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# Application of biofloc technology (BFT) in the aquaculture system

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#### Abstract

The aquaculture industry is growing fast at a rate of ~9% per year since the 1970s. However, this industry has come under scrutiny for contribution to environmental degradation and pollution. As a result, requirement for more ecologically sound management and culture practices remains fully necessary. Moreover, the expansion of aquaculture is also restricted due to land costs and by its strong dependence on fishmeal and fish oil. Such ingredients are one of the prime constituents of feed for commercial aquaculture. Feed costs represent at least 50% of the total aquaculture production costs, which is predominantly due to the cost of protein component in commercial diets. Interest in closed aquaculture systems is increasing, mostly due to bio-security, environmental and marketing advantages over conventional extensive and semi-intensive systems. When water is reused, some risks such as pathogen introduction, escapement of exotic species and discharging of waste water (pollution) are reduced and even eliminated. The environmental friendly aquaculture system called "Biofloc Technology (BFT)" is considered as an efficient alternative system since nutrients could be continuously recycled and reused. The sustainable approach of such system is based on growth of microorganism in the culture medium, benefited by the minimum or zero water exchange. These microorganisms (biofloc) has two major roles: (i) maintenance of water quality, by the uptake of nitrogen compounds generating "in situ" microbial protein; and (ii) nutrition, increasing culture feasibility by reducing feed conversion ratio and a decrease of feed costs. The objective of this chapter is to review the application of Biofloc Technology (BFT) in the aquaculture system and describes the utilization of biofloc biomass as an ingredient for compounded feeds. An addition goal is to clarify the basic aspects of such technology, aiming to encourage further research.

Keywords: Biofloc, microbial protein, C/N ration, micro-organisms and water quality

### Introduction

World Aquaculture is growing at an annual rate of 8.9–9.1% since the 1970s. This high growth rate is needed to solve the problem of shortage of protein food supply and which is particularly situated in the developing countries (Subasinghe, 2005; Gutierrez-Wing and Malone, 2006 and Matos et al., 2006) <sup>[90, 46, 63]</sup>. However, environmental and economical limitations can hamper this growth. Especially intensive aquaculture coincides with the pollution of the culture water by an excess of organic materials and nutrients that are likely to cause acute toxic effects and long term environmental risks (Piedrahita, 2003)<sup>[80]</sup>. High-density culture in intensive systems requires high amounts of feed to be added to the systems. This will cause water quality deterioration due to the high concentrations of organic compounds (Avnimelech, 2007)<sup>[13]</sup>. Since only 20-30% of feed are assimilated in fish and the remaining 70-80% of the feed will be accumulated in the water body as uneaten feed and excretory products (Gross and Boyd, 2000; Avnimelech and Ritvo, 2003) [45]. Ammonia is usually the abundant form of combined inorganic nitrogen in aquaculture pond and it can be rather toxic to animals. Elevated concentrations of ammonia affect growth, oxygen consumption and even can eventually cause mortality of fish. Increased ambient nitrite concentration negatively affects the growth performance and survival of fish. (Colt and Tchobanoglous, 1976; Colt and Armstrong, 1981; Tucker and Robinson, 1990; Mallasen and Valenti, 2006) <sup>[29, 30, 97, 62]</sup> and also inhibits the disease resistance of the cultured fishes (Brock and Main, 1994) [20]. Ammonia-N in water exists in two forms and they are 1) unionised ammonia and 2) ionised ammonium. Among this, unionised ammonia is more toxic when compared to ammonium ion (Boyd and Tucker, 2009) <sup>[21]</sup>. Many researchers made attempt to find a solution to reduce or remove ammonia from aquaculture systems. There are several ways to eliminate ammonia from the aquaculture systems, like exchange and replace the water, use of biofiltration system, Recirculatory

Aquaculture System (RAS), reduce or stop feeding, flush the pond with fresh water, reduce the stocking density, aerate the pond, and reduce the pH level. (Thompson et al., 2002)<sup>[94]</sup>. The use of RAS has the ability to maintain low ammonia and nitrite levels by means of nitrification (Valenti and Daniels, 2000) [99]. However, this is rather expensive and during an imbalance in the process, nitrite levels may rise in water (Russo and Thurston, 1991; Valenti and Daniels, 2000; Jensen, 2003) [84, 99, 53]. Biofloc technology (BFT) is an innovative technology which can solve the above problems. Biofloc is defined as macro aggregates composed of diatoms, macro algae, faecal pellets, exoskeleton, and remains of dead organisms, bacteria and invertebrates. Biofloc technology has become a popular technology in the farming of Oreochromis niloticus, Litopenaeus vannamei, Macrobrachium rosenbergii and Penaeus monodon. It was commercially first applied in Belize by Belize Aquaculture in North America. It also has been applied with success in shrimp farming in Indonesia and Australia (Taw, 2010) [93].

BFT is an aquaculture system which focused on a more efficient use of nutrient input with limited or zero water exchange system. The main principle of BFT is to recycle nutrient by maintaining a high carbon/ nitrogen (C/N) ratio in the water in order to stimulate heterotrophic bacterial growth that converts ammonia into microbial biomass (Avnimelech, 1999)<sup>[11]</sup>. The microbial biomass will further aggregate with other microorganisms and particles suspended in the water forming what has been called "biofloc", which eventually can be consumed in situ by the cultured animals or harvested and processed as a feed ingredient (Avnimelecah 1999; Avnimelech 2007; Crab et al., 2007; De Schryver et al., 2008; Kuhn et al., 2008; Kuhn et al., 2009; Kuhn et al., 2010)<sup>[11, 13,</sup> <sup>31, 37, 57-59]</sup>. BFT is therefore considered as a promising system for a sustainable and environmentally friendly aquaculture system. The term of "biofloc" applies to a compound made out of 60 to 70% of organic matter, which includes a heterogeneous mixture of microorganisms (fungus, algae, protozoan, and rotifers) and 30 to 40% of inorganic matter such as colloids, organic polymer, and dead cell. They can reach a size up to 1000µm, irregular shape, full of the pore, and allow the pass of fluids (Chu and Lee, 2004)<sup>[27]</sup>. In BFT, natural productivity plays an important role in recycling nutrients and maintaining the water quality (Ray et al., 2010; McIntosh et al., 2000) [81, 66]. The consumption of biofloc by shrimp or fish has demonstrated numerous benefits such as improvement of growth rate, (Wasielesky et al., 2006) [104] decrease of FCR and associated costs in the feed. (Burford et al., (2004) [24]. Growth enhancement has been attributed to both bacterial and algae nutritional components and which can lower the conventional feeding ration upto 30% due to the consumption of biofloc (Panjaitan, 2004)<sup>[78]</sup>. Burford et al., (2004) <sup>[24]</sup> reported that more than 29% of daily food consumed for L. vannamei could be biofloc. In tilapia, Avnimelech et al., (1994)<sup>[9]</sup> estimated that feed utilization is higher in BFT than fish reared in conventional waterexchange systems. Nursery phase is defined as an intermediate step between hatchery-reared early post larvae and grow-out phase (Mishra et al., 2008) [69]. Such phase presents several benefits such as optimization of farm land, increase in survival and enhanced growth performance in grow-out ponds (Apud et al., 1983; Arnold et al., 2009)<sup>[2, 3]</sup>. Emerenciano et al., (2011) [43] observed that presence of bioflocs resulted in the increases of 50% in weight and almost 80% in final biomass of early larval stage of fish when compared to the conventional clear-water system. Cohen *et al.* (2005) <sup>[28]</sup> and Mishra *et al.* (2008) <sup>[69]</sup> reported higher survival rates in the rearing of fish larvae in BFT system. The growth and survival of cultured fish were not affected by the higher stocking density of 2500 and 5000 nos/sq.m and which resulted in the greater production outputs (Arnold *et al.*, (2009) <sup>[3]</sup>. Biofloc technology facilitates intensive culture while reducing investment and maintenance costs and incorporating the potential to recycle feed. The technology is based upon zero or minimal water exchange to maximize biosecurity while minimizing external environmental effects.

## History of biofloc technology

The scientific and practical concepts of BFT evolved concurrently and independently at about the same time by Steve Hopkins and co-workers at the Waddell Mariculture Center, South Carolina and by Avnimelech and co-workers in Israel (Avnimelech, 1993; Hopkins et al., 1993; Avnimelech et al., 1994; Chamberlain and Hopkins, 1994) [8, 51, 9, 25]. The pioneers of this technology include Steve Surfling, who established Solar Aquafarms in California, a shrimp and fish farm based upon developing active microbial suspension and Hepher, Schroeder, Moav and Wohlfarth in Israel, who developed the concept of the "Heterotrophic feed web" (Hepher, 1985) <sup>[50]</sup>. In both cases, zero and minimal water exchange was practised. Organic residues accumulating in the pond under such conditions degrade and ammonia - N is nitrified or assimilated, by an intensive microbial community. This series of processes replace the conventional external biofilter or high water exchange systems. In essence, the microbial processes within these ponds serve as the pond water quality treatment system and microbial protein serves as a feed additive.

## Uses of biofloc technology in aquaculture

The current worldwide growth rate of the aquaculture business (8.9-9.1% per year since the 1970s) is needed in order to cope with the problem of shortage in protein food supplies, which is particularly situated in the developing countries (Subasinghe, 2005; Gutierrez-Wing and Malone, 2006; Matos *et al.*, 2006) <sup>[90, 46, 63]</sup>. However, environmental and economical limitations can hamper this growth since intensive aquaculture corresponds with the pollution of the culture water through the addition of anexcess of organic materials and nutrients that are likely to cause acute toxic effects and long term environmental risks (Piedrahita, 2003) <sup>[80]</sup>. The common method for dealing with this pollution has been the use of continuous replacement of the pond water (Gutierrez-Wing and Malone, 2006)<sup>[46]</sup>. However, the water volume needed for even small to medium aquaculture systems can reach up to several hundreds of cubic meters per day. A second approach is the removal of the major part of the pollutants in the water as it performed in recirculating aquaculture systems (RAS) with different kinds of biological water treatment systems (Gutierrez-Wing and Malone, 2006) <sup>[46]</sup>. The amount of water that needs to be replaced on a daily basis generally is reduced to about 10% of the total water volume (Twarowska et al., 1997) [98]. However, RAS technique is a costly one in terms of capital investment and operational energy and labour costs (Gutierrez-Wing and Malone, 2006) [46].

## **Biofloc system**

The bioflocs are conglomerates of microbes, macro

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aggregates composed of diatoms, microalgae, protozoa, faecal pellets, exoskeleton, and remains of dead organisms, dead

organic particles, bacteria and invertebrates.



Coscinodiscus sp

Brachionus sp

Cyclidium sp



Paramecium sp

Triceratium sp

Desmodesmus sp

Bioflocs found in ponds are porous, light and have a diameter of 0.1 to a few millimeter. The basic requirements for biofloc system operation include high stocking density with 130-150 nos /m<sup>2</sup> and high aeration of 28 to 32 Hp/ha with correct paddlewheel position in ponds. Ponds must be lined with concrete or high-densitypolyethylene (HDPE), and pelleted grain and molasses are added to the culture water. A maximum fish production of nearly 50 mt/ha was achieved in small ponds in Indonesia. The microorganisms play major roles with respect to natural productivity, nutrient cycling, water quality and the nutrition of the cultured animals (Moriarty 1997; McIntosh et al., 2000) [66]. Control of the predominantly heterotrophic bacterial community over autotrophic microorganisms is achieved by the use of high carbon to nitrogen ratios (Avnimelech et al., 1989; 1992; 1994; Kochva et al., 1994; Avnimelech, 1998; 1999) [6, 7, 9, 56, <sup>10, 11]</sup>. The uptake of ammonia by bacteria improves water quality and increases microbial biomass production (Avnimelech, 1999)<sup>[11]</sup>. These processes serve as fuel for operating the "floc system" (Burford et al., 2004; Cohen et al., 2005) [24, 28]. Furthermore, a nutrient-rich feed source is available 24 h per day and could reduce artificial feed inputs and costs (Browdy et al., 2001; Avnimelech 2007 <sup>[22, 13]</sup>; Samocha et al., 2007) [87]. Avnimelech, (2007) [13] and Hari et al., (2004 and 2006) [48, 49] opined that the theoretical and practical calculation leads to the opinion that BFT could reduce the feed cost. The survival rate of cultivable finfish in BFT culture system is higher than that of in recirculating aquaculture system and control culture system (Suresh and Lin, 1992; Azim and little, 2008) <sup>[92, 14]</sup>. Craig and Helfrich (2002) <sup>[35]</sup> observed that biofloc do not allow for a complete replacement of traditional fish feed but still can bring about a substantial decrease in the total production costs. Tacon et al. (2002) <sup>[95]</sup> also found that biofloc can reduce the production cost through reduction in the feed cost and in addition to that it brings other beneficial effects such as better water quality

which leads to an environmental friendly aquaculture practices and increase biosecurity. Biofloc technologies (BFT) were developed to minimize effluent discharge, protect the surrounding water resources and improve farm biosecurity (Weirich *et al.*, 2002; Burford *et al.*, 2003; Avnimelech 2007) <sup>[105, 23, 13]</sup>. Large microbial clusters are formed due to the flocculation of cells with feed particles and which favors subsequent uptake by fish. Avnimelech *et al.*, 1982; Beveridge *et al.*, 1989; Beveridge and Baird, 2000) <sup>[16, 15]</sup>. Avnimelech (2007) <sup>[13]</sup> confirmed the uptake of biofloc by Mozambique tilapia using stable nitrogen isotope labelling technique.

#### **Biofloc- Microbial communities**

When fishes are reared at high densities without water exchange biofloc develop in the ponds that have several advantages for culture. The diverse microbial community in the biofloc dominated systems was thought to increase competition with potentially pathogenic microbes including Vibrios, reducing problems with non-excludable pathogens (Bratvold and Browdy, 1999) <sup>[18]</sup>. In addition, the natural productivity associated with these bioflocs has been shown to provide growth-enhancing factors which improve shrimp production (Moss et al., 1992; Moss, 1995; Decamp et al., 2002; Moss, 2002; Burford et al., 2004; Wasileski et al., 2006) <sup>[72-74, 95, 75, 24]</sup>. Additional research has been conducted on the dynamics of the microbial communities in these systems, methods for measuring microbial activity and some techniques for manipulation of the make-up of these communities (Bratvold and Browdy, 1998; 1999, 2001; Browdy et al., 2001a; Burford et al., 2003; Decamp et al., 2003; Ebeling, 2006) <sup>[19, 22, 17, 18, 23, 41]</sup>. Various factors like salinity, light, and type of culture system affect the microbial composition of biofloc (Anand, et al., 2013)<sup>[1]</sup>. Anand et al. (2013) <sup>[1]</sup> observed that the major microbial community developed in the biofloc comprise of Vibrionaceae, Bacillus

*sp* and *Lactobacillus sp* with the majority of gram-negative bacteria.

#### **Biofloc -Microbial Nutrition**

The microbial protein, aggregated in microbial flocs serves as a rich source of amino acids and growth factors for fish and shrimp, leading to significant recycling of protein and higher utilization of feed (Avnimelech *et al.*, 1994; Chamberlain *et al.*, 2001b; Tacon *et al.*, 2002) <sup>[9,95]</sup>. Microbes proved to be an effective source of proteins, vitamins, and essential fatty acids

for shrimp and several fish species (Tacon*et al.*, 2002) <sup>[95]</sup>. It was shown that proteins can be recycled through the activity of aerobic microorganisms and that protein utilization is doubled in BFT ponds (Avnimelech *et al.*, 1994; McIntosh, 2000b) <sup>[9, 65]</sup>. Nitrogen produced in the aquatic system could be controlled by feeding the heterotrophic bacteria with carbohydrate and which can synthesis microbial protein through the uptake of inorganic nitrogen from the water. (Megahed 2010; Panjaitan, 2011; Rostikarita and Liliwalim, 2012; Liu *et al.*, 2014; Rostika, 2014; Ogello *et al.*, 2014) <sup>[67, 79, 82, 61, 83, 76].</sup>



Imhoff Cone

Use of Organic carbon in Microbes

Avnimelech (1999) [11] calculated the carbohydrate requirement (20g) to immobilize 1.0 g of N, based on a microbial C/N-ratio. A concentration of about 10 mg ammonia -N per liter could almost completely be removed within 5 h after the addition of glucose at C/N ratio of 10 without the accumulation of nitrite and nitrate in the culture pond (Avnimelech, 1999)<sup>[11]</sup>. This can be achieved by adding different types of organic carbon source, resulting in a production of microbial proteins that could be reused as fish food. Hari et al. (2006) [49] stated that carbohydrate addition in combination with a decreased dietary protein level improved the sustainability of shrimp farming in extensive shrimp culture systems through increased nitrogen retention in harvested shrimp biomass, reduced demand for feed protein, reduced concentrations of potentially toxic TAN and nitrite-N in the system, and reduced water based nitrogen discharge to the environment. If carbohydrate was added to the water column to enhance heterotrophic bacterial protein production, the protein level in the diet could be reduced from 40% to 25%, without compromising fish production. The control of inorganic nitrogen accumulation in the pond is based on carbon metabolism and nitrogen immobilization into microbial cells (Avnimelech, 1999) [11]. Bacteria and other microorganisms use carbohydrate as a food, to generate energy and to grow. The addition of carbohydrates is a potential means to reduce the concentration of inorganic nitrogen in intensive aquaculture systems. By adding carbohydrate to the water, microbes are forced to immobilize any inorganic nitrogen present in the water, preferably to immobilize TAN. This process is relatively fast if the availability of the carbonaceous substrate added to the culture pond is high (Avnimelech, 1999)<sup>[11]</sup>.

#### **Biofloc-Disease management**

The heterotrophic microbial biomass is suspected to have a controlling effect on pathogenic bacteria (Michaud *et al.*, 2006). Short chain fatty acids as bio-control agents against

Biofloc Tank

pathogenic diseases are of particular interest. It was reported that the application of 20 mm of butyric acid to the culture water of Artemia franciscana resulted in the protection of these organisms against pathogenic Vibrio campbellii (Defoirdt *et al.*, 2006)<sup>[39]</sup>. In this respect, research concerning certain special components in microbial cells is warranted. Emphasis can be put on the organic storage product poly- $\beta$ hydroxybutyrate (PHB). This is an intracellular biodegradable polymer produced by a wide variety of micro-organisms and is involved in bacterial carbon and energy storage (Defoirdt et al., 2007) <sup>[40]</sup>. It is considered to be depolymerised in the gut of higher organisms and has also been shown to act as a preventive or curative protector of A. franciscana against Vibrio infections (Defoirdt et al., 2007)<sup>[40]</sup>. The accumulation of PHB by mixed cultures in BFT can occur under specific conditions determined by the presence of a growth limiting factors such as nitrogen and the presence of an excess carbon source (Salehizadeh and Van Loosdrecht, 2004) [86]. Upon release from the bacterial cell, e.g. in the case of cell death and lyses, degradation of PHB is performed by the activity of extracellular PHB depolymerase enzymes which are widely distributed among bacteria and fungi (Jendrossek and Handrick, 2002)<sup>[52]</sup>. This results in the release of 3hydroxybutyrate into the surrounding environment (Trainer and Charles, 2006) <sup>[96]</sup>. As such, PHB might offer a prebiotic advantage for aquaculture. It was observed that the regular addition of carbon to the culture is known to select for polyhydroxyalkanoate (PHA) accumulating bacteria (Salehizadeh and Von Loosdrecht, 2004) [86] such as Alcaligene seutrophus, Azotobacter vinelandii, Pseudomonas oleovorans and others that synthesise PHA granules. Such granules are synthesised under conditions of nutrient stress thatis when an essential nutrient like nitrogen is limited in the presence of an excess carbon source (Avnimelech, 1999)<sup>[11]</sup>. Enzyme hydrolysis is generally carried out by PHA depolymerase produced by various micro-organisms such as Aspergillus fumigatus, Pseudomonas fluorescens, and others (Khanna and Srivastava, 2004)<sup>[55]</sup>. Chemical hydrolysis can

be carried out by treating the PHA with NaOH (Yu *et al.*, 2005) <sup>[106]</sup>. Defoirdt *et al.*, (2007) <sup>[40]</sup> reported that commercial polyhydroxy butyrate (PHB) particles or PHB accumulating bacteria offered a preventive and curative protection to Artemia against luminescent vibriosis. They observed complete protection at a level of 1000 mg/l of commercial PHB or 10<sup>7</sup>cells/ml of PHB accumulating bacteria. Recently, Rostika (2014) <sup>[83]</sup> observed that the biofloc production in the BFT system has the ability to synthesis compound like biopolymer, bacteriocins against bacterial pathogen and this observation confirms the statement documented by Crab *et al.* (2012) <sup>[34]</sup>.

# **Biofloc - Water quality Management**

Crab et al. (2012) <sup>[34]</sup> stated that BFT is a technique of enhancing water quality in aquaculture through balancing C/N ratio in the system. Nitrogen control is stimulated by feeding bacteria with carbohydrates, and through the subsequent uptake of nitrogen from the water, by the synthesis of microbial protein. The relationship between adding carbohydrates, reducing ammonia -N and producing microbial proteins depends on the microbial conversion coefficient; the C/N ratio in the microbial biomass and the carbon contents of the added material (Avnimelech, 1999)<sup>[11]</sup>. The C/N ratio has been widely used as an index of the rate at which organic matter decomposes. If the organic matter is low in nitrogen content (i.e. a high C/N ratio), some of the nitrogen for microbial growth will be obtained from the water column and will become immobilised as microbial protein. The microbial population comprising of heterotrophic bacteria in BFT culture tank consume ahigher level of dissolved oxygen as much as 77% of the total oxygen consumption (Olah et al., 1987; Visscher and Duerr, 1991; Avimelechi et al., 1992; Sun et al., 2001) <sup>[77, 103, 7, 91]</sup>. However, Avnimelech, (1999) <sup>[11]</sup> stated that the C/N ratio in the pond should be 10.75, to eliminate inorganic nitrogen. Crab et al., (2009) [32] observed that carbon rich and low protein feed (in experiment feed) could sustain 1:20 C/N ratio and limited the presence of inorganic nitrogen species even at high stocking densities of 20 kg/m<sup>3</sup> at harvest. If carbon and nitrogen are well balanced in the system, ammonia-N in addition to organic nitrogenous waste will be converted into bacterial biomass (Schneider et al., 2005) [88]. By adding carbohydrates to the pond, bacterial growth is stimulated and nitrogen uptake takes place through the production of microbial proteins (Avnimelech, 1999)<sup>[11]</sup>. This promoted nitrogen uptake by bacterial growth decreases the ammonia-N concentration more rapidly than nitrification (Hargreaves, 2006)<sup>[47]</sup> and immobilization of ammonia-N by heterotrophic bacteria occurs much more rapidly because the growth rate and microbial biomass yield per unit substrate of heterotrophs are a factor 10 higher times than that of nitrifying bacteria. The microbial biomass yield per unit substrate of heterotrophic bacteria is about 0.5 g biomass C/g when substrate C was used (Eding et al., 2006)<sup>[42]</sup>. Uptake of the bio-flocs by fish depends most probably on the fish species and feeding traits, fish size, floc size and floc density (Avnimelech, 2007)<sup>[13]</sup>. The fluctuation in the level of total alkalinity was very much in biofloc fish culture tank, while it was stable in control tanks (Azim and little, 2008)<sup>[14]</sup>.

## Biofloc as a feed for fishes

Recent studies have indicated that controlled application of nutrient-dense high-protein feeds can improve growth performance when compared to lower protein diets offered on

an iso-nitrogenous basis (Kureshy and Davis, 2002; Davis and Venero, 2005; Venero et al., 2007, 2008) [60, 36, 101, 102]. The formulation of feeds can have an important indirect effect on the system microbial community. Inputs of carbon and nitrogen must be considered as part of overall management of the bio-floc communities upon which system stability depends. Thus some researchers suggest a lower protein feed formulation strategy to encourage heterotrophic bio-floc production (Avnimelech, 1999; McIntosh, 2000a; Ebeling et *al.*, 2006) <sup>[11, 64, 41]</sup>. The contribution of microbial metabolism to fish nutrition was defined by a group of investigators in Israel (Moav et al., 1977; Schroeder, 1978) [70, 89] as the heterotrophic food web. According to this concept, fish can feed, directly or indirectly, on primary producers, yet they can also feed on bacteria degrading residues, applied to the pond, such as manure. Kuhn *et al.* (2009)<sup>[58]</sup> reported that microbial floc incorporated fish diet enhanced that mean growth up to 65%, when compared with the control diets and Rostikarita and Liliwalim, (2012) [82] observed ahigher rate of survival through the incorporation of biofloc at 10% level in the feed. The average protein conversation ratio (PCR) in fish or shrimp production is 4. Protein utilisation is significantly higher in BFT systems. Adding carbonaceous substrates causes microbes to harvest the excreted nitrogen and produce microbial proteins that are then consumed by the fish. It may be said that the fish eat the protein twice: once with the feed and then as microbial proteins. The PCR obtained in BFT commercial systems is about 2, whereas average protein utilisation is 46%, twice as high as in conventional systems (Avnimelech et al., 1994; Chamberlain et al., 2001a)<sup>[9, 26]</sup>. Protein efficiency in experimental tanks, as high as 65% was demonstrated by Velasco et al. (1998) [100]. The bacterial protein and new cells (single-cell protein) synthesised by the heterotrophic bacterial population are utilised directly as a food source by the cultured organisms (carp, tilapia, shrimp), thus lowering the demand for supplemental feed protein (Avnimelech, 1999) <sup>[11]</sup>. Hariet al. (2004) <sup>[48]</sup> reported that culture fish could effectively utilise the additional protein derived from the increased bacterial biomass as a result of carbohydrate addition. Burford *et al.* (2004)<sup>[24]</sup> suggested that "flocculated particles" rich in bacteria and phytoplankton could contribute substantially to the nutrition of the cultivable fish in intensive fish ponds.

## **Biofloc as feed**

Several studies have been conducted using biofloc technology for the culture of tilapia (Azim and Little 2008; Asaduzzaman et al., 2009) [14, 4] and shrimps (Kuhn et al., 2008, 2010; Asaduzzaman *et al.*, 2009; Ray *et al.*, 2011; Emerenciano *et al.* 2012) <sup>[57, 59 44, 4]</sup>. Kuhn *et al.* (2009) <sup>[58]</sup> evaluated the growth of cultivable fish using microbial floc meal as a replacement ingredient for fish meal and soybean protein and concluded that microbial floc meal outperformed the control diets in terms of weight gain per week without any differences in survival. Further, Anand et al. (2013) <sup>[1]</sup> reported that proximate composition of biofloc contained 24.30% protein, 3.53% crude lipid, 29.24% nitrogen free extract, 31.98% ash content and 10.75% acid insoluble ash content. Rostika, (2014) <sup>[83]</sup> also recorded the proximate composition of flocwith 53.5% protein, 2.6% crude protein, 4.0% crude fibre and 7.5% ash content. While Ogello et al. (2014) [76] stated that biofloc produced in BFT system has adequate protein, lipid, carbohydrate and fatty acids. Ju et al. (2008) [54] stated that chlorophyll-dominated biofloc which contained higher

crude protein content (42%) than flocs dominated by diatoms (26 -34%) and bacteria (38%) and further the has been documented that biofloc is a rich source of bio active compounds like carotenoids, chlorophylls, phytosterols, bromophenols, amino sugars. Kuhn *et al.* (2009; 2010) <sup>[58, 59]</sup> stated that the use of biofloc as a dietary ingredient in feed preparation is found to enhance the growth rate of fish. Azim and Little (2008) <sup>[14]</sup>, found that the BFT treatment contributed 44-46% greater individual weight gain and net fish production than those in the control. Ogello *et al.* (2014) <sup>[76]</sup> recorded that highest growth rate (0.3g/day) and yield (300 tonnes/ha) along with 20% reduction in fish feed in well-managedbiofloc ponds, and this growth is in similar with the observation by Avnimelech *et al.* (1994) <sup>[9]</sup>.

# Conclusion

Biofloc technology brings a major advantage of minimizing consumption and release of water, recycling in situ nutrients, organic matter and introduction of pathogen were reduced, improving water quality of the fish farm. Consumption of microorganisms in biofloc technology reduces FCR and feed cost. Also, microbial community is also able to rapidly utilize dissolved nitrogen leached from fish faeces and uneaten food and convert it into microbial protein. Biofloc technology Highers the stocking density, high yield production so its useful for fish farmers. The BFT system normally provides good health environmental condition and hence the survival rate of the cultivable fish will be in the higher side. This biofloc technology could be made as a cutting edge technology for the benefit of fish farmers so as to increase the fish production.

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