Optimum Replacement of Fishmeal with Soybean Meal in Diet for Macrobrachium rosenbergii (De Man 1879) Cultured in Low Saline Water

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Abstract

A feeding trial of substituting soybean meal (SBM) for fish meal (FM) for the giant freshwater prawn (*Macrobrachium rosenbergii*) was conducted in low saline water ponds. Four different iso-nitrogenous diets (Control, Diet-1, Diet-2, Diet-3) were fed to the juveniles stocked at a rate of $2/m^2$ in each pond. The control diet had 36% FM as a sole source of protein, and Diet-1, Diet-2, Diet-3 were prepared by replacing 30%, 50% and 80% of fish meal protein by SBM protein, respectively. Throughout the experiment, water and soil variables were found to be within the acceptable range for prawns. The maximum average weight (26.51 ± 0.20 g) was observed in prawn fed on Diet-3, while the minimum weight (21.72 ± 0.61 g) was measured for Diet-1. The prawns fed Diet-3 displayed higher specific growth rate ($1.31\pm0.27\%$ /day, SGR) and survival (71.97%) than the Control group. Protein efficiency ratio (1.51, PER) and feed conversion ratio (2.52, FCR) in prawns fed Diet-3 were higher and lower, respectively, but the difference was insignificant compared to the other treatments.

Keywords: growth, survival %, FCR, PER, giant freshwater prawn.

Introduction

Giant freshwater prawn, Macrobrachium rosenbergii, inhabits both freshwater and low salinity waters. It is the largest of the freshwater prawns and has been classified as a benthophagic omnivore based on analysis of foregut contents (Weindenbach, 1982; Cohen and Ra'anan, 1983). It is a prime inland cultured species, which has recently emerged as an important shellfish species for culture in south Asian countries after significant losses of penaeid shrimp culture in mid-1990s due to viral diseases. In the late 1980s, the culture of M. rosenbergii has been commenced extensively in Bangladesh and this country has become the 5th largest producer of prawn in the world (FAO, 2003).

In a successful prawn culture, a great deal of consideration is generally given to feeding and management. Feed costs constitute 40-60% of operational costs in production of the freshwater prawn (D'Abramo and Sheen, 1991). Fish meal (FM) makes up a major part of the formulated feeds for any carnivorous fish or shrimp species as a protein source. However, the recent scarcity, rising cost and uncertain consistency in supply of FM have increased the efforts to search for the alternative protein sources to FM in aquaculture by the fish nutritionists and feed manufacturers. For the purpose of producing low cost effective diets, high protein feedstuffs for FM substitution have been investigated throughout the world.

Soybean meal (SBM) is probably the most promising and most studied alternative protein source to replace FM. Use of SBM as a substitute for FM for many fish and marine crustaceans has been reported (Tacon and Akiyama, 1997; Davis and Arnold, 2000; Webster and Lim, 2002). SBM are widely used as the most cost-effective alternative for high-quality FM in feeds for many aquaculture species due to its high protein content (approximately 48%) and excellent amino acid profile (El-Sayed, 1999), lower cost and availability, as compared to the other plant protein sources (Storebakken *et al.*, 2000). Yet, sometimes, the presence of various anti-nutritional factors such as protease inhibitors, lectins, antigenic or estrogenic factors, oligosaccharides, etc. (Liener, 1989) limit its use in crustacean diets.

Little is known about the feasibility of using extracted SBM as a source of dietary protein in feed formulations for giant freshwater prawn in pond culture (Tidwell *et al.*, 1993). The objective of the present study was to demonstrate the effects of the increasing FM replacement by extracted SBM on the growth performance and feed efficiency in giant freshwater prawn, *Macrobrachium rosenbergii*.

Materials and Methods

118 day grow-out period was carried out under farm conditions in a village of Bagerhat district of Bangladesh. The salinity of the zone ranged from 0 to 5‰.

Pond Preparation

Eight ponds of different size, depth, salinity and location were selected (Table 1). The ponds retained water for 5 to 6 months. No water was exchanged

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throughout the experimental period. Prior to stocking of juvenile prawn, all finfish of the ponds were removed by netting and rotenone was applied at a rate of 0.75-0.9 g/m² to confirm the eradication of all predatory species. One week after the application of rotenone, slacked lime was spread over the pond surface water at a rate of 0.025 kg/m².

Feed Formulation and Preparation

Four different types of iso-nitrogenous and

isocaloric diets containing 25% crude protein were formulated using 'Pearson square' method. The feed ingredients were chosen on the basis of its nutritional status, price and year round availability in the local market. The proximate composition (Table 2) of the ingredients was analyzed according to AOAC (1980). The major source of protein in diet 'Control' was supplied by FM. The FM protein was substituted at a rate of 30%, 50% and 80% with extracted SBM in Diet-1, Diet-2 and Diet-3, respectively (Table 3).

After weighing, the feed ingredients were mixed

Table 1. Physico-chemical description of the ponds selected for the study

Pond No.	Area (m ²)	Depth (m)	Dissolved oxygen (ppm)	pН	Salinity (‰)
Control – 1	602.60	1.1	6.4	8.0	3.0
Control - 2	195.07	1.4	5.3	7.9	0.0
Diet: 1-1	346.02	1.2	6.7	8.0	3.0
Diet: 1 –2	398.63	1.1	8.2	7.8	3.0
Diet: 2-1	602.60	1.2	5.0	8.0	3.0
Diet: 2 –2	153.38	1.3	5.4	8.2	3.0
Diet: 3 –1	200.73	1.3	5.1	7.7	0.0
Diet: 3 –2	167.14	1.2	5.8	7.8	3.0

Table 2. Proximate composition of feed ingredients (values average, n=3)

	Proximate composition (% Dry matter basis)				
Feed ingredients	Protein	Lipid	Ash	Crude fiber	NFE ¹
Fish meal	42	8	35	5	10
Extracted soybean meal	54	3	6	11	26
Wheat flour	16	2	3	9	70
Wheat flour by product	19	5	4	9	63
Rice polish	15	18	14	18	35

 1 NFE = 100 – (protein + lipid + ash + crude fiber) %

Table 3. Formulation of different experimental diets for Macrobrachium rosenbergii

		Diets (% in dr	y matter basis)	
Ingredient	Control (0%)*	Diet-1 (30%)	Diet-2 (50%)	Diet-3 (80%)
Fishmeal	36	25.25	18	7.2
Extracted soybean meal	0	8.83	14.72	23.56
Wheat flour	17	17	17.7	18.33
WFBP ^{**}	21	21	21.16	21.66
Rice Polishing	19.4	21.07	21.32	21.4
Oyster Shell	3	3	3	3
Shark oil	3	3.25	3.5	4.25
Binder ^{***}	0.6	0.6	0.6	0.6
Total	100	100	100	100
	Proximate	composition of the die	ts in dry matter	
Protein	25.14	25.28	25.7	26.8
Lipid	8.98	9.61	10.08	8.04
Ash	9.92	9.20	8.01	7.56
Fiber	8.67	9.02	10.12	10.69
NFE	47.29	46.89	46.09	46.91
GE (kJ/g)****	17.91	18.12	18.27	17.86

*% of fishmeal protein substituted by extracted soybean meal.

** Wheat flour byproduct (fine); ² PEGABIND[®] Bentoli, Inc. USA.

^{***} GE (Gross Energy; kJ/g): Protein = 23.9 kJ; Lipid = 39.8 kJ; Carbohydrate = 17.6 kJ.

together manually until a homogeneous mixture was obtained. Then wet ingredients and water were added into the mixture to obtain dough. Following this, mixture was manually pressed through a locally manufactured feed pelletizer. The pellets were dried in well aerated place under the shade for 2 days until became sufficiently dry. Finally, the pellets were sacked in plastic bags and kept in a cool, dry place until used. The proximate analysis (Table 3) of the diets after preparation was also done according to AOAC (1980).

Stocking and Feeding

Juveniles of 2.51-5.19 g were stocked into ponds at a rate of $2/m^2$ and prawns were fed to satiation twice a day with the experimental (New, 2002). The feeding levels were adjusted by checking the feeding trays two hours after serving the diets.

Sampling

Sampling was carried out fortnightly by catching minimum 10 prawns from each pond by cast netting. Individual prawn weights to the nearest unit were recorded. Final sampling (harvesting) was done by draining the ponds, prior to the arrival of the winter season when the temperature dropped below 20°C. Individual weights of all prawns in each pond were recorded for further statistical analysis.

Specific growth rate (SGR%), survival rate %, feed conversion ratio (FCR), and protein efficiency ratio (PER) were calculated according to De Silva and Anderson (1995).

Water and Soil Quality

Water temperature was measured once a day at 10:00 h by mercury thermometers. Dissolved oxygen was measured once a week at 10:00 h by Winkler method (Azide modification of Winkler's method, APHA, 1992). The water pH was determined once a

week at 10:00 h by using RI 02895 (Woon Socket, HANNA) pH meter. Alkalinity of water measurements (mg/L CaCO₃) was made once a week at 10:00 h according to the APHA (1992). Natural productivity of the ponds was measured fortnightly via measuring the transparency by using a Secchi disc.

The total nitrogen content in soil of experimental ponds was determined fortnightly by micro Kjeldahl procedure according to AOAC (1980). The soil pH from the pond bottom was also measured by pH meter.

Statistical Analysis

Means and standard deviations or ranges were calculated and expressed as mean (\pm SD/range). Significance of variations in the water quality and growth parameters was tested by using one way analysis of variance, ANOVA, which was followed by Duncan's Multiple Range Test for significant values using SPSS software. Values were considered significant at 5% level of significance.

Results

Physico-Chemical Parameters of Water and Soil

Overall mean values and ranges of soil and water quality variables are presented in Table 4. The mean water temperature was observed to be within a range of $29.5-30.3^{\circ}$ C and did not show any significant differences (P>0.05) among the ponds receiving different treatments throughout the study. The mean dissolved oxygen (DO) level ranged from 5.5 to 8.2 mg/L throughout the study period. No significant difference (P>0.05) in DO value was observed between the control and other treatments but D-1 and D-3 were found statistically (P<0.05) different. Mean total alkalinity varied within a range of 186.7-220.0 mg/L, and no significant differences (P>0.05) among the treatments were observed. The salinity was

Table 4. Physico-chemical parameters	of water and soil in experimental ponds	(values are the mean of duplicates and range)
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Parameters	Control	Diet-1	Diet-2	Diet-3
Temperature (°C) ¹	29.5 ^a	30.3 ^a	29.9 ^a	30.0 ^a
•	23.6-35.2	26.1-34.4	25.8-33.5	24.7-34.8
DO (mg/L)	6.7 ^{ab}	8.2 ^b	6.9 ^{ab}	5.5 ^a
	3.4-11.7	5.7-11.6	3.9-10.0	3.0-9.5
Total alkalinity (mg/L, CaCO ₃)	212.6 ^a	213.9 ^a	220 ^a	186.7 ^a
	174.0-251.2	164.9-251.2	164.9-257.6	149.2-231.4
Salinity (‰)	0.3 ^a	0.7 ^a	0.5 ^a	0.4 ^a
	0.0-3.0	0.0-3.0	0.0-3.0	0.0-3.0
Water pH	8.0 ^b	8.0 ^b	8.0 ^b	7.7 ^a
	7.7-8.4	7.7-8.7	7.7-8.4	7.6-8.2
Soil-N (%)	0.12 ^a	0.15 ^a	0.14 ^a	0.12 ^a
	0.07-0.16	0.11-0.19	0.07-0.22	0.09-0.16
Soil pH	7.4 ^a	7.4 ^a	7.4 ^a	7.4 ^a
-	6.9-7.6	7.3-7.6	7.1-7.8	7.2-7.6

¹Same superscript in the same row indicates no significant difference (P>0.05)

observed to range from 0 to 3.0‰ and no significant differences (P>0.05) were observed among the mean values. The water pH varied within a range of 7.6-8.7 with an average of 7.7-8.0 and ANOVA showed no significant differences (P>0.05) among means, except the ponds receiving Diet-3. Pond natural productivity ranged from 31.0 to 33.29 cm and no significant differences (P>0.05) were observed among the mean values.

Soil nitrogen and soil pH of the ponds receiving different diets varied from 0.07-0.22% and 6.9-7.8, respectively. No significant differences among the mean values were observed for both parameters.

Growth Performance

Growth data of the experiment are given in Table 5. The lowest survival rate of prawn was observed in Diet-1 (55.64%) followed by that of diet-2 (64.44%) and the highest was observed in Diet-3 (71.97%) followed by the Control (66.93%). No significant difference in the survival of prawns was observed among the groups fed on different diets. The average final weight of the prawn varied from 21.72 to 26.51 g. The prawns fed on Diet-3 showed almost the highest average weight throughout the 118 days of feeding trial (Figure 1). The prawns that fed on Diet-3 and Diet-2 gained significantly higher (P<0.05) average weight compared to the prawns fed on Control and Diet-1. SGR for the diets varied between

0.91 and 1.31. It was evident that SGR was the lowest in Diet-1 and the highest in Diet-3. No significant differences (P>0.05) in SGR among the diets were observed. The lowest production of prawn in the farmer's pond condition was obtained for Diet-1 followed by the control, and that of the highest was obtained for Diet-3 followed by Diet-2.

It is obvious from the Table 5 that FCR values of the 4 experimental diets varied from 2.52 to 4.48. The highest FCR was observed in Diet-1 and the lowest in Diet-3 but no significant differences (P>0.05) in FCR among the diets were reported. The apparent protein efficiency ratios (PER) of the diets were found to be between 0.95 and 1.51. The lowest PER was obtained in Diet-1 and the highest in Diet-3. There were no significant differences (P<0.05) in PER among the means for the treatments.

Discussion

In general, soil and water quality parameters are the important variables influencing the productivity of a water body and the biological performance of cultured aquatic species. During the study period, all the physico-chemical water parameters of ponds were found to be within the acceptable limits for *M. rosenbergii* reported in various literatures (Daniel, 1981; Sandifer and Smith, 1985; Boyd, 1990; New, 1995; D'Abramo and Brunson, 1996; Hall and Van Hamm, 1998).

Table 5. Growth parameters of Macrobrachium rosenbergii fed on different diets in the experimental ponds

Parameters	Control	Diet-1	Diet-2	Diet-3
Stocking weight (g) ¹	3.66±1.15	5.19 ± 1.15	2.51 ± 1.15	4.46 ± 1.15
Final weight $(g)^2$	$23.35^{b} \pm 0.15$	$21.72^{a} \pm 0.61$	$25.61 \ ^{\circ} \pm 0.38$	$26.51^{\circ} \pm 0.20$
FCR	$2.97^{a} \pm 0.09$	$4.48^{a} \pm 1.66$	$3.09^{a} \pm 0.44$	$2.52^{a} \pm 0.47$
PER	$1.34^{a} \pm 0.04$	$0.95^{a} \pm 0.35$	$1.27^{\rm a} \pm 0.18$	$1.51^{a} \pm 0.28$
SGR (% /day)	$1.16^{a} \pm 0.16$	$0.91^{a} \pm 0.38$	$1.18^{a} \pm 0.32$	$1.31^{a} \pm 0.27$
Survival (%)	$66.93^{a} \pm 13.43$	$55.64^{a} \pm 22.42$	$64.44^{a} \pm 23.22$	$71.97^{a} \pm 23.36$
Production (kg/ha/118 days)	$308.65^{a} \pm 59.91$	$243.31^{a} \pm 98.63$	326.98 ^a ±122.25	$382.43^{a} \pm 92.79$

¹Values are the means ± SD of duplicates

²Same superscript in the same row indicates no significant difference (P>0.05)



Figure 1. Growth performance of *Macrobrachium rosenbergii* at different sampling days (average weight).

The findings of New and Singholka (1985), and Adhikari et al. (2007) do not support the present study which demonstrated that higher alkalinity levels did not adversely affect the prawn growth. The study ponds had water transparency of 31-33.29 cm, which seems to be in the suitable range of 25-40 cm recommended by New (2002). Soil nitrogen and pH were found to be also optimum for the growth of M. rosenbergii. According to Boyd (2003), the soil pH range from 6.5 to 7.5 indicates high potential for aquaculture production. The statistical analysis of the results obtained in the present investigation indicates that there were no significant differences (P>0.05) among the water and soil variables measured in all experimental ponds. Thus, the effects of water and soil quality, as well as natural food on the growth of the prawns can be assumed to be similar to each treatment of the respective experiments.

In the present study, the final weight of prawn was influenced greatly with the inclusion of different levels of SBM in the diet. The highest average weight of prawn was achieved in the groups that were fed on Diet-3 where 80% FM protein was replaced by extracted SBM. Comparatively better FCR, PER and SGR was achieved with Diet-3. The highest survival rate was observed in the prawns fed on Diet-3. The lowest growth performance was achieved in the prawns fed on Diet-1. However, the growth performance of prawn juvenile in the present work was found to be lower, compared to the findings obtained by Hossain and Paul (2007). They reported that a diet containing 30% protein using FM, meat and bone meal, mustard oilcake, sesame meal and rice bran may generate good growth in prawn juvenile reared for 90 days. The dissimilarity could be attributed to the difference in feed ingredients and their nutritive value, and crude protein level in the diet.

SBM contain some antinutritional factors, such as protease (trypsin) inhibitors, phytohaemagglutinin (lectins), anti-vitamins, phytic acid, saponins, and phytoestrogens (El-Sayed, 1999; Francis et al., 2001). Phytates, protease inhibitors, lectins and anti-vitamins all can be removed from SBM by thermal processing. Past studies revealed that SBM can be included in diets fed to rainbow trout at levels from 25-80% of the diet (Tacon et al., 1983; Smith et al., 1988; Refstie et al., 1997). According to Mc Googan and Gatlin (1997) the fish grew successfully with diets in which 90 and 95% of FM protein was replaced by SBM with additions of amino acids. Similarly, Gallagher (1994) found that replacement of up to 75% of FM protein with SBM was possible in the diet of hybrid striped bass with methionine supplements. The results of the present studies partially differ with the findings of Du and Niu (2003), who conducted a study in tank water on M. rosenbergii fed to diets where 0, 20, 50, 75 and 100% of FM is replaced by SBM. They concluded that SBM, without supplementation of amino acids or other additives, is not suitable as a major protein source in freshwaterprawn diets. However, Weidenbach (1980) reported that prawns are able to adjust to the absence of feed pellets by increasing consumption of available vegetation. According to Tidwell *et al.* (1995), prawns may be able to adjust to reductions in the nutritional value of prepared diets (i.e. protein source and vitamin and mineral content) by increasing predation on natural fauna (i.e. macro invertebrates) in the pond. These literatures could support the observations of the current study.

Although Diet-3 had higher content of crude protein (26.8%) and fibre (10.69%), compared to the other diets, the prawns displayed the best performance on this diet. This fact could be associated with the minute inclusion of FM containing some essential amino acids, higher protein and/or fibre contents in the diet. González-Peña *et al.* (2002) showed that the SGR, feed conversion efficiency and PER of small and large adult *M. rosenbergii* improved as levels of dietary fibre were increased from 0.4% to 8%; they concluded that the inclusion of up to 10% of dietary fibre increases growth rates in adult prawns by increasing nutrient residence time, thus increasing absorption.

Moreover, the FM used in this study was obtained from a locally available dried fish, Bombay duck, (*Harpodon nehereus*). The protein availability of FM was assumed to reduce due to un-hygienic storage condition and insect infestation during storage. It was also noticed by personal interview with the enterprise that the fish was dried directly in the sun for the production of fishmeal. Fatty acid oxidation could have taken place at the time of sun drying which may affect its quality and consequently the growth of prawn in the control.

In conclusion, it was revealed based on the above findings that the extracted SBM can substitute encouragingly the Bombay duck fish meal protein by 80%. The low price and local availability of SBM made the Diet-3 more potential for dietary cost savings. However, the maximum inclusion level of SBM and other locally available plant ingredients in an attempt to replace regionally available FM should be studied further to develop an economically viable, user friendly and eco-friendly feed for *M. rosenbergii*.

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