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LUNAR RHYTHM-DEPENDENT SPAWNING AND LARVAL RELEASE IN THE CONTINUOUSLY BREEDING MANGROVE CRAB, *SESARMA QUADRATUM* (DECAPODA: BRACHYURA)

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***Abstract*— The present work is a comprehensive study of embryonic development and hatching rhythms of the mangrove crab, *Sesarma quadratum*. The ovigerous females belong to the cohort of 9 – 22 mm carapace width. The eggs of *S. quadratum* are attached to the fine ovigerous hairs along the setae of the pleopods by a stalk called funiculus secreted by the vitelline envelopes of the eggs. Mean egg count in *S. quadratum* is found to be 9587 ± 3526 per female, signifying the wide variation existing among individuals. Fecundity was positively correlated with female size, an important factor that contributes to the variability in egg production. Six stages of embryonic development were categorized by closely examining the daily progress. On the first day of spawning, the eggs remain uncleaved. Then onwards, as cleavage progresses, it passes through blastula, gastrula, pigmented stage, eyespot stage, appendages stage and eventually, the zoea. The average diameter of an undivided egg is found to be 0.025 mm, which subsequently increased to 0.038 mm as embryogenesis proceeds; water accumulation appears to be the key factor for the egg-size increase during development. Upon completion of the development, the egg capsule breaks and the hatched zoea are released into the ambient water by "special fanning movement" of the female's abdomen. The egg cases are soon removed by the peculiar "flapping behaviour" of the female. Interestingly, spawning in *S. quadratum* is synchronous with lunar cycle; spawning rate is found to peak at the time of full moon/new moon. On an average, each spawned egg in *S. quadratum* would require 14- 15 days for completion of embryogenesis. And the larval release also exhibits strong correlation with lunar rhythms.**

Keywords-(lunar rhythm; Sesarma quadratum ; funiculus; ovigerous; zoea)

I. INTRODUCTION

There exists considerable diversity with respect to the pattern of breeding among crustaceans, which apparently is correlated with the attempt of the species in question to cope with the respective ecosystem. While many of the decapod crustaceans are known to be annual breeders (releasing a single brood per year), others are consecutive breeders (releasing several broods a year). Significantly, populations of the same species, but geographically separated, could show differing patterns of breeding. The Calicut population of the field crab *Paratelphusa hydrodromous,* for example, is an annual breeder, while the Trivandrum population of the same species, is a consecutive breeder, releasing 3-4 broods a year [2];[1]. Breeding activity (spawning for instance) seems to be synchronous throughout the population in several species. Sunset seems to influence the larval release of those species inhabiting along the seashore [5]. Although crustaceans, including the brachyuran crabs inhabiting various ecological niches, exhibit diverse breeding patterns, we are still unable to identify precisely, the factors regulating this important physiological event. Keeping this in view, the present paper depicts the results of our close examination on the events related to spawning, embryogenesis and larval release in the mangrove crab, *Sesarma quadratum*, and its entrainment to lunar rhythms.

II. MATERIALS AND METHODS

Ovigerous females of *Sesarma quadratum* were collected from the mangrove swamps of the intertidal regions of Nileshwar (Kasaragod district, Kerala, India) (12° 15' N; 75° 16' E) by baiting and hand picking. The eggs were observed microscopically for the purpose of ascertaining the stages of embryonic development. The diameter of the eggs was measured by using an ocular micrometer while the volume was measured using the formula $V= 4/3\ \pi\ r^3$. Considering the onset of cleavage and the appearance of pigments, eyespots and appendages, the embryogenesis in *S. quadratum* has been classified into six stages viz., uncleaved stage, blastula, gastrula, eyespot stage, appendages stage and finally, the zoea stage.

III. OBSERVATIONS

The ovigerous females belonged to the cohort of 9 – 22 mm carapace width. Soon after spawning, the eggs are brownish and primarily spherical having yolk distributed in uniform fashion. Eggs at this stage have a mean diameter of 250µm (Fig.1.1) and mean volume of 49.063 mm3. Eggs appear in clusters attached to a common stalk by funiculus, and remain entwined to the setal hairs of the pleopods (Fig. 2). Cleavage begins within 6 – 12 hours after spawning, leading to the appearance of the blastomeres at one pole of the egg (Fig.1.2). The embryo now termed as a blastula, has a diameter of ~275µm. The developing egg remains in blastula stage only for one day.

Morphogenesis starts during gastrula stage, characterized by the appearance of red or brown chromatophores. Yolk gets depleted to about one third of the embryonic volume by the 5th day (Fig.1.3; Fig.1.4)). The gastrula with a diameter of ~287.5µm lasts for three days. By the 6th day, eye spot becomes visible (Fig.1.5). The eyespots get fully developed by the 8th day (Fig.1.6). Yolk depletion continues. The developing egg at the eyespot stage has a diameter of 300 µm and an egg mass index of 14.78. A pulsatile heart appears by the 10th day. Appendages bud out along with the emergence of the cephalothorax (Fig.1.6). The rostral spines also become visible when the appendages appear. Abdomen becomes more prominent (Fig.1.7). Only a few yolk spherules are left at this stage (13th day). The cephalothoracic appendages become more pronounced and the development is completed by the 14th day. By the 14th / 15th day, the fully developed zoea larva, by its characteristic undulatory motion, emerges out of the embryonic case. Larval release occurs at night in the full moon or new moon day. The larvae, as soon as released appear as dazzling structures making rhythmic vertical migrations (Fig.1.8). Zoea is about 1mm long and is characterized by the presence of cephalothorax (315µm in length) with rostral spines (125µm long) and a posteriorly placed abdomen. Well developed cephalic and thoracic appendages are characteristic of zoea. It possesses a 540 µm long bifurcated tail.

Our close examination of the population (with samples observed twice in every week) reveals that spawning is in synchrony with the lunar cycle, with peaks occurring at the time of full moon and new moon. Usually, eggs are released at night in most of the instances. The eggs of *S. quadratum* are attached to the fine ovigerous hairs along the setae of the pleopods by a stalk called funiculus secreted by the vitelline envelopes of the eggs. Mean egg count in *S. quadratum* is 9587 ± 3526 per female. Females with a wide carapace and a broad abdomen produced numerous eggs while that with a small carapace width had small number of eggs. Egg volume increased during embryogenesis, as a result of accumulation of more water. Brooding females show specific reproductive behaviour for ensuring oxygenation to egg mass by regular standing, abdominal flapping and pleopod beating. When development is complete, the egg capsule breaks and the zoea hatch out and are released into water by special fanning movement of females' abdomen. The egg cases are then soon removed by the peculiar flapping behaviour of the female.

To determine the lunar rhythm in spawning and the larval release, the female population was observed for their egg size and development stage, which are in turn traced back to the date of extrusion. The mean number of days required for completing the larval development was determined by rearing the ovigerous females in the laboratory and comparing the data with the same population in the field. Our observations revealed that both the laboratory - maintained ones and the field crabs have synchrony not only in the ovarian development but in embryogenesis and subsequent larval release as well. The ovarian cycle as well as the embryonic development was completed in 14-15 days (Table.1). The spawning and the larval release usually take place in the 0 or ± 1 day of full moon and new moon (Fig.3). The Stage I ovary appears on the 0 or ± 1 day of the full moon (FM)/ new moon (NM); the same animal shows the immediate post spawn also. The first day of the FM/NM is characterized by the presence of blastula and Stage II ovary; this is followed by the gastrulation in the embryonic egg whereby the amount of yolk in the egg gets depleted. By the time the ovary enters the stage III; pigmented structures appear in the embryo and the eyespot start developing. As the ovary attains stage IV, the embryo becomes fully mature with the development of the appendages and the rostral spines.

IV. Discussion

S. quadratum, like many other malacostracans brood their eggs in their abdomen in the pleopods. The eggs enclosed in a capsule remain attached to the pleopods by the funiculus. When embryonic development gets completed, the egg capsule breaks and the zoea larva get released into water. Larval release is made possible by continuous fanning movement of females' abdomen [7]. Egg extrusion and larval release in *S. quadratum* follows a lunar rhythm with the peak releases in ± 2 days of the FM or NM. Both extrusion of eggs and hatching of larvae are generally programmed 'so that brood release occurs at a time appropriate for the growth of the young' [1]. The larval release occurs in the open water so that the planktonic larvae can disperse. The larvae are released into the flooding tide and this is most invariably done at night [3]. Hatching in synchrony with the lunar, tidal or daylight cycles may enhance larval survival through concentration, dispersal or by impeding visual predators, and may be an adaptation to high levels of planktivory [6].

In *S. quadratum* an ovarian cycle is completed in 15 or 16 days. The first stage of the ovary (OD<50µm) lasts for only one or two days while the second stage (OD 51-100µm) takes 3 days during which the ovary attains a yellow hue. Then yolk deposition starts and ovary turns into yellowish brown (OD 101-150µm). It extends to about 5 days. Thereupon a marked increase in the gonad index is registered and the ovary appears brown in colour (OD 151-200µm). This stage lasts for about three days followed by a dark brown stage (OD 200-225µm) for two or three days after which spawning occurs.

While the female is brood carrying a second clutch of eggs starts developing in the ovary and the maturation gets completed as the embryo is about to hatch. In that way, there occurs a synchrony in vitellogenesis and embryogenesis, in *S quadratum.*

Existence of a lunar periodicity in spawning and larval release, as shown by *S. quadratum* (present study) attracts much attention. Although the correlation between lunar periodicity and spawning has been suggested previously, its possible adaptive significance has been hardly discussed. In *S. quadratum*, the spawning is found to be 'timed' with larval release from the brood. Being a continuous breeder, *S. quadratum* would need the brood pouch to be free for the ensuing clutch of eggs to be spawned. This suggests that the synchrony existing between the ovarian growth and embryogenesis in the preceding clutch of eggs as an adaptive significance, a feature hardly reported in other brachyurans.

The egg number and its production periodicity were species-specific factors that reflected the reproductive strategy of every species [4]. The fecundity analysis of the present study did not only estimate the egg number, but it also represented the rhythms in which eggs were produced.

TABLE I. SYNCHRONY BETWEEN STAGES OF EMBRYOGENESIS AND GROWTH OF SUCCESSIVE BROOD

Days	Embryonic stage	Oocyte diameter(µm)in the ovary
0	Uncleaved egg	upto50
1	Blastula	51-100
2-5	Gastrula	100-150
6-10	Eyespot stage	151-200
11-13	Appendages	201-225
14/15	Zoea release	>225

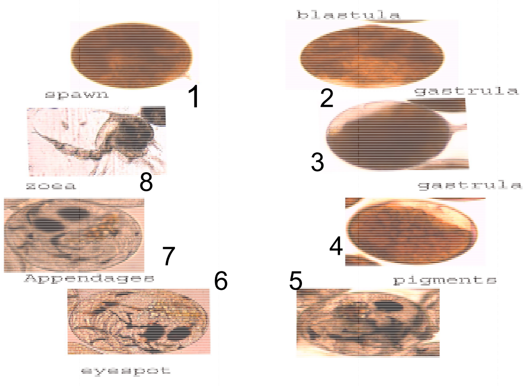

Figure 1. Stages of embryogenesis in *S. quadratum*.

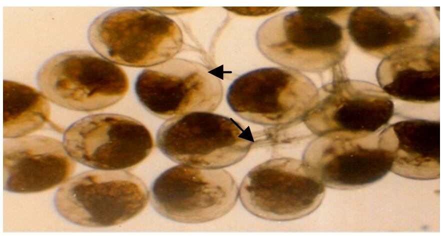

Figure 2. *Gastrula stage eggs with funiculus*

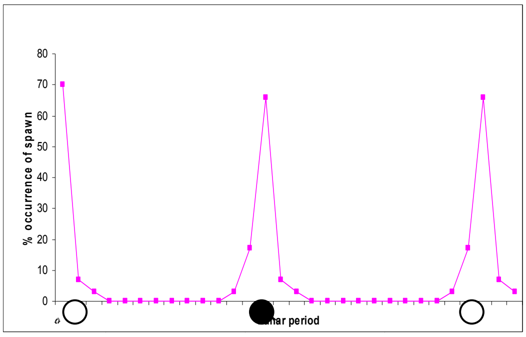

Figure 3. *Semilunar periodicity in spawning in S.quadratum*

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