

Embryology of Commercially Important Portunid Crab *Scylla serrata* (Forsk.)

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Abstract: Embryonic development of the mud crab, *Scylla serrata* (Forsk.) was studied. Five embryonic stages, viz., Blastula, Gastrula, Eye placode, Pigment and Heart beat were identified and described in detail. The size of the developing egg increased at every stage. The colour of the egg was initially yellow and it gradually changed to orange, brown and black colours. The hatching success of freshly hatched I zoea was 80%. The salinity test showed 35 ppt as optimum salinity to rear the larvae of *S. serrata*.

Key words: Blastula, embryonic development, eye placode, gastrula, pigment and *Scylla serrata*

INTRODUCTION

After the disease outbreak in shrimp farming, crabs have become candidate species for aquaculture because of their export potential. The crab culture is presently dependent on wild caught seeds, which are not sufficient (Keenan, 1999; Fortes, 1999). The natural seed availability is also declining due to indiscriminate collection of juveniles for farming. The collected seeds are not of uniform size and availability throughout the year is also doubtful. To solve this problem developing hatchery technology is need of the hour. The mud crabs, *Scylla serrata* and *S. tranquebarica* are commercially important and are available throughout the year along Parangipettai coast (John Samuel, 2003). So far there is no established hatchery for these mud crabs and even if there is one the survival rate is very less i.e., only 10-15% because of mass mortality in the first and second zoeal stages. Therefore a study on their embryology can form the baseline to assess the quality and efficiency of the eggs to hatch into healthy first zoea. So, an attempt has been made here to study the embryonic development of the mud crab, *S. serrata*.

MATERIALS AND METHODS

Healthy gravid females of *S. serrata* (Forsk.) with all the body parts intact and with early broods (yellowish orange coloured eggs) were collected from Parangipettai (Lat. 11° 29'N and Long. 79°46'E) coast. The crabs were given prophylactic dip in 100 ppm formalin for 10 min. and retained in separate cement tanks containing sand-filtered seawater and the tanks were provided with adequate aeration. During the experimental period, optimum environmental parameters were maintained

(salinity 30-35 ppt., pH 7.8-8.2 and temperature of 27 to 30°C). The crabs were fed with mussel and clam meat once a day. Daily colour changes if any, in eggs during incubation period was noted. The diameter of the eggs was measured using a micrometer mounted in the ocular piece of a dissecting microscope. Few eggs were removed from the brood daily, examined under the microscope for colour change corresponding to development and the length of incubation period was noted (Srinivasagam *et al.*, 2000). The quality of newly hatched larvae was assessed through different tests like salinity (30, 35 and 40 ppt) and starvation, by exposing the larvae to different stress tests by sudden changes in the salinity and temperature. After these tests the active zoeae were transferred to the mass culture tanks for further development. Apart from these two tests the egg and larval quality was also ascertained by the following factors, which play an important role in the quality of the larvae.

Fertilisation Success: It is the ratio of developing to dead eggs. This ratio is calculated by removing eggs from the berry and examining them under a microscope.

Rate of Development: is the relationship between egg diameter and temperature of the surrounding environment. This is achieved by measuring samples of eggs daily under a microscope as well as recording the temperature. It can be a useful tool to predict the day of hatching.

Fecundity: The total number of eggs extruded in a single spawning. Fecundity was calculated by weighing the female directly after extrusion and once again after eggs were hatched. From this the total weight of egg mass was calculated as well as the fecundity.

Hatching Success: The total number of healthy larvae produced. This is achieved by calculating the number of larvae per milliliter of water in a known volume of water.

RESULTS

The egg of mud crab passes through different colours with its gradual development. The colour of the egg, immediately after extrusion is yellowish orange and it gradually transforms into orange, brown, and black colours. The incubation period is 7-9 days. The eggs at the time of oviposition were quite distinct and large. They could be divided into five stages, viz., blastula, gastrula, eye placode, pigment and heart beat (Amsler and George, 1984). A gradual change in size of the egg was noted in the five stages (Plate 1a).

Embryonic Developmental Stages:

Stage-I-Blastula: Eggs were round, golden yellow in colour and were undeveloped with mass of undifferentiated cells. Yolk granules were denser. Cleavage and gastrulation were not clear. The diameter of the freshly laid egg was 0.40 mm (Plate 1b).

Stage-II-Gastrula: Eggs were round and deep yellow or yellowish orange in colour. The space between the egg wall and the inner developing embryo was visible. The diameter of the egg was 0.52 mm (Plate 1c).

Stage-III-Eye placode: Eggs were round and orange in colour. Yolk granules were not dense. Segmentation and organogenesis were distinct. The eyespots appeared scarlet crescent. The diameter was 0.60 mm (Plate 1d).

Stage-IV-Pigment: Eggs were brown in colour with slightly elliptical shape. Appendages of embryonic larvae were pigmented. The diameter was 0.65 mm (Plate 1e).

Stage-Heartbeat: The eggs were dark brown or black in colour. Eyes were round in shape. Heartbeat was vigorous. The diameter of the egg was 0.73 mm (Plate 1f).

The larvae after hatching when subjected to different stress tests, showed marked survival rate and success. In the salinity test, the larvae showed good resistance and 100% survival at 35 ppt and reached the second zoea within 3-4 days. At 25, 30 and 40ppt the larvae were active only for 2 days and the survival rate was very less. In the starvation test, all the larvae died within two days. The survival rate in the first day was 70% and in the second day it was 20% and no zoea survived on the third day. The hatching success of the freshly hatched I zoea was 80%.

DISCUSSION

The berried crabs were collected from the marine zone of Vellar estuary where the salinity of the water is

usually high. Spawning occurs in high salinity waters in the lower estuary, tidal passes, and near shore areas. Many, if not all, females spawn twice, which may be during the same season or over two seasons. Development and hatching of eggs takes place under a relatively wide range of salinity and water temperature, but this range narrows down in successive zoeal instars (Lin *et al.*, 1994; Iin *et al.*, 2003)

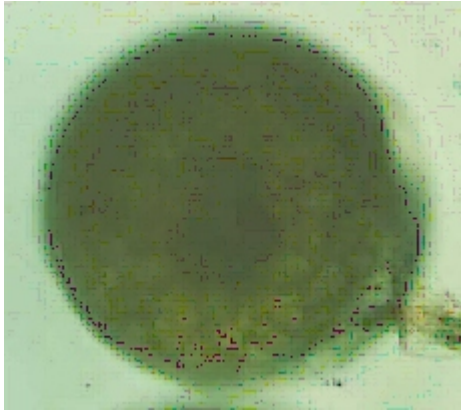
In general Portunids lay around 1 to 6 million eggs per spawning. Fecundity of *S. serrata* ranges from 1 to 3 million eggs whereas in *S. tranquebarica* it is upto 5 million (Srinivasagam *et al.*, 2000). The eggs when deposited are bright orange or yellowish orange, but they become yellow, brown and then dark brown or black before hatching. The colour change is caused by absorption of the yellow yolk and development of dark pigment in the eyes (Sundaramoorthy, 1987; Krishnan, 1987; Vijayakumar, 1992; Veera Ravi, 1994; Parimalam, 2001). Veera Ravi (1994) reported that as the progression of development occurs, the embryo decreases in dry weight, stage by stage, as it utilizes the yolk. Based on the egg size, Shakuntala and Ravichandra Reddy (1982) classified the crustacean embryogenesis into two groups as (1) larger eggs utilizing more lipid than protein and (2) smaller ones utilizing more protein than lipid.

The eggs swell as they develop so that by the time they are ready to hatch, they are roughly double their new-laid volume. During development, the colour of the egg changes through brown to grey as the yolk is used up and the outline of the embryo become visible. The eyes and pigment spots appear first followed by the outlines of the abdomen and cephalothorax (Warner, 1977).

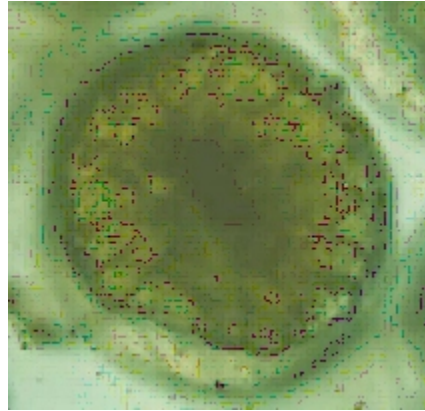
Hatching usually occurs in the early hours of the day in *S. serrata* as all the Portunid crabs generally hatch in the early hours. During the hatching process, the fully developed I - zoea comes out of the egg cases and swims freely in the water column. Davis (1965) reported that in the process of hatching, a period of swelling of the eggs followed by osmotic swelling of the inner egg membrane at the start of hatching. The swelling inner egg membrane then ruptures the chorion by pressure from within the embryo. The inner membrane is subsequently ruptured by mechanical action of the larval abdomen.

The incubation period in the present study took 7-9 days. The reduced incubation period might be due to the good water quality parameters and feed that were maintained during the rearing period. In general, the incubation period for the mud crabs, *Scylla* spp. ranges between 7 and 13 days depending on the conditions maintained.

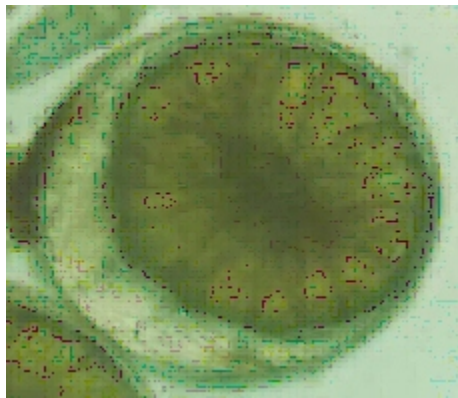
The water quality and feed are important criteria in rearing berried crabs, and if they are not maintained properly disease may attack leading to hatching of unhealthy larvae and mortality in the early stage itself. Iin *et al.* (2003) observed that the egg quality, egg hatching rate and the quality of newly hatched zoea were very good when berried crabs were fed with formulated feeds



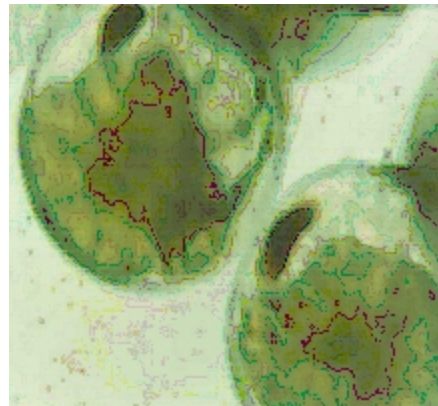
(a) Newly extruded egg



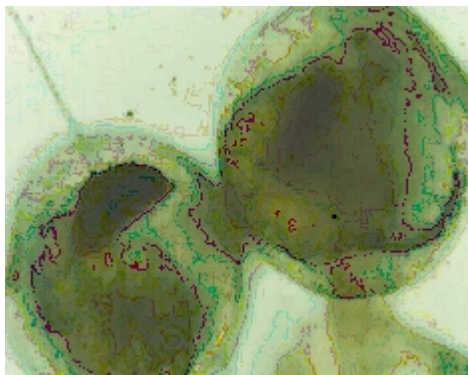
(b) Blastula



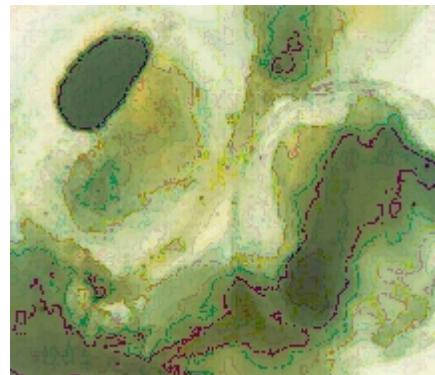
(c) Gastrula



(d) Eye placode



(e) Pigment



(f) Heart beat

Plate 1: Sizes of the egg in different embryonic stages

compared to normal diets. The mortality of eggs has been attributed to fungus, predation, and suffocation in fouled water and changes in temperature. On an average, only one out of a million eggs survives to become a mature adult (Van Engel, 1985; Michael and Dean, 1984).

The quality of eggs produced by individual females has been highly variable. In order to select the best eggs for incubation and subsequent rearing, an estimate of egg quality had to be established and this could be achieved immediately after extrusion. Churchill (2001) reported

that, the newly hatched larvae could be subjected to a variety of stress tests including ammonium, salinity, formalin and starvation and the results of these tests would ultimately be the deciding factors in classifying eggs as either good or poor quality. Rearing of healthy larvae will be possible only if the embryonic development is studied well.

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