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## Effect of Protein Level on Growth Performance of Abalone *Haliotis Squamata*

Zainuddin<sup>a\*</sup>, Siti Aslamyah<sup>b</sup>, Hamzah<sup>c</sup>

<sup>a,b</sup>Faculty of Marine Science and Fisheries, Hasanuddin University, Makassar 90245, Indonesia

<sup>c</sup>Center for Brackish Water Aquaculture Takalar, Indonesia

<sup>a</sup>Email: [zainuddinlatief@gmail.com](mailto:zainuddinlatief@gmail.com)

### Abstract

The effort to find alternative artificial feed for abalone is important to replace the natural feed of seaweed; *Gracillaria* sp., in order that the quantity and quality of abalone feed does not depend on the season. The aim of this study was to evaluate the level of protein in the feed on growth performance of *H. squamata* abalone, through feeding with different levels of protein; 52%, 43%, 32%, and 22% for three months period. Feed ingredients such as tempe powder, fish meal, shrimp head flour, seaweed extract of *Gracillaria* sp. This study was conducted using 240 abalones. The size of abalones ranging from 40.5mm to 45.3mm with an average weight of 12.98 g. The 22% and 43% of protein levels had the same specific weight growth values but differed significantly from other protein levels. Protein levels did not show significant differences in shell length growth and survival rate of abalone.

**Keywords:** abalone; feed; growth; level; protein.

### 1. Introduction

Abalone is one type of mollusca/gastropod that has the potential to be developed. This is due to a tendency to decrease production naturally [1]. Currently abalone is the type of mollusca that began to develop to meet the needs of alternative proteins other than fish. Abalone has high nutritional value with protein content of 54.13%, fat 3.20%, fiber 5.60%, ash 9.11% and moisture content 27.96% [2].

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\* Corresponding author.

In the development of abalone, cannot be separated from various problems. One of the problems faced in the development of abalone is the slow growth [3]. Abalone growth is very slow; not only caused by genetic factor but also caused by the lack of nutrient intake; due to so far it still uses natural food in the form of seaweed. Feeding in the form of seaweed for abalone has weaknesses such as high feed conversion value, besides the amount and quality depends on the season. Efforts to provide artificial feeding in development of abalone in Indonesia are still rare. Several countries such as Japan, Taiwan, the United States, and Australia, artificial feeding for development of abalone has been used. The results of experiments on artificial feeding in Taiwan showed a high rate of abalone growth; 65% faster than abalone that fed with macro algae [4].

The advantages of using artificial feeds other than their nutritional content, this artificial feeds can be prepared based on abalone needs and developments, as well as can also be prepared at any time. Artificial feeding with certain protein values and sources is expected to produce good quality abalone in the form of larger body weights, shell length, relatively high protein content and better taste [5].

Protein in artificial feed is one of the important nutrients because it functions as a building agent and maintains muscle mass. According to Najib [6], the high and low percentage values of the body weight gain is strongly influenced by protein content in a feed, the low amount of protein in the feed will cause slow growth and low weight gain. In a study conducted by Cho and his colleagues [7], feed with 37.6% protein content for *Haliotis discus-hannai* abalone was more efficient in growth than the others.

Although there have been many studies that reveal artificial feeding for abalone, however, so far the optimal level of artificial feed proteins for the growth of *H. squamata* abalone has not been widely revealed. Based on this issue, it is necessary to evaluate feed protein level with growth performance and survival rate of *H. squamata* abalone, through feeding with certain protein content.

## **2. Materials and Methods**

### ***2.1. Feed formulation and preparation***

The test feed used in this study had different protein content depending on the treatment to be tested with the protein content of 52%, 43%, 32% and 22%, respectively. The test feed used in the form of spots with the raw material composition as shown in Table 1 and the feed proximate results are presented in Table 2. The process for making feeds was firstly by grinding all the dry ingredients used. All materials were weighed on the basis of requirements and placed in a plastic bag.

All dried feed ingredients were mixed starting from a small amount of material followed by large amounts of raw material, and then mixing until well blended. All mixed materials were further added with fish oil, vitamins and minerals into the mixture of dry matter. After mixing evenly and then added with warm water into the mixture of feed ingredients until the form of dough.

The feed mixture was mixed until it is not sticky in the hand and was shaped into a fleck/sheet feed. Afterward, the sheet feed were dried to the moisture content at about 10%.

**Table 1:** Composition of the materials at each treatment

Material	Composition (%)			
	A (52%)	B (43%)	C (32%)	D (22%)
Tempe powder	80.00	55.00	12.00	15.00
Fish meal	2.00	10.00	40.00	16.00
shrimp head flour	5.00	10.00	22.00	15.00
Extract of <i>E. cottoni</i>	5.00	17.00	18.00	46.00
Agar flour	1.50	1.50	1.50	1.50
Jelly flour	3.00	3.00	3.00	3.00
Fish oil	1.00	1.00	1.00	1.00
Mineral	1.00	1.00	1.00	1.00
Vitamin	1.00	1.00	1.00	1.00
CMC	0.5	0.5	0.5	0.5
Total	100	100	100	100

**Table 2:** Proximate analysis of the feed used in the study

Feed	Proximate analysis (%)		
	Protein	Fat	Moisture
A	52.16	16.64	7.53
B	43.13	14.73	9.76
C	32.34	4.96	6.77
D	22.16	6.55	7.29

Description: The feed was analyzed in dry weight

## 2.2. Feed trials and design of study

A total of 240 *H. Squamata* abalone was used in this study, with shell length was  $43.28 \pm 1.67$  mm and  $12.98 \pm 1.34$  g of weight. Every 20 abalones were kept in a plastic basket container with size of 60x50x30 cm. All plastic basket containers (12) were put into a 500 L rectangular fiber tub with a circulation system.

The abalones were kept for three months and fed once a day at about 5% of the abalone biomass weight. This study was arranged using completely randomized design (CRD), with four feed protein level treatments with 3 replications.

The treatments consisted of A: 52% protein in the feed; B: 43%; C: 32%; and D: 22%, respectively.

## 2.3. Parameters and Data Analyses

The parameters observed in this study included growth of shell length and the weight of abalones.

Observations and calculations of daily growth rates were performed at the beginning and end of the study with reference to the Corzani and Illanes equations [8].

#### Specific Growth Rate

$$SGR = \frac{W_t - W_0}{t} \times 100\%$$

whereas: SGR = Specific growth rate (%)

$W_0$  = Weight at the beginning of the study (g)

$W_t$  = Weight at the end of the study (g)

t = Study period

#### Specific Shell Length Growth Rate

$$L_s = \frac{L_t - L_0}{t} \times 100\%$$

whereas:  $L_s$  = Specific shell length growth rate (%)

$L_0$  = Shell length at the beginning of the study (mm)

$L_t$  = Shell length at the end of the study (mm)

t = Study period

Survival rate was conducted at the end of the study with refer to the Effendie [9] equation.

$$SR = \frac{N_t}{N_0} \times 100\%$$

whereas: SR = Survival rate

$N_t$  = Number of abalones at the end of the study (ind)

$N_0$  = Number of abalones at the beginning of the study (ind).

Statistical analysis was performed using the statistical package SPSS version 22 for windows. The differences of the growth rate and survival rate of abalones were analyzed by using ANOVA.

### 3. Results

The growth of abalones during the study are shown in Table 3.

**Table 3:** The growth rate of shell length and specific weight of abalones

Protein level of the feed	Growth rate	
	Specific shell length (%)	Specific weight (%)
A (52%)	4.43±0.67 <sup>a</sup>	1.62±0.37 <sup>a</sup>
B (43%)	7.10±0.65 <sup>a</sup>	5.65±0.51 <sup>b</sup>
C (32%)	5.10±3.14 <sup>a</sup>	2.34±1.69 <sup>a</sup>
D (22%)	7.33±1.89 <sup>a</sup>	5.90±1.08 <sup>b</sup>

Description: Similar superscript letter indicates no significant different

Table 3 shows that feed protein level treatment did not show any significant difference ( $P>0.05$ ) on the growth of abalone specific shell length. Meanwhile, feed protein level treatment differed significantly ( $P<0.05$ ) to the growth of specific abalone weight. Treatment of dietary protein level of 32% and 52% was significantly differed ( $P<0.05$ ) with 22% and 43% protein level treatments. Meanwhile, dietary protein levels between 32% and 52% and between 22% and 43% did not showing any significant different ( $P>0.05$ ).

Based on the growth of shell length and weight rates of abalones showed that feed treatment with protein content of 22% had higher than the other treatments. These results indicated that the protein content of 22% prepared from protein sources of tempe flour, fish meal, and shrimp flour was categorized more balanced and the protein content was closely to abalone natural feed of *Gracillariaverucosa* with protein level of 8.0651%.

Survival rate of abalones during study period are shown in Table 4.

**Table 4:** Survival rate of abalones during study period

Protein level of the feed	Survival rate (%)
A (52%)	85.00±0.00 <sup>a</sup>
B (43%)	83.33±2.89 <sup>a</sup>
C (32%)	84.67±2.77 <sup>a</sup>
D (22%)	85.00±0.00 <sup>a</sup>

Description: Similar superscript letter indicates no significant different

Table 4 shows that different protein level treatment did not showing any significant difference ( $P>0.05$ ) to the survival rate of abalones.

#### 4. Discussion

This study shows that dietary protein level difference did not differ significantly to abalone shell growth. Meanwhile, for abalone weight growth showed a significant difference ( $P<0.05$ ). Feed with 22% and 43% protein levels had significantly higher weight growth compared to the feed with 32% and 52% protein levels. These results show that feeds with a balanced composition of protein sources between animal protein and vegetable protein are better than feeds that are composed of only one protein source. Feed treatment with 22% protein level had effect on higher specific weight growth with value of  $5.90 \pm 1.08\%$ . Thus, based on the results of this study it can be concluded that the best feed for growth of *H. squamata* abalone weight is at 22% protein level. In addition, with feeding at 22% protein level will be more efficient than feed with higher protein levels. Similar study was shown on Pacu *Piaractusmesopotamicus* fish with artificial feed with 22%-25% protein level, the result did not significantly affect the growth of Pacufish [10]. Different results, however, were shown in studies of salmon fed with a comparison of protein and fat, where salmon fed with low protein levels of 34% and 36% fat showed significant differences with feeding with high protein levels of 39 % and fat 32% [11].

The mollusca shell composition comprises of 95 - 99% calcium carbonate and the other 5% comprises of organic materials [12]. In this studied it was found that increased protein feed levels did not improve the growth performance of *H. squamata* abalone shells, different results were found in silver spiny fish (*Puntius gonionotus*) in which artificial feeding with protein levels of 20%, 25%, 30%, 35% and 40%, showed the results that the level of 30% had more effective feed growth performance compared with other feed [13].

The results of this study with feeding at 22% protein level had better specific weight growth;  $5.90 \pm 1.08\%$  during three months period. Similar results were also found by Cho and his colleagues [7], that the growth of *H. discus-hannai* abalone fed with 37.6% protein level had better when compared with 38.3% protein level feed; 36.5%; 36.8%; and 10.5%. The composition of feed ingredients for protein level 37.6% ie fish meal (20%), soybean meal (20%), shrimp head meal (13%), dextrin (13.6%), wheat flour (5%), 4%), sodium alginate (22%), vitamins (2%), and minerals (4%). However, a slightly different result is shown in the silverside mexican fish (*Menidianestor*) where in the feeding treatment at protein level of 25%, 30%, 35%, 45%, 50%, and 55%, the best growth and survival were at the level protein between 40% and 50% [14].

The results of this study indicate that survival rate of abalones at 22%, 32%, 43% and 52% protein levels did not differed although the amount of feed proteins consumed was different. This means that abalones can survive with artificial feeding even if their protein levels are low. Thus in development of abalone, the feed can be substituted from natural feed of *Gracillariasp* to artificial feed at certain protein level. Coote and his colleagues [15] suggested that an increase in dietary protein up to 27% level can maximize the growth of *H. leveigata* abalone. Likewise, as stated by Teruel and his colleagues [16] who obtained good growth in abalone *H. asinina*

fed artificially with protein sources derived from animal protein and vegetable protein.

This result also shows that feed with 22% protein level is closer to abalone feed protein content of *Gracillariasp* as stated by Nurfajrie and his colleagues [17], the result of proximate analysis of various types of seaweed, the highest is *Gracillariaverucosa* with protein value of 8.07 %, *Ulvasp* of 6.44%, *Gracillariaarcuata* 7.07% and the lowest on seaweed *E. spinosum* is 4.13%.

The balance of nutrient components of feed such as protein, carbohydrate, and fat should also be considered in artificial feeding for abalone as reported by Mohanta and his colleagues [18], feeding for silver spines at 25% protein and 34% carbohydrate showed high results, however the protein can be in the range of 25% -30% and carbohydrates in the range of 26% -34%.

## 5. Conclusions

Based on the results of this study, it can be concluded that the artificial feeding with 22% of protein level had more efficient for maintaining the abalone. This suggested that feed protein content of 22% is acceptable to support the growth and abalone performance.

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