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The Effectiveness of Three Biofilter Media on Total Ammonia Nitrogen (TAN) Removal and Survival Rate of Tilapia Gift Seeds (*Oreochromis niloticus*) in Recirculation Aquaculture System

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ABSTRACT

Efforts to increase tilapia production are carried out through intensive culture by taking into account various aspects that support fish survival. The culture system that supports intensive culture is the Recirculation Aquaculture System (RAS). The RAS technology has the ability to support aquaculture with very high density and high yields compared to open culture systems. This technology must be added with a biofilter to reduce toxic ammonia because of its high density, so research is carried out using several biofilter media which have become important biofilter media for both RAS in freshwater and seawater. This study aims to determine the effect of different biofilter media in RAS on decreasing Total Ammonia Nitrogen (TAN) concentration and survival rate of tilapia seeds. The method used was experimental with three treatments and four replications. Tilapia with an average individual weight of 3.40 ± 0.15 g were maintained in RAS with three different biofilter media treatments, sand (A), polystyrene microbeads (B) and kaldnes (C). The parameters observed were TAN removal efficiency, specific growth rate (SGR) and survival rate (SR). The results showed that different biofilter media had a significant effect (*P <0.05*) on the TAN removal efficiency value but had no significant effect (*P> 0.05*) on the SGR and SR values. The sand for biofilter media treatment (A) gave the best TAN removal efficiency of 36.61±4.82%.
Key words: kaldnes, polystyrene, sand, TAN, tilapia

ABSTRAK

Upaya peningkatan produksi ikan nila dilakukan melalui budidaya secara intensif dengan memperhatikan berbagai aspek yang menunjang kelangsungan hidup ikan. Sistem budidaya yang mendukung budidaya intensif adalah Sistem Budidaya Resirkulasi (Recirculation Aquaculture System; RAS). Teknologi RAS memiliki kemampuan untuk mendukung budidaya dengan kepadatan yang sangat tinggi dan hasil yang tinggi dibandingkan dengan sistem budidaya terbuka. Teknologi ini harus dilengkapi dengan biofilter untuk mengurangi amonia yang beracun karena kepadatannya yang tinggi, sehingga dilakukan penelitian dengan menggunakan beberapa media biofilter yang telah menjadi media biofilter penting, baik untuk RAS di air tawar maupun air laut. Penelitian ini bertujuan untuk mengetahui pengaruh media biofilter yang berbeda dalam RAS terhadap penurunan konsentrasi Nitrogen Amonia Total (Total Ammonia Nitrogen; TAN) dan kelangsungan hidup benih ikan nila. Metode yang digunakan adalah eksperimen dengan tiga perlakuan dan empat ulangan. Ikan nila dengan bobot individu rata-rata 3,40 ± 0,15 g dipelihara pada RAS dengan tiga perlakuan media biofilter yang berbeda, yaitu pasir (A), polystyrene microbeads (B) dan kaldnes (C). Parameter yang diamati adalah efisiensi penurunan konsentrasi TAN, laju pertumbuhan spesifik (SGR) dan tingkat kelangsungan hidup (SR). Hasil penelitian menunjukkan bahwa media biofilter yang berbeda berpengaruh nyata (P<0,05) terhadap nilai efisiensi penurunan konsentrasi TAN tetapi tidak berpengaruh nyata (P>0,05) terhadap nilai SGR dan SR. Pasir untuk perlakuan media biofilter (A) memberikan efisiensi penyisihan TAN terbaik sebesar 36,61±4,82%.
Kata kunci: kaldnes, polystyrene, pasir, TAN, nila

1. Introduction

Tilapia (*Oreochromis niloticus*) is a freshwater fish species that is tolerant of brackish and marine waters with a salinity of up to 20 ppt. Tilapia production in the world has continued to increase for almost a decade (Arini *et al*., 2018). Tilapia is one of the freshwater commodities that is most in demand by various groups, both local and foreign people (Putra *et al*., 2011; Fadri *et al.,* 2016). Tilapia is one of the important commodities and is widely cultivated in the world (Shiau and Chin, 1999). Tilapia production in the world has increased considerably within 6 years. In 2010, tilapia production was 2,537,445 tons, while at the end of 2016 the production increased to 4,199,556 tons (FAO, 2019). Indonesian tilapia

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production ranks third after China and Egypt (FAO, 2007). Tilapia production in Indonesia has increased every year. Production has almost doubled in 6 years. In 2012, tilapia fish production was 695,063 tons, while in 2017 it reached 1,265,201 tons (KKP, 2018).

Efforts to increase tilapia production are carried out through intensive cultivation by paying attention to various aspects that support fish survival such as water availability, cultivation areas and good environmental quality (Putra *et al*., 2011; Hapsari *et al*., 2020a; Hapsari *et al*., 2020b). Intensive cultivation is characterized by high density and can encourage an increase in feeding which can lead to an increase in waste from leftover feed and fish feces (Harwanto and Jo., 2010; Setijaningsih and Umar, 2015). Intensive fish farming is indeed more efficient in producing fish, but the resulting waste will also increase. Aquaculture activities will produce waste from feed residue and fish metabolism containing ammonia.

A problem that is often encountered in intensive fish farming is the rapid accumulation of waste from feed residue and fish metabolism (Setijaningsih and Umar, 2015; Dwiputra *et al*., 2021). Intensive cultivation also produces chemical wastes such as nitrogen and phosphate elements contained in the culture media water. An increase in density and length of time to keep fish will be followed by an increase in the levels of ammonia in the water (Avnimelech, 2006; Shafrudin *et al*., 2006; Harwanto *et al*., 2010). Fish excrete 80-90% ammonia through osmoregulation, feces and urine (Shafrudin *et al*., 2006). Ammonia that is not oxidized by bacteria for a long time and continuously will be toxic to cultured fish (Chen and Kou, 1993; Benli and Koksal, 2005; Dauhan *et al*., 2014; Norjanna *et al*., 2015).

The cultivation system that can be carried out for intensive cultivation is by applying the Recirculating Aquaculture System (RAS). The RAS technology has the ability to support cultivation activities with a very high density and high yields compared to open cultivation systems (Klanian and Adame, 2013). The water in this system is reused by using waste treatment media such as biological filters to control and stabilize water conditions, reduce the amount of water used and increase fish survival (Martins *et al*., 2010).

Efforts to reduce ammonia in intensive aquaculture with RAS can be done by adding a biofilter to the system to remove ammonia which is toxic to fish. The biological filtration process is the most important thing in recirculation culture systems (Miller and Libey, 1985; Losordo *et al*., 1998; Harwanto and Jo, 2010; Isroni *et al*., 2019, Oktavia *et al*., 2021). Biological filters in the recirculation system should use media that is capable of providing a suitable environment for the growth of nitrifying bacteria. Sand, polystyrene microbeads and kaldnes are generally used as biofilter media and have become important biofilter media, both in freshwater RAS (Harwanto *et al*., 2011a) or sea water RAS (Harwanto *et al*., 2011b).

Sand is a natural biofilter media that has been proven to be successfully used as a biofilter media to maintain the water quality of cultivation media (Shnel *et al*., 2002; Summerfelt, 2006; Harwanto *et al*., 2011a; Mulyadi *et al*., 2014). Polystyrene microbeads have wide commercial use among a wide variety of artificial filter media (Greinner and Timmons, 1998; Malone and Beecher, 2000; Malone and Pfeiffer, 2006; Timmons *et al*., 2006). These filters are easy to find, lightweight and floatable. Kaldnes is an artificial plastic filter media that has been widely researched in the last decade (Kessel *et al*., 2010; Harwanto *et al*., 2011a; Harwanto *et al*., 2011b; Pfeiffer and Wills, 2011; Lebrero *et al*., 2014; Patroescu *et al*., 2015; Elliott *et al*., 2017) and has recently continued to be used (Kholif and Febrianti, 2019; Muhtaliefa *et al*., 2019, Sandy *et al*., 2019), so that kaldnes need to be developed and more introduced as a bofilter media. One of the advantages of this filter media is its specific shape and allows bacteria to grow, but the Kaldnes has a relatively small SSA (Rusten *et al*., 2006) compared to some other media such as sand (Summerfelt, 2006; Harwanto *et al*., 2011a; Harwanto *et al*., 2011b) and polystyrene microbeads (Greinner and Timmons, 1998).

This study was conducted to determine the effectiveness of kaldnes, polystyrene microbeads, and sand in reducing TAN concentration, and further, to determine the effect of biofilter media on the survival and growth of tilapia reared in the RAS. The research was conducted in December 2019 - February 2020 at the Fish Seed Center, Mijen Semarang, Central Java. The research was conducted with a fish maintenance period for 30 days. The TAN analysis was carried out in the water quality laboratory, Faculty of Fisheries and Marine Sciences, Universitas Diponegoro.

2. Materials and Methods

2.1. Materials

Preparation of culture containers

The maintenance containers used were 12 rectangular plastic tubs with a size of 63 ×

46 × 38 cm^3. Filled plastic tube with water a height of 30 cm with a volume of water as much as 80 L. Cleaned plastic tube first before filling it with water. After being filled with water, it is left for 24 hours so that the substances contained in the water are deposited (Cromey *et al*., 2002; Ardita *et al*., 2015).

Preparation of the filter container

The filter container used is a modified 4" PVC pipe with a diameter of 11.4 cm and a height of 43 cm. Each filter container has a filter or buffer. There are 12 filter containers used with the same volume (Greinner and Timmons, 1998; Harwanto *et al*., 2011a; Harwanto *et al*., 2011b). Filter containers with a capacity of 4 L were placed next to each plastic tub for tilapia gift seed rearing. Filled each of the 4 filter containers with biofilter media of sand, polystyrene microbeads and kaldnes which are drained using water pump to grow bacteria. The water pump used for the water rotation process is equipped with a hose that connects the water pump and the water inlet pipe to the filter.

Biofilter conditioning

The biofilter conditioning carried out in this study aim to grow nitrifying bacteria before RAS is used for fish cultivation (Harwanto *et al*., 2011a; Filliazati *et al*., 2013; Hastuti *et al*., 2014). In this study, bacteria were grown using fish culture wastewater at BBI Mijen. Cultivation waste is flowed through the biofilter to form a biofilm layer on the biofilter media. The process of bacterial growth is carried out for two weeks until the microorganism grow on biofilter media (Yani *et al*., 1998; Harwanto *et al*., 2011b; Filliazati *et al*., 2013; Nugroho *et al*., 2014). At this stage, microorganism is naturally attached to biofilter media by flowing wastewater continuously on the filter that already contains the biofilter media (Nugroho *et al*., 2014). If the media has formed a layer of brownish black mucus and is not easily separated from the media, it can be ascertained that microorganisms have grown on the media (Filliazati *et al*., 2013).

Preparation of test fish and acclimatization

The test fish used in this study were 420 healthy gift tilapia fish seeds with an average individual weight of 3.40 ± 0.15 g / fish. The total fish biomass used was 1,428 g. The containers used were 12 plastic tubs and each container will be stocked with 35 gift tilapia seeds with a density of 1,488 g m^{-3}. The stocking density used in this recirculation system is higher than the stocking density used in the study of Kawser *et al*. (2016) who used a density of 567 g m^{-3} with an average individual weight of 3.40 ± 0.3 g / fish. The test fish were acclimatized for seven days and put in the experimental container in the form of 12 plastic tubs with artificial feeding in the form of pellets twice a day.

Weighing absolute weight and absolute length measurement to determine the growth of fish was carried out at the beginning of the study and at the end of the study. After the acclimatization period was completed and before the study began, all tilapia seeds were fasted for one day and weighed (Harwanto *et al*., 2011b). This is also done at the end of the study to determine the growth rate.

Feeding management

During 30 days the maintenance of tilapia seeds, feeding is carried out twice a day, namely at 08.00 and 16.00 (Belal, 1999; Shiau and Chin, 1999; Lim *et al*., 2001; Takeuchi *et al*., 2002; Benli and Koksal, 2005; Adewolu, 2008; Solomon and Boro, 2010; Ayisi *et al*., 2017). Feeding was done using the fix feeding rate method. The amount of feed given was 3% of the weight of tilapia seed biomass per day (Lim *et al*., 2001; Mbahinzireki *et al*., 2001; Takeuchi *et al*., 2002; Adewolu, 2008).

2.2. Methods

Water quality

The parameters of water quality observed were ammonia concentration, temperature, pH and dissolved oxygen (DO) levels. Water quality measurements were carried out at two points in each maintenance container, namely water in the inlet and outlet of the biofilter (Harwanto *et al*., 2011b; Norjanna *et al*., 2015; Muhtaliefa *et al*., 2019). Measurement of temperature, pH and DO is carried out every morning and evening at 08.00 and 16.00 (Karisa *et al*., 2005; SNI.6141 , 2009; Mulyani *et al*., 2014; Norjanna *et al*., 2015; Mulqan *et al*., 2017). Temperature and DO are measured using a DO meter, while pH is measured using a pH meter. Analysis of TAN concentration was carried out every seven days using the spectrophotometric method in the Water Quality Laboratory, Faculty of Fisheries and Marine Sciences, Universitas Diponegoro.

Efficiency removal of Total Ammonia Nitrogen (TAN) concentration

Biofilter efficiency can be determined by calculating the removal in TAN concentration, which is calculated using the formula from Shete *et al*. (2017); Muhtaliefa *et al*. (2019);

Hapsari *et al*., 2020a; Hapsari *et al*., 2020b and Dwiputra *et al*. (2021), i.e:

$$\text{Removal Efficiency} = \frac{\text{TANin - TANout}}{\text{TANin}} \times 100\%$$

Note:
Removal Efficiency = The efficiency of removal TAN concentration (%)
TANin = the concentration of TAN in the biofilter inlet (g m^{-3})
TANout = the concentration of TAN in the biofilter outlet (g m^{-3})

Volumetric TAN Removal (VTR)

Measurement of biofilter performance parameters can be seen from the removal rate of volumetric TAN on culture media which is expressed as grams of oxidized TAN per volume of media per day (g m^{-3} d^{-1}) (Shete *et al*., 2017). Furthermore, Summerfelt and Vinci (2004) stated that the important factors to be considered in the biofilter design are the mass of TAN removed per day and the efficiency of TAN removal from the biofilter.

The VTR calculation formula used by Malone and Beecher (2000), Malone and Pfeiffer (2006), Harwanto *et al*. (2011b), Peiffer and Wills (2011), Shete *et al*. (2017) and Oktavia *et al*., 2021 are as follows:

$$\text{VTR} = \text{Kc} \times (\text{TANin - TANout}) \frac{\text{Qr}}{\text{Vb}}$$

Note:
VTR = The removal rate of TAN (g m^{-3} d^{-1})
Kc = Unit conversion factor =1.44
TANin = concentration of TAN at the outlet biofilter (g m^{-3})
TANout = concentration of TAN at the outlet biofilter (g m^{-3})
Qr = The rate of water flow through the filter (m^3 day^{-1})
Vb = Total volume of filter media (m^3)

Specific growth rate (SGR)

The method used to determine the SGR of reared fish is by calculating the percentage difference between the final weight and the initial weight divided by the length of time for raising fish. Growth is a process of increasing length and weight of an organism which can be seen from changes in length and weight in units of time. The SGR calculation formula used by Tekinay and Davies (2001), Radhakrishnan *et al*. (2016) and Luo *et al*. (2017) are as follows:

$$\text{SGR } (\%\ day^{-1}) = \frac{(\ln Wt - \ln Wo)}{T} \times 100\%$$

Note:
SGR = Specific growth rate (% day^{-1})
Wt = Average fish weight at the end of the study (g)
Wo = average fish weight at the start of the study (g)
T = Length of maintenance (days)

Survival rate (SR)

The method used to determine the SR of reared fish seeds is by comparing the number of fish seeds that are alive at the end of maintenance with the number of fish seeds that are stocked at the beginning of maintenance (Saputra *et al*., 2013). This formula is used to determine the percentage of fish survival and is also used by other researchers such as Tekinay and Davies (2001); Radhakrishnan *et al*. (2016); Luo *et al*. (2017) and Saravanan *et al*. (2018) are as follows:

$$\text{SR} = \frac{\text{Nt}}{\text{No}} \times 100\%$$

Note:
SR = Survival rate (%)
Nt = Number of fish that live at the end of maintenance (fish)
No = number of fish that live at the beginning of the maintenance (fish)

Data Analysis

The results of efficiency of TAN removal data, VTR, SGR and SR data were analyzed using ANOVA analysis variance. Before the ANOVA analysis variance was performed, the data was first tested for normality, homogeneity and additivity tests to determine that the data were normal, homogeneous and additive for further testing, namely ANOVA

Table 1. Water Quality Results of Tilapia Gift (*O. niloticus*) in Cuture Media for 30 Days

Parameter	Water Quality			Tolerance Value
	Filter Media			
	Sand	Polystyrene microbeads	Kaldnes	
Temperature (°C)	24.8-31.5	24.8-31.6	24.8-31.7	25.0-32.0*
pH	6.5-7.5	7.0-7.6	7.0-7.5	6.5-8.5*
DO (mg L^{-1})	3.11-5.84	3.10-5.60	3.10-5.86	≥ 3.00**

*SNI.7550 (2009)
**Crab *et al*. (2007)

Table 2. Results of Total Ammonia Nitrogen (TAN) Inlet and Outlet Value Range for 30 Days

Parameter	Filter Media			Tolerance value
	Sand	Polystyrene microbeads	Kaldnes	
TAN Inlet (mg L^{-1})	0.054-0.278	0.040-0.382	0.054-1.487	< 1.5*
TAN Outlet (mg L^{-1})	0.000-0.239	0.000-0.351	0.034-1.459	

*Crab *et al*. (2007)

Table 3. The Efficiency of Total Ammonia Nitrogen (TAN) Removal Between Treatments

Week	Efficiency of TAN Removal (%)		
	Filter Media		
	Sand	Polystyrene microbeads	Kaldnes
I	29.64	22.33	3.33
II	39.29	16.17	18.24
III	40.30	36.34	15.62
IV	37.22	22.13	12.93
Mean±SD	36.61±4.82[a]	24.24±8.55[b]	12.53±6.51[c]

Note: The mean value for each treatment with different superscripts letter indicates a significant difference ($p < 0.05$).

analysis variance. After the analysis of variance was performed, if a significant effect was found ($P<0.05$), then Duncan's test must be carried out to determine the differences between treatments (Srigandono, 1981). The water quality data obtained were analyzed descriptively.

3. Results and Discussion

3.1. Result

Water quality

The water quality observed in this study were temperature, pH, DO and TAN. Measurements of temperature, pH and DO were carried out every day. The frequency of temperature, pH and DO measurements were carried out twice a day, in the morning and evening. The results of water quality measurements during the observation are presented in Table 1.

The results of water quality measurements in the form of temperature, pH and DO during the observation showed that the temperature values obtained in the tilapia rearing container were in the range of 24.8 to 31.7 ° C, the pH range obtained was 6.5 to 7.6 and DO water is in the range of 3.11 to 5.86 mg L^{-1}. The water quality values of pH and DO in tilapia rearing ponds are in a good range to be used as a medium for tilapia rearing, while the minimum range of temperature values in the rearing medium is not in accordance with the feasibility value.

Based on Table 2, it can be seen that the range value of TAN for each treatment is different. The highest inlet and outlet TAN values were found in the treatment with kaldnes (C) biofilter media, namely 1.487 mg L^{-1} and 1.459 mg L^{-1}, respectively. While the lowest inlet and outlet TAN values were 0.278 mg L^{-1} and 0,239 mg L^{-1}, respectively in the treatment using sand biofilter media (A). Furthermore, the results of the efficiency of TAN removal between treatments are presented in Table 3.

Based on Table 3, it can be seen that the average value of the lowest TAN concentration removal efficiency among the three treatments was obtained in treatment C using a kaldnes biofilter, which was 12.53%. Meanwhile, treatment A had the highest TAN removal efficiency value, namely 36.61%.

Volumetric TAN removal (VTR)

The VTR value was obtained based on the calculation of the TAN removal value from each treatment. Based on Table 4, it can be seen that based on the test results for four weeks, the highest VTR value was found in the treatment using sand biofilter media (A) with a value of 76.85 ± 11.14 g m^{-3} d^{-1}. While the lowest VTR value was 50.75 ± 4.90 g m^{-3} d^{-1} which was found in the treatment using Kaldnes (C) biofilter media.

The highest VTR values at weeks 1, 2, 3 and 4 were also achieved in the treatment using a sand filter (A) with VTR values of

Table 4. Results of Volumetric TAN removal (VTR) Every Week

Week	VTR (g m^{-3} day^{-1})		
	Filter Media		
	Sand	**Polystyrene microbeads**	**Kaldnes**
1	64.46	57.83	45.18
2	72.29	65.66	48.80
3	80.12	72.89	52.41
4	90.53	78.92	56.63
Mean±SD	76.85±11.14[a]	68.83±9.11[a]	50.75±4.90[b]

Note: The mean value for each treatment with different superscripts letter indicates a significant difference ($p < 0.05$).

Table 5. Results of the Specific Growth Rate (SGR) of Gift Tilapia (*O. niloticus*)

Replication	SGR (%day^{-1})		
	Filter Media		
	Sand	**Polystyrene microbeads**	**Kaldnes**
1	2.75	2.84	1.96
2	2.59	1.95	2.22
3	2.73	2.96	2.68
4	1.97	1.91	2.36
Mean±SD	2.51±0.37[a]	2.41±0.56[a]	2.30±0.30[a]

Note: The mean value for each treatment with different superscripts letter indicates a significant difference ($p < 0.05$).

64.46, 72.29, 80.12 and 90.53 g m^{-3} d^{-1}, respectively. While the lowest VTR value was found in the treatment using the kaldnes filter (C) with a value of 45.18, 48.80, 52.41 and 56.63 g m^{-3} d^{-1}, respectively.

Specific growth rate (SGR)

The value of the specific growth rate is calculated based on the added value of tilapia seed weight. There is no significant difference from all these values. The results of SGR during the observation are presented in Table 5.

Based on Table 5, it can be seen that the result of SGR for each treatment is not significant different. The value shows that the SGR in this study was 2.51 ± 0.37 % day^{-1} in A (sand biofilter), 2.41 ± 0.56 % day^{-1} in B (polystyrene microbeads biofilter) and 2.30 ± 0.30 % day^{-1} in C (kaldnes biofilter).

3.2. Discussion

Water quality

Water quality is one of the most influential factors on tilapia seeding activities. Water quality that is not suitable for fish can inhibit aquaculture and fish growth. Based on the observations, it was obtained water quality data during maintenance, namely temperature values of 24.8-31.7 ° C, pH values of 6.5-7.6 and DO 3.10-5.86 mg L^{-1}. According to Sayed and Kawanna (2008), temperature is one of the most important factors affecting the physiology, growth, reproduction and metabolism of tilapia. The results of the minimum temperature limit in this study are lower than the good temperature range for tilapia maintenance according to SNI.7550 (2009), namely 25-32 °C. The temperature of 24.8 °C was obtained in the morning measurement at 08.00. This is presumably due to low weather conditions along with the rainy season. According to Mulyani *et al*. (2014), that water temperature can affect fish appetite. High temperatures will increase the appetite for fish and lower temperatures can cause the digestive process and metabolism of fish to slow down. The pH and DO values in this study are also in accordance with the SNI.7550 (2009) which mentioned that good pH and DO in tilapia maintenance activities are 6.5-8.5 and ≥ 3 mg L^{-1}, respectively. Based on this, the pH and DO values of the study this is suitable for use for tilapia fish maintenance activities.

Intensive cultivation with high stocking density and high feeding can increase the culture waste from feed residue and fish metabolism which can cause the water quality of the culture media to decrease. Based on this study, the highest average total ammonia nitrogen (TAN) inlet and outlet results were obtained in the treatment using Kaldnes biofilter (C). The highest average inlet and outlet TAN in treatment C were 0.760 and 0.735 mg L^{-1}. The lowest average TAN value was found in the treatment using a sand biofilter (A) with an inlet and outlet TAN value of 0.104 and 0.063 mg L^{-1}. Treatment with polystyrene microbeads biofilter media (B) had an average TAN inlet and outlet of 0.112 and 0.072 mg L^{-1}, respectively. This is thought to be related to the SSA value. The greater the SSA of the biofilter media, the more metabolic waste is broken down. This is consistent with Lekang and Kleppe (2000) who reported that filter media that has a large surface area can cause more bacteria to grow. The abundance of bacteria can result in an increase in ammonia reduction per unit filter volume. In addition, a filter media with a large SSA can also be effective because it takes up less space for a large decrease in ammonia.

The results of the highest range of inlet and outlet TAN values were 1.487 mg L^{-1} and 1.459 mg L^{-1} in the treatment using Kaldnes filter (C) at week I or day 7. This is thought to be the cause of the low SR value in treatment C, namely 67.14 ± 8.57%. Except for these values, the TAN values of each treatment in this study were classified as suitable for use in tilapia culture. Losordo *et al*. (1998) stated that the TAN content in recirculation systems using biofilter media should be <1 mg L^{-1}. Furthermore, Crab *et al*. (2007) mentioned that the TAN content in fish culture should not be more than 1.5 mg L^{-1} because it will be toxic to cultured fish.

Volumetric TAN removal (VTR)

The VTR value in this study ranged from 45.18 to 90.53 g m^{-3} d^{-1}. The highest VTR was obtained in treatment using sand biofilter (A). The VTR value at week 1, 2, 3 and 4 were 64.46; 72.29; 80.12, and 90.53 g m^{-3} d^{-1}, respectively. The increase in VTR value at week 3 to week 4 was greater than that of the previous weeks. This is presumably due to the growth of fish, so that the fish weight increases. Along with the increase in fish weight, the feed given increases and the resulting fish metabolism will also increase. This causes increased the TAN and VTR values during maintenance. According to Avnimelech, (2006) and Shafrudin *et al*. (2006), the longer the fish culture time will be followed by an increase in the levels of ammonia in the water. This is in accordance with Shafrudin *et al.* (2006) who stated that fish will release 80-90% of ammonia in waters through the osmoregulation process, through feces and through urine.

The calculation of the VTR value is used as the main indicator to evaluate the performance of one biofilter media with another. The VTR value can also be used to determine the efficiency of TAN reduction which indicates the ability of the biofilter media. Based on the results of the study, the VTR value of 45.18-90.53 g m^{-3} d^{-1}. was lower than that of Summerfelt (2006), which used a sand biofilter with a VTR value ranging from 140-170 g m^{-3} d^{-1}. presumably because the sand biofilter used has a higher SSA, namely 4,000-20,000 m^2 m^{-3}. This is in accordance with Timmons *et al*. (2006) which mentioned that the VTR value is influenced by the SSA size of the biofilter media used. In addition, the TAN concentration in Summerfelt (2006) was also greater, between 1.07-1.68 mg L^{-1}. This is consistent with Malone and Beecher (2000) who reported that the VTR value will increase with increasing TAN concentration.

The VTR results in this study were also lower than that of Harwanto *et al*. (2011b), i.e. 142.6 - 193.8 g m^{-3} d^{-1}. They used the same biofilter (sand, polystyrene microbeads and kaldnes). This is presumably because the sand and polystyrene microbeads media biofilter used have a greater SSA, namely 7,836 and 3,287 m^2 m^{-3}, respectively (Timmons *et al.*, 2006). In addition, the size of the fish used was also larger (29 g per fish), resulting in a higher TAN concentration (Malone and Beecher, 2000).

Specific growth rate (SGR)

Growth is a process of increasing the length and weight of an organism which can be seen from changes in length and weight in units of time. The specific growth rate is a biological parameter that shows the growth rate of the fish being reared. Fish growth can be influenced by internal factors and external factors. Fish growth will only occur if the energy content in feed exceeds the energy needed for body maintenance and replacing damaged cells (Ayisi *et al*., 2017). In addition, the quality of water for good maintenance media can also support the growth of cultured fish optimally (Mulqan *et al*., 2017).

The SGR values of tilapia obtained in the treatment using sand biofilter (A), polystyrene microbeads biofilter (B) and kaldnes biofilter (C) were 2.51 ± 0.37; 2.41 ± 0.56 and 2.30 ± 0.56% day^{-1}, respectively. The three values do not show a significant difference between treatments. This is presumably because the three types of biofilter media are able to maintain water quality with good filtration in the maintenance medium, so that the fish have a high appetite and can use feed energy for their growth. This can be seen from the efficiency value of the reduction in TAN concentration in the three treatments. This is in accordance with Mulqan *et al*. (2017) which mentioned that the optimal filtration process will produce good water quality in tilapia fish culture media and can determine the amount of feed that is suitable for fish needs.

Even though in the first week the TAN value on the C treatment medium exceeded 1.0 mg L^{-1} and approached 1.5 mg L^{-1}, it soon decreased in the following weeks. With the improved water quality, it is suspected that the fish appetite has also improved, which in the end, the SGR value of treatment C is also quite good, the same as the SGR value in other treatments. According to Zonneveld and Fadholi (1991), fish appetite is determined by water quality conditions and fish appetite is also a control factor as well as a major factor for fish growth.

The SGR results obtained in all treatments in this study were better than those of Kawser *et al*. (2016). Their study obtained an SGR of 1.50 ± 0.01% day^{-1} with the same weight of each tilapia (3.40 g) in the recirculation system. The SGR value of this study is similar to that of the study by Mulqan *et al*. (2017), who obtained a value of SGR 2.36 ± 0.79% day^{-1} with a weight of 7.16 g each tilapia seeds in the recirculation system.

Survival rate (SR)

The survival value is the ratio of the number of fish alive from the start to the end of the rearing. Factors that greatly affect the survival of tilapia are feed and environmental conditions. Feeding with the right amount and time must be considered in fish maintenance. This is in accordance with Rejeki *et al.* (2019) which states that in order to prevent a decrease in water quality which will disrupt fish health and cause death, it is necessary to pay attention to the amount and frequency of feeding in fish maintenance. This is also in accordance with Ndome *et al.* (2011) who mentioned that feeding with sufficient quality and quantity as well as good environmental conditions can support the survival of tilapia.

The treatments carried out in this study did not significantly affect the survival value of tilapia seeds. The SR value of each treatment, namely treatment A 84.29 ± 12.01%, treatment B 83.57 ± 10.27% and treatment C 67.14 ± 8.57%. The SR values of tilapia in treatment using sand biofilter (A) and polystyrene microbeads bofilter (B) are in accordance with the provisions of SNI.6141 (2009) which states that the survival of tilapia with a minimum size of 5 cm is 70%. The efficiency value of the reduction in TAN and VTR concentrations affected the SGR and SR yields of tilapia seeds.

The SR values of treatment using sand biofilter (A) is 84.29 ± 12.01% and 83.57 ± 10.27% for using polystyrene microbeads bofilter (B) obtained in this study were better than those of Sayed and Kawanna (2008). In their study, the SR of tilapia seeds obtained in an aquaponic recirculation system with a temperature of 32 ° C was 74 ± 5.04%. However, the survival results obtained in this study were lower than those of Ridha and Cruz (2001) who obtained the SR value of tilapia in the recirculation system of 97.6%; and also, lower than the study of Putra *et al*. (2011) with the best SR value of tilapia seeds obtained was 88% in a recirculation system using zeolite filters, Chinese pond mussel, and lettuce plants.

Treatment C has an SR value of 67.14 ± 8.57% which is not in accordance with the SNI.6141 (2009), namely the SR value of at least 70%. Tilapia in this treatment experienced death in the first week of culture. This is presumably because in the initial week of culture, the TAN value of the culture medium was high (1.487 mg L^{-1}) so that the water quality decreased and resulted in the death of the fish. This is in accordance with Rejeki *et al.* (2019), Hapsari *et al*. (2020); Oktavia *et al*. (2020) and Dwiputra *et al*. (2021) who mentioned that fish becomes stressed when the water quality decreases, so that fish are prone to death.

4. Conclusion

Based on this research, the use of different biofilter media in the recirculation system had a significant effect on the removal of TAN concentration and had no significant effect on the specific growth rate and survival rate of *O. niloticus* seeds. The sand biofilter showed the highest removal value among other. However, all of biofilter media tested were able to manage the TAN value to remain below 1 mg L^{-1} after the first week, and were able to maintain the water quality at the optimum value for tilapia culture.

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