J. mar. biol. Ass. India, 2002, **44** *(1&2) : 220 - 225*

Studies on optimum dietary protein requirement for *Penaeus semisulcatus* de Hann

S.D. Gopakumar*

Central Marine Fisheries Research Institute, Cochin, India.

Abstract

Optimum protein requirement in green tiger prawn *Penaeus semisulcatus* using different protein diet ranging from 20,25,30,35,40,45% was studied. Since the protein source need appeared to be similar in amino acid profile of the experimental organism viz; *P.semisulcatus,* shrimp meal was used throughout the study. The composition of the shrimp meal was 74.5% moisture and 18.2% protein in live weight. The protein content of the dried shrimp was 71.3%. The study indicated the optimum dietary protein requirement for the species as 40%. Since the percentage weight at 35% protein diet was close to that of 40%, a range between 35-40% was taken as optimum for growth.

Proteins which form 65-85% of dry matter in the body are vital as are the functions they perform. In aquaculture the cost of feed is about 60% of the total expenditure, where in cost of protein source is the single expensive item. Protein carries out three important functions *viz.* 1. maintenance - making good of wear and tear and replacement of aged tissues; 2. growth - formation of newer tissues and 3. physiological - (i) serves as energy source on being deaminated (ii) in systhesis of varied protein and non protein biochemicals - coenzymes, hormones, immunochemicals of lubricants (mucins, mucus), antifreeze substances, toxins and venoms (iii) in osmoregulation (iv) in body fluids and tissue pools aiding in the transport of molecules.

Protein cannot be stored the way carbohydrate is stored as glycogen or lipid as fatty adipose tissue. However, presence of protein reserves as metabolic pools of free amino acids have been identified in crustaceans.

Optimal dietary protein requirement

In 1958 De long *et al.* first proposed that a minimum quantity dietary protein is required for giving optimum weight gain for Chinook salmon. This concept was further improved by Ogino and Saito (1970) who first elaborated this concept in their study on carp. They found that weight gain was directly proportional to dietary protein level upto an optimum which was 38% for carp; and when fed with zero protein diet, protein efficiency ratio and net protein utilization decreased linearly with increase in dietary protein level; and when fed with zero protein diet, protein efficiency

* Present address: Lecturer, Centre for Aquaculture Studies, Sacred Heart College, Thevara, Cochin

ration and net protein utilization were negative, approximating endogenous nitrogen excretion. Even when protein was not included in the feed, animal need to break down its body protein to meet metabolic needs. Thus the minimum protein requirement is just met, without resorting to degrowth (Suresh Kumar, 1985). In crustaceans the rate of consumption of food has been found to be high when protein content was low in the diet and also when protein was above the optimal level. In the former, higher consumption rate was coupled with higher protein assimilation efficiency while in the latter assimilation of portein was low. The latter was, therefore, termed as 'the gluten effect'. Thus when protein was present in the diet above the level of optimal requirement, assimilation and growth were not proportionate to consumption (Easterson, 1987). The survey of optimal dietary protein requirement in shrimps indicates, that it varies between 36-50%, averaging around 40%. In the present communication the optimum dietary protein requirement in the green tiger prawn (*P.semisulcatus*) is discussed.

The author is very much thankful to the Director, CMFRI for providing facilities and Shri. D.C.V. Easterson for his suggestions.

Material and methods

The live *Penaeus semisulcatus*, were collected from shore seines being operated at Vellapatti, north of Tuticorin in Gulf of Mannar. Healthy shrimps of uniform size 2 to 3 g were collected and stocked at Tuticorin R.C. of CMFRI. The meal of the same or related species with similar body composition and amino acid profile is considered a suitable standard protein source as it gives the best growth. (Deshimaru and Shigeno, 1972). For the preparation of shrimp meal, shrimps of all sizes were also collected from here. Further, samples of *P.semisulcatus* required for the analysis of body protein composition and amino acid profile were also collected from this centre.

Feeds similar in composition for all other components but variable in percentage protein content of 20,25,30,35,40 and 45 were prepared (Table-1).

In the feed prepared, the carbohydrate content was kept at 12 % which was mostly met by the shrimp meal itself and whenever if fell short, the remaining required portion out of 12 % was met by adding required quantity of potato starch containing 92% digestible carbohydrate. Cholesterol and essential steroid for crustaceans was also added in all the diets at 0.5%. Mineral mixture according to USP XIV supplied by SRL, Bombay was used. To bring the composition to 100% an 'inert' compound alpha cellulose was used as filler.

From the feeding experiments conducted for a period of 30 days with *P.semisulcatus*, in triplicates for each of the diet groups having 20,25,30,35,40 and 45% protein the optimal dietary protein level was arrived at.

During the experiment in each basin of 35 litre capacity 5 numbers of healthy

Table-1. *Consumption of feeds having different protein levels*

Ingredients	20%	25%	30%	35%	40%	45%
* Prawn meal	26.6	35.7	42.9	50.0	57.02	64.4
$ Potato starch	8.7	3.3	2.2	1.1	0.1	0.0
Shark liver oil	7.5	7.5	7.5	7.5	7.5	7.5
Vitamin mixture	10.0	10.0	10.0	10.0	10.0	10.0
Mineral mixture	6.0	6.0	6.0	6.0	6.0	6.0
Cholesterol	0.5	0.5	0.5	0.5	0.5	0.5
Agar as binder	6.5	6.5	6.5	6.5	6.5	6.5
Alpha Cellulose as filler	32.2	30.5	24.4	18.4	12.3	5.1

* Contained 70% protein and 14% carbohydrate

$ Contained 92% digestible carbohydrate.

P.semisulcatus of uniform size acclimatized to laboratory conditions were taken. The medium of filtered sea water was changed every day in the morning, after the collection of faecal matter. During the experimental period the salinity was constant at 35‰ and water temperature varied between 29-31°C. To maintain dissolved oxygen at optimum, filtered air was constantly bubbled with least disturbance to the bottom sand. The pH of water was near constant at 8.5. The shrimp being a nocturnal feeder and burrowing in habit a sand bed 3 cm thickness was provided. Sufficient pelletised feed kept on empty shells was given every day at 6 pm and left over collected after 8 hrs and dried. The quantity of left over was deducted from that of food offered to estimate the quantity for food consumed. Faecal matter collected in the morning were carefully washed off the salt and kept separately for each experiment in pre-weighed vials. The experimental animals were carefully weighed before and at the end of the experiments.

All the biological samples were dried in hot air oven kept at 65°C. Finely powdered samples of shrimp, feed and faecal matter were used for analysis. Protein was estimated following Kjeldahl method (Strickland and Parsons, 1972; AOAC, 1975) and carbohydrate by anthrone-sulphuric acid method (Umbreit *et.al.* 1959). Factor 6.25 was used to calcultate the value of protein from nitrogen.

Following formulae according to Winberg (1956) were used.

From the values of consumption and growth the quantity of protein consumed and gained (added to body) were calculated.

$$\text{Digestibility of protein} = \frac{\text{Protein consumed (g)} - \text{Protein in faceces (g)}}{\text{Protein consumed (g)}} \times 100$$

Results

The analysis of samples of the dried and powdered prawn *P.semisulcatus* indicated that the moisture content was 74.5% and the protein present in the dry matter accounted for 71.3%. The protein content in live prawn accounted to 18.2%.

Optimal dietary protein requirement

Studies conducted to elucidate optimal quantity of protein required to be present in the feed for better growth, with diets containing 20,25,30,35,40 and 45% protein gave 2.4, 3.4, 7.8, 10.1, 11.9 and 8.6% weight gain. Thus dietary protein level of 40% was taken as optimal. Since the percentage weight gain at 35% protein level was close to that of 40% calculations were also made for 35% dietary protein level (Table 2).

The quantity of food consumed for 40% protein group ranged from 30.4 to 46.2g with a mean of 37.3g while for 35% protein group it was 30.1 to 38.6g with a mean value of 35.58g which in terms of quantity of protein consumed worked out to 12.4g (range 10.5 to 13.6g) for 35% dietary protein group and 14.9g (range 12.2 to 18.5g) for 40% protein group.

The quantity of faeces voided for the period of 30 days varied from 12.2 to 15.5g in 35% protein group and 16.4 to 24.4 g for 40%. The mean values respectively were 14.3 and 20.0g. The protein content in the faeces varied from 1.7 to 2.2 g (mean of 1.9g) and between 1.84-2.74g (mean of 2.2g) in 35% and 40% dietary protein groups.

The digestibility of protein between 35% and 40% dietary protein groups was found to be 84.0% (range 83.6 to 84.5%) and 84.9% (range 84.7 - 85.2%) respectively.

The quantity of food consumed per day ranged from 7.1-7.6% of body weight (mean 7.4%). The protein consumed per day by a

Table 2. *Optimum protein requirement for Penaeus species*

Penaeus spp	Size of shrimps. weight in gms. (g)	% Protein studied	Recommended % protein	Authors
P. aztecus	0.02, 0.14	40-48	40	Venkataramiah *et al*, 1975
	4.0, 10.0, 15.0	22-36	30-36	Smith *et al*, 1985
P. californiensis	10	25-40	35	Colvin and Brand, 1977
P. setiferus	4	14-52	28-32	Andrews *et al*, 1972
	3.7, 9.8, 14.7	22-36	30	Lee and Lawrence, 1985
P. vannamei	0.03	25-40	30	Colvin and Brand, 1977
	4.0, 9.8, 20.8	22-36	30	Smith *et al*, 1985
P. japonicus	5.3	63-76	>60	Deshimaru and Shigeno, 1972
	4.2	2-66	52-57	Deshimaru and Yone, 1978
P. monodon	0.5, 1.8	2-62	45-50	Lee, 1971
	1.3	25-60	40	Alava and Lim, 1983
P. indicus	1	21-53	43	Colvin, 1976
P. semisulcatus	2.4	20-45	35-40	Present study

Table 3. *Parameters of consumption and growth calculated for feeds having optimal dietary protein*

Dietary Protein level	Initial Wet weight (g)	Initial Dry weight (g)	Initial Protein (mg)	Final Wet weight (g)	Final Dry weight (g)	Final Protein (mg)	Growth Wet weight (g)	Growth Dry weight (g)	Growth Protein (mg)	Consumption Dry weight (g)	Consumption Protein (mg)	Faeces Dry weight (g)	Faeces Protein (mg)	Digestibility Protein (mg)	Digestibility Protein %	C Dry weight (g)	C Protein (mg)	G Wet weight (g)	G Protein (mg)
35% A	12.83	3.27	2.33	15.48	3.95	2.81	2.65	0.68	0.48	30.09	10.53	12.17	1.69	8.84	83.95	7.09	2.48	0.612	111
B	15.9	4.05	2.89	17.9	4.56	3.25	2	0.51	0.36	37.45	13.11	15.47	2.15	10.96	83.6	7.39	2.59	0.394	72
C	16.4	4.18	2.98	17.6	4.49	3.2	1.2	0.31	0.22	38.6	13.51	15.12	2.1	11.41	84.46	7.59	2.65	0.235	43
X	15.04	3.83	2.73	16.99	4.33	3.09	1.95	0.5	0.36	35.38	12.38	14.25	1.98	10.4	84	7.36	2.58	0.395	72
40% A	19.07	5.02	3.58	22.6	5.76	4.11	2.9	0.74	0.53	46.24	18.5	24.42	2.74	15.76	85.19	7.29	2.92	0.457	83
B	20	5.1	3.64	21.9	5.58	3.98	1.9	0.48	0.34	35.13	14.05	19.16	2.15	11.9	84.7	5.59	2.24	0.302	55
C	12.9	3.29	2.35	14.3	3.65	2.6	1.4	0.36	0.25	30.38	12.15	16.4	1.84	10.31	84.85	7.42	2.97	0.343	62
X	17.5	4.46	3.18	19.6	5	3.57	2.1	0.54	0.39	37.25	14.9	20	2.24	12.66	84.97	6.69	2.68	0.377	69

A,B and C are triplicates of experimental animals.
X Mean values of experimental animals.
C Consumption per day % body weight
G Growth per day in % body weight

100g live *P.semisulcatus* worked out to a mean value of 2.6g (2.5 - 2.7g) in those fed with diets having 35% protein. On the other hand for those fed with 40% protein in diet the quanitity of protein ingested for 100g body weight varied between 2.2-2.9g with a mean value of 2.7g.

Live weight gain in percentage body weight ranged between 0.235 to 0.612 with a mean value of 0.395 for 35% dietary protein groups. The quantity of protein gained or added thereby as growth for these shrimps was between 43 to 111mg with a mean value of 72mg for 100g body weight. For the shrimps fed with 40% protein in the diet the growth in percent body weight ranged between 0.302 to 0.457 with a mean of 0.377. The quantity of protein gained here worked out to 55-83mg with a mean value of 69mg (Table 3).

Discussion

The earliest compounded diet formulated for *Penaeus japonicus* was by Kanazawa *et al,* (1970) who proposed a diet comprising of brine shrimp, salmon and silkworm pupae as an alternative to short-necked clam meat that was becoming scarce. It has about 55% protein in it and produced a growth rate around 72% (New, 1976). Lee (1971) found that increase in dietary protein levels up to 45.8% in *P.monodon* resulted in better growth, while below 18.3% resulted in loss of weight. He also found that the growth rate was much better at 45.8% level than at 62.1%. Lee's work was followed by many study on dietary optimal protein level for different penaeids. Review of the selected results

given in Table 2 indicate that optimal values ranged from 28% for *P.setiferus* (Andrews, *et al.*, (1972) to above 60% for *P.japonicus* (Deshimaru and Shigeno, 1972). But the optimum value in most of the cases lies between 35% and 40%, as in the present study on *P.semisulcatus*. The mean values of weight gain at 35% protein level in the diet in the first and the second experiments were 7.8 and 12.9, while for 40% it was 10.1 and 12.0, whereby indicating the dietary optimal value for protein being 35-40%.

Optimal protein values have been reported to be high whenever crystalline amino acids or protein hydrolysates have been used in the diet (Deshimaru and Kuroki, 1975; Chen, 1993).

Further, defective processing of feed would make the amino acids only partially available to the organism leading to short supply of proteins. Further, utmost care was taken while feed processing to avoid oxidation. Since the protein source used being shrimp meal comprising mostly of *P.semisulcatus, P.indicus and Metapenaeus dobsoni,* it was similar in biochemical composition especially in amino acid profile with the experimental organism *viz. P.semisulcatus.*

References

Alava, V.R. and C.Lim. 1983. *Aquaculture,* **30**: 53.

Andrews, J.W., I.V. Sick and G.J. Baptist. 1972. *Aquaculture,* **1**: 341-347.

Chen, Houng-Young, 1993. *J. World Aquacul. Soc.,* **24** (2) : 231-240.

Colvin, P.M., 1976. *Aquaculture,* **7** : 317.

Colvin, L.B. and C.W. Brand. 1977. *J. World Maricult.Soc.* **8**: 821.

DeLong, D.C., J.E. Halver and E.T. Mertz. 1958. *J. Nutr,* **65**: 589-599.

Deshimaru, O and K. Shigeno 1972. *Aquaculture,* **1** : 115-133.

---------- and K.Kuroki 1975. *Bull. Japanese. Soc. Sci. Fish.,* **41**: 301-304.

---------- and Yone, Y., 1978. *Bull. Japan. Soc. Sci. Fish.* **44**: 1395.

Easterson, D.C.V. 1987. *Proc. Summer Inst. Recent Adv. Finfish and Shellfish Nutr.,* Tech. Paper 5, Pp.20.

Kanazawa, A., M. Shimaya, M. Kawasaki and K.Kashiwada. 1970. *Bull. Japanese Soc. Sci. Fish.,* **36**: 949-954.

Lee, D.L. 1971. *Aquaculture,* **1** : 1-13.

Lee, P.G. and A.L. Lawrence. 1985. *J. World Maricult. Soc.* **16**: 275.

New, M.B. 1976. *Aquaculture,* **9**: 101-144.

Ogino, C., and Saito 1970. *Ibid.,* 36(3) : 250-254.

Smith, L.I., P.G. Lee, A.L. Lawrence and K. Strawn. 1985 *Ibid.* **46** : 85

Strickland, J.D.H., and T.R. Parsons. 1972. *Bull. Fish. Res. Bd. Canada,* **167**: 143-147.

Suresh Kumar, K. 1985. Study on the protein budget in different size groups of *Penaeus indicus.* M.Sc., Thesis, University of Cochin, Cochin. pp.48.

Umbreit, W.W., R.H. Burris and T.F. Stauffer. 1959. *Manometric Techniques. Burgess Pub. Co., Minaneapolis.* **V Edition.** 387 Pp.

Venkataramiah, A., G.J. Lakshmi and G.Gunter. 1975. *Aquaculture,* **6** : 115.

Winberg, G.G. 1956. *Trans Ser., Fish. Res. Bd. Cananda No.***194**: 202 Pp.