Influence of initial weight of seeds in variation of growth and carrageenan content of *Eucheuma spinosum*

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Abstract. Seaweed growth can be influenced by internal and external factors. One of which possibly could affect the growth of seaweed in cultivation area is weight of seed during cultivation process. The aim of this research was to understand the seed weight impact on growth and carrageenan content of *Eucheuma spinosum*. The study was conducted during 3 months (January – March 2014). The research used Complete Random Design with 3 treatments and 3 replications. First treatment was seed weight of 50 g, second treatment seed weight of 100 g, and the third treatment seed weight of 150 g. The results showed that the highest specific growth rate (SGR) was found in treatment of 50 g of seed weight with 4.14%/day, followed by 100 g of seed weight with 3.24%/day, and at the lowest with 150 g seed weight treatment with 2.98%/days. The highest *E. spinosum* carrageenan content was found within the initial seed weight of 50 g with 50.03%, followed by initial seed weight of 150 g with 49.51% and at the lowest at 100 g seed weight with 46.96%. The variance analysis result showed that distinctive initial seed weight treatment affects (sig<0.05) the specific growth rate and carrageenan level of seaweed *E. spinosum*.
Key Words: seaweed, specific growth rate, seed weight, growth, thallus.

Introduction. *Eucheuma spinosum* is one of seaweed type which has important economic value as a main resource of carrageenan and becomes primary commodities in Indonesia. *E. spinosum* is the most popular second type of cultivated seaweed after *Kappaphycus alvarezii. E. spinosum* is a macroalgae family that can be found naturally around Indonesian coastal water area specifically in Eastern Indonesia. The contained carrageenan type is iota which makes it extremely valuable economically. The growth of *E. spinosum* can be diverse depends on the internal and external factors. The external factor is water quality and the internal factor is the thallus health condition. In some *E. spinosum* cultivation areas around Indonesia, the influence of the initial seed weight is still conducted. Besides, the cultivation method could also affect the seaweed growth. The method that is being used by farmers is longline and floating cage. One of the prevention from pest attack is through floating cage method application Kasim & Mustafa (2017). Floating cage method is one of the cultivation method which utilized a special designed tool and protected in order to give total protection of the seaweed from pests (Kasim et al 2016; Kasim & Mustafa 2017). The growth and health of the thallus of *K. alvarezii* is strongly influenced by the quality of seedlings and cultivation methods. Thus, good growth will greatly affect the carrageenan content. Beside that new method innovation development to achieve seaweed production target in Indonesia, the usage of floating cage is necessary to be considered (Kasim et al 2017b). Apart from the cultivation methods, initial weight from every seaweed thallus in the cultivation activity is also important.

The determination of initial seed weight needs to be evaluated in order to create an optimal seaweed growth. Recently, there is no specific study which focused on research regarding the optimal seed weight that can produce the best seaweed quality. Therefore, this research focused on the growth of *E. spinosum* seaweed with distinctive initial seed weight using floating cages. The aim of this research is to understand the impact of initial seed weight towards the growth and carrageenan content of seaweed *E. spinosum* cultivated in floating cages.

Material and Method. This research was conducted from January to March 2014, in Tanjung Tiram, Kendari city, Southeast Sulawesi Province, Indonesia. Carrageenan content was analyzed in Laboratory of the Faculty of Fishery and Marine Science Halu Oleo University, Indonesia. In order to eliminate herbivore attack, we performed this research by using floating cages (Kasim et al 2017a). The seaweed plantation inside floating cages uses different seed weight: 50 g, 100 g, and 150 g/treatment. After weight determinationt, seaweed seeds were tied in each group and spread inside the floating cages with colored marks. Inside the floating cage, there were 9 plots. In every plot different treatment and repetition was applied. The research used Complete Random Design with three treatments (first treatment = 50 g seeds weight; second treatment = 100 g seeds weight; and thirds treatment is 150 g seed weight) and three times repetition. The observed parameter was the specific growth rates (SGR). SGR was measured in every 9 days of interval, for 45 days, counted 5 times of measurement until the end of the research. This SGR can be calculated with a formula of Luhan & Sollesta (2010):

$$SGR = (\ln W_t - \ln W_0)/t \ X\ 100$$

Where:

SGR = specific growth rate (%/day)
W_t = seaweed weight in the end (g)
W_0 = initial seaweed weight (g)
t = maintenance process (day)
ln = natural logarithm

For the determination of carrageenan content the Muñoz et al (2004), formula was applied:

$$\text{Carrageenan content} = \frac{Wc}{Wm} \times 100\ \%$$

Where:

Wc = weight of extracted carrageenan (g)
Wm = weight of dried seaweed (g)

To identify every treatment towards observed variable, the variance analysis (ANOVA) was performed. If the analysis showed a real effect, further the Duncan double distance test will was applied. All of the data's were analyzed through statistic software SPSS version 24.

Results and Discussion. During our research, it was shown that the seed grown from an initial eight of 50 g to 254 g, from the initial weight of 100 to 344 g and from the initial weight of 150 g to 440 g (Figure 1).

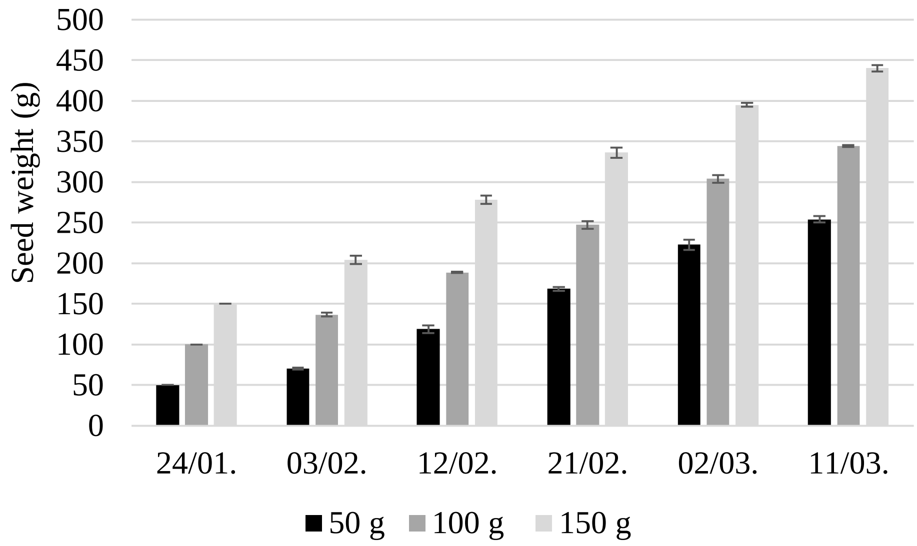

Figure 1. Initial growth of fresh weight of *Eucheuma spinosum* seed during research study.

Specific growth rate (SGR). According to 45 days of measurement shows that the highest SGR result occurred on 50 g seed weight which enriched 3.58–4.95% with an average SGR value of 4.17%, followed by 100 g seed weight with 2.74-3.66% with an average SGR of 3.24%, and the lowest SGR occurred on 150 g seed weight with a range of 2.37–3.73% with an average SGR value of 2.98% (Figure 2).

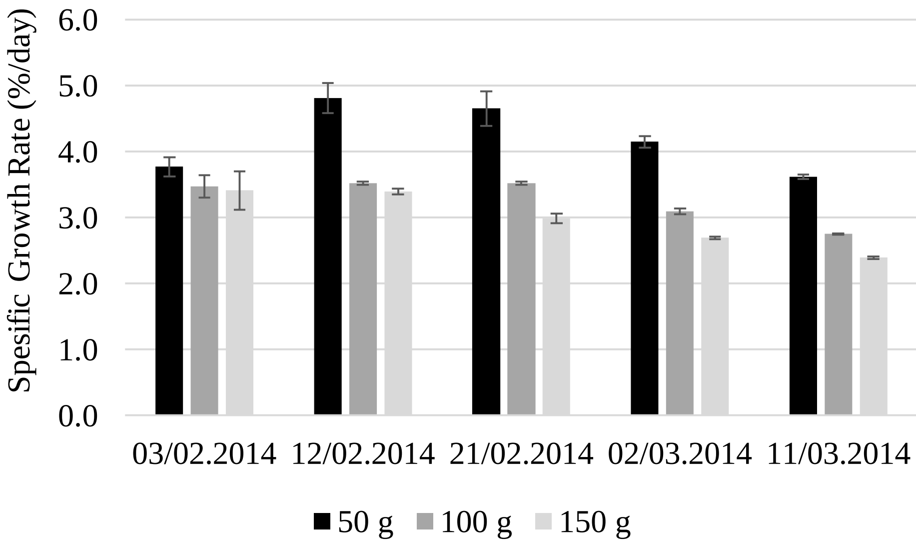

Figure 2. Specific growth rate (SGR) of *Eucheuma spinosum* during research study.

Table 1

Result of analysis variance in every treatment of SGR of *Eucheuma spinosum*

Specification	*Sum of squares*	*df*	*Mean square*	*F*	*Sig.*
Between groups	2.339	3	1.169	279.186	.000
Within groups	.025	9	.004	-	-
Total	2.364	12	-	-	-

* The mean difference is significant at the 0.05 level.

Variance analysis result (ANOVA) shows that the treatment of different initial seed weight gives real impact to the specific growth progress on seaweed *E. spinosum* (sig<0.05) and every further Duncan double distance test shows that seaweed SGR is significantly different between each treatment.

The carrageenan content of *Eucheuma spinosum* according initial seed weight is shown in Figure 3.

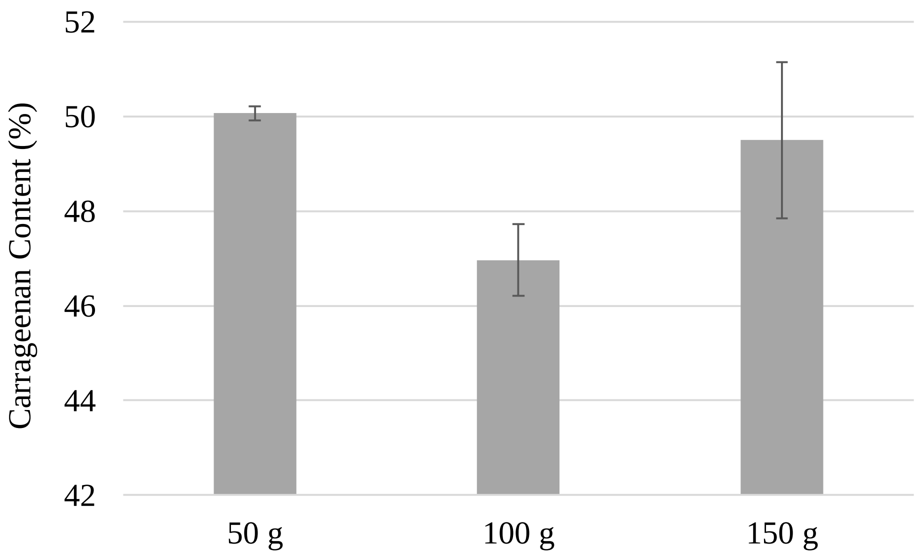

Figure 3. Carrageenan content of *Eucheuma spinosum* according initial seed weight (Asjan 2014).

The highest carrageenan concentration occurred on treatment of 50 g initial seed weight (50.03%), followed by treatment of 150 g initial seed weight (49.51%), and the lowest carrageenan concentration occurred on treatment 100 g initial seed weight which is (46.96%).

Distinctive initial seed weight shows a real impact towards seaweed *E. spinosum* SGR. It is presumed that the determination of initial seed weight really affects the growth progress and nutrients and sun light absorbance of the seaweed. Yusuf (2004) reported that the increase of growth is caused by thallus weight which can develop well and therefore nutrients absorbance process is enhanced by the thallus in Buton coast area. Our research results showed that the highest SGR of seaweed *E. spinosum* is attained from the 50 g initial seed weight treatment, and it is presumed that the cultivated seaweed seed is very young therefore thallus regeneration still continue rapidly and nutrient absorbance occurs optimally so it facilitate new branch development. Indriani & Sumiarsih (2003) reported that nutrient fulfillment affects seaweed growth. Increased growth of *K. alvarezii* is also possible due to the distances between the thallus planted in a planting area and its absorption of nutrient availability in the waters column.

The disadvantage in SGR of seaweed *E. spinosum* within 100 and 150 g initial seed weight is caused by the increasing population of thallus due to maintenance aging and presumed because the space between seeds cannot support thallus growth, so that thallus numbers is denser causing a competition for nutrients, space and sun light absorbance for photosynthesis. The decreasing SGR is caused by rapid cell divide boredom. Seaweed that undergone an adaptation process then face a cell growth swift phase will grow slowly due to decreasing cell development (Yusnaini et al 2000).

The highest seaweed *E. spinosum* SGR average occurred on 50 g initial seed weight treatment with 4.17%, followed by the 100 g initial seed weight treatment with 3.24%, and the lowest within the 150 g initial seed weight treatment with 2.98%. Our result is different compared to Suhardi (2013) which stated that the highest average SGR of *E. spinosum* seaweed cultivated in floating cages at Lakeba coast, Indonesia attained of 4.26% was obtained from 100 g initial seed weight, compared with initial seed weight of 40, 50, and 60 g. Erpin (2012) reported that initial seed weight of 50, 60, and 70 g with the highest mark using long line method at Staring bay, Indonesia was on 60 g seed with 2.52% and the lowest on 50 g with 1.99%. This shows that the initial weight of seeds of each seedlings planted in the cultivation area will affect the growth of seeds in accordance of the environmental qualities. Size of seaweed seed really affects the growth of the seaweed. Besides, daily water quality parameters fluctuation which significantly caused the seaweed thallus becomes yellowish pale which cannot develop adequately so that it reduces the growth progress (Supit 2005).

SGR result on seaweed *E. spinosum* increases for all treatments in the beginning of the plantation at week 1-2. It caused by the age of 9 days when *E. spinosum* goes on logarithmic growth phase (rapid growth phase) and presumed to have no competition to achieve nutrients and having young cells that can regenerate to grow more rapidly. In week 3-4 occurs a decrease in specific growth progress that is presumed to be caused by a maximum range of growth so that eventually the growth will be reduced due to a maintenance time (sigmoid). Basically the growth curve of seaweed will increase along with the availability of environmental support (Mubarak et al 2001).

High and low fluctuation of seaweed growth is presumed to have an impact with the harvest season. According to Paula & Pereira (2003), there is seasonal impact towards the growth of seaweed which is the highest on summer (January-May) and the lowest in winter (June-December). In the present research the average SGR of *E. spinosum* seaweed ranges between 2.98-4.17% in the period of January-March 2014. Abdan (2013), reported that the highest average SGR of seaweed *E. spinosum* is in the range of 3.31-4.26%/day which started in June-July. This is in accordance with Hayashi et al (2007), who states that the higher SGR starts in February-May, and start to go down on July-December. Hurtado et al (2008), stated that during 60 days of the lowest SGR maintenance found in July-August and the highest in January-February.

The present research findings shows that the highest carrageenan concentration of *E. spinosum* seaweed is attained in case of 50 g initial seed weight treatment with 50.03%, followed by 150 g initial seed weight with 49.51% and the lowest in 100 g initial seed weight with 46.96%. Carrageenan level concentration of *E. spinosum* seaweed in 50 g initial seed weight gives the highest response. Growth of *K. alvarezii* thallus occurs due to the absorption of nutrients and impacts on the growth of new thallus that will continue to emerge. Besides, it is presumed that when the thallus is still young, the only energy available is used for growth. After the growth reaches its maximum point, then the remaining energy is used to form the carrageenan. It is in accordance with Supit (2005) which stated that inside young sprouts the percentage of carrageenan is smaller than in older thallus.

The highest carrageenan level during the present research is in the treatment of 50 g initial seed weight, with 50.03%. Abdan (2013), reported that the highest carrageenan level on cultivated *E. spinosum* seaweed with long line method (49.44%). However, Erpin (2012) stated that the highest carrageenan level of *E. spinosum* seaweed cultivated with long line method was observed in case of 50 g initial seed weight with 45.13%, and the lowest in case of 70 g initial seed weight (45.00%). Suhardi (2013), found that the highest carrageenan level in *E. spinosum* seaweed cultivated with floating cage method occurs in treatment of 100 g initial seed weight with 33.44% and the lowest in treatment of 40 g initial seed weight with 27.67%. Thus different carrageenan value is caused by different cultivation season, but the differences do not have a distinct variance value. This is in accordance with Hung et al (2009), that that the increase in carrageenan is caused by increased growth in certain seasons particular in November and December. The differences are also caused by maintenance time, plant distance, extraction method, and extraction raw material. Hayashi et al (2007), stated that the best carrageenan condition could be achieved if the seaweed is cultivated for 45 days. Freile-Pelegrin et al (2006) stated that factors which could affect carrageenan quality are, season, light, nutrient, temperature, and salinity which also could reduce seaweed quality.

The growth of seaweed correlates with its carrageenan level, during the growth the carrageenan level increasing. Besides, the carrageenan level difference could also have caused by pigment contained in seaweed and which is also important for photosynthesis process to form carrageenan. Munaeni (2010) stated that photosynthesis is sun energy absorbance by plant cells which support seaweed optimal growth including the carrageenan level. *E. spinosum* has two life cycle phases which are the vegetative and the generative phase. On the vegetative phase, the energy is distributed for growth and carrageenan production, and then it continues on generative phase where the energy for carrageenan production is reduced for a generative process so that the concentration level is decreased while the growth still occurs reaching a maximum point. The differences in carrageenan levels could be caused by different extraction methods (Hayashi 2007).

Environmental parameters. The water quality in this research was analyzed to understand the quality range which can be tolerated and supports the life and growth of *E. spinosum* seaweed. Based on 45 days' measurement, the temperature in the research location ranges 28-31^{o}C. The results showed a tendency of increasing temperature from day 1 until day 45. The water temperature was relatively stable with not so drastic increase between morning and afternoon. That condition occurs because of the water location which has sun light exposure reaching down to the bottom of the ocean. The recorded temperature ranges was 28-31^{o}C which is suitable for the *E. spinosum* needs to grow optimally. Kordi (2010) stated that water temperature which is suitable for *Eucheuma sp.* seaweed is between 20 and 30^{o}C. Water temperature measurements showed that water condition in Konawe coast, Souteast Sulawesi, Indonesia is appropriate for *E. spinosum* seaweed growth. Salinity could also affect seaweed growth in the cultivation environment. Salinity correlates with osmotic pressure between seaweed and water environment in nutrient absorbance. Water salinity measurement results during the 45 days experiment revealed a range of 31-34 ppt. It showed a relatively good environment for seaweed growth. It is presumed that when the research was conducted, it was during drought season which basically has relatively low rain intensity. Besides that, the research location was far away from human population where the land activity influence is very low. Seaweed growth is high in tropical area with salinity level of 31-34 ppt (Kadi (2006). Sea current has big impact towards aeration, nutrient transportation, and water stirring, which affects the *E. spinosum* growth. Too strong current flow could provoke mechanical damage in thallus, so the cultivation location of *E. spinosum* should be always apart from current flow and violent waves (Sulma & Manoppo 2008). Current velocity speed during the research was in the range of 0.31-0.42 cm/second. Atmadja et al (1996) explained that ideal ocean current for *E. spinosum* cultivation is around 0.33-0.66 cm/second. Ocean water transparency correlates with how far the sun light can penetrate the water for photosynthesis process. Transparency measurements revealed that sun light can penetrate the ocean up to 5-8 m deep. From that finding, was showed that the transparency level is suitable for seaweed growth, which is presumed as uncontaminated water condition.

Water depth is also an important factor on seaweed growth. The lack of seaweed growth due to the increasing of water depth could be caused by few things such as sun light capability to penetrate the ocean water, and the lack of oxygen circulation on water column (Aditya et al 2001). The depth measurement has been done in the morning and afternoon for 45 days on tide of 7 m and when recedes on 5 m, the fluctuations are caused by tidal wave cycle. Seaweed should grow well in 4-17 m depth, so that nutrient absorbance is still continuing and seaweed does not damaged by direct sun light (Poncomulyo et al 2006). The nitrate concentration analysis was in the range of 0.0060-0.0104 mg/L. The high concentration was caused by land activity which produces organic and household pollutants. The current flow carrying dissolved organic substance so it affects seaweed distribution. Kordi (2010) stated that naturally the nitrogen dissolved into coastal water is carried by several components such as river surface currents, result of nitrogen fixation, perspiration and upwelling. Phosphate is one of the parameter which is needed for seaweed growth and basically forms orthophosphate. Phosphate recorded in the present research was in the range of 0.0019-0.0042 mg/L. Phosphate concentration is a pivotal factor for seaweed survival and growth rate. Phosphate and nitrogen is needed for seaweed for its growth; phosphate concentration which is appropriate for seaweed is 0.01-0.067 mg/L Effendie (2003). Basically phosphate that could be absorbed by seaweed is orthophosphate, while nitrogen is still absorbed as a form of nitrate or ammonium. Patadjai (2007) stated that phosphate needs for optimal growth for algae is influenced by a nitrogen substance. Highest phosphate concentration will be lower when it is in the form of ammonium salts. In contrast, if the nitrogen becomes nitrate, the phosphate concentrate which is required is much higher. Phosphate concentration for optimal seaweed growth is in the range of 0.18-0.90 mg/L and availability concentration in water column is in the range of 8.90-17.8 mg/L. Phosphate concentration in water column were from various source such us pollutant industry, domestic waste, organic raw breaks, and phosphor minerals. Nutrients become major and important component for seaweed growth as nitrate (NO_3) and phosphate (PO_4).

Conclusions. The growth of *E. spinosum* is different for each of its part of thallus. The younger thallus with lower weight has a relatively fast growth compared to the old thallus. Thallus with high growth has higher carrageenan content compared to other thallus. The results highligthed that distinctive initial seed weight affects (sig<0.05) the specific growth rate and carrageenan content of seaweed *E. spinosum.*

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