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# Effect of Dietary Protein and Feeding Time on Growth Performance and Feed Utilization of Post Larval Freshwater Prawn *Macrobrachium rosenbergii* (de Man 1879)

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Abstract: A growth trial was conducted to investigate the effect of different dietary protein and feeding times on growth performance and feed utilization of freshwater prawn Macrobrachium rosenbergii Post-Larvae (PL). The experiment was conducted in outdoor concrete pens (6 m<sup>-3</sup>) for 84 days. Post-larvae with an average weight of 20.8±0.17 mg were stocking at 30 PL m<sup>-3</sup> in each pens. Two isocaloric (~15.0 MJ DE kg<sup>-1</sup>) test diets were formulated to contain two different dietary protein 35 and 40%. The daily feeding level was divided into equal two amounts and fed twice a day at three different feeding times (9:00; 12:00 h); (9:00; 15:00 h) and (9:00; 18:00 h). The highest survival rate, weight gain and specific growth rate (p≤0.05) were observed for PL fed diet with 35% CP. The same trend was observed for PL fed with feeding time 9:00, 15:00 h. The PL fed diet containing 35% protein showed the highest (p≤0.05) protein efficiency ratio, protein productive value, fat retention, energy retention and the better feed conversion ratio. Meanwhile, the PL fed at feeding time 9:00, 18:00 h recorded the better feed conversion ratio. No significant difference was showed for the effect of dietary protein on whole body moisture and protein contents. The highest whole body contents of crude fat and gross energy were observed for prawn fed the diet with 35% CP. However, PL fed at 9:00, 15:00 h had the highest whole body protein and lower fat contents (p≤0.05). Irrespective of dietary proteins or feeding times, M. rosenbergii male recorded the better growth performance and feed utilization than female in all scenarios. The obtained findings revealed that M. rosenbergii PL fed the diet containing 35% CP at feeding time 9:00; 15:00 h is recommended to obtain optimum growth performance and feed utilization.

**Key words:** Protein level, feeding time, freshwater prawn, post larvae, growth indices, feed utilization

# INTRODUCTION

World production of freshwater prawn *Macrobrachium rosenbergii* increased from <50,000 MT (metric tons) in 1995 to >280,000 MT in 2003 (New, 2005; FAO, 2005) and has become an important part of the rice-fish or small scale carp polyculture ecosystem in many developing countries (Giap *et al.*, 2005; Hossain and Islam, 2006).

The information of optimum dietary protein of freshwater prawn Post Larvae (PL) in outdoor culture are limited and contradictory. Most of the studies have been carried out on nutritional

requirements of sub-adults and adults (Gitte and Indulkar, 2005). Hari and Kurup (2003) reported that cost of an aquaculture feed, which is directly related to protein content, appears to be the major decisive factor governing the economic feasibility of farming. The expensive protein fraction of feeds should, therefore, be optimally utilized to achieve faster growth rate and feed utilization of the cultured freshwater prawn (El-Sayed, 1997).

Previous early studies on the grow-out phase of M. rosenbergii showed an optimum dietary protein level ranged from 15 to 50%. Bartlett and Enkerlin (1983) reported that 15% dietary protein was sufficient for optimum growth of M. rosenbergii. On the contrary, D'Abramo and Reed (1988) found that 15% dietary protein resulted in a significantly poor performance, while 33-35% protein was optimal. Heinen and Mansi (1991) showed that the higher protein levels (57%) were recorded for maximum growth of M. rosenbergii PL, while Koshio et al. (1992) not found any significant differences in the growth rates, feed utilization and survival of M. rosenbergii juvenile, reared on diets containing 30-50% protein. Mitra et al. (2005) reported that diets containing 35-40% protein was optimum for M. rosenbergii PL reared in free natural food water. Recently, Teshima et al. (2006) showed that nitrogen (N) needed for their maintenance of freshwater prawn M. rosenbergii was lower than that needed for the marine prawn M. japonicus and fish such as earp, Cyprinus carpio and rainbow trout, Salmo gairdneri. However, estimated dietary protein requirements for M. rosenbergii in these above results are fall over a wide range (15-57%). These conflicting results may attribute to the variation of many factors were showed greatly affect on freshwater prawn growth performance and feed efficiency including, the variation of environmental culture conditions (e.g., controlled laboratory conditions, vs. pond culture conditions), water quality (e.g., green water ponds vs. clear water in glass aquaria or fiberglass tanks) and feed processing (pelleted vs. extruded), feeding regimes (feeding level, feeding frequency and feeding time) and protein levels and sources, therefore, further in-depth studies are required for its precise dietary protein optimization for M. rosenbergii (Tacon, 1996; Mendoza et al., 2001; Amaya et al., 2007).

The utilization of dietary protein for freshwater prawn can change depending on feeding level, feeding frequency and feeding time of day (Teshima *et al.*, 2001). Meyers (1987) reported that understanding feeding behavior of cultured species can assist in the development of feeding regimes that reduce the metabolic energy costs in feeding and encourage consumption. Since, feeding regimes (feeding frequency and feeding time) largely influence nutrient utilization of prawn, the determination of optimal feeding time of day for prawn PL is need for maximized their culture production profit. The relatively long larval period (18-35 days) and low larval survival are disadvantages to commercial culture of this species (Keysami *et al.*, 2007). In Egypt, the cost of producing nursed juveniles has emerged as a major factor influencing the profitability of the nursery operators and prawn farmers. However, further in depth information for relevant practical diets to freshwater prawns are need to be developing to sustain higher growth, survival and feed efficiency of prawn PL. Therefore, in the present study, an attempt was made to evaluate the effect of dietary protein levels and different feeding times on growth performance and feed utilization of freshwater prawns *Macrobrachium rosenbergii* post larvae rearing in outdoor cement pens.

### MATERIALS AND METHODS

### **Experimental Design and Culture Techniques**

A grow-out experiment of freshwater prawn *M. rosenbergii* PL was conducted at the Fish Farm Research Station, National Institute of Oceanography and Fisheries (NIOF), Cairo, Egypt for 84 days. In July 28, 2005 the PL of freshwater prawn were obtained from Savt-Kalid prawn hatchery, General Authority for Fish Resources Development (GAFRD), El-Bhira Governorate, Egypt. The prawns

were acclimated for two weeks in one concrete pond (8×4×1 m). During acclimation period, PL was fed on diet containing 35% CP at a level of 10% of body weight. The daily ration was divided into two equal amounts and offered two times a day (9:00, 12:00 h).

At the end of the acclimation period, PL of *M. rosenbergii* with an average weight of 20.8±0.17 mg were stocked into two cement pond (each of 40 m<sup>-3</sup>), where a two cm thick layer of clean, fine sand was spread evenly on the bottom of each ponds. Each cement pond was divided into nine equal pens by nets (6 m<sup>-3</sup>) and stocking with PL at a density of 30 PL m<sup>-3</sup>. Experiments were carried out in triplicates for each treatment. Each culture pen was provided four 30 cm long 16 mm diameter black polyvinyl chloride (PVC) pipe to minimize the cannibalism during the molting. The pond was supplied with fresh water from the Darawa Irrigation Branch, Kalubiya Governorate. The turnover rate of water was 75% day<sup>-1</sup> pond<sup>-1</sup>.

At the beginning of the experiment, three healthy groups of 40 prawns PL were randomly sampled, weighed, immediately anaesthetized with t-amyl alcohol and sacrificed for determination of initial whole body proximate composition. Also, at the termination of the feeding trial, 10 prawns were randomly selected from each pen replicate and anaesthetized with t-amyl alcohol, sacrificed and homogenized in a blender, to determine the final proximate whole body composition. Another five males and five females were obtained from each treatment replicate to be used for the final carcass analysis. The prawns were pooled for each pen replicate, oven-dried, ground and stored at 20°C for subsequent analysis.

### **Experimental Diets**

Two isocaloric (~15.0 MJ DE kg<sup>-1</sup>) test diets were formulated to contain different dietary protein 35 and 40% (Table 1). Post larvae were fed their respective diets at feeding rate of 10% from 0.2 to 1.0 g prawn, 7% for 1.0-5.0 g prawn and 5% for greater than 5.0 g prawn according to Poadas (2004). The daily ration was divided into two equal amounts and fed twice a day at three different feeding times (9:00, 12:00 h); (9:00, 15:00 h) and (9:00, 18:00 h). During the 84 days experimental periods, PL were fed 7 day's a week<sup>-1</sup>. From each experimental treatment replicate, PL was weighed collectively every two weeks, average PL weight was calculated and the amount of daily diet was adjusted accordingly.

All diets formulated mainly by fish meal, poultry by-product, soybean meal, yellow corn, wheat bran, vitamins and minerals premix. Fish meal (999 Con-Kix Fish Meal, Triple Nine Fish Protein a.m.b.a., Thyborøn, Denmark). The feed ingredients poultry by-product, soybean meal, yellow corn, wheat bran, vitamins and minerals premix were purchased from a commercial feed manufacturer (Animal Production Islamic Company (APICO), Dokki-El-Giza, Egypt). Soybean and linseed oils were purchased from local markets. The diets were processed by blending the dry ingredients into a homogenous mixture and then the mixture was passed through laboratory pellet mill in the National Institute of Oceanography and Fisheries, Cairo, Egypt (a California Pellet Mill Co., San Francisco, California).

# **Growth Indices**

Initial and final weights of individual prawns were taken by an electronic balance (±0.001 g). During each sampling, ten males and ten females from each treatment were randomly selected and weighed thereafter. The first sampling was conducted on the 30th day after stocking and at 15 days interval thereafter. The quantity of daily diet was adjusted after each sampling. Survival Rate (SR), Weight Gain (WG), Specific Growth Rate (SGR), Feed Conversion Ratio (FCR), protein efficiency ratio, Protein Productive Value (PPV), Fat Retention (FR) and Energy Retention (ER) were calculated using the following equations:

Table 1: Composition and proximate analysis of the experimental diets (% air dry-basis)

	Experimental diets (%)	
Ingredients	35	40
Fish meal	10.00	10.00
Poultry by-product meal	28.00	33.00
Soybean meal	23.00	28.00
Wheat bran	16.00	14.00
Yellow com	13.00	9.00
Soybean oil	4.00	2.00
Linseed oil	4.00	2.00
Vitamin and Mineral premix <sup>1</sup>	2.00	2.00
Proximate composition		
Dry matter (%)	8.90	9.10
Crude protein (%)	35.40	39.30
Lipid (%)	11.60	10.40
Total carbohydrate (%)	39.40	35.10
Ash (%)	4.70	6.10
Gross energy <sup>2</sup> (MJ kg <sup>-1</sup> )	19.70	19.41
Digestible energy <sup>3</sup> (MJ kg <sup>-1</sup> )	15.06	15.06
Protein: Energy ratio (mg CP kJ DE <sup>-1</sup> )	23.50	26.07

 $<sup>^1</sup>$ : Vitamin and mineral mixture each 1 kg of mixture contains: 4800 I.U. Vit A, 2400 IU cholecalciferol (vit. D), 40 g Vit E, 8 g Vit K, 4.0 g Vit B<sub>12</sub>, 4.0 g Vit B<sub>2</sub>, 6 g Vit B6, 4.0 g Pantothenic acid, 8.0 g Nicotinic acid, 400 mg Folic acid, 20 mg Biotin, 200 gm Choline, 4 g Copper, 0.4 g Iodine, 12 g Iron, 22 g Manganese, 22 g Zinc, 0.04 g Selenium, 1.2 mg folic acid; 12 mg niacin; 26 mg d-calcium pantothenate; 6 mg pyridoxine HCl; 7.2 mg riboflavin; 1.2 mg thiamin HCl; 3077 mg sodium chloride (NaCl, 39% Na, 61% Cl); 65mg ferrous sulfate (FeSO<sub>4</sub>-7H<sub>2</sub>O, 20% Fe); 89 mg manganese sulfate (MnSO<sub>4</sub>, 36% Mn); 150 mg zinc sulfate (ZnSO<sub>4</sub>-7H<sub>2</sub>O, 40% Zn); 28 mg copper sulfate (CuSO<sub>4</sub>-5H<sub>2</sub>O, 25% Cu); 11: mg potassium iodide (KI, 24% K, 76% I); 1000 mg Celite AW521 (acid-washed diatomaceous earth silica).  $^2$ : Calculated using gross calorific values of 23.62, 17.15 and 39.52 kJ g $^{-1}$  for protein, carbohydrate and fat, respectively according to Brett (1973).

 $SR = Final No. of prawn larvae/Initial number of prawn larvae <math>\times 100$ 

WG = Final body weight (g) - Initial body weight (g)

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

# Where:

FBW = Final Body Weight (g)

IBW = Initial Body Weight (g)

In = Natural logarithmic

t = Time in days

FCR = Feed intake (g)/Weight gain (g)

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

 $PPV = Protein gain (g)/Protein intake (g) \times 100$ 

FR = Fat gain (g)/Fat intake (g)  $\times$  100

ER = Energy gain (kJ)/Energy intake (kJ) × 100

### **Proximate Analysis**

Diet, ingredients and whole body composition were determined according to proceeding of AOAC (1995). Moisture was determined after drying the samples in an oven (105°C) for 24 h. Ash by

 $<sup>^3</sup>$ : DE was calculated using values 20.91, 8.36 and 37.63 kJ  $g^{-1}$  for protein, carbohydrate and fat, respectively according to Das et al. (1996)

incineration at 550°C for 12 h. Crude protein was determined by micro-Kjeldhal method, N%  $\times$ 6.25 (using Kjeltech autoanalyzer, Model 1030, Tecator, Höganäs, Sweden) and crude fat by soxhlet extraction with diethyl ether (40-60°C). Gross energy content of diet and whole body samples was calculated according to the gross caloric values by applying the factor 23.63, 39.52 and 17.15 kJ g<sup>-1</sup> of crude protein, crude fat and total carbohydrate, respectively (Brett, 1973). Dietary Digestible Energy (DE) was estimated using values of 20.91, 37.64 and 8.36 kJ g<sup>-1</sup> for crude protein, crude fat and carbohydrate, respectively according to Das *et al.* (1996).

# Statistical Analysis

The data were statically analyzed by ANOVA using MSTAT-C version 4 software (MSTAT-C, 1987) in a factorial design manner ( $2\times3$ ). Duncan's multiple range test was used to compare differences between treatment means when significant F values were observed (Duncan, 1955), at p≤0.05 level. All percentage data were arc-sign transformed prior to analysis (Zar, 1984). However, data are presented untransformed to facilitate comparisons.

### **RESULTS**

All conditions of the experimental evaluation in the present study were apparently satisfactory and fell under the optimal standards defined for nutritional evaluations in freshwater prawn, M rosenbergii PL. There was no significant difference between treatments in measured water quality variables. Overall means±SD for water quality variables were as follows: temperature:  $28.2\pm0.3^{\circ}$ C; dissolved oxygen:  $5.8\pm0.2$  mg L<sup>-1</sup>; pH:  $8.9\pm0.1$ ; ammonia  $0.28\pm0.1$  mg L<sup>-1</sup> and alkalinity  $1.77\pm5.1$  mg L<sup>-1</sup>.

The higher survival rates were observed for *M. rosenbergii* PL fed the diet containing 35% CP and feeding time at 9.00, 15.00 h (Table 2). Growth in terms of Final Body Weight (FBW), Feed Intake (FI), Weight Gain (WG), Specific Growth Rate (SGR) and feed utilization in terms of Feed Conversion Ratio (FCR), Protein Efficiency Ratio (PER), Protein Productive Value (PPV), Fat Retention (FR) and Energy Retention (ER) are shown in Table 2. Concerning the two different dietary protein levels, the highest values of FBW, WG and SGR were observed for prawn fed diet with 35% CP ( $p \le 0.05$ ). The same trend was observed for PL fed diet at feeding time (9.00, 15.00 h), while, the lower value was recorded at feeding time 9.00, 18.00 h. The PL fed diet containing 35% CP recording the better FCR, meanwhile prawn fed at feeding time 9.00, 18.00 h showed significantly better FCR. The highest PER, PPV, FR and ER were recorded for the PL fed 35% CP diet. A same tendency was observed for PER, FR and ER for PL fed with 9:00, 18:00 h feeding time. Whereas, the highest value of PPV was observed for PL fed the diet at feeding time 9:00, 15:00 h.

Table 2: Effect of different dietary protein and feeding time irrespective each other on growth performance and feed utilization of Freshwater prawn Macrobrachium rosenbergii post larvae

	Dietary p	rotein levels		Different fee			
Growth							
parameters	35% CP 40% CP		$MSE\pm$	9:00, 12:00	9:00, 15:00	9:00, 18:00	MSE±
Initial body weight (mg prawn <sup>-1</sup> )	20.80	20.80	0.17	20.80	20.80	20.80	0.17
Final body weight (g prawn <sup>-1</sup> )	7.25°	6.70 <sup>b</sup>	0.45	7.08 <sup>b</sup>	$7.19^{a}$	6.67°	0.56
Survival rate (%)	76.00ª	72.50°	0.50	68.50 <sup>b</sup>	71.50a	$70.00^{ab}$	0.75
Feed intake (g prawn <sup>-1</sup> )	16.00	17.58	1.65	17.16°	17.49ª	15.73°	1.42
Weight gain (g prawn <sup>-1</sup> )	7.23ª	6.68 <sup>b</sup>	0.65	7.06⁰	7.17ª	6.65°	0.12
Specific growth rate (% day)	6.97ª	6.88 <sup>b</sup>	0.55	6.94 <sup>ab</sup>	6.96ª	6.87°	0.18
Feed conversion ratio	2.21 <sup>b</sup>	2.63ª	0.54	2.43ª	2.44ª	2.37°	0.20
Protein efficiency ratio	$1.40^{a}$	$1.18^{b}$	0.30	$1.22^{b}$	$1.20^{\rm b}$	$1.24^{a}$	0.01
Protein productive value	19.09ª	15.81 <sup>b</sup>	2.66	$14.46^{\circ}$	18.12ª	17.07°	1.12
Fat retention	25.03ª	18.56°	2.66	22.26°	19.28°	23.57ª	2.32
Energy retention	18.56ª	10.78 <sup>b</sup>	1.25	11.73 <sup>b</sup>	12.23 <sup>b</sup>	13.01ª	1.36

Mean in the same row sharing the same superscript are not significantly different (p≤0.05)

Irrespective of different dietary protein or feeding time, *M. rosenbergii* male recorded the highest values of growth indices in terms FBW, WG, SGR and FCR than female in all scenarios (Table 3). The same trend was observed for feed utilization induces.

Proximate body compositions of *M. rosenbergii* PL are presented in Table 4. No significant difference (p≤0.05) was observed in whole body moisture and protein contents between the dietary protein treatments. However, higher body fat and gross energy contents were observed when PL fed the diet containing 35% CP. Meanwhile, prawn fed at feeding time 9:00, 15:00 h showed the highest whole body protein and lower fat contents.

Irrespective of dietary feeding time, PL female fed the diet containing 35% CP recorded the highest (p<0.05) values for whole body fat, while the PL male fed 40% CP diet showed the lowest significant (p<0.05) values of whole body fat content (Table 5). The same trend was observed for the whole body gross energy (23.92 and 20.33 kJ g<sup>-1</sup>). However, the highest (p<0.05) value of whole

Table 3: Effect of different dietary protein and feeding time irrespective each other on growth performance and feed utilization of male and female Freshwater prawn Macrobrachium rosenbergii post larvae

	Di etary	protein l	evels			Different feeding times (h)						
	35% CP 40% CP					9:00, 12:00		9:00, 15:00		9:00, 18:00		
Growth	rowth											
parameters	ď	φ	ď.	φ	MSE ±	ਂ	\$	ਂ ਂ	₽	්	φ	MSE±
Initial body weight												
(mg prawn <sup>-1</sup> )	20.80	20.80	20.80	20.80	0.17	20.80	20.80	20.80	20.80	20.80	20.80	0.17
Final body weight												
(g prawn <sup>-1</sup> )	8.62°	5.89°	7.85 <sup>b</sup>	5.54°	1.65	8.48*	5.67 <sup>b</sup>	8.56*	5.80°	8.67ª	5.68 <sup>b</sup>	1.88
Feed intake (g prawn <sup>-1</sup> )	18.06	$13.00^{\circ}$	18.99°	15.05°	1.65	19.19°	15.27°	19.03°	15.90°	17.52°	13.29°	1.33
Weight gain (g prawn-1)	8.60°	5.87 <sup>b</sup>	7.83°	5.52 <sup>b</sup>	1.65	8.46*	5.65 <sup>b</sup>	8.54*	5.78 <sup>b</sup>	8.65	5.66°	1.85
Specific growth rate												
(% day)	7.18°	6.72 <sup>b</sup>	7.06°	6.65°	0.11	7.16*	6.68 <sup>b</sup>	7.17*	$6.70^{\circ}$	7.18°	$6.68^{\circ}$	0.21
Feed conversion ratio	$2.10^{b}$	2.22 <sup>b</sup>	2.43°	2.73°	0.31	2.27 <sup>b</sup>	$2.70^{\text{ab}}$	$2.23^{\text{ab}}$	2.75°	2.03°	2.35 <sup>sb</sup>	0.69
Protein efficiency ratio	1.48°	1.40°	$1.16^{bc}$	1.03°	0.21	1.27 <sup>b</sup>	1.05°	1.30 ab	1.04°	1.42°	1.21 <sup>b</sup>	0.11

Mean in the same row sharing the same superscript are not significantly different (p≤0.05)

Table 4: Effect of different dietary protein and feeding time irrespective each other on chemical body composition of Freshwater prawn Macrobrachium rosenbergii post larvae

	Dietary p	rotein levels		Different feeding times (h)					
Growth									
parameters	35% CP	40% CP	MSE ±	9:00, 12:00	9:00, 15:00	9:00, 18:00	$MSE \pm$		
Moisture (%)	75.47	76.26	1.88	75.50	75.83	75.006	1.00		
Crude protein (%)	13.62	13.47	0.65	11.92°	$15.06^{a}$	12.82 <sup>b</sup>	0.60		
Crude fat (%)	6.02ª	$4.62^{b}$	0.45	5.67ª	4.46⁰	5.60°	0.80		
Ash (%)	4.89 <sup>b</sup>	5.66°	0.70	6.71ª	4.65°	6.52 <sup>b</sup>	0.65		
Gross energy (kJ g <sup>-1</sup> )	22.54ª	$21.04^{b}$	1.80	21.45	22.00	21.91	1.05		

Mean in the same row sharing the same superscript are not significantly different (p  $\leq$  0.05)

Table 5: Effect of different dietary protein and feeding time irrespective each other on chemical body composition of male and female Freshwater prawn Macrobrachium rosenbergii post larvae

	Dietary	protein le	vels			Differen	t feeding	times (h)							
	35% CI	)	40% C	P		9:00, 12:00 9:00, 15:00 9:00, 18:0		3:00	0						
Growth															
parameters	ď	φ	ď	φ	MSE ±	E σ'	Ŷ	ਂ	Ŷ	ਂ	φ.	MSE±			
Moisture (%)	75.00°	76.60°	75.91 <sup>b</sup>	76.62°	1.99	76.77*	76.02 <sup>b</sup>	$76.05^{\rm b}$	75.61 <sup>b</sup>	73.43°	76.69°	1.22			
Crude protein (%)	13.78°	13.65 <sup>b</sup>	13.26 <sup>b</sup>	13.51 <sup>b</sup>	0.65	11.76*	12.08d	15.13°	14.98⁵	13.68°	13.70°	0.88			
Crude fat (%)	5.16 <sup>b</sup>	6.61*	4.44 <sup>b</sup>	4.75 <sup>b</sup>	0.73	4.89 <sup>b</sup>	6.48°	$3.72^{\circ}$	5.22 <sup>b</sup>	5.90⁵	5.29 <sup>b</sup>	0.85			
Ash (%)	6.06 <sup>sb</sup>	4.14°	6.36°	5.12 <sup>b</sup>	0.93	$6.58^{ab}$	5.42 <sup>b</sup>	$5.10^{bc}$	4.19 d	6.99°	4.32 <sup>cd</sup>	0.64			
Gross energy (kJ g-1)	21.16 <sup>b</sup>	23.92°	20.32°	21.70 <sup>b</sup>	2.13	20.28°	22.58ab	$21.08^{b}$	22.96°	20.95 <sup>b</sup>	22.88*	1.80			

Mean in the same row sharing the same superscript are not significantly different (p≤0.05)

body protein was observed for PL male fed at feeding time 9:00, 15:00 h, associated with the lower whole body fat content. The PL female fed at feeding time 9:00, 12:00 h was recorded the highest values of whole body fat.

### DISCUSSION

Over the duration of the present study, all water quality indices represented suitable conditions for prawn culture (Boyd and Zimmerman, 2000). The higher survival rates shown in the present study for PL fed the diet containing 35% protein may be attributed to the fulfillment of dietary protein requirement. Moreover, the PVC pipe used in the present study may have a successful method to help the prawn to overcome molting stress and to avoid cannibalism (Mariappan and Balasundaram, 2004). Survival rates of prawns ranged from 68 to 78%, which similar to that reported by Ali *et al.* (2003) and Hossain and Islam (2006). Molting of decapod crustaceans is affected by external environmental factors such as temperature, salinity, light intensity, pollutants and also by internal factors associate with nutritional state and hormones (Cheng *et al.*, 2003). However, un-suitable feeding diets have been mentioned as a major reason for increasing mortality during the first molt stress and late stages (Yao *et al.*, 2006). Therefore, it is necessary to develop relevant feeding diets suitable for grow-out of prawns farming.

The results of the present study showed that 35% protein diet was recommended to achieve the optimum growth indices of freshwater prawn *M. rosenbergii* PL. The present results agree with the findings of Teshima *et al.* (2006), who reported that protein sparing effect of high protein diet (>35% CP) shows an increase in fat content in *M. rosenbergii* compare to the prawn fed diets of 35% CP. This is indicative that the increase in dietary protein levels exceeding the minimum requirement results in the loss of expensive protein in *M. rosenbergii* diets and accumulation of fats. Nevertheless, the protein requirement of the freshwater prawn *M. rosenbergii* PL is considered to be lower than other prawn species and this may be due to less protein requirement for maintenance compared to carnivorous marine prawn species (Koshio *et al.*, 1992; D'Abramo and Sheen, 1994). Moreover, in present study, higher growth of male prawn was observed in all treatments irrespective of crude protein content or feeding time. This is similar to the findings of Nair *et al.* (2006).

Higher FCR registered at dietary containing 40% protein indicate that protein was used as energy when the supply exceeded the requirement for prawn growth. Higher FCR was also observed by Cortes-Jacinto *et al.* (2003) for juvenile freshwater crayfish, *Cherax quadricarinatus* fed high protein diets. The FCR values in the present study are similar to the values reported by Hossain and Islam (2006). New (1988) reported that for *M. rosenbergii*, an FCR of 1.8-3.0 may be acceptable for dry diets. Moreover, the mean values of FCR in the present study are better than those obtained by Ashmore *et al.* (1985) and Hari and Kurup (2003). This difference may attribute to the substantial contribution of certain type of algae present in outdoor pond to the experimental animal's nutrition. In the pond culture study, natural food items in the pond may have satisfied a part of the protein requirement of omnivorous *M. rosenbergii* PL. However, it is difficult to quantify the contribution of natural foods to the diet of prawn, as this is beyond the scope of the study.

Currently, much attention has been conducted for the influence of feeding schedule in terms feeding levels, frequency and feeding time on fish and prawn growth indices as an important component of successful larval culture (D'Abramo, 2002). Wilkinson (2003) reported that the time of the day that feed is delivered associate with feeding organisms behavior that could be affect growth performance. Several authors reported that there are certain times of the day at which feeding results in better growth and feed efficiency in some fish species (Webster *et al.*, 1992; Boujard and Leatherland, 1992; Velasco *et al.*, 1999; Petkam and Moodie, 2000). In contrast, to our knowledge, no such data is available for freshwater prawn *M. rosenbergii*. However, in the case of crustaceans, it is

generally difficult to measure appropriate feed intake in feeding experiments because of the slow feeding habit of these organisms resulting in the leaching of some nutrients. D'Abramo (2002) showed that freshwater prawn would feed better during their preferred phase according to their feeding behavior. Teshima and Kanazawa (1987) reported that for the prawn *M. rosenbergii*, about 36-40% of given diets was found to be lost to the water and not ingested, compared to marine prawn *M. japonicus*. Therefore, when prawn was fed at preferred time an improvement was showed in feed conversion ratio and growth indices (Boujard and Leatherland, 1992).

The results of present study shows that feeding time has a great effect on growth performance and feed utilization of freshwater prawn M. rosenbergii PL. The highest ( $p \le 0.05$ ) growth indices was observed for M. rosenbergii PL fed the diet at feeding time 9.00, 15.00 h compare to the PL fed at 9.00, 12.00 h or 9.00, 18.00 h. The result agree with the finding of Robertson  $et\ al.$  (1993) who reported that small shrimp metabolize their food faster than large shrimp and generally require more feedings per day. Post-larval shrimp are not able to store much feed in their guts. Ideally, post-larvae should be fed every 3-5 h. Longer intervals between feedings may result in heavy losses due to cannibalism.

In the present study, protein and energy retention seemed low compared to the findings in fishes e.g., rainbow trout 40-60% (Bolliet *et al.*, 2000). However, prawns like any crustacean release cast at each molt, which can represent an energetic expenditure of 26% of the overall inter-molt energy gain (Read and Caulton, 1980). Cuzon *et al.* (2004) reported that protein requirement is probably not as highly correlated with protein accretion compared to vertebrates due to chitin synthesis, chitin is a polysaccharide containing nitrogen equivalent to 43% crude protein.

In conclusion, the obtained findings revealed that *M. rosenbergii* PL fed the diet containing 35% CP at feeding time 9:00, 15:00 h is recommended to obtain optimum growth performance and feed utilization. Generally, *M. rosenbergii* male obtain higher growth indices than female either different dietary protein or feeding times.

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