Optimization of *Anguilla bicolor* oxygen consumption in alkalinity culture media

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**ABSTRACT**

One potential aquaculture commodity that can be developed is *Anguilla bicolor*. One effort that can be conducted to maximize the life and growth of *A. bicolor* by manipulating water quality through alkalinity parameters. Alkalinity is a buffer capacity for changing in water pH, and osmotic pressure of water. Alkalinity levels in culture media can be managed by the addition of calcium carbonate (CaCO\(_3\)), calcium oxide (CaO), or calcium hydroxide (CaOH) with a certain dose to the culture media. This study aimed to determine the effect of alkalinity on aquaculture media based on the level of oxygen consumption (OC) that related to growth value. This research was conducted with an experimental design using a completely randomized design (CRD) with four treatments, namely the addition of CaCO\(_3\) to maintenance media with different doses (A 0 mg L\(^{-1}\); B 50 mg L\(^{-1}\); C 100 mg L\(^{-1}\); D 150 mg L\(^{-1}\)). The results showed that the treatment B (50 mg L\(^{-1}\)) showed an OC value (0.15 mg O\(_2\) g\(^{-1}\) h\(^{-1}\)) and an optimal Growth Rate (GR) (3.75 g d\(^{-1}\)), while the Survival Rate (SR) parameter, the treatment had no significant effect. The conclusion was 50 mg L\(^{-1}\) of calcium carbonate (CaCO\(_3\)) having the optimum alkalinity level in *A. bicolor* culture media. At the additional dose of CaCO\(_3\), the growth value (GR), and the level of eel fish oxygen consumption were optimal.

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1. Introduction

One of fishery commodities cultivation that has a potential and can be developed is *Anguilla bicolor*. According to Affandi (2017) and Monticini (2014), *A. bicolor* has economic value and good enough for the high demands in the national and even international market. That demands show in
increasing each year but is not able to be met overall. For example, Japan, one of the biggest importer countries, has the market demands of 130,000 t y\(^{-1}\), but only about 21,000 t are available. Based on it, there is still needed an effort to improve the production of \textit{A. bicolor} such as controlling the water quality through alkalinity manipulating, which is the most ideal for Anguilla growth.

Alkalinity is a part of chemical parameters on the water quality of aquaculture. According to Effendi (2003), alkalinity is defined as water capacity to neutralize acid known acids neutralizing capacity (ANC) or quantity of anion in water that can neutralize the cation of hydrogen. It also can be simply defined as the buffer capacity changes to pH. According to Affandi & Tang (2017), alkalinity influences osmotic pressure on the water, which affects the aquatic organism’s osmoregulation activities. Low or high alkalinity in water relates to an increase in fish osmoregulation that needs activation much energy. The more energy is used for osmoregulation, the less energy that can be used for their growth. A review by Effendi (2003), the concentration of alkalinity comes from several anions in the water such as bicarbonate (HCO\(_3^-\)), carbonate (CO\(_3^{2-}\)), hydroxide (OH\(^-\)), Borat (H\(_2\)BO\(_3^-\)), silicate (HSiO\(_3^-\)), phosphate (HPO\(_4^{2-}\) and H\(_2\)PO\(_4^-\)), sulfide (HS\(^-\)), and ammonia (NH\(_3\)). Of all components, the significant parts are anion bicarbonate (HCO\(_3^-\)), carbonate (CO\(_3^{2-}\)), and hydroxide (OH\(^-\)). Scabra et al. (2016) stated that those components could be obtained by adding the number of calcium carbonate (CaCO\(_3\)), calcium oxide (CaO), or calcium hydroxide (CaOH) with a precise dosage in water media.

This study aimed to evaluate the influence of alkalinity on oxygen consumption of \textit{A. bicolor} related to their growth rate. Based on the previous survey by Abidin (2011), and Hastuti et al. (2014), the addition of calcium carbonate (CaCO\(_3\)), calcium oxide (CaO), or calcium hydroxide (CaOH) could optimize alkalinity levels to the cultivation media. The optimum alkalinity concentration effect on fish metabolisms could be illustrated in the oxygen consumption of fish. Gracia-López et al. (2004), the rate of metabolism organisms waters describe the value of the level of oxygen consumption. More optimal of alkalinity, more optimal of oxygen consumption of fish, which relates to their growth.

2. Material and methods

This study was conducted by the experimental design that applied complete random design with four treatments (addition of diverse concentration of CaCO\(_3\)). Each treatment had three times of repetition, so that the number of all treatment was twelve.

\begin{itemize}
  \item A : 0 mg L\(^{-1}\) of CaCO\(_3\)
  \item B : 50 mg L\(^{-1}\) of CaCO\(_3\)
  \item C : 100 mg L\(^{-1}\) of CaCO\(_3\)
  \item D : 150 mg L\(^{-1}\) of CaCO\(_3\)
\end{itemize}

2.1. Preparation

The container as a culture media used 12 units aquarium with a recirculation water system. The size of an aquarium was 100 × 50 × 40 cm\(^3\), with the maintenance part 90 × 50 × 40 cm\(^3\) and the filter of 10 × 50 × 30 cm\(^3\). Application of filter was intended to serve as the physics filter, chemical filter, and biology filter that was consisted of synthetic cotton, carbon active, zeolite, coral, and bio balls. On a system recirculation, water from an aquarium entered through a filter by gravity
way. Water was discharged directly into the filter system. Water has passed through the screen in the block of water shelter. Then the water was pumped to aquariums through an inlet pipe before it was used to an aquarium, washed, and dried. Aquarium was added with 90 liters of water and added to different doses of NaCl to produce salinity media according to each treatment.

*A. bicolor* (10 ± 1 g) was obtained from the mouth of the Serayu river, Kabupaten Cilacap, Middle Java. The stocking density of Anguilla bicolor was 4 or 360 g of each aquarium and kept for 15 d. The commercial feed with 45 % protein content was converted into a paste form with a percentage of 3 to 5 % of the biomass. The frequency of feed administration was three times a day in the morning (8 am), evening (3 pm) and night (10 pm). Water quality was maintained, with 20 % of the total water volume replaced in maintenance media every day.

2.2. **Oxygen Consumption Rate**

*Gracia-López et al. (2004)* stated that the level of oxygen consumption is a parameter to determine the rate of metabolism of aquatic organisms. The value of oxygen consumption was calculated based on the formula of Liao (1981).

\[
OC = \frac{V (O_1 - O_2)}{W \times t}
\]

OC = Oxygen consumption (mg O₂ g⁻¹ h⁻¹)
O₁ = Dissolved oxygen pre-treatment (mg L⁻¹)
O₂ = Dissolved oxygen post-treatment in some time manners (mg L⁻¹)
V = Volume of water (L)
W = Fish biomass (g)
t = Time of sampling (h)

2.3. **Survival Rate (SR)**

Survival Rate (SR) is a value that describes the survival of the organism at the end of the maintenance period. The Survival Rate (SR) value was calculated based on Goddard (2012):

\[
SR = \frac{N_t}{N_0} \times 100 \%
\]

SR = Survival Rate (%)
Nₜ = Total of fish in t hour
N₀ = Total of fish pre-treatment

2.4. **Growth Rate (GR)**

Growth rate (GR) is a value that describes the absolute growth in organisms. The GR value was calculated based on the Goddard (2012) formula:

\[
SR = \frac{W_t - W_0}{t} \times 100 \%
\]
SR = Survival rate (%)
GR = The absolute growth of fish (g d⁻¹)
Wt = A. bicolor biomass post-treatment (g)
Wo = A. bicolor biomass pre-treatment (g)
t = Day (d)

3. Results and Discussion

3.1. The level of oxygen consumption

The level of oxygen consumption (OC) of A. bicolor is presented in Figure 1. Overall from the beginning to the end of the study, the lowest OC occurred by treatment B, which was 0.15 – 0.70 mg O₂ g⁻¹ h⁻¹, and the highest was treatment D with 0.70 ± 1.55 mg O₂ g⁻¹ h⁻¹. Treatment A showed the lowest OC value only on day 15, but at the other time observations, the lowest OC value was treatment B.

![Figure 1](image)

*Figure 1. A. bicolor OC value by all treatment in all-time manners*

Gracia-López et al. (2004) state that the level of oxygen consumption is a parameter that describes the rate of metabolism of aquatic organisms. The lower OC value indicates the lower metabolic activity of the organism. That metabolic activity relates to the use of metabolic energy budget, the lower metabolic activity, the less energy is used, so that the budgeted energy will be used for growth. According to Li et al. (2007), an unbalanced condition, fish do more mobile activities so that the respiration process will be higher so that its impact on an increase in the level of oxygen consumption. The level of oxygen consumption Rate can also describe the stress response of fish, according to Bonga & Sjoerd (1997), stress conditions will cause a reallocation of metabolic energy from investment activities (growth and reproduction) into activities to improve homeostasis, such as increased respiration, movement more active, hydro – mineral regulation and tissue repair. The level of oxygen consumption is also related to osmoregulation activities. According to Arjona et al. (2009), the fish osmoregulation process requires glucose as an energy source and oxygen as an oxidizer. The lower the osmoregulation activity, the less oxygen is needed as an oxidizer.
Figure 1 showed that treatment B had the lowest OC value, which was $0.15 \text{ mg O}_2\text{ g}^{-1}\text{ h}^{-1}$. It indicated that treatment B showed metabolic activity and the response to stress conditions compared to other treatments. Optimal levels of alkalinity could cause a low-stress response in treatment B. Based on Islama (2014) study, *Barbonymus schwanenfeldii* that was kept in optimum alkalinity media, had a low oxygen consumption activity so that their growth became high. Gracia-López et al. (2004) and Li et al. (2007) explained that the lower metabolic activity and stress response of fish, the lower the oxygen consumption would be. The low oxygen consumption of *A. bicolor* in treatment B revealed a pattern that showed the same as the value of their growth rate (GR).

3.2. Survival rate

Variance analysis test results (ANOVA) showed that the treatment did not have a significant effect on the value of survival rate (SR) ($P > 0.05$). The highest SR value was shown by treatment B with 96.6 %, and the lowest in treatment D was 91.2 %. On the first day 15 of rearing, *A. bicolor* death occurred in all treatments. The number of dead was no significant difference so that it did not affect the total of SR value. *A. bicolor* deaths occurred continuously with small numbers in treatment D (Figure 2).

![Figure 2](image.png)

**Figure 2.** SR value of *A. bicolor* during the study; A (0 mg L$^{-1}$), B (50 mg L$^{-1}$), C (100 mg L$^{-1}$), and D (150 mg L$^{-1}$)

Survival Rate (SR) is a parameter that describes the number of organisms (fish) that live at the end of the maintenance period. Scabra et al. (2016) stated that fish SR is influenced by various factors, one of which is environmental factors. If the environmental conditions are ideal, or as needed, fish can survive. However, if the environmental conditions are not optimal, fish may die (Scabra & Setyowati, 2019).

Figure 2 showed that the survival rate (SR) of *A. bicolor* during the study showed a fairly high number. Based on experience in the field study, *A. bicolor* farming can be said to be successful if the SR value produced reaches 80 % in minimum. It indicated that the addition of calcium carbonate (CaCO$_3$) as a source of alkalinity in *A. bicolor* culture media did not have a negative effect. According to Rahmatullah (2015), the optimal levels of alkalinity play an important role in balancing osmoregulatory performance and keep pH levels in water for fluctuating. A study by Atmawinata (2015), the optimal alkalinity can optimize the survival of *Marcobrachium rosenbergii* de Man pole prawns.
3.3. Growth rate (GR)

Variance analysis test results (ANOVA) showed that all treatments had a significant influence on the value of Growth Rate (GR) \((P > 0.05)\). The highest GR value shown by treatment B was 3.75 g d\(^{-1}\), and the lowest in treatment D was 1.22 g d\(^{-1}\). The growth pattern of *A. bicolor* in Figure 3 showed that on days 15 and 30, the highest GR occurred in treatment A. Day – 45 and to – 60, the highest value of *A. bicolor* occurred in treatment B.

![Growth rate (GR) graph](image)

Figure 3. GR value of *A. bicolor* during the study; A (0 mg L\(^{-1}\)), B (50 mg L\(^{-1}\)), C (100 mg L\(^{-1}\)), and D (150 mg L\(^{-1}\))

Goddard (2012) stated that the growth rate is the difference between the final biomass and the initial biomass to the time of maintenance. The growth rate relates to the efficiency of the final result because the produced product is the total amount of biomass in kilograms. In the present study, the growth of *A. bicolor* optimally occurred in treatment B, namely the addition of calcium carbonate (CaCO\(_3\)) at a dose of 50 mg L\(^{-1}\). According to Muliani & Budiardi (2013), excessive alkalinity due to the addition of CaCO\(_3\) can cause an increase in stress response in fish that decrease the value of the growth rate (GR). A study by Muliani & Budiardi (2013), catfish cultivated in the media with 150 mg L\(^{-1}\) of CaCO\(_3\), showed a higher stress response than maintenance in the media was added 100 mg L\(^{-1}\) CaCO\(_3\).

Conclusion.

Optimum alkalinity levels in *A. bicolor* culture media could be reached by adding 50 mg L\(^{-1}\) of calcium carbonate (CaCO\(_3\)). At the additional dose of CaCO\(_3\), the survival value, growth, and the level of *A. bicolor* oxygen consumption were optimal.

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