



# Effectiveness Test of Aquaculture Systems: Green Water, Biofloc, and Aguaponic Systems on the Stress Response of Snakehead Fish (Channa striata)

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#### **ABSTRACT**

Water quality is a key factor in the cultivation of snakehead fish (Channa striata) because it can influence the level of physiological stress that impacts the health and growth of the fish. Three cultivation systems commonly used to manage water quality sustainably are the green water system, biofloc, and aquaponics. All three have different approaches to recycling waste and maintaining the balance of the cultivation environment. This study aims to compare the effectiveness of the three systems in maintaining water quality and reducing physiological stress in snakehead fish. The method used was a Completely Randomized Design (CRD) with three treatments and three replications. The parameters observed included levels of ammonia, nitrite, nitrate, dissolved oxygen (DO), and blood glucose levels as an indicator of stress. The results showed that the biofloc system produced the lowest blood glucose level (54 mg/dL), as well as the lowest nitrite and nitrate concentrations compared to the other treatments. Meanwhile, the biofloc system green water produces ammonia and dissolved oxygen (DO) levels, but biofloc is the most stable in maintaining water quality and the health of snakehead fish.

### INTRODUCTION

The fisheries sector plays a significant role in providing food and animal protein for the community (Dwinafiah & Hasan, 2023). One way to strengthen aquaculture is by developing local fish that have high economic value and are easy to cultivate. Snakehead fish (Channa striata) is a popular freshwater fish, both for fresh consumption and as a raw material for the food industry (Mijani et al., 2014). This potential makes snakehead fish a leading commodity for cultivation.

Aquaculture is the primary solution to ensuring a sustainable supply of snakehead fish, but its implementation still faces several technical and environmental challenges. One challenge in intensive aquaculture is the high production of aquaculture waste, particularly organic waste and inorganic nitrogen, such as ammonia. Ammonia is a toxic compound produced from leftover feed, feces and fish metabolism (Hapsari *et al.*, 2021). High ammonia concentrations can disrupt the balance of the surrounding ecosystem (Futri *et al.*, 2025), endanger fish health and even cause fish mortality (Nugroho & Rivai, 2019).

Increased ammonia concentrations can trigger physiological stress in fish, characterized by increased cortisol levels (Tang *et al.*, 2018), blood glucose levels (Parikesit, 2022), and hematocrit levels (Wahyu *et al.*, 2015). Stressed fish will experience physical and behavioral changes, such as decreased appetite and darkening of body color (Firdaus *et al.*, 2018) and become more susceptible to disease due to a decreased immune system (Pratama *et al.*, 2022). Therefore, innovative aquaculture systems are needed that not only increase productivity but also reduce waste and control stress in fish. Several waste assimilation-based cultivation systems have been developed as environmentally friendly solutions, such as green water systems, biofloc, and aquaponics.

The application of green water system cultivation technology utilizes the presence of microalgae such as phytoplankton to absorb waste compounds in water (Treece, 2019), the biofloc system uses heterotrophic bacteria that process organic waste into additional nutrients that can be reused by fish (Roy et al., 2022), and the aquaponic system combines fish and plant cultivation, where plants utilize cultivation waste as a natural fertilizer to support their growth (Roy et al., 2020). These three systems are able to reduce ammonia concentrations and improve water quality (Rizki et al., 2024), thus potentially suppressing stress responses in fish. However, no research has yet observed and compared the effectiveness of green water, biofloc, and aquaponic systems on the stress response of snakehead fish. Therefore, this study aims to determine the effectiveness of these three systems in improving water quality through the parameters of ammonia, nitrite, nitrate, and dissolved oxygen (DO), as well as observing the stress response of snakehead fish (Channa striata) by measuring blood glucose levels.

#### **METHODS**

## Place and Time of Research

The research was conducted in the Outdoor Laboratory of the Fisheries Science Study Program, University of South Sumatra, in Sungai Dua Village, Banyuasin Regency, in February 2024.

# **Tools and Materials**

The tools used in this research included a digital blood glucose tester, spectrophotometer, DO meter, aeration kit, pump, net, container box, scale, ruler, measuring cup, sieve, PVC pipe, net pot, rockwool, and hose. The materials used included snakehead fish fry, commercial feed, spirulina seeds and liquid organic fertilizer, probiotics containing *Lactobacillus casei* and *Saccharomyces cerevisiae* bacteria, molasses, and bok choy (bok choy) seeds.

This study used a Completely Randomized Design (CRD) with three treatments: green water, biofloc, and aquaponics, each consisting of three replications, with snakehead fish (*Channa striata*) measuring 6–7 cm at a stocking density of 2 fish/L.

## **Research Procedures**

## Establishing a Green Water System

The process of establishing a green water system begins with the addition of 8 g/m³ of urea fertilizer and 200 g/m³ of dolomitic lime to the rearing tank. The pond is then filled with

20 liters of clean water and 1,000 mL of cultured *Spirulina platensis* seeds are added, or approximately 10–20% of the water volume, according to Buwono & Nurhasanah (2016). The pond is provided with stable aeration and left for 7 days to allow optimal microalgae growth (Buwono & Nurhasanah, 2018).

# 2. Establishing a Biofloc System

The biofloc system is prepared by adding 10 mL/L of probiotics and 200 mL/L of molasses as a carbon source to a 20-liter container of water. The mixture is then allowed to stand for 7 days to allow floc formation (Putra *et al.*, 2017). This process also involves adding nitrogen and phosphorus sources from feed, as well as heterotrophic bacteria from probiotics. Pond designs for green water and biofloc systems are shown in Figure 1.

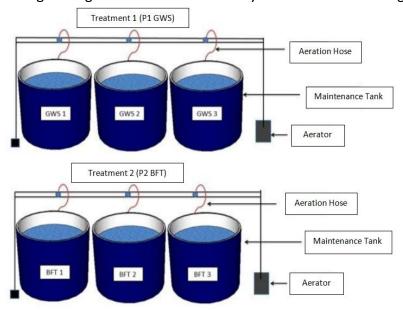


Figure 1. Green Water and Biofloc System Pond Design

## 3. Making an Aquaponic System

The aquaponics system was prepared by installing a water recirculation system using pipes and hoses designed with 18 holes for net pots in each pond, arranged in a tiered manner. Pot trays were prepared as seedling containers, filled with moistened rockwool pieces. Then, well-grown seedlings were selected and transferred to the net pots. Water was supplied using a 20-liter pump. The pond design is shown in Figure 2.

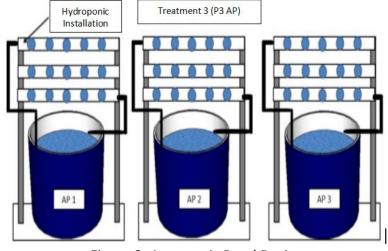


Figure 2. Aquaponic Pond Design

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#### 4. Acclimatization

The prepared media is then used to spread 6-7 cm snakehead fish seeds with a density of 2 fish/L. Acclimatization period lasts for 5 days (Niti *et al.*, 2024).

# 5. Stress Observation and Response (Blood Glucose)

Observations included stress responses and water quality. Parameter observations were conducted over 60 days of maintenance. Feeding was carried out at a rate of 5% of the total biomass per day, three times daily at 8:00, 12:00, and 16:00 WIB. DO measurements of the maintenance water were carried out daily in the morning and afternoon at 6:00 and 17:00 WIB. Meanwhile, ammonia, nitrite, and nitrate levels were measured every 10 days at 17:00 WIB.

Stress response testing through blood glucose testing was conducted at the beginning and end of the study. Blood glucose measurements were performed using a digital blood glucose meter. The device inserted a glucose strip into the device, then waited for the blood image to appear. A blood sample was then dripped onto the strip and the results, expressed in mg/dl, were displayed on the screen (Panuntun, 2023).

# **Data Analysis**

Blood glucose levels, an indicator of fish stress response, were analyzed using analysis of variance (ANOVA) at a 95% confidence level. If results showed significant differences, further testing was performed using the Tukey test. Water quality data were analyzed descriptively and processed using Microsoft Excel. The input data were then displayed in graphical diagrams. Data analysis used Microsoft Office Excel and SPSS 23.0 software.

#### **RESULTS**

# **Blood Glucose**

The results of statistical analysis using ANOVA showed that the use of different cultivation systems had a significant effect (p<0.05) on the stress response of snakehead fish based on blood glucose levels. The best treatment was obtained in the biofloc system with the lowest blood glucose levels, indicating the lowest level of fish stress. The graph of the results of blood glucose level measurements as an indicator of fish stress is shown in Figure 3.

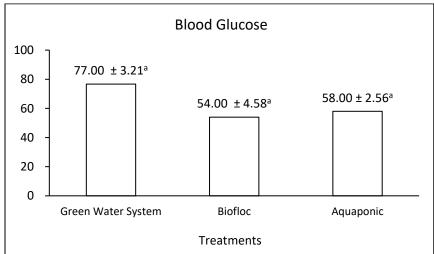
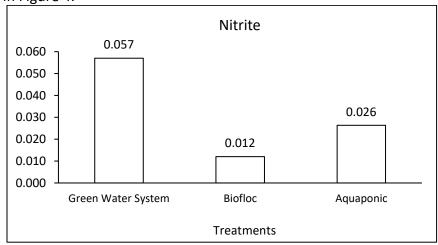


Figure 3. Blood Glucose

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# Water Quality Nitrite

Nitrite measurements indicate that differences in culture systems influence nitrite levels in the rearing medium, which impacts the stress response of snakehead fish. The biofloc system performed best with the lowest nitrite levels, indicating the lowest stress levels in the fish. A graph of the results of nitrite measurements, as a contributing factor to stress in fish, is presented in Figure 4.



Picture 4. Nitrite Levels

## **Nitrate**

Nitrate measurements indicate that differences in culture systems influence nitrate levels in the rearing medium, which impacts the stress response of snakehead fish. The biofloc system performed best with the lowest nitrate levels, indicating the lowest stress levels in the fish. A graph of the nitrate measurement results, as a contributing factor to stress in fish, is presented in Figure 5.

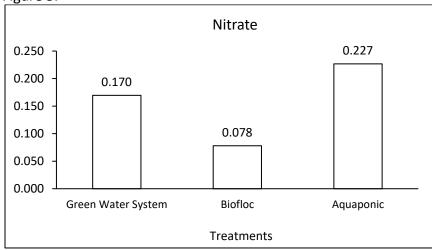


Figure 5. Nitrate Levels

### **Ammonia**

The results of ammonia measurements indicate that differences in cultivation systems affect ammonia levels in the rearing medium, which impacts the stress response of snakehead fish. Green water showed the best results with the lowest ammonia levels, indicating the lowest levels of fish stress. A graph of the results of measuring ammonia levels as a factor causing stress in fish is presented in Figure 6.

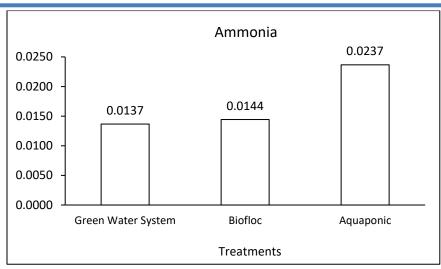


Figure 6. Ammonia Levels

# **Dissolved Oxygen (DO)**

The results of dissolved oxygen level measurements indicate that differences in cultivation systems affect the availability of oxygen in the rearing medium, which impacts the stress response of snakehead fish. Green water showed the best results with the lowest dissolved oxygen levels between the biofloc and aquaponic cultivation systems. The graph of the results of measuring dissolved oxygen levels as one of the factors causing stress in fish are presented in Figure 7.

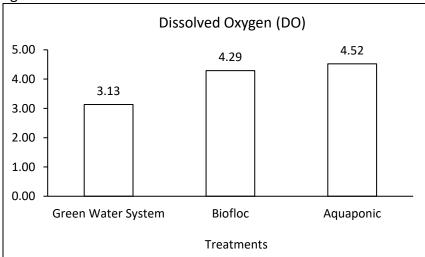


Figure 7. Dissolved Oxygen (DO) Levels

#### DISCUSSION

The results of the study showed that differences in green water, biofloc, and aquaponic cultivation systems significantly affect water quality and the physiological stress response of snakehead fish (*Channa striata*). Blood glucose levels are used as a physiological indicator to assess the level of stress experienced by fish (Yunus *et al.*, 2020). The higher the glucose level, the higher the fish's stress response to environmental conditions. This is supported by Nasichah *et al.* (2016) who explained that when fish experience stress, stimuli are received by sensory receptors and transmitted to the hypothalamus, which then triggers chromaffin cells to release catecholamine hormones. This hormone inhibits insulin secretion, so that glucose

cannot enter cells optimally and causes an increase in blood glucose levels. The results of the glucose level study showed the highest glucose level in the green water system, namely 77.00 mg/dL, indicating that the fish experienced higher stress than the other two systems. The biofloc system showed the lowest glucose level, namely 54.00 mg/dL, indicating that the cultivation environment in the biofloc system is relatively more stable and supports optimal physiological conditions of snakehead fish.

The physiological stress level of fish is greatly influenced by water quality, especially the content of nitrogen compounds such as nitrite ( $NO_2^-$ ) and ammonia ( $NH_3/NH_4^+$ ) which are toxic and dangerous for fish (Dewantoro *et al.*, 2020). In this study, the lowest nitrite levels were found in the biofloc system, at 0.012 mg/L, while the highest levels were found in the aquaponics system, at 0.044 mg/L. The low nitrite levels in the biofloc system indicate that the inorganic nitrogen conversion process by heterotrophic microorganisms is optimal. Bacteria such as *Nitrosomonas* and *Nitrobacter* has an important role in nitrification which converts ammonia into nitrite and then into nitrate (Fadillah *et al.*, 2022).

Nitrate levels, the end result of the nitrification process, were also found to be lowest in the biofloc system at 0.078 mg/L and highest in the green water system at 0.219 mg/L. Nitrate levels are less hazardous than nitrite and ammonia, but high concentrations over the long term can still have harmful effects on fish. The high nitrate levels in the green water system are thought to be caused by the slower nitrate uptake by phytoplankton compared to ammonia and nitrite. Nitrate serves as the primary nutrient source for phytoplankton growth and is used in the formation of organic matter through photosynthesis (Yuliana *et al.*, 2012).

Ammonia is a highly toxic compound to fish, even at low concentrations. Research shows that the green water system has the lowest ammonia levels at 0.0137 mg/L, followed by the biofloc system at 0.0144 mg/L, and the aquaponics system at 0.0237 mg/L. Although the green water system is chemically more effective in suppressing ammonia accumulation, fish stress levels in the green water system are higher. This is likely due to unstable water quality fluctuations and uncontrolled microalgae growth, which affect the stability of the microenvironment in the culture medium. In aquaponics systems, ammonia accumulation can occur due to an imbalance between ammonia production from fish metabolism and the plant's ability to absorb it as a nutrient. When the number of plants is too small or their growth rate is slow, ammonia utilization is not optimal, resulting in the accumulation of ammonia compounds in the culture medium.

Based on the analysis results in Figure 7, the highest dissolved oxygen (DO) value was found in the aquaponics system, at 4.52 mg/L, which is thought to be due to the plants' contribution to oxygen production through photosynthesis. This indicates that water quality cannot be determined by a single parameter alone, but rather by a combination of various chemical and biological factors. This is in line with research by Effendi (2003) in Mado *et al.* (2023) which states that dissolved oxygen levels will fluctuate daily and seasonally, depending on the mixing and movement of water masses, photosynthetic activity, respiration and waste entering the water body.

Overall, the biofloc system showed the most effective cultivation in reducing stress in snakehead fish and maintaining more stable water quality compared to the green water and aquaponics. The effectiveness of the biofloc system comes from its ability to utilize the activity of heterotrophic and autotrophic microorganisms to convert organic waste into floc and use it as fish feed (Wanja *et al.*, 2020), as well as increasing the efficiency of nutrient utilization and reducing environmental stress through more stable water quality control (Zaidy & Eliyani, 2021).

### **CONCLUSION**

This study demonstrated that the biofloc system was most effective in improving water quality by reducing ammonia, nitrite, and nitrate levels and maintaining stable DO levels. This system also produced the lowest stress response in snakehead fish, as indicated by lower blood glucose levels compared to green water and aquaponic systems.

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