice in the world and for Bangladesh also. Research on the green synthesis of nanoparticles can be taken as it is safe for use in the biological environment. On a commercial scale, fish feed formulation with nanoparticles is absent or not yet seen in Bangladesh as chemically synthesized nanoparticles are very costly. But at the experimental level fish feed and fish medicine are being formulated with nanoparticles for activities that affect different native fish species (Fig. 1). Nanoparticles have different activities on microbes, especially

bacteria and fungi. Different nanometals have different impacts on different microbes, they can promote or resist or even annihilate the microbes. The isolated nanoparticles can be used to examine their impacts on different pathogenic microbes of fishes and use to see the growth of gut microbiota. Also, they can be used as water treatment in various diseased fish ponds and evaluate their impacts on diseased fish. Production of fungicides and bactericides can reduce the most discussed problem "Antibiotic Resistance". Resistance to antibiotics in aquaculture has prompted researchers to investigate NPs as novel antimicrobial compounds (Gunalan *et al*., 2012; Swain *et al*., 2014). The effects of some nanoparticles in aquaculture and their impact are summarized in Table 1.

Fig. 1. Types of nanoparticles and their application in the fisheries sector.

Effects of silver nanoparticles (Ag-NPs)

Silver Nano compounds are the widely studied nano-antibacterial agent in the scientific literature. They combat microorganisms in a multitude of ways. Aquaculturists face frequent bacterial disease outbreaks in their aquaculture system in our country, which causes heavy economic loss. Researchers can uptake nanotechnology considering its outstanding functions. Silver nanoparticles provide strong antimicrobial activity toward multidrug-tolerant microbial strains, according to Prakash *et al*. (2013). Ag nanoparticles were also found to kill methicillin-tolerant *Staphylococcus aureus* bacteria (MRSA) (Ayala-Nunez *et al*., 2009). Long-term medication with silver NPs reduced mortality in *Vibrio harveyi* infected juvenile marine shrimp (*Feneropenaeus indicus*) by 71% at greater dosages of silver nanoparticles, according to a study (Vaseeharan *et al*., 2010). Umashankari *et al.* (2012) employed the leaf bud essence of the mangrove *Rhizophora mucronata* to biologically synthesize Ag-NPs, which were subsequently shown to have antibacterial properties over *Pseudomonas fluorescens*, *Flavobacterium* species, and *Proteus* species. When tested against exceptionally virulent Gram positive and Gram negative pathogenic bacteria, as well as some extremely virulent fungal phytopathogens, AgNPs displayed outstanding antibacterial efficacy (Islam *et al*., 2018). Hoque *et al.* (2021) found that, the pathogenic fish bacterium (*Stenotrophomonas maltophilia* stain EP10), the oomycete (*Phytophthora cactorum* strain P-25), and the two different strains of pathogenic strawberry fungus, BRSP08 and BRSP09 (*Collectotrichum siamense*),

are all susceptible to Ag nanocomposites' antipathogenic properties. According to Chhanda *et al*. (2019) *Staphylococcus* spp. and *Salmonella* spp. caused ulcerative symptoms in Thai koi (*Anabas testudineus*). As Ag-NPs are able to kill such bacteria, they can be used as a medication for Thai koi in the culture water of our country which may eradicate the ulcerative symptoms. Moreover, using Ag-NPs in culture water for a long time will help to maintain the water quality as water will be free from harmful bacteria.

Ag-NPs (500 mg/kg meal) were given to zebrafish (*Danio rerio*), and no lesions in the intestinal epithelial or alterations in the microbial population were noticed (Merrifield *et al*., 2013). This nanoparticle can be used as a feed additive for our native *Puntius* species to improve their growth by improving their gut microbiota. Silver nanoparticles had a high antagonistic effect against *Candida* spp as an antifungal drug, equal to the conventional antifungal Amphotericin B (Sanjenbam *et al*., 2014; Mallmann *et al*., 2015). Ag NPs are also presented to have antifungal action over dermatophytes (Kim *et al*., 2008). Antiviral capabilities of Ag-NPs include the ability to attach *in vitro* conditions with HIV-1 viral proteins (Elechiguerra *et al*., 2005). Shabuj *et al*. (2015) reported that environmental changes cause yellow head viral disease (YHD), white spot syndrome viral (WSSV), vibriosis, fusarium, and protozoan in Shrimp species were recorded from the Bagerhat district, Bangladesh during the experimental period. Islam *et al.* (2020) also reported the same disease occurrence in the southwestern part of Bangladesh. So, it indicates that disease management should be remoulded, in that case, Ag-NPs can be used as a trial to treat

the viral, fungal, and protozoan diseases of cultured Shrimp species of the southwestern part of Bangladesh after considering the effectiveness of Ag-NPs.

Effects of gold nanoparticles (Au-NPs)

Owing to its low toxicity to eukaryotic cells, there is currently a greater extant curiosity in studying gold nanoparticles for their antibacterial action (Li *et al*., 2014). Bactericidal actions of gold NPs supported zeolite over *Escherichia coli* and *Salmonella typhi* has been studied (Lima *et al*., 2013). Gold nanoparticles have been presented to have antifungal action over *Candida* species. Smaller gold nanoparticles exhibited stronger antifungal activity than larger gold nanoparticles (Ahmad *et al*., 2013; Wani and Ahmed, 2013). *Heterosporis saurida* was discovered in lizardfish, which affects the kidney, skeletal muscle, body cavity, and intestinal tissue forming whitish lobules with several spores rendering the species unfit for human food (Al-Quraishy *et al*., 2012). After that Saleh *et al*. (2016) studied the antimicrosporidial capacity of Au-NPs toward *H. saurida* in the laboratory. Fungal diseases mostly occur in Indian major carp, Tilapia, and Climbing perch rendering huge losses to the aquaculturists of our country. So, the use of Au-NPs can reduce mortality due to fungal attacks and keep the culture water free from harmful bacteria.

Ava *et al.* (2020) conducted research work to assess the incidence of *Salmonella* and *Escherichia coli* (*E. coli*) contamination in different fish farms and fish markets in the Dinajpur district of Bangladesh. Their results indicated that all of the collected samples

were contaminated with *Salmonella* and *E. coli* bacteria to an unacceptable limit. Another study conducted by Faridullah *et al*. (2016) to determine the level of contamination by indicator organisms (*Salmonella* and *Escherichia coli*) in shrimp (*Penaeus monodon*) farms, depots, and processing plants of Cox's Bazar, found the same alarming result. Hossain *et al.* (2013) documented the occurrence of *Salmonella* and their antibiotic resistance pattern in shrimp from different markets in Dhaka city, the results suggest that shrimps consumed in Dhaka city may be contaminated with multi-drug resistant plasmid-harbouring *Salmonella*. Which is very much alarming for human health. After considering the beneficial and sustainable features of Au-NPs, it urges the use of this nanoparticle in the culture ponds to minimize the bacterial load from culture water as well as from fish. Proper initiative should be taken and more research should be done to make them available at the farmer level.

Effects of zinc oxide nanoparticles (ZnO-NPs)

The antibacterial and antifungal properties of ZnO-NPs have attracted huge attention (Gunalan *et al*., 2012; Swain *et al*., 2014). The particles' effectiveness stems against bacteria from disrupting the microbial cell wall, causing cytoplasm contents to seep out of the cell (Liu *et al*., 2009). *Edwardseilla tarda, Aeromonas hydrophila, Citrobacter spp, Flavobacterium branchiophilum, Staphylococcus aureus, Vibrio species, Bacillus cereus*, and *Pseudomonas aeruginosa* can all be inhibited by ZnO-NPs when used as fish medicine (Swain *et al*., 2014). Ramamoorthy *et al*. (2013) reported on the

antimicrobial properties of ZnO-NPs towards the pathogenic *Vibrio harveyi* bacterium and found that nanoparticles had stronger bactericidal effects than bulk ZnO. Above mentioned microbes cause serious diseases such as Edwardsiellosis, Motile Aeromonas Septicemia (MAS)- Dropsy, Vibriosis, and Columnaris disease (Tail and Fin root disease) in our aquaculture fishes (Carp, Tilapia, Eel, Catfish). So, researchers can also give it try using ZnO-NPs to treat such diseases in a more effective and sustainable way.

Grass carp (*Ctepharyngodon idella*) feed was supplemented with nano zinc oxide (nZnO). The percentage weight gain (%WG), specific growth rate (SGR), and feed conversion ratio (FCR) of fish fed 30 mg/kg nZnO mixed feed were found to be considerably greater (Faiz *et al*., 2015). In *Macrobrachium rosenbergii* PL, a feed augmentation of nanoscale Zn and Cu was used, resulting in improved survival and growth rate (Srinivasan *et al*., 2016). Green synthesized ZnO can be a sustainable and cost-effective source of feed additive for Carp and Prawn species in the aspect of our country most of the expanses in fish culture are done for feed and medicine purposes. Proper utilization of ZnO can help to improve the production rate.

Effects of iron nanoparticles (Fe-NPs)

They had been administered in the diet of young carp and sturgeon (Handy *et al*., 2012; ETC 2003), Young carp, *Carassius auratus*, and sturgeon, *Acipenser gueldenstaedtii* (Srinivasan *et al.*, 2016) they showed a positive result, had rapid growth. In the case of carp 30% and in the case of sturgeon 24% faster growth was observed than fish feeding

with a normal diet. Iron and Selenium NPs mixed diet was applied to *C. auratus gibelio* and the result showed faster growth in the young fish (Fonghsu, 2008; Frederick *et al*., 2010). The 60-day trial of Fe-NPs at 40 mg/kg in the feed for Bagridae catfish *Clarias batrachus* produced the best growth and feed utilization results. Substantially greater protein and fatty acid profile of fish muscle, and blood total protein, cholesterol, and triglycerides level were shown to rise in a manner which was dependent on doses, indicating improved health of fishes given a Fe-NPs supplemented feed (40 mg/kg). However, compared with the untreated group, stress markers (alanine aminotransferase, ALT; aspartate aminotransferase, AST; and alkaline phosphatase, ALP) were observed to rise with increasing Fe-NPs concentration. With changing the amount of Fe-NPs in diets, the buildup of Fe-NPs in tissue, liver, and serum increased in all fish groups, with a greater concentration in fish serum (Akter *et al*., 2018). The same kind of experiment was done in our country by using chemically synthesized nanoparticles (Fe, Zn, Cu, and Se), to produce a nano-nutrient complex (NNC) and were used in the diet of tilapia (*Oreochromis niloticus*) for 60 days at 30 mg/ kg (NNC30) and 60 mg/kg (NNC60) rate. When compared to the basal diet, the fish fed the NNC60 diet had significant changes in ultimate weight and length. Furthermore, the muscles of fish-fed nano diets showed a high level of nutritional content. The NNC60 treated fish had considerably higher levels of protein, amounts of fat, vitamin C, and

essential amino acids than the other groups (Khan *et al*., 2020). As positive results are found from these several studies, Fe-NPs can

be used as a feed additive to our aquaculture species; for improving their immunity, growth and disease resistance. In Bangladesh growth performance of local *Anabas testudineus* is not good though seed production technology has already been achieved. The use of nanoparticles with fish feed can be in the trail to check the growth performances.

Effects of selenium nanoparticles (Se-NPs)

Nano selenium was used in various fish species from Crucian carp (*C. auratus gibelio*) where it boosted fish weight, relative gain rate, and fish antioxidant status, and increased the function of glutathione peroxidase and concentration of selenium in muscle (Handy *et al*., 2012; Bhupinder, 2014) to Nile tilapia (*Oreochromis niloticus*) at 2.5 mg/kg dose and there was a 51.9 percent rise in growth rate above the control group (Deng and Chen, 2003). Nano-Se and selenomethionine were used in Gibelium as a complex mixed diet which boosted final weight, relative gain rate, GSH-Px activity, and muscle Se concentrations (Albrecht *et al*., 2006; Zhou *et al*., 2009). Whereas nano-selenium (nano-Se) and vitamin C complex induced in the diet of Mahseer fish (*Tor putitora*) increased percent weight gain (%WG), feed conversion efficiency (FCE %), and specific growth rate (SGR) of fish significantly ($p \le 0.05$). When near 0.68 mg n-Se/kg dry feed was utilized, the feed conversion ratio (FCR) was significantly reduced ($p < 0.05$) in supplemented diet fish. And when they were applied to Common carp (*Cyprinus carpio*) at a 1 mg/ kg rate in comparison to the control, it reflected in superior growth performance (in terms of final weight and weight gain), increased selenium contents in the liver and muscle, and

lead to increases $(p < 0.05)$ total protein and globulin (Khan *et al*., 2017). Tilapia, mahseer, and common carp are widely cultured fish in Bangladesh and the inclusion of nano selenium in the feed of these fishes can be a step towards sustainable fisheries.

Effects of copper nanoparticles (Cu-NPs)

Cu-NPs (500 mg/kg meal) changed the microbial population in zebrafish (*Danio rerio*). The richest and most diverse bacterial species were found in fish subjected to Cu-NPs, while certain essential species were absent from the population in zebrafish introduced to Cu-NPs. *Cetobacterium somerae*, a common part of the bacterial communities in the fish intestine, was not found in zebrafish fed a Cu-NPs-containing diet (Merrifield *et al*., 2013). *C. somerae* is a beneficial microbe for zebrafish as well as other cyprinid fishes, it improves carbohydrate utilization of fishes when supplemented as a probiotic for fish (Wang *et al*., 2021). So in this case, more specific research is needed about the use of Cu-NPs as a feed additive, as it may alter the intestinal microbial community by removing beneficial bacteria.

Effects of chitosan nanoparticles (C-NPs)

The optimum amount of 1.0 g C-NP/kg food considerably increased the performance of Nile tilapia (*Oreochromis niloticus*). Due to C-NP supplementation, malondialdehyde levels reduced dramatically, whereas catalase, superoxide dismutase, lysozyme, and respiratory burst activities were significantly improved. C-NP in the diet has substantial immune-modulatory capabilities and improves performance and health greatly

(Abdel-Tawwab *et al*., 2019). Dawood *et al*. (2020) found the same result as fish-fed chitosan nanoparticles at a 1 g/kg diet had relatively higher immune and anti-oxidative responses. It indicates that chitosan NPs need to be used in a dose-dependent manner. All the utilization of nanoparticles need to be checked at the field level with detailed experiment and after the validation, the commercial feed industry can cope with this nanotechnological benefit with the inclusion of fish feed.

The disease is considered one of the important factors in the decrease in fish production, both in the farming system and in wild conditions. Large-scale mortality of fish often occurs in ponds due to environmental stress followed by parasitic invasion and bacterial, fungal, protozoan and monogenean infections (Hossain *et al*., 2011). Aftabuddin *et al*. (2016) reported that most aquaculturists face some common problems such as; bad water quality, diseases, poor quality seeds/fry, availability of quality feed, etc. As per the report, the farmers use chemicals and antibiotics to solve those problems, but the chemicals and antibiotics have a serious negative impact both on the environment and human beings. Green synthesized nanoparticles (Ag, Au, Cu, Zn, etc.) can be a solution to inhibit pathogenic invasion of aquaculture as they have an antimicrobial effect against most pathogens and improve growth and immunity by incorporating NPs with feed. Worldwide nanoparticles are being adopted to control fish diseases and disease-causing pathogens. Many fish hatcheries are adopting nano copper, and nano silver technology to destroy the virus, bacteria, protozoa, and other disease-causing microbes through five-layered filtration systems (Fishtech Hatchery limited). They are

successfully producing shrimp brood and PL which are 100% organic and antibiotic-free.

In terms of the fisheries sector of Bangladesh, the natural habitat is decreasing drastically day by day. People are moving towards culture fisheries and adopting more intensive to superintensive methods of fish culture. Due to the overcrowding of fin fish and shellfish, farm disease outbreak and large-scale mortality, and product quality loss is occurring. Different types of new technology or methods are being adopted to face the problem. Countries like and China, Japan, India are looking forward to nanotechnology and nanoparticles to enhance and secure farm production. It is high time for Bangladesh to adopt these technologies and relocate resources in this new area of science to formulate a new diet or medicine which can help to sustain the intensive and super-intensive fish farming system and secure food supply to the people and make sure exportability. There is a great prospect of using nanotechnology or nanoparticles in shrimp farms in the seaside region of Bangladesh, especially in the case of white spot disease. By doing research on various nanoparticles, it will be possible to produce a vaccine, practice medicine, or reduce the severity of this disease. It will be commercially more viable as prawns and shrimp are high-value fish and bring large quantity foreign currency to our country. To maintain seed supply, research should also be conducted to assess the effects of various diets rich in nanoparticles on the survival and disease resistance of fish fry and fingerling. Some research should be done to formulate the complex feed. Initiatives and resource allocation must be done now at University and Government levels.

Conclusion

With the revolutionary utilization of nanoparticles in fish medicine and diet, nanotechnology has evolved as a significant prospect for the development of aquaculture. Pure metallic nanoparticles have antibacterial properties and are used in aquaculture to avoid bacterial tolerance. Nanoparticles with antimicrobial capabilities have been used to inhibit bacterial growth in aquaculture systems. Several researchers have explored the impact of nanomedicines as antifungal and antiviral drugs in depth. Many fish disease-causing bacteria were found to be resistant to silver, gold, and zinc nanoparticles. Nanoparticle supplementation should be commercially adopted in fish feed companies especially for young fish like fry or fingerling to increase their survivability, rapid growth and ensure early health well-being and create disease resistance. In the case of nanotechnological applications in fish therapeutics, there are numerous research gaps. Nanoparticles' antiviral and antifungal properties in fish illnesses are needed to be investigated. If nanotechnological technologies are to be extensively used, more research into vaccine development and diet composition in fish is required.

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