

OBSERVATIONS ON THE MATING BEHAVIOR, SPAWN MASS AND LARVAL DEVELOPMENT OF *HYDATINA PHYSIS* LINNE, 1758 (CEPHALASPIDEA: HYDATINIDAE) FROM KARACHI

Itrat Zehra and Rukhsana Perveen
Centre of Excellence in Marine Biology, University of Karachi,
Karachi-75270, Pakistan.

ABSTRACT: Reproduction of *Hydatina physis* was studied in a population from Karachi, Pakistan, including mating and egg laying behavior, spawn characteristics, and development. Individuals first appear in the field in October and remain until March. The spawning occurs from mid-November till mid-February with a peak in December. During this period the individuals were also observed pairing.

In captivity, mating lasts for 30 minutes, second mating occurs two days later. Oviposition occurs in a very interesting and unusual manner. The mother turns "up-side down" with its foot fully expanded and the shell completely hidden underneath, the expanded foot serves as protective cover to the eggs. Eggs are deposited in a complexly folded mass with a short stem and an adhesive disc. Capsules, arranged in a single layer, contain 4-6 eggs each of which is 70 μm in diameter. Development is planktotrophic and veligers hatch after 14 days at a temperature of 26-28°C.

KEY WORDS: *Hydatina physis* - mating - spawn mass - larval development.

INTRODUCTION

Hydatina physis has a broad distribution, including South Africa, New Zealand, Japan, Hawaii, Caribbean and Canary Islands (Rudman, 1972). On the coast of Karachi, *H. physis* inhabits the muddy sand substrata among loose rocks, in association with cirratulid polychaetes, upon which it feeds. Some aspects of *H. physis* have been reported by Ostergaard (1950) and Baba and Hamatani (1956). However, informations on the mating behavior, oviposition and the embryonic development of this species are lacking. Here we present data on the reproduction, mating behavior, deposition of egg masses and the embryonic development of *H. physis* from Pakistan, and compare these data with those reported for other populations.

MATERIALS AND METHODS

A study of the populations of *Hydatina physis*, was conducted from September 1986 to August 1987. Samples of mature animals were collected from subtidal and intertidal rock pools in Karachi at Buleji (66°50'E; 24°N). Observations were also made on the specific substrata occupied by adults and the associated fauna and flora.

In laboratory, individuals of *H. physis* were maintained in glass aquaria (30x18x18cms) until they acclimated to ambient temperature. After acclimation the

individuals were transferred in pairs to glass bowls (20cm in diameter) filled with seawater, (changed daily) from the sampling site.

Cirratulid polychaetes, the adult's prey, were supplied in abundance daily. Assorted items (bivalve shell, small rocks, the seaweed *Sargassum muticum*), of almost equal dimensions, were also placed in the rearing bowls to provide a choice of substrata for spawning adults, in addition to the greater surface area offered by the bowl. Freshly laid spawn masses were transferred to glass dishes filled with continuously aerated seawater which was changed twice in a day for the first three days, and thereafter once in a day, until hatching occurred. Spawning adults and spawn were kept under continuous aeration.

Capsular morphology and dimensions of eggs were measured immediately after spawning. The spawns were kept under continuous observations at a room temperature of 26-28°C, until the veligers hatched. The drawings were made from live/fresh materials with the aid of camera lucida.

RESULTS

BREEDING HABITS:

The breeding season of *H. physis* extends from October to mid February. Prior to the onset of the breeding season the individuals aggregate (25/1.5m²) in the intertidal pools near low water mark. Most individuals were observed copulating during this time. Spawns were found from November to mid-February, with a peak abundance in December. However, the aggregations of the individuals last until mid-March.

The process of egg laying is very unusual and interesting as the animals lie on their backs instead of sitting on the foot. This up-side down position for spawning was observed both in the field and in the laboratory. However in the field the spawns were always found attached to *Sargassum muticum* whereas in the laboratory they were always found attached to the glass bottom.

MATING BEHAVIOR AND OVIPOSITION:

Mating was observed three times in captivity and 12 times in the field. No courtship behavior was noted. In captivity adults readily mated, when placed together in a glass bowl. As soon as an adult was added in the bowl containing another adult, the resident adult moved across and attached itself to the wall of the container. In the next stage, the other individual moved toward the resident one and oriented itself adjacently and in an opposite direction. Within ten minutes, each individual protruded its penis to enter into the genital opening of the other. The mating process lasted for about 25-30 minutes. A second mating occurred after an average of two days. No protective concealment behavior was observed. In the laboratory the copulating pairs died after 24-48 hours following the second mating.

At the time of egg laying, the ovipositing individual moved toward the water surface. After about 10 minutes, the foot was completely expanded, the animal turned and laid on its back with its foot facing upwards. The egg ribbon was deposited with its foot facing upwards. The egg ribbon was deposited ten minutes following this up-side down posture. The whole ribbon was deposited in about 30 minutes. On completion

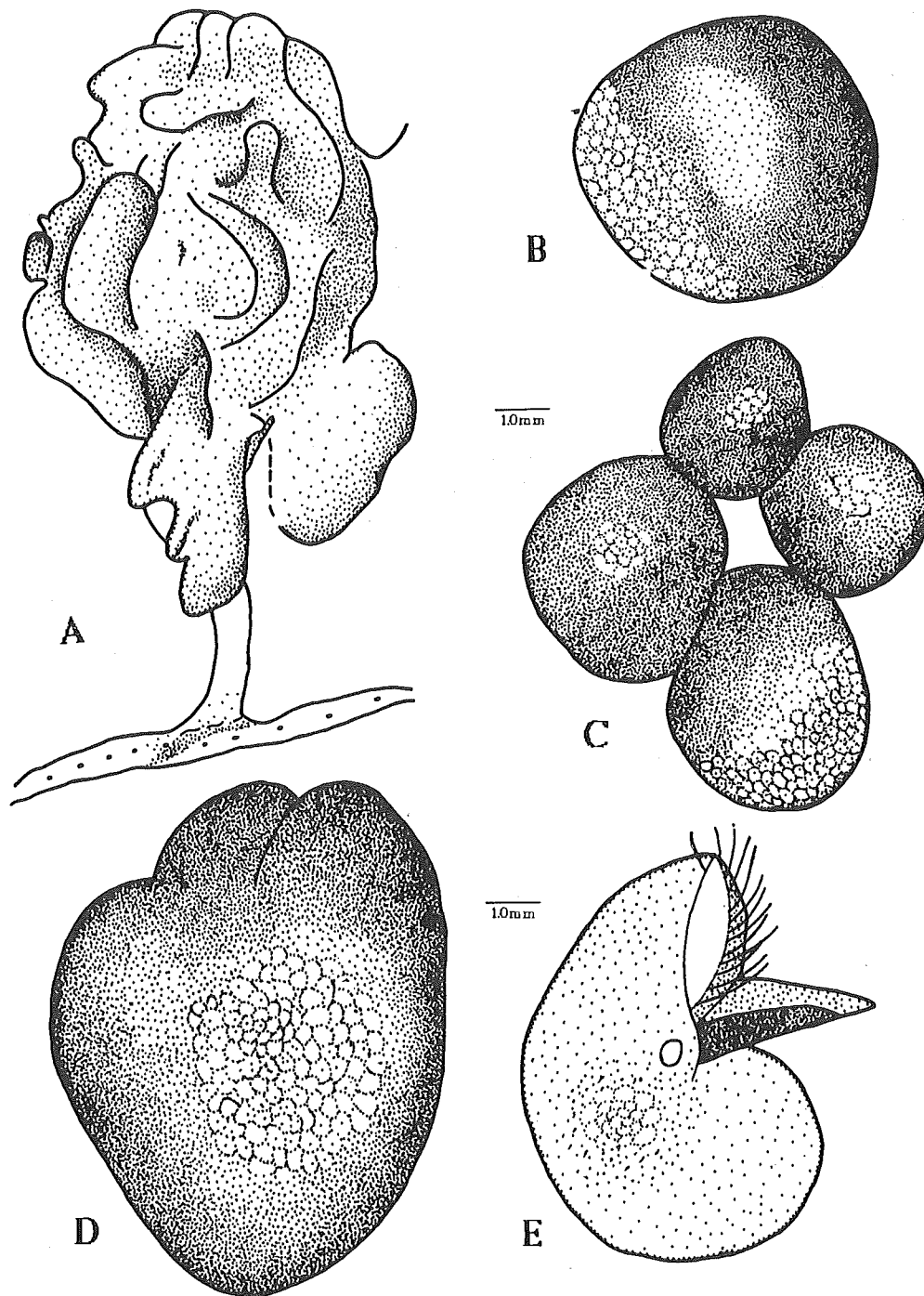


Fig.1. *Hydatina physis*. A. spawn mass; B. single ovum; C. second cleavage stage; D. gastrula stage; E. veliger larval shell just after hatching.

of spawning the spent individual moved to bottom where it stayed most of the time. A marked preference for egg laying (80%) was noted for the glass bowl over the other substrata.

The freshly spawned egg ribbon floats for a while and then adheres to the substratum with its broad disc.

EGG RIBBON AND EGGS:

The snow-white egg ribbon of *H. physis* (Fig.1-A) is broad and intensely folded. As in other hydatinids, the eggs are enclosed in a single layer of hyaline capsules. The milky white eggs (4-6/capsules) are suspended in an albuminous fluid. On an average, a single egg ribbon contains 10100 eggs at the peak of the breeding season. Details of the spawning are summarized in Table I. Characteristics of the egg ribbon, capsules and the eggs are given in Table II.

Table I: Reproductive data for *Hydatina physis* from Pakistan (Data refers to mean value for five cultures, at 26°C).

Mother size (mm)	35 \pm 2.4
Dimension of egg ribbon (mm)	20.2 \pm 0.4 X 10.0 \pm 0.1
Egg capsule size (μ m)	300 \pm 10
Number of capsules per spawn mass	2020 \pm 404
Egg diameter (μ m)	70 \pm 1.2
Number of eggs per capsule	5 \pm 1
Number of eggs per spawn	10100 \pm 200
Incubation period	14 \pm 0.9
Developmental stage at hatching	Veliger
Larval shell height at hatching (μ m)	120 \pm 1.9
Type of sculpture	Non-sculptured
Larval tinge	Pinkish brown

EGG DEVELOPMENT AND HATCHING:

Development to hatching takes 13-14 days at 25-26°C. During this time the embryo passes through a number of stages (Figs.1B-E). The cleavage is spiral, and the first vertical division occurs about 7.5 hours, after oviposition. Gastrulation takes place by invagination about 62 hours, after oviposition and the first sign of rotation occurs on the fifth day. The veliger stage forms after 12 days of incubation. The larvae possess a pinkish brown tinge and a transparent, colourless shell of 1.5 whorls. After fourteen days, the entire ribbon acquires a transparent hue and the larvae escape as free swimming veligers. The whole ribbon evacuated within 0.5 hour. In our study although all the eggs in a hyaline capsule develop into veligers, only 62% successfully

Table II: Comparison between spawning season, egg characters and larvae of *Hydatina physis* from Pakistani, Japanese and Hawaiian waters.

Characters	Pakistani (Present study)	Japanese (Baba and Hamatani)	Hawaiian (Ostergaard)
Spawning period	November-Feb.	May-June	November
Spawn length (mm)	20.2±0.4	25	-
Egg capsule size	300±10	-	-
Egg size (µm)	70±1.2	-	-
Number of eggs per capsule	4-6	7-16	5-6
Incubation period	14±0.9	-	-
Developmental stage at hatching	Veliger	-	-
Larval shell height at hatching (µm)	120±1.9	-	-
Type of sculpture	Non-sculpture	Non-sculpture	-
Larval tinge	Pinkish brown	-	Faint green

hatched out. The empty shells of the other could be seen through the transparency of the ribbon.

DISCUSSION

Reproduction of *Hydatina physis* from Karachi coast of northern Arabian Sea has been investigated. The species inhabits the seaward side of loose rocks and crevices of the sub-tidal zone. *H. physis* is a restricted and eurythermal reproducer, as it does not reproduce round the year and spawning occurs during the season of relatively low temperature covering a long period of four months (October to mid of February). A similar relationship between temperature and spawning has also been established by Amio (1963). The migration and aggregation of adults from their sub-tidal habitats, prior to the onset of breeding season, in the tidal pools of low water mark is an indication of migratory as well as aggregative behavior of breeding. Previous reports of mating behavior for hydatinids are non-existent. No courtship behavior was observed. During the breeding season, adults readily mated when placed with each other. Copulation is reciprocal and simultaneous. No cryptic behavior was observed.

The typical egg laying behavior exhibited by *H. physis* may be true for the other hydatinids as well. As described for *H. vesicaria*, the egg stalks of *H. physis* were not

fixed into the substratum since the eggs are firmly fixed to the substratum by their stalks. Our observations on egg laying behavior support Winner's hypothesis that it is expedient for hydatinids to lay eggs in this position, however more work is needed to confirm this.

It seems that no major difference occur in the capsular morphology. As for other hydatinids reported from other parts of the world (Ostergaard, 1950; Baba and Hamatani, 1958; Winner, 1985) the eggs are enclosed in single layer of hyaline capsules.

A direct relationship exists between fecundity and the breeding season, the highest number of eggs are found at the peak of the breeding season, irrespective of their size (Table I). Whether spawning occurs, once or more than once a year, remains unknown.

A comparison of the reproductive data (Table II) from Hawaiian and Japanese waters, shows that *H. physis* from Pakistani waters produces slightly smaller egg ribbons with fewer eggs per capsule. While no difference was noted in the capsular size, the newly hatched larval shells of Pakistani *H. physis* are slightly bigger than those of *H. physis* from other localities.

Differences are also noted in the colouration of larvae. It might possible, that, the pinkish brown tinge of Pakistani *H. physis* larvae be the result of adult feeding on cirratulid worms, which have orangish brown colouration.

The embryological development in *H. physis*, is typical of related opisthobranchs. Although, no cannibalism or nurse-egg feeding, was observed among sibling veligers, only 66 ± 4 of total hatchings, emerged as free swimming veligers, after 14 days of incubation, at 26°C in laboratory conditions. The situation may vary in the field.

ACKNOWLEDGEMENTS

The authors wish to thank Prof. Dr. Nasima M. Tirmizi, Former Director, Centre of Excellence in Marine Biology, University of Karachi, presently Dean, Faculty of Science, for providing facilities to work at the Centre. We would like to thank Dr. Robert S. Prezante, Department of Biology, Indiana University of Pennsylvania, Indiana USA, and two anonymous reviewers for their helpful comments and suggestions. Thanks are also due to UGC for the financial support extended to the junior author (R.P.).

REFERENCES

- Amio, M. 1963. A comparative embryology of marine gastropods, with ecological considerations. *Journal of the Shimonoseki University of Fisheries* 12: 1-358.
- Baba, K. and I. Hamatani. 1956. Observations on the spawning habits of some of the Japanese Opisthobranchia (II) *Publication of the Seto Marine Biological Laboratory* 5:209-220.
- Ostergaard, J.M. 1950. Spawning and development of some Hawaiian marine gastropods. *Pacific Science* 4: 75-115.

- Rudman, W.B. 1972. The anatomy of the opisthobranch genus *Hydatina* and the functioning of the mantle cavity and alimentary canal. *Zoological Journal of the Linnean Society* 51 :121-139.
- Winner, B.E. 1985. Observations on *Hydatina vesicaria* laying eggs. *Bulletin of Conchologists of America* 12:58-59.