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# Megalopa production of commercially important long eyed swimming crab *Podophthalamus vigil* (Fabricius)

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### ABSTRACT

Seed production technology is very much essential to introduce a new crab to aquaculture. In the present study a mass larval culture experiment was tried with commercially important long eyed swimming crab Podophthalamus vigil. The complete larval development of P. vigil consisted of five zoeael and one megalopal stages. Total duration for zoea I to III required 3-4 days, while zoeae IV and V took 2-3 days and V zoeae required 3 days to metamorphose into megalopa stage. The complete larval development took a span of 12-15 days. The survival rate was drastically decreased from I zoea to megalopa and the final survival was estimated as 3%. The maintenance of good water quality and hygiene during the larval culture is essential to improve the survival of P.vigil.

Key words: Podophthalamus vigil, Artemia nauplii, Brachionus plictilis, zoea, megalopa

#### INTRODUCTION

The focus of the present day aquaculture was increasingly shifted towards the development of hatchery technology and crab farming to meet the human protein demand. In India the shrimp culture is suffering by numerous problems especially viral diseases and pollution. So the farmers are not showing any interest towards shrimps but turn to other crustacean. Even though most of the crabs are high potential for aquaculture. Still the crab farming is an infancy stage; the obvious reason is lack of viable hatchery technology. Mass larval culture experiments were carried out on both commercial and non-commercial crabs all over the world. But, the percentage of survival at the end of the larval cycle is very low. Studies on the mass seed production technology were under experimentation for mud crabs with low survival rates, but such studies are scanty in the sea crabs (1,2) in general and commercially important long eyed swimming crab *Podophthalamus vigil* in particular especially in Indian perspective. So in the present study larval culture experiment was made in *P.vigil*. Since this species available through out the year along Parangipettai coast (3).

#### MATERIALS AND METHODS

Healthy gravid females of *P. vigil* were collected from the Parangipettai coastal waters. Crabs were acclimatized in holding tanks at a salinity  $35\pm1\%$ , pH 8.2 $\pm0.1$ , temperature  $27\pm2$ °C and photophase 12 hours light and 12 hours darkness with continuous aeration. Filtered seawater was used for the entire operation and 50% of water is exchanged every day. Polychaete worms were given as a feed to the brooders. Broodstock with gray eggs were transferred to the hatching tanks. Eggs hatched during the early hours of the day were collected and stocked (50 zoeae/litre) in the larval rearing tanks (50 litres). The entire larval cycle (zoeae I to V and megalopa) was carried out

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at 35ppt filtered seawater. During the experimental period, salinity, dissolved oxygen, pH and temperature were recorded by using a Centuary Water Analyzer Kit Model CK 711. Newly hatched zoeae I was fed three hours after stocking. Larvae were fed twice daily in the morning (8:00 AM) and evening (5:00 PM). All 5 zoeal and megalops stages were fed with rotifers (*Brachionus plicatilis*) and *Artemia* nauplii (OSI Brine shrimp eggs, USA) daily. In the morning larvae were fed with *B. plicatilis* at the rate of 5-15 per ml to zoeae I & II, 15-25 per ml to zoeae III, 25-40 per ml to zoeae IV&V and 70-80 per ml to megalopa. In the evening thawed *Artemia* nauplii, 2-20 per larvae for zoeae I, II& III and 20-50 per larvae for zoeae IV&V and megalopa were provided. The zoeae V on reaching the megalopa stage were provided with pebbles as substrate, oyster shells were suspended by nylon ropes as hide in the rearing tanks. Daily the rearing water was exchanged up to three fourths of the tank capacity. Dead larvae and exuviae were siphoned out during this time to prevent contamination. A cloth sock was used to prevent the loss of live zoeae. Mild and continuous aeration was provided (10 to 15 bubbles per minute) to the rearing tanks. Larval numbers were estimated daily. The experiment was terminated once V zoeae were metamorphosed into megalopa.

#### RESULTS

The regular monitoring of water quality parameters in the culture medium did not show much variation. Parameters like salinity-  $35\pm1ppt$ , dissolved oxygen-  $5\pm1$  O<sub>2</sub> ml/lit, temperature  $-30\pm1$ °C and pH - 7.3 to 8.2 were recorded daily. The complete larval development of *P. vigil* consisted of five zoeal and one megalopal stages. Total duration for zoeae I to III required 3-4 days, while zoeae IV and V took 2-3 days and V zoeae required 3 days to metamorphose into megalopa stage. The complete larval development took a span of 12-15 days. The survival rate was drastically decreased from I zoeae to megalopa and the final survival was estimated as 3% (Table 1).

Table 1 Intermoult duration and survival of	of different larval stages
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Larval stage	Intermoult duration (Days)	Survival(%)
I Zoea to II Zoea	3.28±0.32	70.11±1.18
II Zoea to III Zoea	3.15±0.39	48.3±2.15
III Zoea to IV Zoea	2.71±0.62	33.7±3.65
IV Zoea to V Zoea	2.56±0.13	10.1±3.15
V Zoea to Megalopa	3.25±0.40	313±0.12
Total days	12 - 15	

### DISCUSSION

In order to introduce a new species to aquaculture one of the important constraints is lack of proper seed production technology (4). To date broodstock development and hatchery seed production of crabs (in terms of percentage of survival) have been experimental, though the technology has developed for the production of crab seeds in many countries. Several studies related to the survival of the commercial portunid crab larvae have used brine shrimp, rotifers and algae as food, since the nutrition turns to be vital to the larval survival. Apart from the live feed, the water quality parameters such as salinity and temperature will also play an important role in the larval growth and survival during mass culture experiments.

Water quality (temperature, salinity, nutrient and hygiene) is a significant factor in larval survival (5). Survival and longevity of marine invertebrate larvae are influenced by abiotic factors such as water temperature and salinity, and by biotic factors such as food availability, food quality and predation. In the present study the water salinity was maintained at 35 ppt since the spawning, embryogenesis and hatching of eggs generally takes place in coastal regions. The results of the present study indicate that the most suitable range of temperature for crab larvae was found to be 30 to 32.5°C. The higher mortality rate of the zoea I, especially during the first three days of culture might be due to fluctuations in water temperature in brooder's tank and the mass culture tanks. Mann *et al.* (6) have also been noticed that the temperature shock caused larval stress and mortality. The better survival evidenced in the present study with the larvae of *P. vigil* might also be due to the higher temperature ( $30\pm1^{\circ}$ C). The previous studies on the effect of temperature on larval rearing revealed that the larvae could not survive in low temperatures. The low survival rate was evident when the larvae of *S. serrata* reared at low mean temperatures, *i.e.*, at 27.5°C by Ong (7,8), at 24°C by Du Plessis (9), at 22°C by Brick (10) and at 27°C by Haesman and Fielder (11). Temperature had a strong influence on the survival of *P. pelagicus* larvae, with marked negative effects at 22.5°C.

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The amount and quality of food supply are the chief factors regulating the duration of larval development (12,13). Less diet will prolong larval development, thus, increasing the risks of larval mortality due to predation and starvation. Suitable diet for larvae should meet three general criteria: namely, they should be an appropriate size for easy capture and consumption, they should be present at an adequate concentration, and they should contain essential dietary nutrients (13). The seed production of aquatic species is almost entirely depending on the successful production of live food organisms, principally rotifers, followed by *Artemia*. The superiority of the live food organisms in larval nutrition over existing compounded diet is partly due to the availability of exogenous enzymes through the live food, which in combination with endogenous enzymes of the animal lead to efficient digestibility (14). Young animals with less developed digestive system benefit more from exogenous enzymes than do adults. The exact quantity of food required at each stage cannot prescribe as it depends on the utilization of the feed by the larvae and must be judged visually by the operator.

In the present study the zoeae were initially fed with *B. plicatilis*, since the small size of first zoeae refused to feed on *Artemia* nauplii. *B. plicatilis* is smaller in size and can be ingested completely by small decapod crustacean larvae. Rotifer gut is usually filled with bacteria and algae, which could provide additional nutrition for the larval forms of decapods. Emmerson (15) reported that caloric content of rotifers per gram ash dry weight is not significantly different from that of *Artemia* nauplii. Sulkin (16) reported that the smaller size and slower swimming speed of *B. plicatilis* apparently allow their capture and manipulation by small crab zoea. Soundarapandian *et al.* (17) observed that the *Macrobrachium malcolmsonii* early larval stages apparently graze on the appendages of *Artemia* nauplii but could consume entire rotifers.

Combination of *Artemia* nauplii and rotifer obtained mixed results when fed to the crab larvae by different authors. Brick (10) showed that mud crab larvae fed on *Artemia* nauplii alone had a higher survival rate than those fed on rotifers. He suggested that the addition of rotifers might have contributed to the deterioration of the culture medium, through oxygen consumption or release of metabolites, without providing any nutritional benefit for the larvae. McConaugha (13) reported that *Rithropanopeus harrisii* larvae fed on rotifer could not metamorphose due to low lipid content and low feeding efficiency. Baylon and Failaman (18) demonstrated that the rotifers are more important than *Artemia* nauplii for maintaining the survival rate of the first and second zoeal stages, where as supplying *Artemia* or rotifers as the sole prey failed to maintain the survival rate of mud crab. In most of the previous studies, successful seed productions obtained when rotifer and *Artemia* nauplii were used as feed (1). Successful seed production was reported in *P. trituberculatus* offered with rotifer and *Artemia* nauplii (2,19, 20, 21,). Minagawa and Murano (22) recommended mixed diets (*Artemia* nauplii + rotifer) for mass seed production of frog crab, *Ranina ranina*. In the present study, both rotifer and *Artemia* nauplii have been offered to the larvae of *P. vigil* as in the previous study. However, the survival rate is not encouraging. Various reasons are attributed for the lower survival even though standard live foods were used.

The final survival rate of *P. vigil* larvae in the present study was 3%. The survival rate is comparatively higher than that reported in *S. serrata* (13). The reasons could be that the feeding schedules, incorporated combinations of *B. plicatilis* and *Artemia* nauplii. Similarly, Zainoddin (23) also pointed out that the combination of *B. plicatilis* and *Artemia* nauplii served as feed gave better survival. The survival rate in the zoeal stages (I to IV zoeae, 65 to 35 per cent) achieved by Haesman and Fielder (13) on the larvae of *S. serrata* was due to high concentration of *Artemia* nauplii (5 to 30 nos. per ml) used once in a day. Larvae of the related family *P. trituberculatus* have been reared with success (over 60%) on the same species of rotifer in combination with *Artemia* by Hue *et al.* (19). Minagawa and Murano (22) recommend a combination of diets to mass rear the larvae of *R. ranina*.

In the present study mortality in the first two zoeal stages of *P. vigil* regardless of the feeding regimes is quite comparable with the results obtained by Soundarapandian *et al.* (1) in *P. pelagicus* and Nunnam John Samuel *et al.* (2) in *P.sanguinolentus*. They point out from their experiments that initial mortality during the first two days of the experiment occurred often and relatively high regardless of feeding regimes and are rather unpredictable. They related the mortality to the low viability of the individual larva. Similarly mortality during the zoea V and Megalopa stages was either before moulting, during moulting (includes incomplete moulting) in *P. vigil* larvae. The possible reason cited by Anger *et al.* (24) is that mortality due to depletion of reserves resulting in larval inability to catch the prey. Similarly, Rosenberg and Costlow (25) suggested that the majority of the larval population is preparing for the premetamorphic moult to Megalopa. Likewise Costlow and Bookhout (26) and Christiansen and Costlow (27) have observed high mortalities in the larvae of *R. harrisii* at the premetamorphic stage. They attribute two reasons for

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such mortality -1. The larvae at this premetamorphic stage are extremely susceptible to unfavourable environmental conditions at this time of life cycle and 2. The metabolic cost of metamorphosis is very high and appears to decrease the capacity of larvae to counteract these unfavourable conditions. Cannibalistic tendency was observed from megalopa onwards and it was the main reason for the higher mortality. The complete morphological changes from V zoeae to megalopa also one of the reason for high mortality.

The maintenance of good water quality and hygiene during the larval culture results in higher survival percentages. The hygiene begins with the preparation of the broodstock and continues up to the metamorphosis of megalopa to crablets. The aim is to restrict the growth of potential pathogens, including bacteria, fungi, viruses and protozoa in the culture system. It can be safely assumed that all inputs into a culture tank are potential sources of infection that may reduce rates of larval survival and metamorphosis. All tanks and equipment used in the culture must first be effectively sterilized following standard methods before use as a simple precautionary measure.

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