The impact of fresh and artificial diet on growth and survival rate of spiny lobster, *Panulirus homarus* reared in floating net cage

Ni Wayan Widya Astuti[1*], *Sudewi* Sudewi[1], *Bejo* Slamet[1], *Fahrudin* Fahrudin[1], and *I Nyoman Adiasmara* Giri[1]

[1]Research Center for Fishery, National Research and Innovation Agency, Cibinong, Bogor, Indonesia

Abstract. Some research found that the growth of lobster was higher when fed with fresh diet than artificial diet. Therefore, the growth of lobster that is fed with combination of fresh and artificial diets need to be observed. This study aimed to evaluate different kinds of feed and its combination on growth and survival rate of spiny lobster *Panulirus homarus*. This research was designed with three treatments and three replicates, namely: lobster fed with fresh diet (A), lobster fed with artificial diet (dry pellet) (B), and lobster fed with combination of fresh and artificial diets (C). Lobsters with an average weight 100.30 ±0.79 g were reared in floating net cage with dimension 2×2×2 m^3 at density 40 lobsters/cage. Lobsters were fed twice a day at 08.30 am and 03.30 pm. Fresh diets that were given contained fresh fish, shrimp, crabs and mussels (3:1:1:1). The observation was conducted for 18 weeks. Results of the experiment were lobsters fed with fresh diets (A) and combination of fresh and artificial diets (C) gave higher specific growth rate (0.62±0.03 %/day and 0.61±0.03 %/day) than lobster fed with artificial diets (B) (0.33±0.01 %/day). There were not significantly differences in survival rate of lobster ($p<0.05$).

1 Introduction

Lobsters are known as a valuable crustacean that has high economical value in the market. The demand of lobsters are high not only in domestic, but also in international market [1]. To fulfil the demand of the market, lobster supply still depend on capturing activities. Because of the unselective fishing potentially could reduce lobster population in Indonesia, The Ministry of Marine Affairs and Fisheries published a regulation about the restriction of lobster (*Panulirus* spp.), crabs (*Scylla* spp.) and swimming crab (*Portunus pelagicu*s spp.) in 2015. That regulation leads aquaculturist to concern about lobster farming and increase the production of lobster from aquaculture [2].

Lobster farming in Indonesia develop as grow-out system, which the seed suplies come from capturing activity. The undersized lobster that are caught by the fisherman are reared in floating net cage until meet the market size. Lobster farming in Indonesia are develop in

* Corresponding author: nw.widyaastuti@gmail.com

Lombok, Bali, Java, Sumbawa, Sumatera and Sulawesi [3]. Lobster that are caught in Indonesia are *Panulirus ornatus*, P. *homarus*, P. *polyphagus* and P. *versicolor*. The main lobster that are captured is spiny lobster (*Panulirus homarus*) [4].

Lobster grow-out in floating net cage depends on fresh fish as diets. Crustaceans and mollusks also could be feed source of the lobster [5]. For sustainable reason, artificial feed for lobster had been developed by some researcher. The artificial feed in form of pellets, were studied to find the right formula to support lobster growth. The challenge in developing lobster feed is artificial feed gives lower growth performance [6]. Therefore, feeding management in lobster aquaculture is essential.

A combination of fresh and artificial feed for grow-out of lobster had been experimented in controlled tanks [1]. The result of this experiment showed that the combination of fresh food and formulated diet gave better growth performance, but the survival rate was better when the lobster were fed formulated diet. Although culturing of lobster in indoor tanks provides advantages in terms of controlling the environmental factors most importantly on maintaining water quality, however, farmers prefer to grow lobsters in floating net cages for economic reasons. Hence, a further study which performed in floating net cages is required. A previous study reported that this method has some challenges, such as fluctuation of water quality, high labour of handling, difficult to control cannibalism and also hard to ensure feed is eaten efficiently by lobster [2]. Consequently, it is important to manage grow-out of lobster in floating net cages mostly the feeding regimes for the efficient use of feeds in order to have optimal growth and survival. Therefore, the present study was designed to evaluate the combination of fresh food with artificial diet for growing-out lobster and its impact on survival rate.

2 Materials and methods

This research was conducted in floating net cage of Institute for Mariculture Research and Fisheries Extension (IMRAFE) in Pegametan Bay-Bali, Indonesia. This stage of research including preparation of artificial diets, feeding trial and parameters measurement and data analysis.

2.1 Preparation of artificial diet

The artificial diets were formulated based on the previous research [1]. The ingredients of the diet showed in table 1. The feed was prepared using pelleting machine with 3 mm of diameter. The pellet was dried in oven with temperature 65°C and stored in refrigerator.

Table 1. Composition of artificial diet

Ingredients	Proportion (%)
Fish meal	78.3
Wheat flour	6
Wheat gluten	6
MOS	0.5
Fish oil	2.6

Astaxanthin	1
Cholesterol	0.5
Lecithin	1.7
Mineral premix	0.6
Vitamin premix	1.1
Stay C	0.4
CMC	1.3
Total	100

2.2 Proximate analysis

Nutrient values of artificial diets and fresh feed were analyzed by proximate analysis. The parameters of proximate that were analyzed i.e.: protein, lipid, ash and moisture. Moisture content was analyzed to count dry matter nutrients

2.3 Feeding trial

Lobsters that were reared in this experiment were captured at Southern Bali Coastal Area. Before stocking in the net cage, lobster was acclimated in fiberglass tank for 7 days. A number of 40 lobsters were allocated into cages of 2×2×2 m. Initial carapace length of the lobster was 6.19±0.04 cm, and the initial weight were 100.3±0.79 g. Floating net cage were equipped with squared parallon with dimension 1×1×0.3 m. Net were added on the top of the parallon. Gracillaria were added on the top of the net in amount 5 kg/cage. This parallon and gracillaria function were as shelter for lobsters (Fig 1).

Fig. 1. Square parallon with net (left) and gracillaria (right) as shelter for lobster

This research used completely randomize design (CRD) with 3 treatments and 3 replicates for each treatments. The feed treatments were (A) lobster fed with fresh food, (B) lobster fed with artificial diet, (C) lobster fed with combination of fresh food and artificial diet. Lobster were fed two times daily, at 09.00 am and 15.00 pm. Feed were allocated 30% from total feed daily in the morning and 70% from total feed daily in the evening. Fresh food were given 10% from biomass, meanwhile artificial diets were given 3% from biomass. For the

combination diets treatments, fresh food were given 5% and artificial diets were given 1.5% daily. The fresh food that was given to the lobster was a mixture of fish, crab, shrimp and mussel with the ratio 3:1:1:1.

2.4 Parameters observed and data analysis

The parameters observed in this research were lobster growth (carapace length and body weight), specific growth rate, survival rate, BRIX Index and total haemocyte count (THC). BRIX index represents serum protein contained in the lobster's haemolymph. Length and weight were observed every 3 weeks, meanwhile Brix index and THC were measure at the end of treatment. The feeding trial was conducted for 127 days. Water quality that were monitored during treatment i.e. temperature, pH, salinity, ammonia, nitrite and phosphate.

Carapace length increment, weight gain, specific growth rate and survival rate were calculated with this formula:

$$Carapace\ length\ increment\ (cm) = L_t - L_0 \quad (1)$$

Where L_0 : initial length (cm)
L_t : final length (cm)

$$Weight\ gain\ (g) = W_t - W_0 \quad (2)$$

Where W_0 : initial weight (cm)
W_t : final weight (cm)

$$Specific\ Growth\ Rate\ (\%/day) = \frac{lnW_t - lnW_0}{t} \times 100 \quad (3)$$

Where W_0 : initial weight (cm)
W_t : final weight (cm)
t : treatment period (days)

$$Survival\ Rate\ (\%) = \frac{N_t}{N_0} \times 100 \quad (4)$$

Where N_0 : initial number of lobster
N_t : final number of lobster

Data were presented in table and analyzed by Analysis of Variance (ANOVA) followed by Duncan test with 95% confidence interval using SPSS 14.0 for windows.

3 Result and Discussion

Feed is energy source for lobster. Lobster is carnivorous crustacean that consume feed which contain high protein, low lipid, moderate to high carbohydrate [7]. In this study, lobster were fed mixture fresh food or artificial diet or combination of the two feed. Nutrient content of the feed presented in table 2.

Table 2. Nutrient composition of artificial diet and fresh food (% dry matter)

Nutrient	Artificial diet	Fresh Food			
		Fish	Shrimp	Crab	Mussel
Protein (%)	58.98	69.63	72.24	51.18	70.55

Lipid (%)	15.65	10.35	6.97	3.08	5.38
Ash (%)	15.45	17.84	11.54	35.27	15.20

The nutrient analysis showed that protein, lipid and ash value are vary between all kind of feed that were given to lobster. But in general, all the feed had high protein content. The highest protein content was found in shrimp and the lowest was found in crab. The highest lipid content was found in artificial diet, and the lowest was found in crab. Ash analysis showed the opposite than protein, the highest ash content was found in crab, and the lowest was found in shrimp. Crab that were given to lobster was not only the meat, but also some shells. The shells contain mineral, that caused high value of ash. Highest lipid content in artificial diet could be come from fish meal and fish oil.

Growth of lobster in this experiment presented in table 3. Different feed resulted different lobster growth. Carapace length grew from 6.19 cm in average to 7.01±0.07 cm in treatment artificial diet (A), 7.90±0.11 cm in treatment fresh food (B) and 7.92±0.11 cm in treatment combination of fresh food and artificial diet (C). The highest carapace length increment was found in treatment C (1.76±0.14 cm) and the lowest was found in treatment B (0.83±0.05 cm). Carapace length increment in treatment A were statistically different with treatment B but was not statistically different with treatment C.

The same pattern also found in parameter weight gain and specific growth rate. Lobster final weight were higher in treatment A (120.0±8.2 g) and C (118.0±8.4 g) and lower in treatment B (51.3±2.3 g). Specific growth rate was statistically different between treatment A (0.62±0.03 %/day) with treatment B (0.33±0.01 %/day), but was not statististically different with treatment C (0.61±0.03 %/day). This result showed that lobster growth was better while fed fresh food or combination between fresh food and artificial diet.

Table 3. Lobster growth performance.

Parameters	**A**	**B**	**C**
Initial carapace length (cm)	6.24±0.03	6.18±0.05	6.16±0.05
Final carapace length (cm)	7.90±0.11	7.01±0.07	7.92±0.11
Carapace length increment (cm)	1.66±0.14^{a}	0.83±0.05^{b}	1.76±0.14^{a}
Initial weight (g)	101.2±1.0	99.7±2.2	100.0±1.7
Final weight (g)	221.1±7.3^{a}	151.0±4.0^{b}	217.9±7.7^{a}
Weight gain (g)	120.0±8.2^{a}	51.3±2.3^{b}	118.0±8.4^{a}
Specific growth rate (%/day)	0.62±0.03^{a}	0.33±0.01^{b}	0.61±0.03^{a}

Note: different superscript letters in the same column show significant differences ($P<0.05$)

Lobster growth were higher when the feed contains fresh food and grew slowly when fed artificial diets [3]. This result also reported in lobster with the same feed that were reared in fiber tank with controlled environment [1]. Juvenile *P. argus* that fed with artificial diets showed lower growth rate than lobster that fed with fresh squid [8]. High level of chemo-attractant substances are found in fresh ingredients, which could promote feeding responses in crustaceans [9]. Higher growth in treatment combination of feed could be caused by this

treatment has better nutrient balance that come from artificial feed and fresh food [1]. For better growth and survival rate, a study of feed for lobster suggested to include fresh fishery products in formulating diets [10].

BRIX index of lobster that were fed with artificial feed (12.95±0.31 %) is lower than the lobster that fed with fresh food (17.60±0.63%) and combination of fresh food and artificial diet (18.02±2.03%) (table 4). BRIX index describes sugar content in aqueous solution, and it is correlated with serum protein consentration in blood. Nowadays, this measurement widely used in laboratory or industry to asses health status of lobster [11]. The high BRIX index in treatment fresh food and combination of fresh food and artificial diet correlated with better growth. Presumably it is because the lobster got better nutrient and could metabolized the nutrient especially protein so the growth could be better than the one that fed with artificial feed. The high BRIX index showed higher protein level in haemolymph and also correlated with molting process. The BRIX index will reach the maximum value before the molting period, and then will decrease after molting [12].

There was no significant different in all the treatment for total haemacyte count. THC for treatment fresh food, artificial diet, and combination of fresh food and artificial diet were 83.67 $\times 10^6$ cell/mL, 96.83 $\times 10^6$ cell/mL, and 97.33 $\times 10^6$ cell/mL respectively (table 4). THC is associated with health status and immunity of lobster [13]. High THC value represented good immune status. In normal condition, THC concentration of *P. cygnus* was in the range 5.6±0.7 x 10^6 cells/mL [13]. In this research, THC value in all treatment are higher. It means the immunity respons of the lobster were active to protect lobster from stress condition. Stress status in lobster can be caused by several condition, i.e.: environmental factors, handling and bacterial infection [14]. Increasement of THC concentration indicated an immune response to defend the lobster from disease [15].

Table 4. BRIX Index and Total Haemocyte Count of lobster at the end of treatment.

Parameters	A	B	C
BRIX Index (%)	17.60 ± 0.63^a	12.95 ± 0.31^b	18.02 ± 2.03^a
Total Haemocyte Count ($\times 10^6$ cell/mL)	83.67 ± 17.97^a	96.83 ± 3.88^a	97.33 ± 6.25^a

Note: different superscript letters in the same column show significant differences ($P<0.05$)

THC value was related with survival rate of lobster. Lobster mortality were very low until 63 days of treatment, and increased especially in treatment fresh food until lobster were treated for 127 days (fig. 2). Survival rate (SR) of lobster was not different significantly among treatments. SR values for treatment fresh food, artificial feed and combination of fresh food and artificial feed were 73.3±29.0%, 82.5±9.0%, and 85.0±0% respectively.

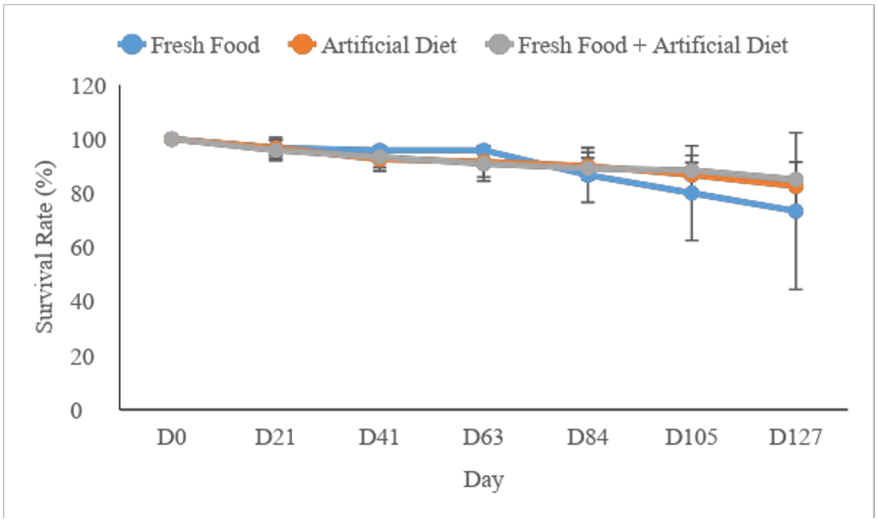

Fig. 2. Survival rate of lobster fed fresh food and artificial diet

Mortalities of lobster in this treatment were caused by cannibalism and disease. The disease that infected lobster after D63 was milky haemolymph disease (MHD). MHD is categorize as fatal disease for lobster farming [16]. MHD is caused by rickettsia like bacteria which spreading in hemolymph and show milky appearance [17]. Fresh food could be disease agent that caused higher mortalities in treatment fresh food. There was addition of vitamin, mineral and other nutrient in artificial feed. This nutrition resulted in better nutritional condition in treatment artificial feed and combination of fresh food and artificial feed.

In floating net cage, the water distribution relative similar from one cage to another. Water quality in floating net cage cannot be controlled as in the tanks. Water quality values during treatment were shown in table 5. Compared with another research, this range of water quality classified as optimum for lobster rearing [1, 3, 17, 18, 19, 20]

Table 5. Water quality in floating net cage during treatment

Parameters	**Values**	
	Result	**Optimum**
Dissolved oxygen (mg/L)	5.05-5.28	2.7-5.4
Temperature (°C)	28.9-29.5	27-32
Salinity (‰)	32-33	30-35
Ammonia (mg/L)	0.0592-0.0621	<1
Nitrite (mg/L)	<0.0087	<5
Phosphate (mg/L)	0.0059-0.00425	<0.015
pH	8.09-8.21	7.0-8.5

4 Conclusion

This study found that fresh diets (A) and the combination of fresh and artificial diets (C) resulted in higher specific growth rate (0.62±0.03 %/day and 0.61±0.03 %/day) than artificial diets (B) (0.33±0.01 %/day). While the survival rates of lobster were not different between the treatments.

Acknowledgement

This study was funded by Ministry of Marine Affairs and Fisheries of The Republic Indonesia in 2020. The author would like to thank to Selamat, Mujiono, Made Sulandra, Made Sutama, Ida Putu Suta, Sumardi, Made Suparya

References

1. Sudewi, N.W.W. Astuti, Fahrudin, I.N.A. Giri, B. Slamet, Growth and survival of spiny lobster, Panulirus homarus fed fresh food and formulated diet, in Proceedings of the 2nd International Conference on Fisheries and Marine, ICFM, 15 July 2021, Ternate, Indonesia (2021)
2. R.Y. Subhan, E. Supriyono, Widanarni, D. Djokosetiyanto, Jurnal Akuakultur Indonesia **1**7 1, 53-60 (2018)
3. N.W.W. Astuti, Sudewi, N.A Giri, Fahrudin, Y.N. Asih, B. Slamet, Fish and shrimp hydrolysate as additives in lobster feed formulation: effect on growth, survival, feed consumption of spiny lobster (Panulirus homarus), in Proceedings of the International Seminar Marine Biodiversity. Utilization, Conservation and Management, MarBioUtiCoM, 8-9 August 2022, Bogor, Indonesia (2023)
4. C.M. Jones, T.L. Anh, B. Priyambodo, Lobsters: Biology, Fisheries and Aquaculture, eds E. Radhakrishnan, B. Phillips, G. Achamveetil, Singapore, 541-570 (2019)
5. B. Slamet, I. Rusdi, N.A. Giri, Haryanti, Post puerulus of scalloped spiny lobster, Panulirus homarus (Linnaeus 1758) rearing in floating net cage with different artificial diet, in Proceedings of the Ecological intensification: A new paragon for sustainable aquaculture, 28-30 October 2019, Bogor, Indonesia (2020)
6. E. Perera, C. Simon C, Rev. Aquacult. **7**, 243-261 (2014)
7. K.C. Williams, Aquac. **263**, 1–14 (2007)
8. L. Rodriguez-Viera, E. Perera, Revista de Investigaciones Marinas **32** 1, 9–15, (2012)
9. S.J. Irvin, S Shanks, Tropical Spiny Lobster Feed Development: 2009 – 2013, ACIAR Proceeding 145, Canberra, Australia (2015)
10. G. Wang, I.J. Mc Gaw, J. Shellfish Res. **33** 3, 805-813 (2014)
11. K. Vidya, S. Joseph, Indian. J. Fish. **59**, 113-118 (2012)
12. H.M. Sang, R. Fotedar, Fish Shellfish Immunol. **28** 3, 483-489 (2010)
13. J. Jusilla, J. Jago, E. Tsvetnenko, B. Dunstan, L.H. Evans, Mar. Freshw. Res. **48**, 863-868 (1997)
14. S. Lorenzon, M. Martinis, D. Borme, E.A. Ferrero, Fish. Res. **137**, 9-17 (2013)
15. E. Supriyono, R. W. Prihardianto, K. Nirmala, AACL Bioflux **10** 2, 147-155 (2017)
16. Sudewi, Z. Widiastuti, B. Slamet, K. Mahardika, Indones. Aquac. J. **13**, 31-40 (2018)
17. B. F. Phillips, J. Kittaka, Spiny lobster: fisheries and culture (Osney Mead, Blackwell Science, 2000)
18. M. Junaidi, N. Diniarti, N. Cokrowati, A. Mukhlis, B. H. Astriana, F. D .L. Conway, Ilmu Kelautan: Indonesian Journal of Marine Science 27 4, 315-329 (2022)

19. J. F. Wickins D.O. C. Lee, Crustacean farming ranching and culture 2nd edition (United Kingdom, Blackwell Science Ltd, 2002)
20. Drengstig, A. Bergheim, Aquacult. Eng **53** 14-18 (2013)