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Effectiveness of Feeding Trash Fish and Spinach Extract on Mud Crab (Scylla Serrata) Feed for Molting Acceleration With the Popeye Method

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Abstract. One of the fishery commodities in the mangrove forest area that has the potential to be cultivated in the mangrove crab (Scylla serrata). The local and international market demand for soft shell mud crab from year to year tends to increase. Most of the efforts to meet the needs of softshell crab still rely on catches from nature where availability is uncertain. The purpose of this study was to identify the effect of the long molting time of mud crab (Scylla serrata) fed trash feed and feed fed with spinach extract using natural methods and popeye. This research is experimental using a completely randomized design. The treatments consisted of the natural method with trash feed, the natural method with spinach extract feed, the popeye method with trash feed, and the popeye method with spinach extract feed. The crabs used are mud crabs with the hard carapace. The test parameter is the length of time for mud crab molting, which is calculated starting from the hard-shelled crab to molting. The data obtained in the form of differences in the length of time for mud crab molting using various treatments were analyzed descriptively quantitatively. The results of this study indicate that the popeye method with spinach extract feed is the treatment that produces the best molting time, which is 14-21 days compared to other treatments.

1. Introduction

One of the fishery commodities in the mangrove area that has the potential to be cultivated is mangrove crab (Scylla serrata) (Nirmala, 2011). The local and international market demand for mud crab from year to year tends to increase. According to Mirea and Moksnes (2013), mangrove crab is a commodity that has the opportunity to be cultivated because besides having a fast growth rate, it also has high economic value. The selling price of other types of crabs that live in mangrove forests is not as high as mangrove crabs (Scylla serrata). Apart from being popular with consumers because it is easy to consume, the protein content of soft-shelled mangrove crabs is also quite high, which is about 20 percent of the total nutritional content of crab meat (Mirera, 2011 and Dumas et al., 2012).

One of the important breakthroughs in soft shell crab cultivation that has been developed by Fujaya et al. (2012) found a molting stimulant derived from extracts of spinach (Amaranthaceae tricolor) called vitomolts and ecdysteroids. The application of spinach extract by injection is proven to accelerate molting, not cause death, and increase crab growth. On the other hand, the use of injection is considered less efficient for mass production of molting crab. Efforts that can be done are using artificial feed as a medium for the application of spinach extract. Fujaya et al. (2012) have conducted trials and proven that spinach extract can be given through artificial feed, and it is effective in accelerating molting and increasing growth. This study aimed to identify the effect of the long molting

time of mud crab (Scylla serrata) fed trash feed and feed fed with spinach extract using natural methods and popeye.

2. Materials and Methods

This research was conducted at the Mangrovesari Dukuh Pandansari Nature School, Kaliwlingi Village, Brebes District, in April - May 2020. The test animals used were mangrove crabs (Scylla serrata) with carapax length 6–8 cm/head and bodyweight 80–90 grams / 60 tails with carapace condition were kept in a basket with a size of 20 cm x 15 cm x 10 cm and carried out in mangrove ponds. This research was conducted experimentally, using a completely randomized design (CRD), with 4 treatments with 3 repetitions each. The layout of the treatment container was completely randomized. The treatment doses in the study were as follows: A1B1: Provision of 100% trash fish feed at a dose of 5% of the biomass weight using natural methods; A1B2: Provision of 100% artificial feed given spinach extract at a dose of 5% of the biomass weight using natural methods; A2B1: Administration of 100% trash fish feed at a dose of 5% of the biomass weight using the popeye method, and A2B2: 100% feed given spinach extract at a dose of 5% of the biomass weight using the popeye method.

The dose of feed is 5% of total body weight and feed is given once a day in the morning at 07.00, referring to Avianto's research (2011). Physico-chemical parameters of water (temperature, pH, and salinity) were observed every 3 days, morning at 06.00, afternoon at 14.00, and evening at 18.00. The length of time for mud crab molting was observed from the first day of treatment until the crab was released from the old carapace. Data analysis of the length of time for mud crab molting was carried out descriptively.

3. Results and Discussion

3.1. Absolute Individual Weight Growth

Absolute individual weight growth which is the difference in weight at the end of the study is presented in Table 1 and Figure 1.

Table 1. Growth of Absolute Individual Weight (gr) Mud Crab (Scylla serrata)

Repeat	A1B1	A1B2	A2B1	A2B2
1	46,40	44,00	45,80	48,40
2	44,20	43,80	42,40	53,60
3	49,00	47,80	43,20	49,20
Average	46,53±2,40	45,20±2,25	43,80±1,97	50,40±2,80

Source: Research results (2020)

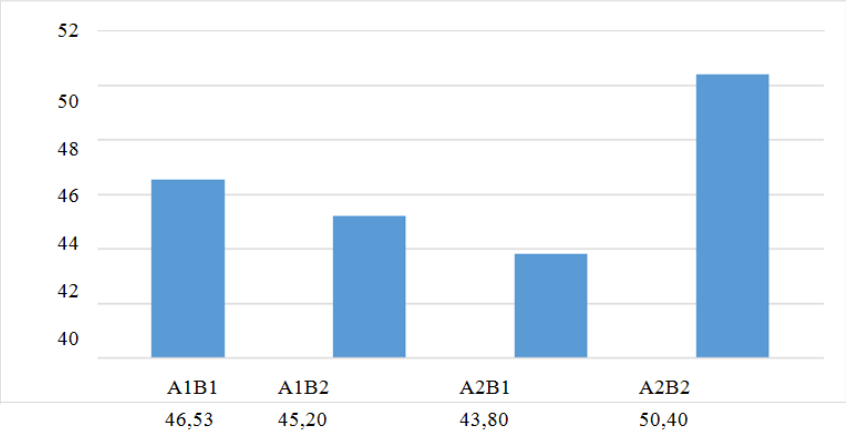

Figure. 1. Growth of absolute individual weight (gr) of mud crab (Scylla serrata)

Bodyweight growth ranged from 43.80 to 50.40 grams with a standard deviation ranging from 1.97 to 2.80. The results of the normality and homogeneity tests on the absolute weight growth data of mangrove crabs (Scylla serrata) show that the data are normal and homogeneously distributed. The results of the analysis of variance showed that the treatment is given had a significant effect (P <0.05) on the weight growth of mud crab seeds (Scylla serrata). There was a significant difference in weight between treatment methods and differences in feed given (Sig = 0.44; p> 0.05). The highest body weight growth of tested animals was in A2B2 treatment (50.4 grams), which was followed by A1B1 treatment (46.53 grams), A1B2 treatment (45.2 grams), and A2B1 treatment (43.8 grams).

3.2. Absolute Individual Length Increase

The feeding of trash feed and artificial feed is given spinach extract using natural methods and popeye to the test crabs are presented in Table 2 and Figure 2.

Table 2. Growth of absolute individual length (cm) of mangrove crabs (Scylla serrata)

Repeat	A1B1	A1B2	A2B1	A2B2
1	1,5	2,2	1,4	3,4
2	2,0	2,0	2,0	3,5
3	2,2	2,7	2,1	2,9
Average	1,9±0,36	2,3±0,36	1,8±0,38	3,2±0,32

Source: Research results (2020)

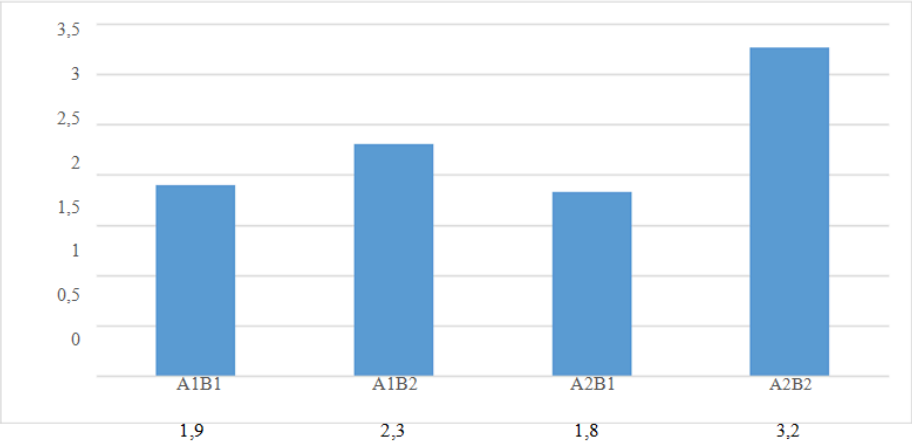

Figure. 2. Increase in length (cm) of mangrove crabs (Scylla serrata)

The results of the normality and homogeneity test on the data on length increase of mud crab seedlings (Scylla serrata) showed that the data were normally distributed and homogeneous. The results of the analysis of variance showed that the treatment given was significant (P <0.05) on the length growth of mangrove crab seedlings (Scylla serrata). There was a significant difference between the length of the crab carapace due to differences in the treatment of the feed given (Sig = 0.022; p <0.05). The highest increase in carapace length of the tested animals was obtained in treatment A2B2 (3.26 cm), then treatment A1B2 (2.3 cm), treatment A1B1 (1.9 cm), and treatment A2B1 (1.83 cm).

3.3. Molting Percentage

The molting percentage of mud crab was significantly different from the different treatment of trash feed and artificial feed is given spinach extract with natural and popeye methods as presented in Table 3 and Figure 3. The molting percentage of mangrove crabs observed weekly is presented in Figure 4.

Table 3. Molting percentage (%) of mangrove crabs (Scylla serrata) during the study

Repeat	A1B1	A1B2	A2B1	A2B2
1	80,00	80,00	80,00	60,00

2	100.00	80,00	80,00	80,00
3	60,00	80,00	60,00	80,00
Average	80,00±11,54	80,00±0,00	73,33±11,54	73,33±11,54

Source: Research results (2020)

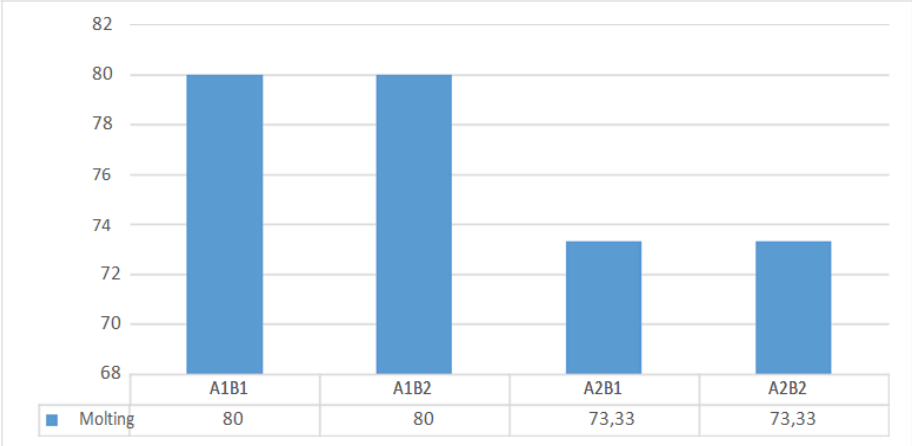

Figure. 3. Molting percentage (%) of mangrove crabs (Scylla Serrata) during the study

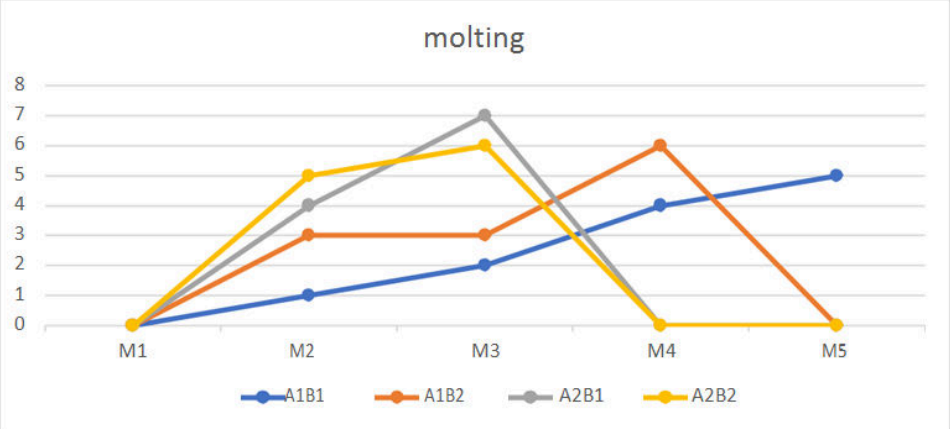

Figure. 4. Molting graph of mud crab (Scylla serrata) per week (%)

The molting percentage produced in this maintenance ranges from 73.33 - 80%. The results of the normality and homogeneity test of the mud crab molting data showed that the data were normally distributed and homogeneous. The results of the analysis of variance showed that the treatment is given had a significant effect (P <0.05) on molting mud crabs (Scylla serrata). There was a significant difference in molting between the given feeding treatments (Sig = 0.166; p <0.05). The highest percentage of molting was produced in the A1B1 and A1B2 treatments (80%), followed by A2BA and A2B2 treatments (73.33%).

3.4. Daily Growth Rate

Data on the percentage of daily growth rates for each research treatment are presented in Table 4 and Figure 5.

Table 4. Percentage of daily growth rate for mud crab (Scylla serrata)

Repeat	A1B1	A1B2	A2B1	A2B2
1	0,828	0,992	0,964	0,692
2	1,228	1,098	0,960	1,018

3	0,920	1,218	0,978	1,066
Average	0,992±0,209	1,102±0,113	0,967±0,094	0,925±0,203

Source: Research results (2020)

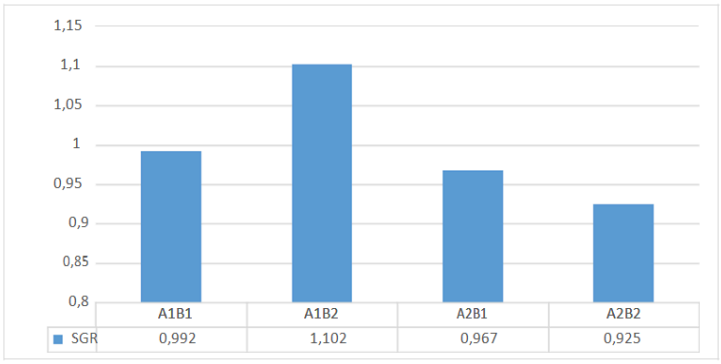

Figure. 5. Percentage of daily growth rate (%) mud crab (Scylla serrata)

The results of the normality and homogeneity tests on the daily growth rate data of mangrove crab seeds (Scylla serrata) showed that the data were normally distributed and homogeneous. The results of the analysis of variance showed that the treatment given gave a significant response to the growth rate of mangrove crab (Scylla serrata) (P <0.05). There was a significant difference in daily growth rates in response to different feeds given (Sig = 0.727; p> 0.05). The highest daily growth rate was obtained in A1B2 treatment (1.102 cm), then A1B1 treatment (0.992 cm), A2B1 treatment (0.967 cm), and A2B2 treatment (0.925 cm).

3.5. Survival

Data on the percentage of survival for each treatment can be seen in Table 5 and Figures 6 and 7.

Table 5. Percentage of survival (%) mangrove crabs (Scylla serrata)

Repeat	A1B1	A1B2	A2B1	A2B2
1	0,828	0,992	0,964	0,692
2	1,228	1,098	0,960	1,018
3	0,920	1,218	0,978	1,066
Average	0,992±0,209	1,102±0,113	0,967±0,094	0,925±0,203

Source: Research results (2020)

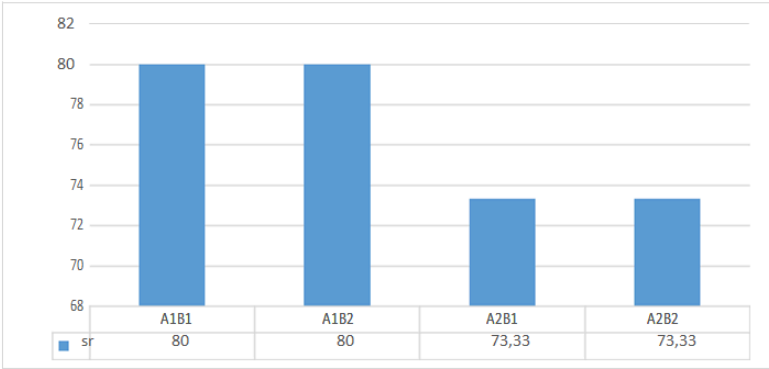

Figure. 6. Percentage of survival (%) mangrove crabs (Scylla serrata)

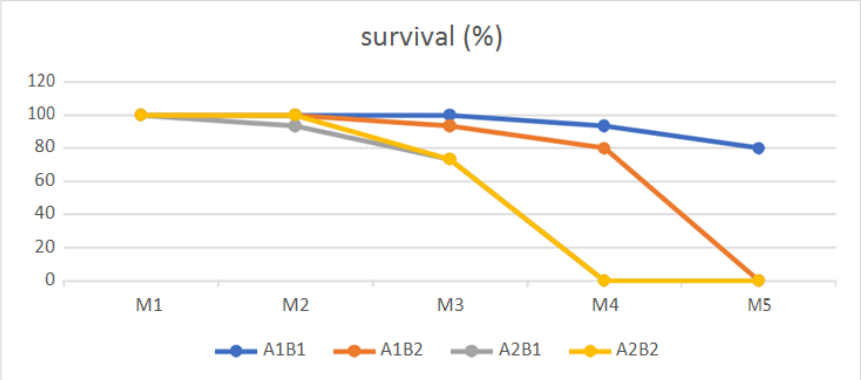

Figure. 7. Graph of survival of mud crab (Scylla serrata) per week

The results of normality and homogeneity tests on the survival data of mangrove crabs (Scylla serrata) showed that the data were normally distributed and homogeneous. The results of the analysis of variance showed that the treatment is given had a significant effect (P <0.05) on the survival of mangrove crab seedlings (Scylla serrata) and there was a significant difference between the survival of mangrove crabs between the different feed treatments (Sig = 0.166; p> 0.05). The highest survival was obtained in the A1B1 and A1B2 treatments (80%), then the A2B1 and A2B2 treatments were (73.33%).

3.6. Relative Growth Rate (RGR)

Percentage data on relative growth rates in each treatment can be seen in Table 6 and Figure 8.

Table 6. Percentage of relative growth rate (%) mud crab (Scylla serrata)

Repeat	A1B1	A1B2	A2B1	A2B2
1	80,00	80,00	80,00	60,00
2	60,00	80,00	60,00	80,00
3	100,00	80,00	80,00	80,00
Average	80,00±20,00	80,00±0,00	73,33±11,54	73,33±11,54

Source: Research results (2020)

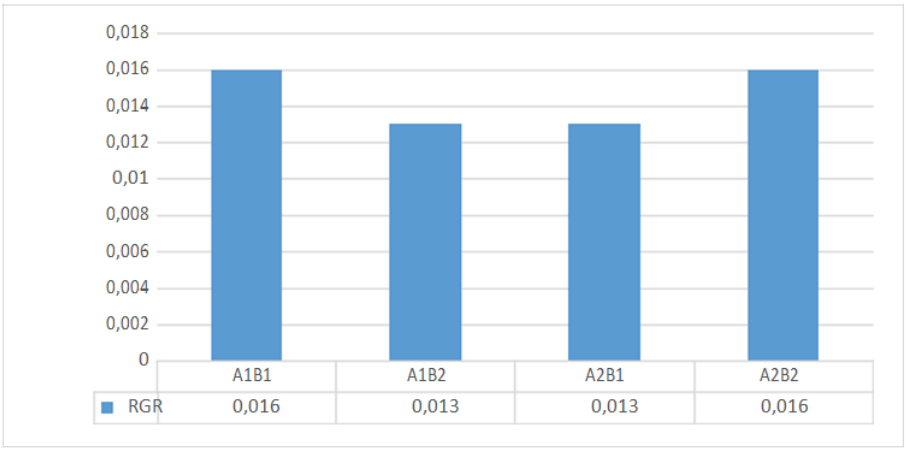

Figure. 8. Percentage of relative growth rate (%) of mangrove crabs (Scylla serrata)

The results of the normality and homogeneity tests on the daily growth rate data of mangrove crab seeds (Scylla serrata) showed that the data were normally distributed and homogeneous. The results of the analysis of variance showed that the treatment is given had a significant effect (P <0.05) on the daily growth rate of mangrove crab seeds (Scylla serrata) and there was a significant difference in the

growth rate of the length between the different feed treatments given (Sig = 0.213; p> 0,05). The highest daily growth rate was obtained in the A1B1 and A2B2 treatments, namely (0.016%), then the A1B2 and A2B1 treatments were (0.013%).

3.7. Mortality

The data on the percentage of mortality in each treatment can be seen in Table 7 and Figure 9.

Table 7. Percentage of mud crab (Scylla serrata) mortality

Repeat	A1B1	A1B2	A2B1	A2B2
1	20,00	20,00	26,67	26,67
2	40,00	20,00	40,00	40,00
3	0.00	20,00	20,00	20,00
Average	20,00±20,00	20,00±0,00	26,67±10,18	26,67±10,18

Source: Research results (2020)

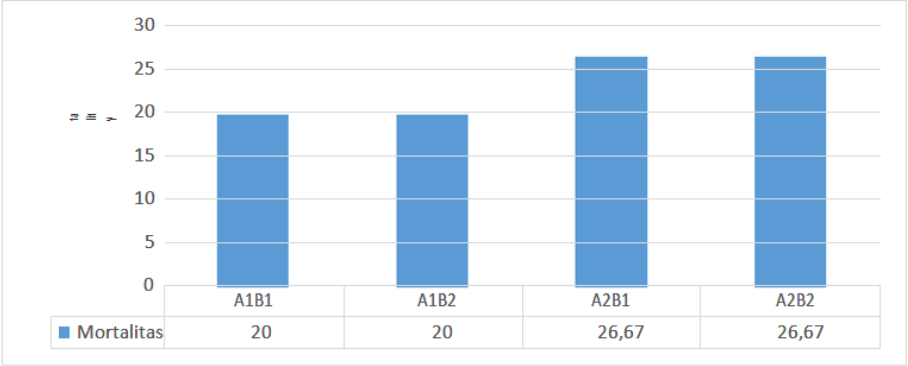

Figure. 9. Percentage of mortality (%) mud crab (Scylla serrata)

The results of the normality and homogeneity tests on the mortality data of mangrove crabs showed that the data were normally distributed and homogeneous. The results of the analysis of variance showed that the treatment is given had a significant effect (P <0.05) on the mortality of mangrove crabs (Scylla serrata). There was a significant difference in mortality between treatment methods and differences in feed given (Sig = 0.166; p> 0.05). The percentage of mortality resulting from this breeding ranges from 20-26.67%. The highest percentage of mortality resulted in treatment A2B1 and A2B2 (26.67%), followed by A1B1 and A1B2 treatments (20%).

Mangrove crabs reared using the popeye method and feeding spinach extract requires the fastest molting time, which is around 14-21 days compared to those given other treatments, namely the natural method by feeding trash fish as presented in Table 8.

Table 8. The length of time for molting mud crabs after being treated

Treatment	Average of molting (days)
A1B1	30-33
A1B2	18-30
A2B1	16-25
A2B2	14-21

Source: Research results (2020)

The results of measurements of several important physicochemical parameters of water, including air temperature, water temperature, pH, and salinity include air temperature, water temperature, pH, and salinity including air temperature, water temperature, pH, and salinity are presented in Table 9. In general, the water chemistry-physics parameters are still in a sufficiently suitable range for mangrove crab cultivation.

Table 9. Water Quality Measurement Results

Standard	Range Field Measurement	Range Parameters (Ikhwanudin, et al., 2012 and Hastuti, et al., 2019)
Air temperature (°C)	27 – 33,2	23–32
Water temperature (°C)	27–37	23–33
Salinity (‰)	23,9 – 30	10–30
pH	7,5 – 8,8	7,0 - 8,5

Source: Research results (2020)

4. Discussion

Based on the results of observations on the growth of mangrove crabs during 35 days of rearing, the success rate of molting in mangrove crabs resulted in different times. Growth is influenced by two factors, namely internal factors which include heredity, age, sex, reproduction, disease resistance, and the ability to utilize feed, while external factors include water quality, density, and feed (Hubatsch, et al., 2015). A2B2 treatment (feed fed with spinach extract and using the popeye method) was the best treatment during this study. The absolute high growth rate in A2B2 treatment is suspected because the popeye method and spinach extract are more effective in accelerating ecdysteroid hormone stimulants. Spinach extract is a molting stimulant that contains a molting hormone (phytoecdysteroid). Ecdysteroid is the main steroid hormone in arthropods and crustaceans which functions as a molting hormone, besides that it also regulates physiological functions, such as growth, metamorphosis, and reproduction (Djunaedi, et al., 2018).

Fast growth also indicates the availability of food and other favorable environmental conditions, while slow growth indicates the opposite. Generally, fish have very rapid long growth in the first few months or years of their life, until maturation. Furthermore, additional energy is used for the growth of somatic and gonadal tissues, so that the growth rate of mature fish is slower than immature fish (Septian, et al., 2013).

According to Avianto (2011) growth in mangrove crabs is characterized by changes in shape and size due to differences in growth rates of different body parts. The amount of growth experienced by crabs depends on the increase in length and weight of each molting crab. The frequency of molting varies depending on the size and stage of the crab. Based on the research results, the highest percentage of molting was shown by the A2B1 and A2B2 treatments at 80%. Spinach can shorten the duration of molting because it contains an ecdysteroid.

According to Rusmiyati (2011) and Aslamyah and Fujaya (2011), external factors that affect molting include water quality and the type of feed consumed. These factors will affect the brain and stimulate the Y-organ to produce an ecdysteroid or molting hormone. Ecdysteroids are hormones that play a role in controlling molting in Arthropods and Crustacea. Ekdysteroid is secreted by the Y organ in the form of ecdysone. In the hemline, this hormone is converted into an active hormone, 20-hydroxyecdysone, by the enzyme 20-hydroxylase, which is found in the epidermis of the organ (Fujaya et al., 2012). According to Habibi, et al. (2013) the molting process begins when epidermal cells respond to hormonal changes through the rate of protein synthesis. The increase in protein synthesis rate due to stimulation of the molting hormone causes apolysis (physical separation between the epidermis and the endocuticles). Next, the epidermal cells fill the gap with an inactive molting solution and then secrete a special lipoprotein or cuticulin layer. The cuticulin layer will form part of the new apiculate. After the formation of the cuticulin layer, the molting solution becomes active and the chemical substance will digest the endocuticles of the old exoskeleton. The cuticulin layer will produce amino acids and microfibrils which are then recycled by the epidermal cells and secreted under the cuticulin layer as new soft and wrinkled procuticles. When the new exoskeleton is ready, muscle contraction and air filling cause the body to swell so that the old exoskeleton cracks along the ecdysteroid sutures, and finally the body with the new exoskeleton leaves the old exoskeleton. After that, the new exoskeleton, which is still soft and wrinkled, will stretch out after being filled with water so that the size of the crab increases after molting (Hastuti, et al., 2016).

In addition to the addition of spinach to feed, the cutting technique of the legs can increase the production of soft shell because cutting the legs can stimulate the release of the hormone exdecis which triggers the rapid molting of mangrove crabs, thereby increasing the growth and survival and production of mangrove crabs will also increase with the correct technique. Septian, et al. (2013) stated that changes in body shape are influenced by the regeneration of missing or broken limbs. If any part of the crab's body is missing or broken, the energy for growth is more focused on forming new tissues of missing or broken limbs. Furthermore, it was also stated that the cutting technique of the walking leg on the mangrove crab which was tested had a positive effect on organ X which produced MOIF (mandibular organ-inhibiting factor) which served to inhibit the performance of the mandibular organ to produce MIH (molecular inhibiting hormone) which inhibited organ Y and produce MF (methyl farnesol) which stimulates the work of Y organ. Y organ produces ecdysteroid, and the activation of the ecdysteroid triggers skin molting (molting). The popeye method (cutting/mutilation of all walking legs) showed a higher growth value than the other treatments. This is presumably because the mangrove crabs that are given mutilation treatment of all walking legs can still grab the feed and consume the feed as energy for growth. According to Hubatsch, et al. (2015), mangrove crabs utilize fat, protein, and carbohydrates as energy supplies for embryo development, however, most of this energy is used for shell formation.

Age and mortality are good predictors of relative growth rates, whereas absolute growth rates are strongly influenced by environmental factors. Furthermore, internal factors such as heredity, age, and ability to utilize feed in this study are also quite difficult to equate because the mangrove crabs used as test animals are obtained from natural catches of fishermen, of course from different parents (Saidah and Leila, 2016).

5. Conclusions and Suggestions

5.1. Conclusion

Based on the research results, it can be concluded that:

a. Different methods and different feeding affect absolute individual weight (gram), absolute individual length (cm), molting percentage (%), daily growth rate (grams), survival (%), relative growth rate (grams), and mortality to accelerated mud crab molting (Scylla serrata).
b. The survival rate of mangrove crabs (Scylla serrata) during the study ranged from 73.33% - 80%.
c. A2B2 treatment (popeye method and artificial feed with spinach extract) was the best treatment during the study.
d. Water chemical characteristics during the research were still feasible for mangrove crab (Scylla serrata) cultivation.

5.2. Suggestion

Cultivation of softshell mangrove crabs should be done using the popeye method. Further studies are needed regarding the optimal levels of spinach extract contained in a feed formulation for mud crabs (Scylla serrata).

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