

Growth and Body Composition of Juvenile Freshwater Prawn, *Macrobrachium rosenbergii*, Fed Different Dietary Protein / Starch Ratios

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Abstract: Two feeding experiments were carried out on the freshwater prawn *Macrobrachium rosenbergii* in order to determine the requirement from dietary protein and energy levels in term of growth, weight gain, % weight gain, specific growth rate and protein efficiency ratio. The highest growth and survival rates were recorded in group of prawn fed diets with P/S ratio of 1:1. The highest protein efficiency ratio was found in group fed diet P/S ratio of 1:3. The protein requirement which give the best results was 25 to 35%. In experiment 2 a purified diet with constant P/S ratio (1:1) was used and the effect of dietary energy level were studied. As the energy level increased from 339 to 429 Kcal/100 g, the weight gain, protein efficiency ratio and specific growth rate were improved. High cellulose in the diet seemed to induce retardation in growth and depression of locomotive activity. Body composition of prawn muscles (crude protein, lipid, ash and moisture) at the end of experiments were determined. It was concluded that freshwater prawn required dietary protein level ranged from 25-35%. Growth improved as the energy level increased from 339 to 429 Kcal/100 g.

Key words: *Macrobrachium rosenbergii* • Protein • Energy • Freshwater prawn

INTRODUCTION

In recent years formulation of artificial diets for crustaceans has progressed remarkably. Typical examples are shown in the improvement of pellets for Kuruma prawn, *Penaeus japonicus* [1], mud crab, *Scylla serrata* [2] and for red claw, *Cherax quadricarinatus* [3]. However, in the giant freshwater prawn *Macrobrachium rosenbergii* (De Man 1879), an artificial diet has not been established precisely. Empirically to a certain degree, the culturists of the giant freshwater prawn have taken advantages of the low dietary protein requirement and high availability of dietary carbohydrates. Carbohydrates together with proteins and lipid form dietary sources of energy. They are also important in storage of dietary energy in the synthesis of chitin, steroid and fatty acids [4].

Requirement of protein has been studies in post larval and juvenile scampi like other crustaceans, the optimum dietary crude protein level ranges from 30-45% [5] under controlled laboratory conditions the utilization of dietary protein is mainly affected by its amino acid composition, level of protein intake, calorie content of the diet, digestibility of the protein, physiological state of the species, water temperature and size of prawn [4].

In regard to nutritional studies of this prawn, low dietary protein requirement and amino acids metabolism have been determined. Information on the carbohydrate nutrition however, is scantier than that of protein nutrition [6].

In the analysis of digestive enzymes of the prawn the high availability of various carbohydrate has been suggested and the ratio of dietary carbohydrate to lipid has been correlated with growth rates [7]. Although the presence of phospholipids in the diet has been shown to be indispensable for the growth of *P. japonicus*, [8] determined the non essentiality of supplemental lecithin in the diet of the giant freshwater prawn.

The aim of the study was to determine the effects of several combinations of dietary protein and starch on growth and body composition in order to get information in formulation of artificial diets for the freshwater prawn.

MATERIALS AND METHODS

An experiment was conducted at the Invertebrate Laboratory, Fish Research Station, National Institute of Oceanography and Fisheries, El-Qanater El-Khairya, Cairo Egypt, during the period from 30 June to 30 August 2007.

Experimental Aquaria and Animal: Glass aquaria (each of 60x40x30 cm.) were filled with dechlorinated tap water (50 L/aquarium and was continuously aerated by means of an electric compressor. PVC tubes and some stones were put on the bottom of each aquarium as a shelter for prawn to minimize cannibalism.

Giant freshwater prawn *M. rosenbergii* were purchased from Saft Khaled Hatchery Etay El Baroud (El-Behera Governorate). Body weight ranged from 0.24- 1.21 g, were acclimated to laboratory conditions during one week in an indoor tank and were fed with clam meat *Gafrarium pectinatum* prior to feeding experiments.

In each experiment 20 prawn were assigned in each aquarium, 10-20% of the rearing water was changed daily with fresh dechlorinated tap water. The water temperature ranged from 27-30°C with mean of 28.5±1.0°C Duplicate aquaria were set for each test group.

Diets: The purified diets were prepared taking as references the formulation by Gomez *et al.* [7]. In experiment 1, the prawn were fed with the diets containing 5 different ratios of protein /starch (P/S), namely 3:1, 1.6:1, 1:1, 1:1.6 and 1:3 (Table 1, groups 1-5); group 6 was solely fed with meat clam and was kept as the control group.

Based on the results of the experiment 1, experimental diets with constant P/S ratio (1:1) were prepared for experiment 11 with the aim of examining the optimal dietary energy level, as a means of regulating the gross energy level. In this experiment, the mortality of the prawn was wholly attributable to cannibalism. Diets used in this experiment listed in Table 2, the dietary protein and

starch which remained in a constant ratio (P/S ratio 1:1) were replaced by cellulose to regulate the gross energy level of the diet. The prawns were fed daily an amount of diet equivalent to 10% of the body weight and residual food was weighed.

Analysis: Muscles were taken from ten prawn in each group and submitted to proximate analysis. Moisture and crude ash were measured by ordinary methods. Crude protein was determined by the Kjeldahl method. Lipid content was extracted by method of Bligh and Dyer [9].

Growth Measurements: Weight gain (WG), % WG, specific growth rate (SGR), protein efficiency ratio (PER) and survival rate were calculated using the following equations:

$$WG = \text{Final body weight (g)} - \text{Initial body weight (g)}$$

$$\% WG = \text{Weight gain} / \text{Initial weight (g)} \times 100$$

$$SGR = (\ln FBW - \ln IBW) / t \times 100$$

Where:

FBW= Final body weight (g)

IBW= Initial body weight (g)

T= Time in days

PER= Weight gain (g) / protein intake (g)

Survival rate = Final number of prawn /Initial number of prawn x100

Table 1: Dietary composition of prawn in experiment 1

Ingredients	Diet numbers					
	1	2	3	4	5	6
Casein (vitamin free)	50.	40	30	20	10	--
Wheat gluten	5.	5	5	5	5	--
Gelatin	5.	5	5	5	5	--
Potato starch	24.	34	44	54	64	--
Others*1	16.	16	16	16	16	--
Clam meat	--	--	--	--	--	100
Total*2	100	100	100	100	100	100
P/S ratio*3	3:01	1.6:1	1:01	01:01.6	1:03	--
Energy*4	473	451	429	407	385	365*5
Proximate composition						
Crude protein	34.6	26.5	24.5	18.4	12.5	15.0
Crude lipid	1.3	2.0	1.7	1.8	1.4	0.6
Crude ash	3.7	3.5	3.4	3.1	3.0	2.3
Moisture	48.9	45.9	45.5	48.3	49.4	81.9

*1 CaHPO₄, 2H₂O, 5; Spirulina powder, 1; Pollack liver oil, 5; cholesterol, 1; mineral mixture 2; vitamin mixture 2

*2 Contains trimethylamine hydrochloride as attractant. *3 Protein/starch ratio.

*4 Gross energy (Kcal/100g dry matter), measured by bomb calorimetry

*5 Calculated on the basis of 4 Kcal/g of protein and 9 Kcal/g of lipid (Atwater coefficient)

Table 2: Dietary composition of prawn in experiment 11

Ingredient	Diet No.				
	7	8	9	10	11
Casein	20.0	22.5	25.0	27.5	30.0
Wheat gluten	5.0	5.0	5.0	5.0	5.0
Gelatin	5.0	5.0	5.0	5.0	5.0
Potato starch	34.0	36.5	39.0	41.5	44.0
Cellulose* ²	20.0	15.0	10.0	5.0	0.0
Others* ¹	16.0	16.0	16.0	16.0	16.0
Total	100.0	100.0	100.0	100.0	100.0
Energy	339.0	361.0	384.0	406.0	429.0
Proximate composition (%)					
Crude protein	18.4	19.9	23.6	24.4	27.3
Lipid	1.2	1.3	1.5	1.1	1.3
Crude ash	3.2	3.2	3.2	3.3	3.4
Moisture	45.3	45.9	46.2	45.9	45.9

*1 CaHPO₄. 2H₂O, 5; Spirulina powder, 1; Pollack liver oil, 5; cholesterol, 1; mineral mixture 2; vitamin mixture 2

*2 Cellulose was considered as non digestible for the calculation of energy

Energy Measurement: Dietary energy values were calculated from the entropy of combustion values measured by bomb calorimetry for each of the dietary ingredients. The energy values of clam meat were calculated on the basis of 9 Kcal/g for lipid and 4 kcal/g for protein or carbohydrate according to Atwater coefficient [7].

Statistical Analysis: The obtained data were statistically analyzed using One Way Analysis of Variance and Duncan's multiple range [10].

RESULTS

Effects of Dietary Protein Level: The feeding effects of diets with different P/S ratio are shown in Table 3, weight gain was quietly high on the diet with the P/S ratios 3:1, 1:1 and 1:3 represented by 1.2, 1.19 and 1.02 (g), respectively. While group 6 fed with clam meat as control had the lowest value in weight gain (0.86 g). The protein efficiency ratio (PER) increased with the higher starch ratio in the diet (Table 3). The specific growth rate (SGR) was the highest (2.86) for prawn fed on diet 3, while prawn fed diet 6 (control diet) represented the lowest value for SGR (1.63). Clam meat showed the lowest effects on growth, PER and SGR. The survival surpassed the 50% value in the groups 1-5 but was markedly low in the group fed with the clam meat (control group) represented by only 50%.

Table 4 shows the proximate composition of the abdominal muscle of the prawn. There were no marked differences, except for the lipid content. The group 6 represented the highest lipid level (8.9%), while the group 1 (P/S ratio of 3:1) contained only 4.5%. The lipid level tended to increase as dietary starch ratio increased, but excessive starch seemed to decrease it.

Effect of Dietary Energy Level: Table 5 show that the weight gain and SGR increased as the energy level increased. Prawn fed with the diet of low energy level (339) presented low weight gain, PER and SGR which represented by 0.48 g, 0.869 and 1.12, respectively. As the energy level increased the weight gain, PER and SGR were improved the best PER(1.39) was obtained in the diet 10 (of which gross energy was 406 Kcal (Table 5).

The survival rate was favorable in the groups fed on diets with high gross energy (groups 10 and 11). Table 6 shows the proximate composition of the muscle of prawn fed different diets (7-11), it was found that the highest crude protein% (89.1) was for group of prawn fed diet 10 while the lowest one was for group fed on diet 9. There was a random variation of lipid level among different groups ranged from 4.2% for group fed diet 8 to 7.3% for group fed on diet 9.

DISCUSSION

The current study aimed to determine the effect of diets with different protein/starch ratio and energy

Table 3: Effect of protein -starch ratio (P/S) on growth of prawn (Mean ± SE)

Diets	P/S ratios	Mean body weight (g)		*WG (g)	% weight gain	*PER	*SGR	Survival (%)
		Initial	Final					
1	3:01	0.28±0.035	1.48±0.158 ^a	1.2	428.60	2.07	2.77	65
2	1.6:1	0.24±0.032	1.18±0.161 ^a	0.94	391.67	2.46	2.65	67
3	1:01	0.26±0.047	1.45±0.228 ^a	1.19	457.70	3.11	2.86	70
4	01:01.6	0.30±0.079	1.31±0.142 ^b	1.01	336.67	3.05	2.46	58
5	1:03	0.26±0.047	1.28±0.193 ^b	1.02	450.00	5.20	2.66	60
6	Clam meat	0.52±0.089	1.38±0.187 ^b	0.86	165.38	1.84	1.63	50

*PER=Protein efficiency ratio, *SGR=Specific growth rate, * WG= Weight gain(g)

Means with different superscripts in the same column are significantly different at least at P<0.05

Table 4: Proximate composition of abdominal muscle of prawn fed different protein/starch ratios (Mean±SE)

Diets	Percent dry basis			
	Moisture	Crude ash	Crude protein	Lipid
1	77.2±5.41	5.9±0.46	89.6±6.99	4.5±0.36
2	77.8±4.89	6.2±0.69	87.4±7.22	6.4±0.52
3	77.5±6.70	6.0±0.92	87.5±6.25	6.5±0.74
4	77.4±3.99	6.1±1.02	86.6±5.89	7.3±0.48
5	78.2±5.11	5.8±0.32	88.7±5.15	5.5±0.36
6	77.6±5.29	6.3±0.61	84.8±4.14	8.9±0.65

Table 5: Effect of different energy levels on growth of prawn(Mean±SE)

Diets	Energy levels	Mean body weight (g)		WG* (g)	% weight gain	PER*	SGR*	Survival (%)
		Initial	Final					
7	339	0.50±0.11	0.98±0.19 ^a	0.48	96.00	0.869	1.12	48
8	361	1.21±0.24	3.15±0.27 ^a	1.94	160.33	1.33	1.58	20
9	384	0.77±0.09	2.23±0.01 ^b	1.46	189.60	1.32	1.76	50
10	406	0.85±0.09	2.55±0.23 ^b	1.70	200.00	1.39	1.85	58
11	429	1.08±0.16	3.22±0.02 ^a	2.14	198.00	1.15	1.82	58

*PER=Protein efficiency ratio *SGR=Specific growth rate *WG= Weight gain(g)

Means with different superscripts in the same column are significantly different at least at P

Table 6: Proximate composition of abdominal muscle of prawn fed diets with different energy levels (Mean±SE)

Diets	Percent dry basis			
	Moisture	Crude ash	Crude protein	Lipid
7	80.5±5.61	7.9±0.57	86.00±5.73	6.1±0.48
8	78.2±4.56	7.2±0.66	88.6±6.25	4.2±0.33
9	79.1±6.77	7.2±0.59	85.5±5.52	7.3±0.56
10	79.5±7.12	6.2±0.52	89.1±6.76	4.7±0.37
11	77.3±6.23	6.6±0.47	86.9±6.62	6.5±0.52

levels on growth performance of the freshwater prawn, *M. rosenbergii*. Growth as well as protein efficiency ratio could be improved by supplementation of starch in the

diet. Best weight gain and survival occurred when the prawn was fed the diet P/S ratio1:1 in this dietary regime the results from the present study indicated that a high starch ratio seemed to promote PER, suggesting that dietary protein was spared by supplementation of starch. While Balazs and Ross [11] concluded that higher protein contents produced larger prawns. High availability of dietary carbohydrate and low protein requirement of the species were reported by Cliffired and Brick [12]. Also Goda [13] concluded that higher growth, survival and feed utilization were for prawn (*Macrobrachium rosenbergii*) fed protein level 300 gKg⁻¹diet and protein efficiency ratio (PE) of 17 mg CK⁻¹g⁻¹.

In *Penaeus monodon* juveniles, protein utilization was improved by increment of the available energy level

in the diet [14]. However, [15] found that the carbohydrate energy associated with the protein in the diet played an important role in the survival rates. A minimum supply of carbohydrate is necessary to assure a metabolic energy pool that will allow the prawn to utilize the protein in the diet efficiently. However, an excess of carbohydrate energy also proves to be an obstacle to the development of the post larvae when the quantity of the ingested protein is insufficient.

Other studies with the freshwater crayfish, *Cherax tenuimanus* have indicated that a dietary protein concentration of 30% may be adequate to maintain maximum rates of growth and higher protein content diets caused insignificant changes [16]. Studies recorded by [17] with the crayfish, *Procambarus clarkii* revealed that the optimum nutrient levels for its growth are 22-26% protein, 6% lipid and 36-41% carbohydrate, with 10-13 MJ/Kg (2.4-2.9 Kcal/g) of energy content. While, [18] on crayfish, *Astacus astacus* suggested that commercial feeds for juvenile of this species should contain about 30-35% protein, 20-25% carbohydrate and not more than 10% lipid.

The sparing effect of dietary protein by replacement with cellulose was observed by Fair *et al.* [19]. Diets containing up to 30% of cellulose did not suppress the growth of the prawn juvenile (*M. rosenbergii*) but actually accelerated it. [20] reported that *M. rosenbergii* was expected to consume dietary fiber to certain extent. However, the results of the present study did not support those findings. The high proportion of cellulose which replaced protein and starch in the diets 7 and 8, resulted in physiological abnormalities in spite of evaluation of growth and survival. Hence, the study not neglect the possibility that a diet with insufficient energy might induce cannibalism. High cannibalism consequently might influence not only on survival rate, but also PER and growth. Cannibalism is inevitable in crustacean culture. Therefore, survival cannot be always considered to be directly originated from the nutritional conditions. Specifically, in the group 7 reared with low gross energy the relatively high survival reflected activity of the prawn insufficient to prey one on another. As a result, mean body weight ranged wider as the cannibalism increased among groups 8-11.

Results of the present study agree with those cited by Gomez *et al.* [7] who found that increasing the dietary energy level improved growth and feed efficiency of *M. rosenbergii* also high cellulose in the diet seemed to induce not only retardation of growth, but also depression of locomotive activity.

In conclusion, the growth performance of the freshwater prawn was improved when fed diet have protein/ starch ratios 3:1, 1:1. The protein requirement for the best prawn growth ranged from 25 -35%. Weight gain and feed efficiency increased as the energy level increased. Prawns fed the diet of low energy level presented the lowest weight gain, PER and SGR.

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