Journal of Ecobiotechnology 2011, 3(1): 14-23
ISSN: 2077-0464
www.scholarjournals.org

Journal of Ecobiotechnology
www.journal-ecobiotechnology.com

Growth Performance of the Monsoon River Prawn *Macrobrachium malcolmsonii* on Formulated Feeds with Combinations of Pulses and Cereals along with Groundnut Oilcake and Soya Meal

P. Saravana Bhavan*, S. Radhakrishnan and C. Seenivasan

Department of Zoology, Bharathiar University, Coimbatore–641046, Tamilnadu, India

Article Info

Article History

Received : 19-11-2010
Revised : 29-12-2010
Accepted : 02-01-2011

*Corresponding Author

Tel : +91-4222428495
Fax : +91-4222425706

Email:
bhavan@buc.edu.in
psbhavan67@gmail.com
bhavanps1967@yahoo.in

Abstract

This study was conducted to categorize the growth performance of feeds formulated with combinations of pulses and cereals (horse gram and ragi, green gram and pearl millet, and cow gram and wheat) along with groundnut oilcake and soya meal on the post larvae (PL) of the monsoon river prawn, *Macrobrachium malcolmsonii*. This is to promote the inland aquaculture of this economically important species. Two types of feeds with different proportions of pulse and cereal (type-A: 25% pulse + 25% cereal; type-B: 40% pulse + 10% cereal) were prepared in each combination along with equal proportion of groundnut oil cake and soya meal (20% each). Tapioca flour (5%) and egg albumin (4%) were used as binding agents. Vitamin B-complex (1%) was also mixed. These feeds were fed to *M. malcolmsonii* PL for a period of 45 days. The efficacy of these feeds on growth performance and biochemical constituents were assessed. Commercially available standard Scampi feed was served as control. The overall influence of these feeds on growth, and concentrations of total protein, amino acid, carbohydrate and lipid of PL were found to be the best in scampi feed followed by the feed rich in pulse (type-B) and the feed with equal proportion of pulses and cereals (type-A). Among three combinations of pulses and cereals were tested, the feed formulated with cow gram and wheat showed the best overall performance, followed by green gram and pearl millet, and horse gram and ragi. All the results were confirmed through paired sample t-test, DMRT, and two-way ANOVA. The survival rate was found to be better in formulated feeds fed PL when compared with control. It is assumed that the additive/ preservative added in the commercially available scampi feed may be the reason for the lower survival rate recorded. Moreover, the commercially available scampi feed is not always affordable to small farmers. Therefore, the farm made feeds with locally available commodities of pulses and cereals can be prepared and used in a sustainable manner for healthy promotion of *Macrobrachium* culture.

Key Words: *Macrobrachium malcolmsonii*, Growth, Pulses, Cereals, Protein, Amino acids, Carbohydrate, Lipid

Introduction

Aquaculture is one of the fastest growing food producing sectors. It is recognized as a viable and profitable enterprise will presumably continue to grow and supply an increasingly larger quantum of nutritious products. This development is real and visible in support of rural livelihoods, improved food security and export earnings [1]. Prawns constitute an important and nutritious delicacy for human consumption and, hence, there is a universal stress on the need for its culture fishery [2]. The global annual production of freshwater prawns (excluding crayfish and crabs) in 2003 was about 280,000 tons, of which China produced some 180,000 tons, followed by India and Thailand with some 35,000 tons each [3]. Next to the giant freshwater prawn *Macrobrachium rosenbergii* (de Man), the monsoon river prawn *Macrobrachium malcolmsonii* (H. Milne Edwards) is a potential species for culture [4, 5]. The commercial farming of this species is still in infancy due to non-availability of seed in adequate quantities and the viable culture technology. However, in a recent report it has been stated that under monoculture system production levels of 750-1,500 kg prawns/ ha/ 8 months are achieved. Further, it is a compatible species for polyculture along with Indian major carps and Chinese carps, which may yield 400 kg prawns and 3000 kg carps/ha/year [5] in a culture system under which water quality, prawn growth and health is generally not monitored. In a semi intensive system with a stocking density of 30,000-40,000 prawns/ ha, under which fertilization and supply of a balanced feed ration are possible, the water quality, prawn health and growth rate are monitored, and the system is made free from predators and competitors. Thus, the production was achieved in the range of 500-1000 kg/ ha/ year [5]. The following range of water quality is prescribed as favoured for the culture of *M. malcolmsonii*: temperature, 26-32^{o}C; transparency, 30-60 cm; pH, 7.0-8.5; dissolved oxygen, 5 mg/l; free CO_2, 8 mg/l; hardness, 100-50 mg/l; alkalinity 80-150mg/l phosphorus, 80-150 mg/l; NH_4-N, 0.02-0.20 mg/l; Calcium, 30-80 mg/l; phosphorus, 0.01-0.90 mg/l; nitrogen, 0.05-90.5 [5]. Since the seed requirement for the commercial farming of this species is not met from the natural resources, large-scale seed production under controlled conditions for year-round supply is extremely important. The technologies of large-scale seed production and grow-out culture have led to increased attention of the farmers and entrepreneurs for diversification of their culture practice. In south India, the economically important *M. malcolmsonii* is found in abundance and constitutes a major fishery in the Cauvery River [6]. Culture of *M. malcolmsonii* depends on the use of a well-balanced nutritional and economic diet [7].

The route to the economic farming of freshwater prawns lies in determining the precise nutritional requirements at all stages of development and the development of an inexpensive artificial diet from readily available foodstuffs. A formulated diet can be well balanced and contain all the essential dietary nutrients, it produce desired results. The true nutritive value of a formulated diet therefore ultimately depends on the bioavailability of the ingredients, and not purely on diet composition [8]. Artificial diets have been increasingly used in semi-intensive and intensive late nursery and on-growing phases to achieve fast growth and high survival [9, 10]. However, commercially available artificial feeds are highly expensive and are not always affordable to small farmers. The expensive protein sources can be limited by including adequate level of carbohydrate, which in turn enhances the protein sparing effect of a particular diet on growth [11-13] and decreases the accumulation of nitrogen waste in culture ponds [14]. Soybean meal is one of the most promising alternatives because of its availability, reasonable price, high digestibility and amino acid profile [15]. There are many other chiefly available plant materials have to be checked their suitability for protein and carbohydrate sources. The increasing pressure on global fish meal stocks has generated interest on uses of plant products [16].

Most grains exerted some functional properties, which is unknown as to whether the protein or the carbohydrate component of the product possesses this property [17-19]. In the present study, basal nutrients (protein, carbohydrate and lipid) for the growth of *M. malcolmsonii* PL were fully relied on carbohydrate rich basal ingredients, such as pulses (horse gram, green gram and cow gram) and cereals (ragi, pearl millet and wheat), groundnut oilcake and soya meal used as regular protein rich ingredients, and tapioca flour and egg albumin were used as binding agents. With these ingredients no additional lipid source was added. This work was carried out to categorize the growth promoting potential of these locally available commodities with an aim for promoting the inland aquaculture of *M. malcolmsonii*.

Materials and Methods

The post larvae/ early juveniles of *M. malcolmsonii* were collected from Lower Anicut of the Cauvery River (Kumbakonam, Tanjavoor District, Tamilnadu, India). They were safely brought to the laboratory in well-oxygenated plastic bags. They were stocked in a large cement tank (1000 L capacity) and acclimatized for two weeks in ground water (pH, 7; total dissolved solids, 0.9 g L^{-1}; dissolved oxygen, 7.2 mg L^{-1}; BOD, 30.0 mg L^{-1}; COD, 125.0 mg L^{-1}; ammonia, 0.028 mg L^{-1}). During which they were fed *ad libitum* with boiled egg albumin, beef liver, commercially available scampi feed and rice alternatively twice a day, and the medium was adequately aerated. On daily basis three fourth of the water was renewed by siphoning method causing minimum disturbance to the prawns. The unfed feed, excreta, and exuvia if any were removed.

In this study, eight groups of 60 PL each (length: 0.8 -1.0 cm; body mass: 0.08-0.10 g) was taken. Each group was equally divided and housed in three aquaria of 20 L capacity. One group served as initial control. After initial morphometric measurement from randomly taken ten individual the entire PL in this group were sacrificed to estimate the initial proximate composition of total protein [20], amino acid [21], carbohydrate [22], lipid [23], and ash [24]. The muscle tissue was pooled together and each initial biochemical parameter was analyzed three times.

The remaining seven groups (each in three aquaria represents replicates) were taken for feeding trial. One group served as control and remaining six groups were served as experiments. The control group was fed with standard scampi feed and the experimental groups were fed with six different feeds formulated by using combination of pulses and cereals alongwith groundnut oil cake, soya meal, tapioca flour and egg albumin (Table-1).

Table 1: Formulated feeds and their protein, carbohydrate and lipid ratio

Basal Ingredients	Feed -A (pulse-25% + cereal-25%) + (GOK-20% + SM-20%) + (EA-4% + tapioca-5%)			Feed –B (pulse-40% + cereal-10%) + (GOK-20% + SM-20%) + (EA-4% + tapioca-5%)		
	Protein %	Carbohydrate %	Lipid %	Protein %	Carbohydrate %	Lipid %
Horse gram + Ragi;	5.5+1.7=7.2	14.2+18.0=32.2	0.12+0.25=0.37	8.8+0.7=9.5	22.8+7.2=30.0	0.2+0.1=0.3
Groundnut oilcake + Soya meal;	9.4+11.8=21.2	4.0+2.0=6.0	1.6+2.8=4.4	9.4+11.8=21.2	4.0+2.0=6.0	1.6+2.8=4.4
Egg albumin + Tapioca	0.4+0.0	0.0+4.0	Nil	0.4+0.0	0.0+4.0	Nil
Total	*28.8*	*42.2*	*4.7*	*31.1*	*40.0*	*4.7*

Green gram + Pearl millet;	6.0+3.0=9.0	15.7+16.7=32.4	0.25+1.25=1.50	9.6+1.2=10.8	25.2+6.7=31.9	0.4+0.5=0.9
Groundnut oilcake + Soya meal;	9.4+11.8=21.2	4.0+2.0=6.0	1.6+2.8=4.4	9.4+11.8=21.2	4.0+2.0=6.0	1.6+2.8=4.4
Egg albumin + Tapioca	0.4+0.0	0.0+4.0	Nil	0.4+0.0	0.0+4.0	Nil
Total	*30.6*	*42.4*	*5.9*	*32.4*	*41.5*	*5.3*
Cow gram + Wheat;	6.2+3.0=9.2	14.0+17.7=31.7	0.25+0.5 =0.75	10.0+1.2=11.2	22.4+7.1=29.5	0.4+0.2=0.6
Groundnut oilcake + Soya meal;	9.4+11.8=21.2	4.0+2.0=6.0	1.6+2.8=4.4	9.4+11.8=21.2	4.0+2.0=6.0	1.6+2.8=4.4
Egg albumin + Tapioca	0.4+0.0	0.0+4.0	Nil	0.4+0.0	0.0+4.0	Nil
Total	*30.8*	*41.7*	*5.1*	*32.8*	*39.5*	*5.0*

GOK, groundnut oilcake; SM, soya meal; EA, egg albumin
Protein: 28.8 - 30.8 (Feed – A); 31.1 - 32.8 (Feed – B); 29-33%
Carbohydrate: 41.7- 42.2 (Feed – A); 39.5-41.5(Feed – B); 42-40%
Lipid: 4.7 - 5.9 (Feed – A); 4.7 - 5.3 (Feed – B), 6-5%

The feeding trial was conducted for a period of 45 days. Three different combinations of pulses and cereals (horse gram + ragi; green gram + pearl millet; cow gram + wheat) each with two different ratios (pulse: 25% & 40%; cereal: 25% & 10%) along with groundnut oil cake (20%), soy meal (20%), egg albumin (4 ml) and tapioca flour (5%) were taken. The pulses and cereals are used as major carbohydrate sources and minor protein sources. The groundnut oil cake and soy meal were used as major protein sources and minor carbohydrate sources. The egg albumin and tapioca flour were used as binding agents.

A mixture of these ingredients except egg albumin was stream cooked for 15 minutes and cooled in room temperature, then egg albumin was added. Vitamin B-complex (1%) was also mixed. A semi solid paste was prepared and then made to form pellets (3-5 mm) using a manual pelletizer. These pellets were sun dried for 48 hrs. Water stability of the feed was assessed and the leaching was not exceeded 10% loss of dry matter per eight hours. Exactly 1 g pellet was taken in a glass beaker and soaked in known quantity of water for eight hours. After soaking the samples were drained on pre-washed Whatman filter paper and dried in an oven at 40°C for 24 h. The leaching rate was found to be less than 100 mg.

On the final day of feeding trial, morphometric data were taken. The nutritional indices, such as survival rate, weight gain, biomass index, specific growth and condition factor were calculated [25, 12].

$$\text{Survival Rate (SR)} = \frac{\text{No. of live prawns}}{\text{No. of prawns introduced}} \times 100$$

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

$$\text{Biomass Index (BI)} = \frac{\text{Final weight (g)} - \text{Initial weight (g)}}{\text{Initial weight (g)}} \times 100$$

$$\text{Specific Growth Rate (SGR)} = \frac{\log \text{ of Final weight (g)} - \log \text{ of Initial weight (g)}}{\text{No. of days}} \times 100$$

$$\text{Condition Factor (CF)} = \frac{\text{Final weight (g)}}{\text{Final length}^3 \text{ (cm)}} \times 100$$

Fifteen prawns from each aquarium were sacrificed (15x3=45 from each group) for estimating the proximate composition of total protein, amino acid, carbohydrate, lipid and ash. From each aquarium tissue from a group of three PL was pooled to constitute a single observation of each parameter and five such pooled sampling was made. Thus, three such observations were made for each parameter (3x5=15x3=45). Data between control and experiments and between experiments were subjected to statistical analysis: paired samples t-test, two-way ANOVA and DMRT were adopted by using SPSS, version-13.0 of IBM software.

Results and Discussion

Growth and Survival

Table 2 represents the morphometric data and nutritional indices in *M. malcolmsonii* PL fed with formulated feeds prepared from three different combinations of pulses and cereals. The initial body length and weight of PL were recorded to be 1.03 cm and 0.092 g respectively. Significant increase in weight gain was recorded when comparing the final weight with initial weight ($P<0.002$-0.013). The increase in final weight was found to be higher in the scampi feed (control) fed PL followed by feeds contained higher pulses ratio (feed-B), and feeds prepared with equal proportions of cereals and pulses (feed-A). Therefore, the weight gain and specific growth rate recorded were in the order of control > feed-B > feed-A. Among three types of feeds formulated, the feed contained cow gram and wheat showed the best performance, the feed prepared with green gram and pearl millet produced better result, and the feed with horse gram and ragi yielded good growth. These were further confirmed through comparison of DMRT mean significance (Table 2a). The values calculated for condition factor was the least in the best feed that was formulated with cow gram and wheat, less in better feed (green gram + pearl millet) and the low in good feed (horse gram + ragi). These were further confirmed through comparison of DMRT mean significance (Table 2a). However, control feed showed very least condition factor, which reflects its supreme quality when compared with experimental feeds. In two-way ANOVA, the F-values obtained for weight gain, specific growth rate and condition factor were found to be significant (either at 5% or 1% level) when compared between treatments, between feed types and between combinations of pulses and cereals (Table 2a). However, the F-values obtained for these parameters were found to be not significant when different combinations of pulses and cereals were compared with feed types (Table 2a).

The most important observation is the survival rate, which was found to be higher in formulated feeds when compared with control. Among formulated feeds, the elevated survival rate was the highest with cow gram and wheat (94%), followed by the feed prepared with horse gram and ragi, and with green gram and pearl millet ($P<0.074$-0.478). This was also confirmed through two-way ANOVA, the F-values obtained were found to be significant ($P<1$), when compared between treatments, between different combinations of pulses and cereals, and between feed types and different combinations of pulses and cereals (Table 2a). However, no difference was recorded in survival rate between feed types A and B (Table 2). This was confirmed through statistically insignificant F-value obtained (Table 2a).

Growth is an index of water quality and nutrients provided. As far as freshwater prawn culture is concerned there are many factors relates the growth and feeding activity [26], which include a functional digestive system to efficiently utilize the nutrients present in the food offered [11] and the physiological conditions, and the rearing environment [8]. Growth of prawn is normally very fast during the early life and slows down during adult; the survival rates are also very high during the early life and fell subsequently [27]. In the present study, the overall performance of growth and survival were found to be appreciably good in PL fed with formulated feeds as seen in control feed fed PL. Therefore, the selected pulses (horse gram, green gram and cow gram) and cereals (ragi, pearl millet and wheat) can be included in feed formulation for *Macrobrachium*.

Table 2: Morphometric data and nutritional indices of *M. malcolmsonii* PL fed with different formulated feeds

Parameters		Prepared Feeds	Control (Scampi feed)	Feed - A (pulse-25% + cereal-25%) + (GOK-20% + SM-20%) + (EA-4% + tapioca-5%)	Feed - B (pulse-40% + cereal-10%) + (GOK-20% + SM-20%) + (EA-4% + tapioca-5%)	Rank
Morphometric Data	Initial length (cm)	Control (Scampi feed)	1.03 ± 0.15	1.03 ± 0.15	1.03 ± 0.15	
	Final length (cm)	Horse gram + Ragi	2.23 ± 0.20	1.96 ± 0.15 (0.011)	2.10 ± 0.10 (0.046)	III
		Green gram + Pearl millet	2.23 ± 0.20	1.93± 0.15 (0.009)	2.13 ± 0.20 (*)	II
		Cow gram + Wheat	2.23 ± 0.20	2.03 ± 0.15 (0.020)	2.16 ± 0.15 (0.136)	I
	Initial weight (g)	Control (Scampi feed)	0.092 ± 0.002	0.092 ± 0.002	0.092 ± 0.002	
	Final weight (g)	Horse gram + Ragi	0.149 ± 0.003	0.130 ± 0.003 (*)	0.137 ± 0.003 (*)	III
		Green gram + Pearl millet	0.149 ± 0.003	0.132 ± 0.003 (*)	0.141 ± 0.003 (*)	II
		Cow gram + Wheat	0.149 ± 0.003	0.138 ± 0.002 (0.003)	0.144 ± 0.003 (*)	I
Nutritional Indices	Weight gain (BI, Biomass Index)	Horse gram + Ragi	0.057 ± 0.002 BI=61.95	0.038 ± 0.002 (*) BI=41.30	0.045 ± 0.003 (0.002) BI=48.91	III
		Green gram + Pearl millet	0.057 ± 0.002 BI=61.95	0.040 ± 0.004 (0.005) BI=43.47	0.049 ± 0.001 (0.005) BI=49.39	II
		Cow gram + Wheat	0.057 ± 0.002 BI=61.95	0.046 ± 0.004 (0.011) BI=50.00	0.052 ± 0.003 (0.013) BI=56.52	I
	Specific growth rate	Horse gram + Ragi	0.465 ± 0.013	0.332 ± 0.029 (0.005)	0.388 ± 0.028 (0.012)	III
		Green gram + Pearl millet	0.465 ± 0.013	0.342 ± 0.029 (0.006)	0.415 ± 0.027 (0.025)	II
		Cow gram + Wheat	0.465 ± 0.013	0.391 ± 0.021 (0.004)	0.436 ± 0.023 (0.037)	I
	Condition factor	Horse gram + Ragi	1.121 ± 0.088	1.423 ± 0.042 (0.008)	1.308 ± 0.104 (0.002)	III
		Green gram + Pearl millet	1.121 ± 0.088	1.341 ± 0.087	1.233 ± 0.058	II

			(0.000)	(0.023)	
	Cow gram + Wheat	1.121 ± 0.088	1.169 ± 0.056 (0.122)	1.134 ± 0.059 (0.519)	I
Survival rate (%)	Horse gram + Ragi	91.0 ± 3.00	93.00 ± 2.00 (0.074)	93.00 ± 2.00 (0.074)	II
	Green gram + Pearl millet	91.0 ± 3.00	92.00 ± 1.00 (0.478)	92.00 ± 2.00 (0.225)	III
	Cow gram + Wheat	91.0 ± 3.00	94.00 ± 1.00 (0.122)	94.00 ± 1.00 (0.122)	I
Rank		I	III	II	

GOK, groundnut oilcake; SM, soya meal; EA, egg albumin
Each value is mean ± SD of triplicate observations.
Significance (P<) of paired samples t-test are given in parentheses (* the correlation and t cannot be computed because the SE of the difference is '0').

Table 2a: Two-way ANOVA and DMRT values of morphometric data and nutritional indices of *M. malcolmsonii* PL fed with different formulated feeds

Parameter	Two-way ANOVA						DMRT				
	SV	DF	SS	MS	F- value	CV %	(Mean Significance)				
Length	Treatment	8	0.322	0.040	1.33 ns		--	T1	T2	T3	G mean
	Type (T)	2	0.301	0.150	4.98*		G1	a	a	a	a
	Grains (G)	2	0.011	0.0056	<1	8.2	G2	a	a	a	a
	T x G	4	0.0099	0.0024	<1		G3	a	a	a	a
	Error	18	0.545	0.0302	--		--	--	--	--	--
	Total	26	0.867	--	--		--	--	--	--	--
Weight	Treatment	8	0.001288	0.00016	19.01**		--	--	--	--	--
	Type (T)	2	0.0011	0.00055	65.49**		G1	a	b	b	b
	Grains (G)	2	0.000114	0.000057	6.75**	8.2	G2	a	b	ab	b
	T x G	4	0.000064	0.000016	1.89 ns		G3	a	a	a	a
	Error	18	0.000152	0.0000084	--		--	--	--	--	--
	Total	26	0.000143	--	--		--	--	--	--	--
Weight gain	Treatment	8	0.0012	0.00016	21.56**		--	--	--	--	--
	Type (T)	2	0.0011	0.00055	74.28**		G1	a	b	b	b
	Grains (G)	2	0.00011	0.000057	7.66**	5.6	G2	a	b	ab	b
	T x G	4	0.000064	0.000016	2.15 ns		G3	a	a	a	a
	Error	18	0.00013	0.0000074	--		--	--	--	--	--
	Total	26	0.00141	--	--		--	--	--	--	--
Specific growth rate	Treatment	8	0.063	0.0079	15.40**		--	--	--	--	--
	Type (T)	2	0.054	0.0272	52.50**		G1	a	b	b	b
	Grains (G)	2	0.0059	0.0029	5.69*	5.5	G2	a	b	ab	b
	T x G	4	0.0035	0.00088	1.71 ns		G3	a	a	b	a
	Error	18	0.0093	0.00051	--		--	--	--	--	--
	Total	26	0.073	--	--		--	--	--	--	--
Condition factor	Treatment	8	0.309	0.0386	6.52**		--	--	--	--	--
	Type (T)	2	0.162	0.0814	13.74**		G1	a	a	a	a
	Grains (G)	2	0.093	0.0468	7.91**	6.3	G2	a	a	ab	a
	T x G	4	0.527	0.0131	2.22 ns		G3	a	b	b	b
	Error	18	0.106	0.0059	--		--	--	--	--	--
	Total	26	0.416	--	--		--	--	--	--	--
Survival rate	Treatment	8	36.000	4.500	<1		--	--	--	--	--
	Type (T)	2	24.000	12.000	2.57 ns		G1	a	a	a	a
	Grains (G)	2	8.000	4.000	<1	2.3	G2	a	a	a	a
	T x G	4	4.000	1.000	<1		G3	a	a	a	a
	Error	18	84.000	4.666	--		--	--	--	--	--
	Total	26	120.000	--	--		--	--	--	--	--

** - Significant at 1% level; ns: not significant
* - Significant at 5% level
T1, Control; T2, Feed type-A; T3, Feed type-B; G mean, between grains.
G1, Horse gram + Ragi; G2, Green gram + Pearl millet; G3, Cow gram + Wheat.
DMRT significant: a, not significant at 5% level; ab, slightly significant; b, significant

Protein and Amino acids

Nutrition is regarded as a key factor controlling survival and growth of crustacean culture. Table 3 represents contents of biochemical constituents in PL fed with formulated feeds. The content of total protein in PL fed with different types of feeds was in the order of scampi feed > feed-B (formulated with higher proportion of pulses) > feed-A (contained equal proportion of pulses and cereals). Among three different combinations of pulses and cereals tested, the protein level in PL fed with feed prepared from cow gram and wheat was found to be higher followed by feed prepared with green gram and pearl millet, and horse gram and ragi. Therefore, total protein value observed in PL fed with the feed formulated by using cow gram and wheat, particularly feed type-B was very nearest to control. The similar trend was recorded in the content of total amino acid of PL fed with different types of feeds formulated with combinations of different pulses and cereals (Table 3). These results were confirmed through paired samples t-test (protein: P<0.000-0.141; amino acid: P<0.001-0.946) and DMRT (Tables 3 and 3a). When two-way ANOVA was applied, the F-values obtained for all the combinations of variables were found to be significant at 1% level except total protein between different combinations of pulses and cereals, and total amino acid between feed types and different combinations of pulses and cereals were compared (Table 3a).

Prawns are rich in variety of organic and inorganic constituents. One of the major requirements of prawn culture is the transformation of dietary protein into tissue protein for normal body function, growth and maintenance. Therefore, protein constitutes the most expensive ingredients in artificial feed. In the present study, the quality and quantity (or the nutritive/ biochemical value) of dietary protein offered with formulated feeds, particularly by feed type-B were appreciably good as control feed. Further, among three different B-type feeds, PL fed with cow gram and wheat showed comparatively equal performance with control. The protein requirements in animals are influenced by various factors, such as body size, water temperature, feeding rate, availability and quality of foods, over all digestible energy content of the diet and the quality of protein [28]. In general, larval and juvenile animals have greater protein requirement than adults [29] because the former usually have faster growth rates and higher metabolic rates. In the present study, the early PL has efficiently utilized the nutrients available in formulated feeds, particularly with cow gram and wheat. Therefore, this feed produced appreciable growth as control.

The body amino acid concentration reflects the dietary amino acid concentration [30, 31]. Amino acids participate in osmoregulation and in the control of cell volume [32-34], which represents the body mass. In the present study, the biomass in PL fed with formulated feeds, particularly with cow gram and wheat was equally good as control feed.

Carbohydrate

The content of total carbohydrate in PL fed with different feeds was in the order of control > green gram and pearl millet > horse gram and ragi > cow gram and wheat. Therefore, the content of total carbohydrate was found to be lower in PL fed with formulated feeds when compared with control. As for as feed types are concerned, in contrast to total protein, the level of total carbohydrate was found to be higher in feed type-A than that of the feed type-B. This is because to the increased proportion cereals and decreased proportion of pulses in feed type-B (Table 3). These results were confirmed through paired samples t-test (P<0.000-0.231) and DMRT (Tables 3 and 3a). When two-way ANOVA was applied, the F-values obtained for all the combinations of variables were found to be significant either at 1% level or 5% level except comparison between feed types and different combinations of pulses and cereals were made (Table 3a).

Generally, carbohydrates in tissues of crustaceans exist as protein–bound sugars and glycogen. It is well known that the sugars serve as energy reserve for the metabolic process. Carbohydrates play an important role in the synthesis of chitin, fatty acids and steroids. Carbohydrates are considered to be the first among the organic nutrients can be utilized to generate required energy. Carbohydrates are the inexpensive source of energy supplying nutrients and serve as precursors for the dispensable amino acids and metabolic intermediates necessary for growth [35]. In the present study, the formulated feeds seemed to be contained enough quantity of carbohydrates. Actually feed type-B contained less quantity of total carbohydrate when compared with feed type-A because of less proportion of cereals and more proportion of pulses. However, feed type-B produced better results because it contained more protein. Moreover, among three different formulations, the feed, which contained cow gram and wheat was produced better growth. As all the feed types and feed combinations formulated contain adequate quantity of carbohydrate. It is suggested that the available protein was spared for growth rather than utilized for maintenance.

Lipid

In the feed type-A, the content of total lipid was found to be in the order of control > green gram and pearl millet > cow gram and wheat > horse gram and ragi. Whereas, in the case of feed type-B is concerned, the content of total lipid was found to be in the order of green gram and pearl millet > control > cow gram and wheat > horse gram and ragi. As for as experimental feeds fed PL are concerned, the total lipid content was found to be the highest in green gram and pearl millet fed PL, followed by cow gram and wheat, and horse gram and ragi. Similar to total protein level, the level of total lipid was also found to be higher in feed-B than that of the feed-A (Table 3). These results were confirmed through paired samples t-test (P<0.000-0.005) and DMRT (Tables 3 and 3a). When two-way ANOVA was applied, the F-values obtained for all the combinations of variables were found to be significant at 1% level (Table 3a).

Table 3: Concentrations of biochemical constituents in *M. molcolmsonii* PL fed with different formulated feeds

Parameters	Prepared Feeds	Initial	Control (Scampi feed)	Type - A (pulse-25% + cereal-25%) + (GOK-20% + SM-20%) + (EA-4% + tapioca-5%)	Type - B (pulse-40% + cereal-10%) + (GOK-20% + SM-20%) + (EA-4% + tapioca-5%)	Rank
Protein (mg g^{-1} wet wt.)	Horse gram + Ragi	27.13 ± 4.16	55.8 ± 1.62	50.47 ± 1.55 (0.000)	52.13 ± 0.61 (0.024)	III
	Green gram + Pearl millet	27.13 ± 4.16	55.8 ± 1.62	51.80 ± 0.91 (0.010)	53.80 ± 1.63 (0.000)	II
	Cow gram + Wheat	27.13 ± 4.16	55.8 ± 1.62	52.40 ± 0.52 (0.033)	54.40 ± 0.60 (0.141)	I
	Rank	--	I	III	II	
Amino Acid (mg g^{-1} wet wt.)	Horse gram + Ragi	15.22 ± 8.27	34.53 ± 1.44	28.02 ± 2.62 (0.011)	30.72 ± 1.21 (0.001)	III
	Green gram + Pearl millet	15.22 ± 8.27	34.53 ± 1.44	28.30 ± 1.24 (0.000)	33.53 ± 1.44 (*)	II
	Cow gram + Wheat	15.22 ± 8.27	34.53 ± 1.44	32.25 ± 0.34 (0.070)	34.48 ± 0.62 (0.926)	I
	Rank	--	I	III	II	
Carbohydrate (mg g^{-1} wet wt.)	Horse gram + Ragi	13.07 ± 1.90	28.93 ± 0.64	27.00 ± 0.23 (0.015)	25.80 ± 0.72 (0.000)	II
	Green gram + Pearl millet	13.07 ± 1.90	28.93 ± 0.64	27.86 ± 1.73 (0.231)	26.93 ± 0.64 (*)	I
	Cow gram + Wheat	13.07 ± 1.90	28.93 ± 0.64	26.20 ± 0.72 (0.000)	25.20 ± 0.72 (0.000)	III
	Rank	--	I	II	III	
Lipid (mg g^{-1} wet wt.)	Horse gram + Ragi	2.0 ± 0.11	3.7 ± 0.16	2.6 ± 0.24 (0.002)	3.1 ± 0.20 (0.001)	III
	Green gram + Pearl millet	2.0 ± 0.11	3.7 ± 0.16	3.2 ± 0.22 (0.005)	4.0 ± 0.15 (0.000)	I
	Cow gram + Wheat	2.0 ± 0.11	3.7 ± 0.16	2.8 ± 0.11 (0.001)	3.4 ± 0.17 (0.001)	II
	Rank	--	I	III	II	
Ash (%)	Horse gram + Ragi	5.70 ± 0.30	7.56 ± 0.20	8.20 ± 0.30 (0.008)	8.46 ± 0.50 (0.035)	I
	Green gram + Pearl millet	5.70 ± 0.30	7.56 ± 0.20	7.63 ± 0.15 (0.136)	8.10 ± 0.20 (*)	II
	Cow gram + Wheat	5.70 ± 0.30	7.56 ± 0.20	7.20 ± 0.20 (*)	7.43 ± 0.20 (*)	III
Rank		--	I	III	II	

GOK, groundnut oilcake; SM, soya meal; EA, egg albumin

Each value is mean ± SD of triplicate observations.

Significance (P<) of paired samples t-test are given in parentheses (* the correlation and t cannot be computed because the SE of the difference is '0').

Table 3a: Two-way ANOVA and DMRT values of concentrations of biochemical constituents of *M. malcolmsonii* PL fed with different formulated feeds

Parameter	Two-way ANOVA						DMRT				
Protein	SV	DF	SS	MS	F- value	CV %	(Mean Significance)				
	Treatment	8	95.513	11.939	7.28**		--	T1	T2	T3	G mean
	Type (T)	2	81.357	40.678	24.80**		G1	a	a	a	b
	Grains (G)	2	9.359	4.679	2.85 ns	2.4	G2	a	a	a	ab
	T x G	4	4.795	1.198	<1		G3	a	a	a	a
	Error	18	29.526	1.640	--		--	--	--	--	--
	Total	26	125.039	--	--		--	--	--	--	--
Amino acid	Treatment	8	173.92	21.740	10.50**		--	--	--	--	--
	Type (T)	2	117.50	58.75	28.37**	4.5	G1	a	b	b	b
	Grains (G)	2	32.41	16.20	7.83**		G2	a	b	a	b
	T x G	4	24.00	6.001	2.90 ns		G3	a	a	a	a

	Error	18	37.27	2.070	--		--	--	--	--	--
	Total	26	211.20	--	--		--	--	--	--	--
Carbohydrate	Treatment	8	49.141	6.142	8.86**		--	--	--	--	--
	Type (T)	2	40.376	20.188	29.12**		G1	a	ab	ab	ab
	Grains (G)	2	5.804	2.90	4.19*	3.0	G2	a	a	a	a
	T x G	4	2.960	0.740	1.07 ns		G3	a	b	a	b
	Error	18	12.478	0.693	--		--	--	--	--	--
	Total	26	61.620	--	--		--	--	--	--	--
Lipid	Treatment	8	5.226	0.653	20.54**		--	--	--	--	--
	Type (T)	2	3.406	1.703	53.55**		G1	a	b	b	b
	Grains (G)	2	1.166	0.583	18.34**	5.3	G2	a	a	a	a
	T x G	4	0.653	0.163	5.13**		G3	a	b	b	b
	Error	18	0.572	0.0318	--		--	--	--	--	--
	Total	26	5.799	--	--		--	--	--	--	--
Ash	Treatment	8	4.069	0.508	7.60**		--	--	--	--	--
	Type (T)	2	0.920	0.460	6.87**		G1	a	a	a	a
	Grains (G)	2	2.065	1.032	15.43**	3.3	G2	a	b	a	a
	T x G	4	1.083	0.270	4.05*		G3	a	b	b	b
	Error	18	1.204	0.066	--		--	--	--	--	--
	Total	26	5.274	--	--		--	--	--	--	--

** - Significant at 1% level; * - Significant at 5% level; ns, not significant.
T1, Control; T2, Feed type-A; T3, Feed type-B; G mean, Between Grains.
G1, Horse gram + Ragi; G2, Green gram + Pearl millet; G3, Cow gram + Wheat.
DMRT significant: a, not significant at 5% level; ab, slightly significant; b, significant.

In this study, the total lipid level was found to be increased in PL fed with green gram and pearl millet when compared with control. This increase in lipid level seems to have less influence on growth of PL, since lipids are reported to only serve as an alternate source of energy in crustaceans [36]. Lipids provide a source of indispensable nutrients and they also act as carriers of certain non fat nutrients notably the fat soluble vitamins like A, D, E and K [37]. Lipids are extremely important in maintaining structural and physiological integrity of cellular and sub cellular membranes. The dietary lipid level as low as 5% can be provided the sufficient levels of essential fatty acids. It has been reported that the freshwater prawns contain more level of total lipid than marine shrimps (3.18 Vs 1.33%) due to the presence of higher levels of triglycerides in the former category [38]. The importance of lipid in aquaculture has been stressed in several culture species of marine crustaceans: the cholesterol supplementation in diets improves biological performance of the prawn, *Penaeus japonicus* [29]; the mud crab, *Scylla serrate* [39]; and the tiger shrimp, *Penaeus monodon* [40]. The importance of lipid in growth promotion of *Macrobrachium* needs to be studied thoroughly.

The level of ash in PL fed with different formulated feeds was in the order of horse gram and ragi > green gram and pearl millet > control > cow gram and wheat. Therefore, the content of ash was found to be the highest in PL fed with horse gram and ragi, followed by green gram and pearl millet when compared with control. Whereas it was found to be lower in PL fed with cow gram and wheat when compared with control. These results were confirmed through paired samples t-test ($P<0.008$-0.136) and DMRT (Tables 3 and 3a). When two-way ANOVA was applied, the F-values obtained for all the combinations of variables were found to be significant either at 1% level or 5% level (Table 3a). Similar to that of total protein, amino acid and lipid levels, the proximate composition of ash was also found to be higher in feed-B than that of the feed-A (Table 3). In this study, the ash content in PL fed with different formulated feeds indicates their mineral contents.

In this study, *M. molcolmsonii* uses dietary carbohydrate as a major energy source since its availability was plenty. Therefore, proteins sparing by lipid is not considered to be crucial. The given protein was fully utilized for growth as plenty of carbohydrate and lipid were available. Therefore, diet with adequate levels of carbohydrate and lipid will enhance the protein sparing effect on growth. Since, the values of nutritional indices and analyses of proximate composition in PL fed with formulated feeds showed that the overall production quality was the best in scampi feed, better in feed type-B (contained higher pulses ratio) and good in feed type-A (contained equal proportion of pulses and cereals). Among different combinations of pulses and cereals used, cow gram and wheat showed the best growth performance on *M. malcolmsonii*, followed by better performance of green gram and pearl millet, and good performance of horse gram and ragi. Therefore, locally available low cost commodities of pulses and cereals can be used as feed ingredient to promote inland aquaculture of *Macrobrachium*.

Acknowledgement

The University Grants Commission, Government of India, New Delhi is gratefully acknowledged for the financial support provided. The first author is expressing his sincere thanks to Mr. G. Prakash, R. Vivek, and J. Rekha, 2008-2009 batch project students for initiating this work.

References

[1] Whitmarsh, D. and M.G. Palmieri, 2008. Social acceptability of marine aquaculture: The use of survey-based methods for eliciting public and stakeholder preferences. *Marine Policy*., 33: 452–457.

[2] Bhavan, P.S., S. Radhakrishnan, C. Seenivasan, R. Shanthi, R. Poongodi, and S. Kannan, 2010a. Proximate composition and profiles of amino acids and fatty acids in the muscle of adult males and females of commercially viable prawn species *Macrobrachium rosenbergii* collected from natural culture environments. *Int. J. Biol.*, 2: 107-119.

[3] FAO, 2007. FAO Yearbook. Fishery statistics: Aquaculture Production 2005. www. Fao.org/figis.

[4] Kutty, M.N., F. Herman, and H.L. Menn, 2000. Culture of other prawn species. (ed. by M.B. New & W.C. Valenti), pp. 393–410. Freshwater Prawn Culture: the Farming of *Macrobrachium rosenbergii.* Blackwell, Oxford.

[5] Radheysham., 2009. Farming the freshwater prawn *Macrobrachium malcolmsonii*. Aquaculture Production and Environment Division, Central Institute of Freshwater Aquaculture, Kausalyagang, Bhubaneswar-750002, Orissa, India. *Aquaculture Asia Magazine*, pp. 29-32.

[6] Bhavan, P.S., C. Yuvaraj, M. Leena, and M. Sangeetha, 2008. Concentrations of total protein, lipid, and carbohydrate in juveniles and sub adults of the prawn *Macrobrachium malcolmsonii* collected from the Cauvery River. *Indian J. Fish.*, 55: 323-325.

[7] Kanuajia, D.R. and A.N. Mohanty, 1996. Prospects of both mono and mixed culture of *Macrobrachium malcolmsonii*, Fishing Chimes (March, 1996), XVI, 33-35.

[8] Lee, P.G. and A.L. Lawrence, 1997. Digestibility. In: *Crustacean Nutrition* (ed. by L.R. D'Abramo, D.E. Conklin, & D.M. Akiyama), Advances in World Aquaculture, vol. 6. World Aquaculture Society, Baton Rouge, LA, pp. 194– 260.

[9] Piedad-Pascual, F., E.M. Cruz, and A. Sumalangcay Jr. 1990. Supplemental feeding on *Penaeus monodon* juveniles with diets containing various levels of defatted soybean meal. *Aquaculture* 89: 183-191.

[10] Lee, D.O.C. and J.F. Wickins, 1992.Crustacea farming, Blackwell Scientific Publications, London p.392.

[11] Anderson, T. and S. De Silva, 2003. Nutrition. In: *Aquaculture; Farming of Aquatic Animals and Plants* (ed. by J.S. Lucas, & P.C. Southgate). Blackwell Publishing, Victoria, Australia, pp. 146–171.

[12] Bhavan, P. S., S. Anjalin Ruby, R. Poongodi, C. Seenivasan, and S. Radhakrishnan, 2010b. Efficacy of cereals and pulses as feeds for the post-larvae of the freshwater prawn *Macrobrachium rosenbergii*. *J. Ecobiotechnol.*, 2/5: 09-19.

[13] Bhavan, P.S., V.G. Devi, R. Shanthi, S. Radhakrishnan, and R. Poongodi, 2010c. Basic biochemical constituents and profiles of amino acids in the post larvae of *Macrobrachium rosenbergii* fed with *Spirulina* and Yeast enriched *Artemia. J. Scientific Res.,* 2: 539-549.

[14] Raj, A.J.A., M.A. Haniffa, S. Seetharaman, and S. Appelbaum, 2008. Utilization of various dietary carbohydrate levels by the freshwater catfish *Mystus montanus* (Jerdon). *Turkish J. Fish. Aqua. Sci.,* 8: 31-35.

[15] Floreto, E.A.T., R.C. Bayer, and P.B. Brown, 2000. The effects of soybean-based diets, with and without amino acid supplementation, on growth and biochemical composition of juvenile American lobster, *Homarus americanus*. *Aquaculture,* 189: 211–235.

[16] Gimenez, A.V.F., A.C. Dia, S.M. Velurtas, and J.L. Fenucci, 2009. Partial substitution of fishmeal by meat and bone meal, soybean meal, and squid concentrate in feeds for the prawn, *Artemesia longinaris*: Effect on digestive proteinases. *The Israel J. Aqua.* – Bamidgeh, 61: 48-56.

[17] Thomas, M. and A.F.B. Van der Poel, 2001. Functional properties of diet ingredients: manufacturing and nutritional implications. In: *Advances in Nutritional Technology* (ed. by A.F.B. Vander Poel, J.L. Vahl, & R.P. Kwakkel), pp 109-122. Wageningen Pers, Wageningen, Netherlands.

[18] He, T., I.M. MeCullum, G.L. Campbell, D.L. Thiessen, and R.T. Tyler, 2002. Potential replacement of fish meal with feed pea *Pisum sativum* and pea protein fractions in practical diets for juvenile coho salmon, *Oncorhyncus kisutch*, pp. 8-10. *Proceedings of the Fourth Canadian Pulse Search Workshop*, Edmonton, Zee.

[19] Glencross, B.D., W.E. Hawkins, D. Evans, P. McCafferty, K. Dods, R. Mass, and S. Sipsas, 2005. Evaluation of the digestible value of lupin and soya bean protein concentrate and isolates when fed to rainbow trout, *Oncorhynchus mykiss*, using either stripping settlement faecal collection methods. *Aquaculture,* 245: 211-220.

[20] Lowry, O.H., N.J. Rosebrough, A.L. Farr, and R.J. Randall, 1951. Protein measurement with Folinphenol Reagent. *J. Biol. Chem.,* 193: 265-276.

[21] Moore, S. and W.H. Stein, 1948. Photometric ninhydrin method for use in the chromatography of amino acid. *J. Biol. Chem.,* 176: 367-388.

[22] Roe, J.H., 1955. The determination of sugar and blood and spinal fluid with anthrone reagent. *J. Biol. Chem.,* 212: 335-343.

[23] Folch, J., M. Lees, and G.H. Sloane-Stanley, 1957. A simple method for the isolation and purification of total lipids from animal tissues. *J. Biol. Chem.,* 226: 497-508.

[24] APHA., 2005. Standard methods for examination of water and wastewater, 21st Edn. American Public Health Association, Washington, DC.

[25] Tekinay, A.A. and S.J. Davies, 2001. Dietary carbohydrate level influencing feed intake, nutrient utilization and plasma glucose concentration in the rainbow trout, (*Oncorhynchus mykiss* Walbaum, 1792). *Turkish J. Vet. Anim Sci.,* 25: 657-666.

[26] New, M.B. and W.C. Valenti, 2000. Freshwater prawn culture: the farming of *Macrobrachium rosenbergii.* Oxford, England, Blackwell Science.

[27] Miller, G.C., 1971. Commercial fishery and biology of the freshwater shrimp, *Macrobrachium* in the Lower St. Paul River, Liberia, 1952-53. P. 13. U.S. Department of Commerce, *Natl. Mar. Fish. Serv., Spec. Sci. Rep. Fish.* 626.

[28] Watanabe, T., 1988. Fish nutrition and Mariculture (ed. By T. Watanabe), JICA Textbook, The general aquaculture course, Kanagawa International Fisheries Training Centre Tokyo University of fisheries, Tokyo, Japan.

[29] Teshima, S.I. and A. Kanazawa, 1984. Effect of protein, lipid, and carbohydrate levels in purified diets on growth and survival rates of the prawn larvae. *Bull. Jap. Soc. Sci. Fish* 50: 1709-1715.

[30] Cheng, Z.J., K.C. Behnke, and W.G. Dominy, 2002a. Effect of feather meal on growth and body composition of the juvenile pacific white shrimp (*Litopenaeus vannamei*). *J. Appl. Aquaculture* 12: 57-70.

[31] Cheng, Z.J., K.C. Behnke, and W.G. Dominy, 2002b. Effect of poultry by-product meal as a substitute for fish meal in diets on growth and body composition of the juvenile pacific white shrimp (*Litopenaeus vannamei*). *J. Appl. Aquaculture* 12: 71-83.

[32] Gilles, R., 1982. Osmoregulatory processes in mollusks and crustacean from media with fluctuating salinity regime. *Biol. Fisiol. Animal*. USP 6: 1–36.

[33] Chang, E. and J.D. O'Connor, 1983. Metabolism and transport of carbohydrates and lipids. In: *The Biology of Crustacea. Internal Anatomy and Physiological Regulation, 5* (ed. By L.H. Mantell), pp. 263–287. Academic Press, New York.

[34] Schein, V., Y. Waché, R. Etges, L.C. Kucharski, A.V. Wormhoudt, and R.S.M. De Silva, 2004. Effect of hyper osmotic shock on phosphoenolpyruvate carboxykinase gene expression and gluconeogenic activity in the crab muscle. *FEBS Letter* 561: 202–206.

[35] NRC (National Research Council), 1993. Nutrient requirements of fish. p.114. Committee on Animal Nutrition, Board on Agriculture, National Research Council, National Academy Press, Washington DC, USA.

[36] Gillbert, L.E. and J.D. O'Conner, 1983. Chemical Zoology (ed. By V.M. Florin & B.T Sheer), pp. 229. Academic press. New York.

[37] New, M.B., 1986. Aquaculture diets of post larval marine fish of the super - family Percoidae, with special reference to sea bass, sea breams, groupers and yellow tail: a review. *Kuwait bullet. Mar. Sci.* 7: 75-151.

[38] Chanmugam, P., J. Donovan, C.J. Wheeler, and D.H. Hwang, 2006. Differences in the lipid composition of freshwater prawn (*Macrobrachium rosenbergii*) and marine shrimp. *J. Food Sci.* 48: 1440-1441.

[39] Sheen, S.S., 2000. Dietary cholesterol requirements of juvenile mud crab *Scylla serrata*. *Aquaculture* 189: 277–285.

[40] Sheen, S.S., S.J. Chen, and Y.S. Huang, 1994. Effect of dietary lipid levels on the growth response of tiger prawn, *Penaeus monodon*. *J. Fish. Soc. Taiwan* 21: 205-213.