

Nile tilapia (*Orechromis niloticus*) productivity performance in small-scale recirculating aquaculture systems (RAS) with different biofilter media

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Abstract. To increase sustainable aquaculture production, recirculating aquaculture systems (RAS) are among the most widely used aquaculture production systems. This research aims to determine the performance of small-scale RAS with different biofilter media. The current research was carried out in 4 sets of RAS, with different treatments. The treatment is differentiated based on the biomedia used, namely: pumice, moving bed bioreactor (MBBR), bioball, and no media as control treatment. Nile Tilapia (*Orechromis niloticus*) are used as testing organisms. Fish performance will be measured by analyzing the specific growth rate (SGR), the food conversion ratio (FCR), and the survival rate. The water quality data was descriptively analyzed to see trends in the recirculating system. The results showed that the MBBR treatment provided the best values for the parameters reflecting the performance of fish growth (SGR $3.24\pm0.26\%$ and FCR 1.30 ± 0.18), the water quality, and the degree of ammonia purification (4.16%), which were significantly better than in the control treatments. It is recommended to use MBBR in the biofilter for the best performance in small-scale RAS for fish farming. This research will be the first step in developing small-scale RAS designs, especially for aquaculture activity in developing countries. **Key Words**: aquaculture production system, biofilter, purification, water quality, sustainable.

Introduction. Currently, the world population is continuously growing. By 2050, the world population is estimated to reach almost 10 billion people (United Nations 2022). With the increase in the world population, if the ability to produce more food does not increase by up to 50%, it is estimated that in the future our food will not be able to meet the needs of the human population (Searchinger et al 2018). Among the foods currently needed, protein is one of the important nutrients. Based on (Boyd et al 2022), fish is a source of good quality protein in quantity and quality. In general, fish commodities can come from capture fisheries and aquaculture activities. However, due to pollution, overfishing, etc., currently captured fisheries are considered to be less sustainable. On the other hand, aquaculture is known to continue to grow and, in proportion, it already produces more than 50% of the total fish consumed worldwide (FAO 2022). Moreover, aquaculture production is currently predicted to increase more than 2 times by 2050, compared to the aquaculture production in 2010 (Searchinger et al 2018).

Aquaculture, although it is one of the best alternatives for fish production, also has several problems. For example, in terms of excessive use of water resources and wastewater. Globally, aquaculture requires more than 100 km³ year⁻¹ of water (McNevin & Boyd 2014). Aquaculture waste, which is dominated by organic materials, is also

known to pollute the environment (Ahmad et al 2022). Therefore, because the need for intensification and expansion of aquaculture is urgently needed, innovation in terms of the aquaculture production system must be developed to reduce the drawback of conventional aquaculture activities.

A closed aquaculture system is a cultivation system that separates the environment of the cultivation container from the outside environment (Oyinlola 2019). This system can be a solution to conventional aquaculture because it tends to be more efficient in water use and limits the volume of aquaculture waste (Ghamkhar et al 2021). Currently, the most widely applied closed aquaculture systems are biofloc technology (BFT) and recirculating aquaculture systems (RAS). Biofloc technology utilizes heterotrophic bacteria to recycle nitrogen waste in aquaculture ponds. However, BFT requires high technical capabilities and still experiences problems with a high total suspended solid (TSS), which in some cases has a negative impact on the cultured organism.

Unlike BFT, RAS is an aquaculture production system that reuses cultivation water through the filtration process (Badiola et al 2012). So, in practice, the water filtration system is one of the things that needs to be focused in this system. One of the problems often faced with RAS is the high investment cost compared to other aquaculture production systems (Timmons & Ebeling 2010; Badiola et al 2012). This will be a significant problem, particularly in developing countries, where aquaculture activities are mostly carried out on a small-scale.

In general, the most important thing in the aquaculture system is the management of nitrogen/ammonia waste, which is very toxic to the cultured organism. Ammonia can cause physiological changes, oxidative stress, inflammatory, and immunosuppressive effects in farmed fish (Elshopakey et al 2023). In RAS, ammonia waste is handled using the biofilter (Suriasni et al 2023). Biofilters are used to increase nitrifying bacteria, which convert toxic ammonia into less toxic nitrate, through the nitrification process (Gonzalez-Silva et al 2016; Suriasni et al 2023). In practice, the requirement for a biofilter is the availability of media with a large enough specific surface area (SSA), as an accessible pass through for water (Kordkandi et al 2018).

In this research, a small-scale recirculating aquaculture system was built, that can be implemented with lower investment costs and can be applied to the urban farming. This research aimed to determine the performance of RAS with different biofilter media, based on fish growth and water quality. This research will be the first step in small-scale RAS design, which can be an alternative technology that is accessible to all groups, including fish farmers in developing countries.

Material and Method

RAS design. This research was carried out at the Teaching Farm of the Department of Fisheries, Faculty of Agriculture, Universitas Gadjah Mada, Indonesia. This research was carried out in 4 sets of RAS. Each set consists of four fish maintenance tanks (500 L each); these four maintenance tanks have an outlet with a central drain design, which is then connected to the set of filter tanks by a pipe (2 inch). The filter consists of four smaller tanks (each 120 L) with a specific purpose. The first tank is a radial flow settler tank, followed by a mechanical filter tank with 5 kg of fish nets as its main media, followed by a biofilter tank, and ends up in the sump tank. The water from the sump tank is then flowed back into the maintenance tank using the water pump (Luckiness 107 pump). Each maintenance container will receive water from the sump tank with a discharge value maintained at 3.6 L minute⁻¹. The existence of filter tanks will manage the water quality conditions to meet the water needs of tilapia. The treatment that will be used in this research is based on the different types of biomedia in each biofilter. The first RAS set functions as a control treatment (without adding biomedia). Then there are three other treatments using pumice stone (Pumice), Kaldnes moving bed bioreactor (MBBR), and bioball as biomedia. All biomedia were maintained in a volume of 30 L. The RAS design is presented in Figure 1.

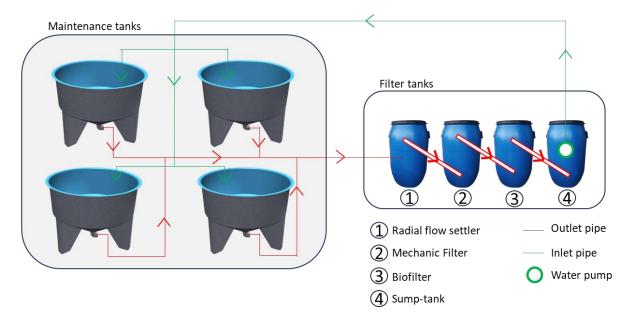


Figure 1. Scheme of a set of recirculating aquaculture system.

Test specimens and their maintenance. A total of 40 individual Nile tilapia (7.20 \pm 0.6 cm; 6.80 \pm 1,4 g) were placed in each maintenance tank. The tilapia was obtained from a local farm originating in the Special Region of Yogyakarta, Indonesia. Tilapia was chosen because it represents a freshwater commodity that is widely traded worldwide and also the main local commercial commodity. Before being distributed to the maintenance tank, the tilapia fish are first acclimated in a cage placed in a large concrete pond for 7 days. The tilapia fish are then fed ad satiation during the acclimatization process. The research was carried out for 56 days and tilapia was given food 5% of its biomass per day. The feeding frequency is maintained two times a day, at 08:00 and 16:00. The fish are weighted every 20 days to adjust the daily feeding dose.

Measured parameters. The performance of the recirculating system is reviewed from several parameters, including the degree of purification, which can be calculated using the formula below (Timmons & Ebeling 2010):

$$C_p = \frac{C_{in} - C_{out}}{C_{in}} \times 100\%$$

Where:

Cp - the degree of purification;

C_{in} - Value of water quality parameters (TAN and Turbidity) entering the filter chambers; C_{out} - Value of the water quality parameters that exit the filter chambers.

Water quality parameters such as ammonia (NH_3), total ammonia nitrogen ($TAN=NH_3+NH_4$), dissolve oxygen, pH, temperature, ammonia, and turbidity will be measured every 7 days to determine the performance of the system. Water quality measurement is done using an Aquatroll 500 multiparameter sonde. The fish growth performance will be measured by analyzing the specific growth rate (SGR), the Food Conversion Ratio (FCR) and the survival rate of tilapia.

Data analysis. The water quality data was descriptively analyzed to see trends in the recirculating system. The values for the degree of purification and tilapia performance were then tested for normality and homogeneity of the data. When the data met the requirements for homogeneity and normality, a parametric ANOVA test was performed to see if there are differences between the treatments. When the data did not meet homogeneity and normality, it was analyzed using the nonparametric test of Kruskal-

Wallis. The statistical analysis was carried out using the GraphPad Prism software (Version 9.5.1).

Results

Growth, food conversion ratio and survival rate. Based on the SGR value, it can be seen that tilapia in MBBR treatment generally had the highest growth $(3.24\pm0.26\% d^{-1})$ compared to other treatments. Meanwhile, the lowest growth rate was in the control treatment (2.60 \pm 0.40% d⁻¹). Statistical ANOVA followed by the Tukey test revealed a significant difference between the SGR value of the control and MBBR treatment (P=0.035), while the pumice and bioball treatments were not significantly different from the other treatments. The same pattern can also be seen in the FCR value, where MBBR treatment generally has the most effective feed utilization compared to other treatments, with an average FCR value reaching 1.30±0.18. Meanwhile, the average FCR in other treatments was greater than 1.5, indicating an inefficient use of the feed. According to the ANOVA test, there were significant differences in FCR values between treatments (P=0.022), in particular between control and MBBR treatments. Meanwhile, the pumice and bioball treatments generally did not show significant differences compared to the other treatments. Based on survival rate, there was no significant difference in value between treatments based on the nonparametric Kruskal Wallis test (P=0.916), with values ranging between 96-98%. The growth parameters, feed conversion ratio and survival rate can be seen in Table 1.

Table 1

Growth parameter,	food	conversion	ratio	and	survival	rate value
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Parameters	Control	Pumice	MBBR	Bioball
Initial weight (g)	6.9±1.03	6.5±0.91	6.88±0.21	6.88±0.57
Final weight (g)	29.50 ± 4.08^{a}	36.42±3.92 ^{ab}	42.73±7.06 ^b	37.75±3.83 ^{ab}
SGR (% d ⁻¹)	2.60 ± 0.40^{a}	3.07±0.25 ^{ab}	3.24±0.26 ^b	3.04 ± 0.14^{ab}
FCR (ratio)	1.98±0.30ª	1.6 ± 0.28^{ab}	1.30 ± 0.18^{b}	1.58 ± 0.25^{ab}
Survival rate (%)	96.88±3.15	97.50±2.89	98.13±2.39	96.25±4.33

Differences in superscripted letters on the same row indicate significance based on the parametric ANOVA test and followed by the Tukey test, at a 95% confidence level.

Water quality. On average, the TAN concentration was highest in the control treatment, with an average of 1.81 ppm, a lowest concentration of 0.02 ppm (day 1) and a highest one of 3.23 ppm (day 56). This was then followed by pumice (1.56 ppm), Bioball (1.56 ppm) and MBBR (1.49 ppm) treatments. On the basis of the temporal trend, TAN concentration tends to increase as the number of days of treatment increases. However, we can see several decreasing TAN concentration trends in each treatment, which happened on day 14 to day 21 and day 35 to day 42. The similar temporal trend was also found in the ammonia parameter, where the concentration tends to increase in the beginning of treatments but decreases in the middle of treatments. Based on the average concentration, the treatment with the highest ammonia content was also in the control treatment with an average concentration of 0.15 ppm (and values ranging from 0.00 to 0.2 ppm), followed by bioball (0.15 ppm), MBBR (0.13 ppm) and pumice (0.12 ppm).

DO concentrations, in general, had a decreasing trend. The highest average DO concentration was in MBBR treatment, with an average of 6.84 ppm and values ranging from 6.42 to 7.20 ppm. The other treatments, ordered by decreasing values of the oxygen content in a decreasing order are: bioball (6.76 ppm), pumice (6.60 ppm) and control (6.51 ppm). Temporal trends in the form of decreasing values can also be found in the water pH. This is indicated by the range of pH values that were alkaline at the beginning of the study (8.6) and gradually became slightly more acidic, close to 7.9–8.0, in almost all treatments.

There is a tendency for the temperature to increase with the increase of the number of treatment days, but the range is not too large, only 25-28°C. A temporal

increase trend can also be seen in the turbidity value, which generally ranged from 0.6 NTU on the first day to 8 NTU at the end of the study. In general, the highest average turbidity was in the control and bioball treatments, with a value of 3.7 NTU, and the lowest was in the MBBR treatment, with an average turbidity of 2.45 NTU. The temporal trend of the water quality parameters can be seen in Figure 2.

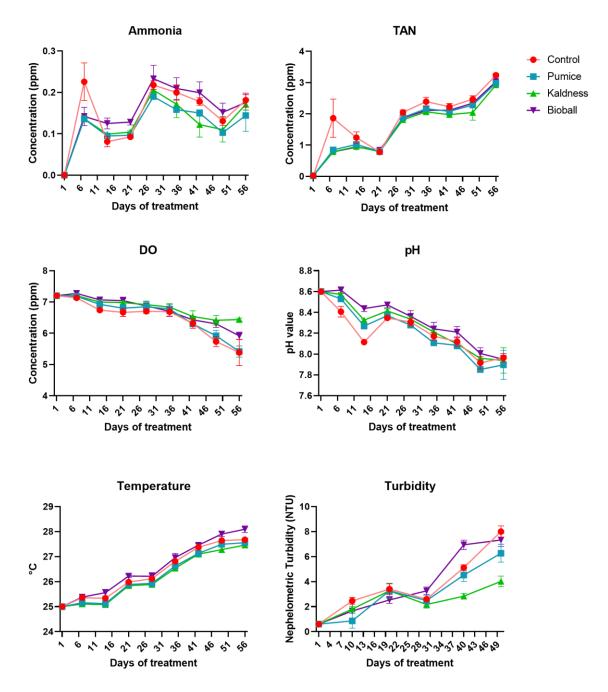


Figure 2. The temporal trend of water quality in maintenance tanks.

Degree of purification. Based on turbidity purification efficiency, it can be seen that MBBR treatment provides the highest average degree of purification (17.24%) compared to the other treatments. Meanwhile, in the control treatment, in general there is no reduction in the turbidity value, so the control treatment was the only treatment that gave a negative value for the degree of purification (-15.38%). Meanwhile, for the degree of purification in TAN, a similar pattern was also observed where MBBR treatment gave the best degree of purification (4.16%) compared to other treatments, while the

control treatment also provided the lowest degree of purification (0.40%) compared to the other treatments.

Davi		Degree of purification (%)							
Day —	Control	Pumice	MBBR	Bioball					
	Turbidity								
d10	-31.03	2.33	16.90	12.99					
d20	-14.56	15.90	30.88	3.22					
d40	-20.37	-3.54	21.13	25.73					
d50	4.46	14.04	0.06	15.36					
Mean	-15.38±14.88	7.18±9.34	17.24±12.86	14.33±9.24					
	Total ammonia nitrogen								
d7	1.14	6.62	3.39	1.34					
d14	-0.3	6.01	3.91	5					
d21	0.06	2.68	1.38	0.25					
d28	0.87	3.9	4.25	1.31					
d35	0.41	3.54	8.32	3.12					
d42	0.05	2.91	4	0.24					
d49	0.59	3.81	6.58	1.59					
d56	0.42	1.2	1.46	1.14					
Mean	0.40 ± 0.47	3.83±1.76	4.16±2.36	1.75±1.59					

The value of degree of purification based on turbidity and TAN on RAS

Table 2

Discussion. The recirculating aquaculture system is currently one of the production systems that has been developed to overcome classic aquaculture problems (Badiola et al 2012; Shitu et al 2022). The RAS is currently considered as very efficient in terms of water use. In recent years, climate change and variability have become one of the most important environmental issues (Froehlich et al 2022). For example, Indonesia, as one of the world's leading producers of aquaculture commodities, is impacted by the El Nino phenomenon. Therefore, various locations in Indonesia have a tendency to experience drought phenomena (Avia et al 2023), worsened by the existence of another environmental problem, namely water sources pollution in Indonesia (Wicaksono et al 2016; Wicaksono et al 2021; Wicaksono 2022). Thus, the water resources that needed for aquaculture purposes are deficient both in terms of quantity and quality. The development of an aquaculture production system that is efficient in water use is important.

In this research, a small-scale RAS was designed that can be used as a solution to develop an aquaculture production system. The variety of biomedia in the RAS design is one of the main focuses in this research. Biomedia efficiency is important due to the accumulation of TAN as aquaculture waste (Shitu et al 2022). The presence of TAN, in general, can cause several adverse impacts on the fish production (Elshopakey et al 2023). This waste can be neutralized by changing the form of TAN to nitrate, which is a less toxic compound to the aquatic environment. The process of changing TAN to nitrate is called the nitrification process, and this process occurs in the biomedia that exist in the biofilter tank (Gonzalez-Silva et al 2016). Therefore, looking for the best biomedia can be the first step in developing a small-scale RAS for aquaculture production.

In this study, it can be observed that the RAS with the best performance, in terms of SGR and FCR, was in the MBBR treatment, significantly better than in the control, according to the ANOVA statistical test. This indicates that MBBR treatment generally leads to a better overall health of the tilapia. Consequently, fish do not need to spend more energy for adaptation and homeostasis to the environment, so the energy can be used for growth purposes. This can also be seen in terms of water quality: the MBBR treatment has the better water quality, indicated by lower ammonia, TAN and turbidity values. This is also supported by the tendency for oxygen to be higher in MBBR treatment compared to other treatments. Based on the temporal trend in the ammonia and TAN parameters, it can be seen that there was an increase in concentration after adjustments were made to the biomass calculation in the pond. Therefore, on days 20 and 40 there is an increase in the food dose given to the tilapia, leading to an increase in the ammonia and TAN values in the days afterward. However, the biofilter's ability to carry out nitrification also tends to increase in the following days after the trend of increasing ammonia concentration. One of the factors that can speed up the nitrification process is the presence of the ammonia concentration itself (Dodds et al 2017). When ammonia is available in large quantities, it becomes an energy source for nitrifying bacteria to reproduce, and this phenomenon will increase the effectiveness of nitrification process. This condition leads to a decreasing trend in ammonia and TAN to be seen after an increasing trend occurs around days 20 and 40. The concentration of ammonia in the form of NH₃ also tends to decrease due to the decreasing trend of pH as maintenance time increases (Boyd & Tucker 1998). A decrease in pH will determine the form of nonionized ammonia (NH₃) to shift to the NH₄ form. Ammonia in the form of NH₄ tends to be less toxic to fish(Suriasni et al 2023).

Based on the temporal trend, it can also be seen that there is an incrementing trend of temperature as the maintenance time goes by. This could be caused by the air temperature in the research area. The experiment was carried out during the dry season. Even though the maintenance tank is located semi-outdoor, an increase of air temperature can still happen, and lead to an increment of water temperature. Increasing the temperature of the water can also have an impact on the increase in metabolism in fish, which can then lead to an increased oxygen consumption and CO₂ release in the water (Islam et al 2019). This can then have an impact on the decreasing trend of oxygen and pH in the water. Overall, an increase of 10°C in water temperature, will double the metabolism process. A decrease in oxygen concentration in water can also be caused by an increase in the tilapia biomass during maintenance, so that the volume of oxygen consumed also becomes larger and the oxygen concentration in the tank becomes lower.

Based on the degree of purification, MBBR treatment provides the best purification efficiency among other treatments, both based on turbidity and TAN value. Although the purification value was still lower compared to the research carried out by Pfeiffer & Wills (2011), which has a TAN removal efficiency of on average 12-14%, a high waste load is still entering the system. Biomedia stimulates the development of nitrifying bacteria colonies by providing media with a large Specific Surface Area (SSA) value (Jiang et al 2019). Therefore, in practice, increasing the volume of biomedia can be an alternative to increase the degree of purification value because there will be more space for the nitrifying bacteria to settle. In this study, the SSA value in the MBBR biomedia can reach up to 690 m² m⁻³, higher than in pumice (225 m² m⁻³) and in bioball (210 m² m⁻³) (Santos-Pereira et al 2019; Shehab et al 2021). The MBBR treatment also has the advantages of a dynamic biomedia (always moving). This movement can prevent solid waste from settling in the biomedia. In practice, preventing the accumulation of solid waste on the biomedia may also cause a decrease in turbidity, thus, the degree of purification value in the MBBR treatment tends to be higher compared to the other treatments. The positive degree of the purification value, in terms of turbidity, in the bioball and pumice treatments shows that biofilter media can reduce turbidity, to a certain degree. However, more research is needed to verify the role of biofilters in reducing water turbidity.

Conclusions. Based on the results obtained, the MBBR treatment provides the best performance for fish growth, water quality, and degree of purification, among all treatments. Therefore, it is recommended to use a biofilter using the MBBR method to provide optimal results in the preparation of a small-scale RAS design for fish farming.

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