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Microbubble Aeration in A Recirculating Aquaculture System (RAS) Increased Dissolved Oxygen, Fish Culture Performance, and Stress Resistance of Red Tilapia (*Oreochromis* sp.)

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Abstract

Microbubble aeration has been recognized as a tool to improve dissolved oxygen and fish culture performance in limited water aquaculture. This research aims to investigate the effect of microbubble aeration on increasing dissolved oxygen, fish culture performance, and stress resistance of red tilapia. The study used 3 treatments of aerations namely microbubble, blower, and without aeration with 3 replicates each. Red tilapia weighing 115.27±3.93 g were stocked in a plastic tank of 800 L water volume at density 50 individual/tank and cultured for 50 days in a recirculating aquaculture system (RAS). Dissolved oxygen, fish culture performance, and stress resistance were evaluated. Dissolved oxygen was monitored daily. In the middle of the research period, a stress test was performed using different salinity at 12 and 24 ppt. Statistical analysis was subjected to water quality, fish culture performance, and stressed resistance. The microbubble aeration was able to stabilize DO level on 4.28 mg/L until the end of the experiment and suppressed the CO_2 and ammonia content. Fish biomass was higher in microbubble treatment, with a lower FCR value, lower stress level indicating fish in good health and promoting good culture performance.

Keywords: Blood, Growth, Survival, Water quality

Introduction

The production of tilapia decreased by 1.3 % in the Asian market in 2020 [1]. The Covid 19 pandemic has caused a decrease in aquaculture and fish processing activity and a significant drop in market demand. Furthermore, the FAO has targeted a more rapid increase in tilapia production starting in 2021. Indonesia, the world's 2nd-largest tilapia producer [2], can increase aquaculture production from this particular sector.

Currently, aquaculture faces many problems, i.e., limited land and water, aquaculture waste fish diseases [3]. Recirculating aquaculture systems (RAS) offers a better solution to water quality in aquaculture systems to increase fish biomass with limited land and water resources and minimize water pollution [4]. Therefore, it is urgent to develop aeration techniques in RAS to maintain favorable water quality and fish resistance in aquaculture systems [5]. Increasing oxygen concentration in stagnant culture systems eases usually using a conventional blower/aerator. Aeration microbubbles are innovative technology as a tool to increase oxygen concentration, therefore fish biomass increases.

The microbubble aeration technology has been shown to improve the efficiency of oxygen mass transfer between the gas-to-water phases [6] and increase the dissolved oxygen (DO) in saturation level [7-9]. The DO is a critical factor of water quality in RAS and directly affects fish survival and growth [10,11]. The higher DO level maintains the stability of aquatic life. In contrast, in fish culture systems, low DO level directly affects the reduction of the metabolism process, suppresses growth [12], increases stress hormone and alters the function of immunity [10], and a total of plasma protein (TPP) [13], as well as causes the

change in hematological parameter [14,15]. Low water quality control, limited water changes, and aeration stimulate stress conditions that reduce growth and production [16]. An increase in glucose content in fish blood is one of the stress indicators in fish, as its mechanism in overcoming homeostasis due to stress against physiological change [17]. Therefore, the RAS system using microbubble aeration promises a method to overcome the problem of aquaculture problems.

Previous studies found that microbubble aeration increases the growth rate of red sea bream culture [18], red tilapia [19], koi [20], the culture of oyster in Hiroshima, shells in Hokkaido, and pearl in Mie Prefecture, Japan [21]. Although microbubble generator technology has been developed since a few years ago, application in intensive fish culture is still limited. In addition, comprehensive research on the effect of microbubble aeration on oxygen concentration, fish growth performance, and stress resistance of red tilapia in RAS has not been conducted. This research will compare microbubble aeration and conventional blower aeration and without aeration on oxygen concentration. The DO increase is expected to reduce fish stress and subsequently improve its health, and the negative impact of waste control [22]. The hematology parameter is important for assessing physiological status [23] and monitoring fish stress [24,25]. The microbubble aerator is a high-potential technology to improve performance in an intensive aquaculture system without extensive land and water usage.

Material and methods

This research was carried out from August to October 2020 at the Aquaculture Laboratory, Department of Fisheries, Faculty of Agriculture, University of Gadjah Mada (UGM), Yogyakarta (7°46'1.5' S, 110° 22' 54' E) Indonesia.

Fish preparation

The red tilapia strain (Nilasa) was obtained from a local seed producer in Sleman, Yogyakarta. The fish were acclimated in 9 containers with a volume of 1 m^3 each and provided with blower aeration for 15 days. During acclimatization, the fish was fed with a commercial pellet HI-PRO-VITE 781, with contents of crude protein (32 %), fat (3 - 5 %), fiber (4 - 6 %), ash (10 - 13 %), and water (11 - 13 %)

The aerator installation

The nozzle of a microbubble aerator with a type of orifice and porous pipe / multi-fluid was installed with a pump of Yamano WP-106 brand, a maximum capacity discharge of 4 m^3 /h and a maximum head of 4 m. The blower aerator CE used was Resun LP 100, with a capacity of gas flow of 140 L/min. A flow meter was used to regulate the flow rate of air entering the aerator and microbubble at 3 L/min to the tank culture each. A debit of water recirculation system was maintained of 1.8 L/min for each treatment.

Experimental procedures

The experiment was conducted using a completely randomized design with 2 treatments each, i.e. aerator microbubble, aerator blower, and without aeration in RAS each, with a fish stocking density of 50 individuals with the size of 115.27±3.93 g each tank. The fish were reared in a tank with a water volume of 800 L. Fish were cultivated in a container under a semi-open room for 50 days with RAS. During cultivation, these fish were fed using a commercial feed of HI-PRO-VITE 781 with commercial pellets 2 times a day at satiation.

Dissolved Oxygen concentration and temperature were measured daily, while pH, total dissolved solids (TDS), CO_2, alkalinity, biological oxygen demand (BOD), nitrite, nitrate and total ammonia were measured at initial, 25 and 50 days of fish culture. The day after water quality was monitored, individual fish measured the weight, length. At the end of the experiment, the number and total weight biomass were calculated. The water quality measurement procedure followed SNI 06-6989.9-2004, and American Public Health Association (APHA) 2017 method.

Hematology tests including hematocrit (Hct), hemoglobin (Hb), red blood cell (RBC) count, white blood cell (WBC) count, total plasma protein (TPP), and glucose were performed every 17 days (for all 4 tested densities at Day-0 (D-0), 17 (D-17), 34 (D-34) and 50 (D-50) [26]. The stress tests were carried out on day 25^{th}. The stress tests were carried out on day 25. The test exposed the fish to elevated salinity levels of 12 and 24 ppt for 24 h (5 fish/tank with 2 replications). Blood glucose levels, Hb, Hct, and cortisol levels were checked at the 0^{th} and 24^{th} hours.

The fish was drugged with clove oil with a concentration of 0.05 mL/L before blood sampling. The sampled blood was put into an Eppendorf tube to observe RBC count, WBC count, TPP, glucose, Hb, Hct, and cortisol. RBC count and WBC count were calculated using the double-improved Neubauer

hemocytometer. The content of Hb and Hct was measured using an automatic blood cell analyzer Dr. Hb MHD-1 (Korea). The glucose level was measured using the Gluco Dr test strips model AGM-2100 (Korea). Plasma cortisol content was measured using radioimmunoassay enzyme (Cortisol ELISA), and the TPP was measured using a colorimetric method, glucose concentration was measured using the solution of Bradford protein test kit (Sigma).

Calculating data

Fish biomass was calculated based on the fish's total weight at the harvest. The specific growth rate (SGR) was calculated using a formula of (27), feed intake formula (FIperc), and food conversion ratio (FCR) following the formula of [28]. Whereas the protein efficiency ratio (PER) follow the formula [29];

$$SGR\ (\%\ d^{-1}) = \frac{(logn_{Wf} - logn_{Ws})}{T} \times 100\ \% \quad (1)$$

$$FI_{perc}(\%d^{-1}) = \frac{FI}{BWg} \times 100\ \% \quad (2)$$

$$BWg = \exp\{1/2[\ln(Ws)+\ln(Wf)]\} \quad (3)$$

$$FCR = \frac{FItot}{Wf-Ws} \quad (4)$$

$$PER = \frac{weight\ gained\ (g)}{weight\ of\ protein\ consumed\ (g)} \quad (5)$$

Where: Wf = final weight of fish; Ws = weight of fish at Start; T = duration of cultivation, FI_{perc} = body weight percentage, FI = average feed intake per fish (g $fish^{-1}$ d^{-1}), BW_g = geometric body weight (g), FI_{tot} = the total feed intake per fish during the experimental period (g)

Statistical analysis

Oxygen concentration during fish culture in each treatment was analyzed descriptively and presented as a daily trend. Data sets of water quality parameters, fish size, biomass, and hematology were analyzed using a statistical correlation test and analysis of variance (ANOVA) at a confidential level of 95 %.

Results and discussion

Water quality

The daily average DO concentration during the experiment in each treatment was given in **Figure 1**. The DO concentration showed a decrease after stocking. The initial DO at microbubble treatment was 7.64 mg/L decrease to 4.28 mg/L at the 2nd week, and it was relatively stable until the end of the experiment. The treatment of aeration blowers shows a similar pattern. Initially, oxygen concentration 6.83 mg/L was reduced to 1.86 mg/L at the 2nd week and stable until the end of the experiment. Meanwhile, at the control treatment, the oxygen concentration initially was 5.41 mg/L decreased to 0.61 mg/L in the 2nd week, causing 85 % of fish mortality. After the fish death, the value of DO increased to 4.44 mg/L until the final experiment showing no oxygen consumption. The value of temperature, TDS, pH, alkalinity, CO_2, BOD, nitrite, nitrate, and ammonia each treatment was presented at **Table 1**. The temperature fluctuated in a range of 25.0 - 28.5 °C, and with no difference between the 3 treatments ($p > 0.05$). TDS was significantly different ($p < 0.05$) among treatments, i.e. 0.370±0.002 mg/L (microbubble), 0.307±001 mg/L (blower) and 0.23±0.001 mg/L (control). The pH of microbubble 6.33±0.01, blower 6.35±0.01, and control 6.34±0.01 were not significantly different ($p > 0.05$) among treatments until the final experiment.

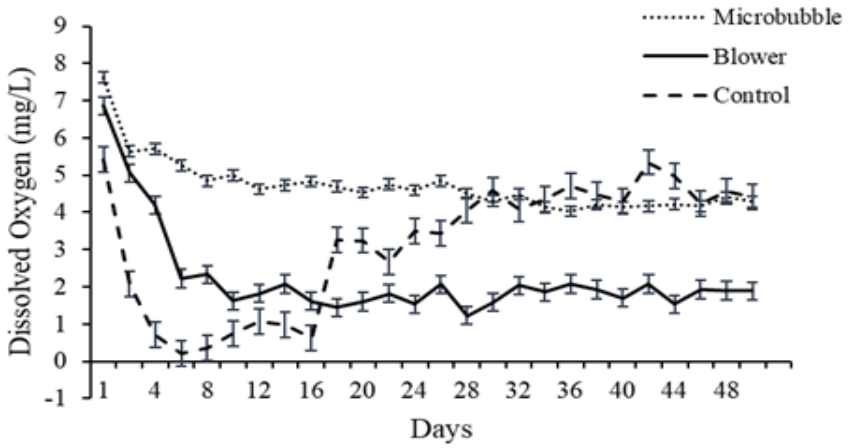

Figure 1 Parameter dissolved oxygen (mg/L) in tanks during the 50-day trial.

The alkalinity and CO_2 level of the microbubble aeration treatment were significantly higher than the blower treatment ($p < 0.05$). BOD levels and nitrite level among the treatments was not significantly different ($p > 0.05$). Nitrate level was increased in all treatments until the end of the experiment, with the highest level in blower treatment. Ammonia levels rose at the end of the culture, and there were significant differences ($p < 0.05$) between blower, microbubble, and control.

Table 1 The value of temperature, TDS, pH, alkalinity, CO_2, BOD, nitrite, nitrate, and ammonia of the microbubble, blower, and control aeration treatment at initial, middle, and the final experiment (D-0^{th}, D-25^{th}, D-50^{th}) (Mean ± SE).

Parameter	D-0	D-25			D-50		
		Microbubble	Blower	Control	Microbubble	Blower	Control
Temperature (°C)	25.41	26.23±0.07	26.07±0.02	26.27±0.16	27.12±0.08	26.55±0.08	26.59±0.1
TDS (mg/L	0.23	0.28±0.002	0.29±0.001	0.29±0.003	$0.37±0.002^a$	$0.31±0.003^b$	$0.30±0.003^b$
pH	7.33	6.4±0.02	6.36±0.08	6.43±0.001	6.51±0.1	6.41±0.2	6.77±0.013
Alkalinity (mg/L)	131	$121.33±0.67^a$	$103.33±3.33^b$	$82.66±3.71^c$	$100.67±5.8^a$	$81.33±1.3^b$	$152.00±3.06^c$
CO_2 (mg/L)	3.47	$6.00±0.29^a$	$10.53±0.29^b$	$7.20±0.52^c$	$14.00±1.57^a$	$26.67±0.67^b$	$7.33±0.88^c$
BOD (mg/L)	0.77	$1.43±0.29^a$	$1.0±0.25^a$	$2.67±0.38^b$	2.40±0.81	2.60±1.12	1.80±0.42
Nitrite (mg/L)	0.001	$0.066±0.003^a$	$0.059±00^a$	$0.083±0.003^b$	0.81±0.02	0.78±0.01	0.81±0.012
Nitrate (mg/L)	0.61	1.64±0.27	3.19±0.72	1.68±0.08	$4.01±0.14^a$	$5.68±0.08^b$	$2.23±0.12^c$
Ammonia (mg/L)	0.002	$0.06±0.002^a$	$0.067±0.005^a$	$0.19±0.026^b$	$0.65±0.06^a$	$1.16±0.06^b$	$0.64±0.02^c$

(Value with a different superscript at the same row show significant difference at 95 % level)

Hematology parameters

Hemoglobin (Hb) levels in fish blood (**Figure 2(A)**) increased until the end of the experiment, measuring at 9.86 g/dL (blower), 8.47 g/dL (microbubble), and 8.57 g/dL (control). Values in all treatments were significantly different ($p < 0.05$). The value of hematocrit (Hct) for fish under all treatments increased until the end of the experiment (microbubble: 28.3 %, blower: 29.58 %, and control: 28.63 %) (**Figure 2(B)**) with all treatments showed statistical difference ($p < 0.05$). At the end of the experiment, the RBC count (**Figure 2(C)**) of fish under aeration blower treatment were $(375.0 \pm 1.2)\times10^3$ cells/mm, control $(360.0±1.1)\times10^3$ cells/mm and microbubble treatment $(353.0±3.1)\times10^3$ cells/mm ($p < 0.05$). The WBC (**Figure 2(D)**) at the of the experiment of at aeration blower were 8350.33±68.39 cells/mm, microbubble

8330 ± 33.33 cell/control ($p < 0.05$), and control 8130 ± 66,67 cell/mm was not significantly different ($p < 0.05$). The TPP value of fish (**Figure 2(E)**) in blower treatment was the highest (58.06±0.86 mg/mL) at the end of the experiment and significantly different ($p < 0.05$) with control (50.06±0.98) and microbubble treatment (48.34±1.32) mg/L. Blood glucose level (**Figure 2(F)**) was significantly different on the 34th day in all treatments. However, the end of the experiment indicates insignificant blood glucose levels for all treatments (microbubble: 84.00±2.00 ng/L; blower: 94.00±6.43 ng/L; control: 77.00±5.50 ng/L).

The correlation between DO with glucose, WBC, RBC, TPP, Hb, and Hct was given in **Figure 3**. The correlation between DO and WBC count was 74.3%. There was a significant correlation between DO and RBC count of fish at all treatments, approximately 72.5 %. The correlation between DO and TPP was significant, i.e. 71 %. DO levels were significantly correlated with hemoglobin by up to 90 %. There was a significant correlation of about 90 % between DO and hematocrit.

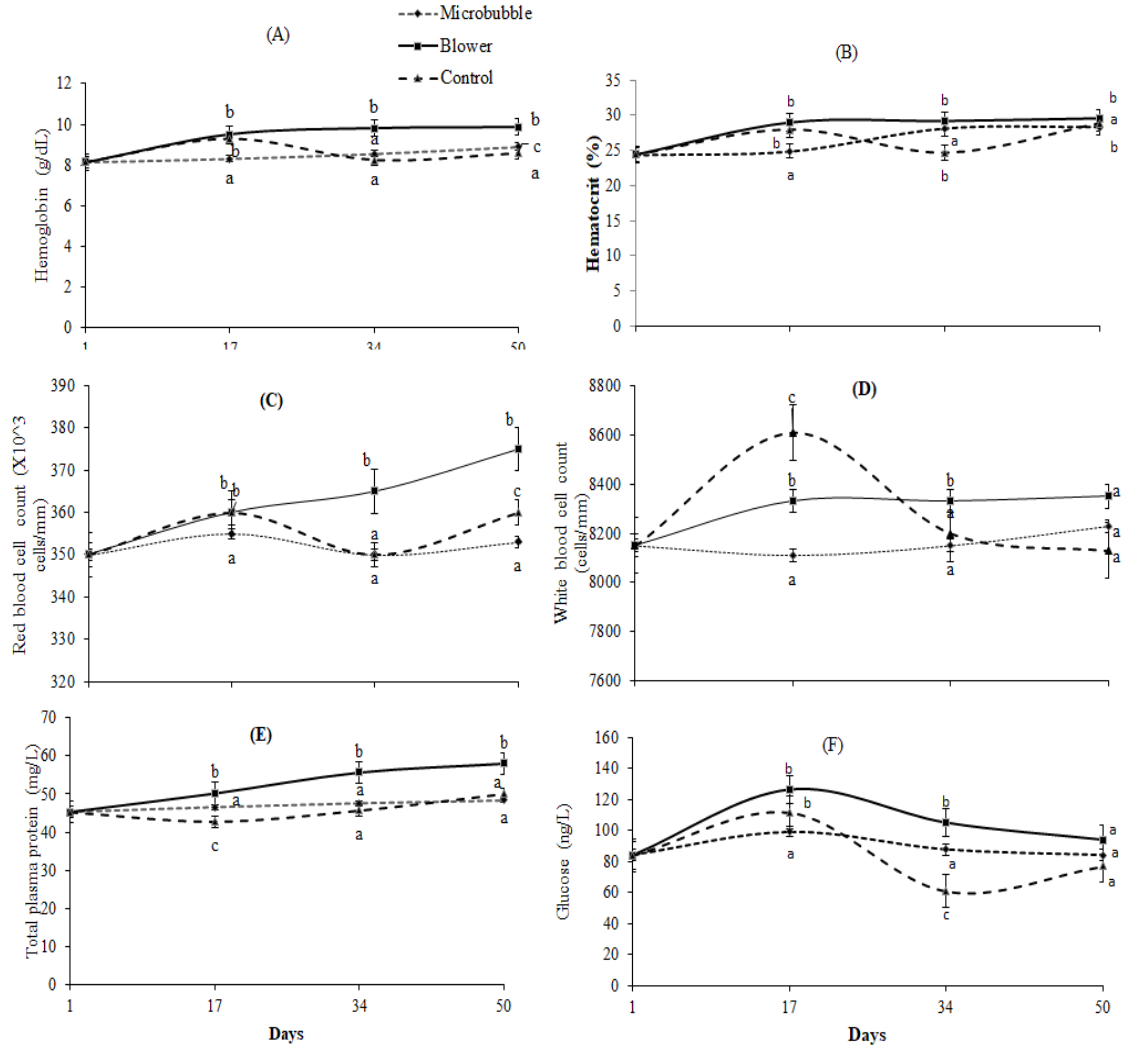

Figure 2 Hematological parameters: (A) Hemoglobin (g/dL); (B) Hematocrit (%); (C) Red blood cell count (x10^3 cells/mm); (D) White blood cell count (cells/mm); (E) Total plasma protein (mg/L); (F) Glucose (ng/L) of fish in microbubble aeration treatment, blower and control were observed every 17 days. (Mean ± SE).

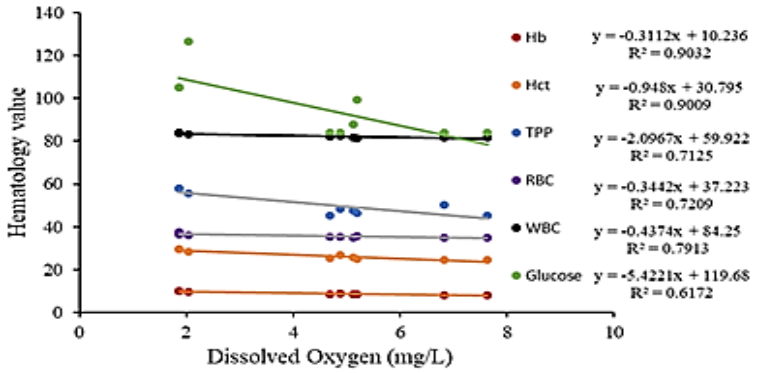

Figure 3 Correlation between DO and glucose (ng/L), WBC count (×10^2 cells/mm), RBC count (×10^4 cells/mm), TPP (mg/L), Hb. (g/dL), Hct. (%).

Fish stress assay

The fish stress test using salinity treatment was presented in **Table 2**. The fish subjected to stress in microbubble under both salinity of 12 and 24 ppt exhibited significantly ($p < 0.05$) lower glucose values than those under blower treatment. On one hand, the value of fish cortisol under microbubble treatment at the salinity of 12 ppt was significantly different ($p < 0.05$) lower than that under blower treatment. However, under salinity of 24 ppt, fish cortisol at 2 treatments was not significantly different ($p > 0.05$). Meanwhile, Hb and Hct did not show significant difference ($p > 0.05$) under the 2 treatments.

Table 2 The value of glucose (ng/L), cortisol (mg/L), hemoglobin (%), and hematocrit (%) of fish stress under salinity test (Mean±SE).

Parameter	1st salinity (0.22 ppt)		2nd salinity (12 ppt)		3rd salinity (24 ppt)	
	Microbubble	Blower	Microbubble	Blower	Microbubble	Blower
Glucose (ng/L)	97.0±2.08[a]	110±2.52[b]	159.33±14.95[a]	203.0±19.77[b]	174.67±9.96[a]	278.67±10.97[b]
Cortisol (mg/L)	36.46±2.45	46.46±9.19	100.88±9.48[a]	132.83±6.32[b]	160.77±15.68	171.32±12.53
Hemoglobin (%)	9.67±0.22	10.40±0.61	8.50±0.35	8.50±0.56	8.0±0.12	8.57±0.23
Hematocrit (%)	29.00±0.66	31.20±1.82	23.50±2.25	25.50±1.67	24.00±0.35	25.7±0.73

(Value with a different superscript at the same row show significant difference at 95 % level)

Fish performance

The fish performance from 3 treatments are presented at **Table 3**. The final weight of the fish under microbubble treatment was significantly higher ($p < 0.05$) than that of blower treatment. The fish in microbubble aeration also had significantly ($p < 0.05$) higher specific growth rate (SGR), feed intake (FI_{per}), protein efficiency ratio (PER) and biomass compared to the other 2 treatments. FCR in microbubble treatment was significantly ($p < 0.05$) lower than blower treatment.

Table 3 Initial weight (g), final weight (g), biomass (g), FIperc (%/day), PER, SGR (%), and FCR fish in the experiment (Mean±SE).

Parameter	Treatment		
	Microbubble	Blower	Control
Initial weight (g)	114.55±1.49	113.33±1.71	113±0.86
Final weight (g/fish)	202.03±0.96[a]	148.83±1.45[b]	156.33±3.19[c]
Biomass (g)	10076.2±55.40[a]	7293.32±137.59[b]	1200.17±74.56[c]
FIper (%/day)	1.49±0.06[a]	0.91±0.03[b]	1.67±0.12[a]
PER	2.41±0.07[a]	1.68±0.1[b]	–16.76±1.23[c]
SGR (%)	1.14±0.02[a]	0.55±0.03[b]	0.63±0.05[b]
FCR	1.30±0.04[a]	1.87±0.11[b]	1.39±0.05[b]

(Value with a different superscript at the same row show significant difference at 95 % level)

The treatment of different aeration devices, namely microbubble and blower, in this research influence DO concentration and impact to the variation of other water quality parameters. DO levels in the present study decreased after 2 weeks and were relatively stable until the end of the experiment, except in control. DO is the most critical parameter, and its concentration is generally considered as a limiting factor for fish survival [10], which means that low DO can affect the mortality rate [22], and it happened in fish of the control treatment. The drastic decrease of oxygen concentration below 1 mg/L in the non-aerated system affects hypoxia in fish [30], so the fish mortality in control treatment reaches 85 %. This observation suggests that aeration is a vital tool since it impacts survival and growth of fish [31]. It shows that with only 15 % initial number, then the DO level of control increased to 4.44 mg/L.

The DO concentration in microbubble aeration at the end of the study was higher than the blower treatment. The microbubble produces smaller bubbles [7] with sizes less than 40 µm [8] cause slow rise velocity, low buoyancy force, high dissolubility, high interfacial area, high inner pressure of the air. Therefore, microbubble aeration affects oxygen steady in the water column evenly [6,9]. While in the blower aerator produced larger bubbles with size of 80 - 100 µm [6] and limited area caused the bubble release from water surface easily.

Though the DO is influenced by temperature [22], in this study, the water temperature was quite constant. The optimum temperature attained for the best growth performance and feed efficiency of red tilapia [32]. Beside temperature, pH value and BOD did not show significant differences amongst the treatments, and it was still under the optimum range for tilapia [33,34]. Other water parameters show significant differences at the end of the experiment, namely CO_2, alkalinity, nitrite, nitrate, and ammonia. It indicates that microbubble aeration revealed better quality of the water culture (**Table 1**). One of the highest stressors on the tilapia culture is increasing ammonia concentration and its derivatives in the fish culture [35], i.e. nitrite and nitrate. In this study, the aeration microbubble could suppress the ammonia, which was oxidized to nitrite and nitrate compared to the aeration blower treatment (**Table 1**).

Appropriate management of water quality in fish culture is essential to maintain the good health of fish [18]. Hematology parameters indicate fish health in response to changing water quality [19]. There was a significant correlation between DO and all parameters of fish hematology (**Figure 3**). Since the initial application of microbubble aeration, there was no significant alteration in the value of hematology and fish glucose (**Figure 2**). This observation suggests that fish are in good health. Meanwhile, fish on aeration blower treatment experienced an increment in hematology and glucose. Fish glucose in aeration blower treatment increased reaching 126.33 ng/L on the 17^{th} day indicating a stress condition on fish. Then, it decreased to 94 ng/L at the final experiment, closing the normal limit of fish glucose at 40 - 90 mg/L [36]. The alteration of the Hb value and the increase in glucose highlighted the stress condition in fish [19,37, 38]. In addition to DO, exposure to ammonia also stimulate a significant decrease in hematological parameters [39], which occur in blower treatment with ammonia concentration exceeding 1 mg/L. The increase of RBC count (**Figure 3(C)**) of fish on blower treatment was a homeostasis effort of fish in reproducing Hb to bind oxygen due to the DO of blower treatment was less than 2 mg/L, indicating hypoxia on fish. Hypoxia is a critical issue in aquaculture, affecting metabolism, and significantly increases plasma cortisol and glucose [6]. Some bony fish have been proven to maximize blood-carrying oxygen capacity in response to hypoxia by increasing the content of Hb and Hct so that they could alternate cell volume and synthesize new erythrocytes from limp [40]. In this experiment, there was a correlation between DO and Hb about 90 %. It meant that the oxygen in the waters was significantly influencing the change of fish Hb. Decrease in DO forces fishes and aquatic organisms to either hard to uptake oxygen from water or reduce their rate of energy expenditure [30,41].

Besides glucose, cortisol might be a stress indicator for fish [7,25]. Fish cortisol with a salinity test of 12 ppt in microbubble treatment was significantly lower whereas that of 24 ppt under both treatments showed insignificant difference. The cortisol value culture in aeration microbubble was lower, indicating more resistance against the pressure of salinity stressor. This finding is supported by [42], suggesting that 16-ppt salinity concentration is suitable for red tilapia culture as the fish are more adaptive to salinity conditions without changes in their growth rate. It was proven in this study that when the stress test used salinity 24 ppt, the test fish experienced an increase in glucose and cortisol and 40 % mortality in both treatments.

The value of fish hematology in aeration microbubble was better. It impacts fish physiology in microbubble treatment, affecting higher feed intake compared to that in blower aeration. The temperature and DO were the main factors influencing appetite, metabolism, and fish growth [43]. A previous study found that ammonia had an impact on feed intake [44]. The critical negative consequences of the high ammonia in red tilapia culture are the substantial decrease in body growth rate, alteration in hematological nature, increased cortisol and glucose in the blood to manage with ammonia toxic effect [35]. This can be

seen in blower treatment, which experienced growth retardation and decreased fitness. In addition to DO and ammonia, CO_2 has also been considered a factor affecting feed intake and fish growth. However, previous research found that CO_2 did not harm fish in an intensive aquaculture system, except that the concentration reached up to 100 mg/L [45]. In this study, the CO_2 value on all treatments was below 100 mg/L, so that it did not negatively impact the feed intake of fish. The low feed intake of fish in blower treatment was caused by a high ammonia content and a low level of DO. The lack of oxygen in fish affected the activity and interrupted the digestion metabolism, decreasing their appetite of the fish [46]. Consequently, it influenced the fish's growth.

The growth of red tilapia in running water, according to Soderberg [47], should have a minimum oxygen tensity of 60 mm Hg (equal to 2.9 mg/L at 32.7 °C). However, Boyd [22] pre required above 5 ppm, or 4 mg/L [3], and DO level should be maintained above 2 mg/L [48]. Meanwhile, DO in blower treatment was less than 2 mg / L, which indicates hypoxia. Hypoxia causes physiological disturbances and negatively affects behavior, physiology, immunology, and growth at the individual and population [30].

In addition to water quality, hematology, and level of feed consumption, the growth of fish was also determined by fish capability in utilizing protein in feed (PER). The fish culture in microbubble aeration had a higher PER value, so the SGR of the fish in the microbubble treatment was also higher. It had a good impact on the FCR value of the fish in aeration microbubble (**Table 4**).

The strong relationship of DO with other parameters produced the performance in individual size and biomass of fish in this study. Previous studies found that microbubble aeration increases the growth rate of red sea bream fish [22], red Nile [23], koi [24], oyster cultivation in Hiroshima, shells in Hokkaido, and pearl in Mie Prefecture, Japan [25]. Similar results also found in this study, the fish treated with microbubble aeration improved to 75 % from the initial biomass, while the biomass of the fish treated with blower increased to 30 %. This result might indicate that a lack of feed in blower aeration fish causes reduced muscle mass [49]. The aeration microbubble produced DO that is capable of maintaining the digestion metabolism of fish so that they could perfectly convert the feed into biomass [47]. According to [31,50], it was parallel that increased aeration and proper culture management could overcome the impact of CO_2 and other water quality parameters as the main restricting factors. The aeration microbubble could maintain a high concentration of oxygen, so the mass of fish was increasing up to 200 g, and its biomass reached 10 kg/tank.

The microbuble aeration can be applied to increase the growth of slow-growing commercially grown fish [19]. The aeration can also reduce land use for ponds and recirculating system, due to its capacity to increase oxygen saturation in narrower and deeper container without sacrificing fish production. It could also benefit industrial-sized fish farm, by lowering energy cost for increasing DO [19].

Conclusions

The application of microbubble aeration significantly improved the overall water quality parameters in the recirculating aquaculture system. The microbubble aeration was able to stabilize DO level on 4.28 mg/L until the end of the experiment and suppressed the CO_2 and ammonia content. The strong relationship of DO with other parameters produced the performance in size and biomass of fish. Fish biomass was higher in microbubble treatment, with a lower FCR value than other treatments. The fish hematology parameters were more stable that indicate a lower stress level. Stable physiological conditions and low stress implied that the cultivated fish is in good health, promoting good performance. Overall, microbubble aeration was proven to improve the quantity and quality of fish produced in RAS.

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