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CENTRAL MARINE FISHERIES RESEARCH INSTITUTE (Indian Council of Agricultural Research) P. B. No. 2704, E. R. G. Road, Cochin-682 031, India

31. REPRODUCTION IN EDIBLE BIVALVE SHELLFISHES OF RATNAGIRI COAST

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ABSTRACT

Clams, oysters and mussels are regularly fished and marketed at different places in Ratnagiri district throughout year, rarely in peak of monsoon. The annual reproductive cycles of three species of edible clams *Paphia laterisulca, Katelysia opima* and *Meretrix meretrix,* the oysters *Saccostrea cucullata* and C. *grypholdes* and the green mussel *Perna virldis* of the Ratnagiri coast have been studied. Maximum gonad index occurs during June-August and November-February in *P. laterisulca,* June-September and January-February In *K. opima* and February-August In *M. meretrix.* The histological studies revealed spawning m *P. laterisulca* during mid September-Match with two peaks • once in October-November and another In February-March. Spawning in *K. opima* occurs during October-November and March-April. *M. meretrix* spawns only during October-November. *S. cucullata* spawns during October-January, and C, *grypholdes* during October-November. *P. virldis* spawns during late July-early September. The difference In such spawning periods of these shell-fishes have be^on attributed to the change^{*} In salinities in the respective habitats with the secondary cffecta of temperature on the gonads- The results are discussed in the light of species adjustment to the fluctuating environment and biochemical-phyliology,

INTRODUCTION

Research work on the biology and fishery of edible bivalve shellfishes contributing to the substantial catches was started comparatively recently in India. There have been considerable J X .. J • . ^_ preliminary and some detailed investigations on various species of local importance, and the results obtained are of some help to make effective attempts in management and conseri»!. 4 ui е. vation measures. Some of the notable contributions from various parts of the Indian coast are on clams (Rao 1952; Abraham 1953; Durve 1964a; Alagaraswami 1966; Rao, 1967), on mussels (Paul 1941; Jones 1951; Rao et al 1975), and rock oysters (Paul 1942; Venkataraman and Chari 1951; Rao 1951,1 956, Rao and Navar 1956; Durve and Ball 961; Durve 1964b, 1965) and on cockles (Narasimham 1969).

Along the coast of Maharashtra State with its vast coastal areas and creeks, muddy bays, rocky inshore regions, estuaries and backwaters, edible shellfishes are common and offer an attractive field for fishery enterprise. Clams, oysters and mussels form regular fishery of considerable local importance along the coast. In Ratnagiri, along the coast. the following species of bivalve shellfishes are fished throughout different seasons.

the clams	— Paphia laterisulca, Katalysia
	opima and Meratrix meretrix'.
the oysters	— Saccostrea cucullata and C.
	grypholdes:
~~~ ~~~~~	_ p^^^^ ^.^j^]^.
, <b></b> ,,	
the cockles	— Anadara granosa.

The Kalbadevi, Sakhartar, Sh.rgaon and <u>Bhatiaestuar.es</u> and creeks, and Ratnagir. bay on the outskirts of Ratnagiri are excellent shell-

Considering the abundant distribution and local market in Ratnagiri we have undertaken to study several aspects of eco physiology and neuroendocrinology of these shellfishes since ^g^Q (pjagabhushnam et al., > 1972; Mantale ^^ g, 1972; Nagabhushanam and Mane 1973, .,975 1973; Mane 1974 a, b, 1975 a, b, 1978; Mane and Nagabhushanam 1975, 1976, 1979, 1983; Mane and Bidarkar 1976; Talikhedkar et al 1976; Mane and Dhamne 1980), Here we report comparative data on the reproductive biology of these edible shellfishes from Ratnagiri coast,

#### MATERIAL AND METHODS

*P. laterisulca* (35-45 mm), *K. opima* (32-37 mm) and *M. meretrix* (45-50 mm) from Kalbadevi

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estuary, C-cucw//«fa (35-40 mm) from Shirgaon creek, C. *gryptioides* (80-90 mm) and *P. viridis* (70-80 mm) from Bhatia creek were collected monthly at regular intervals. The gonads of the animals were fixed in Bouin's fluid and histological sections taken as described elsewhere (Mane 1973; Bidarkar 1974). The gonads of the clams, *P. laterisulca, K. op/ma and M meret-nx* were studied by the determination of gonad indices by volume displacement method or on wet weight basis (Giese 1969). During the collection of shellfishes from the respective beds salinity and temperature data were collected.

#### RESULTS

The shellfish in spawning condition are plump and in prime condition for use. After spawning the gonad is much reduced in size and once again occupies much of the visceral mass. Thus it is natural that as the size of the gonad increases with the ripening of the sex products, the gonad is expected to vary in size and weight. Therefore, the gonad indices have shown that the maximum increase in the index occurs from June to August and again from November to February in *P. laterisulca,* from June to September and January to February in *K. opima,* and from February to August in *M.* 

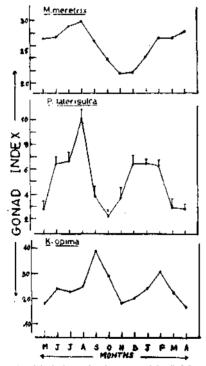


Fig. 1. Variations In the gonad indicM of the dams from Kaibadevi estuary, Ratnagiri.

*meretix.* In the other periods the index for each species remains low (Fig. 1).

. , u ^ The histological preparations of the gonad tissues of each species show following conditions:

۸ Spawning in this clam starts ^^^ representing partial shedding of ^^^ ^ November spawning is more ^^ ,^^ complete as majority of the clams have 3.^{^^} gametes showing spawned-out condition m m gm unspawned gametes left in the follicles after spawning undergo cytolysis ^"'^ ^^^ follicles start budding young developing sex cells in December. From the beginning of January till February few clams show active gametogenetic activity in gonads. A part of population reaches mature condition in March and begin to spawn till April. Those spawned show cytolysis of unspawned gametes. From June to September the gonads show development of the sex cells in follicles. Later almost all clams become sexually mature and ready to spawn in October,

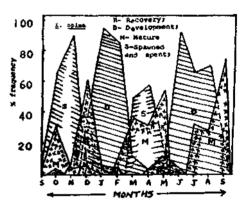


Fig. 2. Ssasonal gonadal changes In K, opima.

*P. laterisulca* (Fig. 3) This clam has a prolonged spawning season extending from mid September to the end of March with two peaks - one in October-November and the other in February-March. Active gametogenesis takes place in May and June, and by the end of June the whole population reaches peak of maturity which is maintained till mid September. The spawned out individuals from the first peak immediately enter into a phase of redevelopment in December and attain maturity within 4 to 6 weeks i. e. upto January. A few clams Continue to spawn in December and January but at low intensity

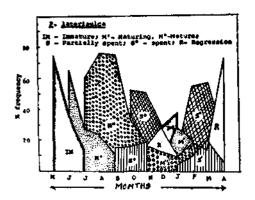


Fig. 3. Seasonal gonadal changes in P. laterisulca.

and immediately after spawning they enter into a phase of rematuration. After second peak of spawning majority of the clams pass into recovery condition in April before entering in gametogenetic activity.

*M. meretrix* (Fig. 4) Immediately after the close of monsoon i. e. in September the clams begin to spawn and majority of the clams are spawned-out in October and November. After the spawning, the gonads undergo recovery representing the cytolysis of unspawned gametes left in the follicles. Gametogenetic activity in the

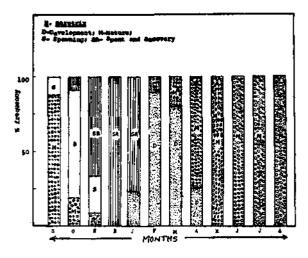


Fig. 4. Seasonal gonadal changes in M. meretrix.

gonad tissue begins in January and continues till February. Further development leads to the mature condition of the sex products in April. The clams maintain this condition till August end, i. e. throughout the monsoon and begin to spawn in September.

C. cucullata (Fig. 5) After completion of spawning in January, both the sexes pass in The follicles begin to to recovering stage. shrink in size and get replaced by the connective tissue in February. The phagocytic cells appear in large number within the follicles. The follicles disappear rapidly and the gonad tissue enter neutral condition (difficult to differentiate the sexes) in March. When the reorganization of the gonad tissue begins, a few 'oysters in April show reappearence of the follicles with stem cells which are going to develop the young sex cells. The gametogenetic activity continues in May and June and in July both the sexes become sexually mature showing follicels fully

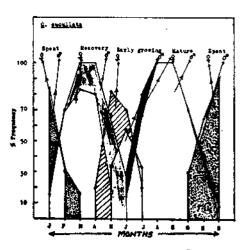


Fig. 5. Seasonal gonadal changes in C. cucullata.

enlarged and packed with reproductive elements. The interfollicular vesicular tissue as well as the connective tissue between the gonadial layer and the gut are reduced to the bearest minimum. In some of the oysters maturing condition still persists showing continuous budding of the sex cells. In August and September both the sexes remain in mature condition and almost entire population becomes sexually mature. Spawning begins in October, a few oysters showing partially spawned-out condition of the From November onwards till gonad tissue. January many oysters spawn showing spawnedout condition. Immediately, after spawning, the sign of disintegration of the reproductive elements appears in the population of the oysters in February. This ultimately leads to the neutral condition as described earlier.

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C. fifAyp/»o/£/es (Fig. 6) Most of the oysters close to the follicle walls and the walls of the become sexually mature in June but still in a few oysters the gametogenetic activity progresses. Gradual development leads to the extensive proliferation of the follicles accompanied by rapid growth of the sex products in large numbers. By July all the oysters reach a fully mature condition which is maintained by rapid growth of the sex products in large numbers.

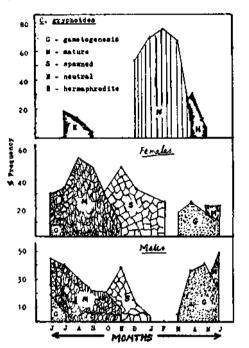


Fig. 7. Seasonal gonadal changes in C. gryphoides.

till September. During mid-monsoon (first fortnight of July) a few hermaphrodite indivi-The germinal epithelium produals appear. liferates fresh oogonia in all the follicles and lumina contain almost ripe sax products. Spermatogonia and spermatocytes are not seen. In a few oysters, the follicles contain maturing oocytes and traces of mature sperms in the lumen of the follicles. Thus, hermaphroditism inthis oyster is regarded as the transitional phase m the change of sex from male to female. immediately, at the close of monsoon (in the first fortnight of September) a few ovsters begin to spawn. More than half of the population spawns in October. By November oysters spawn more or less completely. From the first fortnight of December the follicles and the gonad ductules start shrinking considerably. The vesicular tissue cells surrounding the follicles become large, flattened and bladder like with hyaline cytoplasm but some of them

ductules are much condensed in size. The cells in amoeboid form with granular cytoplasm take up phagocytic function. These cells infliter through the walls of the follicles and accumulate in large number within the lumina. With subsequent development in the gonad the oysters pass to neutral condition (difficult to differentiate sexes) in February. Indistinguished sexes at the early gametogenetic activity appear in the first fortnight. During the second fortnight sex differentiation is marked by the slight expansion of the gonad tissue and appearance of oogonia and spermatogonia in females and males, respectively. With the development of the sex products, the gonads of the oysters become fully mature by May. Few females show unspawned eggs in February. The relict female gametes instead of undergoing resorption still remain in the lumina of the follicles and the follicle walls start proliferating fresh oogonia i.e. the sex products of the opposite sex which in turn, by series of divisions, give rise to spermatocytes and spermatids. In April these hermaphrodites occur in the samples and show gradual degeneration of unspawned sex products in follicles. It can be said that as spawning in female oysters is either late or prolonged (in partial State) and the vigourous differentiation of the male phase (some what premature) results in the occurrence of bisexual condition. In this hermaphroditism the change of sex is from female to male,

 $p ^{j,,-/g}$  (pjg 7) j^e disintegration of the unspawned gametes of the previous spawning ^tgrts in March showing recovery in gonad, *i*^^^ characteristic of sex differentiation is lost 3,[^] the follicles begins to shrink in size. They 3,3 simultaneously replaced by the developing r.u • • connective t.ssue. Phagocytes appear m large """"er w.thm the follicles. Thus the follicles fapidly disappear and the mussels enter in neutral stage. Most of the mussels are in neutral stage in April. In May very clear hermaphrodites are seen. The histological sections show well developed ova and sperms. In the sections of one of the mussels the gonadal follicles close to the periphery have shown large ripe ova, oogonia and developing ova of small

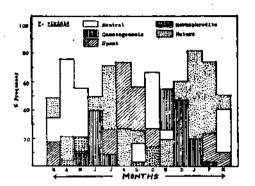


Fig. 7. Seasonal gonadal changes in P. viridis.

size in connection with the follicle layer. In the deeper regions the ova gradually diminish in size. The germinal cells actively proliferated fresh oogonia in all the follicles. The lumen was almost packed with well developed spermatozoa but there were no spermatocytes close to the follicle wall. In the sections of other few mussels, the formation of ova had taken place to a greater extent than in the proceeding one. Well developed ova were seen not only alog the peripheral region but also deeper down the follicles. The sperms were either altogether absent as in the follicles of the top layers or very sparse deeper down the follicles. The active proliferation of the female sex cells and the entire absence of the fresh spermatocytes in these mussels show that the change of sex is from male to female. The rare occurrence of the individuals with the reproductive elements of both the sexes is an indication that hermaphroditism is not a regular feature. As the germ cells stop proliferating components of one sex and give rise to those of other sex, hermaphroditism is said to be a purely transitional phase. Gametogenetic activity in males commences by about end of May and in females by beginning of June. lt reaches its peak in both the sexes towards the end of June resulting rn full ripe gonad follicles. In the beginning of July the gonads of both the sexes become fully enlarged and packed with reproductive elements. The interfollicular vesicular tissue as well as the connective tissue betweenthe gonadial layers get considerably By third week of July spawning reduced. commences. A few partially spent oysters show

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shrinking of gonad follicles and the vesicular tissue surrounding them enlarges Within the follicles small amount of unspawned gametes are retained. Samples collected subsequently during August and early September showed increasing number of fully spawned-out mussels. In late September and October for the second time the disintegration of unspawned gametes takes place, vary similar to that had taken place in early summer, resulting in occurrence of large number of mussels in neutral stage. Few mussels in hermaphroditic condition are observed in late October. The presence of large unspawned eggs in the lumen of the Collides and numerous spermatogonia along */® follicular wall suggest a change of se;^ from female to male. Reorganization of gonad tissue starts by the beginning of November with very fapid proliferation of oogonia in some and spermatogonia in others. As a result follicles are found to spread out and the vesicular tissue reduces leading to mature condition of the sexes The number of fully ripe males and females increase in January. In February a few mussels show the beginning of spawning showing the occurrence of partially spawnedo"* males and females. This spawning can be said to be a minor one since only a few mussels >n the population release gametes partially In March disintegration of the reproductive products begins in the population of mussels leading ultimately to the condition described earlier in March.

#### USSION

From Indian waters both continuous and discontinuous spawning seasons in bivalves have been reported and regarding the nature of their reproductive cycles even the same species from one locality is known to differ from those of different locality in their reproductive seasons. For example, *Meretrix casta on* the east coast spawns twice in a year during April-May and again in September (Hornell 1922). whereas the same species spawns several times in a year in Adyar estuary (Abraham 1953) and Mandapam Camp (Durve 1964a). On the other hand, another species *M. meretrix* in Kalbadevi estuary, Ratnagiri,

along west coast begins to spawn in September and continues till November (according to the present study). Donax cuneatus on Madras coast and those in the Palk bay has only single reproductive cycle, although in the former locality the breeding is relatively much longer (Nayar1955). In contrast to this, Rao (1967) in the same wedge clam reported prolonged spawning from January to June on Madras coast. Alagaraswami (1966) in Donax faba also reported prolonged spawning on Mandapam Camp coast. Donax cuneatus in Mirya Bay (Talikhedkar 1975) shows close similarity in the reproductive cycle with the same species in Palk Bay (Nayar 1955) and on Madras coast (Rao 1967), although the clams from later locality are known to breed for a much longer period. In Katelysfa op/ma only one spawning season occurs in Adyar estuary, Madras (Rao 1951) but the same species in Kalbadevi estuary, Ratnagiri spawns twice in a year.

One of the most important questions in the problem of reproductive cycles is, what mechanism regulate gametogenesis and spawning in Indian marine bivalve molluscs? The studies on the environmental factors, like salinity and

temperature in the reproductive study areas by the above workers and' many others have shown that under tropical' conditions of Indian

coast, the water temperature does not fall as m waters and the temperate temperatures remain comparatively high throughout the year except for few degrees drop in winter. The complex of physical variables in the environmem influence and the timing of events in the reproductive cycles of marine invertebrates .s well documented (G.ese 1959). In terriperate regions gonad development and reproduction in marine bivalves have been correlated with the wide range of temperature fluctuations (Posgay 1930; Loosanoff and Nomejko 1951; Loosanoff and Davis 1952; Dickie 1953; Wi son and Hodgkin 1967). On the other hand, in tropical regions the reproductive cycles of many marine bivalves have been correlated with "fluctuations, in salinity (Paul 1942; Rao 1951; 'Durve '1'965; Alagaraswami 1966).

The low salinity in monsoon in Kalbadevi estuary affects the activities of the clams. The 172

filtration rate and oxygen consumption decrease because for most of the period tliey remain with shell valves closed, though they could tolerate considerable low salinity in monsoon than summer (Mane 1974a, 1975a, b; Dhamne and Mane 1976; Mane and Dhamne 1980). Due to unfavourable conditions gametogenesis and maturation take place at slow rate, and through monsoon the gonads of the clams reach plump condition. As the salinity of the estuary increases with the temperature after the close of monsoon, the plump clams receive favourable environment and spawning begins. Spawning continues till almost all the gametes are released, With further increase in temperature and salinity followed by rematuration process in the later season the gonads of clams reach plump condition in early summer - the period when the clams are subjected to optimum temperature when spawning begins. It is of course very difficult to determine all the environmental factors con-^/o^////Q he reproduction in bivalves in natural conditions, but it is the salinity change playing an important role in determining the physiolog'^a' activity of the clams with other factors counteracting on them.

, Regarding ovisters from Indian coast, the first record by 'Hornell '(1910) showed that spawning in Madras back water oyster coincides ^ ^ ^ north-east monsoon which begins in October. Later in 1921, he suggested that as the rainy season differs on the two coasts of India, there is a corresponding divergence in the spawning maxima (Hornell 1922, R30 (1951, 1955) found the optimum ^// requirements of 22.26°Uor the develop-^.ent of eggs under laboratory conditions and f.^her confirmed this by field observations. He mm suggested that the spawning in C. madra-• j ". * • b, 1 1 »u-sens/s does not take place unless the optimum Salinity IS reached by the influx of rainwater, by the evaporation of sea watër or by the opën-x.u. in 1951 ingofthebar in Ennore backwater. In 1951, he indicated that besides the attainment of the salinity to the optimum level, there may be other unknown environmental'factors or the presence of some suitable 'chemical influencing spatfall in case of *C. madrasensis.* Durve (1965) observed that spawning in C. gryphoides in CMFRI

Kelwa backwaters, Bombay, did not take place at 28.58°/oo but following heavy rain the salinity decreased to 13.15 7,,, and the oysters began to spawn in July. He considered that the optimum level of salinity reuqirements for spawning in C. gryphoides might be between 28.58 and 13.15°/, o and as the spawning season advances (in monsoon) the oysters become more responsive to stimulation and spawn even in lower salinities than in the beginning of the spawning but the reason for this, he was unable to ascertain. His observations have further shown that spawning in females was more related to the lowering of the salinity than in males, as the females spawned earlier than males, which spawned till November when the salinity increased appreciably. According to our present findings, C. cucullata in Shirgaon creek spawns with the rise in salinity (after rainfall) and moderate temperature. The spawning reaches its peak in November and December. In February and March with the rise in temperature and salinity, the spawned out gonads recover at a faster rate and gametogenesis proceeds actively to reach mature condition by July. This condition is retained till the close of monsoon. The present findings on C. gryphoides in Bhatia creek shows difference in the spawning periodicity from that of the same species in Kelwa backwater on Bombay coast. This may be due to the physiologically different races of the oysters along the west coast, as suggested by Loosanoff (1933) for C. virginica from Long Islands Sound and Texas. During August the salinity of the Bhatia estuary lowers as much as 18°/,,,, at which time C. gryphoides do not show any spawning activity. Further in September as the salinity raises to 23.5 7[^], spawning commences and continues till November with the rise in salinity to 28.7 700 at the time majority of the oysters spawn. Those oysters, in particular females, which spawned in late October do have December when not spawn any more in the salinity increases to 31 700 and the temperature of the water increases. Thus it can be seen that spawning in these oysters takes place only when the salinity ranges between 23.5 to 31 7,,,... Regarding the temperature of the creek water, the fluctuation is ony between 22" to 33° C the lowest being in January and highest in May. During the spawning period (from September to November) the temperature fluctuates from 27° to 28.5°C and during the course of prespawning, at the time when all the oysters are fully mature, the temperature ranges from 29°C in June and 26.5°C in August. This indicated that temperature might not have any influence on maturation and spawning in these oysters. These observations are in agreement with the earlier findings of Rao ^'951; on *C.madrasensis* of Madras and of Durve ^1965^ on *C. gryphoides* from Bombay coast.

Thus once a shellfish population has reached maturity, spawning may be triggered through interaction between the organism and external factors that induce gamete release, if all mem. bers of the populations are mature and react simultaneously to the factors inducing spawning, gametes may be released synchronously and a short breeding period results (in oysters, Rao 1956; Durve 1965; in mussels, Parulekar 1982; from present study). Therefore, synchrony in spawning appears to depend on a critical state of maturity within an individual and among members of the population, as well as their representatives to the exogenous factors inducing gamete release. Once animals are mature a variety of factors seem to induce spawning.

Studies on *Perria viridis* from Goa coast by Rao (1975) have shown that in the green mussel breeding takes place throughout the year with maximum larval abundance in October and November and with lesser abundance again in March. Our study on green mussel from Bhatia creek indicates spawning from July to early September. The mussels on Goa coast also spawn during this time. Since many mussels in Bhatia spawn during this period they pass in to neutral stage in the subsequent period. Those mussels reaching fully mature condition in the gonads in January respond to high salinity and temperature, and spawn from February to beginning of March. Many mussels fail to spawn perhaps due to considerable increase in salinity and temperature unfavourable to them to spawn. On the Goa coast the temperature of the sea water is somewhat lower in summer than at

Ratnagiri though the salinities in both the areas remain high. The study by Rao (1975) on Goa coast is based on the older and younger mussels of the population separately. They have found restricted breeding in both; in older ones taking place from July to December and in smaller ones from January to April. Thus the population as a whole, the mussels on Goa coast are said to have breeding almost round the year with maturation process continuing almost simultaneously. In our study only the adult mussels belonging to 70 to 80 mm in shell length have been studied. It would be interesting to study breeding in the older and smaller populations of mussels of Ratnagiri coast separately.

However, the physiological variations in populations of a species exposed to different environments could either be a phenotypic response of a single genotype or could be truely genetic (Prosser 1955). Comparision of physiological responses of geographically separated populations of marine animals have shown variations in the adaptive capacity but in many cases the difference has been phenotypic. Hence variations in the reproductive activity, if any, may therefore be adaptive for continuity of populations in geographically separated areas. Spawning is a protracted process in a number of species of bivalve shellfishes. Several peaks within breeding periods lead to an extended breeding seasons. When different age groups in a population spawn at different times, an extended breeding season is observed. More data on the influence of various exogenous and endogenous factors controlling gametogenesis, growth and maturation of gonads and spawning are needed to understand the reproductive cycle.

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