Growth, Biomass Production, and Carrageenan Yield of Kappaphycus alvarezii (Doty) Cultivated in Deep Seawater of Saparua Bay in Central Maluku

Petrus A. Wenno^{*)}, Matheus Ch.A. Latumahina, Sven R. Loupatty, Agustina W. Soumokil, and Endang Jamal

Faculty of Fisheries and Marine Science, Pattimura University, Ambon, Indonesia *)Corresponding author: petrawenno@gmail.com

Received: 05 December 2017; Accepted: 02 May 2018

Abstract

Petrus A. Wenno, Matheus Ch.A. Latumahina, Sven R. Loupatty, Agustina W. Soumokil, and Endang Jamal. 2018. Growth, Biomass Production, and Carrageenan Yield of *Kappaphycus alvarezii* (Doty) Cultivated in Deep Seawater of Saparua Bay in Central Maluku. *Aquacultura Indonesiana, 19* (1): 28-32. Cultivation of *Kappaphycus alvarezii* has been carried out by inverted pyramid method in the deep water of Saparua Bay. This study aims to analyze the daily growth rates (DGR), biomass productions (Y) and carrageenan yields (YC) of the green and brown strain of *K. alvarezii* with different water depth by inverted pyramid method in deep seawaters. *K. alvarezii* with an initial weight of 100 g were planted successively at the water depths of 1, 3, 5, 7 and 9 m during four growing seasons that last for 49 days in every season. The results showed that the highest daily growth rate and biomass production were on the green strain at the depth of 1 m and brown strain at the depth of 3 m, which were 4.18% and 749.29 g/m2 then 4.19% and 754.51 g/m2, respectively. Both of DGR and Y in brown strain was higher than the green strain at the surface layer. While the highest carrageenan yield were on the green and brown strain at the depth of 9 m, which were 16.53% and 14.85%, respectively. Seaweed cultivation in deep waters has a positive impact on carrageenan yields in line with the increasing depths while the growth rate and the biomass production can be achieved higher at the lower depth.

Keywords: Biomass production; Carrageenan yield; Growth rate; Inverted pyramid; Kappaphycus alvarezii

Introduction

Seaweed cultivation in deep waters has not been an option, since shallow waters are still available (Dahuri, 2012). However, the use of shallow water in large and continuing scales can lead to the loss of nutrients (Teichberg *et al.*, 2008; Xu *et al.*, 2001) that will have an impact on long-term harvests.

Utilization of deep waters is important to replace shallow waters utilization which continue to increase following the increase of national target of seaweed production. Hurtado-Ponce *et al.* (1996) showed that the production of seaweed biomass in deep waters by hanging long line method was lower than shallow waters on bottom method. On this condition can cause shallow waters more attractive than deep waters for cultivation. But not all regions especially in some parts of Eastern Indonesia which dominated by deep waters has sufficient shallow waters for seaweed cultures. Therefore, the use of deep waters for seaweed culture to replace shallow water should be considered to meet the increasing demand for seaweed in the future (Navar and Bott, 2014). An inverted pyramid method is developed from a hanging raft method (Wenno et al., 2015) and a hanging long line method (Hurtado-Ponce et al., 1996), since both methods are less practical to apply to mass scale of seaweed cultivation. The hanging raft method requires a number of PVC pipes and cement-sand mixtures for building the construction below the water surface. On the other hand, nylon ropes as the main materials of the hanging long line method are sometimes tangled and lifted to the water surface if there were a lot of waste at the cultivation site. An inverted pyramid method is believed to overcome the weaknesses of both methods as a concerned in this research.

Seaweed production can be enhanced through the expansion of cultivated area (Neish, 2009), but the application of an inverted pyramid method can reduce shallow waters needed. Hurtado *et al.* (2008) suggested that growth rates affect the biomass and the carrageenan content. The objective of this study was to determine the daily growth rate, biomass production and carrageenan yield of *K. alvarezii* cultivated with inverted pyramid method in deep seawaters.

Materials and Methods

Study Site

This research was carried out in deep water of Booi Village, Saparua Bay of Central Maluku district during April to August 2017. The activity took place in deep seawater area which about 50 m away from the coral reef edge or 150 m from the coastline (Figure 1).

Seedling Preparation and Planting

Kappaphycus alvarezii were cultivated in deep seawater area of Booi Village using seeds from the Buru Island. Seeds were selected according to Neish (2005), which was a young tip which still quite sharp and in conical form were separated from their parent colonies. The seeds were used with the initial weight 100 g because in a previous study it gives better growth (Wenno *et al.*, 2015). Each strain was grown at the depth of 1, 3, 5, 7 and 9 meters during four growing seasons that last for 49 days in every season and the distance between two consecutive seasons is 28 days. Setting time for planting and sampling occurred in the same day.

Farming Construction

Inverted pyramid developed from hanging rafts (Wenno *et al.*, 2015), and hanging long line (Hurtado and Agbayani, 2002) (Figure 2). A main frame (W x L = 12 x 12 m) was designed as rectangular shape with the help of large diameter nylon ropes that can accommodate as many as 36 small inverted pyramids (W x L = 2 x 2 m) (Figure 2).



Figure 1. Location of the experiment, Booi village (Arrow) at Saparua bay in Central Maluku (http://www.websitesrcg.com



Figure 2. A scheme of the inverted pyramid

An inverted pyramid hangs on a small rectangular frame $(2 \ x \ 2 \ m)$ and becomes a pyramid base at the top of construction. The pyramid is composed by rectangular frame $(2 \ x \ 2 \ m)$ that functions as a pyramid base at the top and four ropes hanging from each corner of the pyramid base. The lower end of the rope are united and got a concrete weight so the joined rope did not have to lose its appearance when there is water agitation from outside.

Cultivation equipment requires anchors and buoys to maintain their position on the water surface. Buoy is very important, made from styrofoam board (H x W x L = 40 x 40 x 40 cm) and have a buoyancy to lift the entire equipment including the protective net to prevent herbivore and drifting garbage. The main frame (W x L = 12 x 12 m) can accommodate as many as 36 small inverted pyramid which are submerged below the water surface. A buoy load becomes lighter as the colony grows larger and has its own lift. Hanging ropes of inverted pyramid is a main place to tie seeds according to tie-tie technique (Msuya and Salum, 2007).

Daily Growth Rate, Biomass and Carrageenan Measurement

The weight of *Kappaphycus alvarezii* measured each week and lasts for 49 days. Daily growth rates (DGR) were measured and expressed as the percent increase in fresh weight per day (Hung *et al.*, 2009):

$$DGR = [(Wt / Wo)^{1-t} - 1] \times 100$$

where:

DGR = daily growth rate

Wt = fresh weight (g) at day t

Wo = initial fresh weight (g)

T = time interval (7 days)

The production of biomass (Y) measured at the end of research and expressed as fresh weight per unit culture area (g/m^2), and calculated with the formula suggested by Hurtado *et al.* (2001):

$$\mathbf{Y} = (\underline{\mathbf{Wt} - \mathbf{Wo}})$$

where:

Y = production of biomass (g/m^2)

Wt = fresh weight (g) at day t

Wo = initial fresh weight (g)

A = area of 1 m^2 of culture basin.

The carrageenan yield performed as followed (Hung *et al.*, 2009).

$$YC = (WC / Wdw) \times 100$$

where:

YC = carrageenan yield (%)

WC = weight of carrageenan extract (g)

Wdw = dried weight of analyzed thallus (g).

Data Analysis

Data was processed with Minitab 17. Daily growth rate (DGR), biomass production (Y), and carrageenan yield (YC) were analyzed in response to the depths and strains using one way ANOVA. Differences among treatments were analyzed with two-way ANOVA. The level of significant is P< 0.05.

Results and Discussion

The result of data showed that strain and depth of water affects daily growth rate (DGR), biomass production (Y), and carrageenan yield (YC). Both green and brown strains have little effects to DGR and Y, but have different influences on carrageenan yields (YC) (Table 1).

Table 1. The average daily growth rate (DGR), biomass production (Y), and carrageenan yield (YC) of green and brown strains of *Kappaphycus alvarezii*

Strain K. alvarezii	Depth(m)	DGR (%/day)	Y (g/m ²)	YC (%)
Green	1	$4.18\pm0.03^{a(**)}$	749.29±10.55 ^{b (**)}	$13.40\pm0.49^{i(ns)}$
	3	4.16±0.06 ^{ab (**)}	742.98±19.27 ^{c (**)}	$14.08\pm0.34^{f(*)}$
	5	3.96±0.05 ^{cd (*)}	671.84±14.85 ^{g (*)}	14.74 ± 0.40^{d} (*)
	7	$3.81 \pm 0.04^{f(*)}$	629.19 ± 12.52^{i} (*)	15.33 ± 0.42^{b} (*)
	9	3.69±0.05 ^{g (ns)}	593.46±13.69 ^{j (ns)}	16.53 ± 0.42^{a}
Brown	1	4.11 ± 0.08^{b} (**)	724.44±28.18 ^{e (**)}	$12.61\pm0.42^{j(ns)}$
	3	4.19 ± 0.09^{a} (**)	754.51±32.19 ^{a (**)}	$13.57 \pm 0.26^{g (ns)}$
	5	4.11 ± 0.08^{b} (**)	727.16±29.82 ^{de (**)}	$13.52 \pm 0.38^{h (ns)}$
	7	4.00 ± 0.08^{c} (*)	$688.03 \pm 27.82^{f(*)}$	14.45 ± 0.36^{e} (*)
	9	3.86±0.07 ^{e (*)}	$642.47 \pm 23.72^{h(*)}$	14.85±0.3 ^{c (*)}

Note :

The same superscript letter in the same column indicates no significant difference (P>0.05).

(**) = higly significant; (*) = significant; (n^{s}) = not significant

The average DGR of green *K. alvarezii* was recorded slightly lower than the brown strain. The highest DGR of the green occurred at 1 m (4.18%) and the brown at 3 m (4.19%). However, the green and the brown showed the increase of weight according to the decrease of water depth. It means that the cultivation of plants can be achieved higher at the lower depth. Following Duncan's test, both strains can be grouped to the depths of 1, 3 and 5 m; depths of 5 and 7 m; and depths of 7 and 9 m. These groups can be used to determine condition of waters that may support growth of seaweed in deep waters. The ANOVA was conducted to determine the effect of water depths to DGR, Y and YC.

Daily Growth Rate

The ANOVA result of DGR at green and brown strains of *K. alvarezii* tends to decrease according to increasing depths. The effectiveness of photosynthesis at the surface layer of deep waters is supported by the role of chlorophyll-a as main pigment which dominates green plants (Ishida and Green, 2002). It was also occurred at the brown which showed an increase in DGR inversely proportional to the water depth.

The DGR of green and brown strain showed a significant interaction between strain and depth (P < 0.05). The DGR of both strains tends to be higher at the surface. Highest DGR of the green occurred at a depth of 1 m (4.18%) and the brown at a depth of 3 m (4.19%). These are related to the density of colony (Hurtado et al., 2008) that leads to rapid growth. Highest DGR is affected by the interaction among sunlight, temperature and water movement (Santelices, 1999), which is always took place on the water surface. It is also suggested that highest DGR at lower depth is associated with water current (Harrison and Hurd, 2001; Santelices, 1999) and nutrient uptake (Barr et al., 2008). According to Neish (2005), water movement at the lower depth is turbulent therefore it reduces nonmixed water thickness at the border layer (Neish, 2005). This can cause nutrients in the lower depth to be absorbed faster than those at the higher depth. Glenn and Doty (1990) reported that nutrients uptake when they rapidly flow among the colony, and absorb as much ammonium at a lower depth and nitrate at a higher depth (Bracken and Stachowicz, 2006; Taylor et al., 1998). The highest DGR of the green at the lowest depth as indicated in this study is influenced by solar radiation that causes water movements (Neish, 2005). The highest DGR of the brown strain at the depth of 3 m (4.19%) was possibly due to the faster absorption of nutrients by its thicker thalli than the thinner thalli of the green.

Biomass Production

A biomass production (Y) of *K. alvarezii* is affected by water depths. The highest Y of the green occurred at a depth of 1 m (749.29 g/m²), and the brown (754.51 g/m²) at a depth of 3 m. The Y relatively has the same pattern of change in comparison to DGR based on the data used.

The highest Y is related to colony densities (Hurtado *et al.*, 2008) that affect circulation of nutrients as it was to DGR. High Y is caused by the utilization of bigger water column of deep waters. The majority of seaweed cultivation that already spread throughout the Indonesia waters until now is rely on shallow water, meanwhile the water column below the surface layer of deep waters is relatively untapped. This may be a great loss for Indonesia seaweed farmers if the deep waters are not put to good use in the future. This is in accordance with Hurtado and Agbayani (2002) who reported that the Y obtained from seaweed by utilizing water column in deep water is higher than longline method applied on surface layer.

The Yield of Carrageenan

Unlike green strain, brown strain of *K. alvarezii* has photosynthetic pigments which dominated by phycobiliprotein and chlorophyll-b that always increased according to depth (Ramus *et al.*, 1976). It can maintain photosynthesis rate in deep waters by increasing phycobiliprotein and chlorophyll-b ratio against chlorophyll-a. In this research the YC of both strains increased with the increasing depth. This finding was different to Hurtado *et al.* (2008) that the increase of YC is in line with DGR when cultivating in shallow water. It is also against the finding of Hayashi *et al.* (2007) that DGR is closely related to the amount of carrageenan produced.

Conclusion

Kappaphycus alvarezii of green strain has different characteristics to brown strain. Both of DGR and Y in brown strain was higher than the green strain at the same surface layer. Green strain has an ability to produce more carrageenan than the brown strain. It is important to analyze factors affecting growth rate, biomass production and carrageenan yield in deep waters, as well as the possibility to use off shore territories as largescale cultivation area in the future.

Acknowledgement

This study was supported by grant of Ministry of Research, Technology and Higher Education of Republic of Indonesia by contract number 090/SP2H/LT/DPRM/IV/2017.

References

- Barr, N.G., A. Kloeppel, T.A.V. Rees, C. Scherer, R.B. Taylor, and A. Wenzel. 2008. Wave surge increases rates of growth and nutrient uptake in the green seaweed *Ulva pertusa* maintained at low bulk flow velocities. *Aquat. Biol.*, 3:179-186.
- Bracken, M.E.S. and J.J. Stachowicz. 2006. Seaweed diversity enhances nitrogen uptake via complementary use of nitrate and ammonium. *Ecology*, 87(9): 2397-2403.
- **Dahuri, R**. 2012 Cetak Biru Pembangunan Kelautan dan Perikanan. Menuju Indonesia yang Maju, Adil-Makmur, dan Berdaulat. Roda Bahari, Bogor, 134 pp.
- Glenn, E.P. and M.S. Doty. Growth of the seaweeds Kappaphycus alvarezii, K. striatum and Eucheuma denticulatum as affected by environment in Hawaii. Aquaculture, 84 (3-4): 245-255
- Harrison, P.J. and C.L. Hurd. 2001. Nutrient physiology of seaweeds: Application of concepts to aquaculture. *Cah. Biol. Mar.*, 42: 71-82.
- Hayashi, L., E.J.D. Paula, and F. Chow. 2007. Growth rate and carrageenan analyses in four strains of *Kappaphycus alvarezii* (Rhodophyta, Gigartinales) farmed in the subtropical water of Sao Paulo State, Brazil. J. Appl Phycol., 19: 393-399.
- Hung L. D., K. Hori, H.Q. Nang, T. Kha, and L.T. Hoa. 2009. Seasonal changes in growth rate, carrageenan yield and lectin content in the red alga *Kappaphycus alvarezii* cultivated in Camranh Bay, Vietnam. *Journal of Applied Phycology*, 21: 265-272.
- Hurtado A.Q., A.T. Critchley, A. Trespoey, and G. Bleicher-Lhonneur, 2008 Growth and carrageenan quality of *Kappaphycus striatum* var. Sacol grown at different stocking densities, duration of culture and depth. *Journal of Applied Phycology*, 20: 551-555.
- Hurtado A.Q. and R.F. Agbayani. 2002 Deep-sea farming of *Kappaphycus alvarezii* using the multiple raft, long-line method. *Botanica Marina*, 45: 438-444.
- Hurtado, A.Q., R.F. Agbayani, R. Sanares, and M.T.R. Castro-Mallare, 2001. The seasonality and economic feasibility of cultivating *Kappaphycus alvarezii* in Panagatan Cays, Caluya, Antique, Philippines. *Aquaculture*, 199: 295-310.

- Hurtado-Ponce A.Q., R.F. Agbayani, and E.A.J. Chavoso. 1996. Economics of cultivating *Kappaphycus alvarezii* using fixed-bottom line and hanging-long line methods in Panagatan Cays, Caluya, Antique, Philippines. *Journal of Applied Phycology*, 105: 105-109.
- Ishida, K. and B.R. Green. 2002. Second- and third hand chloroplasts in dinoflagellates: phylogeny of oxygen-evolving enhancer 1 (PsbO) protein reveals replacement of a nuclear-encoded plastid gene by that of a haptophye tertiary endosymbiosis. *Proc. Natl. Acad. Sci., USA* 99: 9294-9299.
- Msuya, F.E. and D. Salum. 2007. Effect of cultivation duration, seasonality, nutrients, air temperature and rainfall on carrageenan properties and substrata studies of the seaweeds *Kappaphycus alvarezii* and *Eucheuma denticulatum* in Zanzibar, Tanzania. WIOMSA/MARG I no 2007-06. 36pp.
- Nayar, S. and K. Bott. 2014. Current status of global cultivated seaweed production and markets. *World Aquaculture*, 45: 32-37.
- Neish, I.C. 2009. Tropical Red Seaweeds as a Foundation for Integrated Multi Tropic Aquaculture (IMTA) Four Propositions and an action plan for this major opportunity in the Coral Triangle. SEAPlant.net Monograph no. HB2E 1209 V3 IMTA. December, 2009.
- Neish I.C. 2005 The *Eucheuma seaplant* handbook. Volume I, Agronomics, Biology and Crop System. Sea Plant Net Technical Monograph No. 0505-10A, Makassar.
- Ramus, J., S.I. Beale, D. Mauzerall, and K.L. Howard. 1976. Changes in photosynthetic pigment concentration in seaweeds as a function of water depth. *Marine Biology*, 37(3): 223-229.
- Santelices, B. 1999. A conceptual framework for marine agronomy. *Hydrobiologia*, 398/399: 15-23.
- Taylor, R.B., J.T.A. Peek, and T.A.V. Rees. 1998. Scaling of ammonium uptake by seaweeds to surface area : volume ratio : geographical variation and the role of uptake by passive diffusion. *Mar. Ecol. Prog. Ser.*, 169: 143-148.
- Teichberg, M., S.E. Fox, C. Aguila, Y.S. Olsen, and I. Valiela. 2008. Macroalgal responses to experimental nutrient enrichment in shallow coastal waters: growth, internal nutrient pools, and isotopic signatures. *Mar. Ecol. Prog. Ser.*, 368: 117-126.
- Xu, Z., X. Lin, J. Lin, L. Xie, and C. Huang. 2001. The effects of nutrient availability on the uptake of nitrogen and phosphorus by *Gracilia tenuistipitata* var. *liui* Zhang et Xia. *Acta Ecol. Sin.*, 22: 366-374.
- Wenno, P.A., R. Syamsuddin, E.N. Zainuddin, and R. Ambo-Rappe. 2015. Cultivation of red seaweed Kappaphycus alvarezii (Doty) at different depths in South Sulawesi, Indonesia. AACL Bioflux, 8(3):468-47