

## Biochemical and texture property changes during molting process of tiger prawn, *Penaeus monodon*

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### Abstract

Texture and biochemical changes, specifically in moisture, protein content and amino acid profile were studied throughout the molt cycle of *Penaeus monodon*. This study was initiated to investigate changes that may occur in mass and tissue composition during different molting stages. Molting of *Penaeus monodon* are classified into 3 main stages; postmolt, intermolt and premolt. Result of biochemical analysis, shows that moisture varies from 78.02-80.88%, indicating a maximum value during postmolt and minimum value during premolt. Total protein content was found to be higher during intermolt (23.48%) and postmolt (22.27%) compared to premolt (23.10%). The result of SDS-PAGE electrophoresis shows a higher intensity of protein band during intermolt which corresponds to higher protein content. Thus, this electrophoresis method is useful as an alternative to determine different stages of shrimp molting. This is based on intensity of the protein band which is referring to protein content of the shrimp at its different stages. The amino acid profile showed that arginine, alanine, glutamic acid, glycine, lysine, and threonine increased during premolt whilst isoleucine and phenylalanine higher during postmolt. Aspartic acid and histidine was higher in intermolt than in premolt and postmolt. Texture analysis carried out give an elastic modulus value that is high during premolt compared to postmolt and intermolt. There are significant changes in texture and biochemical composition through elastic modulus value and protein composition of *Penaeus monodon* during each stages of molting cycle.

### Keywords

Texture  
 biochemical composition  
 molting  
*Penaeus Monodon*

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### Introduction

Growth and development of crustacean animal appear to be discontinuous processes associated with successive molts (Skov and Hartnoll, 2001). Crustacean's physiology, behaviour and reproduction is intrinsically linked to the molting cycle. Weight of the organisms in crustacean can be affected by molting process. (Xu *et al.*, 1993). Crustacean's molting process consisted of different stages including postmolt, (A-B stages), intermolt (C-Stage), premolt (D-Stage) and ecdysis (E-Stage). The cuticle of crustacean is still thin and water is taken up in the early stage (A-C), so that the larval body expands and rapidly develop its final size and shape. In late stages A-C, the epidermis shows a conspicuous tissue growth, while larvae reinforce the cuticle. During premolt stage (D), occurs a initiation in formation of new setae and appendages, and retraction of the epidermis from the cuticle (apolysis). Stage E is a very short process. There is a cuticular rupture between

the cephalothorax and the pleon, followed by a rapid retraction of the pleon from the old exoskeleton (Wang *et al.*, 2007).

Even though there are extensive studies in molting cycle of adult Decapoda and other crustaceans, it is still much less information for larval stages, mainly due to practical problems related to small body size, a thin and hardly structured integument, short moulting cycles, and restricted availability of materials with precisely known age within a moulting cycle (Anger, 2001). Drach and Tchernigovtzeff (1967) founded the criteria for characterization of different molting stages in crustaceans. The methodology used for molt staging generally can be carry out by observations of the degree of hardness of the exoskeleton and microscopic examination of the transparent edge of the uropods or pleopods (where epidermal with drawl and development of new setae can be observed) (Yamaoka and Scheer, 1970).

Molt cycle is of immense physiological importance; hence some study was initiated to evaluated changes

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that may occur in mass and tissue composition during different molting stages. Expression of results is a critical importance when comparing the changes in the biochemical composition of animals relative to various physiological phenomena (Read and Caulton, 1980). Study on biochemical changes in the species of crustacean during molting cycle is not conducted for overall major constituent. Success of molting cycle can be evaluated by measuring cyclical variations in metabolites such as glycogen in the hepatopancreas, and glucose and hemocyanin in the hemolymph. Difference in energy requirements throughout the molt cycle can be an indicator of variations in glucose concentrations. Hemocyanin was used as an indirect indicator of the energy consumed by shrimp in various molt processes that is related to the osmotic capacity. (Galindo *et al.*, 2009).

HPLC analysis of amino acids founded higher levels of essential amino acids, such as phenylalanine, leucine, tyrosine, isoleucine, tryptophan, methionine, valine, threonine, arginine, histidine, lysine in female prawns when compared to the male prawns. Furthermore, GC analysis of fatty acids showed both polyunsaturated and saturated fatty acids levels were revealed to be higher in female prawns when compared to the males (Bhavan *et al.*, 2010).

Studies founded that there are changes in the proximate chemical composition, fresh mass, water content, ash content, organic constituents, lipid and protein contents and energy levels of penaeid prawn, *Penaeus monodon* during different reproductive stages (Suneetha *et al.*, 2009). Hemolymph osmolality was low at post-molt stages A and B, and increased gradually until intermolt stage C0, reaching a maximum at stage D2 in the giant freshwater giant prawn, *Macrobrachium rosenbergii*. Levels were lowest at the ecdysis (E) stage (Wilder *et al.*, 2009). Lipids, in specific phospholipids, are shown to be the main important source of energy catabolised during the starvation period, which was greater than 62% reduction in total lipid during 30 days of starvations. Proteins were also catabolised during starvation, decreasing by 34% in total content 25 days of starvation that much lesser than lipid. (Limbourn *et al.*, 2008). Research shows a significant increase of copper and proteins levels in the hepatopancreas and hemocyanin concentration in haemolymph form postmolt to premolt stages. This suggests that copper is an crucial element in the development molting of crustaceans (Sreenivasa Rao *et al.*, 2008).

Knowledge of the biochemistry and texture changes during the molt cycle are essential for a complete understanding of molting cycle processes and indirectly can contribute vital information on

nutritional feed development and application. Hence, present study was conducted to find out the changes in the biochemical composition and its texture of penaeid prawn, *Penaeus monodon* during the molting process.

## Material and Method

### Collection of sample and molt stages determination

*Penaeus monodon* prawn taken from prawn tank reared at Lembaga Kemajuan Ikan Malaysia, Kuala Muda, Kedah. Size of prawn taken was range between 8-15cm. Method of molting cycle stage detection is by touching outer shell hardness, prawn behaviour observation and microscopic observation. End of light uropods or pleopods are cut and place onto slide and observe with microscope (10x magnification) Transparent appendage and developing satae (setogenesis) have been used as the observation criterion (Longmuir, 1983).

### Texture analysis

10 prawn sample from each postmolt, intermolt, and premolt stage was prepared. Prawns were cooked into 500mL of boil water for 4 minutes. Then, each sample cooled down in cool water at 22°C for 5 min. After cooking, shell of prawn taken off and the meat are cut into cube shape (1cm x 1cm). Each sample cut at same segment. Instron testing machine 1140 was used to compress the prawn until 75% of its length before. Crosshead pressure was at 100mm/min and chart pressure was at 400mm/min. 10kg load cell was used (Brauer *et al.*, 2003).

### Moisture analysis

Moisture analysis was carried out by drying method. 5 g of sample was place onto aluminium dish that been weight. Aluminium dish with the sample were then place in an oven with 105°C for 4 h. After drying the sample been cooled and weight. Drying steps repeated until constant reading (Heaton *et al.*, 1975).

$$\text{Moist weight} = \text{wet sample weight} - \text{dry sample weight}$$

$$\% \text{ moisture} = \frac{\text{wet weight} - \text{initial weight}}{\text{initial weight}} \times 100$$

### Protein analysis

Protein analysis carried out by Kjeldahl method. 2 g of duplicate prawn sample was place into digestion tube. 0.3 g of  $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$ , and 0.2 g of  $\text{CuSeO}_3$  catalyst were added and followed by 15 ml of sulphuric acid. Tubes were mixed. Blank tube was prepared only with 2 g sucrose with same catalyst and sulphuric acid. Those tubes were place in digestion block. Each tube was covered and water was left

to continuously flow. After 10 min, water flow was off slowly. Complete digestion can be indicated by clear mixture and no black spot formation in that tube. Tubes were cool off under running tap water. 250 ml boric acid was prepared as an indicator in conical flask. This conical flask place under distilled condenser unit with the end of the pipette reach boric acid. Digestion tubes were prepared in distilled unit. NaOH (60%) were slowly added and steam was supplied. Distilled end were collected for 125 ml and run for titration with 0.2N HCl (AOAC, 1980).

1ml 0.2N HCl = 2.8 mg Nitrogen  
 % nitrogen content in sample  

$$\%N = \frac{(\text{ml HCl titration} - \text{ml HCl blank}) (2.8) \times 100}{\text{sample weight (g)} \times 1000}$$

#### *Protein analysis: Electrophoresis SDS-PAGE*

##### **Sample and reagent preparation**

Same segment of prawn sample from postmolt, late postmolt, intermolt and premolt stages were prepared. Phast gel media were prepared; Stain, Destain 1, Destain 2, Destain 3, and Preserving solution. Buffer prepared from 10 mm Tris/HCl, 1mm EDTA with pH 8.0. 2.5%, SDS (Sodium Dodecyl Sulphate) and B-mercaptoethanol were added and followed by 0.01% Bromophenol blue as tracking dye.

##### **Extraction of protein**

One gram of prawn sample was taken for each prawn and place into test tube and labelled. Buffer solution was added to each tube and homogenise at 13,000rpm/min for 3 min. Sample was then centrifuge for 15-10 min at 200 rpm. Supernatant of the solution is diluted with buffer solution with ratio 1:6 (v/v) and prepared for electrophoresis.

##### **Electrophoresis**

Protein content for each extract was analysed by Phastsystem machine. Separation technique using SDS-PAGE (Sodium Dodecyl Sulphate-Polycrylamide Gel Electrophoresis) and PhastGel gradient 10-15 was coloured by coomassive blue. Low molecular weight protein and high molecular weight protein were used for analysis from Pharmacio. These proteins are phospholipase b (94000), albumin (67000), ovalbumin (43000), carbonic anhydrase (30000), trypsin inhibitor (20100), and  $\alpha$ -lactalbumin (14400). This gel were dried in an oven at 30°C for 30 minutes and covered and kept in room temperature. Movement distance of the protein were observed as clear line for each sample and compared with protein marker. R value for each band calculated by;

$R_f = \frac{\text{band distance from first distance}}{\text{band of first distant to reference point}}$

Value of reference point was the final point that been detected by coloured protein. A graph of  $R_f$  value of standard protein with log molecular weight for each protein were formed. Molecular weight of each sample of protein can be defined by calibration curve based on  $R_f$  value of sample (Hoffman, 1977).

#### *Amino acid analysis with High Performance Liquid Chromatography*

Analysis of amino acid content were analyse by PICO-TAG method. This method involves protein hydrolysis, sedimentation process, and HPLC analysis. Fresh prawn sample of each molt stage were prepared for amino acid content analysis. 0.2105 g of prawn sample was weight after mixing. 20.0 ml of 6N hydrochloric acid added to the sample and place into an oven at 110°C for 24 h. The samples were then pour into 50 ml volumetric flask and added with HPLC solution until volume 50.0 ml. The solution were mixed and sieved. This process was specifically for analyse methionine and cystine content. This is because during 6N HCl hydrolysis, both of this amino acid was denatured. 2.0 ml hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) and 18.0 ml HCOOH (99.9%) were added into tube with solution before and placed in room temperature for 1 h. The solution was then kept in refrigerator until use. 0.2105 g sample of prawn was placed into test tube and placed into beaker filled with ice cube. After then, cold 4.0 ml performic acid was place into each contained prawn sample and these tubes were kept into refrigerator for 16 h. After 16 h, 0.4ml HBr (5°C) added into each tube in laminar flow and kept back into refrigerator for half an hour. Drying method was carried out by rotary evaporator at 65°C, and followed by hydrolysis. 10  $\mu$ l of sieved sample were taken from steps before and placed into small tubes. These small tubes contained each postmolt, premolt and intermolt stage sample place in dryer PICO-TAG and vacuum at 100 millitorr. From mixture solution methanol: water (1:1), 1.0 ml was taken and pour into test tube and followed by 0.25 ml TEA (triethyl amine). The mixture solution was mixed well. 25  $\mu$ l of mixture solution was taken and placed into drying sample tube before. The tubes were then mixed for 10 minutes and repeated drying process for 100 mtorr. 400 $\mu$ l mixture methanol:water (7:1), 50 $\mu$ l TEA and 50 $\mu$ l PITC were mixed in test tube and mixed tube and mixed slowly. From mixed tube, 25 $\mu$ l of the sample was taken and placed into sample tube to repeat drying. Tube were mixed for 10 min and followed by drying at 100 mtorr. These tube samples



were placed in refrigerator until use. 100 µl diluents sample added to each sample tube and mixed. 20 µl were collected from this mixture with syringe and injected into HPLC (Kan and Shipe, 1982).

#### Statistical analysis

All the analysis was carried out triplicate. Data was expressed as the means of measurements. The experimental data were analyzed (p-value) using One way Anova at confidence level of  $\alpha = 0.05$ .

### Result

#### Moisture and protein analysis

Result of moisture and protein analysis of postmolt, intermolt and premolt sample as in Table 1 below;

#### Texture analysis

Elastic modulus values of *Penaeus monodon* at postmolt, intermolt and premolt stage results show in Table 1.

#### Electrophoresis SDS-PAGE

Result of electrophoresis analysis is shown by electrophoretogram in Figure 1. In running gel, protein marker is injected together.

#### HPLC Analysis

HPLC results of amino acid content in *Penaeus monodon* have shown in Table 2 below:

### Discussion

#### Moisture

This study shows an average percentage of moisture value in tiger prawn, *Penaeus monodon* as 80.68%, 79.12% and 78.02% during postmolt, intermolt and premolt respectively. Statistical analysis has found that there was significance difference in moisture percentage for the three samples. Study by Simpson *et al.* (1998) found that there is 79.69% of moisture in fresh shrimp (*Pandalus borealis*) meat. It is generally understood that the increase in size at ecdysis in crustacea is due to water uptake (O'Connor and Gilbert, 1969). Travis (1957), clearly demonstrated that during molt cycle of *Panulirus argus*, the ratio of water to fresh mass is greatest at ecdysis and during A stage and this proportion declines to early premolt before rising again at late pre-molt. A similar pattern is observed in *P. monodon* with a 76% water content at stage A steadily declining to 73 % at D. The water content appears to be minimum during post molt stages A and B. But the water content of the prawn decreases from stage A to B (Suneetha *et*

*al.*, 2009).

#### Texture analysis

Our study have shown that premolt sample have the highest elastic module value (4.44 Ncm<sup>-2</sup> c) and followed by intermolt (2.08 Ncm<sup>-2</sup> b) and postmolt (1.85 Ncm<sup>-2</sup> a) sample. Elastic module value shows the ability of a product to deform. Brauer *et al.*, have found that elastic module value for cultured white shrimp (*Penaeus vannamei*) is 4.0 Ncm<sup>-1</sup>. For meat or food that is made from meat, the properties of highest elastic value show a high quality of a product (Nute *et al.*, 1987). In this analysis, elastic module value is found from ability-deform curve with 75% compressed of sample method. Average value of elastic module for prawn at intermolt stage is 2.08 Ncm<sup>-2</sup>. Prawn is still soft at early intermolt stage, and body tissue is gradually formed at final intermolt. Prawns at premolt stage have hard outer shell. The shape of this prawn are consistent and not easily destroy after been processed. This stage has taken 76% of the whole stages. From the texture analysis been carried out, we have found that elastic module value are highest at this stage and the meat is more hard and elastic. From our results shows the highest moisture content of postmolt stage shrimp has the hardest elastic module value. On the contrary, the lowest moisture content of premolt stage shrimp has the softest elastic module value.

#### Protein

Average percentage of protein in prawns are 22.27%, 23.48%, 23.10% during postmolt, intermolt, and premolt respectively. Statistical analysis have found that there is no significance different in percentage of protein (at  $p < 0.05$ ) throughout molt cycle. Study showed that there is 85.81% protein content in fresh shrimp meat (*Pandalus borealis*) (Simpson *et al.*, 1998). The hepatopancrease periodically accumulates and releases copper during molting and starvations (Arumugam, 1989) and it has been shown to be the site of haemocyanin synthesis (Spindler *et al.*, 1992). Hemocyanin is a copper containing, multi-subunit protein; it has evolved to carry out the specialized functions of oxygen transport in arthropods and molluscs. The oxygen molecule is bound to two copper ions, each of which is coordinated by three histidines. Crustacean hemocyanins are highly variable in quaternary structure. Hemocyanins also gave rise to non-respiratory proteins, which most likely have storage function (Burmester, 2004). From this analysis, we have found that highest percentage of total protein is during intermolt stage. This showed by well tissue generation throughout intermolt stage.

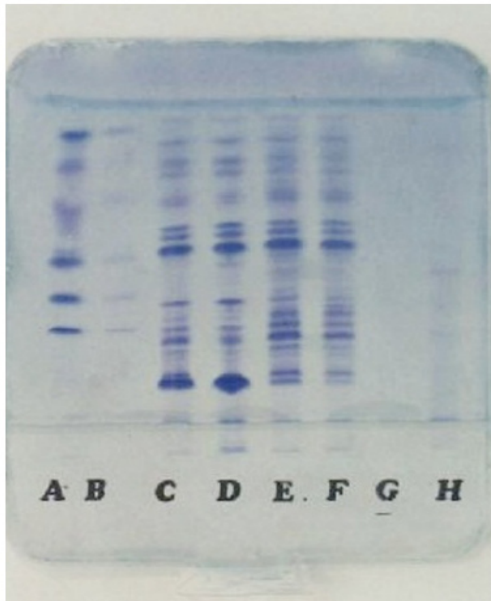


Figure 1. *Penaeus monodon* electrophoretogram where A : Low molecular weight protein marker, B: No sample, C: Postmolt sample, D: Late postmolt sample, E: Intermolt sample, F: Premolt sample, G: No sample, H: High molecular weight protein marker

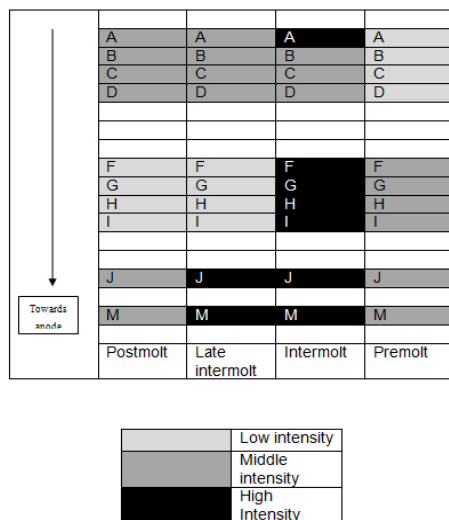


Figure 2. Difference in intensity of band with different molting stages

Low of total protein content during postmolt and premolt are caused by low feed eaten by the prawn. In the study of changes of lipid and protein in starving tiger prawn, *Penaeus esculentus* have found that protein is the main energy sources for the prawn. There is a loss of 550 mg protein after the prawns are starving for 14 days compared to loss of 84 mg lipid. Low protein content during premolt and postmolt are maybe because of prawn haven't eat and protein sources are used as energy supply for the body.

**Electrophoresis SDS-PAGE**

Electrophoresis SDS-PAGE has been used in determines protein content quantity in different molting stages of white shrimp for this study. The

basic principle; every individual protein has a specific charge that depends on pH. If there is mixed of protein placed in electrical charge area and each protein will be move depends on its charge. Movement of different size of protein are depends to charge-weight ratio. Result of sample with protein band has been shown in electrophrogram diagram (Figure 1). Sample of postmolt and late postmolt have found to show a band that is similar to all other band. An obvious similarity is at 0.3488, 0.4106, and 0.5814. From the protein curve, those proteins have molecular weight of 94000, 77108, and 98388 respectively. Analysis of the protein band of intermolt sample shows that no band at Rf value between 0.3256 to 0.4302 (d and e). There is clear band form in line f,g,h with Rf value 0.4302, 0.4651, and 0.5116. No band of d and e can differentiate the stages from postmolt, late postmolt and premolt stage or identification of g and h band that are protein with molecular weight between 54137 to 63095 maybe can be used to identify of prawn at intermolt stage. Band of premolt sample is less clear than intermolt sample. This shows that the protein content at this stage are less and this proven by analysis of protein by Kjeldahl method where there is more protein content during intermolt than during premolt stage. Increase of protein content during intermolt shows a better formation of tissue. Hence as overall, there is no significance different in protein value of these two stages. Band that form in Rf value of 0.4186 to 0.5116 are missing. Two band that are clearly formed at Rf value of 0.4302 and 0.4187, while band of postmolt and late postmolt sample are not forming. a,b,c,d and e band shows the same Rf value for these four sample that been analyse. k and l band have not found at intermolt only. As overall, band of intermolt sample is the clearest then followed by premolt and postmolt sample.

Movement during electrophoresis depends on size and charge particle because of amphoteric protein are natural (Mackie, 1974). At pH value above the isoelectric protein point will have the negative charge and move towards anode. While, if this is happen to pH value lower than isoelectric protein point, positive charge will create a movement towards negative electrode. Figure 2 below shows band of electrophoretic sample that been used in this study.

**Amino acid content analysis (HPLC)**

High Performance Liquid Chromatography has been commonly used in amino acid analysis because of it is faster and more accurate. Protein that been analyse will be hydrolysed with 6N hydrochloric acid (HCl), with standard hydrolysis temperature 110°C

Table 1. Average of moisture, protein content and elastic modulus value of *Penaeus monodon*

Sample	Moisture(%)	Protein(%)	Elastic modulus, Nem <sup>-2</sup>
Intermolt	79.18a	23.48a	2.08b
Postmolt	80.88b	22.27b	1.85a
Premolt	78.02c	23.10a	4.44c

average a,b,c with different letter shows significance difference between sample (p<0.05)

Table 2. Amino acid content in *Penaeus monodon* (gram amino acid/100g protein)

Amino acid	Postmolt	Intermolt	Premolt
Alanine	0.45	0.31	0.49
Ammonia	0.43	0.43	0.43
Arginine	3.92	4.52	6.49
Aspartic acid	2.99	4.93	3.74
Cystein	0.90	1.09	1.09
Glutamic acid	2.76	3.05	3.84
Glycine	1.64	1.77	2.41
Histidine	2.23	3.31	3.13
Isoleucine	5.34	3.86	4.43
Leucine	1.97	2.44	2.43
Lysine	5.12	4.72	6.54
Methionine	22.12	20.37	21.63
Phenylalanine	5.58	5.33	4.55
Proline	1.51	1.50	1.48
Serine	2.76	3.12	3.73
Threonine	16.76	17.56	18.93
Valine	2.49	3.78	2.79
Tyrosine	0.45	0.49	0.55

Table 3. Comparison between *P. monodon* and *P. aztecus*

Amino acid	postmolt	intermolt	premolt	<i>P.aztecus</i>
Alanine	0.0009	0.0008	0.0011	0.0122
Arginine	0.0088	0.0111	0.0149	0.0111
Aspartic acid	0.0068	0.0129	0.0086	0.0350
Cystine	0.0020	0.0026	0.0025	0.0072
Glutamic acid	0.0021	0.0075	0.0089	0.0292
Glycine	0.0037	0.0045	0.0056	0.0124
Hystidine	0.0051	0.0081	0.0072	0.0068
Isoleucine	0.0125	0.0094	0.0102	0.0007
Leucine	0.0044	0.0056	0.0056	0.0054
Lycine	0.0128	0.0129	0.0151	0.0157
Proline	0.0034	0.0036	0.0034	0.0122
Serine	0.0068	0.0075	0.0092	0.0180
Threonine	0.0378	0.0435	0.0438	0.0115
Methionine	0.0498	0.0498	0.0492	0.0159
Phenylalanine	0.0126	0.0130	0.0140	0.0074
Tyrosine	0.0027	0.0031	0.0036	0.0059
Valine	0.0086	0.0092	0.0098	0.0190

Postmolt (gram amino acid/gram sample)

Intermolt (gram amino acid/gram sample)

Premolt (gram amino acid/gram sample)

Juvenile *Penaeus aztecus* (g AA/g sample) (Shewbart et al., 1972)

for 24 h. After hydrolysis, the sample will be dry and redry. Drying steps at low temperature carried out using vacuum agar. Table 2 shows 18 individual amino acid content found from HPLC analysis been carried out.

In this experiment, retention time been used as indicator for type of amino acid present. Quantity of each amino acid is calculated based on volume of peak. We have found that there is a difference in some type of amino acid during molting process. Amino acid content is mostly high during premolt stage and low during postmolt. There is exception for isoleucine content that is high during postmolt at 5.43g/100g while 3.86 g and 4.43 g for other amino acid during intermolt and premolt.

Amino acid profile during postmolt stage shows alanine, ammonia, cystine, and brosin are at lowest and same during intermolt and premolt. Arginine and cystine are found to reduce almost 50% from amount during premolt. There is an obvious reduction between premolt stage and postmolt stage compared to intermolt stage.

Main physiology changes for most decapods happens when Y-organ taken out from X-organ inhibition during late intermolt, C4 and this continuously with synthesize and molting hormone excretion. There are not much morphology changes during early premolt stage Do, but there is where most biochemical processes happen. Glycogen starts to accumulate in epithelium and subepithelial connective tissue (Passano, 1960).

Amino acid content increase during intermolt stages compared during postmolt stages. Cystine content increase than its content during postmolt while lysine at the same amount for both stages and increase 30% during premolt. Ammonia content remain same throughout moltcycle process.

There is no obvious difference of amino acid content between intermolt and premolt sample compared to postmolt and premolt sample. Robertson et al., (1987) explained that intermolt stage have taken 3.5-4.5 days from whole complete moltcycle (20 days) for penaeid prawn. Premolt stage is 70% of a complete moltcycle. Three layers of thin epicuticle, thick calcium exocuticle and endocuticle are form during intermolt stage and completely form during premolt. Epidermis cell is for biochemical storing.

Ammonia chloride, cystine, and leucine content are same in premolt sample and intermolt sample. Aspartic acid, histidine, phenylalanine, proline, and valine shows a little reduce during premolt compared to during intermolt. Total content for three sample are 88.68g, 82.37g and 79.46g amino acid for each 100 gram protein for premolt, intermolt and postmolt sample.

Study on *Penaeus esculentus* have found that total protein amount in blood is not different significantly throughout moltcycle. Concentration of protein decrease during postmolt stage and increase



until maximum during premolt because of changes in volume of blood. Prawn has still remain concentration of some constituent for constant even there is no enough feed that effect its growth (Skinner,1966; Barnes and Blackstock,1973)

This explained some on amino acid that are remains throughout moltcycle such as ammonia chloride. Increase of some amino acid during premolt can also be explained with molting phenomena. Decapods cuticle contained 20-40% protein (Richards, 1951) and increase of amino acid content during proecdysis is for synthesis of cuticle by epidermis.

Table 3 have shown comparison of *Penaeus monodon* with juvenile *Penaeus aztecus*. From table 3 have found that alanine and aspartic acid content in these three samples postmolt, premolt and intermolt lower compared to *Penaeus aztecus*. Arginine, histidine, serine, lysine, tyrosine and valine have in same range for both species. While, cystine and glutamic acid are in ratio 1:3 for postmolt of *Penaeus monodon* compared to *Penaeus aztecus*. Threonine and methionine content have shown 4 times more from value compared. Simpson *et al.* (1998) have found that there is a high level of glycine, proline, arginine, and valine amino acid in fresh *Pandalus borealis* shrimp meat.

## Conclusion

In summary, this study has demonstrated changes in texture and biochemical composition through elastic modulus value and protein composition of *Penaeus monodon* during moltcycle. Our study provides some significance information on biochemical composition changes in protein and amino acid at different stages, premolt, intermolt and postmolt. We have found that electrophoresis SDS-PAGE can be a method to determining stages of prawn in moltcycle. Further research, however, recommended to deeper investigate on other biochemical composition such as carbohydrate and lipid that may have correlation in prawn growing. This information also can be used in determining nutritional component needed by prawn specifically at different stages as they grow up.

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