Original article

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The addition of calcium oxide with different doses in the recirculation system to improve the abalone *Haliotis squamata* seed production

Penambahan kalsium oksida dengan dosis yang berbeda pada sistem resirkulasi untuk meningkatkan produksi benih abalon *Haliotis squamata*

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ABSTRACT

Information about abalone growth is necessary to overcome the abalone culture sustainability. Water quality parameters are aspects that need to be reviewed, one of which is the calcium level in the water. The aim of this study was to determine the optimal calcium dose in recirculation system for abalone *Haliotis squamata* growth. The study was conducted with an experimental method with four treatments (calcium oxide with doses of 0, 15, 30, 45 mg/L) and three times replications. Abalone with 2.3 g body weight, 2.3 cm shell length, and 1.2 cm shell width was reared for 60 days in recirculation system and fed with *Gracilaria verrucosa*. The results showed that the best abalone treatment medium with the addition of calcium oxide to increase the abalone seed production was 15 mg/L CaO with 100% survival rate, 31.57 ± 2.82% feed efficiency, and 0.56 ± 0.06%/day specific growth rate.

Keywords: Abalone, calcium, organ composition, production, recirculation

ABSTRAK

Informasi tentang pertumbuhan abalon diperlukan untuk mengatasi keberlanjutan budidaya abalon. Parameter kualitas air merupakan aspek yang perlu ditinjau. Kadar kalsium dalam air menjadi salah satu aspek yang dapat ditinjau.Tujuan dari penelitian ini adalah menentukan dosis kalsium optimal melalui sistem resirkulasi untuk pertumbuhan abalon *Haliotis squamata*. Penelitian ini dilakukan dengan metode eksperimental dengan empat perlakuan 3 ulangan penambahan CaO dengan dosis 0, 15, 30, dan 45 mg/L. Secara singkat, abalon dengan bobot badan 2,3 g, panjang cangkang 2,3 cm dan lebar cangkang 1,2 cm dipelihara selama 60 hari dengan sistem resirkulasi dan diberi makan dengan *Gracilaria verrucosa*. Hasil penelitian menunjukkan bahwa media perlakuan abalon dengan penambahan kalsium oksida terbaik untuk meningkatkan produksi abalon adalah CaO 15 mg/L dengan tingkat kelangsungan hidup 100%, efisiensi pakan 31,57 ± 2,82 %, dan laju pertumbuhan spesifik harian 0.56 ± 0.06%/hari.

Kata kunci: Abalon, kalsium, komposisi organ, produksi, resirkulasi

INTRODUCTION

Abalone *Haliotis squamata* is a herbivorous marine gastropod. Abalone is one of great demanded marine commodities because of its high nutrient contents, distinct taste, and health benefits obtained (Sarifin *et al*., 2011; Nurfajrie *et al*., 2014). High market demand of abalone from 134,000 tons in 2015 increased by 150,000 tons in 2016 (FAO, 2017) with the selling price reaches 1,000,000/kg. The abalone supply from culture production has not been maximized, causing an increased natural exploitation, resulting a decreased total population of abalone seeds in the original habitat. The improved abalone seed production through culture can be performed by a flow-through system (Nur, 2020). This system can improve the cultured organism survival rate and growth. Nevertheless, this system has disadvantages, namely, requiring a large amount of water, difficulties in water control treatment, and other biological impacts (Snow *et al*., 2020). Therefore, a more efficient alternative technology is necessary for obtaining a high seed production that can fulfill the market demand.

Recirculation system is a system that reutilizes water by continuously turning the water around through a filter or into a tank (Fauzzia *et al*., 2013; Thesiana & Pamungkas, 2015). This system is commonly applied in a region far from abundant water source, therefore preserving the water and land use. Recirculation system has an effect on the culture environment claimed to more environmentally friendly due to maintaining the water waste from the culture tank and returning it to the tank with the quality closed to its previous. The waste management in recirculation system is performed by applying the mechanical and biological filters (Aji, 2012). Irvandi (2017) conducted a study of abalone stocking density in a filtrated recirculation system at 1.5 g/ind. size with 100, 200, and 300 ind/m^2. This study resulted an insignificant difference among different stocking densities on length growth and shell width. Decreased several production parameters were suspected due to the unoptimized rearing media quality. Moreover, a certain compound addition is thought to help improve the length growth and shell width.

Abalone requires some minerals for the metabolic process, membrane permeability, and shell formation process as the biggest part of the abalone body. Calcium is a mineral ion that plays a major role in bone and teeth formation, body skeleton rigidity maintenance, blood thickening, skeletal, heart, other tissue muscle regulations, muscle relaxation and contraction, osmotic balance maintenance, and shell formation (Zainuddin, 2010; Damayanti *et al*., 2018). Calcium in calcium carbonate is a biomineral that is mostly found in mollusk and crustacean body (Džakula *et al.,* 2013; Purba *et al*., 2015). Based on the results of Rahayu *et al*. (2015), the crushed clam shell produced as a powder had a calcium content of 92%. Abdullah *et al*. (2010) also added that calcium was needed in large amount to form a shell, therefore the calcium addition from the outside should be considered. The shell formation process will be more effective in culture media has a sufficient Ca^{2+} ion as capable of helping the homeostatic process (Hastuti *et al*., 2012). The abalone culture technology development related to the calcium content in culture media for seed production improvement needs to be performed. Therefore, the aim of this study was to determine the best dose of calcium oxide (CaO) addition in the filtrated recirculation system to improve the abalone seed production.

MATERIALS AND METHODS

Tank preparation

This study used aquaria with 80 cm×30 cm×40 cm, which were initially cleaned from wastes. Aquaria were disinfected using a 30 g/L calcium hypochlorite and rinsed with a clean water, then stood for 24 hours. Aquaria were filled with marine water at 30 g/L salinity as much as 15 L. In this step, a recirculation system was made for each treatment. Each aquarium was placed three shelters from polyvinyl chloride (PVC) pipes at 10 cm diameter with both hole sides were enclosed with gauze. A water pump was set for each recirculation system. Each rearing tank was set two aerations for oxygen stabilizing maintenance.

Experimental media preparation

The CaO lime (calcium oxide) was bought in a powder form and kept in a dry place. Before being used for treatment, the CaO lime was initially mashed before measured based on treatment doses, namely the addition of CaO with 15 mg/L, 30 mg/L, and 45 mg/L. The measured CaO limes based on the treatments were initially dissolved in 240 mL water referred to the measuring glass size used, then poured in media based on the treatment doses.

Experimental fish preparation

The experimental fish used were 10 abalone *Haliotis squamata* for each replication. The abalones were obtained from Gondol Main Center for Marine Culture Research and Development, Bali. The initial weight of abalone was 2.3 g with 2.3 cm shell length and 1.2 cm shell width.

Experimental feed

The feed used was seaweed *Gracilaria verrucosa* obtained from Karawang. The seaweed was cleaned using a freshwater. The seaweed was then moved into the prepared and aerated aquaria. The seaweed remains were placed in a container to keep the feed in a wet condition. The feed was given *ad libitum* with twice a day frequency (Kuncoro *et al*., 2013).

Sampling

Rearing was performed for 60 days. Samples were taken as much as 12 abalones once in 15 days from each treatment to measure their shell length, shell width, and weight. The dead abalones were measured each day and accumulated once in 15 days.

Parameters

Survival rate

The number of abalones was calculated at the initial study period. At the final rearing, the number of surviving abalones were calculated and divided by the number of abalones at the initial rearing. The calculation results of initial and final number of abalones were calculated using the formula of Effendie (1997):

$$SR\ (\%) = \frac{N_t}{N_0} \times 100$$

Note :
SR = Survival rate (%)
Nt = The final number of experimental animals
No = The initial number of experimental animals

Feed efficiency

To identify how much abalone consumed the feed and digested it, abalone was measured and noted at the initial and final study. The number of dead abalones during the study were measured and noted. The feed weight given from initial to final study was measured and noted. The calculation of feed efficiency was based on Tacon (1993):

$$EP = \frac{(W_t + D)}{F} - W_0 \times 100$$

Note :
EP = Feed efficiency (%)
Wt = Experimental animal biomass at the final study (g)
D = Dead experimental animal weight during the study (g)
Wo = Experimental animal biomass at the initial study (g)
F = Amount of feed given in wet weight (g)

Specific growth rate

The specific growth rate (SGR) can be defined as the fish weight change in weight, size, or volume along with the time change. The specific growth rate (SGR) was calculated based on Huisman (1987) formula:

$$SGR = \frac{(\ln W_t - \ln W_0)}{t} \times 100$$

Note:
SGR : Specific growth rate (%/day)
Wt : Average abalone weight at the final rearing (g/ind)
Wo : Average abalone weight at the initial rearing (g/ind)
t : Rearing period (day)

Length growth rate

The length growth rate (LPP) is the length change from the initial to the final study. The length growth rate was calculated based on the formula (Allen *et al*., 2006):

$$LPP = \frac{(SL_t - SL_0)}{t} \times 10000$$

Note :
LPP = Shell length growth rate (µm/day)
SLt = Final shell length (cm)
SL_0 = Initial shell length (cm)
t = Period (day)

Data analysis

The survival rate, specific growth rate, and length growth rate were processed using an analysis of variance at 95% degree of confidence level. When there was a significant difference, data were continuously processed with a Tukey test. The data analysis was performed using Microsoft Excel 2010 and SPSS 21 software.

RESULTS AND DISCUSSION

Result

Survival rate

The abalone survival rate in a treatment without CaO addition was 63.33%. The survival rate percentage in 15 mg/L CaO treatment reached 100%. However, this percentage decreased gradually along with the addition of CaO, namely, 80.00% in 30 mg/L treatment and 73.33% in 45 mg/L treatment.

Feed efficiency

Based on the calculation results (Figure 2), the feed efficiency of abalone showed a significant difference (P<0.05) result among treatments. The treatment dose of 15 mg/L calcium oxide (CaO) obtained the highest feed efficiency, namely, 31.57 ± 2.82% and control treatment (0 mg/L) obtained the lowest feed efficiency, namely, 23.18 ± 1.37%.

Specific growth rate (SGR)

The different lime dose addition was significantly different in the specific growth rate of abalone (P<0.05). The highest specific growth rate value was obtained from 15 mg/L treatment, namely, 0.56 ± 0.06%, while the lowest was obtained from 45 mg/L treatment, namely, 0.39 ± 0.01% (Figure 3).

Length growth rate

The different lime dose addition was significantly different on the shell length growth (P<0.05). Based on the shell length growth calculation (Figure 4), the highest was obtained from B treatment with 151.89 ± 11.00 µm, while the lowest was obtained from D treatment with 111.75 ± 2.82 µm.

Discussion

The survival rate is one of the main parameters that indicates the success of an aquaculture

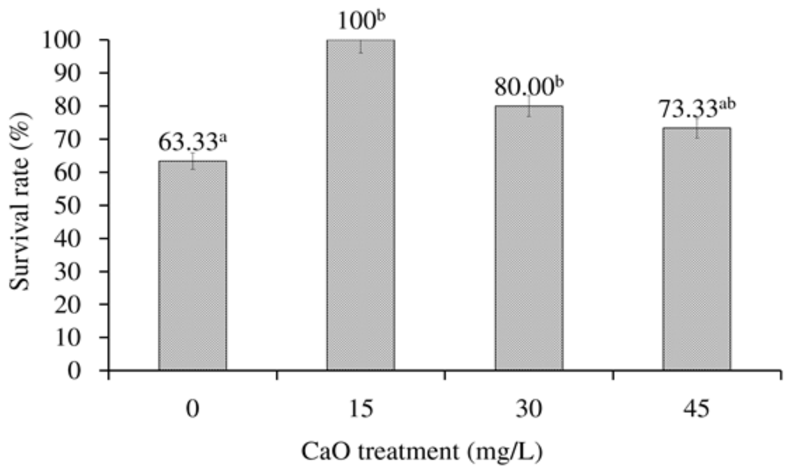

Figure 1. The survival rate of abalone. Different letters on the bars show a significant difference at 5% degree level (Tukey test).

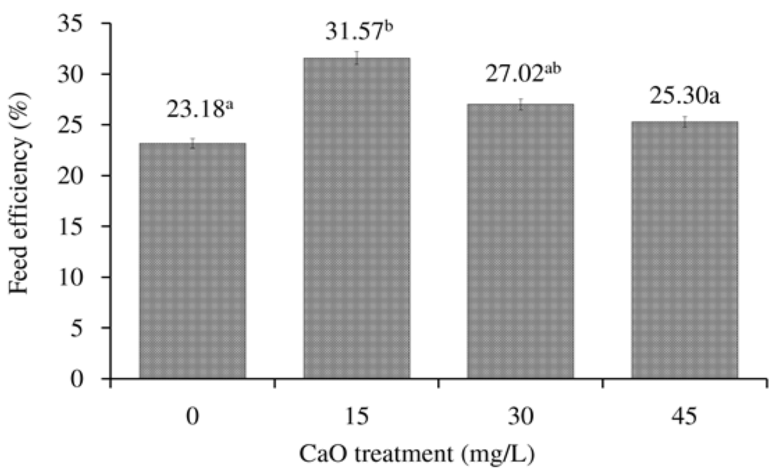

Figure 2. The feed efficiency of abalone. Different letters on the bars show a significant difference at 5% degree level (Tukey test).

commodity maintenance (Rachimi *et al*., 2016). The biotic and abiotic factors directly influence the growth and survival rate. As shown in Figure 1, the survival rate of abalone was significantly different among treatments ($P<0.05$). The 15 mg/L CaO treatment produced the survival rate of 100% at the final rearing. This study supported the statement of Kelabora (2014) that the survival rate increased after the optimal calcium addition in the media and decreased after high calcium addition in the media. This condition indicates that the optimal culture media will support the abalone life.

The low survival rate level in 0 mg/L was thought due to lack of Ca^{2+} ion and water quality reduction (water pH) that caused abalone stress (Karim *et al*., 2017). The stressed abalone is characterized as strongly attached to the substrate when being touched, inactive movement, or moving to the pond side enclosed to the water surface (Lockman *et al*., 2017). Abalone is very sensitive against friction, therefore necessary to be carefully handled (Rejeki *et al*., 2014; Hayati *et al*., 2018).

Feed efficiency (FE) is the ratio of body weight increment produced and the feed consumed. Figure 2 shows that the highest FE value was obtained from 15 mg/L CaO dose treatment with 31.57 ± 2.82%. High FE indicates that the energy utilization obtained from feed was more used for growth and survival than for osmoregulation or daily maintenance. This surely implicates on the increased specific growth rate and shell length. Meanwhile, lower feed efficiency in 0 mg/L treatment with 23.18% was thought due to the abalone seeds were in the hyperosmotic condition, which utilized more energy to sustain the ion balance in the body and its environment. Thereby, the feed utilization became less efficient. Feed containing protein, lipid, and minerals, especially calcium, plays a role in supporting the abalone growth (Susanto *et al*., 2010). Mineral contained in feed is used in the shell biosynthesis process, while mineral in the red blood cells are used for sustaining the osmotic pressure, besides becoming enzyme components (Affandi *et al*., 2011).

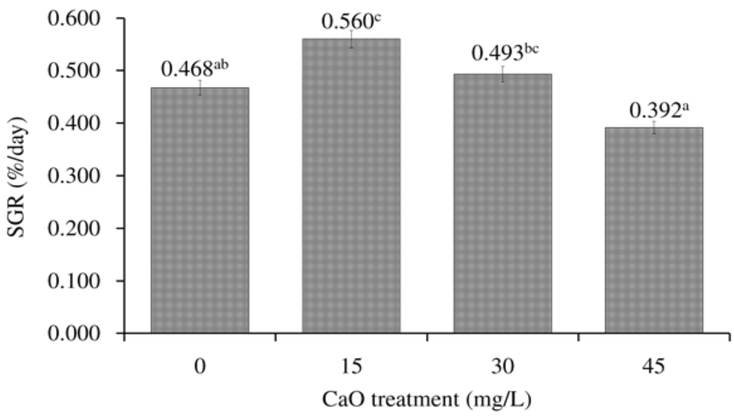

Figure 3. The specific growth rate (SGR) of abalone. Different letters on the bars show a significant difference at 5 % degree level (Tukey test).

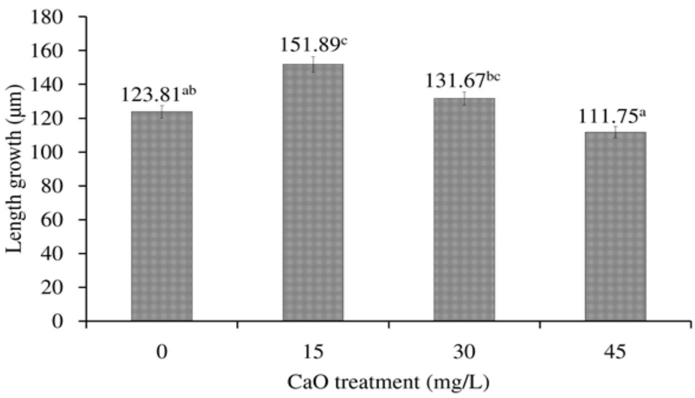

Figure 4. The length growth value of abalone shell. Different letters on the bars show a significant difference at 5 % degree level (Tukey test).

Growth is an important component in the productivity. Growth will perform well whenever the fish body approaches the isoosmotic condition (Diansyah *et al*., 2014). This condition causes the physiological process can perform well, therefore the available energy used for daily maintenance remains low and energy allocation for growth is maximized. The specific growth rate of abalone during rearing showed that the 15 mg/L CaO treatment had the highest growth compared to other treatments with 0.56 ± 0.06%/day. High growth rate indicates that the osmotic pressure in rearing media and abalone body approach the isoosmotic condition. The ion concentration is relatively balanced; therefore abalone does not need to allocate an extra energy for balancing the osmotic pressure inside and outside of the body. Thereby, abalone can utilize the feed well for its growth. The study results of Nurussalam *et al*. (2017) showed that crabs reared in media with calcium and magnesium content more than 30 mg/L for once in 15 days could cause stress. When there is a high osmotic pressure, then the energy allocation for osmoregulation will be greater. This condition can end up to a minimum energy utilized for growth (Pamungkas, 2012; Yuliani *et al*., 2018).

The clamp growth is divided into two parts, namely, flesh growth and shell growth (Runtu *et al*., 2016). The length growth rate of abalone shell in CaO addition treatments (15, 30, and 45 mg/L) during the study ranged between 0.01–0.02 cm/day. The low growth value in 45 mg/L CaO treatment was suspected due to high Ca^{2+} ion availability in the water that was unable to be absorbed well by abalone. This condition may occur due to calcium in the water was bound to other matters. Calcium in marine water can bind to CO_2 for forming $CaCO_3$ (Bogart, 2016). The shell length growth is correlated to the weight growth. This condition is characterized as a symmetric shell and flesh growth of abalone. The value of abalone shell length growth in 15 mg/L CaO treatment (151.89 ± 11.00 µm/day) showed a better growth and significantly different from 45 mg/L CaO treatment (111.75 ± 2.82 µm).

CONCLUSION

The rearing media with the calcium oxide addition of 15 mg/L was the best treatment to significantly improve abalone *Haliotis squamata* production (P<0.05).

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