

UDC 639; DOI 10.18551/rjoas.2023-07.24

#### INFLUENCE OF WATER QUALITY ON GROWTH OF STRIPED SNAKEHEAD FISH (CHANNA STRIATA BLOCH, 1793) FRY USING BIOFLOCS AND CONVENTIONAL METHODS

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## ABSTRACT

The potential for the development of Snakehead farming (Channa striata Bloch, 1793) is still very large, not only by using simple technology (conventional) but also using the biofloc method. This study aims to determine the effect of water quality parameters on the growth of snakehead fry that is reared using biofloc and conventional methods. This research was conducted by comparing snakehead fry rearing using the biofloc method and conventional method with details of treatment 1 (P1) using the biofloc method with as many as 6 replicates and treatment 2 (P2) using the conventional method as many as 6 replicates. DO variable does not affect length gain in biofloc and conventional treatments (p>0.05). The pH variable did not affect weight gain in biofloc and conventional treatments (p>0.05). The variables of pH, DO, Temperature, and ammonia together affect the length gain in bioflok and conventional treatments (p<0.05). The variables of pH, DO, temperature, and ammonia together affect the weight gain in biofloc treatment (p<0.05), but the variables of pH, DO, temperature, and ammonia together do not affect the weight gain in conventional treatment (p>0.05). Based on the mean comparison test (independent t-test), the mean water quality in terms of pH and DO in the biofloc and conventional treatments were different, with the mean pH in the conventional treatment higher than in the biofloc treatment, while the mean DO in the biofloc treatment was higher than the conventional treatment. Based on the mean comparison test (independent t-test), the mean temperature, ammonia, length, and weight growth of snakehead fry in biofloc and conventional treatments are the same.

### **KEY WORDS**

Snakehead fish, rearing, biofloc technology (BFT), conventional system.

The increasing consumption of snakehead fish, especially from natural catches, causes its population to decrease day by day (Fitriliyani, 2005; Muslim, 2007), so an intensive cultivation effort is needed to increase the production (Yulisman *et al.*, 2011; Kordi, 2011; Muslim & Syaifudin, 2012). Domestication of snakehead fish has been successfully carried out by the Mandiangin Freshwater Aquaculture Center (KKP, 2014) by producing a fry that has been able to consume the commercial feed. The current survival rate of snakehead fish fry is still quite low (Ramli & Rifa'i, 2010, Extrada *et al.*, 2013; Haiwen *et al.*, 2014). Intensification of aquaculture with high density and nutrients are obtained almost entirely from the high protein feed given (Aji *et al.*, 2014), but fish only utilize 16.3%-40.87% of feed protein (Hari *et al.*, 2004) and the rest will be disposed of in the form of excretion, feed residues and feces (Pillay, 2004) and ammonia which is nitrogen waste (Stickney, 2005). Ammonia-nitrogen can be converted into microbial biomass (Ebeling *et al.*, 2006) with the increasing number of heterotrophic bacteria; it can reduce total ammonia-nitrogen, nitrite, and nitrate in aquaculture (Hari *et al.*, 2004; Ekasari, 2008; De Schryver & Verstraete, 2009).



Biofloc utilize ammonia, and nitrogenous organic waste to be converted into heterotroph bacterial biomass provided there is a balance between organic carbon and nitrogen (Schneider *et al.*, 2005). According to Crab *et al.*, (2007), biofloc technology is a cultivation technology with the addition of organic carbohydrates into the rearing container which aims to increase the C/N ratio and stimulate the growth of heterotrophic bacteria that can assimilate inorganic nitrogen into bacterial biomass (Schneider *et al.*, 2005) so that the addition of organic carbon sources such as molasses, wheat flour, and tapioca starch can maximize the growth of heterotrophic bacteria through increasing the C/N ratio (Avnimelech, 1999; Hari *et al.*, 2004; Ebeling *et al.*, 2006). Hari *et al.*, 2004; Avnimelech, 2009; Crab *et al.*, 2007; Ekasari, 2008; Jung *et al.*, 2020 stated that several types of fish and shrimp can consume biofloc-containing protein. The nutrients contained in floc are protein (19%-58%), carbohydrates (27%-59%), fat (2%-39%), and ash (2%-17%) which are ideal for fish growth (Verstraete *et al.*, 2008; Crab *et al.*, 2009; Yu *et al.*, 2023).

Utilization of flocs formed in culture containers can be applied to reduce the amount of culture waste, water use, and the amount of commercial feed compared to the conventional method of adding manure to grow natural feed as a food source (Qin *et al.*, 1997, Alam *et al.*, 1993; Kumar *et al.*, 2005; Mollah *et al.*, 2009; War & Altaff, 2011). In addition to using the biofloc method, snakehead fish rearing is also carried out using conventional methods, namely growing natural food (plankton) by adding organic fertilizer (poultry manure) to the cultivation container (Ray & David, 1969; DePauw *et al.*, 1980; Srivastava *et al.*, 2006; Altaff & War, 2010; War & Altaff, 2010; 2011). This study aims to determine the dynamics of water quality (pH, DO, temperature, and ammonia) in snakehead fish fry rearing both with biofloc and conventional methods on the growth of the fry.

# METHODS OF RESEARCH

This study was conducted at the Mandiangin Freshwater Aquaculture Center (MFAC) for 35 days.



Figure 1 – Location map of the Mandiangin Freshwater Aquaculture Center (MFAC) (https://www.google.com/maps/place/Balai+Perikanan+Budi+Daya+Air+Tawar+Mandiangin)

Twelve 40x30x25 centimeter plastic tubs were utilized as raising containers. Each container for animal rearing holds 20 L of river water that has been deposited there for four days. Aeration for each container came from a Techno Takatsuki® HP-150 Hiblow Air Pump Blower (Japan) that had aeration stones that rose 5 cm above the bottom of the rearing container.



The utilized fry were nine days old. The two broodstock that were employed spontaneously spawned in pairs and had mean weights of 242 g and mean standard lengths of 25.4 cm.

Heterotrophic bacteria, a commercial product (Biomin Aquastar® Pond), were utilized in this investigation at a concentration of  $1.0 \times 10^9$  CFU mL-1 (Runa, 2019). The probiotic is available as a powder. According to the instructions for usage, 1.2 mg of probiotics are added to a volume of 20 L of water since the dose supplied to the water is 1 mL 50 L<sup>-1</sup> of water.

Molasses, a byproduct of the sugar industry, is employed as a carbon source (Miao *et al.*, 2017; Chang Liu & Kai Cheng, 2022). Molasses has a 37% carbon content (Suastuti, 1998). Each maintenance container requires 2 mL of molasses because the amount used is equivalent to 1 mL per 10 L of water. Tapioca starch, up to 1 g in 20 L of water each, was added to the water after the first carbon (K. K. Prajith, 2012).

This study consisted of two treatments (Miao et al., 2017), namely:

- Treatment 1 (P1): Rearing using the biofloc method with 6 replicates;
- Treatment 2 (P2): Rearing using the conventional method with 6 replicates.

The preparation stage includes disinfection of containers and tools to be used, rearing media, water treatment, and adaptation and acclimatization of the fry.

In this study, preparation comprises preparing containers and raising water. Before usage, the raising containers were cleansed and dried. Suprapto & Samtafsir (2013) state that starters included commercial probiotics:  $10 \text{ mL m}^{-3}$  (0.2 g 20 L<sup>-1</sup>),  $100 \text{ g m}^{-3}$  dolomite lime (2 g 20 L<sup>-1</sup>),  $3000 \text{ g m}^{-3}$  salt (60 g 20 L<sup>-1</sup>), and 100 mL m<sup>-3</sup> molasses. For bacteria to grow in the media, it was also left out for seven days.

After the media preparation process was finished, which took place 5 days after the addition of floc growth material in treatment 1 and dried poultry manure in treatment 2, fish stocking was carried out (Setyawan *et al.*, 2014). According to the predefined treatment, a stocking density of 40 fish 20 L<sup>-1</sup>, or 2 fish L<sup>-1</sup> was used (Purnamawati *et al.*, 2017). The stocked snakehead fry were 9 days old, measuring  $6.5\pm0.52$  mm in length and  $3.03\pm1.45$  mg in weight. Fish rearing was for 35 days.

A commercially produced feed in the form of flour (Fengli 0®, Matahari Sakti, Indonesia) and small granules (Prima Feed 500®, Matahari Sakti, Indonesia) was fed at 8% of the biomass. The minimum amount of nutrients in the powdered commercial feed was 41% protein, 7% fat, 3% fiber, and up to 13% ash. 39–41% of the protein in the tiny granular commercial feed is protein. At the start of stocking, natural food is provided in the form of tubificid worms (Tubifex tubifex) (Helkianson *et al.*, 2020). Two times every day, in the morning and evening, food is given out evenly.

Probiotics and molasses were given every day during the rearing time after feeding as much as 0.4 mL.

Sampling was done every five days to measure the growth of length and weight of the fry. Length measurements were taken using millimeter blocks.

Parameters observed during the study included growth (length and weight), survival rate, and water quality.

Length growth according to Effendie, (1997) was measured using the formula:

$$L = Lt - LO$$

Where: L = Length growth (cm); Lt = Length growth after rearing (cm); L0 = Length growth before rearing (cm).

Absolute growth was calculated using the Effendie formula, (1997) as follows:

$$W = Wt - W0$$

Where: W = Absolute weight growth (g); Wt = Weight of fish fry at the end of rearing (g); Wo = Weight of fish fry at the beginning of rearing (g).



Survival rate (SR) was obtained based on the equation proposed by Zonneveld *et al.*, (1991) and Effendie, (1979), namely:

$$SR = \frac{Nt}{N0} x \ 100\%$$

Where: SR = Survival rate (%); Nt = Number of fish at the end of study (fish); No = Number of fish at the beginning of the study (fish).

Total ammonia, pH, dissolved oxygen, and temperature were the water quality indicators measured in this investigation. An on-site Celsius thermometer (Lutron®, USA) was used to measure the water's temperature. A digital pH meter (Lutron®, USA) was used to measure the pH of the water on-site. The spectrophotometer-vis technique (HACH DR 1900, USA) with a wavelength of 640 nm was used to measure the ammonia content (APHA, 2005).

Using SPSS (Inc., Chicago, IL, USA), research data on growth and water quality were statistically examined. Several growth parameters, including weight growth and length growth in both the biofloc (P1) and conventional (P2) methods, were tested for relationships between water quality parameters and growth using regression analysis, partial test (t-test), simultaneous test (F-test), and coefficient of determination analysis (R2). An independent t-test with a 95% confidence level was used to finish the calculation (p<0.05).

#### **RESULTS OF STUDY**

Although the difference at the ninth sampling was only 0.08 cm, treatment 1 had higher sampling averages for total length than did treatment 2. In contrast to the average weight calculation, it appears that treatment 2's average calculation is higher when compared to treatment 1's average calculation; however, the difference is just 0.033g in the ninth sample.

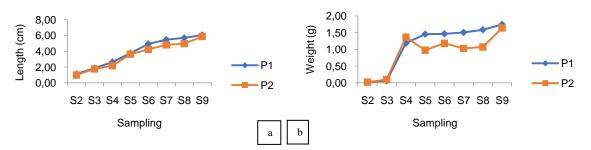


Figure 2 – Length (a) and weight (b) measurements in P1 and P2

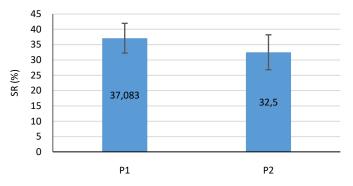
Figure 1 shows a growth graph for length and weight based on the average of each sampling, beginning with the second sampling. From the second sampling to the ninth sampling, there was an increase in the length and weight of snakehead fry. Except for the third and fourth weight samples, which show that treatment 2 is generally superior to treatment 1, sampling both length and weight in treatment 1 is higher. This indicates that the fry reared using the biofloc method have a greater increase in length. In P1, the average weight gain is 1.741 g, and the average length gain is 5.41 cm. When compared to treatment 2 (P2), which has 5.237 cm for length growth and 1.637 g for weight growth, both numbers are higher.

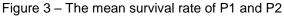
The mean survival rate in treatment 1 was 37.083% higher than in treatment 2 which was 32.5%.

Water quality parameters observed in this study were total ammonia, acidity (pH), dissolved oxygen, and temperature. Water quality measurements based on the length of rearing (Figure 4), there was a spike in ammonia after the addition of floc-forming materials in P1 and the addition of poultry manure in P2 (Figure 5).



RJOAS: Russian Journal of Agricultural and Socio-Economic Sciences ISSN 2226-1184 (Online) | Issue 7(139), July 2023





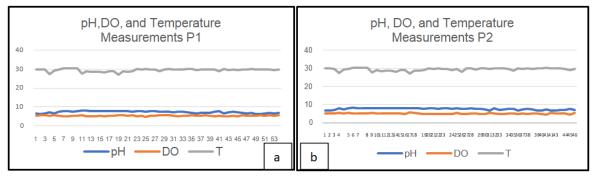
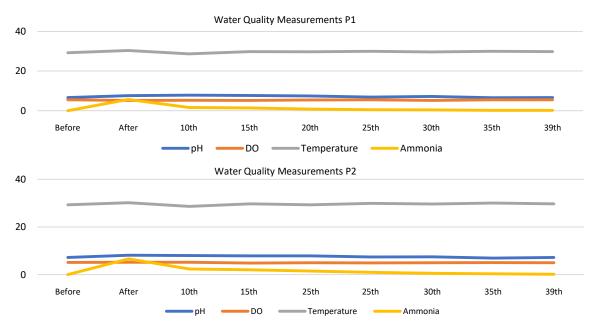
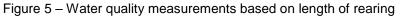


Figure 4 – Water quality measurements: a) Daily data of pH, dissolved oxygen, and temperature P1; b) P2





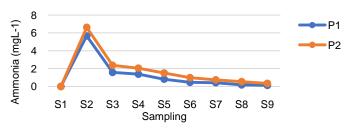


Figure 6 – P1 and P2 ammonia measurements from sampling 1 to 9



There was a spike in ammonia levels in the above graphs in both treatments 1 and 2 increasing from the first sampling to the second sampling, namely after the addition of flocforming materials in P1 and the addition of poultry manure in P1. The addition of organic materials, both floc-forming materials and poultry manure, contributes to the N element and there has been no N breakdown by both heterotrophic bacteria (biofloc) and autotrophic bacteria (conventional).

### DISCUSSION OF RESULTS

When compared to conventional treatments, the biofloc treatment's total length calculation yields longer results. When compared to biofloc (P1), the conventional treatment (P2) calculates the average weight at a higher rate. The overall weight calculation revealed variations, particularly in treatment 2, or the fourth through ninth samplings, which were influenced by water quality parameters. According to Qin *et al.* (1998), stocking density is another environmental element (together with water quality) that affects growth and survival rates; the higher the stocking density, the greater the impact on these parameters. A high stocking density will restrict movement space, according to Hidayatullah *et al.*, (2015), which supports this claim.

The biofloc method used in P1 was able to maintain ammonia limits within certain levels that can be tolerated by snakehead fish fry when compared to conventional methods that are prone to uncontrolled ammonia fluctuations (Avnimelech, 1999; Ekasari, 2014). The results of the calculation of survival (SR) in both treatments 1 and 2 are closely related to ammonia levels. Ammonia (NH<sub>3</sub>) was a waste product of protein metabolism and was toxic to fish (Boyd & Tucker 1998). Because too high ammonia can inhibit its excretion through the gills, resulting in accumulation in the blood and reduced affinity of haemoglobin to oxygen, causing death (Nirmala *et al.*, 2012), as well as an increase in white blood cells (Supriyono *et al.*, 2010; Supriyono *et al.*, 2011). Although snakehead fish tend to be more tolerant of ammonia levels above the threshold (Bijaksana, 2014; Qin *et al.*, 1997), it will affect their appetite which has an impact on growth.

For snakehead culture, the water quality parameters of pH, dissolved oxygen, and general temperature continue to fall within the normal range (KKP, 2014). The link between the two parameters is directly proportional, meaning that the ammonia level rises with increasing acidity and vice versa. The pH level can influence the ammonia concentration. According to Surbakti (2015), the optimal pH for snakehead fry growth and survival was 6–6.5. The inclusion of probiotics, which can break down ammonia into nitrite before turning it into nitrate (Ekasari, 2014; Hartini *et al.*, 2013), had an impact on the reduction in treatment 1. According to Avnimelech. (1999) and Ekasari. (2014), protein breakdown from leftover feed and fish waste caused the ammonia that was present in the aquaculture environment.

The growing biofloc were expected to be able to assimilate ammonia waste so that the ammonia in the cultivation media becomes low and suitable for the growth of cultured fish. Figure 5 shows the highest ammonia and increased sharply at the  $2^{nd}$  sampling, namely after the provision of materials to grow flocs and grow natural food before stocking fish, namely 5.66 mg L<sup>-1</sup> in treatment I and 6.617 mg L<sup>-1</sup> in treatment 2, then gradually decreased to the lowest point at the 9<sup>th</sup> sampling with a value of 0.122 mg L<sup>-1</sup> in P1 and 0. 337 mg L<sup>-1</sup> in P2 a higher ammonia value was seen in P2, namely in the treatment that was not inoculated with commercial heterotrophic bacteria (only relying on bacteria that naturally exist in the media), while the P1 treatment was added to the inoculation of commercial heterotrophic bacteria with a dose added to the water of 1 mL 50 L<sup>-1</sup> water so that for a volume of 20 L of water, 1.2 mg of probiotics were added.

The addition of floc-forming materials in P1 and poultry manure in P2 contributed to the increase in ammonia, where the breakdown of N organic matter in the cultivation media into ammonia was quite high and has not been balanced with the rate of ammonia assimilation by heterotrophic bacteria which causes the ammonia in the media to increase sharply. Taking the 10<sup>th</sup> sample, the ammonia value in P1 is 0.078 mg L<sup>-1</sup> which was a value close to the required standard of <0.02 mg L<sup>-1</sup>, while in P2 it is still above, namely 0.132 mg L<sup>-1</sup>. The



same thing was also revealed by Usman *et al.*, (2011) which revealed a spike in Total Ammonia Nitrogen (TAN) and the increase was higher in the treatment without heterotrophic bacteria when compared to the treatment that added heterotrophic bacteria.

Treatment 1 with the addition of heterotrophic bacteria showed lower ammonia levels compared to treatment 2, indicating the effect of heterotrophic bacteria can contribute to the decrease in ammonia (Badjoeri & Widiyanto, 2008). The results of the analysis showed that ammonia influenced the length gain in treatment 1 (P1) by 62.9%, while in treatment 2 (P2) by 67.5% higher treatment 2 (P2) when compared to treatment 1 (P1) and together pH, dissolved oxygen, temperature, and ammonia although not significant, influenced the weight gain in treatment 2 by 74.4%, higher treatment 1 (P1) when compared to treatment 2 (P2), although the ammonia value in some sampling was above the required value, according to Wise (2010); Qin *et al.*, 1998; Crab *et al.*, 2012 snakehead fish had a fairly high tolerance level to ammonia levels that are above the average threshold. The tolerance value of snakehead fish to dissolved ammonia levels in water at different pH is at ammonia concentrations of more than 0.54 mg L<sup>-1</sup> at pH 8.0 to 1.57 mg L<sup>-1</sup> at pH 10.0 (Qin *et al.*, 1997 in Extrada *et al.*, 2013).

From the second sampling to the third sampling, there are variations in the temperature measurement graph. The weight gain graph (Figure 1) illustrates how this is impacted by temperature, which accelerates metabolism and impacts fish hunger, which in turn affects fish weight. Following that, a study by Qin *et al.*, (1998) found a significant correlation between temperature and the eating behavior of snakehead fry, with warmer temperatures resulting in more active eating than cooler temperatures.

Fluctuations in dissolved oxygen levels were quite high; it was strongly influenced by the stocking density and microorganisms that consume oxygen. The results of the analysis showed that the dissolved oxygen variable did not affect length gain in treatment 1 (P1) or treatment 2 (P2) (p>0.05). Snakehead fish can take oxygen directly from the surface (Bijaksana, 2014), so low dissolved oxygen content did not have a significant impact on the survival of the fish. These results are following research conducted by Purnamawati *et al.*, 2017, namely snakehead fry without aeration provides growth results, survival rates, and even higher feed efficiency when compared to aeration. According to Kordi (2010), the ideal dissolved oxygen level for snakehead fish was 3-6 mg L<sup>-1</sup>. The results of data analysis using a partial test (t-test) showed that the dissolved oxygen variable did not affect length gain in treatment 1 (P1) and treatment 2 with (p>0.05), while pH, temperature, and ammonia had affected on length gain in treatment 1 (P1) and treatments 1 (P1) and 2 (P2) (p<0.05), while dissolved oxygen, temperature, and ammonia affect weight gain in treatments 1 (P1) and 2 (P2) with (p>0.05).

Simultaneous test (F-test) on water quality parameters showed that the parameters of pH, dissolved oxygen, temperature, and ammonia together affect the length gain in treatment 1 (P1) and 2 (P2), and affect the weight gain in P1 but not in P2. The coefficient of determination test (R<sup>2</sup>) on water quality parameters pH, dissolved oxygen, temperature, and ammonia affects the length gain in treatment 1 (P1) of the fry respectively by 46.7%, 12.3%, 4.3%, and 62.9%, while in treatment 2 (P2) respectively by 48.1%, 9.9%, 14.1%, and 67.5%. Together pH, dissolved oxygen, temperature, and ammonia affect the increase in length in treatment 1 by 90.7% and treatment 2 by 95.2%.

Water quality parameters namely pH, dissolved oxygen, temperature, and ammonia affect weight gain in treatment 1 (P1) of snakehead fry respectively by 26.7%, 14.8%, 1.9%, and 63% while together pH, DO, temperature, and ammonia although not significant, affect weight gain in treatment 1 by 81%, while in treatment 2 (P2) respectively by 21.2%, 4.8%, 0.6%, and 50.5% and together pH, dissolved oxygen, temperature, and ammonia, affect weight gain in treatment 2 by 74.4%.

Based on the independent t-test on water quality in terms of pH and DO, it shows that the mean water quality in terms of pH and dissolved oxygen in treatments 1 and 2 was different, with the mean pH in treatment 2 higher than in treatment 1, while the mean dissolved oxygen in treatment 1 was higher than treatment 2, these results was following the



research conducted by Shamsuddin *et al.*, (2022). Water quality in terms of temperature and ammonia levels showed that the mean water quality in terms of temperature and ammonia levels in treatments 1 and 2 were the same. The length gain of the snakehead fry shows that the mean length growth of the fry in treatment 1 and treatment 2 was the same, while the weight gain of the fry shows that the mean weight in treatment 1 and treatment 2 was the same.

## CONCLUSION

pH, dissolved oxygen, temperature, and ammonia together affect the length gain of the snakehead fry in biofloc and conventional treatments and affect the weight gain only in biofloc. Dissolved oxygen did not affect the length gain of the fry in both biofloc and conventional treatment. pH did not affect the weight gain of the fry in both biofloc and conventional treatment The biofloc method was able to suppress the increase in pH. Rearing of snakehead fry using biofloc and conventional methods resulted as no significant difference in mean temperature, ammonia, length, and weight growth.

## ACKNOWLEDGMENTS

Thank you to the Mandiangin Freshwater Aquaculture Center for help during this research.

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