**A Major Step Towards the Sustainability in Aquaculture Practices Through Biofloc Culture Technology**

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**------------------------------------------------------------------------------------------------------------------Introduction**

 In the current Indian economy, aquaculture is thought to be the sector with the quickest rate of growth. It is established that the quantity produced by the Indian fisheries industry increased from 2.4 million tons in 1980 to 14.1 million tons in 2019–20, with an average growth rate of 4.35% for both capture and culture. It has been observed that over 50% of India's fish production comes from states like Andhra Pradesh, Tamilnadu, Odisha, and West Bengal. However, on the other hand, the increasing population and rising demand for food push farmers toward intensive aquaculture practices rather than extensive or semi-intensive ones, which has an impact on the sustainability of natural resources like land and water, which are essential to aquaculture. The goal of aquaculture growth should be to increase the production of aquaculture products while keeping the operation's reliance on natural water and land sources relatively small (Avnimelech, 2009). A technology known as closed recirculatory aquaculture systems (RAS) has been created in order to prevent the future scarcity of natural resources. The creation of environmentally safe, sustainable aquaculture systems is the second objective (Naylor et al., 2009). The third goal is to create systems that offer a neutral benefit rate in order to promote social and economic sustainability. With biofloc technology, all three of these requirements for sustainable cultivation can be satisfied. According to Debbarma et. al, (2021), the biofloc is defined as macro-aggregates of diatoms, macro-algae, fecal pellets, exoskeleton, remnants of deceased creatures, bacteria, protozoa, and invertebrates. Using the extra nitrogen waste from the uneaten feed and faces of the cultured species, biofloc technology uses the microbial community to manage the suspended solids in the culture system rather than eliminating them through other processes.

**Compositions of Biofloc:**

 Algae, bacteria, and protozoans combine with some particulate organic debris, such as faces and uneaten feed, to form the biofloc. The floc is confined by the filamentous microorganisms that live there and kept together by the mucus that the bacteria secrete. Additionally, certain grazers of floc, such as nematodes and some zooplanktons, are present. In green water BFT, flocs range in size from 50 to 200 microns.

**Different kinds of Biofloc Technology**

1.**The outdoor culture unit:** which is exposed to the natural light, the outside lined raceways for shrimp culture in green houses, as well as lined ponds or tanks for tilapia or shrimp. Concentrated water, like that found in the green water biofloc system, contains a full combination of bacteria and algae.

2**. The indoor culture unit:**  which isn’t exposed to the natural sunlight, only bacteria regulate the water quality in closed indoor systems, which are run as "Brown-Water" biofloc systems.

 To keep the particles afloat in the water column, constant, intense aeration is required; otherwise, the biofloc would settle out of suspension and create piles that would devour the adjacent dissolved oxygen level. The very hazardous gases hydrogen sulphide, methane, and ammonia are released by these anaerobic zones into the shrimp culture. Since the water respiration rate in the biofloc culture unit is 60% higher than in other conventional methods, the aeration system must be kept running in order to maintain the proper quantity of dissolved oxygen. Small tanks or raceways are comparatively much easier to create turbulence in than big outdoor culture units since the solids must be removed by periodic flushing and pumping sludge from the pond center, by operating the paddle wheel aerator.

**Nutritive value of biofloc**

 The high cost of aquaculture feeds is one of the major problem faced the aquaculture industry. Aquaculture feeds must have sufficient amounts of protein and an appropriate balance of amino acids because these nutrients are vital to the growth and general health of aquatic animals. But because these nutrients are a costly part of the feeds, their price affects the feeds' market value. For tilapia, feeding, for instance, can represent 50% of the operating costs and may even escalate in the case of high-protein diets or insufficient proteins (Hisano et al., 2020). Feeding tilapia different feed sources, i.e. algae, zooplankton, and phytoplankton, their nutrient content would improve fish growth, survival, and productivity (Narimbi et al., 2018). According to Avnimelech et al. (2009), tilapia can absorb up to 240 mg N kg−1 of biofloc, or 25% of the protein contained in fish diets. Additionally, bioflocs can contain 25–50% protein, 0.5–5% fat, and the essential amino acids lysine and methionine, along with various vitamins and minerals, including phosphorus, as a result, it offers raised aquatic species an alternate nutrition source.

 It was noted in the previous study, the microbial community that inhabits bioflocs determines its nutritional value. Additionally, the biochemical makeup of bioflocs is influenced by specific elements such as carbon sources and C:N ratio. According to Moreno-Arias et al. (2018), the mixed feed used has an impact on the fatty and amino acid content of shrimp and biofloc cultivated in BFT systems. According to Mugwanya et al., (2021), the plant-based protein diets are thought to be more ecofriendly and sustainable, and thus are more suitable for use in biofloc systems. Consequently, the release of phosphorous and nitrogenous wastes in the aquatic ecosystem and the reliance on overfished marine resources,

Most of the current research on probiotic and prebiotic bioflocs focuses on how microbes proliferate to create floc. Probiotics are beneficial bacteria that are either naturally occurring in the BFT system or given to it in order to boost the immune system of the raised species against both biotic and abiotic stress. Prior research has found and extracted a number of beneficial bacteria from the shrimp culture BFT system, including members of the Bacillaceae family (Ferreira et al., 2017).

**Microorganisms in biofloc culture systems**

 An essential component of any aquaculture system are microorganisms. The BFT system's microorganisms play a variety of vital roles since they are diverse and important. The importance of biofloc organisms in biological filter technology, the variables influencing their population compositions, the effects of BFOs on water quality, and their usage as a food source for aquaculture. Photoautotrophic (such as microalgae), chemoautotrophic (such as nitrifying bacteria), and heterotrophic (such as fungus, protozoans, ciliates, and zooplankton, such as rotifers, copepods, and nematodes) species are frequently found in the BFT system. The density, quality, and diversity of BFOs are influenced by variables such salinity, carbon supply type, carbon-nitrogen ratio, aeration, light, stocking density, and total suspended solids (Khanjani et al., 2022). Diverse microorganisms exhibit distinct functional traits and carry out three primary roles in any aquatic farming system, they serve as an additional food source, produce probiotic qualities, and aid in the removal of inorganic nitrogen compounds (bioaccumulation, bio-assimilation, nitrification, and de-nitrification).The heterotrophic bacteria, which is the nitrogen conversion agent fascinates like other nitrogen conversion mechanisms such as nitrites (Ekasari, 2014), take floc as substrates and help in Sludge degradation (Debbarma, R., Biswas, P and Singh S. K. (2021), the heterotrophic bacteria involve in the nutrient recycling by taking inorganic phosphorus (Kirhman, 1994), which reduces the discharged phosphorus with enhancing the availability of nutrients. Come to phototropic, they help in nitrogen uptake (Khanjani *et al*., 2022), and de-nitrifications and it will depend on the environmental conditions.

 **Monitoring & control of the growth of biofloc development:**

 The number and prevalence of microbes, according to Debbarma et al. (2021), aid in the interplay of many biotic and abiotic elements, displaying an ecological succession across time. A useful method for keeping an eye on this bio floc system is to use the medium's color. Primarily photoautotrophic in the first stage, lasting up to three weeks following the addition of carbohydrates There is heterotrophic dominance. Additionally, nitrifying bacteria were discovered in that biofloc system 6–8 weeks after stabilization.

**The species, which can be cultured in BFT:**

 It has been shown that the probiotic effect of bioflocs, either in situ or added to the diets of different aquatic animals in biofloc systems, improves the growth and well-being of those species. Aquaculture species that are omnivorous and have a high tolerance and high suspended solids levels are the best suitable to benefit from these probiotic effects on aquaculture practices. According to Luo et al,( 2014) and Xu & Pan, (2014) Tilapia (*O. niloticus*) and White shrimp are the most typical animals suitable for biofloc systems respectively. However, other species also have been successfully cultivated in biofloc systems in recent years, including *Macrobrachium rosenbergii*, Common carp, African catfish, Golden crucian carp (*Carassius auratus*) and Indian major carps. Therefore, it is proved that there is a possibility to culture some species, those who can tolerate some suspended solids in the water column with zero water exchange.

**Carbon–Nitrogen ratio:**

 The carbon-nitrogen ratio is crucial in the aquatic environment for preventing the build-up of harmful inorganic nitrogen compounds and converting them into helpful microbial biomass that could feed the raised species. The C/N ratio can be obtained by altering the feed's carbohydrate content or by adding an external carbon source to the rearing water, which will allow microorganisms to absorb waste ammonium and produce microbial biomass. Azaduzzaman et al*.,* 2010, recommended that the increasing the C/N ratio from 10 to 20 significantly increased the bio volume of [phytoplankton](https://www.sciencedirect.com/topics/agricultural-and-biological-sciences/phytoplankton), crustaceans and rotifers in the water column by 15%, 6% and 11%, respectively. The bio volume of periphyton was 50% higher in treatment CN20 compared to treatment CN10. Increasing the C/N ratio from 10 to 20 raised the bio volume of total heterotrophic bacteria (THB) in the water column (70%), deposition (36%) and periphyton (40%). In another study Pérez-Fuentes et al. (2016), reported in the setup of, under high-density cultivation of *O. niloticus* in a BFT system, C/N ratios exceeding 15:1 increases the production of dissolved salts and settled biomass, which is directly impacted the growth performance of fish. The most of researchers recommended a C/N ratio of 10:1 as the optimum condition for the production of *O. niloticus* reared under similar conditions. In another study, (Silva et al., 2017) also found poor water quality due to high TSS, turbidity, alkalinity, and settle able solids because increase the C/N ratio of 20:1, which affected the growth performance of *O. niloticus*. Similar results have been reported in *Clarias gariepinus*. However, Yu et al., (2020), Haghparast et al *(2020*), and Wang et al., (2015) reported better growth performance and immunity in carp at high C/N ratios (20:1 and/or 25:1) cultured in BFT. The disparity between the species and the organic carbon source may be the cause of the inconsistent outcomes. For a biofloc-based system, a C:N ratio of 10 to 20 is advised based on species preferences. The goal of the current investigation is to measure the individual and combined impacts of adding substrate and adjusting the C:N ratio on fish output and aquaculture sustainability.

 Carbon sources are more effective than others at encouraging floc formation. The effective development of flocs by various carbon sources mostly depends on their carbon concentration and rate of degradation. Lower the ammonia concentrations and a faster rate of beneficial microbial biomass growth are indicators of enhanced water quality, which is generally caused by simple sugars like molasses breaking down more quickly than complex carbohydrates like cassava starch (El-Sayed et al., 2021). Khanjani et al.*,* (2021a) found that simple sources of carbon in starch and molasses were led to advance densities of heterotrophic bacteria as compared to complex sources like barley, flour, corn etc. According to Mugwanya et al., (2021), molasses is the most extensively used carbon sources in BFT systems during larval, nursery, and grow-out stages due to their effectiveness in improving water quality for the sustainable production of aquatic species. The factors determining carbon addition are, protein level present in feed, ease of availability and cost, carbon content of the source and Production efficiency. e. g. 1.49g of sucrose can produce 1g of floc

**Conclusion**

 Based on the aforementioned explanations, it can be concluded that BFT is a culture-friendly technique it can be significantly contribute to upholding the idea of sustainability in aquaculture. Even though BFT systems are becoming more and more popular, still more research is needed to optimize the system's functional parameters like energy requirements at a lower cost of materials to ensure that small-scale aquaculturists and those in developing nations can use and find them acceptable. Alternative energy sources including wind turbines, gas, and solar power systems should be taken into account. Furthermore, since in present situations used BPT techniques for certain species are important aspects is water exchange or clear water systems, more research on this situations regarding the water quality of cultivable species reared in BFT under marketable settings is necessary for this system to produce a further sustainable impact in production.

**REFERENCES**

1. Asaduzzaman, M., Rahman, M. M., Azim, M. E., Islam, M. A., Wahab, M. A., Verdegem, M. C. J., & Verreth, J. A. J., Effects of C/N ratio and substrate addition on natural food communities in freshwater prawn monoculture ponds. *Aquaculture*, *306*(1- 4) ,2010, pp 127-136.
2. Avnimelech, Y.; Kochba, M. Evaluation of nitrogen uptake and excretion by tilapia in bio floc tanks, using 15N tracing. *Aquaculture* **2009**, *287*, pp 163–168.
3. Avnimelech, Y. *Biofloc technology: a practical guidebook*. World Aquaculture Society.,2009.
4. Debbarma R , Biswas P & Singh S. K. An integrated biomarker approach to assess the welfare status of Ompok bimaculatus (Pabda) in biofloc system with altered C/N ratio and subjected to acute ammonia stress. *Aquaculture*, *545*, 2021, pp.737184.
5. Ekasari, J. *Biofloc technology as an integral approach to enhance production and ecological performance of aquaculture* (Doctoral dissertation, Ghent University), 2014.
6. El-Sayed, A.F.M. Use of biofloc technology in shrimp aquaculture: A comprehensive review, with emphasis on the last decade. *Rev. Aquac.* **2021**, *13*, pp 676–705.
7. Ferreira, M.G.; Melo, F.; Lima, J.V.; Andrade, H.A.; Severi, W.; Correia, E.S. Bioremediation and biocontrol of commercial probiotic in marine shrimp culture with biofloc. *Lat. Am. J. Aquat. Res.* **2017**, *45*, pp 167–176.
8. Hisano, H.; Parisi, J.; Cardoso, I.L.; Ferri, G.H. and Ferreira, M.F. Dietary protein reduction for Nile tilapia fingerlings reared in biofloc technology. *J. World Aquac. Soc.* **2020**, *51*,pp 452–462.
9. Haghparast, M.M.; Alishahi, M.; Ghorbanpour, M.; Shahriari, A. Evaluation of hemato-immunological parameters and stress indicators of common carp (*Cyprinus carpio*) in different C/N ratio of biofloc system. *Aquac. Int.* **2020**, *28*, pp 2191–2206.
10. Kirchman, D. L. The uptake of inorganic nutrients by heterotrophic bacteria. *Microbial Ecology*, *28*, 1994,pp 255-271.
11. Khanjani, M. H., Mohammadi, A., & Emerenciano, M. G. C. Microorganisms in biofloc aquaculture system. *Aquaculture Reports*, *26*,2022, 101-103.
12. Luo, G., Gao, Q., Wang, C., Liu, W., Sun, D., Li, L., & Tan, H. Growth, digestive activity, welfare, and partial cost-effectiveness of genetically improved farmed tilapia (Oreochromis niloticus) cultured in a recirculating aquaculture system and an indoor biofloc system. *Aquaculture*,2014, *422*, pp 1-7.
13. Mugwanya, M., Dawood, M. A., Kimera, F., & Sewilam, H. Biofloc systems for sustainable production of economically important aquatic species: A review. *Sustainability*, *13*(13),2021, 7255.
14. Moreno-Arias, A.; López-Elías, J.A.; Martínez-Córdova, L.R.; Ramírez-Suárez, J.C.; Carvallo-Ruiz, M.G.; García-Sánchez, G.; Lugo- Sánchez, M.E.; Miranda-Baeza, A. Effect of fishmeal replacement with a vegetable protein mixture on the amino acid and fatty acid profiles of diets, biofloc and shrimp cultured in BFT system. *Aquaculture* **2018**, *48,* pp 53–62.
15. Narimbi, J. Mazumder, D.; Sammut, J. Stable isotope analysis to quantify contributions of supplementary feed in Nile Tilapia *Oreochromis niloticus* (GIFT strain) aquaculture. *Aquac. Res.* **2018**, *49*, pp 1866–1874.
16. Naylor, R. L., Goldburg, R. J., Primavera, J. H., Kautsky, N., Beveridge, M. C., Clay, J. & Troell, M. Effect of aquaculture on world fish supplies. *Nature*, *405*(6790), 2000, pp 1017-1024.
17. Pérez-Fuentes, J.A.; Hernández-Vergara, M.; Pérez-Rostro, C.I.; Fogel, I. C:N ratios affect nitrogen removal and production of Nile tilapia *Oreochromis niloticus* raised in a biofloc system under high density cultivation. *Aquaculture* **2016**, *452*,pp 247–251.
18. Silva, U.L.; Falcon, D.R.; Pessôa, M.N.D.C.; Correia, E.D.S. Carbon sources and C:N ratios on water quality for Nile tilapia farming in biofloc system. *Rev. Caatinga* **2017**, *30*, pp 1017–1027.
19. Wang, G.; Yu, E.; Xie, J.; Yu, D.; Li, Z.; Luo, W.; Qiu, L.; Zheng, Z. Effect of C/N ratio on water quality in zero-water exchange tanks and the biofloc supplementation in feed on the growth performance of crucian carp, *Carassius auratus*. *Aquaculture* **2015**, *443*,pp 98–104.
20. Xu, W. J., & Pan, L. Q. Dietary protein level and C/N ratio manipulation in zero‐exchange culture of Litopenaeus vannamei: Evaluation of inorganic nitrogen control, biofloc composition and shrimp performance. *Aquaculture Research*, *45*(11),2014, pp 1842- 1851.
21. Yu, Z.; Li, L.; Zhu, R.; Li, M.; Duan, J.; Wang, J.Y.; Liu, Y.H.; Wu, L.F. Monitoring of growth, digestive enzyme activity, immune response and water quality parameters of Golden crucian carp (*Carassius auratus*) in zero-water exchange tanks of biofloc systems. *Aquac. Rep.* **2020**, *16*, 100283.