# International Journal of Advanced Research in Biological Sciences ISSN: 2348-8069 www.ijarbs.com Coden: IJARQG(USA)

**Research Article** 

Histological study of reproductive cycle of estuarine clam, Katelysia opima

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#### Abstract

The reproductive cycle of *Katelysia opima* was studied for a 1-year period by monthly observation of gonadal changes. The gonadal maturation is dependable on the environmental factors and availability of food material. Clams become sexually mature at different ages and sexual maturity is a function of size rather than of age. *K. opima* was located at the intertidal zone of estuary. On the account of present investigation, the clam *K. opima* breeds twice in a year.

Keywords: K. opima, Kalbadevi estuary, gonadal development, mollusks, gamatogenesis.

# Introduction

Reproductive phase such as gonad development, spawning and fertilization are coordination with seasonal environmental changes. The timing and duration of reproductive activity may be determined through the interaction between endogenous and exogenous factors. Reproduction in Indian bivalve molluscs has been studied extensively by Sastry, A. N. (1979), much of literature is concern with reports on annual breeding periods. Reproduction is cyclical and it may be annual, biannual or continuous.

Spawning period of mollusca depends on the species, in some biannual period of spawning, while in other species annual patterns was observed by some workers. Hornell (1922) and Abraham (1953) observed the spawning in *Meretrix casta* was twice in year. Nagabhushanam and Mane (1975), observed that, *Katelysia opima* spawns twice in a year. In *Ostrea madrasensis*, Rao (1951), observed continuous breeding under marine environment. Mikhailov *et. al.* (1995), has studied sexual differentiation of reproductive tissue in bivalve molluscs. Kim, *et. al.* (1998) studied annul and biannual changes in gonadal development of bivalve molluscs. Stead *et.al.* (2002), showed that, gonadal index cycle was similar for two species of bivalves. Al–Mohanna *et.al.* (2003), investigated morphology of the cell during oogenesis in *Amiantis umbonella*. The available food was directly related to the rate of the gonadal development and with the total quantity of gonad generated. Equally, food restrictions limited gonadal recovery after spawning episodes (Marina Delgado *et. al.*, 2005).

In view of the fact that environmental factors those seasonally fluctuated affect the gonadal development in bivalves, our principal objective was to histologically study sexual maturation in *Katelysia opima*.

## **Materials and Methods**

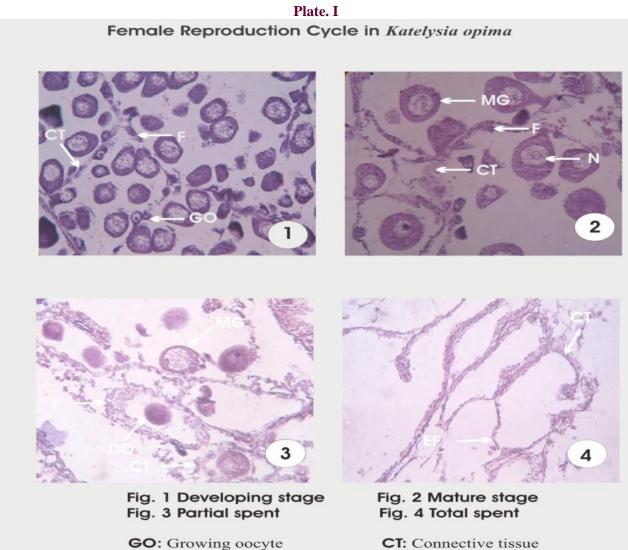
On every fort night of study period clams were collected during low tide of every new moon and full moon days from Kalbadevi estuary, Ratnagiri, Maharashtra, India. The clams were then brought to the laboratory and the shells were cleaned. These clams were sacrificed and gonads were dissected out and fixed in Bouins preservative, prepared in sea water. After 48 hours of preservation, the tissues were further processed for the block preparation in paraffin wax. These paraffin blocks were sectioned at 5-6 µm thickness on microtome and stained with haematoxylene and eosin stain. All the observations for microphotography were made under the Olympus CH 20i (U.S.A.) microscope.

# **Results**

### a) Female reproductive cycle (Plate. I)

During January, the clams showed active gametogenesis in gonads. In mid of January, small oocytes appeared to develop along the base of follicle wall representing the developing stage (Fig-1). In February, the developmental stage showed half grown oocytes attached to the follicle walls by slender peduncles representing the late growing stage. Eggs

have vitelline membrane and nucleus present in the centre. A few primary germ cells were also observed proliferating from the wall. In March, gonads with many mature ova free in the lumen of the follicles having a large nucleus at the centre of each egg representing mature stage (Fig-2). In April, some clams were found the spawning activity, which was reached its peak (Fig-3). In May the unspawned ova underwent cytolysis representing the recovery stage in gametogenesis (Fig.-4). In August, clams attaining maturation phase and in September almost all clams represent the mature stage. In October, the mature clams showed the evidences of spawning. In November, clams having a few follicles were also observed with unspawned ova. In December, these ova in the follicles underwent cytolysis, representing the recovery stage.



F: Follicle MG: Mature gamate N: Nucleus

**CT**: Connective tissue

**EF:** Empty follicle **DO:** Degenerating oocytes

#### b) Male reproductive cycle (Plate- II)

The male reproductive cycle also follows the same stages of development as that of female clams. The gametogenesis was started in January. The primary and secondary spermatogonia were developed along the base of the follicle wall representing the developing stage. In February, the growing stage was started, in this stage the follicles showed centripetal arrangement of the bands of spermatozoa, spermatocytes and spermatids with few spermatogonia (Fig-1). In March, the follicles enlarged with their lumen packed with spermatozoa forming plums at many places, while few follicles showed partially spent condition, this represent the mature and spawned stage. Regular spawning was observed in the next month and by the April almost all clams showed spent condition i.e. spent stage (Fig-2). In May, some spermatozoa was unspawned and they were retained in

the follicles underwent cytolysis and the follicles have been recovered representing the developing stage to give rise again to germ cells protruding from the walls. In June, the gonads showed early development stage, showing spermatogonia and stage of maturity (Fig.-3). In July, the clams showed advance stage of maturity. In August the clams those remained immature showed developing stages representing well defined growing and mature stages (Fig-3). In September, the clams reached in mature stage. In October, male gonads showed partially spawned out condition representing spawned stage, the rows of sperm were separated and more random in their arrangement and majority of follicles showed few spermatozoa and spermatids. In November, the follicles were empty and their shape was changed but, some retained their shape with residual spermatozoa and spermatids. In December, both recovery and developing stages were appeared; this was the stage of developing gonad (Fig.-4).

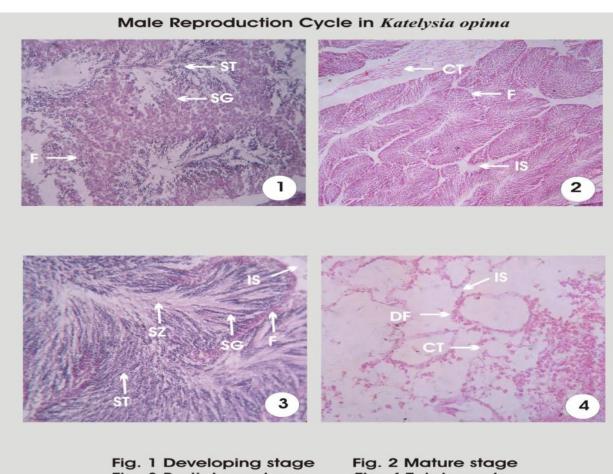


Plate II

# Fig. 3 Partial spent

**SZ:** Spermatozoa SC: Spermatocytes F: Follicle **IS:** Interstitial spaces

# Fig. 4 Total spent

SG: Spermatogonia ST: Spermatid **DF**: Degenerating follicle A large number of marine organisms found in the range of intertidal zone are subjected to wide variations in salinity, temperature, action of waves, and availability of oxygen as well as food in the environment. Rate of growth depends largely on geographic location. Clams become sexually mature at different ages and sexual maturity is a function of size rather than of age.

*K. opima* was located at the intertidal zone of estuary. On the account of present investigation, the clam *K. opima* breeds twice in a year. The first spawning period was observed in the month of April to June and second one took place in October to December.

In Arca granosa showed continuous breeding with peak periods in April to May, September to October and from December to January and the period from June to September was resting period in this clam (Maske et. al., 2015). Fluctuation in environmental factors affects the spawning period. In summer, salinity and temperature was high, which triggers the spawning. In monsoon, due to the flood and rain water, the salinity decreases in majority of period. Due to influx of rain water the turbidity has increases, and valves of bivalves remained closed, so that metabolic rate slows down (Mane, 1974). At the end of September, majority of clams undergo maturation phase. In October, salinity and temperature rises which triggers the spawning process in clams, and they receives favourable environment to spawn. In the month of October, the salinity increased considerable. The rapid increase in metabolism make gonad to undergo active gametogenesis and it reaches full maturation stage by the end of March. Spawning takes place in March and April. The K. opima is being primarily marine, when the temperature and salinity are on the optimum level the clams spawns (Nagabhushanam et. al., 1975). Spawning was maximum in month of March to June, might be due to longer day length, gametogenesis in Argopecten irradians from Massachuset was correlated with day length (Sastry, 1970).

There are many evidences that a complex of physical variables in the environment influences the sequence and timing of events in reproductive cycle of marine invertebrates. The factors inducing spawning may be quite different from those inducing annual reproductive cycle (Giese, 1959). The gametogenesis is controlled by many exogenous and endogenous factors among which temperature has received much attention (Mackie, 1984). The temperature may not

influence the spawning of marine bivalves of tropical waters, but salinity has been considered as important factor in the initiation of spawning in bivalves of tropical area (India). Lowest temperature in winter, gametogenesis was slowed down but, not entirely arrested. From Indian waters, both continuous and discontinuous spawning seasons in bivalve molluscs were reported (Abraham, 1953 and, Rao, 1967). Along Indian coasts, the water temperature does not fall below the optimum requirements of many molluscs as in temperate waters and the temperature is comparatively high throughout the year except for few degree decrease in winter.

The fluctuation in temperature may trigger the spawning of clams. In Katelysia opima two spawning periods were observed throughout the study period. Meretrix casta on the east coast spawns twice in a year during April-May and again in September (Hornell, 1922), whereas Meretrix casta of Adiyar estuary and Mandappam fish farm spawns several times in a year (Rao, 1951 and Durve, 1964). Spawning period in Meretrix casta does not remain constant from year to vear in the same environmental condition neither it coincides in any two different environments, and found that low salinity in Adiyar estuary was the cause of spawning in the clam (Rao, 1951). Gametogenic cycle may occur in population of a species on an annual, semiannual or continuous basis. The timing and duration of the gametogenic cycle and the number of cycles within a year may be characteristic of specific populations. Gametogenesis in some species appears to be restricted to certain months in the year, while in others it occurs continuously throughout year.

Due to unfavorable conditions, gametogenesis and maturity in clams takes place at slow rate through monsoon. Optimum salinity requirements of 22.26 %0 for the development of eggs of oysters under laboratory conditions, further confirmed by field observations (Rao, 1951). Some aspects of ecophysiology of estuarine clams have been worked out from the west coast. The studies reveal that, the low salinity in monsoon affects the activity of the clams and oysters (Mane, 1974 and 1978). Field observations concluded that, the optimum level of salinity requirements for spawning Crassostrea graphoides in Kelwa backwaters on Bombay coast may be between 28.58 and 13.15 %0 and as spawning advances in monsoon, the oyster become more responsive to stimulation and spawn even in low salinities (Durve, 1964).

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Other factors such as mechanical stimulation (wave shock, pressure changes, and tidal cycle) are not yet confined to induce spawning in some lamellibranches from Indian coast. Possible effect of pressure changes accompanied by hauling on the spawning of K. opima, beside the influence of salinity and temperature in Kalbadevi estuary (Mane, 1973). Temperature acts as a triggering stimulus for initiation of the oocytes growth phase Scallops maintained at temperature exceeding certain maximum  $(25^{\circ} \text{ and } 30^{\circ})$  also fail to initiate gonad growth and gametogenesis (Sastry, 1970 and 1966). The reproductive cycle of M. mactroides followed an annual cyclicality, which was significantly correlated to monthly mean sea surface temperatures (Marko Herrmann et. al., 2009).

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