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Sustainable Aquaculture through Recycling of Waste Nutrients using Biofloc Technology

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Abstract

Development of a sustainable aquaculture industry is a challenging task particularly due to the limited availability of natural resources as well as the impact of pollutants in the environment. With these limitations in mind, the development of sustainable aquaculture should focus on the conceptualization of systems aiming high productivity and profitability with minimal utilization of resources including water, space, energy and eventually capital costs. To overcome these challenges, the best environmentally acceptable “Biofloc technology” (BFT) has been developed. BFT is mainly based on the principle of minimal or zero water exchange which recycles the waste nutrients, in particular nitrogen, into microbial biomass that can be used by the cultivable animals. Heterotrophic microbiota are stimulated to grow by steering the C/N ratio in the water through the modification of the carbohydrate content in the feed or by the addition of an external carbon source in the water, so that the bacteria can quickly assimilate the waste ammonia for new biomass production. Hence, ammonia-N can be maintained at a low and nontoxic concentration so that water replacement is no longer required. BFT has been applied to culture various shrimps and fin fishes. Not all species are candidates to BFT. Desirable characters are filter feeding habit, omnivorous, and digestive system adaptable to assimilate the microbial protein. Thus, this technology serves as environment friendly approach reducing pollution, water resources, improving biosecurity, etc. The present paper addresses the biofloc technology and its potential uses to achieve sustainable aquaculture.

Key words: Sustainable aquaculture, biofloc technology, filter feeders, environment friendly.

1.0. Introduction

Aquaculture continues to grow faster than other major food production sectors in the world (FAO, 2018). It has been developing, expanding and intensifying in almost all the regions of the world. However, its sustainable development was challenged by limited sources of land, water, environmental and social issues. As a result, the aqua industry has come under scrutiny for contribution to environmental degradation and pollution. Extensive method of finfish or shellfish farming is practiced in a natural habitat with little inputs and no supplementary feeding has minimum impact on the environment. However, high-quality artificial feeds and chemicals are

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used in semi-intensive and intensive culture practices (Arvanitoyannis and Kassaveti, 2008) are lead to the production of large quantities of solid and nutrient wastes into the environment. Generally, the effluents discharged from finfish/shellfish farming mainly contain nitrogen and phosphorus which cause serious environmental problems. Hence, to reduce the impact of wastewater on the environment, more ecologically sound management and culture practices are necessary (Emerenciano et al., 2013). Fishes excrete nitrogenous waste products by diffusion and ion exchange through the gills, urine and feces (Turcios and Papenbrock, 2014). Ammonia is the chief excretory product of fishes and is toxic at higher concentrations. Due to the toxicity of ammonia and nitrite, and the chance of hypertrophication of the environment by nitrate, the decomposition and reuse of these nitrogenous compounds using a suitable wastewater treatment technology is especially important in aquaculture (Brown et al., 1999). The development of wastewater treatment technology in aquaculture industry will minimize the ecological and social problems, and provide greater long-term economic safety for operation of the industry (Doupe et al., 1999). "Biofloc Technology" (BFT) is an alternative and gaining its own importance as eco-friendly aquaculture system since the nutrients are continuously recycled and reused (Emerenciano et al., 2013; Martinez et al., 2017). In addition, BFT will reduce the pollution of pond water by lowering the concentration of toxic ammonia, nitrite and hydrogen sulphide and improves the water quality under zero/minimal water exchange system (Luo et al., 2014). Hence, the technology has recently gained attention as a sustainable method to control water quality, with the added value of producing proteinaceous feed *in situ* (Crab et al., 2012). Hence, the present attempt has been made to give an overview of the biofloc technology and its beneficial effects to achieve sustainable aquaculture.

2.0. Biofloc:

Biofloc is an aggregate of heterogenous bacteria, algae, fungi, protozoans, metazoans, copepods, particulate organic matter such as uneaten feed, faeces and detritus (Crab et al., 2009; Ballester et al., 2010; Ray et al., 2011; Hargreaves, 2013 and Ahmad et al., 2017). All these are held together in a matrix by electrostatic attraction or by the mucus secreted by bacteria (Hargreaves, 2013). Typical flocs are irregular have a broad distribution of particle size, are fine, easily compressible, and highly porous and are permeable to fluids (Chu and Lee, 2004). Bioflocs are generally microscopic in size, mostly around 50-200 μ m and easily settles in calm water, though the typical bioflocs could be larger and visible to naked eye (Hargreaves, 2013). Moreover, they have low sinking rate, keep floating on water thus increasing the opportunity to derive nutrients.

2.1. Principle of Biofloc Technology (BFT):

BFT is the technique of enhancing water quality through the addition of external carbon to the aquaculture system (Crab et al., 2012). The basic principle of the technology is to recycle nutrients and nitrogenous wastes by maintaining a high carbon/nitrogen (C/N) ratio in the water to stimulate the growth of heterotrophic bacteria (Avnimelech, 2006). BFT is based on the maintenance of high levels of microbial floc in suspension by constant aeration and addition of carbohydrate source that allows aerobic decomposition of the organic material (Avnimelech and



Weber, 1986). By the addition of carbohydrates, growth of heterotrophic bacterial is stimulated and microbial protein production takes place through nitrogen uptake (Avnimelech, 1999). Thus, the technology is efficient and environment-friendly as the nutrients are recycled and reused within the system itself (Emerenciano et al., 2013).

2.2. Types of BFT:

Depending upon the constituents, two types of biofloc systems have been used in commercial aquaculture. The systems that are exposed to natural light in outdoor, lined ponds or tanks for the culture of shrimp or tilapia, and lined raceways for shrimp culture in greenhouses are called “Extensive or green-water” biofloc systems, where a complex mixture of algal and bacterial processes control water quality. Most biofloc systems in commercial use are green-water (Hargreaves, 2013). The other biofloc systems (raceways and tanks) which are operated in closed buildings with no exposure to natural light are called “Intensive or brown-water” biofloc systems, where only bacterial processes control water quality.

2.3. Adaptable Species for BFT:

Species selection is one of the prime criteria for the successful use of biofloc technology as it cannot be applied to all aquatic species (Avnimelech, 2007). Desirable characters of a species for selection are tolerance to high solids concentration and poor water quality, and physiological adaptations that allow them to consume biofloc and digest microbial protein (Milstein et al., 2001; Hargreaves, 2013). BFT has been used successfully in culturing different crustacean species such as Pacific white shrimp, *Litopenaeus vannamei* (Khanjani et al., 2017), pink shrimp, *Farfantepenaeus duorarum* (Emerenciano et al., 2014), red shrimp, *Farfantepenaeus pauliensis* (Emerenciano et al., 2011), tiger shrimp, *Penaeus monodon* (Shyne et al., 2017), giant freshwater shrimp, *Macrobrachium rosenbergii* (Prajith and Madhusoodana, 2011), and Finfish such as Pacu, *Piaractus brachypomus* (Poleo et al., 2011), golden carp, *Carassius auratus* (Castro et al., 2016), Bocachico, *Prochilodus magdalenae* (Pertúz-Buelvas et al., 2016), African cichlid, *Pseudotropheus saulosi* (Harini et al., 2016), tilapia, *Oreochromis niloticus* (Luo et al., 2017), *Oreochromis mossambicus*, *Oreochromis andersonii* (Day et al., 2016) and channel catfish, *Ictalurus punctatus* (Green & McEntire, 2017).

2.4. Nutritional quality and measurement of biofloc:

Biofloc contributes nutritional standards and serves as an aquaculture feed. The nutritional value depends on different factors such as food preference, floc density, and capability of the animal to ingest and digest microbial protein (Hargreaves, 2006). Research has shown that the nutritional properties of the flocs are influenced by the type of carbon source used to produce the flocs and the capacity of the technique to control the water quality in the culture system (Crab, 2010). Different organic carbon sources stimulated the growth of specific bacteria, protozoa and algae and hence influenced the microbial composition and community organization of the bioflocs and their nutritional properties (Crab, 2010). Biofloc has 38% protein, 3% lipid, 6% fiber, 12% ash,



and 19 kJ/g energy (on dry matter basis) (Azim and Little, 2008). Azim and Little (2008) reported 50% crude protein, 2.5% crude lipid, 4% fiber, 7% ash, and 22 kJ g⁻¹ energy and revealed that the quality of biofloc is does not depend up on the quality of feed used for biofloc production (35 and 22% crude protein). Ballester et al. (2010) reported that when carbon sources like wheat bran and molasses were used, bioflocs contain 30.4% crude protein, 4.7% crude lipid, 8.3% fiber, 39.2% ash and 29.1% nitrogen free extract on dry matter basis. Thus, the nutritional composition and quality index of the floc changes on the basis of the carbon source added. Besides these characteristics, the type of carbon source also influences the palatability and digestibility of the cultured organisms (Crab et al., 2009; Crab, 2010). Generally, biofloc can be measured by using imhoff cones, desirable volume ranges between 1-40 ml/l for fish and 2-15 ml/l for shrimp culture (Suneetha et al., 2018). Besides the volume, the colour, physico-chemical properties and nutritional value have also been used to characterize the floc (Avnimelech, 1999).

2.5. Carbon and Nitrogen Ratio:

In the aquatic environment Carbon-nitrogen ratio (C/N) plays an important role in the immobilization of toxic inorganic nitrogen compounds into useful bacterial cells (single-cell protein) acts as a direct food source for the cultured organisms (Avnimelech, 1999). The alteration in the C/N ratio may result in a shift from an autotrophic to a heterotrophic system (Avnimelech, 1999; Browdy & Bratvold, 2001). As the C/N ratio of bacterial cells is 5:1 (Rittmann & McCarty, 2001) and the conversion efficiency of bacteria is 40–60%, C/N ratio of 10 or more is required for the growth of heterotrophic microorganisms (Avnimelech, 1999). The carbon sources used in BFT are the by-products which are derived from human and/or animal food industry. The selection of the carbon sources can be done by considering the cost, local availability, biodegradability and efficiency to support bacterial community (Emerenciano et al., 2013). Generally, wheat flour, starch, cellulose, cassava meal, corn flour, sorghum, molasses can be used as carbon sources (Avnimelech, 1999; Emerenciano et al., 2013).

3.0. Mechanism of wastewater treatment:

A major task of water quality management in any aquatic animal production system is maintaining ammonia concentration below toxic levels. In biofloc systems, the three main processes that control ammonia are i) algal uptake ii) bacterial assimilation and iii) nitrification.

3.1. Algal Uptake:

Any nutrient rich aquatic system when exposed to sunlight is prone to develop a dense algal bloom. Nutrients are enriched in the water by the addition of fertilizers and decomposition of organic matter in the form of faecal solids, uneaten feed, dead organisms, etc. These nutrients are readily taken up and stored in the algal cells resulting in bloom formation. The rate of algal uptake also depends on the intensity of light. Generally, at daily feeding rates less than 300 kg/ha (30 g/m²), algal activity is the major factor controlling water quality (Hargreaves, 2013).



3.2. Bacterial Assimilation:

In intensive biofloc system, supplementary carbohydrate source is added to stimulate the growth of heterotrophic bacteria. By the manipulation of C:N ratio, heterotrophic bacteria demand for nitrogen in the form of ammonia because organic carbon and inorganic nitrogen are generally taken up in a fixed ratio depending up on the requirement of the bacterial cell. Thus, ammonia is immobilized and packed as protein in the bacterial cell. This microbial protein in the form of heterotrophic bacteria in flocs can serve as a supplemental source of nutrition for fish and shrimp.

3.3. Nitrification:

The oxidation of ammonia to nitrite and then to nitrate is called nitrification. Ammonia is converted into nitrite and then to nitrate by the nitrifying bacteria. So, waste nitrogen is repeatedly cycled between dissolved ammonia and solids of algae or bacteria. If solids are removed in the form of sludge, a significant amount of nitrogen can be taken out of the system. The dynamics of ammonia in biofloc systems are complex, involving interplay among the algae and bacteria that compete for ammonia. Thus, there will be less ammonia present in the system.

4.0. Potential uses of BFT:

4.1. Zero or minimal water exchange:

A major challenge in aquaculture system is the requirement of periodical water exchange to maintain water quality and the resulting huge nutrient and organic matter discharge (Moss et al., 2001). The sustainable approach of this system is based on the minimum or zero water exchange. As a closed system, BFT has an advantage of releasing minimum water into rivers, lakes and estuaries containing escaped animals, nutrients, organic matter and pathogens. Also, surrounding areas are benefitted by the “vertically growth” in terms of productivity, preventing coastal or inland area destruction, induced eutrophication and loss of natural resources. In conventional methods, drained water from harvested ponds and tanks often contain relatively high concentrations of nitrogen and phosphorous as well as limiting nutrients that induce algae growth, which may cause severe eutrophication and anaerobic conditions in natural waterbodies. In BFT, minimum water discharge and reuse of water prevent environmental degradation and make the system “environment friendly” with a “green” approach (Suneetha et al., 2018). Minimum water exchange maintains the heat and prevent temperature fluctuations (Crab et al., 2009) and allowing growth of tropical species in cold areas. As there is a lot of scarcity for water, the application of biofloc technology may significantly reduce the quantity of water used in aquaculture. An intensive zero exchange lined shrimp pond requires water of 1–2.26 m³ kg⁻¹ shrimp, whereas a conventional system with regular water exchange may require water up to 80 m³ kg⁻¹ (Hargreaves, 2006). In addition, Luo et al. (2014) noted that water consumption of biofloc-based tilapia culture system was 40% lower than that of recirculating aquaculture system (RAS).



4.2. A tool for nutrient recycling and water quality management:

Maintaining and monitoring water quality parameters such as temperature, dissolved oxygen, pH, salinity, dissolved and suspended solids, alkalinity, etc. are essential in sustainable aquaculture. Microorganisms present in the biofloc play a key role in the maintenance of good water quality. By maintaining a high C:N ratio, nitrogenous by-products are taken up by heterotrophic bacteria. The understanding of water quality parameters and their interactions in biofloc are crucial in the maintenance of the production cycle (Emerenciano et al., 2017). The macroaggregates (biofloc) are rich in proteins and lipids which are available “*in situ*” 24 hours per day (Avnimelech, 2007). In the water column, a complex interaction occurs between organic matter, physical substrate and large range of microorganisms such as phytoplankton, free and attached bacteria, aggregates of particulate organic matter and grazers such as rotifers, protozoan ciliates and flagellates, and copepods (Ray et al., 2010). They play an important role in recycling of nutrients and maintaining the water quality (McIntosh et al., 2000; Ray et al., 2010). Bacteria and other microorganisms act as very efficient “biochemical systems” to degrade and metabolize organic residues (Avnimelech, 1999). They recycle both organic and inorganic matter (un-consumed and non-digested feed, metabolic residues and carbon sources) into new microbial cells.

4.3. Enhances the feed utilization efficiency:

In situ utilization of microbial flocs generated in biofloc systems and their utilization as a feed ingredient has been well documented (Kuhn et al., 2009; Anand et al., 2014). Ju et al. (2008) demonstrated that the concentrations of free amino acids such as alanine, glutamate, arginine and glycine, which are known attractants in shrimp diet (Nunes et al., 2006), are present in bioflocs. Da Silva et al. (2013) reported that the application of biofloc technology on Pacific white shrimp super-intensive culture considerably enhanced N and P utilization efficiency upto 70% and 66% respectively, relative to conventional intensive culture systems with regular water exchange. Biofloc technology increases the feed utilization efficiency and reduces the feed cost. It increases the feed quality and feeding strategy in a way that the nutrients can be efficiently delivered and finally utilized. It reutilizes the nutrient waste through modifications in the culture system. The nutrient waste that was generated from unconsumed feed, digestion and other metabolic processes was converted into microbial biomass that may eventually consumed by the cultured animal itself or other animal as their food source. As the floc volume increases, the amount of feed to be supplied to the animal gradually decreases thus feed can be reduced to 30% of conventional feeding ration due to biofloc consumption in shrimp (Avnimelech et al., 1994). Burford et al. (2004) reported that more than 29% of daily food consumed for *L. vannamei* could be biofloc. Avnimelech et al. (1994) estimated that feed utilization in tilapia is higher in BFT than conventional water-exchange systems.



4.4. Probiotic effect:

Release of pathogen contaminated water into natural water bodies creates serious health issues both for human and animals. The post harvested water in BFT is safe because biofloc itself acts as a probiotic. Bioflocs contain a wide range of bacteria, algae, protozoa and other zooplankton organisms. Studies revealed that there are as many as 2000 operational taxonomic units [OTUs] present in the system. It is a novel strategy for disease management in contrast to conventional approaches such as antibiotic, antifungal, probiotic and prebiotic application. The continuous addition of carbon source develops polyhydroxy alkanoates (PHA) accumulating bacteria and other groups of bacteria. Poly hydroxyl butarate (PHB) is produced by a wide variety of microorganisms such as *Bacillus* sp., *Alcaligenes* sp., *Pseudomonas* sp. from soluble organic carbon and is also involved in bacterial carbon metabolism and energy storage (Sinha et al., 2008). These granules are synthesized under conditions of physiological and nutrient stress, where nitrogen is limited in the presence of an excess carbon source. These polymers are degraded in the gut and could have antibacterial activity. The breakdown of PHA can be carried out *via* chemical and enzymatic hydrolysis. Enzyme hydrolysis is generally carried out by extracellular depolymerases activities which are widely distributed among bacteria and fungi, acting as a preventive or curative protector against *Vibrio* infections and stimulate growth and survival of shrimp and fish larvae (De et al., 2012). Studies revealed that the immune system of shrimp can be enhanced in the BFT and there is a lower incidence of diseases among shrimp grown in biofloc system.

4.5. Aquaponics:

The most conflictory issue is about the discharge of harvested water into agricultural lands and other natural water bodies. The harvested water of BFT is highly nutritious that can be reused for aquaponics. Aquaponics is a sustainable production system that combines aquaculture with hydroponics. The waste excreted from aquaculture species and uneaten feed is converted into beneficial nutrients in water. These aquaculture waste waters are directly pumped to raceways of hydroponics vegetables. Typical plants raised in aquaponics include lettuce, shurd, tomato and fruits such as passion fruit, strawberry, watermelon etc. The presence of rich microbiota and variety of micro and macronutrients especially Nitrate and other beneficial nutrients in the water are absorbed by the vegetables and fruits in a “natural fertilization way”.

5.0. Conclusion and perspectives of BFT:

The aquaculture practices have become a major problem by generating a lot of waste effluents, which will damage the aquatic environment at the global level. The intensification of aquaculture will lead to environmental degradation, and socioeconomic conflicts. The requirements for sustainable and eco-friendly aquaculture development can be fulfilled by the use of BFT. A variety of beneficial features can be ascribed to this technology as a sustainable tool to simultaneously address its environmental, social and economical issues concurrent with its growth. However, research should be focused on the optimal way to manage biofloc in aquaculture ponds with



respect to optimal floc morphology, compositional and nutritional value. Implementation of this technology by convincing the farmers is an additional challenge. Therefore, educating and sharing technical knowledge emphasizing its potential economic benefits to the farmers is necessary to implement BFT in future aquaculture systems.

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