Note

Amino-acid and biochemical composition of whole body and molt of the Indian white shrimp

J. SYAMA DAYAL, S. AHAMAD ALI AND C. SARADA

Central Institute of Brackishwater Aquaculture, 75, Santhome High Road, Raja Annamalai Puram, Chennai 600 028, India.

ABSTRACT

The amino acid and biochemical composition of molt (exuvia) and whole body of the shrimp, *Fenneropenaeus indicus* were analysed along with morphometric relations. The wet weight of exuvia $(0.22 \pm 0.07 \text{ g})$ is nearly 4.8% of the whole shrimp weight $(4.50 \pm 1.42 \text{ g})$. The goodness fit statistics R² revealed highly significant relationship in weights of wet exuvia- wet shrimp, dry exuvia- wet shrimp and dry exuvia- dry shrimp with values 0.90, 0.89 and 0.88, respectively indicating that more than 88% of variation of the data explained by the models. The crude protein and crude fat contents of whole shrimp expressed on dry matter basis were 3-5 times higher than those found in exuvia. However, the exuvia of the shrimp had more than twice the dry matter and about four times ash contents when compared to that of whole shrimp body. The amino-acid composition of the whole shrimp body and the exuvia expressed on dry matter basis were 63.37 and 11.94 per cent, respectively. Glutamic acid, leucine and aspartic acids in the whole shrimp and glutamic acid, glycine and alanine in the exuvia were the predominant amino acids.

The integument of crustaceans consists of basement membrane. epidermis and cuticle. The cuticle of crustacea acts as an exoskeleton providing mechanical support for their body preventing from physical injury and protecting them against extreme ionic and osmotic changes (Travis, 1960). This exoskeleton is periodically shed through the process of molting. Molting is one of the most important physiological processes for crustacean growth since increase in body size occurs in a series of steps associated with the castings of the old exoskeleton (Passano, 1960). Frequency of molting depends not only on species but also on the growth stages. Penaeids molt more frequently, once every few days or weeks while, large

crustaceans such as crabs and lobsters may molt only once or twice a year in their adult stages (Dall *et al.*, 1990).

The molt of crustacea consists of both organic and inorganic materials such as protein, chitin and minerals (Travis, 1960) and their composition varies greatly from species to species (Welinder, 1974). As a result of molting, crustaceans loose considerable amounts of these nutrients and these should be replenished.

Crustaceans are having the habit of eating their exuvia after ecdysis (Forster, 1976; Thomas *et al.*, 1984). If this nutrient source is not accounted for, it could lead to errors in nutritional studies, especially those relating to dietary requirements of protein, amino acids and minerals. If the composition of this exuvia is known, the amount of these nutrients provided by ingested molts can be determined. Most of the works on these aspects are limited to relationships between exuvia and the total body length and weight only (Dall *et al.*, 1990). The present study is aimed at determining the biochemical composition along with amino acid profiles of exuvia and the whole body in the Indian white shrimp, *F. indicus*.

Juveniles of F. indicus weighing 4.5 $g \pm 1.42$ (mean \pm S.D.) were obtained from the wild and maintained in a 1000 l fibre reinforced plastic tank. One shrimp per tank were randomly distributed in twelve 50 l fibre reinforced plastic tanks. During the experimental period, the water temperature was $29 \pm 1^{\circ}$ C, salinity at 33 $\%_{00}$ and dissolved oxygen 7 mg/l. Eighty per cent of the water in the tanks was exchanged daily. The animals were fed *ad libitum*, twice a day with a standard formulated feed having 39.5 % crude protein.

The molting of the shrimp was closely monitored and the exuvia and freshly molted shrimp were removed from the tanks. Empty tanks were cleaned and restocked with new, randomly selected shrimps from holding tank. The trial was continued for 30 days to collect exuvia and molted shrimp. Three shrimps from holding tank and exuvia samples were analyzed for biochemical composition (AOAC, 1990). Amino acids were analyzed after sealed tube hydrolysis with 6 N HCl (Spackman et al., 1958; Finlayson, 1964) in Shimadzu HPLC system using dye as fluorescent detector post-column derivatization with O-Pthaladehyde. Each analysis was done in duplicate.

The functional relationship between

shrimp weight and exuvia weight was determined by the following regression equation

$$Y_i = \beta_0 + \beta_1 X_i + \varepsilon_i$$

Where Y_i is the response variable, X_i is a regressor variable, β_0 and β_1 are unknown parameters to be estimated, and is an error term (Gomez and Gomez, 1984). As mentioned above, regression function is fitted to the data. The following stochastic regression equations are considered.

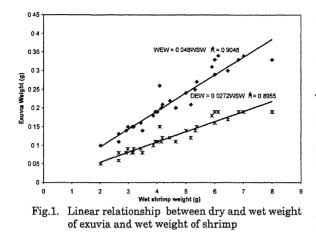
$$WEW = aWSW + \varepsilon$$
 ²

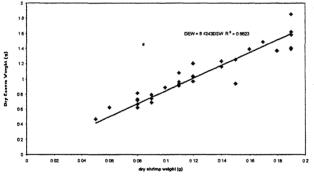
$$DEW = aWSW + \varepsilon$$
 .3

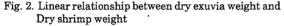
$$DEW = aDSW + \varepsilon$$

Where WEW is wet exuvia weight (g), WSW is wet shrimp weight (g), DEW is dry exuvia weight (g), DSW is Dry shrimp weight (g), 'a' is parameter to be estimated and ε is error term. The constant term is not included in all the equations 2, 3 and 4 because if input equals to zero then one would expect the output to be zero as well.

The average weight of shrimp and exuvia were 4.50 ± 1.42 and 0.22 ± 0.07 g, respectively indicating that the wet weight of exuvia was $4.8 \pm 0.46\%$ of the wet weight of the whole shrimp body. The dry exuvia was $11.74 \pm 1.13\%$ of the dry weight of the whole shrimp body. Kibra (1993) reported that the loss of weight immediately following ecdysis averaged 4.9% in Penaeus monodon, which is linked with the loss of the exuvia. The morphometric relationships (Fig.1 and Fig.2) between wet and dry weight exuvia and wet shrimp weight are highly significant ($R^2 = 0.90$, P < 0.001) indicating strongest relationship in both the cases. Regression calculations







reported earlier (Sarac *et al.*, 1994) in *P.* monodon on wet exuvia weight-wet body weight, dry exuvia weight-dry body weight and dry exuvia weight-wet body weight were 0.75, 0.78 and 0.83, respectively. The relationship between dry weight of exuvia and dry shrimp weight shown similar results ($R^2 = 0.88$, P < 0.001). Comparable relationship between the dry weight of whole prawn body and exuvia ($R^2 = 0.97$, P < 0.001) was reported by Hewitt (1984) in the freshwater prawn, Macrobrachium australiense.

The dry matter content of whole shrimp body was 23.15 ± 0.16 per cent.

Alava and Pascual (1987) and Sarac et al. (1993)also found that the dry matter was in the range of 23.4 - 26.7 per cent for iuvenile P. monodon. The exuvia had more than half (56.48%) dry matter. Similarly, high dry matter content was reported in other crustacean species, Cancer magister (Allen, 1971). The crude protein value observed (69.94%) in F. indicus in this study was within the range of crude protein reported for juveniles of P. indicus (59.9-64.0), P. monodon (61.8 - 75.81) and Metapenaeus monoceros (62.1-67.81)(Vattheeswaran and 1986: Ahamad Ali. Ahamad Ali, 1996; Alava and Pascual, 1987; Sarac et al., 1993). Alava and Lim (1983) reported that crude protein levels in the body of juvenile P.

monodon were directly related to dietary protein levels. Variations in dietary energy levels significantly influenced the total body lipid levels (5.1-10.7%) of juvenile *M. monoceros* (Kanazawa *et al.*, 1981).

The exuvia of the shrimp had more than twice the dry matter and about four times ash contents when compared to that of whole shrimp body (Table1). However, crude protein and crude fat contents of whole shrimps expressed on dry matter basis were 3-5 times higher than those found in exuvia. The crude protein recorded in exuvia (23.11%) in *F. indicus* is within the range (20 - 40%)

J. Syama Dayal et al.

Composition(%)	Shrimp body	Exuvia	Ratio * (%) 11.74 ± 1.12	
Dry matter	23.15 ± 0.16	56.48 ± 0.84		
Crude protein	69.94 ± 0.14	23.11 ± 0.218	3.87 ± 0.37	
Ether extract	5.48 ± 0.08	1.09 ± 0.14	2.34 ± 0.22	
Ash	16.68 ± 1.01	63.01 ± 1.48	44.34 ± 4.26	

TABLE 1: Biochemical composition[#] of exuvia and whole body of F. indicus.

On Dry matter basis except for dry matter * Component in wet exuvia (g)

— X 100 Component in wet shrimp (g)

TABLE 2: Amino acid composition of whole body and exuvia F. indicus.

Amino acids	Whole shrimp body		Exuvia		Ratio [*] of AA in wet exuvia to AA in wet shrimp body
	% Dry matter basis	Ratio to total AA (%)	% Dry matter basis	Ratio to total AA (%)	
ASP	5.83	9.20	1.01	8.46	2.04
THR	2.66	4.2	0.6	5.03	2.65
SER	2.51	3.96	0.57	4.77	2.67
GLU	10.11	15.95	2.51	21.02	2.92
PRO	3.91	6.17	0.75	6.28	2.25
GLY	5.33	8.41	1.1	9.21	2.42
ALA	4.02	6.34	1.05	8.79	3.07
CYS	0.6	0.95	0.08	0.67	1.57
VAL	2.94	4.64	0.66	5.53	2.64
MET	1.66	2.62	0.11	0.92	0.78
ISO	3.08	4.86	0.28	2.35	1.07
LEU	6.14	9.69	0.6	5.03	1.15
TYR	2.29	3.61	0.53	4.44	2.72
PHE	2.66	4.20	0.8	6.70	3.53
HIS	2.01	3.17	0.11	0.92	0.64
LYS	4.21	6.64	0.33	2.76	0.92
ARG	3.41	5.38	0.85	7.12	2.93
TOTAL	63.37	100	11.94	100	2.21

* Amino acid in wet exuvia (g)

- X 100

Amino acid in wet shrimp (g)

reported earlier (Dall et al., 1990). The crude protein of exoskeleton decreased from 53.5%, just after molting to 30.8%, after14 days in Astacus fluviatilis (Welinder, 1975). Exuvia contain up to 20% chitin (Welinder, 1975) which is a non-protein compound containing 7% N (Muzzarelli, 1977). Further, the exuvia also contains other non-protein nitrogen compounds such as chitosan. Because of this true protein content in exuvia is very less as represented by low amino acid content (Table 2). The ash content of dry exuvia is 63.01%, which is mainly present in the form of calcium salts (Welinder, 1975). Since calcium is a major part of the exuvia, shrimps require high levels of calcium intake, especially after molting. This requirement seems to be met mainly by absorbing Ca available in the surrounding water (Davis and Gatlin, 1991).

The amino-acid composition of the whole shrimp body and the exuvia expressed on dry matter basis is presented in Table 2. Glutamic acid was the predominant amino acid in whole shrimp body (10.11%) and in exuvia (2.51%). This was followed by leucine (6.14%) and aspartic acid (5.83%) in shrimp body and glycine (1.10%) and alanine (1.05%) in exuvia. The amounts of sulphur-containing amino acids, cystine and methionine, were low in the exuvia (0.08 and 0.11%, respectively). The essential and non-essential amino acids constituted 28.72 & 34.65 and 4.34 & 7.6 per cent, respectively in the whole body and in the exuvia. The total amino acids in exuvia had 2.21% of that of the whole shrimp. Glutamic acid, leucine and aspartic acids were also the same three amino acids predominant in Penaeus aztectus (Shewbert et al., 1972). The exoskeleton amino acid reported in Cancer magister by Allen (1971) were valine (1.84%), aspartic acid (1.14%), glutamic acid (1.03%), glycine (0.91%)and alanine (0.69%). The amino acid composition of the exuvia of F. indicus is similar to those reported for other

penaeid shrimps (Sameshima et al., 1973; Welinder, 1974). The loss of essential and non-essential amino acids through ecdysis in F. indicus is 1.81% and 2.45%, respectively. Loss of these nutrients in P. monodon was 1.51% and 1.92%, respectively (Sarac et al., 1994). The study revealed that there is a strong relationship between the weights of exuvia and shrimp body weight. The proportion of dry matter and ash were more in exuvia compared to whole shrimp body, whereas it is vice - versa in case of protein and lipid proportions. The exuvia contains considerable amounts of crude protein and 2.21% of amino acids when compared to that of shrimp. Nutrient consumption from exuvia could be estimated using the regression equations calculated in the present study especially in dietary nutrient requirement studies.

Acknowledgements

The authors express their sincere thanks to Dr. Mathew Abraham, Director and Dr. G.R. M. Rao former Director, Central Institute of Brackishwater Aquaculture, Chennai for their encouragement.

References

- Ahamad Ali 1996. Carbohydrate nutrition under different dietary conditions in prawn Penaeus indicus. J. Aquacult. Trop., 11: 13-25.
- Allen, W. V., 1971. Amino acid and fatty acid composition of tissues of the dungeness crab (*Cancer magister*). J. Fish. Res. Bd. of Canada, 28: (8) 1191-1195.
- Alava, V. R. and C. Lim 1983. The quantitative dietary protein requirements of *Penaeus monodon* juveniles in a controlled environment. *Aquaculture*, **30**: 53 - 61.
- Alava, V. R. and F. P. Pascual 1987. Carbohydrate requirement of *Penaeus* monodon (Fabricus) juveniles. Aquaculture, **61**: 211- 217.
- Dall, W., B.J. Hill, P.C. Rothlisberg and D.J. Staples 1990.The biology of the

Penaidae. Adv. Mar. Biol., 27: 1-489.

- Davis, A. and D.M. Galtin 1991. Dietary mineral requirements of fish and shrimp. In: D.M. Akiyama and R.K.H. Tan (Eds.), Proceedings of the Aquaculture Feed processing and Nutrition workshop, 19 - 25 September 1991, Thailand and Indonasia, p. 49 -67.
- Finalayson, A.J., 1964. Amino acid recovering in the analysis of some feed samples. Can. J. Plant Sci., 45: 184 – 188.
- Forster, J.R.M., 1976. Studies on the development of compound diets for prawns. In: S.P. Kent, W.N. Shaw and K.S. Danberg (Eds.), Proceedings of the First International Conference on Nutrition, Lewes. Delware, University of Delware, New York. P. 229-48.
- Gomez, K. A. and A. A. Gomez 1984. Statistical Procedures for Agricultural Resaerch, 2nd Edition. John Wiley and Sons, New York, p. 8-19.
- Hewitt, D.R. 1984. Growth and bioenergetics of the freshwater prawn, Macrobrachium australiense (Holthuis), Honours Thesis, Townsville, Qld, Australia, 84 pp.
- Kanazawa, A S. Teshima, S. Matsumoto, and T. Nomra 1981. Dietary protein requirements of the shrimp, *Metapenaeus monoceros. Bull. Jpn. Soc. Sci. Fish.*, 47: 1371 - 1374.
- Kibra, G., 1993. Studies on molting, molting frequency and growth of shrimp (*Penaeus monodon*) fed on natural and compound diets. Asian Fisheries Science, 6: 203-211.
- Muzzarelli, R.A. A., 1977. *Chitin*. Pentagon Press, New York, 309 pp.
- Passono, L.M., 1960. Molting and its control. In: *The physiology of Crustacea.*, 1: 473-536, T. Waterman (Eds.). Academic Press, New York.
- Sameshima, M., T. Katayama and O. Deshimaru 1973. Constituents of the amino acids composed of the protein in the exoskeletons of the crustacea. *Mem.*

Date of Receipt : 05-11-2002 Date of Acceptance : 11-04-2005 Fac. Fish. Kagoshima Univ., 22: 33-37.

- Sarac, H.Z., H. Thaggard, J. Saunders, M. Gravel, A. Neil and R.T. Cowan 1993. Observations on the chemical composition of some commercial prawn foods and associated growth responses in *Penaeus monodon. Aquaculture*, 115: 97-110.
- Sarac, H.Z., N.P. McMeniman, H. Thaggard, M. Gravel, S. Tarbett and J. Saunders, 1994. Relationships between the weight and chemical composition of exuvia and whole body of the black tiger prawn, *Penaeus monodon. Aquaculture*, **119**: 249 - 258.
- Shewbart, K.L., W.L. Mies and P.D. Ludwig, 1972. Identification and quantitative analysis of the amino acids present in protein of the brown shrimp *Penaeus* aztecus. Mar. Biol., 16: 64 - 67.
- Spackman, D.H., W.H. Stein and S. Moore, 1958. Automatic recording apparatus for use in the chromatography of amino acids. Anal. Chem., 30: 1190.
- Thomas, M.M., D.C.V. Easterson and M. Kathirvel 1984. Energy conversion in the prawn, *Metapenaeus dobsonii* (Mieps) fed on artificial feed. *Indian J. Fish.*, **31** (2): 309-312.
- Travis, D.E. 1960. Matrix and mineral deposition in skeletal structures of decapoda crustacea (Phylum Arthropoda). In: R.F. Sognnaes (Ed.), *Calcification in Biological Systems*,: 57 - 116.
- Vattheeswaran, S and S. Ahamad Ali, 1986. Evaluation of certain substances as growth-promoting agents for the prawn Penaeus indicus. Indian J. Fish., 33 (1): 95-105.
- Welinder, B.S., 1974. The crustacean cuticle. I. Studies on the composition of the cuticle. Comp. Biochem. Physiol., 47A: 779-787.
- Welinder, B.S., 1975. The crustacean cuticle. II. Deposition of organic and inorganic material in the cuticle of Astacus fluviatilis in the period after molting. Comp.Biochem.Physiol., 51B: 409-416.