Chapter 15
Perspectives of Nanotechnology in Aquaculture: Fish Nutrition, Disease, and Water Treatment

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15.1 Introduction

Globally, food fish demand has been on the rise for the past seven decades (annual consumption rate at 3.1%), at a rate nearly double that of the annual global human population growth (1.6%) (FAO 2020). The sad reality is that the fishing sector (capture fisheries) which has been the main supplier of food fish over the years is alone unable to meet the current and future global food fish demand. To meet the global food fish demand, aquaculture, which is one of the fastest growing food-producing sectors, is believed to be a good opportunity to complement capture fisheries. The stress on aquaculture to close the supply and demand gap of food fish has led to a shift from extensive to intensive methods such as the recirculatory aquaculture systems (RAS). In the intensive systems, fish are stocked at high density, and this has been shown to cause stress in farmed fish, thereby affecting fish performance and welfare (Sneddon et al. 2016; Hoseini et al. 2019). In addition, these systems could also be accumulation grounds for pollutants either from water sources or fish feeds (Wang and Wang 2012; Boonanuntanasarn et al. 2014) and diseases (Romero et al. 2012; Culot et al. 2019). Thus, since the emerging of intensive farming systems, the sustainability of aquaculture has been predominantly criticized.

The benefits associated with aquaculture such as the provision of accessible food, income generation, and community empowerment could have led to the radical search for effective strategies to mitigate negative impacts, instead of discouraging the practice. One of the emerging technologies is nanotechnology, which is defined as the "science and engineering concerned with the design, synthesis, characterization, and application of materials that possess a functional organization on the nano-metric scale (10^{-9} m) (Silva 2010)". Nanoparticles are characterized by higher reactivity and can change the pharmacological properties of active principles (Jiang et al. 2019). This technology is widely researched in aquaculture for various purposes such as vaccine delivery (Rajeshkumar et al. 2009), gene transfer (Murata et al. 1998), drug delivery (Lavertu et al. 2006; Wei et al. 2007), delivery of nutrients (Ashouri et al. 2015), nutraceuticals (Aklakur et al. 2016), and water filtration and remediation (Khosravi-Katuli et al. 2017). Therefore, this chapter reviewed the application of nanotechnology in aquaculture with specific focus on fish nutrition, diseases, and water quality management (Fig. 15.1), presenting trends and perspectives.

15.2 Nanotechnology Application in Fish Nutrition

Traditionally, feeding fish has relied on providing fish with food in the form of a pellet/ bycatch/ fish-offal. The pellet is chiefly formulated based on the daily nutritional fish requirements for major components such as proteins, carbohydrates, fats, minerals, and vitamins. Recently, nutritionists utilized nanotechnology to create various delivery systems such as encapsulation, protection, and controlled release of

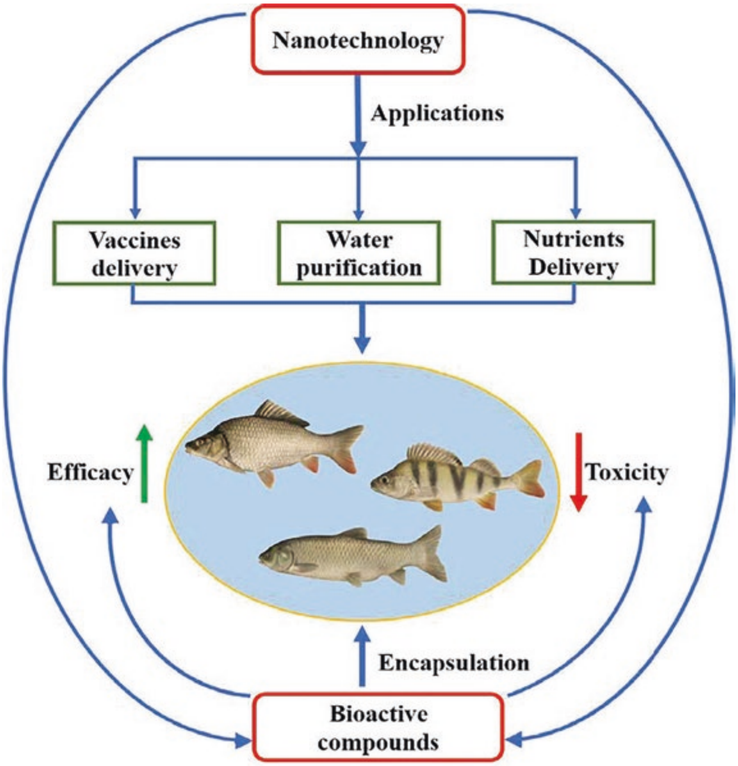

Fig. 15.1 Schematic representation of nanotechnology applications in aquaculture. (Adapted from Shah and Mraz (2020))

micronutrients. Hence, nanotechnology has an important potential to boost nutritional assessment and measures of bioavailability. For instance, ultrasensitive detection of nutrients and metabolites increases the understanding of nutrient and biomolecular interactions in specific tissues.

In nutritional research, gastrointestinal tract has always been the preferred and most important route of feed/food delivery principles including for nanoparticles. Nanoparticles can route to the gastrointestinal tract in many ways such as (1) ingestion or swallow pathway: ingestion directly from food and water and from therapeutic nano-drugs administration; (2) inhalation pathway: inhaled nanoparticles can be swallowed and enter to the gastrointestinal tract following clearance from the respiratory tract; and (3) oral pathway: oral or smart delivery into gastrointestinal tract, in which particle uptake in the gastrointestinal tract depends on diffusion and accessibility through mucus and contact with the cells of the gastrointestinal tract (Hoet et al. 2004). The smaller the particle diameter the faster is the diffusion through gastrointestinal tract mucus to reach the cells of intestinal lining, followed by uptake through gastrointestinal tract barrier to reach the blood (Hoet et al. 2004).

In fish, one important idea is that nanoparticles will enhance aquafeeds by increasing the proportion of fish feed nutrients that pass across the gut tissue and into the fish, rather than passing directly through the fish digestive system unused (Handy 2012). Specifically, the delivery systems of nanoparticles are aimed to improve the bioavailability, bioaccessibility and hence efficacy of the nutrients by improving their solubility and protection of fish gut. The delivery systems specifically consist of micronutrients trapped within nanoparticles that may be fabricated from surfactants, lipids, proteins, and/or carbohydrates (Joye et al. 2014). The small particle size in these systems has several advantages over the conventional delivery systems including improved bioavailability, higher stability to aggregation and gravitational separation, and higher optical clarity (Joye et al. 2014). For instance, immuno-modulatory ingredients such as phenolic compounds, vitamins, and minerals are being increasingly introduced into aquafeeds to improve fish health and growth performance. Nevertheless, incorporating these nutraceuticals into feeds is often challenging due to their low bioavailability, which can be solved by encapsulating the bioactive components. As the size of a particle containing encapsulated bioactive agents decreases their bioavailability increases, enabling their faster digestion and absorption. Besides, nanoparticles can be formulated to survive passage through specific regions of the gastrointestinal tract and then release their payload at a specified point, thus maximizing their potential immune-nutritional benefits (Jafari and McClements 2017).

15.2.1 Nanoparticles' Role in Fish Nutrition

In fish nutrition, nanoparticles are playing an important role in improving growth performance and immuno-biochemical (health) status of fish. They are usually incorporated in little amount, however, at a higher cost. Therefore, intensive care should be taken in their usage to maximize their utilization and avoid wastage (Friends of the Earth 2008). Consequently, many studies have been reported to address the functions and levels of nanoparticles and various methods have been adopted in aquatic animals (Table 15.1):

Sahu et al. (2008) conducted a 60 days experiment to investigate the effect of dietary *Curcuma longa* nanoparticles (0.1, 0.5, 1.0 and 5.0 g kg^{-1} of orally supplemented feed) on enzymatic and immunological profiles of rohu, *Labeo rohita* (Ham.), infected with *Aeromonas hydrophila*. Dietary *C. longa* nanoparticle significantly enhanced lysozyme activity, superoxide anion production, and serum bactericidal activity; and promoted protection against *Aeromonas hydrophila* (Sahu et al. 2008). In vitro and in vivo studies on the effects of dietary curcumin nanoparticles (0.5 & 1%, orally supplemented feed) reported that dietary curcumin nanoparticles (1) significantly enhanced growth, survival rates, and disease resistance; (2) decreased lipid peroxidation product; (3) promoted antioxidant status and protein content; (4) improved liver proactive effects; (5) increased haemoglobin content, RBC count and haematocrit; and (6) enhanced overall growth performance and

Table 15.1 Application of nanotechnology/ nanoparticles role in fish nutrition

Nanoparticles	Function	Fish Species	References
Aloe vera	A diet supplemented with 1% *Aloe vera* nanoparticles significantly promoted the growth parameters of fish in contrast to a control group.	Siberian sturgeon (*Acipenser baerii*)	Sharif Rohani et al. (2017)
Ginger	Fish fed with 1 and 0.5 g ginger nanoparticles per kg feed showed 100% relative percentage survival, whereas fish fed with 0.5 g ginger per kg feed showed 20% mortality rate and 71% relative percentage survival. These findings confirmed that ginger nanoparticles as a successful formulation in the prevention of motile *Aeromonas septicaemia* in common carp fingerlings compared to ginger.	Common carp (*Cyprinus carpio*)	Korni and Khalil (2017)
Azolla microphylla	Significantly ameliorated the levels of metabolic enzymes, hepatotoxic markers, oxidative stress markers, altered tissue enzymes, reduced hepatic ions, abnormal liver histology, etc. Based on those results, it was suggested that *Azolla microphylla* phytochemically synthesized gold nanoparticles as an effective protector against acetaminophen-induced hepatic damage in fresh water common carp.	Common carp fish (*Cyprinus carpio* L.)	Kunjiappan et al. (2015)
Azadirachta indica (neem)	Significantly elevated functional activity of immunological parameters in fish treated with these nanoparticles. It was concluded that they have a potential immunomodulatory and antibacterial activity.	Mrigala carp (*Cirrhinus mrigala*)	Rather et al. (2017)
Curcuma longa	Significantly enhanced lysozyme activity, superoxide anion production, and serum bactericidal activity; and improved protection against *Aeromonas hydrophila.*	Rohu (*Labeo Rohita*)	Sahu et al. (2008)

(continued)

Table 15.1 (continued)

Nanoparticles	Function	Fish Species	References
Curcumin	Enhanced growth, survival rates, and disease resistance of *A. testudineus* (Bloch). Promoted antioxidant status and protein content of the fish. Decreased lipid peroxidation product. Increased haemoglobin content, RBC count, and haematocrit in the fish. Improved over all health status of the fish.	*Anabas testudineus* (Bloch)	Manju et al. (2009, 2012, 2013)
Curcumin	Remarkably minimized CCl4-induced liver damage by upregulating hepatocyte antioxidative capacity and inhibiting NF-kB, IL-1b, TNF-α, and IL-12 expression in Jian carp.	Jian carp (*Cyprinus carpio* var. Jian)	Cao et al. (2015)
Curcumin	Notably improved growth performance, feed utilization, oxidative status, immune responses, and disease resistance of fish. Promoted non-specific immune defense mechanisms against *Vibrio alginolyticus*. Enhanced hepatic lesions in aflatoxin B infected fish.	Nile tilapia (*Oreochromis niloticus*)	Elgendy et al. (2016); Mahmoud et al. (2017); Manal (2018)
Curcumin	Enhanced growth performance and increased disease resistance against *Edwardsiella tarda* infection.	Mrigala carp (*Cirrhinus mrigala*)	Leya et al. (2017)
Curcumin	Promoted performance of catfish and increased their disease resistance, reducing use of antimicrobials in fish farming.	Channel catfish (*Ictalurus punctatus*)	Hafiz et al. (2017)
Curcumin	Enhanced the activities of digestive enzymes. Modulated the expression of GH in brain and growth factors such as IGF-1 and IGF-2 in muscle of *O. mossambicus.*	Mozambique tilapia (*Oreochromis mossambicus*)	Midhun et al. (2016)
Selenium (Se)	Nano-Selenium (Se, 1 mg kg^{-1} diet) showed significant improvement in the growth and antioxidant defense system of common carp in contrast to a control group.	Common carp (*Cyprinus carpio*)	Ashouri et al. (2015)
Selenium (Se), zinc (Zn) and manganese (Mn)	Dietary nanoparticles such as nano-selenium (Se), zinc (Zn), and manganese (Mn) in early weaning diets enhanced stress resistance and bone mineralization of gilthead seabream.	Gilthead seabream (*Sparus aurata*)	Izquierdo et al. (2017)

(continued)

Table 15.1 (continued)

Nanoparticles	Function	Fish Species	References
Iron (Fe)	A diet supplemented with iron (Fe) nanoparticles and *Lactobacillus casei* as a probiotic significantly promoted growth performance of rainbow trout.	Rainbow trout (*Oncorhynchus mykiss*)	Mohammadi and Tukmechi (2015)
Manganese (Mn)	Dietary MnO nanoparticles (16 mg kg^{-1} diet) significantly elevated the growth performance and antioxidant defense system of freshwater prawn.	Freshwater prawn (*Macrobrachium rosenbergii*)	Asaikkutti et al. (2016)
Copper (Cu)	Supplementation of dietary copper (Cu) nanoparticle (20 mg kg^{-1} diet) significantly improved the growth, biochemical status, digestive and metabolic enzyme activities, antioxidant, and non-specific immune response of aquatic animals.	Freshwater prawn (*M. rosenbergii*) and Red sea bream (*Pagrus major*)	Muralisankar et al. (2016); El Basuini et al. (2017)

health status of *A. testudineus* (Bloch) (Manju et al. 2009, 2012, 2013). Cao et al. (2015) studied the effects of curcumin nanoparticles (0.1%, 0.5%, or 1.0% of orally supplemented feed) on antioxidative activities and cytokine production in Jian carp (*Cyprinus carpio* var. Jian) with CCl_4-induced liver damage. Dietary curcumin nanoparticles significantly reduced CCl_4-induced liver damage in Jian carp by upregulating hepatocyte antioxidative capacity and inhibiting NF-kB, IL-1b, TNF-a, and IL-12 expression (Cao et al. 2015). Supplementation of curcumin nanoparticles (0.5, 1, or 2% of diet) significantly improved non-specific immune defense mechanisms of fish against *Vibrio alginolyticus*; promoted hepatic lesions in aflatoxin B infected fish; improved hepatosomatic index (HIS) values; and enhanced growth performance, feed utilization, oxidative status, immune responses, and disease resistance of tilapia, *Oreochromis niloticus* (Elgendy et al. 2016; Mahmoud et al. 2017; Manal 2018). Leya et al. (2017) evaluated the effects of curcumin nanoparticles supplemented diet (0.25, 0.5, 1, 1.5 and 2% of orally supplemented diet) on growth and non-specific immune parameters of mrigala carp (*Cirrhinus mrigala*) against *Edwardsiella tarda* infection. Dietary curcumin nanoparticles improved growth performance and increased disease resistance against *Edwardsiella tarda* infection in *C. mrigala* (Leya et al. 2017). Curcumin nanoparticle supplementation (0.5 & 1% of orally supplemented diet) significantly enhanced performance and increased disease resistance of catfish, *Ictalurus punctatus* (Hafiz et al. 2017). Midhun et al. (2016) evaluated modulation of digestive enzymes, GH, IGF-1, and IGF-2 genes in the teleost, Tilapia (*Oreochromis mossambicus*) by dietary curcumin nanoparticles (0.5 & 1%). Dietary curcumin nanoparticles significantly improved the activities of digestive enzymes and modulated the expression of GH in brain and growth factors such as IGF-1 and IGF-2 in the muscles of tilapia, *O. mossambicus* (Midhun et al. 2016).

Studies in other nanoparticles, Sharif Rohani et al. (2017), evaluated the effects of three different levels (0.5, 1.0, and 1.5% of the diet) of *Aloe vera* nanoparticles on the growth performance, survival rate, and body composition of Siberian sturgeon (*Acipenser baerii*). This study reported that a diet supplemented with 1% *Aloe vera* nanoparticles significantly promoted the growth factors of fish in contrast to a control group but did not find significant difference in body composition of fish (Sharif Rohani et al. 2017). Korni and Khalil (2017) studied the effect of ginger and its nanoparticles on growth performance, cognition capability, immunity, and prevention of motile *Aeromonas septicaemia* in common carp (*Cyprinus carpio*) fingerlings. Fish fed with ginger nanoparticles (1 and 0.5 g kg^{-1} of diet) showed better growth performance, and significantly increased total protein, globulin, and lysozyme of fish; and showed 100% relative percentage survival (RPS) compared to control group (Korni and Khalil 2017). Kunjiappan et al. (2015) investigated the hepatoprotective and antioxidant effects of *Azolla microphylla*-based gold nanoparticles against acetaminophen-induced toxicity in a fresh water common carp fish (*Cyprinus carpio* L.). Their results showed that gold nanoparticles significantly ameliorated the levels of metabolic enzymes, hepatotoxic markers, oxidative stress markers, altered tissue enzymes, reduced hepatic ions, abnormal liver histology etc. Based on those results, it was suggested that *A. microphylla* phytochemically synthesized gold nanoparticles as an effective protector against acetaminophen-induced hepatic damage in fresh water common carp (Kunjiappan et al. 2015). Rather et al. (2017) evaluated the immunomodulatory potential of green synthesis of silver nanoparticles (G-AgNPs) using *Azadirachta indica* (neem) in *Cirrhinus mrigala* fingerlings challenged with *Aeromonas hydrophila*. This study reported that dietary G-AgNPs significantly increased the functional activity of immunological parameters (nitro-blue tetrazolium assay, myeloperoxidase activity, phagocytic activity, anti-protease, and lysozyme activity), enhanced disease resistance and improved survival rate; and it was concluded that biosynthesized silver nanoparticles have immunomodulatory and antibacterial activity (Rather et al. 2017). Formulation of solid lipid nanoparticles-encapsulated 6-coumarin-loaded pectin microparticles showed improved uptake of the compound by two gilthead seabream (*Sparus aurata* L.) cell types compared to a competitor 6-coumarin-loaded pectin microparticles, which makes solid lipid nanoparticles as suitable nanocarriers for the delivery of biologically active substances in fish (Trapani et al. 2015).

Furthermore, dietary nano-minerals or dietary minerals at the nanoscale size may pass into cells more readily than their larger counterparts, and this accelerates their assimilation process into the fish. For example, dietary selenium (Se, 1 mg kg^{-1} of diet) nanoparticles significantly promoted growth and antioxidant defense system of common carp (*Cyprinus carpio*) in contrast to a control group (Ashouri et al. 2015). In rainbow trout, a dietary iron (Fe) nanoparticles and *Lactobacillus casei* as a probiotic significantly improved growth performance and feed utilization, such as weight gain, specific growth rate, daily growth rate, condition factor, and food conversion rate (Mohammadi and Tukmechi 2015). Nanoparticles such as nano-selenium (Se), zinc (Zn) and manganese (Mn) in early weaning diets for gilthead seabream (*Sparus aurata;* Linnaeus, 1758) enhanced stress resistance and bone mineralization (Izquierdo et al., 2017). Dietary copper (Cu) nanoparticle (20 mg kg^{-1} of diet) significantly improved the growth, biochemical status, digestive and metabolic enzyme activities, antioxidant, and non-specific immune response of red sea bream, *Pagrus major* (El Basuini et al. 2017) and freshwater prawn, *M. rosenbergii* (Muralisankar et al. 2016). Supplementation of manganese oxide (MnO) nanoparticles (16 mg kg^{-1} diet) significantly elevated the growth performance and antioxidant defense system of freshwater prawn (*Macrobrachium rosenbergii*) (Asaikkutti et al. 2016).

15.2.2 Nanotechnology Application in the Aquafeed Industry

There are numerous potential applications of nanotechnology in feed industry, including: (i) minor modifications of natural ingredients to enhance taste, palatability and sensory improvement such as flavor, color, and texture; (ii) enhancing nutrition quality of foods by stabilizing active ingredients such as nutraceuticals in feed matrices, packaging, and product innovation to extend shelf-life, (iii) increasing bioavailability of essential nutrients (Food Safety Authority of Ireland 2008). Nano-delivery of bioactive/nutrient in feedstuffs or in vivo in fish is enabled through improved knowledge of feed materials at the nanoscale. The different nanomaterials that have the potential to be used for this purpose are nanocomposites, nanoclays, and nanotubes. The nanoproducts that would find applications are nanosensors, nanoimaging, and nanochips and nanofilters. Similarly, the potential nano-delivery systems are nanocapsules, nanocochleates, nanoballs, nanodevices, nanomachines, and nanorobots (Thulasi et al. 2013).

In aquafeed, nanotechnology may also play significant roles in the delivery of micronutrients to aquatic animals. For instance, nanomaterials can be used to coat nutrients that could normally degrade, such as fatty acids, or have limited assimilation efficiency across the gut of fishes, because they are poorly soluble (i.e. fat-soluble vitamins) (Handy 2012). Nanoencapsulation technology has been suggested for vitamins, minerals, carotenoids, and fatty acids, with increasing bioavailability being the main goal (Acosta 2009; Bouwmeester et al. 2009).

Several vitamins and their precursors, such as carotenoids, are insoluble in water. Nevertheless, nanotechnology helps to address these problems. Specifically, when prepared as nanoparticles, these vitamins and their precursors can easily be homogenized with cold water, which enables to increase their bioavailability. For example, Vitamin B_{12} absorption from the gut under physiological conditions occurs via receptor-mediated endocytosis; and the ability to increase oral bioavailability of various peptides (granulocyte colony stimulating factor, erythropoietin) and particles by covalent coupling to vitamin B_{12} has been reported by Russell Jones (2001) and Russell Jones et al. (1999). Vitamin E is a term describing all tocopherol and tocotrienol derivatives, which exhibit the biological activity of alpha tocopherol. Its structure is sensitive to light, heat, and oxygen; consequently, synthetic versions of vitamin E are less expensive, but have lower biological activity (Thulasi et al. 2013).

Nano-micelles made from casein can be used as a vehicle for hydrophobic ingredients such as vitamin D_2 (Semo et al. 2007).

Nanoscale mineral supplements might provide a source of trace metals, without the extensive faecal losses normally associated with mineral salts (e.g. Fe salts; Carriquiriborde et al. 2004). Nanomaterials may also offer an alternative to organic forms of food supplements, where antinutritional factors (incidental pesticides, toxic metals, etc.) in the ingredient can sometimes be a problem (Berntssen et al. 2010).

Nanomaterials can be used to change the physical properties of aquafeed in addition to enhancing the bioavailability and stability of aquafeed. For example, feed wastage and pollution in aquaculture due to poor feed quality (stability, texture or inappropriate buoyancy of the pellet) is a continuing problem (Handy and Poxton 1993); and small supplementations of nanomaterials can significantly alter the physical properties of these pellets. Specifically, the additions of single-walled carbon nanotubes to trout feed can result in a hard pellet that does not fragment easily in water (Handy 2012). Rainbow trout readily eat feed containing nanomaterials up to 100 mg kg^{-1} TiO_2 nanoparticles (Ramsden et al. 2009) and/or 500 mg kg^{-1} C60 and 500 mg kg^{-1} single-walled carbon nanotubes (Fraser et al., 2010) without loss of appetite or growth rate. Therefore, adding a few milligrams of nanomaterial/nanoparticles to aquafeed modify the physical properties of pellets, which could play important roles in the development of aquafeed industry, ultimately sustainable growth of aquaculture industry.

15.3 Nanotechnology Application in Aquaculture Disease Control

Aquaculture sector (especially intensive and super-intensive commercial farms) has grieved major economic losses because of disease outbreaks caused by several pathogenic agents (i.e. bacteria, viruses, and parasites) (Huang et al. 2015; Shinn et al. 2015; Tandel et al. 2017). Traditionally, these pathogens could be treated with chemical disinfectants and antibiotics either through feed, immersion, or injection. However, the use of these chemicals in aquaculture has been criticised, because, they are no longer effective i.e. several pathogenic bacteria i.e. *Aeromonas hydrophila, A. salmonicida, Yersinia ruckeri, Vibrio, Listeria, Pseudomonas*, and *Edwardsiella* species have been reported to be insensitive against most common antibiotics used in aquaculture (Sørum 2008; Swain et al. 2014). In addition, the excessive use of these chemicals in aquaculture could be toxic to other organisms including humans and the environment (Shah and Mraz 2019; Malheiros et al. 2020). This could have paved ways to the search for better alternative technology to control bacteria, viruses, and parasites in aquaculture. Today, nanotechnology has become the new alternative with potential to be used as antimicrobial agents, vaccines, and diagnosis tools for disease causing agents in fish farming (Shaalan et al. 2016).

15.3.1 Nanoparticles as Antibacterial Agents in Aquaculture

Different metal nanoparticles (biologically or chemically synthesized) have been recommended as alternative antibacterial agents, with potential to eradicate or reduce the use of traditional antibiotics in aquaculture (Gunalan et al. 2012; Shaalan et al. 2016) (Table 15.2). Biologically synthesized metal nanoparticles (derivatives of plants, bacteria and fungi) are more advocated over chemically synthesized ones because of their high antimicrobial activity, environmental friendliness, simplicity and affordability (Kalishwaralal et al. 2008; Gunalan et al. 2012; Prasad 2014; Prasad et al. 2016, 2018; Srivastava et al. 2021; Sarma et al. 2021). Some of the antibacterial metal nanoparticles studied in aquaculture include zinc nanoparticles (ZnNPs), silver nanoparticles (AgNPs), copper oxide (CuONPs), gold nanoparticles (AuNPs), and titanium dioxide (TiO_2NPs) (Swain et al. 2014), with AgNPs, ZnNPs, and AuNPs being the widely studied nanoparticles. These nanoparticles could be used either alone or in combination with each other (Venegas et al. 2018). This section reviews the commonly reported metal nanoparticles as antibacterial agents in aquaculture (Table 15.2).

Zinc Nanoparticles (ZnNPs) These nanoparticles are gaining popularity due to their multifunctional properties; antibacterial and antifungal properties (Wang et al. 2008; Di Cesare et al. 2012). Zinc-oxide (chemically synthesized) reportedly showed broad spectrum antibacterial activity against *Aeromonas hydrophila*, *Edwardsiella tarda*, *Flavobacterium branchiophilum*, *Vibrio* sp., *Staphylococcus aureus*, *Bacillus cereus*, and *Citrobacter* sp. (Swain et al. 2014), which are some of the important pathogenic bacteria in aquaculture. Remarkably, ZnO-NPs synthesized with aloe extracts showed high broad antibacterial activity when compared to the chemically synthesized ZnO nanoparticles (Gunalan et al. 2012). Similarly, ZnO-NPs synthesized with *A. hydrophila* showed antibacterial activity against *Enterococcus faecalis*, *Pseudomonas aeruginosa*, *Candida albicans*, *Escherichia coli*, and *Aspergillus flavus* (Jayaseelan et al. 2012). In addition, dietary supplementation of ZnO-NPs reportedly enhanced resistance of *Labeo rohita* (Swain et al. 2019), and *Oreochromis mossambicus* (Anjugam et al. 2018) against *A. hydrophila*. Generally, the antibacterial mechanisms of nanoparticles are fairly understood. The active oxygen species generated by the metal oxide particles is considered the main mode of action, thereby these particles inhibit bacterial proliferation by disrupting the bacterial cell membrane, hence destroying the cell content (Liu et al. 2009; Gunalan et al. 2012; Bhuyan et al. 2015).

Silver Nanoparticles (AgNPs) They have been widely reported to elicit antibacterial activity against a broad spectrum of pathogenic bacteria of economic importance in aquaculture. Silver nanoparticles are reported to inhibit bacterial growth through different mechanisms: Ag+ binds to the bacterial cell membrane proteins resulting in the distraction of the membrane (Lara et al. 2010; Aziz et al. 2014, 2015, 2016, 2019), and by disrupting the cell division or bacteria reproduction process (Huang et al. 2011). A study by Elayaraja et al. (2017) demonstrated that silver nanoparticles synthesised using bacterial cellulose (Ag-NPs-BC) had high

Table 15.2 Some nanoparticles studied as antibacterial agents in aquaculture

Nanoparticles (NPs)	Screened bacteria	Antibacterial activity (Yes/No)	References
Zinc nanoparticles (ZnNPs) Zinc oxide (ZnO) (chemical)	*Aeromonas hydrophila; Edwardsiella tarda; Flavobacterium branchiophilum; Vibrio* sp.; *Staphylococcus aureus; Bacillus cereus; Citrobacter* sp.;	Yes	Swain et al. (2014)
ZnO (chemical) Bulk-ZnO (chemical)	*Vibrio harveyi*	Yes No	Ramomoorthy et al. (2013)
ZnO (Chemical) ZnO-*Aloe vera* (biological)	*Serratia marcescens; S. aureus; Proteus mirabilis; C. freundii*	Yes	Gunalan et al. (2012)
ZnO-*A. hydrophila* (biological)	*Enterococcus faecalis; Pseudomonas aeruginosa; Candida albicans; Escherichia coli; Aspergillus flavus*	Yes	Jayaseelan et al. (2012)
Dietary ZnONPs	*A. hydrophila*	Yes	Anjungam et al. (2018)
ZnO (chemical)	*A. salmonicida; Yersinia ruckeri; Aphanomyces invada*	Yes	Shaalan et al. (2017)
ZnO-Ag (mixture) (chemical)	*Pseudomonas* spp.	Yes	Venegas et al. (2018)
Silver nanoparticles (AgNPs) AgNPs (chemical)	*A. salmonicida* subsp. *Salmonicida* *S. aureus;* *E. coli* O157:H_7; *Streptococcus pyogenes;* *V. fluvialis*	Yes Yes Yes Yes	Shaalan et al. (2018) Ayala-Nunes et al. (2009) Lara et al. (2010) Meneses-Marquez et al. (2019)
Ag-TiO_2(chemical)	*A. hydrophila; E. tarda; F. branchiophilum; Vibrio sp; S. aureus; Citrobacter* sp.	Yes	Swain et al. (2014)
AgNPs-*Citrus limon* (biological)	*E. tarda; S. aureus*	Yes	Swain et al. (2014)
AgNPs-Tea leaf	*V. harveyi*	Yes	Vaseeharan et al. (2010)
AgNPs-*Calotropis gigantea* extracts (biological)	*V. alginolyticus*	Yes	Baskaralingam et al. (2012)
AgNPs-BC (biological)	*V. harveyi; V. parahaemolyticus*	Yes	Elayaraja et al. (2017)
AgNPs-red algae (biological)	*V. harveyi; V. parahaemolyticus;* *V. alginolyticus;* *V. anguillarum*	Yes	Fatima et al. (2020)

(continued)

Table 15.2 (continued)

Nanoparticles (NPs)	Screened bacteria	Antibacterial activity (Yes/No)	References
Gold nanoparticles (AuNPs) Fucoidan-AuNPs (biological)	*A. hydrophila*	Yes	Vijayakumar et al. (2017)
Acanthophora spicifera-AuNPs (biological)	*V. harveyi* *S. aureus*	Yes No	Babu et al. (2020)
Herbal extracts-AuNPs (biological)	*A. hydrophila* *S. agalactiae*	Yes Yes	Fernando and Cruz (2020)
Anacardium occidentale-AuNPs (biological)	*A. hydrophila*	No	
	A. bestiarum	Yes	Velmurugan et al. (2014)
	P. fluorescens *E. tarda*	Yes No	
AuNPs-zeolites	*E. coli; Salmonella typhi*	Yes	Lima et al. (2013)
Nigella sativa essential oil-AuNPs (NsEO-AuNPs)	*S. aureus; V. harveyi*	Yes	Manju et al. (2016)
AuNPs (chemical)	*V. parahaemolyticus*	Yes	Tello-Olea et al. (2019)

Yes = inhibited bacterial growth; No = did not inhibit bacterial growth

bactericidal activity against *V. parahaemolyticus* and *V. harveyi*, which are some of the deadliest bacterial pathogens in shrimp aquaculture. Similarly, biologically synthesized Ag-NPs were recommended as alternative antibiotics in controlling *S. aureus* and *E. tarda* (Swain et al. 2014), and *V. harveyi* infection in *Feneropenaeus indicus* (Vaseeharan et al. 2010). Interestingly, AgNPs demonstrated effectiveness against multi-drug resistant bacteria such as methicillin-resistant *S. aureus* (MRSA) (Ayala-Nunez et al. 2009), ampicillin-resistant *E. coli* O157:H$_7$, and erythromycin-resistant *Streptococcus pyogenes* (Lara et al. 2010). This is indeed an indication that these nanoparticles have the ability to eradicate the use of ineffective antibiotics to fight bacterial diseases in aquaculture.

Gold Nanoparticles (AuNPs) They are one of the emerging nanoparticles, and they can be more preferred mainly because of their less toxicity to animals (Li et al. 2014). Different gold nanoparticles have been reported to possess antibacterial properties, with the potential to eliminate bacteria responsible for huge production and economic losses in aquaculture (Table 15.2). A study by Vijayakumar et al. (2017) demonstrated that fucoidan (marine polysaccharide)-coated gold nanoparticles (Fu-AuNPs) inhibited the biofilm of *A. hydrophila*, and reduced mortality in *A. hydrophila*-infected *Oreochromis mossambicus* juveniles. *Acanthophora spicifera* (marine red algae)-mediated gold particles (As-AuNPs) exhibited the highest antibacterial activity against *V. harveyi* than *S. aureus* (Babu et al. 2020). Gold nanoparticles reportedly act against bacterial pathogens via a number of pathways such as their ability to collapse the bacterial membrane potential, inhibit ATPase activities, and subsequently the ATP level; and inhibit the subunit of ribosome from binding tRNA (Cui et al. 2012). In addition, AuNPs synthesized with crude herbal extracts reportedly inhibited *A. hydrophila* biofilm formation via the disruption of their quorum sensing ability (communication between cells) (Fernando and Cruz 2020). The communication between bacterial cells has been the target to control bacterial virulence for promising antibacterial agents (Rasmussen et al. 2005).

15.3.2 Nanoparticles as Vaccine/Drug Delivery Vector

In aquaculture, drugs are traditionally administered through feed, injection, or immersion. The traditional drug delivery methods are considered to be ineffective for several reasons such as poor bioavailability and absorption of the drugs to the targeted cells (Moges et al. 2020). Recently, the use of nanoparticles in drug formulation and delivery has gained attention in the fight against pathogens in aquaculture (Table 15.3). With this technology, the compound of interest (i.e. antibiotics, vitamins, vaccines, probiotics) is encapsulated into a compound of the nanoscale, thereby increasing absorption of the compound to targeted region, because nanoparticles are able to penetrate through cellular barriers (Sivakumar 2016; Moges et al. 2020); hence better protection against pathogens compared to traditional drug delivery methods.

Chitosan (Chit.) (polysaccharides) and poly-lactic glycolipids acid (PLGA) (copolymer) nanoparticles are the widely studied nanoparticles for drug delivery. These nanoparticles are commonly used due to their outstanding physiochemical properties such as biocompatibility, bioactivity, non-toxicity, and biodegradability (De Jong and Borm 2008; Lü et al. 2009). Chitosan nanoparticles combined with infectious salmon anaemia virus (ISAV) gene as an adjuvant were used to develop a DNA vaccine to control ISAV in Atlantic salmon culture (Rivas-Aravena et al. 2015). Chitosan nanoparticles-based vaccine was developed for *Lates calcarifer* against *V. anguillarum* (Rajesh Kumar et al. 2008). In addition, PLGA nanoparticles loaded with rifampicin were reported to show efficacy against *Mycobacterium marinum* in zebra fish larvae (Fenaroli et al. 2014). The use of chitosan and PLGA nanoparticles in combination were also reported in aquaculture. For instance, a plasmid DNA vaccine (pDNA) combined with PLGA and chitosan nanoparticles complex (pDNA-PLGA-Chit-NPs) significantly activated immune parameters in *Labeo rohita* and increased their survival after *Edwardsiella tarda* infection (Leya et al. 2020). This is said to be attributed to the ability of the complex vaccine to act synergistically to provide the host with amplified protective immunity against pathogens (Leya et al. 2020).

Table 15.3 Some chitosan and Poly lactic-co-glycotic acid nanoparticles studied as drug delivery agents in aquaculture

Nanoparticles (Chit-PLGA)	Pathogens	Fish species	References
pDNA-PLGA-Chit-NPs pDNA-PLGA-NPs PLGA-NPs Chit-NPs	*Edwardsiella tarda*	*Labeo rohita*	Leya et al. (2020)
Chit-ISAV	Alphavirus	Atlantic salmon	Rivas-Aravena et al. (2015)
Chit-DNA (pVAOMP38)	*Vibrio anguillarum*	*Lates calcarifer*	Rajesh Kumar et al. (2008)
Chit-DNA (pEGFP-N2OMPK, pDNA)	*V. parahaemolyticus*	*Acanthopagrus schlegelii* Bleeker	Li et al. (2013)
Chit-inactivated *E. ictaluri* and infectious spleen and kidney necrosis virus.	*E. ictalurid*	*Pelteobagrus fulvidraco; Siniperca chuasi*	Zhang et al. (2019) Zhu et al. (2019)
Chit-*Piscirickettsia salmonis* membrane	*Piscirickettsia salmonis*	*Dario rerio*	Tandberg et al. (2018)
PLGA-rifampicin	*Mycobacterium marinum*	*Dario rerio*	Fenaroli et al. (2014)

Chit Chitosan, *PLGA* Poly lactic-co-glycotic acid, *NPs* Nanoparticles, *p* plasmid; *ISAV* Infectious Atlantic salmon anaemia virus

15.4 Nanotechnology Application for Water Quality Management in Aquaculture

In aquaculture, animals are fed with high-protein feeds, and fertilizers, especially in semi-intensive systems, are used to stimulate natural feeds to sustain the growth of farmed animals and stimulate production. However, the challenge is in the handling/management of uneaten feed and waste products, which often contribute to the culture water quality (Ninh et al. 2016). Consequently, water in poorly managed aquaculture systems may be enriched with nutrients and organic and suspended matter (Boyd 2001; Sikder et al. 2016) which are associated with negative effects on fish growth, increased fish stress, and high risks of infectious diseases (Boyd and Tucker 1998; Boyd 2001). Water quality management is, therefore, vital for aquaculture operations.

Contrary to conventional wastewater treatment methods such as chemical treatment, filtration, and ion exchange (Muzammil et al. 2016), aquaculture effluents are treated via sedimentation, constructed wetlands, and water treatment reservoirs (Boyd 2001; Kerepeczki et al. 2011). However, these techniques are said to be ineffective in the complete removal of contaminants (Le et al. 2019). Therefore, the use of nanomaterials has been recommended as the best alternative technology in the purification of water either for human consumption (Gehrke et al. 2015) or fish culture (Sichula et al. 2011). This section outlines the use of nano-catalysts and nano-adsorbents for wastewater treatment in aquaculture.

15.4.1 Nanocatalysts and Nanoadsorbents in Aquaculture

Nano-catalysts are being employed in wastewater treatment for the chemical oxidation of organic and inorganic pollutants (Muzammil et al. 2016). Titanium oxide (TiO_2) and Zinc oxide (ZnO) are some of the widely used nanoparticles in photocatalysis. Their efficiency depends on the interaction with light energy and presence of metallic nanoparticles/semi-conductor metals (Acheampong and Antwi 2016). For instance, titanium oxide (TiO_2) was reported to remove bacterial cells (Litter 2015). This is because, TiO_2 possess high antimicrobial abilities that permits its use in inactivating pathogenic organisms such as bacteria found in wastewater (Wu et al. 2014; Amin et al. 2014). In another study, TiO_2 was reported to reduce the viability of several waterborne pathogens such as protozoa, fungi, *E. coli*, and *P. aeruginosa,* after 8 hours of simulated solar exposure (Amin et al. 2014). In addition, titanium was able to remove heavy metals such as chromium and arsenic from wastewater (Litter 2015).

A study by Le et al. (2019) tested the removal of heavy metal ions using rod-shaped ZnO particles under utraviolet light and visible light. This study observed that ZnO nanoparticles could remove heavy metal ions such as Cu(II), Ag(I) and Pb(II) at an efficiancy rate greater than 85%, but not very efficient at removing Cr(VI), Mn(II), Cd(II), and Ni(II) ions, regardless of the light source used. Similar to $TiO_{2,}$ ZnO nanoparticle produced by solution combustion method (SCM) has also demonstrated effectiveness in removing *E. coli* from water (Masoumbaigi et al. 2015). Another important nano-catalyst in wastewater treatment is nanosilver. These nanoparticles synthesized with fungal species have been reported to remove *E. coli, Staphylococcus* sps, and *Pseudomonas* sps in wastewater (Moustafa 2017), which are some of the pathogenic bacteria in aqauculture as demostrated above in Sect. 15.3.

In addition to nanocatalysts, nanoadsorbents are likewise impressive water treatment methods, used to remove heavy metals, nutrients, and microbes from water (Thines et al. 2017). One such nanoadsorbent is activated charcoal (AC). A study by Aly et al. (2016) and Sichula et al. (2011) indicated that AC successfully removed ammonia from aquaculture production systems and reduced unionized ammonia concentrations in *O. niloticus* culture respectively. The application of nanocatalysts and nanoadsorbents is, therefore, a promising approach for the management of water quality in aquaculture production systems. By adopting nanotechnologies, microbial and heavy metal contamination can be adressed in aqaculture and in so doing, manage the challenges of nutrient accumulation, ultimately disease proliferation.

15.5 Conclusion and Future Perspectives

Challenges associated with increased intensification in aquaculture such as poor feed quality and utilization, increased disease outbreaks, and poor water quality cannot be overemphasised. This chapter has provided substantial evidence that nanomaterials have the potential to enhance feed quality (nutritional and physical properties), feed utilization, drug formulation and delivery, disease treatment, and water quality management in aquaculture; hence improving fish growth and better economic return. Despite promising research findings, the speed of implementation of this technology in aquaculture is still limited. One of the limitations is the complex manufacturing process of nanomaterials, which requires expensive equipments and services, and this could directly influence the cost of nanoproducts. Therefore, small-scale fish farmers may be financially limited to participate in the manufacturing process of the nanomaterials. Another limitation is that some nanoparticles, particularly the chemically synthesized ones, are toxic to animals at higher dosages and may negatively affect the development of animals (Verma et al. 2017, 2018).

Moving forward, there is a need to adopt nanoparticles manufacturing approaches such as biological methods, which are described to be simple and produce nontoxic, environmentally friendly, and affordable products (Kalishwaralal et al. 2008; Prabhu and Poulose 2012; Thangadurai et al. 2020, 2021; Maddela et al. 2021). This way, aquaculture farmers at all levels (from small scale to commercial) would be able to harness the benefits associated with nanotechnology. All in all, nanotechnology is playing important roles in the sustainable development of aquaculture.

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