Growth and survival of spiny lobster, *Panulirus homarus* fed fresh food and formulated diet

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Abstract. To date, lobster aquaculture relies on fresh food for feeding; however, the availability of fresh food is limited and its excessive use causes environmental degradation. Therefore, formulated diet is required to establish sustainable lobster aquaculture. Concerning this issue, this study was carried out to assess the growth and survival of spiny lobster *Panulirus homarus* fed fresh food and formulated diet. The study was conducted in a completely randomized design (CRD) with 3 groups of treatments and 3 replicates for each treatment. Feeding experiment was performed in a flow-through water system using 9 fibre tanks, 4000 L in volume. Each tank was stocked with 40 lobsters, with initial weight of 78.08 ± 0.22 g. Each group of lobsters was fed with fresh food (A), formulated diet (pellet) (B) and a combination of pellet and fresh food (C). The fresh food was a mixture of fish, crabs, shrimp and small mussel (3:1:1:1). Feeding experiment was done for 15 weeks. Results of the experiment showed that the highest specific growth rate of lobster was obtained from lobsters fed with the combination of pellet and fresh food (0.52 ± 0.02 %/day) and the lowest growth was resulted from lobsters fed formulated diet (0.16 ± 0.03 %/day). In contrast, the highest survival was achieved in lobsters fed formulated diet (51.67%). Whereas the lowest survival was found in lobsters fed fresh food (10%). This study indicated that good growth of lobster was resulted from feeding with formulated diet combined with fresh food, while good survival was supported by feeding with formulated diet.

1. Introduction

Lobsters are highly desired marine products in both domestic and international markets. It is not only as an important fishery commodity, but also as a symbol of a prestigious for consumers. Lobster species that commonly found in Indonesian waters are *Panulirus homarus, P. longipes, P. penicilatus, P. polyphagus, P. versicolor* and *P. ornatus*. The high market demand, and price of lobster which is around IDR. 300,000 to IDR. 1,000,000 (depending on species and size) lead to increased exploitation [1].

An effort to meet the high market demand of lobster is through aquaculture industries, though aquaculture of lobsters is still in its infancy. Supply of seed for grow-out of lobster yet depends on wild capture. In addition, feeding for lobster relies on fresh fishery product, commonly bycatch. A study stated that lobsters prefer fresh food to frozen food. However, the availability and nutritional values of fresh fishery product varies and its excessive use causes environmental pollution [2].

Therefore, development of formulated diet containing proper nutritional values for lobster aquaculture is necessary [3]. Research on the development of formulated diet for lobsters had been...
carried out for more than 30 years. However, formulated diet has low performance in promoting growth of lobster. This is the main challenge in developing a commercial diet for lobster aquaculture [4]. A previous study suggested to include fresh fishery products in formulating diet to increase feed intake for better growth and survival of lobster [5]. Presumably that high feed intake could be achieved as well by feeding lobsters with a combination of formulated diet (pellet) and fresh food such as fish, shellfish, shrimp and crabs. Therefore, this study was performed to determine the growth and survival of lobster *P. homarus* fed formulated diet and fresh food.

2. Materials and methods

2.1. Experimental diet preparation

The ingredients of the formulated diet are presented in table 1. All the ingredients were mixed thoroughly, and added with enough water to make a dough. The dough was extruded through a pelletizing machine to make a 1-cm-long pellet with a diameter of 3 mm. The pellets were dried in an oven at 70°C and subsequently stored in a refrigerator for the feeding experiment.

| 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. Feeding experiment

Lobsters used in this study were obtained from the catch of fishermen in Jembrana, southern Bali. The lobsters were transported to the lobster research facility of Institute for Mariculture Research and Fisheries Extension (IMRAFE) Gondol-Bali by dry method. Lobsters were acclimatized to the diet and experimental conditions for two weeks. Feeding experiment was carried out from April to August 2020. The experiment used 9 fiber tanks, 4000 L in volume. The study was designed using a completely randomized design (CRD) with 3 treatments and 3 replicates for each treatment. Each tank was stocked with 40 lobsters with an initial weight of 78.08 ± 0.22 g. The experiment was done in a running water system, each tank was equipped with aeration as oxygen supply and shelters for hiding. The treatments tested in this study were differences in feeding composition, namely fresh food (A), pellet (B) and a combination of fresh food and pellet (C). The fresh food was a mixture of fish, crabs, shrimp and small mussel at a ratio of 3:1:1:1. Feeding was done two times per day with a pellet dose of 5% of biomass/day, while fresh food was 10% of biomass/day.
2.3. Data collection and analysis

Parameters observed in this study were growth performance which included weight gain and specific growth rate, survival, total haemocyte count (THC), and BRIX index which represent serum protein of lobster’s haemolymph. Weight and carapace length of lobsters were measured every 3 weeks. Measurements of THC and BRIX index were carried out at the end of the experiment. Water quality analysis included temperature, dissolved oxygen, pH, salinity, ammonia, nitrite and phosphate. The weight gain, specific growth rate and survival were calculated as follow:

- Weight gain (WG) (%): \(100 \times \frac{(W_t - W_o)}{W_o}\)
- Specific growth rate (SGR) (%/day): \(100 \times \frac{\ln W_t - \ln W_o}{t}\)
- Survival rate (SR) (%): \(100 \times \frac{N_t}{N_o}\).

Where \(W_t\) and \(W_o\) are the lobster’s weights at the final and the initial of the feeding experiment, \(t\) is the experimental period (days), and \(N_t\) and \(N_o\) are the number of lobsters at the final and initial of the experiment. Analysis of proximate was performed to determine crude protein, lipid, water and ash content of the experimental diets by standard methods [6]. Data were reported in tables and figures and analyzed by analysis of variance (ANOVA) and followed by Duncan Test at 95% confidence intervals by SPSS 14.0 program.

3. Results and discussion

3.1. Growth of lobster

Different feeding in this study resulted in different growth of lobster among group of treatments. At the end of the experiment, pellet resulted in the lowest final weight (92.81 ± 5.38 g), whereas fresh food combined with pellet resulted in the highest final weight (136.03 ± 6.39 g). The difference in body weight was seen since the 9th week of rearing. Differences in growth of lobsters fed with different compositions of diet were not only seen in body weight, but also in carapace length (table 2). The combination of pellet and fresh food resulted in a higher carapace length than the fresh food as well as pellet only. At the end of the study, carapace length of lobsters fed fresh food, pellet and the combined diet were 6.58 ± 0.41 cm, 6.07 ± 1.17 cm; and 6.89 ± 0.10 cm, respectively. These results suggested that the differences in types of feed in this study resulted in the same growth pattern between the weight gain and the increase in carapace length of the lobsters.

The results of this study showed that the differences in feeding composition for lobster \(P.\ homarus\) had a significant effect on growth (p < 0.05). The weight gain of lobsters fed the combination of pellet and fresh food (73.85 ± 4.77 %) was significantly higher than that of lobsters fed pellet (19.20 ± 4.87 %), but was not significantly different from that of lobsters fed fresh food (56.11 ± 30.30 %). Effects of different feeding composition on the specific growth rate was similar to its effects on the weight gain. Feeding with pellet resulted in low growth (table 2).

| Table 2. Growth of lobster Panulirus homarus fed fresh food and formulated diet (pellet) |
|-----------------------------------------------|-----------------------------------------------|-----------------------------------------------|
|                                               | A (fresh food)                                | B (pellet)                                    | C (fresh food + pellet)                        |
| Initial weight (g)                            | 78.20 ± 0.54                                  | 77.82 ± 1.41                                  | 78.23 ± 2.12                                  |
| Final weight (g)                              | 122 ± 23\(^{ab}\)                            | 92.81 ± 5.38\(^{a}\)                         | 136.03 ± 6.39\(^{b}\)                        |
| Weight gain (%)                               | 56.11 ± 30.30\(^{ab}\)                       | 19.20 ± 4.87\(^{a}\)                         | 73.85 ± 4.77\(^{b}\)                         |
| Specific growth rate (%/day)                  | 0.41 ± 0.18\(^{ab}\)                         | 0.16 ± 0.03\(^{a}\)                         | 0.52 ± 0.02\(^{b}\)                         |
| Initial carapace length (cm)                  | 5.58 ± 0.09                                  | 5.64 ± 0.02                                  | 5.65 ± 0.06                                  |
| Final carapace length (cm)                    | 6.58 ± 0.41\(^{ab}\)                         | 6.07 ± 1.17\(^{a}\)                         | 6.89 ± 0.10\(^{b}\)                         |
| Carapace length increment (cm)                | 1.00 ± 0.50\(^{ab}\)                         | 0.43 ± 0.05\(^{a}\)                         | 1.24 ± 0.05\(^{b}\)                         |

The differences in growth found in this study could be attributed to the differences in the number of moulting occurrences in each group of treatment. Feeding lobsters with fresh food and the combination of fresh food and pellet resulted in a much higher number of moulting than feeding with
pellet only (table 3). Moulting occurrences in the fresh food feeding reached to 62, in the combined feeding achieved to 56, and the lowest was in the pellet feeding which only reached to 21. The low moulting occurrences in the pellet feeding caused in lower growth compared to the other treatments. It was stated that lobster growth occurs through moulting process [7].

**Table 3.** Molting occurrences in rearing of lobster *Panulirus homarus* fed fresh food and pellet

| Replicates | Treatments          | A (fresh food) | B (pellet) | C (fresh food + pellet) |
|------------|---------------------|----------------|------------|--------------------------|
| 1          |                     | 69             | 27         | 60                       |
| 2          |                     | 53             | 10         | 53                       |
| 3          |                     | 64             | 26         | 56                       |
| Average    |                     | 62             | 21         | 56                       |

The difference in growth and the number of molting in each treatment group was likely due to the different amount of feed consumed and the different nutritional values of the feed. Observation during the feeding experiment showed that lobster fed pellet had a low response to feeding. Meanwhile, lobsters fed fresh food had a high response to feeding.

Fresh food which consisted of a mixture of fish, small mussel, crabs and shrimp resulted in a higher number of molting occurrences. Although the number of molting occurrences in the fresh food treatment was higher than in the combined feeding, the growth was seen to be lower. This was probably because of the richness and better nutrients balance in the combination of pellet and fresh food which resulted in better growth. The pellet contains complex ingredients which provided protein, lipid, carbohydrate, astaxanthin, cholesterol, as well as vitamin and mineral premix (table 1). The crude protein and lipid of the pellet were 59.03 % and 15.68 %, respectively (table 4). The crude protein of the pellet was higher than that of crabs, but, lower than that of fish, shrimp, and small mussel. In terms of lipid content, pellet contains the highest crude lipid, whereas crabs had the lowest crude lipid content. Fish, shrimp and small mussel indicated a relatively high lipid content.

**Table 4.** Proximate compositions of the experimental diets

|                 | Pellet | Fish | Shrimp | Crabs | Small mussel |
|-----------------|--------|------|--------|-------|--------------|
| Protein (%)     | 59.03  | 69.73| 72.51  | 50.98 | 70.94        |
| Lipid (%)       | 15.68  | 10.58| 7.07   | 3.14  | 7.88         |
| Ash (%)         | 15.41  | 17.87| 11.64  | 35.52 | 13.50        |

The results of this study showed that feeding with pellet resulted in the lowest growth. The similar feeding response was also reported in juvenile lobster *P. argus* fed fresh squid which had a significantly higher specific growth rate than they fed pellet. The final weight of lobsters was four times higher than those fed dry pellets [8]. Growth of lobster *Thenus australiensis* fed fresh shellfish was two times higher than they fed pellet. The high growth was also reflected by high feed intake in lobsters fed fresh shellfish. These data indicated that low feed intake was the main cause of low growth in lobsters fed pellet [9]. A previous study also reported that *P. homarus* fed pellet experienced difficulties in metabolic and nutrient distribution [10]. This was likely due to the hepatopancreas of lobsters fed pellet showed vacuolation which caused by hypertrophy of B cell and remarkable thickenings of the lumen wall of the hepatopancreas [10].

3.2. **Survival**

Survival of lobsters from the start to the 3rd week of experiment was high, but then decreased until the end of the study. At the end of the study, the highest survival was obtained from feeding with pellet.
that achieved to 51.67%, followed by feeding with the combination of pellet and fresh food at 33.33%, and the lowest survival was obtained from feeding with fresh food which only 10% (figure 1). Feeding with formulated diet resulted in the highest survival rate because of the formulated diet provided a better balance of nutrients compared to fresh food [3]. The low survival in the fresh food treatment was possibly due to the imbalance of nutrients and food crumbs which decreased water quality and lowered the lobster's immune system as indicated by the decrease in the total haemocyte count (THC) (table 5). In addition, some newly molt lobsters were observed, but they subsequently died. The results of this study are in line with a previous study that juvenile *P. homarus* fed trash fish had higher growth than those fed pellet, and the lowest survival rate was found in lobsters fed trash fish [11].

![Figure 1. Average survival (%) of lobster *Panulirus homarus* fed fresh food and pellet](image)

### 3.3. Total haemocyte count (THC) and BRIX index

Data on total haemocyte count and BRIX index at the end of the study showed no differences among feeding treatments (table 5). Although it was not statistically different, feeding lobsters with pellet showed a high THC (105.66 x 10^5/ml), followed by the combination of pellet and fresh food (102.63 x 10^5/ml), and fresh food (101.78 x 10^5/ml). High THC was stated to be associated with high levels of lobster's immunity [12]. Thus, it could be stated that the high THC in lobsters fed pellet was thought to play a role in increasing survival of lobsters. Meanwhile, the low survival in lobsters fed fresh food might be related to the low THC values. THC is an indicator of lobster's health conditions. High THC is consistent with good health. Low nutritional and health conditions are consistent with high mortality rates [13]. Low THC indicates that lobsters are under immune pressure or stress conditions [14] and increases susceptibility to pathogens [15].

BRIX index of lobsters fed fresh food, pellet and the combination of pellet and fresh food were 14.80%; 14.32% and 17.56%, respectively (table 5). The high BRIX index in lobsters fed the combination of pellet and fresh food was assumed to be due to better feed intake and nutrient intake than that of lobsters fed only pellet. Brix index was argued to be correlated with nutritional conditions such as serum protein concentration in lobster’s haemolymph. Lobster *Homarus americanus* fed twice a week had a serum protein concentration of 2.7 times higher than they fed once a month or with limited access to natural food [16]. Lobsters fed with high frequency of feeding showed high feed intake which lead to the high serum protein levels. Although serum protein levels can be used as an indicator of lobster nutrition and quality, careful interpretation of the results is needed because serum protein levels are affected by the molting cycle [16].
Table 5. Total haemocyte count (THC) and BRIX index of lobster *Panulirus homarus* fed fresh food and pellet

| Treatments | THC (x $10^5$ / ml) | BRIX index (%) |
|------------|---------------------|----------------|
| A (fresh food) | 101.78±5.35$^a$ | 14.80±2.86$^a$ |
| B (pellet) | 105.66±5.50$^a$ | 14.32±1.00$^a$ |
| C (fresh food + pellet) | 102.63±36.32$^a$ | 17.56±2.13$^a$ |

Healthy and strong lobsters generally have high serum protein levels, while weak lobsters generally have low serum protein levels [16]. Healthy lobsters with high BRIX values promote the success of molting and increase growth of lobsters as observed in the combined feeding of pellet and fresh food. However, it should be noted that low serum protein does not necessarily mean nutritional deficiencies or unhealthy conditions. This is due to serum protein concentration increases during premolt, reaching a maximum just before molting, then dropping dramatically immediately after molting [16].

3.4. Water quality

Range of water quality which included ammonia, nitrite, phosphate, pH, dissolved oxygen (DO), temperature and salinity was similar between feeding treatments (table 6). This water quality range was not different from the water quality in the study of juvenile *P. ornatus* fed moist pellet, namely, nitrite <0.3 mg/L; ammonia <0.03 mg/L; pH, 7.8 – 8.3 and temperature of 29°C [17]. This range of water quality is classified as optimal for lobster aquaculture [17].

Table 6. Water quality in rearing of lobster *Panulirus homarus* fed fresh food and pellet

| Parameters          | Treatments            | A (fresh food) | B (pellet) | C (fresh food + pellet) |
|---------------------|-----------------------|----------------|------------|-------------------------|
| Ammonia, NH$_3$ (mg / L) | 0.0535 - 0.0687 | 0.0537 - 0.0880 | 0.0542 - 0.1204 |
| Nitrit, NO$_2$ (mg / L) | <0.0087 | <0.0087 | <0.0087 |
| Phosphate (mg / L) | 0.0120 - 0.0441 | 0.0113 - 0.0426 | 0.0100 - 0.0616 |
| pH                  | 7.92 - 8.20          | 7.90 - 8.21     | 7.93 - 8.10 |
| Dissolved oxygen (mg /L) | 5.01 - 5.87 | 4.97 - 5.87 | 5.07 - 5.71 |
| Temperature (°C)    | 28.6 - 29.2          | 28.6 - 29.1     | 28.7 - 29.2 |
| Salinity            | 30 - 31              | 30 - 31         | 30 - 31     |

4. Conclusion

This study found that the highest growth of lobster was obtained from lobsters fed the combination of pellet and fresh food, and the lowest growth was observed in lobsters fed pellet. On the other hand, the highest survival was found in lobsters fed pellet. While the low survival was obtained from lobsters fed fresh food.

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