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Cultivation of red seaweed *Kappaphycus alvarezii* (Doty) at different depths in South Sulawesi, Indonesia

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Abstract. The culture of red seaweed *Kappaphycus alvarezii* in shallow water is at present increased in accordance with the necessity to multiply biomass. This economic commodity is highly demand for its polysaccharide. However, an effort to expand the cultivation area is difficult to several places. This problem may be overcome by employing some hanging rafts at different depths. This research was carried out for 49 days under three different treatments, *i.e.* two morphological types (green and brown), three initial seedlings (50, 100 and 150 g) and five different water depths (1, 2.5, 4, 5.5 and 7 m). The result showed significant differences in growth and biomass among the three treatments (P<0.01). The green strain was influenced by the prolonged period of heavy rain, while the brown one by rainy and hot weather suggesting that this plant may exist along the year. The maximum daily growth rate and biomass was monitored at 100 g initial seed as 5.03% and 10,219 g m^{-2} for green strain and 5.78% and 11,450 g m^{-2} at 150 g initial seed for brown strain. Meanwhile the maximum carrageenan content was observed at higher depth for both green and brown strains.
Key Words: *Kappaphycus alvarezii* farming, deep water, daily growth rate, biomass, carrageenan.

Introduction. The culture of red seaweed *Kappaphycus alvarezii* has been conducted almost around all Indonesian waters recently since it was first introduced (Adnan & Porse 1987). Normally, the culture take place in shallow waters at a depth of about 10 meters, but not all waters can be utilized for culturing. Seaweed biomass obtained through longline technique in shallow waters can produce about 12 tons acre $year^{-1}$ (Dahuri 2012). The product can reach 48 tons acre $year^{-1}$ when vertical technique is deployed in deep water (Hurtado & Agbayani 2002). Deep water area can be used as a productive culture site when meet the requirements of seeds quality, the initial weight and water depth. The utilization of deep waters should be in agreement to the status of coral reefs around the location as an essential requirement for obtaining higher biomass production (Chandrasekaran et al 2008).

High biomass production achieves through deep water culture system (Hurtado-Ponce et al 1996) suggest that the deep water can be used for large-scale cultivation. Sahoo et al (2002) and Sahoo & Ohno (2003) suggest that water comes from deep column is very rich in nutrients and can be used alternatively as laboratory culture media and cultivation media for *Kappaphycus* and other types of seaweed. However, the use of deep water has several constrains related to physical and chemical characteristics, cultivation equipment and proper environment to make it usable. As an important commodity with high market demand, cultivation of *K. alvarezii* requires broader area in order to meet market demand (Nurdjana 2010).

There are few studies related to growth and carrageenan content generating in shallow waters (Ohno et al 1994; Hurtado et al 2008; Naguit et al 2009). However, there were not many studies dealt with growth and the yield of carrageenan in deeper waters. This study was aimed to assess the growth of *K. alvarezii* in deeper waters based on different strains, initial weights and depths of the waters that affect growth, biomass and the yield of carrageenan.

Material and Method. Seedlings of green and brown strains of *K. alvarezii* were obtained directly from the cultivation area in the Takalar Regency of South Sulawesi. These seeds were used after three days acclimatization. Good seeds are the young plants with their tips still sharp and in conical shape (Neish 2005). The seeds were then put at hanging raft (Figure 1) and monitored for 49 days for growth, biomass and carrageenan yield.

Hanging rafts (Wenno 2014) were designed to replace hanging rope techniques (Hurtado et al 2001). Each raft consists of two PVC pipes (L = 200 cm, ø = 5 cm) filled with a mixture of concrete cement which served as the weigh. Both PVC pipes connected by two pieces of wood (H x W x L: 5 x 7 x 400 cm) at the end to form a raft. The total area of raft is 8 m^2 (W x L: 200 x 400 cm). Some nylon strings (ø = 4 mm) with the length of 4 m were used to connect both PVC pipes and functioning as a binding place for seaweed seeds. The distance between the nearest two strings was 20 cm. The raft was hung in the water column with the help of the buoy ropes (ø = 10 mm) and was placed at different depths (1, 2.5, 4, 5.5 and 7 m). The buoy was made of styrofoam (H x W x L: 40 x 50 x 50 cm). During operation, the first raft was connected to the second; the second raft was connected to the third and so on until reach the fifth at a maximum depth of 7 m. The hanging rafts construction was then tied to the anchor ropes (ø = 12 mm), meanwhile the anchor was made from flour sacks filled with sand (Figure 1).

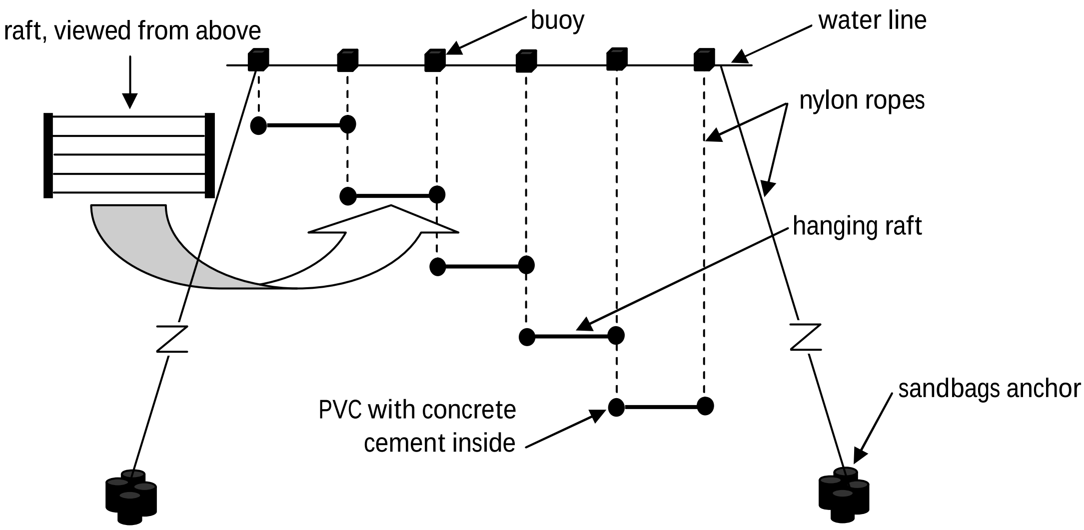

Figure 1. Hanging rafts construction, viewed from frontage (Wenno 2014).

Seaweed seeds were tied to the span ropes according to tie-tie technique (Goes & Reis 2010). Seeds from different initial weights (50, 100 and 150 g) were tied to the twin knot ropes (ø 1 mm). The closest distance between two nodes was 20 cm. Seeds of different strains (green and brown) and different initial weights were placed on each raft at different depths.

Samplings were carried out for seven consecutive weeks. Data were then used to calculate daily growth rate (DGR) which was determined weekly using the following formula of Dawes et al (1993) cited by Hurtado et al (2001) as follows:

$$DGR = \left[L_n\left(W_t / W_o\right)\right]^{\frac{1}{t}} \times 100$$

Where : DGR = daily growth rate (%);

W_t = fresh weight at day t;

W_0 = initial fresh weight;

t = time interval of measurement (7 days).

Seaweed biomass was recorded at the end of experiment and expressed as fresh weight of seaweed per unit culture area (g m^{-2}), and computed with the following formula:

$$Y = (W_t - W_0)/A$$

Where: Y = biomass production;
W_t = fresh weight at day t;
W_0 = initial fresh weight;
A = area of 1 m^2 raft.

The measurement of carrageenan contents (yield) was performed following the formula suggested by Hayashi et al (2007) and Hung et al (2009):

$$Y_C = (W_C / W_{dw}) \times 100$$

Where: Y_C = carrageenan content (%);
W_C = weight of carrageenan extract (g);
W_{dw} = dry weight of analysed thallus (g).

Three factor analysis of variance according to Zar (1999) were used to analyse the experiment data and was performed with SPSS v 21 software. Subsequent analysis with Tukey's HSD test was computed when there were significant differences among treatments with level of significance ($P<0.01$).

Results and Discussions. Daily growth rate of *K. alvarezii* shows the interaction between strain and initial weight, strain and depth, as well as the initial weight and depth which was highly significant ($P<0.01$), but no significant effect between strains, the initial weight and depth ($P>0.05$). Further test showed that the highest daily growth rate of green and brown strains was obtained at the initial weight of 100 g, which tend to be the same (2.84%). It was related to the density of plant (Hurtado et al 2008), leading to rapid growth in the initial weight. Daily growth rate at the initial weight of 100 g was influenced by the interaction between solar radiation, temperature, nutrient and water movement (Santelices 1999), and causing absorption of nutrients faster than other initial weights. Absorption of nutrients was influenced by the density of plants (Azanza-Corrales et al 1996).

Daily growth rate of *K. Alvarezii* of green and brown strains tend to be the same. The highest similarity daily growth rate in green strain was achieved at the depths of 1 and 2.5 m (2.55%), and the lowest one was at 7 m depth (2.23%). The highest similarity daily growth rate in brown strain was also achieved at the depth of 1 and 2.5 m (2.83%), and the lowest one was at a depth of 7 m (2.57%). The highest daily growth rate of green and brown strains associated with the movement of water (Santelices 1999). The movement of the water at that depth was turbulent, consequently reduce the thickness of water that is not mixed in the boundary layer (Neish 2005), and the absorption of nutrients in this depth is faster. Glenn & Doty (1990) suggested that the absorption of nutrients during the fast flowing water between thalli is higher for ammonium at a lower depth than for nitrate at the higher depth. The absorption of ammonium by seaweed is more important than nitrate (Dy & Yap 2001; Raikar & Wafar 2006).

Similarly to daily growth rate, biomass production of *K. alvarezii* showed the interaction between strain and initial weight, strain and depth, as well as the initial weight and depth which was highly significant ($P<0.01$), but not significant by the interaction between strain, the initial weight and depth ($P>0.05$). The highest biomass production of green strain was achieved for the initial weight of 100 g (10,219 g m^{-2}), and the lowest with the initial weight of 50 g (6,709 g m^{-2}). The highest biomass production of brown strain was found for the initial weight of 150 g (11,450 g m^{-2}), and the lowest one with the initial weight of 50 g (7,479 g m^{-2}). The highest biomass production achieved was related to the density of *K. alvarezii* thalli (Hurtado et al 2008) that affect the circulation of nutrients. With the initial weight of 100 g the biomass production of green and brown strains gained was two times higher than the initial weight of 50 g. The biomass production of green strain with the initial weight of 100 g was optimal and may not be increased above this initial weight whilst the biomass production of brown strain can be increased up to the initial weight of 150 g.

The highest biomass production of green strain can be obtained at a depth of 1 m (9,172 g m^{-2}) and of brown strain at a depth of 2.5 m (10,522 g m^{-2}). The similarity of the highest biomass production of both strains obtained at depths of 1 and 2.5 m. The highest biomass production similarity of brown strain was achieved at 1 and 2.5 m depth. This is related to the absorption of nutrients in the lower depths which is faster than that of the higher depth (Neish 2005). Turbulent water movement causes the thickness of the boundary layer between the water and thalli reduced and accelerated the diffusion of nutrients into thalli (Neish 2005). Biomass production of *K. alvarezii* was influenced by the interaction between nutrients and the movement of seawater among thalli (Santelices 1999).

The yield of carrageenan in *K. alvarezii* shows the interaction between strain and initial weight, strain and depth, as well as the initial weight and depth and was highly significant ($P<0.01$), but not significant by the interaction among the strains, the initial weight and depth ($P>0.05$). The higher yield of carrageenan of green and brown strains was obtained with low initial weight. The yield of green strain obtained with the initial weight of 50 g was higher (15.06%) than that of the brown strain (12.33%) of the same initial weight. The yield of green and brown strains was higher at the higher depths when compared to the lower depths. The highest yield of carrageenan in green strain was obtained at a depth of 5.5 m (14.71%) and brown strain at the depth of 7 m (12.18%). The highest yield of carrageenan is the result of the interaction between depth and the initial weight at a depth of 7 m with the initial weight of 50 g (14.70%), followed by the initial weight of 100 g (13.25%) and the initial weight of 150 g (11.95%). The results showed that the yield of carrageenan in both strains was not in line with the daily growth rate. It was found that the yield of carrageenan in this study was contrarily with the finding of Hurtado et al (2008) with a long line cultivation technique, which yields increased in line with the daily growth rate.

In this study, carrageenan yield of *K. alvarezii* green and brown strain increases with the depth and corresponding with the increasing level of carotenoid (0.46 mg/g for green strain and 0.54 mg/g for brown strain) which was obtained at the highest level at a depth of 7 m (16% carrageenan for green strain and 13.39% carrageenan for brown strain). Carotenoid acts as antenna pigments for absorbing light in the process of photosynthesis to produce carbohydrates (Takaichi 2011). However, carrageenan yield in this study is lower than the findings of Hayashi et al (2007) and Distantina et al (2011).

Conclusions. Daily growth rate of *K. Alvarezii* green and brown strains was influenced by the initial weight and tend to be reduced by an increasing of depth. Biomass production was also reduced by the increasing depth with the highest biomass of green strain obtained with lower initial weight, and brown strain with higher initial weight. The yield of carrageenan increased according to depth with the highest yield was observed at green strain compare to brown strain. To obtain higher biomass and carrageenan yield, cultivation should be done at a lower depth for green strain and at a rather lower to higher depth for brown strain.

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