

## Determination of Optimum Male: Female Ratio and Salinity Level for Larval Production of *Macrobrachium rosenbergii* (De Man 1879) under Sri Lankan Conditions

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**ABSTRACT.** *Aquaculture is the fastest food growing activity in the world food sector. The global aquaculture production increased 13 times, from 8.53 million tonnes in 1970 to 64.82 million tonnes in 2007. Total annual global production of all freshwater prawn species was nearly 460,000 tonnes with a value exceeding US \$ 1.8 billion in 2007. Main freshwater prawn species commercially cultured in the world is Macrobrachium rosenbergii. Experiments were conducted at Brackish Water Prawn Breeding Station Pambala, Chilaw. First experiment was conducted to determine the suitable male and female ratio for Macrobrachium rosenbergii under Sri Lankan hatchery conditions. In this experiment males and females in 1:10, 1:8, and 1:5, 1:3 ratios were cultured under artificial conditions in the hatchery. Three cement tanks (8x6x1m) each separated into 4 parts by using plastic mesh frames were used. Each replicate had one blue clawed male and the experiment was repeated. Results revealed that 1:5 ratio gave significantly higher ( $P<0.05$ ) larvae, while in 1:10 ratio both eggs and larvae were in minimum numbers. The second experiment was conducted to determine the optimum salinity level for spawning of gravid females under Sri Lankan conditions. For this experiment, four fiber glass tanks each of 1000 L having two gravid females were used. Salinity was kept at 4, 5, 6, and 7 ppt and a constant temperature of 28°C was maintained. Experiment was repeated three times. Results revealed that 5 ppt salinity gave the optimum conditions. These results could be used to obtain higher production of *M. rosenbergii* seeds under Sri Lankan conditions.*

**Key Words:** *Macrobrachium rosenbergii, Male: female ratio, Salinity level.*

### INTRODUCTION

The giant freshwater prawn (*Macrobrachium rosenbergii* (De Man, 1879) is one of the most economically important species of prawns in the world. World aquaculture production of *Macrobrachium rosenbergii* increased to 118,501 tonnes, having a value US \$ 410 million in 2000. Total annual global production of all freshwater prawn species was nearly 460 000 tons with a value exceeding US \$ 1.86 billion, in 2007. The People's Republic of China is the largest freshwater prawn producer in the world. Fresh water prawns have been reared in captivity, either through introducing wild juveniles or by trapping them, along with other crustaceans and fish in tidal ponds and paddy fields in Indian sub-continent (Wickins, 1976).

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However, modern aquaculture of this species has its origin in early 1960s. Ling (1977) and Ling and Costello (1979) reported that rearing of freshwater prawn larvae required brackish water conditions for survival. Adult prawns living totally in fresh water and streams have been collected from 160 km distance from the sea. The distribution of natural stocks of this species in Sri Lanka is abundant and its exploitation from the wild is in the increase (Samarasinghe, 1983). Expansion of the *Macrobrachium rosenbergii* culture in Sri Lanka is limited mainly due to lack of prawn seeds (4.1million prawn seeds were produced in 2008). Therefore, good breeding techniques have to be developed under hatchery management conditions and upgrading of prawn seed production is essential in increasing freshwater prawn production in the country. Optimum sex ratio for maximum oviposition in *Macrobrachium rosenbergii*, Malecha (1983) suggested a ratio of 1: 4, 1:5, while Verghese *et al.* (1992) recommended 1:3, 1:4 or 1:5, and Sureshkumar and Kuroup (1998) suggested a 1: 4 male: female ratio. Consequently, based on these results for Sri Lankan hatchery conditions 1:3 1:5 1:8 male:female ratios were recommended. Freshwater prawn culture becomes more popular in the inland fisheries sector today. Therefore, development of mass scale seed production techniques is very important for popularizing freshwater prawn production in Sri Lanka. The objective of this study was to determine a suitable male: female ratio and optimum salinity level for spawning and hatching of eggs under Sri Lankan hatchery conditions.

## MATERIALS AND METHODS

Experiments were conducted at Brackish Water Prawn Breeding Center, Pambala, Chilaw. Three cement tanks (6 m x 4 m1m) were partitioned into four equal parts, by using plastic mesh and wooden frames, in order to prevent movement of prawn from one area to the other. Same water quality parameters were maintained in each replicate. Feed was supplied for the broodstock at 5% of body weight and two times per day. Temperature and pH were maintained at 28-30°C, and 7.5-8.0 ranges, respectively. Ammonia level was maintained at less than 0.6mg/L level during the 21 day culture period by supplying running water. To the four replicates of each treatment, randomly stocked one blue clawed male, (TL means 280 mm.) and female (Fig. 1).



**Fig. 1. Blue clawed male (a) and Gravid female (b)**

Mean total length in 1:10, 1:8, 1:5, 1:3 ratios, (TL for male 280 mm, and female 225 mm). Experiment was performed for two culture cycles. Colour of the egg, in each gravid female

was observed. Gravid females were harvested after 21 days and the weight of each prawn, total length and egg weight were obtained. The second experiment was to determine suitable salinity level for gravid females to spawn. In this experiment, four fiber glass tanks 1000L each and salinity levels of 4, 5, 6 and 7 ppt were maintained. Each treatment had six replicates. Temperature was maintained at 28°C using heaters (Model: 66, IMP.GAL.300W, Automatic). Salinity was measured by using a refractometer (type Milwaukee), pH measured using an electronic pH meter and ammonia by using a test-kit (Hatch Test Kit, FF-IA,). A completely randomized design was used for the second experiment. Logistic linear regression model was used to evaluate the hatching probabilities of the different male : female ratios. ANOVA were used for the mean separation of treatments, body weight and eggs weight of the prawns.

## RESULTS AND DISCUSSION

### Experiment 1

Water quality parameters in the treatments always ranged between the optimum values (Table 1).

**Table 1. Water quality parameters measured (Experiment 1)**

Parameter	Mean ± SE	Range
Temperature(water) (°C)	28.7 ± 0.048	28-31
Temperature (air) (°C)	29.1 ± 0.044	28-32
pH	7.8 ± 0.048	7.0-8.5
DO (mg/L)	6.06 ± 0.047	5.0-7.0
Hardness CaCO <sub>3</sub> (mg/L)	87.08 ± 1.018	50-100
Ammonia (mg/L)	0.46 ± 0.036	<0.5

Ammonia concentration was always maintained below 0.5 mg/L by having a running water system. It was essential since *M rosenbergii* was fed with highly protenacious diet, which consisted of oyster mussels, fish and squids. New and Valenti (2000) have also recommended a highly protenacious diet to be given two times a day. New and Singholka (1985) have indicated that pH between 7- 8.5 and temperature between 28-32 °C should be maintained in the hatchery. They have also recommended that total hardness should be less than 100 mg/L CaCO<sub>3</sub> in the water. When 1:10 mal:female ratio was used, the number of eggs and larvae was found to be minimum (13575 ± 24). A significant difference (p <0.05) was observed between 1:8 and 1:3 ratios (Table 2).

**Table 2. Number of eggs and larvae production (Experiment I)**

Treatment	Quantity of eggs	Weight of the eggs (g) SE	Larvae numbers	Total Length of Female (cm) SE
1:10	24171 <sup>b</sup> ± 32	13.5 ± 0.76	13575 ± 24	22.0 ± 0.96
1:8	25636 <sup>b</sup> ± 26	15.5 ± 0.61	17595 ± 20	23.6 ± 0.73
1:5	34294 <sup>a</sup> ± 19	19.1 ± 0.45	24427 ± 16	23.0 ± 0.50
1:3	26280 <sup>b</sup> ± 27	20.0 ± 0.63	16509 ± 21	22.6 ± 0.78

(<sup>a, b</sup> values are significantly different ( p < 0.05 ).

According to results, a significant difference ( $P < 0.05$ ) was observed among the four treatments with respect to total eggs laid (Table 2). Varghese *et al.* (1992) recommended a sex ratio between 1:3.75-1:4.75 for maximum oviposition. Malecha (1983) has suggested 1:4 - 1:5 while Sureshkumar and Kurup (1998) have recommended 1:4 for optimum oviposition and hatching.

New and Singholka (1985), observed that a female *M. rosenbergii* can lay between 80,000 to 100,000 eggs per spawning when they are fully matured. In contrast, in this experiment the number of eggs spawned ranged between 24,171 and 34,294. New and Singholka (1985) have indicated that when *M. rosenbergii* breed for the first time, number of eggs ranged between 5,000 to 20,000. Therefore, prawns used in this experiment cannot be first brooders. Based on the results (Table 2), 1:5 would be the best combination of the male: female ratio to get highest larvae ( $24427 \pm 16$ ). However, the male: female ratio of 1:10 was found to be yield the lowest number of larvae among the treatments used ( $13575 \pm 24$ ).

### Experiment – (2)

Water quality parameters were maintained under optimum conditions in Experiment 2 by having a running water system. Temperature was kept at 28°C by using heaters. Continuous aeration was also provided (Table 3).

**Table 3. Water quality parameters in Experiment 2.**

Parameter	Unit	Mean $\pm$ SE	Range
Temperature (water)	°C	28.0 $\pm$ 0	28-31
pH	-	7.8 $\pm$ 0.05	7.0-8.5
DO	mg/L	6.13 $\pm$ 0.04	5.0-7.0
Hardness	CaCO <sub>3</sub> mg/L	86.11 $\pm$ 1.27	50-100
Ammonia	mg/L	0.43 $\pm$ 0.03	<0.5

Adult prawns were tolerant to a wide temperature range of 18-34 °C (New & Valenti, 2000). New & Singholka (1985) indicated that a pH between 7.0 - 8.5 and temperature of 28-31°C could be maintained in a hatchery. They also recommended a hardness level of less than 100mg/L CaCO<sub>3</sub> in the hatchery.

**Table 4. Numbers of eggs and larvae produced in different salinities**

Salinity Level (ppt)	Mean Egg No.	Egg weight (g)	No of Larvae	Total length (cm)
4	24360 $\pm$ 16	13.6 $\pm$ 1.15	1500 $\pm$ 10	22.5 $\pm$ 0.60
5	24300 $\pm$ 23	13.5 $\pm$ 0.53	17830 $\pm$ 20	22.2 $\pm$ 0.66
6	24709 $\pm$ 23	13.7 $\pm$ 0.53	17052 $\pm$ 19	21.1 $\pm$ 0.69
7	27600 $\pm$ 24	15.3 $\pm$ 0.57	4258 $\pm$ 19	22.4 $\pm$ 0.69

Results of Experiment 2 revealed that a salinity level of 5 ppt and 6 ppt provided optimum conditions for spawning and hatching (Table 4). The number of eggs spawned did not show a significant ( $p > 0.05$ ) difference (4ppt, 5ppt, 6ppt, and 7 ppt) among all the treatments (Table 3), indicating that salinity level does not have a significant effect on spawning. However, a significant difference ( $p < 0.05$ ) was observed between the numbers of larvae produced in the four treatments (Table 4).

Salinity of 4 ppt produced few larvae, while at 7 ppt larval production was less than 5 ppt and 6 ppt levels. According to New & Valenti (2000), hatching occurs naturally under estuarine conditions and egg hatchability is higher in brackish water than in freshwater. Caluwe *et al.* (1995) reported that a combination of 6 ppt salinity with temperature of 26 to 28°C is optimal for the incubation of *M. rosenbergii* eggs. However, Gomez (1987), observed that temperatures above 30°C levels causes problems due to development of protozoa and other microorganisms. According to the results, a male: female ratio of 1:5 and a salinity level of 5-6 ppt provides the optimum conditions for spawning and hatching of *M. rosenbergii* eggs under Sri Lankan hatchery conditions.

## CONCLUSION

Based on the findings of this study, male: female ratio of 1:5 and a salinity level of 5-6 ppt provide the optimum conditions for spawning and hatching of *M. rosenbergii* eggs under Sri Lankan hatchery conditions. Therefore, these results will be important for the mass scale prawn seed production in the country. Moreover, further research is needed in breeding for the development of freshwater prawn culture sector in Sri Lanka.

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