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Impact of Iso-Protein Feed with Different Carbohydrate and Lipid Ratios on Growth and Feed Efficiency of the Mud Crab, *Scylla serrata*

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ABSTRACT

The use of artificial feed in mud crab aquaculture continues to be developed considering that feed has an important role in growth, survival, and production costs. The largest production cost comes from feed, especially feed protein. The utilization of carbohydrates and lipids as feed protein savers in mud crab aquaculture has not been widely studied. This study aimed to analyze iso-protein feed with different carbohydrate and lipid ratios on molting, growth, and feed efficiency of mud crab, *Scylla serrata*. Mud crabs with a weight of 70± 4.7g/ head and a carapace length of 5± 0.5cm were reared using crab boxes and placed in ponds for 60 days. *Pterygoplichthys pardalis* meal and maggot meal were used as feed protein sources. The treatments in this study were 35% iso-protein feeds with different carbohydrate and lipid ratios: treatments A (31 and 10%), B (33 and 9%), C (34 and 7%), and D (35 and 6%). This study used a completely randomized design consisting of four treatments and three replicates. The parameters observed were molting percentage (MP); growth (absolute weight growth "WG" and specific growth rate "SGR"); feed efficiency (protein retention "PR" & lipid retention "LR", feed conversion ratio "FCR", and survival "SR") in addition to the physical-chemical parameters of the research media. Data were analyzed using a completely randomized design, while water physico-chemistry was analyzed descriptively. The results showed that 35% iso-protein feed with different ratios of carbohydrates and lipids had no significant effect on molting, growth, and feed efficiency. However, there is a trend that the quality of crab meat with the carbohydrate and lipid ratio treatment of 31 and 10% is better than the other treatments. Similar research is still needed with more comprehensive studies and conducted in controlled media so that water quality does not experience extreme fluctuations.

INTRODUCTION

The mud crab, *Scylla* spp. is a fishery commodity from the crustacean group other than shrimp that has high nutritional value **(Islam *et al.*, 2022)** and specialty flavors, as well as high prices. Producers' demand for crabs is increasing for food needs **(Cerbule *et al.*, 2023)**. The demand for crabs is obtained from catches and aquaculture activities **(Keenan, 1999)**. Crab culture is a profitable activity if conducted with low production costs **(Apine *et al.*, 2023)**. Feed contributes more than half of the production costs **(Lovell *et al.*, 1993; Holme *et al.*, 2009; Hasnidar *et al.*, 2022)**. Feeding with an appropriate nutrient content can result in good growth and survival **(Chepkirui-Boit *et al.,* 2011)**. The nutritional content of the feed is always related to the materials used **(Manam, 2023)**.

Efforts to obtain cheap and quality feed ingredients are ongoing. Some of the feed ingredients currently developed as a source of feed protein are Amazon sailfin catfish, *P. pardalis* meal (**Castelnau, 1855; Cano-Salgado *et al*., 2022; Hasnidar *et al*., 2022)** and maggot meal **(Herawati *et al*., 2019; Herawati *et al*., 2020)**. Both ingredients are expected to replace the commercial fishmeal, which is increasingly expensive. *P. pardalis* meal has a protein content of 38.6 to 54.48% and fat content of 6.23 to 15.64% **(Panase *et al.*, 2018; Hasnidar *et al*., 2022)**. Its use as a source of feed protein has been reported by several researchers such as in juvenile Mekong giant catfish, *Pangasianodon gigas* **(Panase *et al*., 2018)**, catfish, *Clarias gariepinus* **(Mukhlis *et al*., 2020)** in addition to vaname shrimp, *Litopenaeus vannamei* **(Hasnidar *et al*., 2022)**. Apart from *P. pardalis* meal, maggot, *Chrysomya megacephala* meal can also be used as a protein source **(Siva Raman *et al*., 2022)**. Maggot is a fly larva *Hermetia illucens* or black soldier that has metamorphosed, with a protein content of 43.30 to 46.70% and a lipid of 7.00 to 14.35% **(Fasakin *et al*., 2003)**. Substitution of *P. pardalis* with maggot meal has a significant effect on the growth and feed efficiency of *Salmo salar* **(Belghit *et al*., 2019)**, Atlantic salmon **(Bruni *et al.,* 2020)**, *Clarias gariepinus* **(Fawole *et al*., 2020)**, milkfish, *Chanos chanos* **(Herawati *et al*., 2020)**, *Monopterus albus* **(Hu *et al*., 2020)**, *Oreochromis niloticus* **(Yildirim-Aksoy *et al*., 2020)** and *Micropterus salmoides* **(Peng *et al.,* 2021)**.

Mud crabs are carnivorous organisms that tend to require high-protein feed. The protein requirement of mud crabs is between 32 to 40% **(Catacutan, 2002)**, and 46.90 to 47.03% **(Unnikrishnan & Paulraj, 2010)**. Combining different protein sources, i.e. *P. pardalis* and maggot meal, is expected to complement each other, especially the amino acid content that is lacking. The amino acid profile present in a particular protein varies depending on its source **(Gorissen *et al.*, 2018; Day *et al*., 2022)** in addition to using cheap and quality protein sources for the efficient use of feed, it can also be done by reducing protein content and increasing the carbohydrate and lipid content of feed **(Kabir *et al.,* 2020)**.

Protein is the most expensive feed component compared to carbohydrates and lipids. According to **Suhenda and Samsudin (2008)**, lipids and carbohydrates can be a protein-sparing effect, which is a substitute for protein as an energy source, hence the use of energy from protein can be optimized for growth. The reduction of feed protein content also has an impact on reducing the discharge of nitrogen waste into the environment. According to **Zainuddin *et al*. (2014)**, nitrogen waste discharge is higher at 30% feed carbohydrate content compared to 49% content. Furthermore, increasing feed carbohydrates can increase body glycogen levels which can be used for other metabolic activities at any time. Conversely, high feed fat content will cause fat storage in the body, decreased food consumption and growth, as well as liver degeneration. According to **Catacutan (2002)**, mud crabs grow well when given feed with a lipid content of 6 or 12%.

The utilization of carbohydrate and fat as a sparing effect of feed protein use in crabs is not yet available. Information is still limited to the level of protein and lipid of jambal fish with a ratio of carbohydrate and lipid of 32 and 8% **(Suhenda & Samsudin, 2008)**, 55 and 7% in golden mandarin fish, *Siniperca scherzei* **(Sankian *et al*., 2018)**. Furthermore, in white shrimp, *Litopenaeus vannamei* the use of carbohydrates with a content of 38% resulted in better growth.

MATERIALS AND METHODS

The research was conducted in a community aquaculture area in Minasa Upa Village, Bontoa District, Maros Regency, South Sulawesi, Indonesia. The series of research consisted of preparation, i.e. preparing mud crabs, *Scylla serrata* (Forsskal, 1775) research media, research containers (crab boxes), preparing feed ingredients and making feed (January to February, 2023), conducting research (March to April, 2023) & testing feed chemistry and crabs (May, 2023). The stages of the research are as follows:

Preparation of *P. pardalis* and maggot meal

P. pardalis specimens were collected from Lake Tempe, Wajo Regency, South Sulawesi, Indonesia. The fish were cleaned and gutted & then dried for five days. To reduce moisture during drying, the fish wee then baked at 70°C for 24 hours **(Panase *et al*., 2018)**. The dried *P. pardalis* samples were then ground using a blender, and sieved using a 425µm mesh size sieve. Subsequently, the *P. pardalis* meal was ready for use. Maggots were obtained from a maggot farming in Makassar. Maggot was cleaned, and dried in the sun until dry. After drying, the maggot samples were put in an oven at 60^0C for 12 hours **(Alpionita, 2020)**, then mashed using a blender and sieved using a 425µm-sized sieve. Furthermore, maggot meal was ready for use.

Preparation of feed

Each feed ingredient used was weighed as needed. The ingredients and the amount needed for each treatment are presented in Table (1). The weighed feed ingredients were mixed evenly by stirring manually until the ingredients were homogeneous. Furthermore, small amounts of ingredients, such as fish oil, vitamin premix, and carboxy metil cellulose (CMC) were added and stirred again. After all the ingredients were evenly mixed, warm water was added to the mixture of feed ingredients until it formed a dough. The feed dough was stirred until it did not stick to the hands, then the dough was molded into pellets using a feed printer. The pelleted feed was then spread evenly on a tray and dried in the sun, then the feed was ready to be applied. The proximate test results of the test feed are presented in Table (2).

Table 1. Feed ingredients and quantities required for each treatment

Feed ingredient	Feed formulation (ratio of carbohydrate and lipid content of feed)			
	A (31 and 10)%	B (33 and 9)%	C (34 and 7)%	D (35 and 6)%
P. pardalis meal	30	30	30	30
Maggot meal	30	30	30	30
Soybean meal	24	24	24	24
Bran meal	5	5	5	5
Corn meal	0	2.5	5	7.5
Fish oil	7.5	5	2.5	0
Vitamin premix[1]	3	3	3	3
CMC[2]	0.5	0.5	0.5	0.5
Total	100	100	100	100

1) Vitamin premix (in one kg of diet): vit. A 90.000IU, vit. D3 30.000IU, vit. K3 36mg, vit. E 225mg, vit. B1 90mg, vit. B2 135mg, vit. B6 90mg, vit. B12 90mg, vit. C 240mg, calcium D-pentathenate 120mg, folic acid 45mg, biotin 300mg, inositol 375mg, nicotinamide 600mg, and cholin chloride 450mg.
2) Carboxymethylcellulose.

Table 2. Proximate results of the experimental diets in mud crabs, *S. serrata* (Forsskal, 1775)

	Feed formulation (ratio of carbohydrate and lipid)			
	A(31 and 10)%	B(33 and 9)%	C(34 and 7)%	D(35 and 6)%
Feed proximate test result				
Protein (%)	34.49	34.48	34.96	34.97
Carbohydrate (%)	30.77	33.07	33.55	35.29
Lipid (%)	9.94	8.87	7.30	6.31
Ash (%)	17.21	17.75	17.44	18.25
Water (%)	6.27	6.83	7.15	8.70

Description: Analysis results. Lab. polytechnic tester, Pangkep. 2023. Except moisture content, all fractions are expressed in dry matter. *Carbohydrate: extractable material without nitrogen (BETN) + crude fiber.

Mud crabs, *S. serrata* with a weight of 70± 4.7g/ crab and a carapace length of 5± 0.5cm, were obtained from fishermen collecting in the location of the study site. The culture container used was a crab box measuring length × width × height that was 20 × 15× 15cm as many as 60 pieces, and each crab box was filled with one crab. At the bottom and sides of the crab box, plastic nets with a mesh size of one inch were installed to prevent feed and feed residue from falling through the holes in the crab box.

Furthermore, crab boxes were placed in a row on a maintenance raft made of bamboo, with floats from the used plastic bottles. Water changes in the pond were made once a day. The feed had an iso-protein composition with varying ratios of carbohydrate and lipid, as detailed in Table (1) for each treatment. The crabs were fed 5% of the biomass weight per day, with a frequency of feeding of 2 per day at 7 am and 6 pm during 60 days of rearing.

Observations of the number of molted and dead crabs were made daily, and growth sampling was conducted every 15 days. Dead crabs were recorded and weighed. Crab weight gain was measured using digital scales (O-haus) with an accuracy of 0.1g, and the carapace width was measured using a sliding mister with an accuracy of 0.1mm. Measurement of physical and chemical properties of water, such as temperature, pH, salinity, dissolved oxygen, ammonia, and nitrite were measured throughout the experiment. Temperature, pH, and salinity were measured daily, while measurements for dissolved oxygen, ammonia, and nitrite were measured at the beginning, middle and end of the experiment.

Observations of crab body chemical composition were made on crabs before treatment (initial) and crabs at the end of the experiment. A total of five crabs were taken randomly for body composition analysis. Proximate analysis of feed and crabs was conducted based on the outlines of the AOAC **(Helrich, 1990)**. Dry matter content of samples was obtained by drying them in an oven at 105°C for 16 hours; crude protein was analyzed by micro-Kjeldahl, and fat was determined gravimetrically by chloroform methanol extraction. Crude fiber was analyzed by heating with alternating acid and alkaline washing, and ash by burning in a furnace at 550°C for 24 hours.

Parameters measured were molting (molting percentage "MP"), growth (weight gain "WG", and specific growth rate "SGR"), and feed efficiency (protein retention "PR", lipid retention "LR", feed conversion ratio "FCR", and survival rate "SR"). The formulation used the **Boonyaratpalin (1989)** equation as follows:

$$MP = \frac{number\ of\ molting\ crabs}{initial\ number\ of\ crabs} \times 100$$

$$WG = (final\ weight\ of\ crabs - initial\ weight\ of\ crabs)$$

$$SGR = \frac{Ln\ final\ weight - Ln\ initial\ weight}{cultivation\ time} \times 100$$

$$PR = \frac{increase\ in\ body\ protein\ weight}{feed\ protein\ weight} \times 100$$

$$LR = \frac{increase\ in\ body\ lipit\ weight}{feed\ lipit\ weight} \times 100$$

$$FCR = \frac{dry\ weight\ of\ feed\ given}{(final\ weight + dead\ crab\ of\ weight) - initial\ weight}$$

$$SR = \frac{Final\ number\ of\ crabs}{initial\ number\ of\ crabs} \times 100$$

Statistical analysis

The experimental design used was a completely randomized design, with four treatments and three replications. The treatments consisted of four feed compositions (Table 1). To determine the effect of treatment on the test parameters, an analysis of variance (ANOVA) was performed, followed by Duncan multiple range test (DMRT) at a significance level of 95% ($P< 0.05$). All data were presented as mean and standard deviation. Statistical analysis was performed using SPSS software version 17 for Windows (SPSS Inc., Chicago, USA). The physical-chemical properties of water were analyzed descriptively.

RESULTS

Mud crabs reared for 60 days with feed composition treatment having different carbohydrate and lipid ratios showed no significant effect on MP, WG, SGR, PR, LR, FCR, and SR (Table 3).

Table 3. Molting, growth, and feed utilization response of mud crab, *S. serrata* (Forsskal, 1775) with different carbohydrate and lipid ratios

Parameter	Feed with different carbohydrate and lipid ratios			
	A (31 and 10)%	B (33 and 9)%	C (34 and 7)%	D (35 and 6)%
MP (%)	33.33±11.55[a]	33.33±11.55[a]	26.67±11.55[a]	26.67±11.55[a]
WG (g)	158.18±2.94[a]	158.03±3.63[a]	155.25±2.65[a]	153.54±4.05[a]
SGR (%)	1.79±0.14[a]	1.78±0.05[a]	1.72±0.41[a]	1.71±0.04[a]
PR (%)	17.98±0.49[a]	16.74±0.89[a]	16.43±0.89[a]	16.31±0.38[a]
LR (%)	9.19±0.10[a]	8.97±0.06[a]	8.55±0.71[a]	8.65±0.49[a]
FCR	1.94±0.16[a]	2.03±0.14[a]	2.09±0.12[a]	2.10±1.09[a]
SR (%)	86.67±11.54[a]	80.00±0.00[a]	86.67±11.55[a]	73.33±11.55[a]

- MP (molting percentage), WG (weight gain), SGR (specific growth rate), PR (protein retention), LR (lipid retention), FCR (feed conversion ratio), SR (survival rate).
-The results of the analysis of variance showed that the treatments did not significantly affect (P> 0.05) the test parameters.
- Mean values followed by the same letter in the same row indicate not significantly different (P>0.01) based on Duncan multiple range test (DMRT)

Based on the proximate test results of crab meat before treatment (initial) and after treatment (final), the protein content of the crab increased, but the lipid and carbohydrate contents decreased (Table 4).

Table 4. Chemical/ proximate test results of mud crab meat, *Scylla serrata* (Forsskal, 1775) during initial and final research

	Feed with different carbohydrate and lipid ratios	Proximate composition		
		Protein	Carbohydrate	Lipid
Initial meat		45.98	24.06	2.88
Final meat	A (31 and 10)%	54.20	11.02	2.62
	B (33 and 9)%	52.27	21.43	2.50
	C (34 and 7)%	49.21	23.29	2.47
	D (35 and 6)%	49.17	20.80	2.36

The physicochemical properties of the rearing media during the study and the optimal value for mangrove crab cultivation are shown in Table (5).

Table 5. Physico- chemical variable, measurement result, optimal level, and measuring tools

Physico- chemical variable	Measurement result	Optimal level	Measuring
Temperature	10 to 30^0C	29^0C (Hastuti *et al*., 2019); 20 to 30^0C (Alberts-Hubatsch *et al*., 2016)	Thermometers
pH	6.0 to 8.0	7.75 to 8.50 (Hastuti *et al*., 2016)	pH meters
Salinity	5 to 15 ppt	25 ppt (Hastuti *et al*., 2015); 20 ppt ((Suyono, 2021)	Refractometers
Dissolved oxygen (DO)	3 to 5 mg/l	±5.51 mg/l (Faturrohman *et al*., 2017)	DO meter
Ammonia (NH_3)	0.10 to 0.30 mg/l	< 0.5 (Hastuti *et al*., 2019)	Spectrofotometer
Nitrite (NO_2^-)	0.00 to 0.002 mg/l	<0.05 (Hastuti *et al*., 2019)	Spectrofotometer

DISCUSSION

The results of this study showed that iso-protein diets with different carbohydrate and lipid ratios did not significantly affect the molting percentage, absolute growth, growth rate, protein retention, fat retention, feed conversion, and survival of mud crabs (Table 3). The chemical (proximate) test results of the four feed treatments contained 34.49 to 34.97% protein, 30.77 to 35.29% carbohydrate & 6.31 to 9.94% fat. The low range of feed carbohydrates and fat is thought to be the reason why the treatments did not affect the test parameters. The protein content of the feed in this study was lower than the needs of mangrove crabs according to **Catacutan (2002)** (32 to 40%), and **Unnikrishnan and Paulraj (2010)** (46.90 to 47.03%). Therefore, it is thought to cause molting, growth, and feed efficiency not to be optimal. Feed with the right ratio of carbohydrate and fat is expected to supply the energy needed by the crabs and also have a protein-sparing effect. According to **Furuichi and Yone (1982)**, carbohydrate requirements vary among species; carnivorous species require lower carbohydrates than herbivorous species. Crabs are carnivorous species, presumably less able to metabolize carbohydrates to produce energy that can support molting and better crab growth. According to **Mente (2006)**, protein functions as a source of energy for growth and tissue repair. Body protein is always in a dynamic balance, and protein requirements vary greatly according to age and species **(Radhakrishnan *et al*., 2020)**.

The carbohydrate content of the feed in this study was 30.77 to 35.29%, which is higher than the crustacean requirement level, which is only about 20 to 30% in *Penaeus monodon* (Fabricius) **(Alava & Pascual, 1987)**, *Scylla serrata* **(Truong *et al*., 2008)**, *Litopenaeus vannamei* **(Wang *et al*., 2014; Wang & Chen, 2016)**, and 18 to 20% in mud crab **(Zhang *et al*., 2022)**. The high carbohydrate content in this study aimed to evaluate the optimal carbohydrate requirement to replace protein as a source of energy. The results showed that the treatment did not significantly affect the test parameters. Crab growth is a function of molting **(Tamsil & Hasnidar, 2018)**. The more often a crab molts, the faster it grows. The number of crabs molting in this study was 29.0 to 31.7%. Crabs are less able to metabolize carbohydrates properly, hence molting and growth are less than optimal. The same thing was also stated by **Zhang *et al*. (2022)** who determined that carbohydrate levels of 24.0 to 30.0% in feed negatively affected growth, prolonged the molting cycle, and affected crab health **(Li *et al*., 2013)**. The low number of molted crabs is also thought to be due to the relatively short research time of only 10 weeks. According to **Stevens and Jewett (2014)**, crabs begin physiological preparations for molting about 2 months before releasing their shells. Meanwhile, mangrove crabs grow rapidly through molting occurring every 5- 30 days, depending on the phase of development. Only a few studies have discussed the effect of feed quality on molting and crab growth. However, feed with quantity and quality below the optimum will negatively affect growth **(Nguyen *et al*., 2014)**.

Molting in crabs is a complex interaction between internal and external factors. The molting process consists of four stages, namely pre-molting, molting (ecdysis), post-molting, and inter-molting; the longest stage is the inter-molting. If the energy from food is sufficient for growth, the inter-molting phase will be faster, and vice versa. Maximum weight growth will stimulate the crab to begin the molting process. This process begins with the formation of a new carapace under the old carapace. When the old carapace is detached (ecdysis), the new carapace will replace it with a larger size. Therefore, the increase in crab size only occurs during molting. According to **Stevens and Jewett (2014)**, the increase in size of crab every time it molts is around 20 to 25%. Hence, the growth pattern of crabs (especially the order decapoda) is discontinuous. High molting activity is not only influenced by the quantity and quality of food, environmental conditions but also the durability of the food when it is held by the crab's claws to be inserted into its mouth.

Lipids in crustaceans are assimilated in the hepatopancreas, distributed to other tissues via hemolymph. Lipids function to produce energy for molting, limb regeneration and growth **(Tocher, 2003)**, as well as increase metabolism and respiration in mitochondria **(Yuan *et al*., 2019)**. Feed lipid levels in this study recorded 6.31 to 9.94%, falling within the levels that can support good crab growth, which is 2 to 10% **(Sheen & D'Abramo, 1991; Zhao *et al*., 2014)** or 2 to 12% of the dry weight of food **(Catacutan, 2002)**. The lipid requirements of organisms vary widely depending on species and environmental conditions **(Holme *et al*., 2007)**. **Bowser and Rosemark (1981)**, in their study, denoted that if lipids (phospholipids) are not fulfilled in the feed, it will cause molting syndrome (molting failure) resulting in death in lobster, *Homarus americanus*, as well as inhibiting the development of *Marsupenaeus japonicus* larvae **(Teshima *et al.*, 1986)**. Furthermore, the high feed lipid levels inhibit growth and reduce feed utilization, lipolysis, lipid deposition in the hepatopancreas **(Sun *et al*., 2020)**. Lipid metabolism in crabs still needs to be studied more deeply since there are many factors that affect it.

Protein retention is the amount of protein stored in the body and used to build new body tissues, repair damaged cells, and daily metabolic processes. The results showed that the treatment had no effect on protein retention. Factors that affect protein retention include size, water temperature, feed dosage, quantity and quality of feed, feed energy content and protein quality **(Suhenda *et al*., 2004; Jobling, 2012)**. Due to the low feed protein content compared to the crab's needs, it is suspected that the energy for growth is also reduced.

Lipid retention is the amount of feed lipids that can be utilized and stored in the body during the rearing period. Similar to protein retention, the treatment did not significantly affect lipid retention. Lipids serve as a source of energy, build cell membranes, precursors of physiological components, including sex hormones , molting activity **(Sheen & Wu, 2003)**, reproduction and egg quality **(Islam & Yahya, 2016)**, as well as growth and survival **(Aaqillah-amr *et al*., 2021)**. The lipid content of the feed is appropriate for the crab's needs, but due to the low protein content of the feed, energy from lipids replaces protein energy to survive.

Feed conversion rate (FCR) is a description of the efficiency of feed utilization, the smaller the FCR value, the more efficient the feed utilization of an aquaculture organism. The FCR in this study was 1.94 to 2.10, which is lower than the FCR of mangrove crabs fed with raw fish, which were 7.20 to 21.5, according to **Hastuti *et al*. (2016)**, and 2.57 to 3.60 according to **Hudita *et al.* (2020)**. The difference in FCR value is thought to be due to differences in feed type, namely artificial feed (dry) and loose fish feed (wet), environmental factors and physiological conditions of the test animals. The use of artificial feed in crabs is still being researched in terms of quality, quantity, size, shape and texture of the feed. Crabs have unique eating habits, their claws function as hands to hold the food into the mouth. The feed given must have a compact texture and not easily crushed. The artificial feed in this study has a texture that is not optimal, easily crushed, which is thought to cause high FCR values.

The survival rate of crabs from this study was 73 to 87%, surpassing the findings of previous research by **Hastuti *et al*. (2016)** which is 0 to 83%, and exceeding those reported by **Yulianto *et al*. (2019)**, ranging from 63 to 80%. This difference is thought to be due to several factors, including treatment, internal and external factors of the test animals. The high survival rate of the results of this study is thought to be due to the feed provided produces enough energy to sustain the life of the crab. Environmental conditions, especially salinity, are at a less than the optimal level, but mangrove crabs are still able to survive well. Efforts to maintain the osmotic balance of body fluids and the environment (osmoregulation process) require high energy, which affects the low number of crabs molting and growth.

The results of the initial and post-study crab chemistry tests by feeding according to the treatment (Table 5) showed that the protein content of crab meat tended to increase after the study. Although the feed given did not significantly affect the test parameters, the meat protein content showed an increase. The treatment with a carbohydrate and lipid ratio of 31 and 10% has a higher protein content than the other treatments. This is thought to be ascribed to the fact that crabs are less able to metabolize carbohydrates compared to fats. Furthermore, the carbohydrate and lipid content of crab meat at the beginning and after the study tended to decrease. This may be related to the energy released from carbohydrates and lipids which is used to adapt to the non-optimal physical-chemical environment of water, especially salinity (Table 5). According to **Tseng and Hwang (2008)** and **Wang *et al*. (2012)**, carbohydrates can fulfill the high energy needs of aquatic animals during stressful conditions. Furthermore, **Wang *et al*. (2014)** stated that optimal carbohydrate levels can reduce the adverse effects of ammonia stress and low salinity on vanname shrimp.

CONCLUSION

In conclusion, iso-protein feed with different carbohydrate and lipid ratios did not significantly affect molting, growth, and feed efficiency of mud crabs. However, there is a trend that the treatment with carbohydrate and lipid ratios of 31 and 10% is better than the treatments of 33 and 9%, 34 and 7%, as well as 35 and 6%. Additional research is required to further explore this topic comprehensively, ideally conducted in controlled environments to minimize fluctuations in water quality.

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