Concept of microbial bioremediation in aquaculture wastes; Review

S. N. Musyoka

Musyokasonia@yahoo.com

Tutorial Fellow, Department of Fisheries Management & Aquaculture Technology,

South Eastern Kenya University, Kitui, Kenya

Abstract

The past two decades have seen great intensification of the aquaculture industry. Currently aquaculture production accounts close to 50% of the consumption of fish worldwide. This substantial growth in the aquaculture sector is greatly attributed by the increase in fish demand, declining fisheries, improved technologies among others. To meet this demand, there have emerged intensive culture practices which are associated with both an uncontrolled use of feed and a massive production of waste. For this reason, the aquaculture industry is currently considered to produce an adverse and negative impact in the environment. Most governments have establishmentstrict regulations dealing with the discharge of untreated aquaculture wastes to the environment. Given the risks associated to the use of antibiotics in general and in fish farms in particular, bioremediation therefore remains the most health and efficient way of treating aquaculture waste. Several studies have identified microorganisms as the paramount bioremediators, as they are able to completely remove the wastes generated by the fish-farm facilities. This paper reviews several concepts under which micro-organisms bioremediate aquaculture wastes.

Key words; Bioremediation, wastes, microorganisms, organic, fish farm.

I. INTRODUCTION

Aquaculture waste treatments have been debated widely by scientists, environmentalists and politicians. The health risks associated with the use of chemicals and antibiotics to treat wastes, heightened restrictions on their use and limitations on vaccination and chemotherapy. It is widely accepted that, there is a need to create sustainable aquaculture which will continue its production to cater for the fish demand without having adverse effects on the environment or human health. Several regulations have been set up to control the quality of the discharge derived from aquaculture to the environment. The Environmental Protection Agencies (EPA) has set the effluent discharge guidelines that restrictthe amount of feed or water released to each individual farm, offering management practices and measures for compliance [1].

The risks and impacts from aquaculture waste generate great scientific interests [2; 3; 4] and great advocacy for undertaking mitigation measures [5; 6; 7]. Most studies and research has been focusing on exploring environment friendly, health, efficient and cost-effective methods for improving the quality of aquaculture waste before it is released to the environment [8]. Bioremediation is generally defined as the use of organisms capable to degrade contaminants through their metabolic activity, to solve environmental problems, such as those produced by pollution [9; 10]. The organisms or organism-derived compounds used for this end are known as Bioremediating Agents or Bioremediators [11].

Bioremediation is considered an ancient concept, as the Romans at around 600 B.C. used it for treating wastes. It has been, since then, continually improved, and the use of natural occurring microorganisms and their metabolic activities, has been extended to the use of genetically modified microorganisms with wider metabolic capabilities, and/or the use of the specific enzymes involved in the degradation processes [12; 13]. Indeed, the increasing numbers of known genes that code for contaminant-degrading enzymes are being used to redesign genetically modified microorganisms. This is achieved through the currently available genetic engineering techniques that can act in nature as biosensors and bioreporters for recognizing pollutants [14].

II. RESULTS AND DISCUSION

The use of bioremediation in aquaculture started with the establishment, by different Governments, of strict regulations dealing with the discharge of untreated aquaculture wastes to the environment [14]. These regulations became necessary with the onset of the knowledge about: i) the risks associated to the use of antibiotics in general and in fish farms in particular; and ii) the inorganic particulate matter, living and dead particulate organic matter, dissolved organic matter, ammonia, nitrite, nitrate, phosphate and other potential contaminants that are present in the effluents from aquaculture facilities. Since then, a high number of studies have been developed to search for the most suitable organisms for cleaning the aquaculture effluents. These studies have identified the microorganisms as the best bioremediators, as they are able to completely remove the wastes generated by the fish-farm facilities [15; 16].

Microorganisms can remove the contaminants through different processes such as uptake, adsorption and biodegradation [17]. Particularly, bacteria, among the many diversified groups of microorganisms, have been proved to play a central role in the degradation of fish-farm-derived contaminants [18]. Their functioning in the aquaculture facilities, by simply developing their natural metabolic activities, brought the idea of using them as the main remediating agents for aquaculture wastes [19, 14].

Microorganisms are grouped as autotrophs and heterotrophs; depending on the source of carbon they need for both living and reproducing. The autotrophs, which can be photolitotrophs (those that use the solar light as source of energy) or chemiolitotrophs (those that use the cellular transference of electrons as a source of energy), fix the inorganic carbon (CO_2) present in the environment to generate organic carbon. They are catalogued, therefore, as primary producers of the food chain [20]. As they adsorb and transform the soluble biologically available phosphorus and nitrogen during their own growth process, they play a focal role in the removal of these chemicals, which are highly present in the formula of the nutrients that are used as food in aquaculture facilities and then in their wastes [21; 22]. The heterotrophic degraders destroy and transform/immobilize the non-living organic matter to produce carbon for building their own cells. In return, the cells can act as electron donors catalysing the oxidation of these organic chemicals. The concept applied involves;

A. Bioremediation of the organic compounds

Aquaculture wastes contain, among other chemicals, dissolved and suspended organic compounds. Microbial degraders require organic matter for their own growth [17]. Those that carry the genetic information for oxidizing organic matter, breakdown aquaculture wastes by their metabolic capabilities and transform them, either in a less reduced organic form or in the inorganic compounds CO_2 and H_2O, the end products of the complete oxidation process or mineralization, thus restoring the contaminated sites and controlling further pollution [22].

Bacteria belonging to the gram positive genus *Bacillus* (e.g. *Bacillus subtilis*, *B. licheniformes*, *B. cereus* and *B. coagulans*) and of the genus *Phenibacillus* (e.g. *Phenibacillus polymyxa)* have been known to efficiently breakdown carbonaceous organic matter to CO_2 by using enzymes that help to breakdown proteins and starch to small molecules [23]. However, they are not usually found in the water column but in the sediment, which is their natural habitat. Oxidation of organic matter is aided in this habitat by the activity of burrowing animals such as prawns and shrimps which help oxygen penetration to the pond sediment. The microbial activity drives to the production of microbial biomass, cellular protein, inorganic nutrients and CO_2. The inorganic nutrients and CO_2 can be then used by the phytoplankton while the biomass and the cellular protein are eaten by zooplanktons, prawns, shrimps and fishes [24]. In the pond sediment, the microbial biomass bind with other dissolved matter to form detritus which is food source to the aquatic fish [25].

B. Bioremediation of nitrogenous compounds

The nitrogen compounds (ammonia, nitrite and nitrate) have adverse effects on aquatic ecosystem. The nitrogen compounds in aquatic ecosystem undergo an endless loop, which involves several phases. The first phase is the accumulation of ammonia from metabolic excretion, uneaten food, dead organisms, shell moults, mineralization of the organic matter present in the sediment, and through molecular diffusion from reduced sediment. The ammonia nitrogen compounds are removed by autotrophic nitrification and sometimes by heterotrophic nitrification. The genus *Nitrosomonas, Nitrosovibrio, Nitrolobus, Nitrococcus, Nitrosococcus, Nitrospira* and *Nitrobacter* are among the main autotrophic bacteria genera, mostly responsible for the nitrification process [26]. Some other microorganisms are also involved in the nitrification process, such as heterotrophic bacteria [27; 28], as well as some fungi such as

Aspergillus flavus [29]. In the first step of the nitrification process, ammonia is converted to hydroxylamine, then to nitrite through a complex process using hydroxylamine oxidoreductase and ammonia monooxygenase enzymes [30]. The nitrite formed is the second phase of the nitrogen oxidation cycle, and it is further oxidized to nitrate by nitrite oxidoreductase enzyme [31].

Denitrification is the third phase in which anaerobic bacteria reduces the accumulated nitrate to harmless nitrogen gas which bubbles out of the pond system. It is done by microbial bacteria possessing gene which encode nitrate, nitrite, nitric oxide and nitrous oxide reductase metalloenzymes, which facilitate the process of nitrate reduction to produce molecular nitrogen [32; 33]. This denitrification process takes place under low oxygen conditions, and it is primarily done by various heterotrophic bacteria including *Paracoccus denitrificans* and various species of *Pseudomonas* [34].

The last phase in the nitrogen cycle is the use of the accumulated nitrate by the bacteria, algae and aquatic plants for photosynthesis, thus reducing its accumulation. In return, they are either consumed directly by fish or are first consumed by zooplanktons which are then consumed by fish. When the fish excretes their faeces, the nitrogen component cycle starts again. Sometimes the cycle ends before denitrification, as the nitrate produced by nitrification process is absorbed by the microbes and phytoplanktons thus limiting nitrate for denitrification process.

C. Bioremediation of Hydrogen Sulphide (H_2S)

Heavy feeding in aquaculture ponds leads to excessive accumulation of organic detritus causing severe and prolonged anoxia conditions in sediment bottoms. In aerobic conditions, the organic sulphur decomposes to sulphide, which can then be oxidised to sulphate. This is highly soluble in water, and therefore it gradually disperses from sediments. This process is carried out by different microorganisms. Under anaerobic conditions, sulphate can be used as terminal electron receptor of microbial metabolism, leading to the production of hydrogen sulphide gas, which can be anaerobically metabolised by the photosynthetic purple and sulphur bacteria [23].

D. Bioremediators as biocontrol agents

Bioremediation of aquatic diseases can be completed by some group of bacteria mostly referred to as 'friendly' bacteria. These bacteria are applied as a probiotics to improve the health of the cultured animals. Studies have proven that friendly bacteria produce antimicrobial effect against pathogenic microorganisms, hence favouring the health of the host [35]. These friendly bacteria produce antibiotics, siderophores, bacteriocins, hydrogen peroxide, organic acids and enzymes such as lysozymes and proteases [36], which may have bactericidal or bacteriostatic effect on pathogenic bacteria. They also alter the pH of the surrounded environment thus inhibiting the proliferation of pathogens [37].

In addition, some algal species, such as those belonging to the genus *Tetraselmis*, produces bacteriocins and organic acids which control the growth of some bacterial pathogens, hence preventing the disease [38].

The competition among microorganisms can be also used as biocontrol of pathogens. Different microbes have different natural affinity towards hydrocarbons, which create interference and competitive exclusion of over some others. This could be used to control pathogenic bacteria [22].

Some bacteria act by competing for both nutrients and adhesion sites in the fish intestine, which is necessary for pathogen survival, or have barrier effect and colonization resistance [20]. Reference [11] used some strains of *Bacillus* Spp. to decrease the proportion of luminous *Vibrio* Spp. in the sediments of the ponds in which shrimps were cultured. His results showed that *Bacillus* spp. had an inhibitory activity over *Vibrio* spp., thus helping to increase the shrimp survival in the ponds. Protease and lipase, as well as vitamins produced by other bacteria, aid in the digestive processes of the cultured species, promote their growth, reduce the stress, and increase the reproduction of aquatic animals [33; 39; 40; 41]. Probiotics are for long known to improve and modify the intestinal microbiota balance when given as food supplement [41].

Most commonly used probiotic bacteria are *Lactobacillus, Carnobacterium, Vibrio alginolyticus*, *Bacillus* and *Pseudomonas* [42].

III. CONCLUSION

Biological remediation has been proposed by many authors as the most probable environmental safe alternative for waste treatment. Bioremediation is considered to be an aesthetically pleasing, relatively cost effective alternative, which in addition, is easy to be implemented and maintained, can be performed on-site and/or off-site, and reduces the amount of waste to be land filled.

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