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Original Article

Evaluation of Formulated Inert Larval Diets for Giant Freshwater Prawn, *Macrobrachium rosenbergii* Weaning From *Artemia*

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Abstract

Two larval inert diets were formulated and evaluated in partial replacement for *Artemia* in the larval rearing of *Macrobrachium rosenbergii*. Thirty larvae L^{-1} were stocked in 150-L Fiberglass Reinforced Plastic (FRP) tanks, (Which had central sloping bottom) with a total density of 4500 larvae in each tank. The test diets were formulated by using meat of fish (Diet 1) and shrimp and clam (Diet 2) as major ingredients. Other ingredients used were chicken eggs, agar, milk powder, cod liver oil. The diets were fed to larvae twice daily at 8:30 hrs and 14:30 hrs at a rate of 0.20.5-g/1000 larvae. Larvae fed diet (2) containing shrimp and clam meat showed significantly higher survival than those fed fish meat based diet diet (1). The larvae fed diet (2) was more efficient in larval metamorphosis, larvae took less time to reach the next stage compared to those fed diet (1). The larvae fed diet (2) had significantly higher mean larval stage (MLS) than those fed diet (1). Length and weight measurements were higher for the larvae fed diet (2). Water quality (Water temperature, pH, dissolved oxygen, free carbon dioxide and total ammonia) were not affected by the test diets and the values recorded were in acceptable range for freshwater prawn larval rearing.

Key Words: *Nutrition, Inert diets, Larvae, Freshwater prawn, Macrobrachium rosenbergi.*

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Introduction

The giant freshwater prawn, *Macrobrachium rosenbergii* is a commercially important species in India and South Asian countries. Freshwater prawn farming has the potential to revolution the rural aquaculture, considerable employment and income could be generated, thereby bringing

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prosperity to rural poor (Parameshwaran, 1994). Freshwater prawn farming is environmentally sustainable, since it is practiced at lower grow-out density (New, 1995). A major constraint in the large scale aquaculture of this species in India is the scarcity of seed at required quantities and in all locations. A majority of seed used in grow out farming of *M. rosenbergii* comes from hatcheries (Murthy, et al. 2004). Existing hatcheries in the country are not producing up to their installed capacity due various constraints.

Average survival in commercial hatcheries in India is about 30% (Murthy and Satheesha, 1998; Murthy, et al. 2004). Imported *Artemia* cysts are predominantly used, which are expensive and uncertain in availability. Dependence entirely on *Artemia* as feed not only makes hatchery operations expensive, but also unsustainable. Hence, there is a need to look for alternative acceptable diets to replace *Artemia* and reduce the cost of larval prawn rearing. Accordingly, an attempt was made in the present study to evaluate formulated inert larval diets to replace *Artemia* partially in freshwater prawn hatchery.

Materials and Methods

Two larval diets were formulated using meat of fish (Diet 1) and shrimp and clam (Diet 2), and along with other ingredients namely, chicken eggs, agar, milk powder, cod liver oil... etc (Table 1). One gram of yeast was dissolved in 5ml of warm water and mixed with egg white including yolk. Other feed ingredients were added and blended thoroughly in a kitchen mixer. The diets were cooked on a water bath for 15 minutes, cooled and stored at 4^0 C.

Fiberglass reinforced plastic tanks (150 *L capacity*) with central sloping bottom were employed for larval rearing.

After hatching, the larvae were collected and distributed to larval rearing tanks at a low density of 30 larvae L^{-1} of water with a total density of 4500 larvae in each tank.

Table 1: Composition of formulated test diets.

Composition	g/100g	
	Diet 1	Diet 2
Egg	20	20
Shrimp meat	-	30
Clam meat	-	30
Fish meat	60	-
Milk powder	10	10
Corn flour	06	06
Agar agar	01	01
Vitamin-mineral mix	01	01
Yeast	01	01
Cod liver oil	01	01
Total	100	100

Diets were offered to the prawn larvae twice daily at 8:30 hrs and 14:30 hrs at a rate of 0.2 to 0.5g for 1000 larvae. Before feeding, the diets were passed through desired mesh size sieves (250-1000µ), so that the particle size of the food was acceptable to the prawn larvae. Live *Artemia* were fed once daily at 17:30 hrs at a rate of three organisms per ml of tank water. Biochemical analysis of freshly hatched *Artemia* and formulated diets was carried out (Table 2) following standard methods of AOAC (1995). Water quality was analysed every week. Temperature of water was measured using a mercury thermometer having an accuracy of 0.10^0C. pH was measured using a laboratory lovibond comparator. Salinity of the water was estimated using a refractometer with 1ppt accuracy (Atago make). Dissolved oxygen, free carbon dioxide and total ammonia-N were estimated following standard methods (Clescerl, et al. 1999).

Table 2: Proximate composition of the two test diets and *Artemia* nauplii (% dry weight±SD).

Diet	Protein	Lipid	Ash
Diet 1	49.07±0.42	8.63±0.41	2.90±0.02
Diet 2	50.03±0.80	8.21±0.21	2.92±0.01
Artemia	48.43±1.36	19.00±0.26	7.43±0.21

Variation in size among individuals within each of the first nine stages of *Macrobrachium rosenbergii* larvae was observed following procedure described earlier (Uno and Soo, 1969; Murai and Andrews, 1978). Efficiency of experimental diets was evaluated on the basis of postlarval growth, time required for metamorphosis and related percentage survival. The larvae were observed daily to record the progression of metamorphosis namely, mean larval stage (MLS) and relative percentage survival. Larvae from each tank were collected and studied for morphological characters following the descriptions given by Uno and Soo (1969). Development of larvae was determined by calculating the MLS formulas described by Lovett and Felder (1988):

MLS= (S x PS)

where, S is the larval stage number, and PS is the proportion of the larvae at stage S.

When more than 95% of the larvae in all the tanks had metamorphosed to postlarvae, the experiment was terminated. All the post larvae were harvested from each tank and counted. At the end, 50 postlarvae were randomly taken from each tank to measure individual total length (from tip of the rostrum to the end of the telson) and total weight.

Two way analysis of Variance and Duncan's multiple range test were used to test the significant difference between the treatments (Snedecor and Cochren, 1968; Duncan 1955).

Results and Discussion

The tanks in which larvae were offered diet (2) had higher survival rate (38.9%) than larvae offered diet (1) (31.1%). The larvae fed diet (2) took less time to reach the next stage than those fed diet (1) which indicates efficiency of diet (2). The larvae fed diet (2) had the highest MLS which significantly differed from those of larvae fed diet (1). The length and weight measurements were highest for the larvae fed diet (2) (9.69mm and 9.96mg, respectively) than the larvae fed diet (1) (0.19mm and 9.32mg). Higher survival, MLS, growth and metamorphosis were recorded in the larvae fed diet (2), probably attributed to the diet which contained shrimp meat and clam meat, which are known to contain higher HUFAs, feed attractants and certain unknown growth factors. Shrimp and clam are of marine origin. They contain more of ω_3 HUFA's, which are important for prawn larval growth and survival (Murthy, 1998). The dietary protein and lipid requirements of postlarvae of *M. rosenbergii* have been reported (Indulkar and Belsare, 2003; Murthy, 1998). Lee (1982) reported an average production of 19 PL L^{-1} of *M. rosenbergii* postlarvae by feeding *Artemia* and egg custard diet. Formulated diet that contained varying levels of chicken egg, milk powder, squid meat in combination with *Artemia* yielded better growth and survival of prawn larvae (Rao, 1997). Mohan and Rao (2000) obtained synchronous larval metamorphosis of *M. rosenbergii* by feeding meat of freshwater mussel (*Lamellidens sp*), than by feeding prepared diet and *Artemia.*

Artemia nauplii are the predominant live food used for larval rearing in prawn hatcheries. Though supplementation of *Artemia* with prepared feed in prawn larval rearing have been reported (Sick and Beaty, 1975; Corbin, et al. 1983), no standard substitute for *Artemia* has been developed in freshwater prawn hatcheries. In view of the high cost of cysts and their occasional scarcity, too much dependence on *Artemia* is a major constraint in the expansion of *Macrobrachium rosenbergii* hatcheries (New, 1990). Bacterial degradation of larval exuvia and empty shells of *Artemia* foul the water, accumulated debris entangles larvae and leads to mortality. The cysts which are ingested by the larvae cannot be digested and may cause blockage of the gut and have other deleterious effects (Stults, 1974). There is considerable variation in nutritive quality (Particularly HUFAs) of *Artemia* cysts among different geographical strains, (Murthy, 1998). These are some of demerits of *Artemia* in addition to its cost and availability factors. Long larval phase (Compared to penaeid shrimps) and cannibalistic nature of larvae of freshwater prawn have resulted in poor survival in the

commercial hatcheries in India (Murthy, et al. 2002)

Water quality was found suitable for rearing of freshwater prawn larvae in the present experiment. Water temperature was ranged from 26.4 to 26.8^{O}C. Salinity was maintained at 12±2 ppt and larvae of *Macrobrachium rosenbergii* withstand a salinity range from 6 to 12 ppt (Alam, et al. 1995). The pH values were between 7 and 8.5 and considered ideal for the larval rearing of giant freshwater prawn (Reddy, 1997). The average dissolved oxygen level varied from 5.53 to 5.70mg L^{-1}. Prawn larvae requires more than 4mg L^{-1} of dissolved oxygen (Reddy, 1997). Free carbon dioxide was not detectable in any of the cultured tanks. Ammonia–Nitrogen fluctuated from 0.03 to 0.05mg L^{-1} in the present study. Ammonia–Nitrogen in the range of 0.16-0.18 ppm found harmless for larvae of freshwater prawn (Alam, et al. 1993).

Conclusion

It may be concluded that diet (2), which had meat of shrimp and clam enhanced growth, faster metamorphosis and survival of prawn larvae when fed in combination with *Artemia* than diet (1), which contained fish meat, in the present study.

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