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Evaluation of Marine Fish Meat Incorporated Diets on Growth and Survival of Post-larvae of *Macrobrachium rosenbergii* (de Man)

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Abstract

Post-larvae of *Macrobrachium rosenbergii* (PL-5, average weight-10.8 ± 0.17 mg, average length, 12.1 ± 0.25 mm) were fed with five different marine fish meat incorporated diets, namely, diet-I, diet-II, diet-III, diet-IV and diet-V, prepared using ribbion fish (*Lepturacanthus savala*), croaker (*Johnius dussumieri*), pink pearch (*Nemipterus japonicus*), catfish (*Osteogeneiosus militaris*) and Thryssa (*Thryssa* sp.), respectively. Other ingredients such as prawn shell, groundnut oil cake, rice bran, wheat flour, poultry egg, agar and mineral mixture were similar for each diet. The experiment was conducted in plastic tubs (40 litres) for 30 days. Four replicates were carried out for each treatment. Stocking density was maintained at 2 PL L^{-1} and post larvae were fed 20% of their collective body weight. ANOVA (one-way) showed significantly higher ($p<0.05$) percentage gain in weight (481.3), higher specific growth rate (5.86) and higher protein efficiency ratio (2.0) in post-larvae fed on diet-III. However, there was no significant difference ($p>0.05$) in the growth rate of post-larvae fed on diet-I and diet-III. Significantly lower growth rate was observed in post-larvae fed on diet-II, diet-IV and diet-V as compared to post-larvae fed on diet-I and diet-III. There was no significant difference ($p>0.05$) in survival and food conversion ratio of post-larvae fed any of five different diets.

Introduction

Giant freshwater prawn, "Scampi" (*Macrobrachium rosenbergii*) farming has tremendous potential in India. With the development of hatchery technology for production of seed and its successful demonstration in coastal and inland states of the country, a general awareness for farming of this freshwater prawn among the farmers and entrepreneurs is growing rapidly (Tripathi 1992). Scientific developments in scampi farming and high returns from this have motivated the farmers to undertake monoculture and polyculture with carps in the grow-out ponds. Scampi responds very well for extensive and semi-intensive culture systems (Raje and Joshi 1992). Reddy (1997) reported that scampi can be cultured as an alternative to tiger shrimp (*Penaeus monodon*) in low saline water areas and coastal saline soils as Scampi can grow comfortably in waters having salinity up to 10 g L^{-1}. However, considering the vast potential resources of India for aquaculture and keeping in view the future environmental hazards and the long-term sustainability of the prawn farming. In India, 36,640 ha. are under scampi culture with an annual production of about 39,000 tons from monoculture and as one of the components in polyculture (Lokare and Anis 2000).

It is reported that feed requirements vary from species to species and also among different stages of growth in the same species (Indulkar and Belsare 2001). Two batches of the same species of prawn could have similar initial weight but different in age shows differential growth due to stunting caused by nutritional deficiency in the diet (New 1976). At present, formulated pelletized feeds made out of locally available ingredients are not readily available. New (1998) suggested that the conventional feed ingredients available in each area have to be utilized to meet the varied needs of different species at different sizes of shrimps and prawns and their formulation must adjust according to local manufacturing, storage and distribution facilities.

However, there is limited research on evaluation of diets for indoor or outdoor nursery rearing of post-larvae of *M. rosenbergii* (Briggs et al. 1988; Heinen and Mensi 1991). Most of the studies have been carried out on nutritional requirements of sub-adults and adults. Therefore, information is needed for production of juveniles by minimizing nursery costs, as the cost of producing nursed juveniles has emerged as a major factor influencing the profitability of prawn farmers. Therefore, in the present study, an attempt was made to evaluate the effect of formulated diets prepared from locally, easily available and low value varieties of marine fishes for nursery rearing of post-larvae of *M. rosenbergii* with the intention to develop a low cost and readily available post-larval feed.

Materials and Methods

Five day old post-larvae of *M. rosenbergii* acclimatized to freshwater were procured from the private hatchery located in Sawarde (Chiplun), Dist. Ratnagiri – 415 629 (India). The post-larvae produced from a single female were used for the experiment.

Five compounded flake diets (diet-I, diet-II, diet-III, diet-IV and diet-V) were formulated using different fish meat powders, i.e. ribbion fish (*Lepturacanthus savala*), croaker (*Johnius dussumeri*), pink pearch (*Nemipterus japonicus*), catfish (*Osteogeneiosus militaris*) and Thryssa (*Thryssa* sp.) and other ingredients in each diet keeping 40% protein level (Indulkar and Belsare 2001). All other ingredients were the same in each diet except the fish meat powder. Ingredients in each diet were adjusted so as to provide crude protein in the diet between 40% as the diets with this crude protein level resulted in better growth of post-larvae of *M.rosenbergii* (Indulkar and Belsare 2001). The dried ingredients in powder form were passed through a 0.5 mm mesh size and weighed as per the required quantities and thoroughly mixed. Required quantity of water (500 ml Kg^{-1}) was added and again blended for five minutes. The mixture was steamed until it became slurry. The slurry was cooled at room temperature and then additives (vitamin-mineral mixture) were added as per the requirement. The cooled slurry was spread on a black polythene sheet with the help of a smooth brush in a layer of approximately one mm thickness and sun dried. The sun-dried flakes were removed from the polythene sheet and oven-dried at 60°C for 2 to 3 hours to remove moisture. Oven-dried flakes were cooled to room temperature, packed in plastic bags and stored till used for experimentation. Composition of feed ingredients in each diet and proximate composition of formulated diets (ingredients on % dry weight basis) are given in table 1. Proximate composition such as, moisture, crude protein, crude fat and total ash of diets were analyzed as per the standard methods (AOAC, 1984). The proximate and certain fatty acid profile of marine fish meat powders are given in table 2.

The experiment was conducted for 30 days in 40 litre capacity plastic tubs keeping 20 litre water in each tub. Post-larvae were stocked at 2 L^{-1} (Garces and Heinen 1989). Four replicates were carried out for each treatment. Before starting the experiment, 50 post-larvae were chosen randomly and their initial weight and length (from tip of rostrum to telson) were recorded. Feeding was done at 20% of collective initial body weight of post-larvae (Reddy 1997). The calculated quantity of feed was divided in two parts and given in the morning (0800 hours) and evening (1800 hours). The left over feed and excreta of post-larvae were removed daily. The water siphoned out at every siphoning was replaced immediately with fresh water. Nearly 20 to 30% water from each container was replaced after each siphoning. Experiment was conducted in the laboratory at room temperature. During the experimental period

routine water parameters such as water temperature, pH and dissolved oxygen were recorded daily using standard methods (AOAC, 1984).

Table 1. Composition of feed ingredients in the diets and proximate composition of formulated diets (ingredients on % dry weight basis).

Ingredients	Formulated Diets				
	Diet - I (Ribbon fish)	**Diet - II** (Croaker)	**Diet - III** (Pink perch)	**Diet - IV** (Catfish)	**Diet - V** (Thryssa fish)
Fish meat	31.23	35.89	30.93	40.19	45.65
Prawn shell	13.75	12.82	13.81	11.96	10.86
Wheat flour	13.75	12.82	13.81	11.96	10.86
Ground nut oil cake	13.75	12.82	13.81	11.96	10.86
Rice bran	13.75	12.82	13.81	11.96	10.86
Poultry egg	13.75	82	13.81	11.96	10.86
	Proximate composition (% dry weight basis)				
Moisture	6.2	6.8	5.7	6.9	6.4
Crude protein	41.78	41.07	41.0	38.95	39.6
Crude fat	7.2	7.8	7.0	7.1	6.9
Total ash	13.9	14.4	13.5	15.0	14.1

Table 2. Proximate composition (% value) and certain fatty acids (% of total lipid) of fish meat powders (± : Standard error of mean)

Component	Ribbon fish	Croaker	Pink pearch	Catfish	Thryssa
Protein	77.0 ± 6.2	70.2 ± 5.8	76.7 ± 5.8	65.4 ± 4.5	60.8 ± 3.5
Lipid	9.83 ± 1.7	7.43 ± 1.5	8.5 ± 2.2	9.95 ± 1.8	5.5 ± 1.5
Ash	5.36 ± 1.0	7.83 ± 2.4	6.95 ± 2.8	4.43 ± 2.1	11.56 ± 2.0
Fatty acid					
14:0	5.2	1.7	3.3	2.7	4.7
16:0	28.7	39.3	21.5	22.4	31.7
18:0	9.4	9.0	8.5	11.4	8.9
16:1	7.3	8.6	7.2	7.5	6.3
18:1	17.1	14.0	16.3	17.9	22.7
20:1	2.3	1.2	-	-	-
18:3n-3	0.7	0.4	0.9	2.1	0.5
20:4n-6	3.8	4.1	4.6	4.8	2.3
20:5n-3	5.1	3.3	4.7	5.3	5.1
22:5n-3	2.3	1.7	2.2	2.7	0.1
22:6n-3	14.0	11.0	15.2	12.1	9.8
Total n-3 HUFA	21.4	16.0	22.1	20.1	15.0

At the end of an experiment, prawns in each tub were counted, weighed and their length was measured. The data for growth and survival was analyzed by one-way analysis of variance (ANOVA) to test any significant differences among means. F-value ($p< 0.05$) was employed using Fisher's protected Least Significant Difference (LSD) test to determine significant difference between the diets (Snedecor and Cochran 1967).

Results

The protein level in the various meat powder ranged between 60.8 to 77.0%, lipid level ranged between 5.5 to 9.95% and ash content varied between 4.43 to 11.56%. Ribbion fish had higher protein (77.0%) than other fish meat powder used. Thryssa meat had lowest protein (60.8%), lowest lipid (5.5%) and highest ash content (11.56 %) than other fish meat powders. Pink pearch had higher level of total n-3 HUFA (22.1%) as compared to that of ribbon fish (21.4%), croaker (16.0%), catfish (20.1%) and Thryssa (15.0%).

Results on growth and survival of post-larvae fed on different formulated diets for 30 days are given in table 3.

Table 3. Growth and Survival of post larvae of *Macrobrachium rosenbergii* fed with formulated diets.

Particulars	**Diet I (Ribbon fish)**	**Diet II (Croaker)**	**Diet III (Pink pearch)**	**Diet IV (Catfish)**	**Diet V (Thryssa)**
Initial average weight (mg)	10.8 ± 0.17	10.8 ± 0.17	10.8 ± 0.17	10.8 ± 0.17	10.8 ± 0.17
Initial average length (mm)	12.1 ± 0.25	12.1 ± 0.25	12.1 ± 0.25	12.1 ± 0.25	12.1 ± 0.25
Weight after 30 days	58.2 ± 2.77	54.5 ± 5.31	62.5 ± 1.58	60.5 ± 3.48	45.5 ± 3.20
Length after 30 days	20.6 ± 0.37	18.7 ± 0.32	20.1 ± 0.36	19.5 ± 0.59	19.2 ± 0.51
Gain in Weight (mg)	47.5 ± 2.73	41.8 ± 3.42	51.7 ± 1.58	49.8 ± 3.48	34.7 ± 3.20
Gain in Length (mm)	8.5 ± 0.38	7.3 ± 0.32	8.0 ± 0.36	7.4 ± 0.58	6.3 ± 0.42
Percentage gain in Weight	441.6^{bc} ± 29.8	388.6^{ab} ± 31.8	481.3^{c} ± 14.7	463.2^{bc} ± 32.4	323.3^{a} ± 29.8
Percentage gain in Length	70.5^{b} ± 3.13	60.6^{ab} ± 2.71	66.3^{b} ± 3.03	61.2^{ab} ± 4.89	52.1^{a} ± 3.48
Specific growth rate (% day^{-1})	5.62^{bc} ± 0.16	5.27^{ab} ± 0.21	5.86^{c} ± 0.08	5.49^{bc} ± 0.24	4.82^{a} ± 0.24
Food conversion ratio	1.37^{a} ± 0.08	1.57^{a} ± 0.01	1.25^{a} ± 0.36	1.31^{a} ± 0.10	1.65^{a} ± 0.15
Protein Efficiency Ratio	1.84^{bc} ± 0.12	1.62^{ab} ± 0.13	2.00^{c} ± 0.06	1.91^{bc} ± 0.13	1.35^{a} ± 0.12
Survival (%)	80^{a} ± 2.04	85^{a} ± 4.08	87^{a} ± 1.44	80^{a} ± 3.53	79^{a} ± 4.26

a, b, c : value in a row sharing similar letter do not differ (P>0.05). ± : standard error of mean.

The results showed that the percentage gain in weight of post-larvae were 441.6, 388.6, 481.3, 463.2 and 323.3 in post-larvae fed on diet-I, diet-II, diet-III, diet-IV and diet-V, respectively. ANOVA showed significant difference ($p<0.05$) in percentage gain in weight of post-larvae fed on different formulated diets. By computing LSD (83.49), the maximum percentage gain in weight (481.3%) was observed in post-larvae fed on diet-III followed by post-larvae fed on diet-IV (463.2%) and minimum percentage gain in weight (323.3%) was observed in the post-larvae fed on diet-V. However, there was no significant difference ($p> 0.05$) among the percentage gain in weight of post-larvae fed on the diets I, II and IV; I, III and IV; II and V.

The percentage gain in length of post-larvae after 30 days of nursery rearing was 70.5, 60.6, 66.3, 61.2, and 52.1 for diet-I, diet-II, diet-III, diet-IV and diet-V, respectively. The ANOVA showed significant difference ($p<0.05$) in percentage gain in length. The LSD (10.66) showed the minimum and maximum percentage gain in length of post-larvae when fed on diet-V (52.1%) and diet-I (70.5%), respectively. However, there was no significant difference ($p> 0.05$) in percentage gain in length of post-larvae fed on diets, I, II, III and IV; II, IV and V.

The specific growth rate (SGR) (% day^{-1}) of post-larvae fed for 30 days on different diets was observed to be 5.62, 5.27, 5.86, 5.49, 4.82 for diet-I, diet-II, diet-III, diet-IV and diet-V, respectively. ANOVA showed SGR of post-larvae was significantly different ($p<0.05$) among the diets. The maximum and minimum SGR was observed in the post-larvae fed on diet-III (5.86) and diet-V (4.82), respectively. LSD (0.562) showed significantly higher SGR in diet-III (5.86). However, there was no significant difference ($p> 0.05$) in SGR of post-larvae fed on diets, I, II and IV; I, III and IV; II and V.

ANOVA did not show significant difference ($p>0.05$) in food conversion ratio (FCR) of post-larvae fed on different formulated diets. Better FCR (1.25) was found in diet-III as compared with other diets.

ANOVA showed significant difference ($p<0.05$) in protein efficiency ratio (PER) of post-larvae fed on different formulated diets. LSD (0.35) showed significantly higher PER (2.00) in diet-III. However, there was no significant difference ($p>0.05$) in PER of post-larvae fed on diets, I, II and IV; I, III and IV; I, II and V.

The average survival ranged between 79 to 87%. ANOVA did not show significant difference ($P>0.05$) in the survival of post-larvae fed on different formulated diets.

The initial proximate compositions of post-larvae and after 30 days of rearing are given in table 4. Body composition showed crude protein in the range of 60.5 to 66.2%. Crude protein in post-larvae did not differ significantly ($p>0.05$). Crude fat level in the post-larvae varied in the range of 8.7 to 9.5 per cent, which was also not significantly different ($p>0.05$). Total ash was in the range of 18.6 to 20.4% and did

not differ significantly ($p>0.05$). The initial proximate composition of post-larvae and after feeding on various diets did not differ significantly ($p>0.05$).

Table 4. Proximate composition of post-larvae of *M. rosenbergii* fed on marine fish meat incorporated diets during 30 days of rearing period.

Parameters	Proximate composition (%)					
	Initial	Diet - I	Diet - II	Diet - III	Diet - IV	Diet - V
Protein	60.8 ± 1.3	60.5 ± 2.3	63.4 ± 1.1	66.2 ± 2.0	64.0 ± 1.2	62.9 ± 0.8
Lipid	8.2 ± 0.1	9.2 ± 0.3	9.8 ± 0.8	9.5 ± 1.2	8.9 ± 0.3	8.7 ± 0.4
Ash	18.9 ± 0.5	20.1 ± 0.7	19.4 ± 0.4	18.6 ± 0.4	20.4 ± 0.4	19.3 ± 0.3
Moisture	4.4 ± 0.3	5.5 ± 0.9	4.3 ± 0.7	3.8 ± 0.6	3.5 ± 0.6	3.9 ± 0.4

Values in % dry weight basis ± : standard error of mean.

During the experimental period, temperature varied in the range of 26.0 to 28.5°C, pH varied in the range of 6.5 to 7.5 and dissolved oxygen in the range 5.0 to 6.2 mg L^{-1}. All the water parameters were found to be in the range of tolerance limit of post-larvae of *M. rosenbergii* (New and Singholka 1985).

Discussion

Considering the weight gain, length gain, specific growth rate and protein efficiency ratio of the post-larvae of *M. rosenbergii* fed on various formulated diets during 30 days of indoor nursery rearing, the formulated diet-III resulted in better growth of post-larvae as compared to other formulated diets. The diet-III, in which maximum weight and length of post-larvae were obtained, comprises pink perch meat powder (30.93%), prawn shell powder, groundnut oil cake, wheat flour, rice bran and poultry egg (13.81% each). The proximate composition of diet-III consisted of 41.0% crude protein, 7.0% crude fat and 13.5% total ash. The minimum weight and length of post-larvae were observed in diet-V, which comprised of Thryssa fish meat powder (45.65%), prawn shell powder, rice bran, wheat flour and groundnut oil cake (10.86% each). The proximate composition of diet-V consisted of 39.6% crude protein, 6.9% crude fat and 14.1% total ash. There was no significant difference in the proximate composition of all five diets.

Comparison of the results of the present experiments with the results of other studies (e.g. Sandifer and Smith 1975, 1977; Willis et al.1976; Smith and Sandifer 1979; Smith et al. 1983; Hilton et al. 1984; Behanan et al. 1992) is rather difficult due to varying experimental designs in different regions, differences in stocking size, density, temperature and innumerable possible combinations of ingredients used. Aquacop (1976) showed that when 40% copra meal was added to a formulated

diet as a substitute for local fish meal, there was no adverse effect on the growth of *M. rosenbergii*. Wills et al. (1976) observed significantly greater weight gain in *M. rosenbergii* fed on Trout Chow-2 and the natural diet (consisted of chopped fish, chopped live earthworms, chopped shrimp 25% each and chopped sprouted mung beans, and chopped long grain wild rice, 12.5% each) than fed on marine rations. Fair et al. (1980) reported that a diet containing up to 30% cellulose fed to juvenile *M. rosenbergii* had no detrimental effect on growth. Ashmore et al. (1985) observed higher weight gain in *M. rosenbergii* when fed on diet containing barley than other cereal grains (corn, milo, and wheat). Behanan et al. (1992) reported the diet containing catfish meat (20%), prawn head (10%), gluten (10%) and clam meat powder (60%) gave better survival rate, feed conversion efficiency and protein efficiency ratio than the diet having clam meat alone. Nair and Sherief (1993) observed a diet containing clam meat and tapioca in the ratio of 1:1.5 as a practical and inexpensive diet for feeding *M. rosenbergii* juveniles. Ravishankar and Keshavanath (1988) reported that *M. rosenbergii* utilized feed pellets containing silkworm pupae and shrimp waste more efficiently and gave a higher specific growth rate than diets containing fish meal, silkworm pupae alone or silkworm pupae plus clam meat in combination. Unnikrishnan et al. (1992) were able to substitute extracted silkworm pupae for extracted clam meat for *M. rosenbergii* post-larvae without any detrimental effect on the survival, growth rate or PER. Earthworm (*Eisenia* sp.) have been identified as dietary ingredients for rearing of *M. rosenbergii* (New 1995). Zimmermann et al. (1991) compared the use of marine shrimp meal, fish meal and meat meal in 30% protein, seven per cent lipid in the diets for post-larval of *M. rosenbergii* and obtained the best biomass production with the shrimp meal diet or by rotating the three feeds on a 3-day cycle. The use of the fish meal diet gave an intermediate response while the effect of the diet containing meat meal was very poor. Mazid and Mahmud (1992) reported that a 30% protein diet containing fish meal gave a better growth rate and survival than one containing blood meal. Garces and Heinen (1993) and Molina et al. (1995) evaluated commercial feeds with beef liver supplement as diets for post-larvae of *M. rosenbergii* and reported beef liver supplementation could make a better commercial diet than Purina Trout Chow-2 for indoor nursery rearing. Tidwell et al. (1993) indicated that fish could be partially or totally replaced by soybean meal and distillers by-products in diets for the pond production of freshwater prawns. Law et al. (1990) suggested that copra, soybean meal and wheat flour were good sources of nutrients for prawns. Durairaj et al. (1992) noted an improved growth rate when prawns in manured ponds were fed a pelleted feed containing shrimp head meal (20%) and fish meal (10%) rather than a conventional feed of 1:2:1 groundnut oil cake, rice bran and trash fish. Lobeo et al. (1995) observed that post-larvae of *M. rosenbergii* fed on diet containing fish meal (34.8%), soyameal (34.8%), corn meal (17.9%), shrimp carapace meal (5%), soybean oil (5%), binder (2%), attractive-

stimulant (0.2%) and vitamin-C (0.3%) provided the best development with regard to weight gain and food conversion. Das et al. (1995) reported that best specific growth rate and protein efficiency ratio of *M. rosenbergii* juveniles were achieved when fed with diet having 40% prawn meal. Ali et al. (2000) stated that it is possible to establish silage-based diet for *M. rosenbergii* post-larvae as a possible alternative to fish meal.

It has been reported that n-3 HUFA particularly 20 : 5n-3 and 22 : 5n-3 fatty acids are known to be essential for proper growth, development and survival of marine fishes and crustaceans (Reigh and Stickney 1989). In crustaceans, several PUFA, such as 18: 2n-6, 18: 3n-3, 20: 5n-3 and 22: 6n-3 are essential to increase growth and survival of larvae and juveniles. In addition, their ability to promote ovarian maturation in brood stocks and to promote better quality eggs has been demonstrated (D'Abramo et al., 1994; Querijero et al., 1997; D'Abramo, 1998). Sandifer and Joseph (1976) and Roustaian, et al. (2001) pointed out that the *M. rosenbergii* juveniles are able to desaturate and elongate 18 : 3n-3 and 18: 2n-6 fatty acids. Reigh and Stickney (1989) observed depressed growth in freshwater prawn when fed on 18 : 3n-3 as the only source of dietary lipid. New (1995) reported that 18 : 2n-6 and 18 : 3n-3 fatty acids are required by freshwater prawns. Diet containing either 22 : 6n-3 or 20 : 4n-6 PUFAs were equally effective. It is therefore felt that, high n-3 HUFA content in pink pearch (22.1%) and ribbon fish (21.4 %) might have resulted in better growth and survival of post-larvae of *M. rosenbergii* during this experiment.

The present study indicated that better growth (more than 62.5 ± 1.58 mg in weight and 20.1 ± 0.36 mm in length) and survival (more than 85 %) of post-larvae of *M. rosenbergii* could be achieved within 30 days of rearing period by feeding diet containing combination of pink perch fish meat powder (30.93%), prawn shell, groundnut oil cake, rice bran, wheat flour and poultry egg (13.81 % each) or ribbon fish meat powder (31.23 %), prawn shell, groundnut oil cake, rice bran, wheat flour and poultry egg (13.75 % each) .

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