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COCHIN, INDIA ★ 12 - 18, January 1980

PART 2

MOLLUSCAN CULTURE

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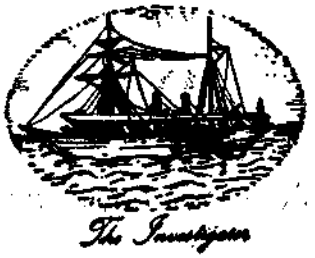

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PROCEEDINGS OF THE

SYMPOSIUM ON COASTAL AQUACULTURE

PART 2 : MOLLUSCAN CULTURE

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Proc. Symp. Coastal Aquaculture, 1983, 2: 427-435

OYSTER CULTURE AT TUTICORIN

K. NAGAPPAN NAYAR* AND S. MAHADEVAN
Central Marine Fisheries Research Institute, Cochin-682 018

ABSTRACT

Oyster culture as a food producing subsystem, can be undertaken effectively and profitably in India. The success of this centure can have far reaching results in solving the demand for cheap animal protein from the sea and as an income generating avocation for fishermen.

Investigations carried out at Tuticorin to find out the feasibility of oyster farming in coastal areas were successful. Effective methods have been developed for large-scale oyster spat collection by using lime-coated country tiles. For growing the oysters to marketable size in short period 'rack culture' technique was effectively employed. Production of 119 tonnes of oysters per hectare could be achieved in 12 months time. Applicability of the technology developed in popularising oyster farming along the Indian coasts is discussed in detail.

INTRODUCTION

RICKER (1969) has stated that by the year 2000 A.D. the total marine capture fisheries yield will be of the order of 150-160 million metric tons which will supply only about 30% of the expected world human protein needs. Fish contribute to a diet from as little as 2.74 gms/day/caput (India) to as much as 76.17 gms/day/caput (Japan) according to Kaul (1978). Hanson (1974) is of the view that agriculture and fisheries are very likely nearing the limits of their practical production capacities. This combined with development in mariculture could make mariculture — a food producing subsystem — an economically important step for augmenting resources. Evaluation of the bioeconomic suitability of marine fishes and shellfishes based on trophic efficiency, reproduction in nature and in captivity, growth of biomass production and life cycle clearly shows that molluscs present fewer problems of culture. In addition to this, another factor in favour of undertaking culture of molluscs is that of all groups advanced technology has been established only for the molluscs, because considerable efforts have been expended on them for several years. Of these, culture of oysters and mussels is labour-intensive and the selection of one or the other or both for culture depends on regional and specific locational factors, micro-oceanographic features and other physical environmental facets like tidal range, state of sea etc. Regional factors are the most pragmatic ones of labour resources, market proximity, public receptivity and other socio-economic aspects.

As far as India is concerned it may be seen from the annual marine fish landing figures for the past several years that molluscan landings form an insignificant percentage (0.8%) of the total. Hornell (1909 a, b, 1910, 1914, 1917, 1922, 1949 a, b, c), Rai (1928), Rao (1941), Rao (1958, 1963, 1966), Jones (1950, 1968 a, b). Jones and Alagarswami (1973), Alagarswami and Narasimham (1973), Mahadevan and Nagappan Nayar (1973), Nagappan Nayar and Mahadevan (1973, 1974), Rao (1974) only

* Present address: Tuticorin Research Centre, Central Marine Fisheries Research Institute, Tuticorin 628 001.

to mention a selected few, have drawn our attention to the immensely rich molluscan resources, with a good variety of commercially important and economically valued species of protein food value, occurring in the coastal waters of India. Their utilisation, especially of the edible varieties, has been static and inadequately attended to till now. While capture fisheries of molluscs is no doubt under exploited a total absence of culture attempts exists in this realm. To fill this deficiency, programmes were formulated to start culture of edible molluscs by developing suitable techniques of collection, growing and farm management. Of this edible oyster culture was chosen as one of the priority areas.

For the success achieved in the development of oyster culture techniques at Tuticorin, the authors owe their debt of gratitude to Dr. E. G. Silas, Director who has been supporting our efforts by his guidance, encouragement and help. The devotion of all other members of the oyster culture team at Tuticorin is also gratefully acknowledged.

Status of Edible Oyster Culture and Resources Availability

Of the 11 species of *Crassostrea* reported from Indian waters (Awati and Rai, 1931), *C. cornucopia*, *C. glomerata*, *C. belcheri*, *C. quercina*, *C. crenulifera*, *C. bicolor* and *C. lacerata* appear to be economically unimportant. *C. cristagalli* and *C. folium* mentioned by Rao (1974) also appear to be of not much economic value. Of the remaining four *C. gryphoides* and *C. discoides* are mostly found in the north-western coast of India, the former being more important, occurring in the coastal areas of North Kanara and Maharashtra in considerable numbers. *C. madrasensis* is known to grow (Pl. I A, B) abundantly in wild state forming extensive beds in the tidal creeks and backwater areas of the east coast. In addition to this settlement of this species in the estuarine regions of many east coast rivers is not uncommon. Unfortunately nowhere in the east coast is this valuable resource commercially fished for food except on a limited scale by the Tamil Nadu Fisheries Department arranging to collect a few thousand oysters every year from the wild stock in Pulicat Lake and Ennore Estuary for supply to a few western-style hotels and westerners in Madras. The wild oysters get stunted in growth because of overcrowding in beds and pose great difficulties in harvesting. The habitat where the oysters thrive in nature, is also looked down upon by fisherfolk. Exploitation from the rugged natural beds is unremunerative considering the time spent in gathering them.

To the developed nations, oyster meat is well known for its epicurean value. Added to this, protein value of oyster meat is high comparable to other shell fishes. These factors and natural resource richness of *Crassostrea madrasensis* in the east coast of India influenced the formulation of a programme of oyster farming at Tuticorin adopting a suitable technique. The strategy was to culture oysters in the coastal area so that once the technology of growing oysters is proven a few thousand hectares in the shallow backwater areas out of an estimated 2 million hectares of such water bodies in India can be profitably utilised for developing and promoting oyster culture industry.

Selection of Farm Site

Before deciding upon the location for the oyster farm, extensive survey was taken up during the years 1975-76 to determine the areas where important oyster beds exist in the long coastal districts of Tamil Nadu. Of the 42 river mouths and backwater areas from Madras to Cape Comorin, 11 were identified as containing sizable population of oysters. The area between Madras to Point Calimere being a cyclone prone one, it was decided to consider initial culture experiments either along the Gulf of Mannar Coast or in the Palk Bay zone. Of

these two areas, the Vaigai Estuary at Attankarai is the only area with pronounced oyster bed formation in the Palk Bay side. The bar mouth here remains closed for most of the period during summer with the result that there is progressive increase in salinity due to solar evaporation and heavy oyster mortality occurs. During the north-east monsoon period following summer months freshets in the river lower the salinity to near freshwater condition which again acts as an adverse factor for oyster growing experiments. In the Gulf of Mannar, three important regions where oyster populations exist over considerable stretch were identified (1) Koothanguli Backwater region where Hanuman Nadhi, a small tributary joins it, (2) Tambaraparani Estuary at Pinnakayal near Dharangadhara Chemical Factory water pumping station and (3) Tidal inlets in the vicinity of Tuticorin. The oyster beds at Tuticorin are situated in tidal inlets leading to considerable expense of shallow water. The shallow water creeks and the adjacent areas were found suitable for conducting experiments. Another important factor was the easy availability of labour, infrastructure facilities and marketing channels. An oyster farm was therefore established at Tuticorin in one of the creeks, the Karapad Creek.

Karapad Creek

The western bank of the shallow Tuticorin bay is deeply indented in two places one at Karapad and the other at Uppar. The Karapad Creek is situated 2 km south of Tuticorin (Pl. I C). This tidal creek, 2 m deep and 30 m broad at its mouth, cuts into the land westwards as a channel for a near 750 m course after which it leads to a waterspread bathing an expansive shallow mangroved-fringed swampy area to the north and meanders further south-westwards to a distance of 2 km (Pl. II A) becoming shallower in many spots wherever sandy shoals intervene. Such areas get exposed during tidal egression.

Natural oyster beds

Extensive beds of oysters exist in the shallow areas of mangrove fringed watermass, while in the proximity of exposed flats and on either bank of the waterspread also patches of live oyster settlement are not uncommon. The oysters are densely settled in several clumps of which the top layers contain mostly dead oysters because of continued exposure during certain seasons of low tidal amplitude after settlement. The oysters in the lower strata are mostly silt-covered (Pl. II B). Both dead and live ones are met with here. The creek area is comparatively free of oyster settlement except for stray settlement in the intertidal area of a few concrete pillars of a make-shift bridge connecting the north and south banks of the channel. The salinity, temperature and other physico-chemical features of the creek water are identical to those of the open bay water since there is a free and uninterrupted ingress and egress of sea water daily in the creek throughout the year. A standing column of 1 m water depth is thus maintained even at the time of the lowest tide. At spring tides the high water level exceeds 2.5 m. The bottom in the channel is of muddy sand while the interior of the creek is muddy and slushy. For the culture experiments the channel area was preferred because of the nature of bottom, free water-flow and uniform depth.

Method of Culture

Of the several culture methods followed in different countries it was felt that in coastal areas where surf does not break, the 'rack-tray' culture method would give desired results. The creek with soft bottom would pose few problems for erecting wooden racks also. Hence this method was chosen.

a. Oyster rack

Twelve numbers of 2.4 m long wooden poles of 7.5 cm diameter, earlier coated with tar, with one end sharpened to enable them to be

easily driven into the sandy bottom 60 cm down, were planted in a line at intervals of 1 m across the creek. Two such equidistant parallel rows were driven to complete a single rack. At least 1.8 m length of these poles project above the bottom. One cut piece each of the same quality wood 1.4 m long and 6-7 cm diameter was tied parallel and across from pole to pole at a height of 0.9 m above the bottom. The finished frame structure served as the platform frame for placing the trays containing oysters. Coir rope was used for fastening the poles although nylon cord can also be used for additional strength. The height of the frame was so adjusted to enable the trays containing oysters resting on them to remain submerged in water during normal low tide periods (Pl. II D). For controlling the settlement of fouling organisms the trays were adjusted to remain exposed during low tides by manipulating the height of platform frame. A platform thus constructed will be able to hold 20 trays each.

b. Trays

The tray fabricated for this purpose was rectangular in shape of 90 × 60 × 15 cm dimensions (Pl. III A). The frame of the tray was of 6 mm welded steel to which 2 mm nylon twine netting of 20 mm mesh size was given on the sides and bottom. The frame of the tray was coated with lacaloid black paint as an anticorrosive measure. The nylon meshed trays are strong enough to bear the weight of the oysters, at the same time allowing free water flow.

Each rack was designed to give an area of 25 sq.m allowing space on all sides for farm work to be carried out using 2.75 m long flat bottomed fibreglass dinghy. The rectangular trays were fashioned to hold 150-200 oysters each. Thus provision was made to accommodate 20 trays to grow 3000-4000 oysters in a 25 sq.m area. By erecting a panel of such racks parallel to the previous ones it was possible to increase the area of cultivation. 30 such racks for experiments were put up.

c. Oyster spat collection

Preparation of lime-coated tiles : Of the several experiments tried with different spat collection materials (details of which form the matter for publication elsewhere), country roofing tiles of 24 × 15 cm were found to be the best suited for collecting large quantities of oyster spat from nature. The tiles were first coated with a layer of lime by dipping them individually in a vat containing sufficient quantity of slaked lime. The tiles were then spread out in shaded area to dry for 24 hours. After this the tiles were again dipped in slaked lime with a mixrure of sand to obtain a second coating with rough surface. The tiles were dried as before and kept stocked for use during collection season (Pl. II C). Setting properly prepared tiles in the right season was considered very important to achieve maximum success.

Gonadial maturity of oysters and spawning season

Regular sampling of oysters from natural bed for a 12 month period to determine the gonadial maturity stages enabled the determination of the right time of spawning. It was noticed that in the natural beds majority of oysters attained maturity in April and August. Ripe female and male oysters discharged the reproductive elements in the first fortnight of April when the temperature was 28.8°C, salinity 34.8‰ and oxygen content 4.5 ml/l (Fig. 1). Spat collectors laid in the natural bed area (during April first week) on low submerged wooden platforms enabled large scale settlement of oyster spat on the tiles. At the time of laying spat collectors the following precautions were taken :

i. The platform on which tiles were arranged always remained submerged in water so that the tiles would not get exposed at any time till they are finally removed for scraping the oyster spat.

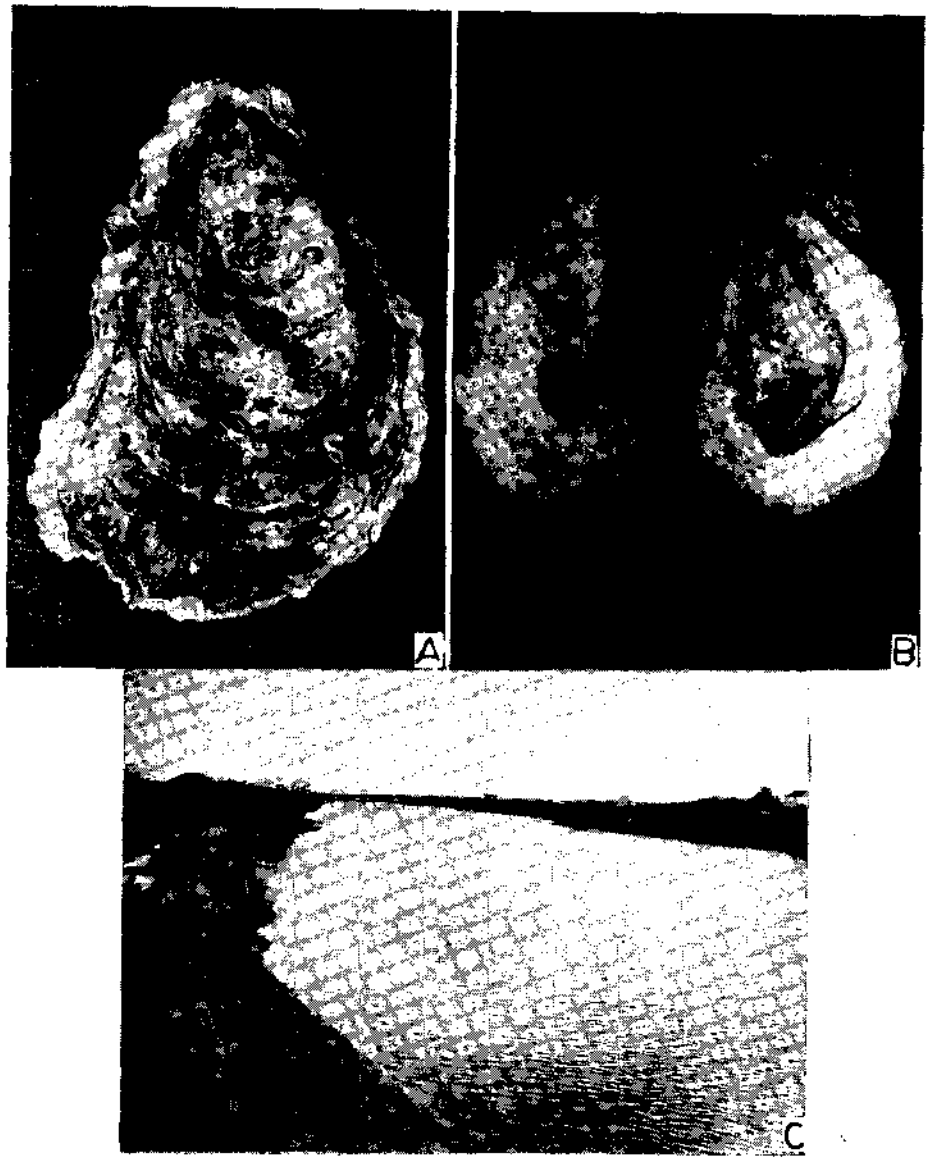

PLATE I. A. The edible oyster *Crassostrea madrasensis*, B. Shell valves of *C. madrasensis* opened to expose the body of the oyster and C. A view of the Karapad Creek at Tuticorin.

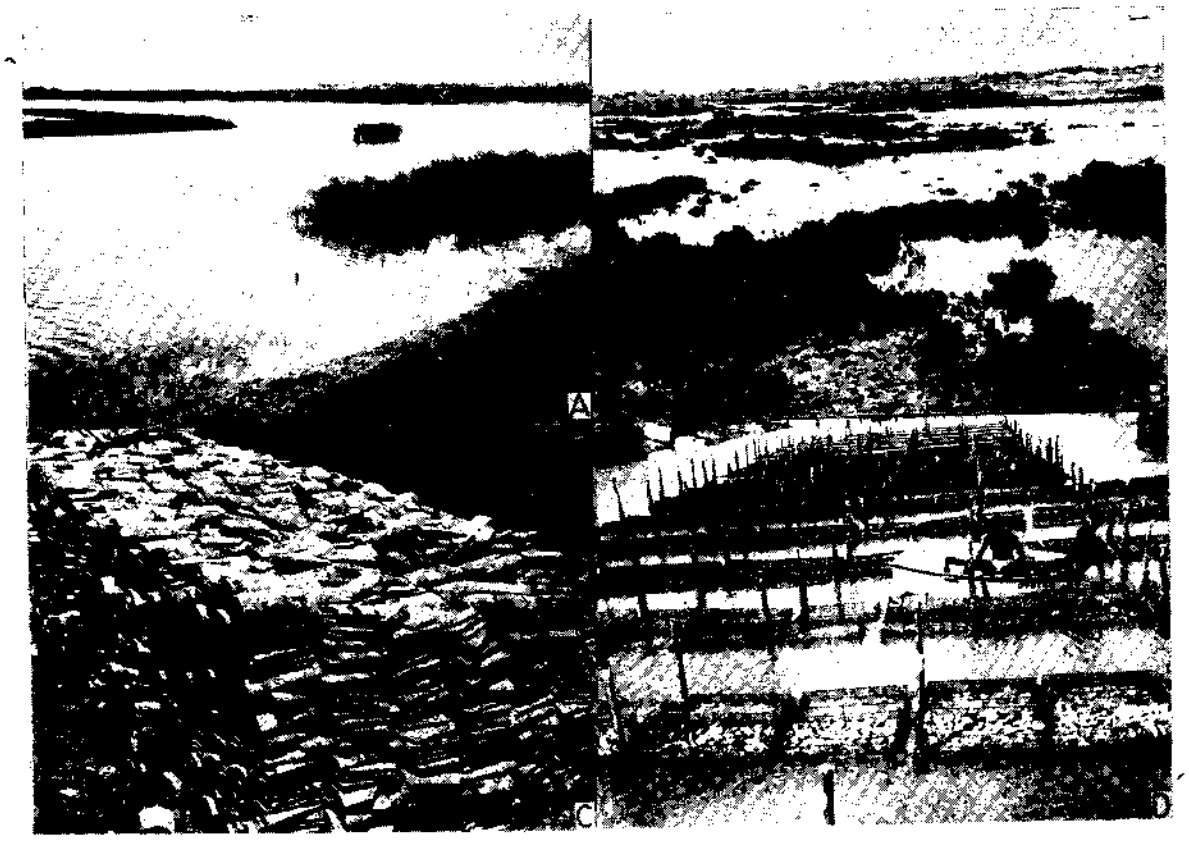

PLATE II. A. A view of the Karapad Creek spreading over mangrove fringed area to the right and continuing southwestward, B. Natural beds of edible oyster exposed partially during low tide, C. Lime-coated tiles used for spat collection and D. The oyster growing racks in the creek (Note the oysters kept in the rectangular trays).

PLATE III. A. Rectangular tray used for growing oysters, B. Box-type cages used for initial growth of scraped oyster spat, C. Oyster spat settled on tiles and D. Scraping of spat in progress.

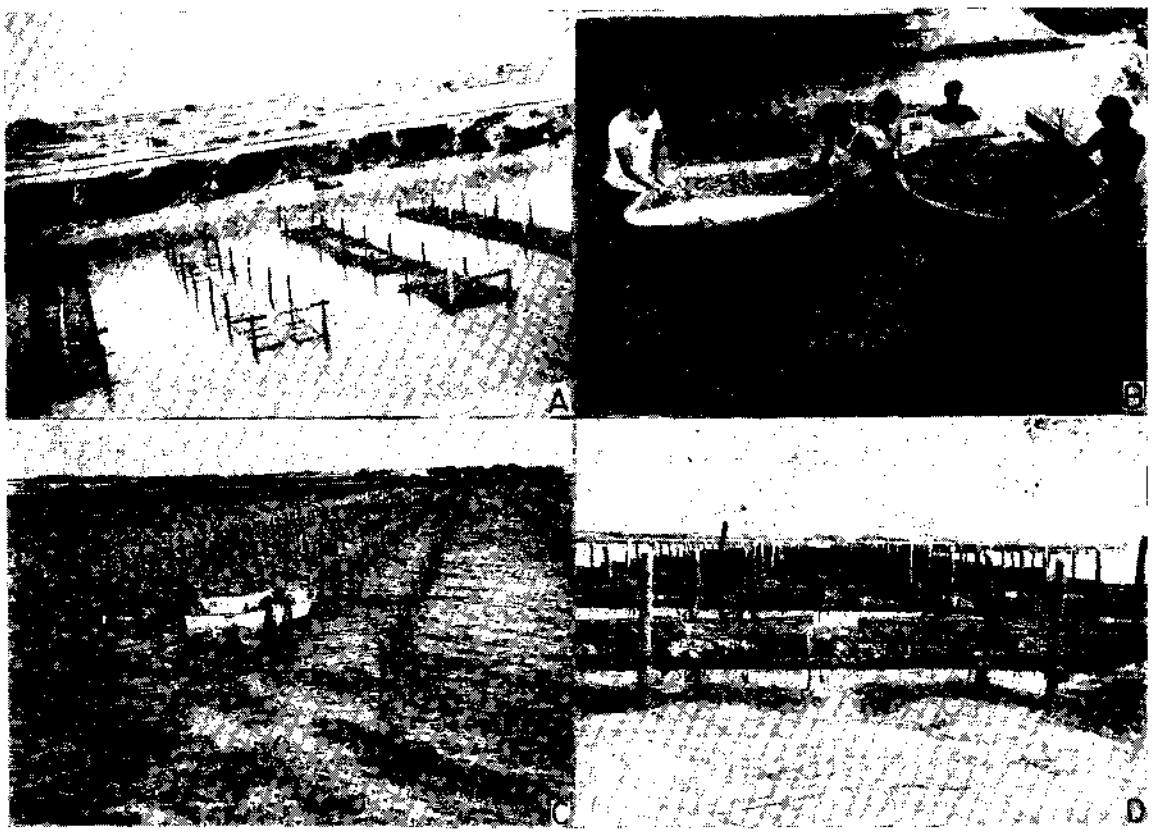

PLATE IV. A. The erosion of the bank of the creek, B. Harvested oysters being taken in fibreglass dinghies, C. A view of the oyster farm in open sea coast and D. A view of the open sea oyster farm at very low neap tide.

ii. The selection of site where spat collectors were laid was free of currents and was a calm area. Spat collectors were laid in different places in the vicinity of natural oyster beds.

iii. The concave side of tiles was kept facing down while being positioned on collection racks since it is this darker side that oyster larvae prefer for attachment.

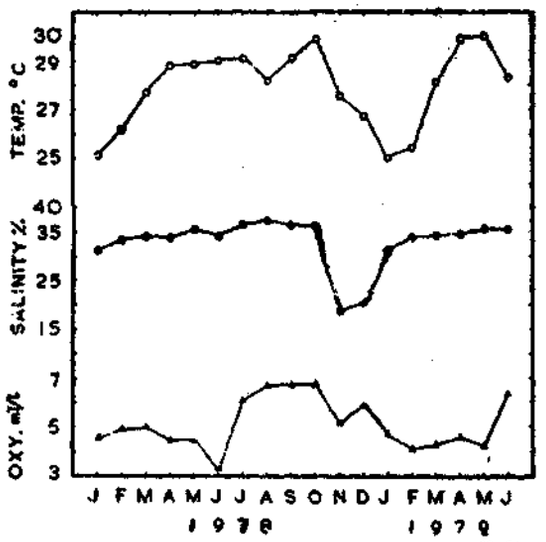

Fig. 1. Seasonal variations in temperature, salinity and dissolved oxygen in oyster culture area at Tuticorin during 1978-79.

iv. Since the time taken for oyster larvae to settle down after fertilisation may vary from 15-20 days or even more depending on environmental conditions, spat collectors were laid only after satisfying about the ripe condition of the gonads. This gave maximum results as otherwise fouling community settlement on tiles effectively reduces or prevents good spat settlement.

Periodically the spat collectors were examined to see the progress of spatfall. Spat settlement ceased by April end. In August when water temperature was similar to that in April, 28.1°C, and salinity and oxygen higher, 37.08 ‰ and 6.7 ml/l respectively spatfall was very less as compared to that in April. On many tiles as many as 100 spat could be collected in April although the average was about 40 per tile (Pl. III C). The spat were allowed to grow on tiles till they attained a size of 30 mm in about 45 days. After this, scraping of spat was done (Pl. III D). Very little force is needed to dislodge the spat from the tiles. During the first season it was possible to collect a total of 600,000 spat by the above method. The spat were then transferred to box type nylon meshed cages for initial growth since the oyster seeds at this size were light and small.

The box type cages were of identical material as the rectangular trays except that these were much smaller of 40 × 40 × 10 cm size with 12 × 12 mm meshed nylon netting and provided with a lid (Pl. III B). The lid ensured that the oysterlings were not thrown out and lost because of wind and wave action. 400-5000 oysterlings were put in each cage and suspended using nylon ropes from the cross poles of the rack, below the surface of water. The cages were often examined and algal filaments clogging the meshes were removed and gentle scrubbing was resorted to for removing barnacles and other foulers. Monitoring of physico-chemical properties of the creek water was done as also the collection and analysis of the planktonic food available. Phytoplankters were found to be present throughout the year with blooms of *Rhizosolenia* spp. and *Chaetoceros* spp. occurring from June to October.

Growth of oysters and production

The study on the growth of the oyster spat in the cages revealed that there was rapid growth in the first three months and a size of 38 mm was attained in 60 days by July, 1978 giving an average of 12.6 mm growth per month. At this stage oysters were transplanted to open rectangular trays for further growth. Manage-

ment of the farm stock after this needed only the examination of oysters once in a month, cleaning of oysters and trays and removal of predators. The monthly rate of growth was less in later months with very poor growth from December to March. At the end of the first year the average growth recorded was 84 mm (Fig. 2) with a total shell and meat weight of 120-130 g per oyster of which the meat weight alone was 8 gm. Mortality out. The deflection of water current by the rack panels caused erosion of the raised channel banks damaging nearby structures on either bank (Pl. IV A). The creek water being the main source of supply of sea water to adjacent salt pans the creek entrance had to be temporarily closed whenever large-scale maintenance and repair works were undertaken by the salt pan owners. This happened once towards the end of the present initial experiments by

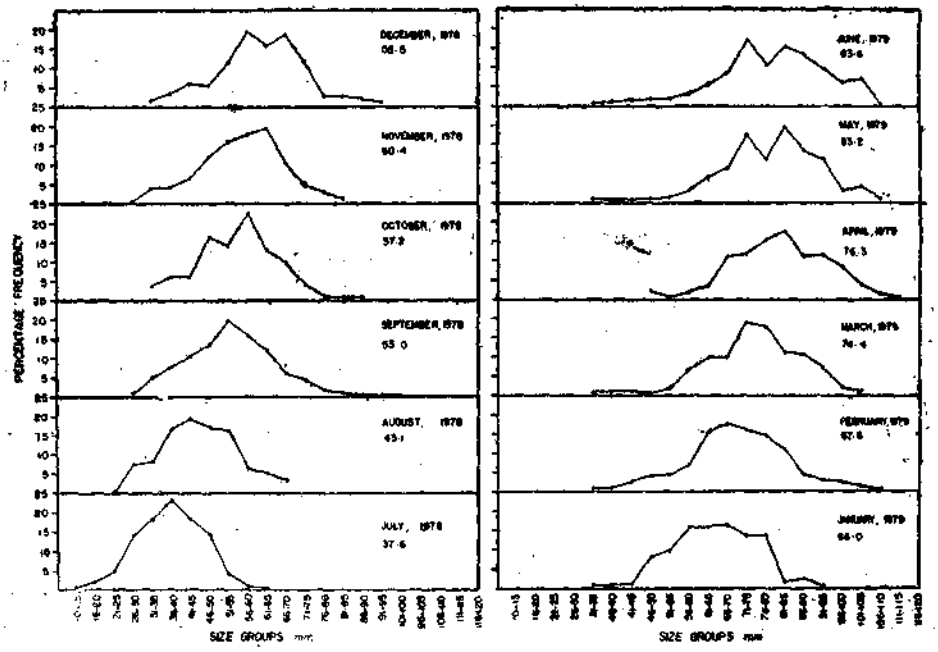

Fig. 2. Size frequency composition and average sizes of cultured *Crassostrea madrasensis* at Tuticorin during 1978-79.

noticed in the farm was very little (5%). The production per rack containing 4,000 oysters (Pl. IV B) at the end of one year was 425 kg (shell on) indicating a production rate of 119 t/ha with a meat yield of 9 t/ha.

Difficulties experienced in farming in the creek

Because of the obstruction to the free flow of water by erecting racks and placing trays silting occurred in the area covered by racks. Desilting operations had to be constantly carried which time we had obtained necessary basic data and information for planning large-scale confirmatory farming work. Because of the problems in the creek the farm site was shifted in 1979 to the shallow Tuticorin Bay on the southern side of the Karapad Creek.

Farm site at Tuticorin Bay

The shore of Tuticorin Bay is shallow with depth ranging from 1.5 to 2.5 m. The nature of bottom is sandy with a shoreward approach

through a flat of inter-tidal mud and sand (Pl. IV C). Roughly 100 hectares of such shallow area available is not subjected to high waves or wind action normally and therefore was considered suitable for establishing the oyster farm. The oysters in the natural bed in the Karapad Creek and the stock held in the oyster farm would produce seeds needed for eventual large-scale farming venture. 80 racks were constructed and 2,50,000 oyster spat collected from the natural bed in April 1978 were grown in the farm at the rate of 3,000 spat per rack (Pl. IV D). The growth rate was very satisfactory although mortality of transplanted stock of size 45-65 mm was noticed in June-August due to predation by one species of gastropod entering the trays. This gastropod was identified as *Cymatium cingulatum* (details of this are reported elsewhere by Thangavelu and Muthiah). The problem was tackled by periodical examination of trays and removing the gastropods daily during this period. The estimated production of oysters from rack and tray system of culture was less in the Tuticorin Bay as compared to that in Karapad Creek and amounted to 90 t (shell on)/ha with a meat production of 7.5 t/ha.

Further intensification of farm work was taken up by the end of 1979 and an additional 90 racks had been put up where the work of oyster growing was entrusted to 15 small-scale fishermen. Orientation training in all aspects of oyster culture was given to them. Spat collected in April, 1979 by using country tile technique enabled to keep a stock of 5,00,000 which were utilised by the fishermen for farming. This new venture is being watched keenly and if successful would add a new dimension in establishing oyster farming as an additional income generating venture.

Discussion

The foregoing account of experimental culture of edible oyster carried out at Tuticorin has shown the feasibility of growing oysters in shallow coastal areas and tidal creeks to marketable size with a production of 90—119 tonnes of oysters/ha with a meat yield of 7.5 9.0 t/ha. This much of production cannot be expected from natural beds due to obvious reasons. In Australia where *rack* system of culture is followed in many areas, 2,000 kg of oyster meat has been produced per hectare (Bardach *et al.*, 1972). In the same country *tray* culture is reported to yield 5,400 kg of oyster meat per hectare. In this context the results achieved at Tuticorin appear to be significant. Although culturists in Japan are able to produce 26 tons of oyster meat by longline system and 20 tons by *raft* culture such attempts do not appear to be immediately possible in the deeper areas of our coast because of the non-existence of sheltered bays. The 'lines' or 'rafts' positioned in the open sea will not only hamper navigation, but also clash with the interests of the traditional fishermen exploiting these areas.

It was Hornell (1910) who first suggested taking up oyster culture in Tamil Nadu and after seven decades we are seriously implementing that idea along with similar attempts in the field of mussel culture, etc. The results achieved are very encouraging and it is hoped that this will find favour with culturists.

Although the present culture experiments at Tuticorin have shown the possibility of a reasonably high output by the 'rack-tray culture' method, ample scope exists to achieve higher production values per hectare by raising the number of oysters in a rack increasing the number of trays per rack and keeping them in a two tier arrangement of 20 trays per tier, the two tiers being one below the other but well above the bottom. Predator problem does not appear to pose difficulties at Tuticorin since vigilence during the period of predator abundance will help to eradicate the gastropod which occurs in the area. It cannot be said that throughout the east coast the same problem alone would confront the farmers. Problems

might differ from region to region and by experience these can be identified, documented and remedial action taken as otherwise mortality of farm stock would result in the venture becoming unproductive and unremunerative.

Keeping the future perspectives in mind it appears necessary that in our effort to establish a permanent oyster industry attention has to be paid to the development of hatchery system for seed production which will reduce our total dependance on nature for seed requirements for the large-scale expansion of this industry. This has been identified, therefore, as a priority area of research.

It is too early to work out the economics of the culture attempted based on the data available. But there is every hope that the margin of profit will be sufficiently attractive to provide necessary incentive to the farmers to come forward to take up oyster farming. Consumer preference and market demand are no doubt the deciding factors, for which systematic extension work is necessary. Although the paper does not deal with the steps proposed to be taken to find marketing avenues, it may be mentioned here that some years ago people never thought much of the potentialities of market for prawns and lobsters, but found themselves in a world wide demand trade for prawn meat which has resulted in keen interest in prawn farming in India. In this context oyster culture should be taken up as a long range development programme initially to cater to local market demands and later for export market.

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OBSERVATIONS ON THE SETTING OF SPAT AND GROWTH OF *CRASSOSTREA MADRASENSIS* IN VAIGAI ESTUARY AT ATHANKARAI

K. SATYANARAYANA RAO, D. SIVALINGAM AND K. A. UNNITHAN
Central Marine Fisheries Research Institute Cochin-682 018

ABSTRACT

The setting of spat of the edible oyster *Crassostrea madrasensis* on different kinds of spat collectors like oyster shells, wooden pieces, concrete pieces, concrete slabs, tiles, bamboo frames and coconut shells kept in Vaigai Estuary at Athankarai was studied. It was observed that there was spatfall on concrete pieces with irregular surfaces given lime coating, on oyster shells given a coating of cement or lime and lime-coated curved tiles. Oyster spat set in stray numbers on bamboo frames, Mangalore tiles and coconut shells, spatfall took place between January and April and generally only small numbers of spat were seen to set on spat collectors in the other periods of the year. Oysters attained an average size of 86.7 mm and a maximum of 110 mm at the end of one year. Average and maximum sizes of 89.6 mm and 130 mm were attained at the end of 14 months and growth of oysters was very much retarded later.

INTRODUCTION

THE BACKWATER OYSTER *Crassostrea madrasensis* is widely distributed in India and occurs in the estuaries and backwaters along the east coast in Orissa, Andhra Pradesh and Tamil Nadu and on the south-west coast in Karnataka and Kerala. For successful culture of oysters, oyster spat have to be collected in large numbers by using appropriate spat collectors and the spat and growing oysters have to be reared to marketable size by spreading them on the bottom or keeping them in trays on racks in unpolluted brackish or sheltered coastal waters.

In Vaigai Estuary at Athankarai about 30 km from Mandapam camp on the south-east coast of India, there are beds of *Crassostrea madrasensis* extending over an area of 1.5 ha (Rao *et al.*, MS). Efforts have been made to collect spat of oysters in the estuary and a preliminary report was given by Rao (1976). Detailed observations on the spatfall on collectors and growth of the oyster are presented in this paper.

*Present address: Tuticorin Research Centre, Central Marine Fisheries Research Institute, Tuticorin 628 001.

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MATERIAL AND METHODS

Spat collectors of nine different types were used to determine their relative efficiency for collection of oyster spat : oyster shells given a coating of cement and fine sand in the ratio 2 : 1 mixed with water, oyster shells given a coating of lime, sand and mud in the ratio 2 : 1 : 1, wood plates 10 × 10 × 2.5 cm in size given lime-coating, lime-coated concrete pieces with irregular surfaces 12.5 × 12.5 × 12.5 cm in size, concrete slabs 30 × 15 × 2.5 cm in size, lime-coated curved tiles 23 × 15 × 2 cm in size, lime-coated Mangalore tiles 33 × 25 × 2.5 cm in size, lime-coated coconut shell halves,

frames of the sizes 1 × 1 m and 1.8 × 1 m made of bamboo strips.

Oyster shells, five each, were tied to bamboo strips and fixed in the estuary. Shell strings were hung from horizontal poles. Shells were also kept in bags of synthetic twine netting or in trays which were placed on racks erected in the estuary. Tiles, curved ones and Mangalore type were kept in trays on racks. Curved tiles were also kept spread on racks. The concrete pieces, concrete slabs and coconut shell halves were hung from horizontal poles. The bamboo frame spat collectors were filed vertically.

The spat collectors were placed in Vaigai Estuary at Athankarai in the vicinity of oyster beds. They were laid in the months December-January every year and replaced by fresh ones if there was fouling. The spat collectors were set about 0.5 m above the bottom. At weekly intervals the spat collectors were cleaned with wirebrush and fouling organisms like algae, barnacles and polychaetes removed. When the juvenile oysters grew to a size of 25-30 mm, they were removed from the spat collectors. The same collectors were used again to study the seasonal variations in spatfall. The dislodged oysterlings were reared in trays kept over the racks. The spat collectors were examined once every week and the number of fresh spat which had set was noted. Growth of spat was studied by their size frequency. Water temperature and salinity data were collected during 1973-78.

Results

It was observed that oyster spat set a minimum of two weeks after laying the spat collectors. A slimy layer on the collector appeared to be favoured by the oyster larva for setting.

Spatfall on various kinds of spat collectors

A. *On oyster shells given cement coating*

Of the 400 shells tied to bamboo strips and placed in December 1972, many of them did not collect any spat. A total of 156 spat and 89 spat were collected in January and February 1973 respectively. Very few spat settled in March, May and June. The mean spatfall was 0.7 spat/shell. Among the 180 shells on strings, 19 spat were observed in January and stray ones during February-May. The mean spatfall was 0.2 spat/shell (Pl. I A).

B. *On oyster shells given lime coating*

On this kind of spat collector tied to 30 synthetic ropes, each with six shells, 166 spat had set in the period January-March, 1974 and 12 in June and July, 1974 with a maximum of 3 spat and an average of 1 spat per shell in the season. On shells tied to 40 ropes the spatfall was low in the first quarter of 1975 being 32 and another 45 spat had set on a total of 50 ropes in July 1975 indicating setting on an average 0.3 spat per shell in the season. In January 1976 a total of forty spat had set and January 1978 only two spat were seen on this type of spat collectors tied to 50 ropes. The poor spatfall in January 1976 is attributed to failure of spawning of oysters. On 480 oyster shells given lime coating and kept in 16 bags of synthetic twine netting only 29 spat had set in January, 1978 and 16 in February, 1978 with an average of 0.1 spat per shell.

On keeping 2,000 oyster shells given lime coating in ten trays placed on a rack in December 1977, 279 spat set in February, 191 in March and 16 in July 1978 with a maximum of 3 spat and average of 0.2 spat per shell.

C. *On wooden pieces given lime coating*

72 ropes with 4 wooden pieces tied to each were kept in the estuary in 1974 and 1975 but only stray oyster spat had set on this spat collector in February and April 1974 and January and February 1975.

D. *On concrete pieces with irregular surfaces given lime coating*

When 30 ropes with a total of 150 concrete pieces were kept, 405 spat had set in the first

quarter of 1974. A maximum of 24 spat with an average of 2.7 spat had set on each concrete piece, setting not taking place on some of the pieces. In 1975, 1976 and 1978 spatfall was very poor in the first quarter but it was better in September and November 1976 when 238 and 125 had set on keeping a total of 30 ropes with 150 concrete pieces with an average of 1.6 spat and 0.8 spat per concrete piece in the two months respectively.

E. *On concrete slabs given lime coating*

34 ropes each tied with three concrete slabs were kept in the estuary from the first quarter of 1975 but only stray spat had set in March 1976.

F. *On curved tiles given lime coating*

1,720 curved tiles were kept in 48 trays on three racks each tray containing 40 tiles in December 1977 (Pl. II B). 158 oyster spat had set in February 1978 and 787 spat in March 1978 with a maximum of 32 spat setting per tile on the concave surface of tiles (Pl. II A). On many tiles spat fall did not take place. In May and June 185 and 162 spat had set and the spatfall was much lower, 15 and 48 in July and August 1978. The average spat which had set per tile was 0.8 in the season.

1,200 curved tiles were kept in six bamboo frames in February 1978 and another 1,200 curved tiles were kept on six additional frames in July 1978. There was good spatfall on tiles kept on bamboo frames, 1,970 spat had set on the spat collectors in March 1978. In May, June, July and August 1978 the numbers of spat which had set were 439, 289, 31 and 76 respectively. A maximum of thirty spat had set on a tile and the average number of spat set on each tile was only 1.2 per tile there being no spatfall on a large number of tiles.

G. *Lime coated Mangalore tiles kept in trays*

100 Mangalore tiles were kept in 10 trays on a rack in January 1978, each tray containing ten tiles. Only stray numbers of spat had set on these tiles in March, July and August 1978. In March 3 spat had set and a total of 25 and 47 spat had set in July and August 1978. A maximum of 4 spat had set on this type of collector per tile with an average of 0.8 spat per tile.

H. *Lime coated coconut shell halves*

500 lime coated coconut shell halves were suspended from 100 synthetic ropes with 5 shells per rope which was weighted with a stone at the bottom. On this kind of spat collector also only small numbers of spat had set. One or two stray spat had set on the inner side of a few shell halves and on an average 0.1 spat had set per shell half(Pl.I B). In April, May and June 1978, 30, 4 and 2 spat had set. The spat adhered firmly to the coconut shells and it was difficult to remove them without injuring.

I. *Frames made of bamboo strips*

Ten frames 1 × 1 m in size were kept fixed vertically in the estuary in November 1973 and ten bigger frames 1.8 × 1.8 m in size in February 1978. In 1974 there was no spatfall and a few stray spat had set on the spat collectors in February 1978.

Growth of spat and oysters

The size range of oyster spat which had set on oyster shell spat collectors was 8.5 to 33.0 mm in March, 1973 with a mode at 15.5 mm (Fig. 1). This mode shifted to 35.5 mm in April, 1973 and 45.3 mm in May, 1973 showing a rapid growth of 20 mm in March. Between April and May the growth of modal size group is only 9.8 mm but the mode at 45.3 mm in May shifted to 65.0 mm in June with a growth of 19.7 mm and the maximum size was 83.0 mm. In July, 1973 the mode was at the same position and it increased to 75.2 mm in August and 75.4 mm in September. A few spat had set in May and June. The modal sizes were 84.8 mm and

PLATE I A. Spat of *Crassostrea madrasensis* set on oyster shell spat collector tied to synthetic rope and B. Oyster spat set on coconut shell halves.

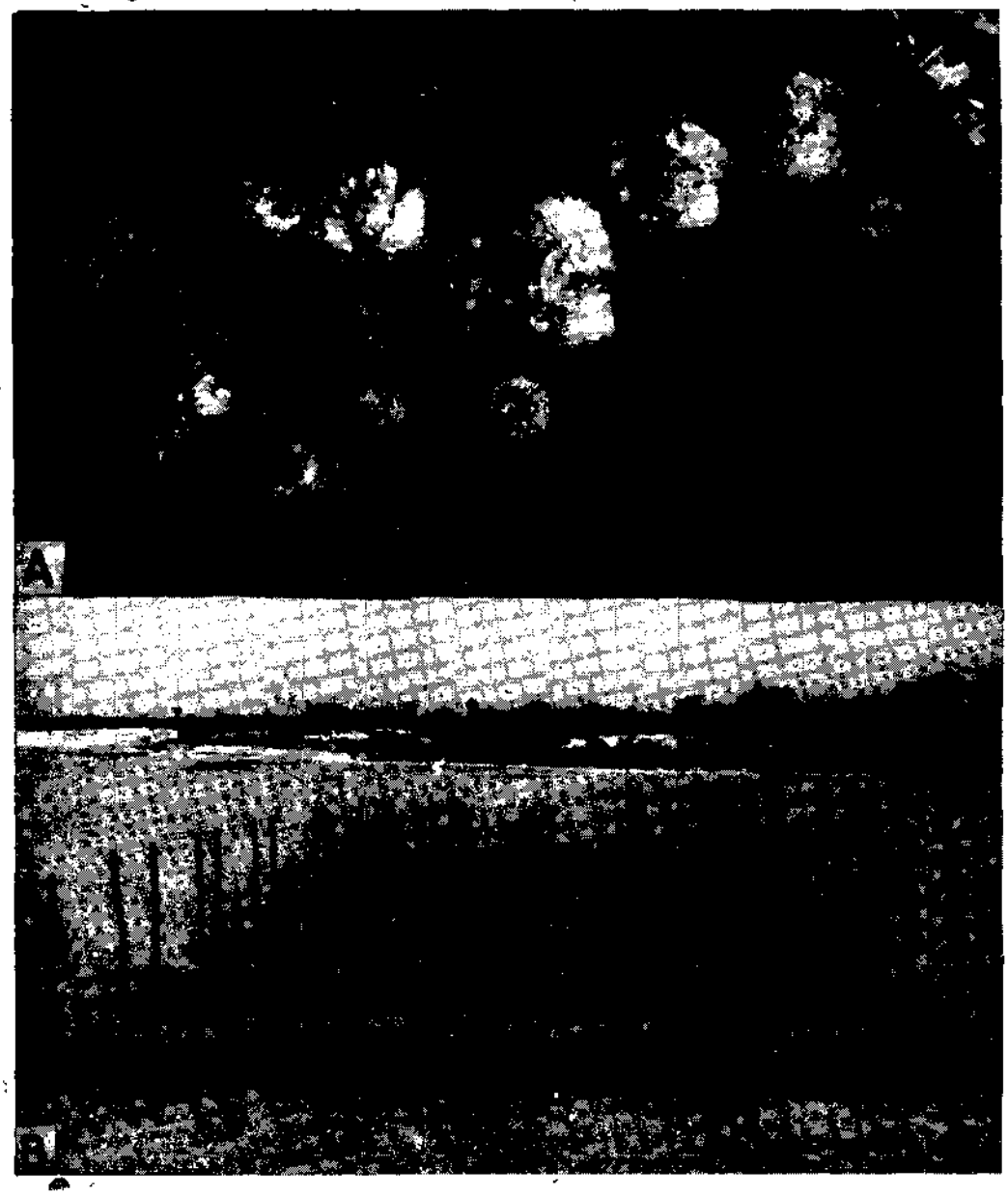

PLATE II A. Oyster spat set on a lime-coated curved tile and B. A view of the racks on which spat-collectors and growing oysters were kept in trays in Vaigai Estuary at Athankarai.

84.0 mm and maximum sizes 107 mm and 111 mm in October and November, 1973 respectively. In January, 1974 the modal size was at 96 mm and the maximum size 110 mm. This mode attained a size of 106.3 mm in October, 1974 126.2 mm in July, 1975 and 129.6 mm in March 1976. The maximum size was 111 mm in November 1973, 119 mm in February 1974, 130 mm in March 1974, 133 mm in July 1975, and 150 mm in March, 1976. It could be considered that the spat 8.5-33.0 mm in size seen in March, 1973 had set in the period January-March. An average size of 86.7 mm and maximum size of 110.0 mm are reached at the

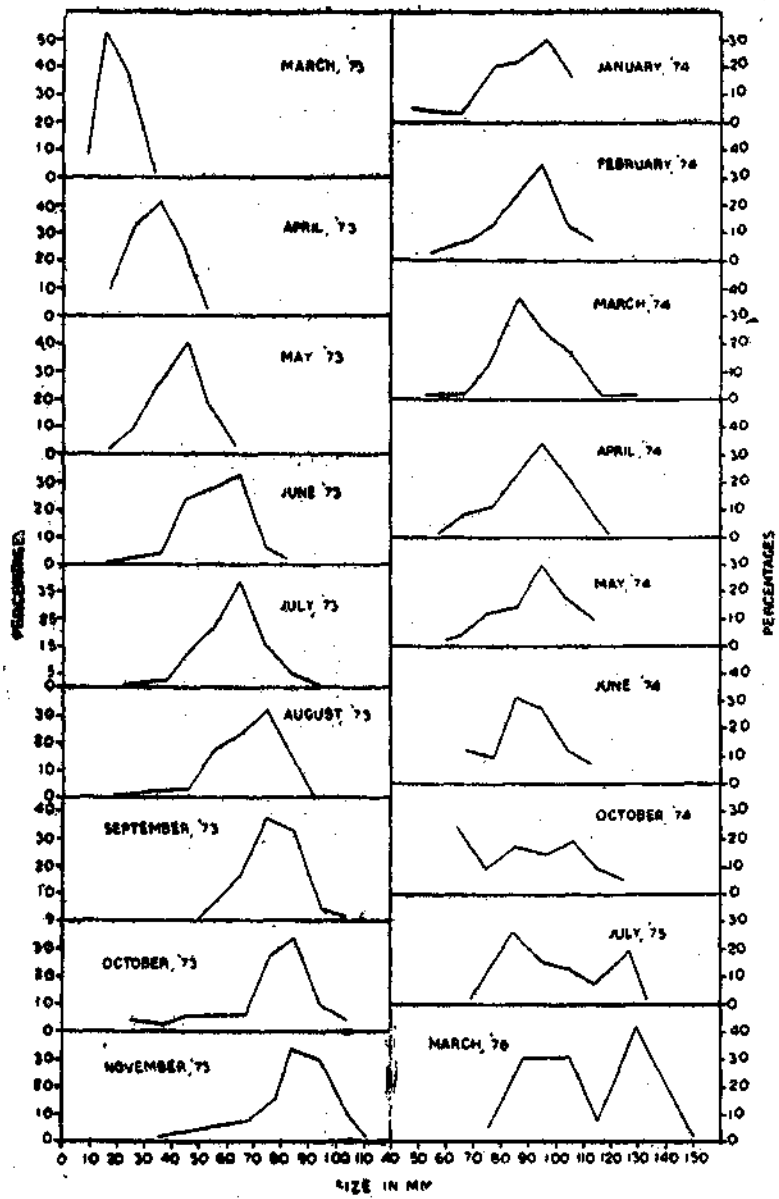

Fig. 1. Size frequency of *C. madrasensis* cultured in Vaigai Estuary at Athankarai.

end of one year in January, 1974. The cultured oysters showed further growth and attained an average size of 89.6 mm and maximum size of 130 mm in March, 1974. Even after another two years the average size attained is only 112.5 mm and maximum size is 150 mm in March, 1976.

Crassostrea madrasensis which set in December - January 1974 showed a mode at 25 mm in January 1974 which shifted to 35 mm in February 1974 and 55 mm in April 1974. A mode at 35 mm in May shifted to 55 mm in August. A single mode was seen at 65 mm in October which increased to 75 mm in December 1974. The maximum size reached at the end of December 1974 was 100 mm.

Oyster spat which had set in November 1975 showed a mode at 25 mm in January 1976 which increased to 35 mm in February, 45 mm in April and 55 mm in May. It was stationary at 55 mm in July and September but increased to 65 mm in October, 85 mm in November and 95 mm in December. The maximum size reached at the end of 1976 was 135 mm.

The 1978 year class showed a mode at 5 mm in February and March and at 10 mm in April and May. In June two modes were seen at 15 mm and 65 mm and these shifted to 25 mm and 75 mm in September.

The 1975 year class oysters grew to an average size and weight of 82 mm and 112 gm and had an average meat weight of 9.3 gm at the end of one year. The 1978 year class oysters which had set in February reached an average size and weight of 76 mm and 48.3 gm and had an average meat weight of 6.2 gm at the end of seven months in September 1978.

Survival

The survival rate of oysters from spat which had set on shell spat collectors with cement coating was 36-41 %, that from shells with lime coating 46-56%, that from concrete pieces with irregular surfaces 76-79%, that from curved tiles 67% and that from Mangalore tiles 70%. The survival rates of oysters set on concrete pieces may be seen to be highest followed by that on Mangalore tiles and curved tiles. Surviavl rates of spat on oytser shells was low.

Hydrological characteristics of Vaigai Estuary at Athankarai

Data on water temperature and salinity in Vaigai Estuary at Athankarai for a typical year, 1973 are given in Table 1. The temperature increased over a limited range from 27.4°C in January to 32.7°C in April after which there was a decrease in the succeeding months. In the other years 1974-78 the pattern was similar

TABLE 1. *Seasonal variations in water temperature and salinity of Vaigai Estuary at Athankarai during* 1973

Months		Temperature °C	Salinity ‰
January	..	27.4	29.98
February	..	28.0	27.98
March	..	30.7	29.82
April	..	32.7	33.66
May	..	32.4	34.81
June	..	31.0	32.46
July	..	30.0	35.53
August	..	29.5	34.98
September	..	29.0	34.07
October	..	28.0	6.94
November	..	27.5	7.20
December	..	data not available	

with lowest temperature of 26°C in January 1976 and a maximum of 33.3°C in April 1974. Unlike temperature, salinity of the estuary varied markedly during the course of a year. It was 29.98‰ in January, 1973 and rose to values ranging from 32.46 to 35.53‰ in the months of April-September. The salinity declined to 6.94-7.2‰ during October-November 1973. Due to rains of north-east monsoon and freshets from the upper reaches of River Vaigai. In the periods July-September and

October-November 1973 when the salinity was high and very low respectively, there was retardation of growth of oysters indicating that high as well as very low salinity conditions are not favourable for growth of the oysters. Rao and Nayar (1956) also observed that constantly high and low salinities slackened the growth of *C. madrasensis* of Adyar Estuary. In Vaigai Estuary at Athankarai very high salinities of 52.35‰, 62.39‰ and 47.41‰ were recorded in August and September 1976 and September 1977 due to evaporation of estuarine water when the estuary was cut off from the sea by the formation of a sand bar.

Pests and Predators

The green alga *Enteromorpha* sp. and the red alga *Polysiphonia* sp. grew over the outer surface of the valves of cultured oysters and formed a covering over them but they did not cause any significant damage. The barnacle *Balanus amphitrite communis* was a serious pest being competitor for space to oyster spat on spat collectors and they were scraped with chisel. The whelks *Thais* and the crabs *Scylla serrata* and *Pagurus* sp. were the predators which occurred in small numbers.

Discussion

It has been observed by Rao (1974) that sexually ripe *Crassostrea madrasensis* are found in Vaigai Estuary at Athankarai almost throughout the year in greater or lesser percentages. The results obtained in the present work indicate that successful oyster spatfall takes place in the estuary following breeding of oysters in the period January to April and on a small scale from June to December. The main spatting season in Vaigai Estuary is January-March. Spatfall takes place when the salinity ranges between 20.96‰ and 27.0‰ and the estuary is connected with the sea. Spatfall continues to take place on a small scale in the later months when the salinity is higher. Hopkins (1931) had considered that in *Crassostrea virginica* of Galveston Bay the salinity should be about 20‰ while Gaarder (1932, 1933) and Gaarder and Bjerkan (1934) have observed that a salinity of 24.0‰ and over with 31-35‰ was optimum for successful growth of larvae of *O. edulis*. Rao (1951) has observed that when Adyar Estuary was connected with the sea and the salinity of the estuary was 27‰ and above spatfall of *C. madrasensis* takes place.

The present work shows that concrete pieces with irregular surfaces given lime coating, oyster shells given lime or cement coating and curved tiles given lime coating are suitable for collecting spat of *Crassostrea madrasensis* in Vaigai Estuary at Athankarai. Among the three kinds of spat collectors the average number of spat setting is highest on concrete pieces given lime coating and the number of spat setting on lime-coated oyster shells and curved tiles is less. The kind of spat collector which is most efficient resulting in setting of large number of spat differs in various areas. Lime-coated curved roofing tiles have been effectively used in Arcachon, Southern France (Hornell, 1910 ; Yonge, 1960). Hornell (1910) pointed out the possibilities for culturing *Ostrea* (=*Crassostrea*) *madrasensis* along the east coast of India by collection of spat by laying lime-coated tiles. Sundararaj and Devanesan achieved limited success in effecting setting of spat of *C. madrasensis* using this type of spat collector. Recently in Tuticorin Bay, Mahadevan and Nayar (1980) obtained a maximum of 97 and average of 34 spat of *C. madrasensis* per lime-coated tile. On the southwest coast, Dhulkhed and Ramamurthy (personal communication) found a maximum of 30 to 40 oyster spat setting on Mangalore tiles kept in Sambhavi Estuary, Mulki.

Varying types of spat collectors are used for oyster culture in various parts of the world. Rens of oyster shells hung from horizontal poles or rafts are used in Japan (Cahn, 1950)

and British Columbia, Canada (Quayle, 1969; 1971). Stick culture is prevalent in British Columbia and Australia (Qualye, 1969; Glude 1976). Wooden veneer rings dipped in cement slurry are used successfully for spat collection in British Columbia (Quayle, 1969). In United States of America oyster culture is largely carried out by the bottom culture method and spat are collected on shells (Bardach *et al.*, 1972). In U.S.A., with the development of hatchery method of production of spat a number of oyster hatcheries have been established which produce and supply spat to oyster farmers (Glude, 1976).

The average growth of 86.7 mm attained by *Crassostrea madrasensis* in Vaigai Estuary at Athankarai is similar to that of 85-90 mm observed by Mahadevan and Nayar (1980) in the same species in Tuticorin Bay. The maximum sizes of 100-110 mm reached by *C. madrasensis* at the end of one year at Athankarai is higher as compared to 84 mm recorded by Rao and Nayar (1956) for the species in Adyar Estuary, Madras. The American oyster *Crassostrea virginica* has been found by Ingle (1950) to grow to a size of 104 mm in seven months itself in the warm Apalachicola Bay in Florida while Menzel (1951) has reported that the *C. virginica* reaches a size of 100 mm only in nine months on Louisiana Coast. A very rapid growth to a size of 80 - 95 mm has been observed by Dhulkhed and Ramamurthy (1980) in *C. madrasensis* in Sambhavi Estuary, Mulki and the authors attributed it to higher productivity of waters on west coast of India than on the east coast. The growth of the above species of *Crassostrea* is very rapid as compared to that of the European oyster *Ostrea edulis* which grows to a maximum size of only 35 mm (Orton, 1937) at the end of one year.

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ON THE SETTING OF SPAT AND GROWTH OF THE EDIBLE OYSTER *CRASSOSTREA MADRASENSIS* (PRESTON) IN COCHIN BACKWATER

K. S. PURUSHAN, U. K. GOPALAN AND T. S. S. RAO*
Regional Centre of National Institute of Oceanography, Cochin-682 018

ABSTRACT

Results of a study on the setting of spat and growth of the edible oyster *Crassostrea madrasensis* (Preston) in Cochin Backwater from September, 1976 to August, 1977 are presented. Of the various cultches such as oyster shells, concrete panels, clay tiles, aluminium sheets, wooden planks, nylon ropes and coir ropes used, the former four were seen to be successful. Among these, clay tiles were the most efficient for spat setting with a maximum of 84 individuals on a tile (0.2 m^2 area). The spatfall commenced in January and lasted till the middle of February. The spat grew on all the cultches from January to July when the individuals on an average attained a length of 61.75 mm (maximum) on the concrete panels and 50.3 mm (minimum) on the oyster shell cultches. Differential growth rate was observed in respect of individual spat as well as in those set on different cultches. Maximum growth was registered in February-March (12.00 — 22.15 mm), when the hydrographic conditions of the backwater were stable. From April to June the growth rate gradually decreased and reached the lowest in July. Subsequently almost all the growing oysterlings died by the end of July or early August when the salinity of the backwater considerably decreased.

Heavy infestation of fouling organisms such as *Spirorbis*, *Modiolus*, *Balanus*, *Corophium*, *Ligia* and *Sphaeroma* were found on all cultches particularly on to shell cultches. High turbidity, silting and other changes arising out of increased human activities in Cochin Backwater together with menace of fouling organisms hinder the settlement, growth and culture of oysters although spat are profusely available during favourable season.

INTRODUCTION

A COMPREHENSIVE account of oyster cultivation in various parts of the world is given by Bardach *et al.* (1972) and Milne (1972). In India studies on oyster culture were initiated by Hornel (1910 a, b) in the beginning of this century and a review of the works on Indian edible oysters since then has been given by Rao (1974). Recently some information on the techniques and possibilities of edible oyster culture has been released by C.M.F.R.I. (1977). *Crassostrea madrasensis* (Preston) tops the list of commercially important edible oysters of India. In Cochin Backwater heavy encrustations of this species are found in the intertidal embankments during the post monsoon and premonsoon months and in the subtidal region they occur year round. Settlement and growth of this species as fouling organisms have been investigated by Nair (1967) and Balasubramanyan and Nair (1970). The dense populations of this species occurring in the intertidal region do not survive the monsoon owing to the adverse effects of fresh water as revealed by the seasonal studies on their production (Purushan *et al.*, 1979). In this background an attempt was made to examine setting and growth of *C. madrasensis* in the comparatively deeper portions of the Cochin Backwater.

* Present address : National Institute of Oceanography, Dona Paula-403 004, Goa.

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Materials and Methods

A raft (Pl. I A) made of bamboo poles floated by means of empty iron drums of 200 litre capacity was anchored at a depth of 5 m, near Ramanthuruth Island (Fig. 1) in Cochin Harbour area in September, 1976. For the fabrication of the raft frame, uniform sized bamboo poles were arranged in a crisscross pattern at intervals for 10 cm and fastened using manila and nylon ropes. The raft had an area of 3.5 m × 3.5 m. Various cultches like oyster shells, concrete panels, clay tiles, aluminium panels, wooden planks, nylon ropes and coir ropes were separately suspended. About 30-32 oyster shells (thoroughly cleaned and dried) were strung along a rope. About 8 shells had a surface area of 0.2 m^2 which was considered as unit area for comparison. Square panels (approximately 0.2 m^2 surface area) made of concrete, clay, aluminium sheets and wooden planks were fastened separately at 0.5 m intervals on nylon ropes to form respective cultches. Each cultch was rigged by knots on nylon ropes passing through the central holes. The nylon ropes, coir ropes, aluminium sheet and wooden plank cultches were suspended by using suitable weights to keep them in an up-

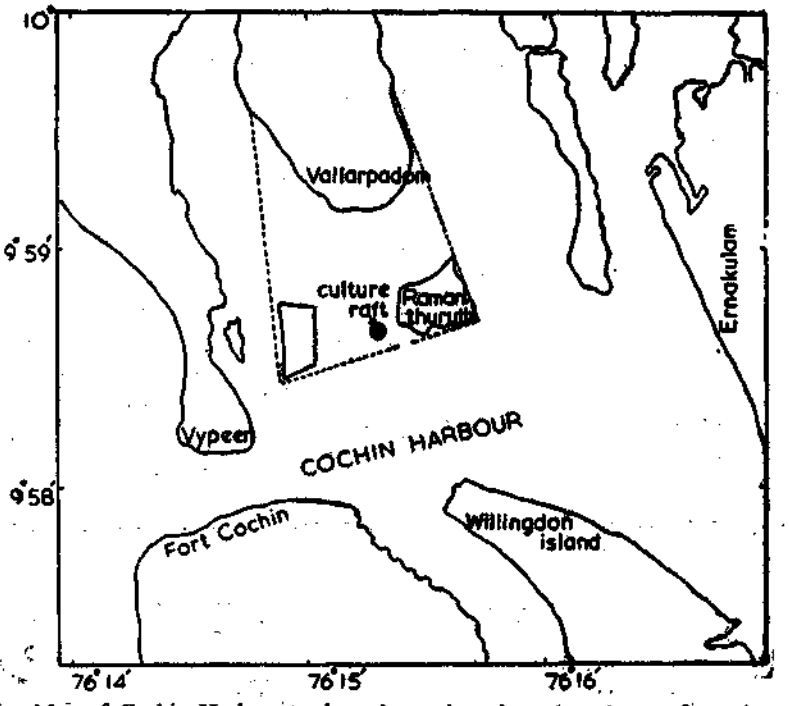

Fig. 1. Map of Cochin Harbour to show the station where the culture raft was installed.

right position. No additional weights were provided to the shell cultch, concrete panels and clay tiles. Three identical sets of cultches were used in each series and the average settlement and growth rate for various cultches has been worked out. At fortnightly intervals the cultches were examined for spatfall, growth

of spat (Pl. I B) and fouling (Pl. I C). The fouling organisms were removed each time before keeping to cultch.

Periodical measurement of spat were recorded and hydrographical parameters such as salinity and oxygen were estimated adopting standard procedures (Strickland and Parsons, 1965). Temperature, light penetration and turbidity conditions were recorded.

Results

The hydrographic conditions during the experimental period are presented in Table 1. The surface temperature was maximum in April (33.0°C) and minimum in July (27.0°C). The salinity values varied between 0.27‰ in July and 30.75‰ in March. The dissolved oxygen values fluctuated between 2.6 ml/l in April and 5.7 ml/l in June. Higher values were obtained during the monsoon period. Light penetration (secchi disc reading) values reached a maximum in February (125 cm.) During December-March the water remained relatively more clear. Thereafter the values decreased considerably and reached the minimum in July (43 cm).

The settlement and growth of spat of edible oyster *Crassostrea madrasensis*/unit area (0.2 m²) on different cultches suspended from raft during 1976-77 are presented in Table 2. From September to December, 1976 there was no spatfall observed in any of the cultches. But spatfall commenced in January and lasted for a few weeks. As shown in Table 2, the size of spat at the beginning of observations ranged between 4 and 20 mm. Though the size of spat on different cultches varied, the modal size (10 mm) was more or less common to most of the cultches. The number of spat had gone up on all cultches during February and comparatively maximum number of spat (84/0.2 m²) had set on tile cultches. A gradual decrease in their numerical abundance was observed from March onwards and only the minimum number of individuals was available during July on all the cultches.

The maximum survival of spat *i.e.*, 28.99% was observed on oyster shell cultch and that of the minimum was on clay tiles, *i.e.*, 26.1%. Thus the maximum survival values were recorded from cultches of minimum settlement and minimum survival values from cultches of maximum settlement. From the earlier stages of observation onwards the mortality of spat decreased with increasing depths and in all cases the maximum survivors were present on all panels close to the bottom. Irrespective of the cultches, very high mortality (71.1-73.9%)

Table 1. *Hydrographic parameters recorded in Cochin Backwater where oyster culture raft was installed during* 1976-77

Months			Temperature (°C)	Salinity (‰)	Oxygen (ml/l)	Secchi disc reading (cm)
1976	September	..	28.3	4.02	4.1	55
	October	..	29.5	11.38	5.2	78
	November	..	29.7	5.25	4.5	80
	December	..	30.1	19.40	5.6	111
1977	January	..	30.0	28.46	4.3	100
	February	..	30.2	28.28	4.8	125
	March	..	32.0	30.75	4.6	100
	April	..	33.0	29.69	2.6	60
	May	..	30.5	22.50	5.2	52
	June	..	27.5	1.10	5.7	45
	July	..	27.0	0.27	5.3	43
	August	..	28.0	1.29	5.4	51

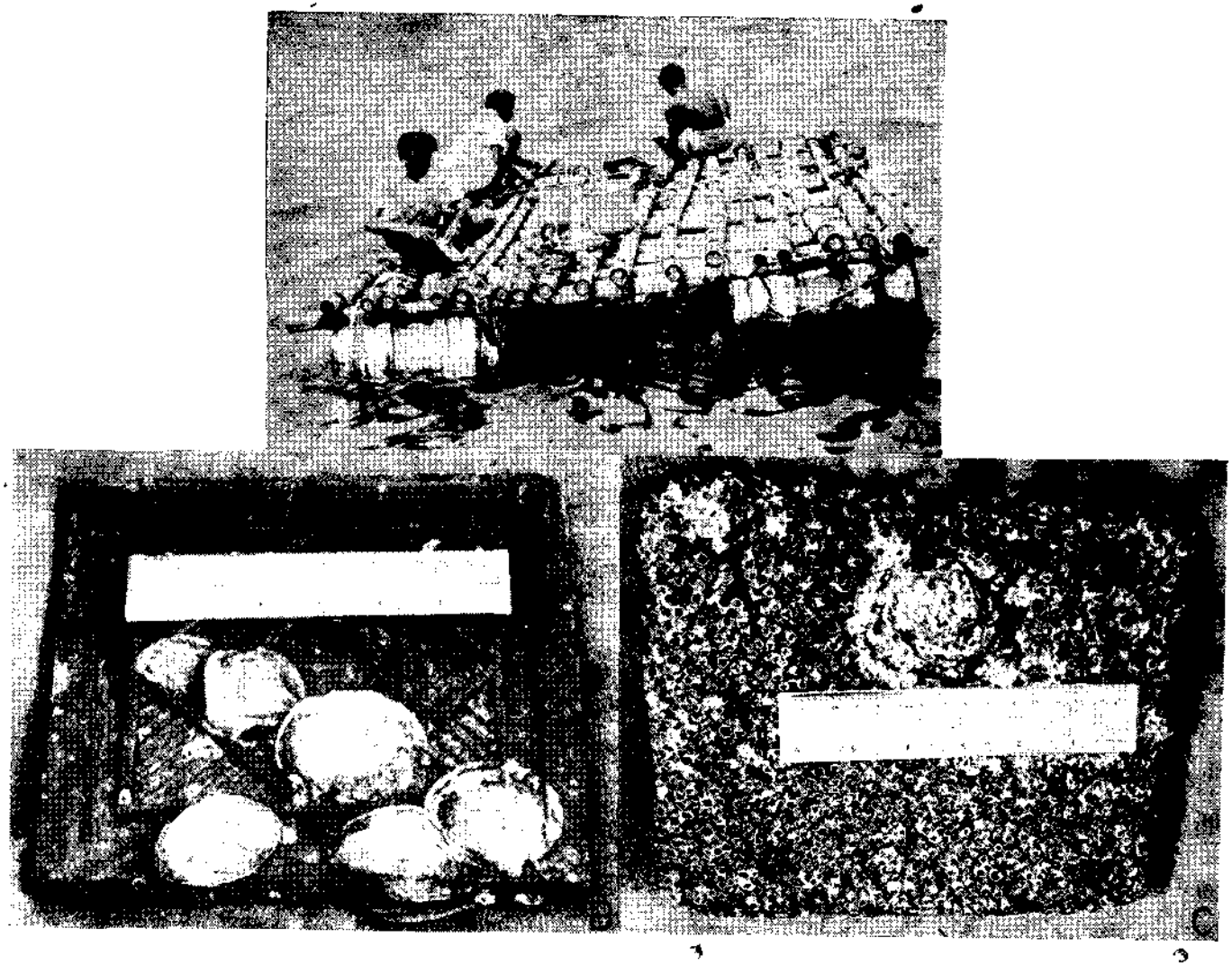

PLATE I A. Floating raft used for suspending cultch ; B. Growing oyster spat in the tile cultch and C. A growing spat surrounded by fouling organisms.

TABLE 2. *Settlement and growth in size of spat of edible oyster* Crassostrea madrasensis, *unit area* (0.2 m²) *on different cultches suspended from raft during* 1976-77

	Months	No. of days passed	Total No.	SIZE Minimum (mm)	SIZE Maximum (mm)	Mean (mm)	Modal (mm)	Total No.	SIZE Minimum (mm)	SIZE Maximum (mm)	Mean (mm)	Modal mm
				Oyster Shell					*Clay Tile*			
1976	Sept-Dec.	..		No spat fall					No spat fall			
1977	January	.. 0	16	4.0	13.0	8.5	10.0	49	5.0	15.0	7.86	10.0
	February	.. 26	38	12.0	29.0	26.28	25.0	84	11.0	34.0	24.6	30..0
	March	.. 69	32	25.0	45.0	42.5	40.0	51	31.0	48.0	42.8	45..0
	April	.. 90	30	34.0	48.0	46.5	45.0	45	33.0	53.0	46.77	50.0
	May	.. 120	27	36.0	53.0	48.3	50.0	39	35.0	57.0	49.2	500
	June	.. 153	20	37.0	54.0	50.2	50.0	31	37.0	59.0	52.8	550
	July	.. 169	11	37.0	55.0	50.3	50.0	22	39.0	62.0	54.0	55.0
	August	..		All dead					All dead			
				Concrete					*Aluminium*			
1976	Sept-Dec.	..		No spat fall					No spat fall			
1977	January	.. 0	41	10.0	20.0	13.42	15.0	36	4.0	14.0	8.6	10.0
	February	.. 26	60	28.0	38.0	35.57	35.0	49	14.0	32.0	28.0	25.0
	March	.. 69	35	40.0	58.0	50.6	50.0	34	22.0	43.0	40.0	35.0
	April	.. 90	28	48.0	65.0	55.8	55.0	26	33.0	45.0	44.5	40.0
	May	.. 120	25	53.0	67.0	59.9	55.0	21	36.0	52.0	48.0	45.0
	June	.. 153	21	56.0	68.0	61.75	60.0	18	38.0	56.0	50.75	50.0
	July	.. 169	17	57.0	68.0	61.75	60.0	14	39.60	57.0	51.0	55.0
	August	..		All dead					All dead			

was observed in July. A conspicuous feature noticed during the experimental period was that almost all the spat were seen dead by the end of July or early August.

The growth of individuals was followed for a period of 169 days when the individuals on an average attained a length of 61.75 mm (maximum) on concrete panels and 50.3 mm (minimum) on oyster shell cultches. The size of spats which was in the range of 10-15 mm on different cultches during January has shifted to the range of 50-60 mm at the end of observations in July.

The periodical growth increments presented in Table 3 reveal that in general the growth was considerably faster during January-March on all the four cultches with the maximum (22.15 mm) on the concrete panel and the minimum (12.0 mm) on the aluminium panel. From April to June the growth slackened (1.8-5.2 mm) and in July very little or practically no growth (0-1.2 mm) was observed. The average monthly growth increments were 6.97, 7.69, 7.97 and 7.00 respectively for shell, tile, concrete and aluminium cultches. Among the four cultches tile was found the best for spatfall, concrete for faster growth and shell for maximum survival.

TABLE 3. *Periodical growth increments (in mm) of spat of* Crassostrea madrasensis *on different cultches during* 1977

Period	Oyster shell	Clay tile	Concrete	Alumi-nium
Jan-Feb. ..	17.78	17.74	22.15	19.40
Feb-Mar. ..	15.22	18.20	15.13	12.00
Mar-Apr. ..	4.00	3.97	5.20	4.50
Apr-May ..	1.80	2.57	4.10	3.50
May-June ..	1.90	3.60	1.85	2.75
June-July ..	0.10	1.20	0.00	0.25
Seasonal average ..	6.97	7.69	7.97	7.00

Fouling organisms began settling on the cultches within a short period after they were kept. Among the foulers, algae Belonging to *Enteromorpha* sp. and *Polysiphonia* sp. were more common on the top-most cultches. Animal foulers showed particular preference for shell cultches. Many hydroids and bryozoans were found settled on all the cultches. Tubiculous polychaetes such as *Polydora* sp. and *Spirorbis* sp. were abundant on the shell cultches. Among the crustaceans, isopods *Sphaeroma* sp., *Ligia* sp. and *Cirolina* sp. occurred in large numbers particularly in January. The genus *Balanus* occurred predominantly on all the cultches throughout the year. The amphipod *Corophium* sp. abundantly occurred on the wooden planks. Among crabs *Viaderiana* sp. was common on all cultches. The mussel *Modiolus* sp. was seen attached in large numbers during November-February.

DISCUSSION

Hornell (1910 a), Paul (1942), Rao (1951) and Rao and Nayar (1956) studied the growth of oysters in the natural environment on the east coast of India. Nair (1967) and Balasubramanyan and Nair (1970) reported the growth of oysters as foulers in Cochin Backwater. In the present work the mean and maximum growth attained by edible oysters were 61.75 mm and 68.0 mm within a period of 169 days. This seems to be better than that reported by Rao and Nayar (1956) in the case of *Crassostrea madrasensis* which attained a mean length of 61.0 mm after 180 days in Adyar Estuary. As revealed by C.M.F.R.I. (1977), the same species attained marketable size (100-110 mm) at Karapad farm in less than 12 months. But in Madras Harbour, Paul (1942) recorded a mean growth of 66 mm for *C. madrasensis* after 243 days which shows that growth in this species was better on rearing in trays in off-bottom culture. The relatively faster growth noticed in *C. madrasensis* in Cochin Backwater indicates that an average size of 60-62 mm (average

meat weight 5 gm, *i.e.* 10% of total weight) is possible within a period of 5-5½ months.

Spat of *C. madrasensis* settled in early January recorded vigorous growth till March. A general decline in April-June was followed by practically very little or no growth in July. Balasubramanyan and Nair (1970) observed in the same species at Cochin varying differences of growth in their shells in every month. As reported by Dame (1972) in the case of *C. virginica* of different sizes, the instantaneous growth rate of *C. madrasensis* at Cochin also showed a reduction with increasing size. Rao and Nayar (1956) have attributed widely fluctuating environmental conditions to the differences in the rate of growth of this species in different periods in Adyar Estuary.

The cessation of growth of *C. madrasensis* during July and subsequent death by August can be ascribed to steep fall of salinity to 0.27‰ and continuous low salinity conditions. There is significant variation in the salinity recorded during the favourable season of growth (28.46-30.75‰ and that prevailed during the unfavourable season (0.27 - 1.29‰. Rao and Nayar (1956) noticed that prolonged immersion of spat and yearlings under very high or very low salinity condition (0.3-41‰) has an adverse effect on the oyster growth rate in Adyar Estuary. Butler (1949) experimentally proved the detrimental effects of salinity on *C. virginica*. Chanley (1958) found juvenile *C. virginica* surviving salinities as low as 5‰, but failed to grow below 5‰. In the case of *C. gigas*, Fujiya (1970) observed that formation of larval shell is retarded in suboptimal salinities. Nair (1967) and Balasubramanyan and Nair (1970) had also noticed the adverse effect of low salinity on the settlement and growth of oysters and other fouling organisms in Cochin Backwater.

It has been observed that the longest survived spat were on the bottom-most cultches. This is an indication of the possibility for the better survival of oysters in still deeper waters where a distinct stratification of saline water prevails even during the monsoon due to the penetration of saline water (Haridas *et al.*, 1973). This reveals the secret of survival of a subtidal population of *C. madrasensis* which in effect is the natural seed stock of the species in this region. This also indicates that salinity is one of the important factors influences the survival of edible oysters in the region.

In the present study the lesser fluctuation noticed in the dissolved oxygen content of the area did not seem to have any appreciable influence on the oysters when compared to salinity. The temperature fluctuated from 27-30°C during monsoon and 30.2-33.0°C during premonsoon.

The adverse effects of fouling organisms on the growth of oysters are quite large. The high settlement of *Balanus* sp. and the incidence of isopods like *Sphaeroma* sp. and *Ligia* sp. results in competition with oysters for space and food. The ill-effects of metallic pollution as revealed by Sankaranarayanan *et al.* (1978) and other unfavourable factors like increased rates of silting, turbidity and organic pollution accelerated by human interference as reported by Purushan *et al.* (1979) negatively influence the growth and survival of oysters in Cochin Backwater.

The present experiment indicates that commercial oyster culture using floating rafts is not viable in Cochin Backwater on an year round basis, as the oysters could not survive the monsoonal influx of freshwater. Even during the period of fast growth, the oysters are largely affected by the menace from fouling organisms coupled with man made environmental alterations. These factors would have played predominant role in the gradual destruction of the once productive 'oyster beds' which sustained a fishery two decades ago in the Cochin Harbour region.

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SOME ASPECTS OF EXPERIMENTAL CULTURE OF THE OYSTER *CRASSOSTREA MADRASENSIS* (PRESTON)

M. MOHAN JOSEPH AND SHANTHA JOSEPH

University of Agricultural Sciences, College of Fisheries, Mangalore-575 002, India

ABSTRACT

Settlement and rate of growth of the oyster *Crassostrea madrasensis* (Preston) were studied at the Mulki Estuary, Dakshina Kannada. The breeding season extends from October to May. Peak Settlement of spat takes place during November-December and March-April. Of the several cultch materials tried, oyster shell, used automobile tyres, rigid PVC, lime-coated tiles and asbestos were found to be suitable. Cultch smeared with crude extracts of oyster tissue supported more spat per unit area than the untreated panels. The spat grew initially at the rate of 2-3 cm per month. Spat transferred to suspended wire bag grew faster than the feral ones. The oysters attained ≥7.0 cm shell-height in about 7 months. The size at first maturity was 12-14 mm for males and 24-26 mm for females. Study of the condition and edibility indices showed that the best season for harvest is May-September.

INTRODUCTION

BECAUSE of their sessile habit and comparatively low position in the food web, the edible oysters form one of the best suited marine animals for large-scale culture. The history of oyster culture dates back to the times of the ancient Romans. However, the high degree of technological proficiency attained in the fields of hybridization, seed production, field culture and disease control is the result of meticulous work carried out during the past few decades. At present several species of oysters belonging to the genera *Ostrea* and *Crassostrea* are profitably mass cultured along the coasts of maritime countries of the world (Korringa, 1976 a, b). Although many species of edible oysters occur in the Indian waters and at least two of these *viz.*, *Crassostrea madrasensis* and *C. gryphoides* have great potential as culturable species, no extensive farming is practised in India due to the very limited demand. In the Kelwa Backwaters near Bombay and Ennur near Madras, small sized oysters are collected and maintained in the field till they attain marketable size (Jones, 1970). Even now, work on the farming of Indian edible oysters is limited to experimental scale and is restricted to a few research institutions and universities. The biology of the species involved is fairly well known and there is considerable scope for undertaking culture of oysters in the backwaters and estuaries of both coasts of India.

The present paper incorporates the findings of a study on the experimental culture of *C. madrasensis* carried out in the Mulki Estuary during 1976-79.

The authors are grateful to Prof. H. P. C. Shetty, Director of Instruction, College of Fisheries for the encouragement and facilities.

CHARACTERISTICS OF HABITAT

The Mulki Estuary (Lat. 13°5'N and Long. 74°46'E) is formed by the confluence of the Shambhavi river and the Pavanje rivers near Mulki town, in the district of Dakshina Kannada, Karnataka. The estuary has a waterspread area of *Ca.* 5 km² and an average

depth of 3 m. The tidal range is about 2 m. The bottom consists mainly of a mixture of silt and sand. The southwest monsoon contributes significantly to the total rainfall in this region. During this season the Dakshina Kannada Coast receives about 3000 mm of total rainfall. The ambient water temperature fluctuates from 25.5° (September) to 32.7°C (March) during the course of one year. During the southeast monsoon months (June-September) the salinity of the water decreases from about 36.0‰ to very low levels (< 1.0‰) while turbidity increases up to 95.5 ppm. The estuarine water does not show wide fluctuations in dissolved oxygen content and pH.

Breeding Season

Our knowledge of the biology of *C. madrasensis* from the west coast of India is meagre. Balasubramanyan and Nair (1970) observed that in the Port of Cochin, the breeding of this species is seasonal, restricted to the periods of high salinity. Menon *et al.* (1977) recognised two peaks for the settlement of oyster larvae in the Netravathi-Gurupur Estuary, *viz.*, March and May. Rao and Menon (1978) reported heavy settlement of *C. madrasensis* spat in the above estuary during March-June. Joseph (1979) observed that in the Mulki Estuary the spawning season is rather prolonged, extending from October to May, with two peaks: December-January and April-May.

Spatfall

The spatfall in the Mulki Estuary was monitored from the settlement data gathered from cultch materials suspended in the estuary. Shells of the green mussel (*Perna viridis*), oyster (*C. madrasensis*), uncoated roof tiles, lime-coated roof tiles, asbestos plates, asbestos plates coated with crude extract of oyster meat, coconut shells, used automobile tyre pieces and rigid PVC pieces were the cultch materials tried in the present study (Pl. I). Of these, shells of *Perna viridis*, uncoated tiles and coconut shells were found to be not suitable, as settlement of spat was negligible. Lime-coated roof tiles were good spat collectors, but the loss of spat due to pealing up of the coating was considerably high, especially during the initial stages after settlement. Both untreated and treated asbestos panels attracted appreciable number of larvae resulting in heavy settlement. Asbestos panels smeared with an extract of oyster meat (prepared by blending the meat of ten mature oysters in 500 ml of sea water) and dried in the sun for two days appeared to support higher spat density than untreated panels. Settlement of spat was poor on freshly laid panels prepared from used automobile tyre. However, moderate to high settlement resulted when these panels were kept after being subjected to about ten days of leaching in sea water. Rigid PVC panels were also found suitable as the fouling intensity on them by barnacles and polychaetes was considerably low. However, the spat of oysters fall off the substratum at the slightest pressure. Compared to all the above materials tried, the oyster shells (upper valves of *C. madrasensis*) appeared to be the best spat collector because of their easy availability, convenient size, good spat density, neglegible market value and reusability (Pl. II). An interesting feature observed during the present study was the effect of spacing of the cultch materials on the settlement density. When cultch was strung close together with very litte (< 2 cm) space in between, the settlement was high. However, when the same cultch materials were suspended at intervals greater than 4-5 cm, the intensity of space settlement was considerably reduced. Polyethylene tubings cut into the required sizes were used as spacers. Probably, as Korringa (1970) opined, prolonged periods of low current velocities are required for good settlement.

Two periods of peak spat settlement were noticed in the Mulki Estuary, the first from November to January and the second from

PLATE I. Different types of culch materials used for collecting spat of *C. madrasensis* : A. Used automobile tyre pieces, B. Oyster shell and C. Lime-coated tiles.

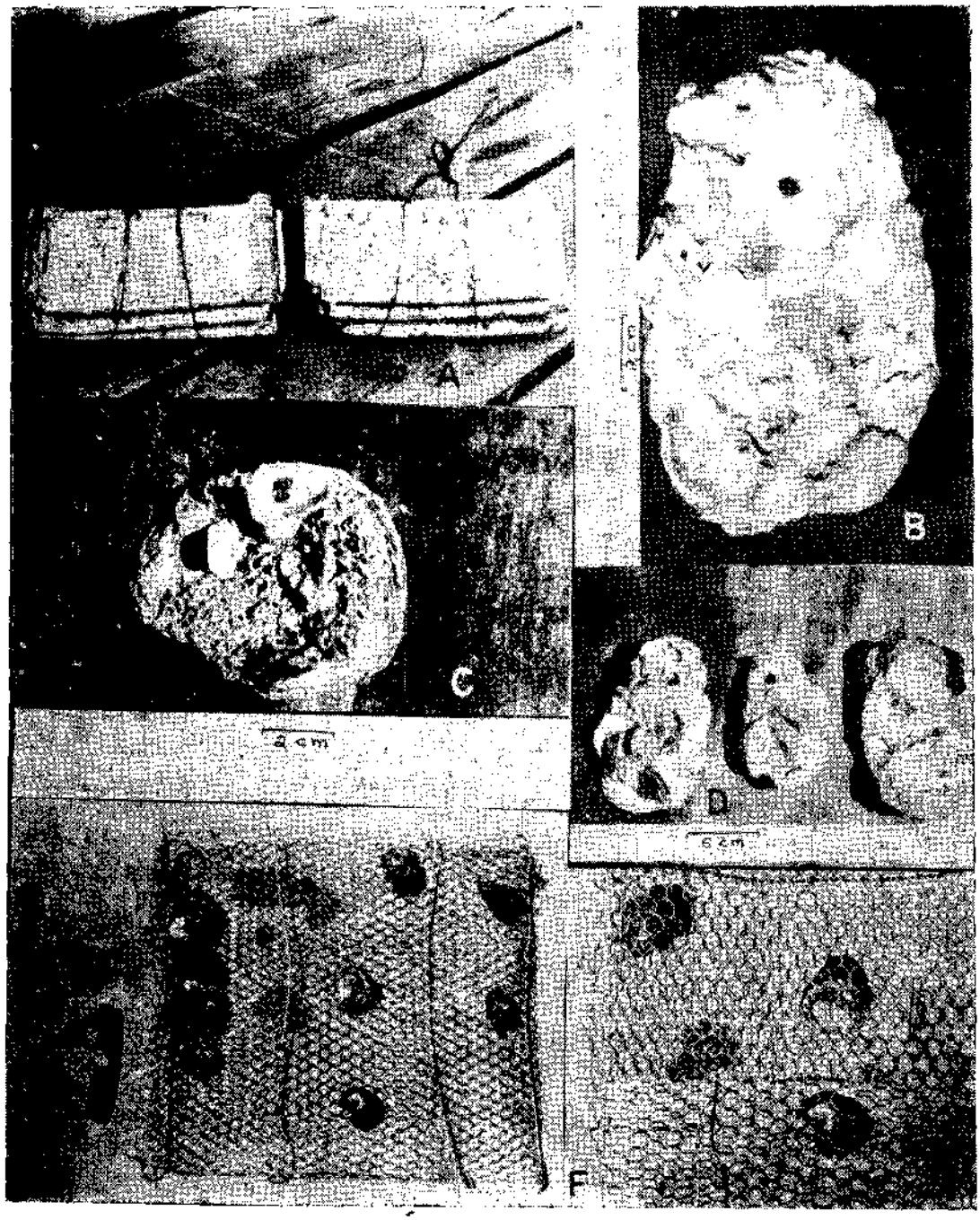

PLATE II. Spat collectors and cultured oysters : A. Lime-coated tiles with oyster spat, B. Oyster shell with heavily settled spat, C. A spat collector heavily fouled by tube-dwelling polychaetes, D. Oyster shell with growing spat and oysterling. Arrows indicate oysters of five months' growth, E. A cage used for suspending culchless spat in the estuary and F. Culchless oysters grown in cages. Age about 3 months.

March to May. There was no settlement of spat during the period extending from August to September. However, the spat of the rock oyster *C. cucullata* settles heavily almost throughout the year in the Mulki Estuary except during July and August and probably forms the toughest competitor to *C. madrasensis* for settlement space, especially at those regions of the estuary where depth is comparatively less.

Growth of spat

When the spat was about 2-3 cm in size (shell-height), the oyster shells used as cultch were removed and polyethylene spacers were introduced in between, so that the interval between the shell pieces was about 6-7 cm. A few of the spat were removed from automobile tyre and rigid PVC cultch materials, placed inside wire cages (size 40 × 40 cm) made out of galvanised iron chicken-wiremesh (Pl. II E) and suspended in the estuary. Rate of growth in length (shell-height) was followed by making periodical measurements with the aid of a vernier calipers. The mean shell-heights from the time of settlement were as follows: 2.10 cm at 1 month; 3.5 cm at 2 months; 4.5 cm at 3 months; 5.5. cm at 5 months; 6.2 cm at 6 months and 7.2 cm at 7 months. From the foregoing it appears that the rate of growth of *C. madrasensis* in the Mulki Estuary is considerably faster than that reported for the same species from the Madras Coast by Hornell (1910), Paul (1942) and Rao and Nayar (1956) and for *C. gryphoides* from the Kelwa Backwaters by Durve and Bal (1962). Joseph (1979) has reported that the feral population of *C. madrasensis* inhabiting the Mulki Estuary grow to a marketable size of *Ca.* 7 cm in 9 months time (mean sizes: 4.70 cm at 3 months; 6.33 cm at 6 months; 7.66 cm at 9 months). In the Netravathi-Gurupur Estuary the maximum recorded size of *C. madrasensis* on glass panels immersed for 20 weeks duration is 7.6 cm shell-height (R. J. Katti, unpublished data). All these evidences suggest that *C. madrasensis* can be grown to marketable size in the Mulki Estuary or similar estuaries of this region within a maximum period of 7 months from the time of settlement. This unusually high rate of growth might be the result of the abundant food supply in the estuary, coupled with other favourable environmental factors experienced during the post and pre-monsson periods. Thus, if cultch is laid in October and quality spat collected, the oysters will be ready for harvest by next April or latest by May before the commencement of the southwest monsoon (Fig. 3).

Condition

Studies conducted on the variations in the condition and edibility indices have shown that in the Mulki Estuary the oysters are in their best condition from late May to September. Condition indices as high as 63.3 have been recorded during this period (Joseph, 1979). Further, during this period, the oysters are in a sexually indeterminate stage and have large quantities of carbohydrates (for example, 68.93% dry weight in the indeterminate gonad) (Joseph, 1979).

Harvesting

Analyses of the data on the annual gonadial cycle, variations in the length-weight relationships, condition and edibility indices and biochemical composition of feral oysters (Joseph, 1979) suggest that the ideal time for harvest is the period extending from late May to September. Along the Dakshina Kannada Coast, the southwest monsoon rains commence by the second fortnight of May. However, intense rainfall is experienced from June onwards only, resulting in heavy flooding of the estuaries. Hence, the harvesting operations can be completed by the end of May. This will result not only in the production of oysters with higher meat content and nutritive value, but also in preventing loss of culture rafts and poles.

Associates, parasites and mortality

A large number of fouling organisms settle on spat collectors and cultured oysters. The most numerous of these are the barnacles (*Balanus amphitrite* var. *variegatus*, *B. amphitrite* var. *communis*, *B. amphitrite* var. *denticulata* and *B. amphitrite* var. *cocgiensis*), tube-dwelling polychaetes (*Hydroides norwegica*, *Pomatoceros triquetor*, *Spirorbis* sp. and *Polydora ciliata*) and the spat of the rock oyster (*C. cucullata* = *S. tuberculata*). Heavy fouling by barnacles and polychaetes results in poor settlement of oyster larvae. Further, fouling impairs the growth of already settled spat. This problem is important as the breeding season of most of the fouling organisms of this region coincides with that of *C. madrasensis*, resulting in intense competition for settlement space (Menon *et al.*, 1977). Further studies are required to overcome the problem created by such heavy fouling on spat collectors. *Polydora cilata* causes extensive mud blisters on the shells of *C. madrasensis* in the Mulki Estuary (Stephen, 1978). The larval trematode *Bucephalus* sp. also infests oysters, leading to parasitic castration of the host (Joseph, 1978). However, both these species do not seem to pose any serious threat to the cultured oysters which live in the estuary only for about 7 months. During the southwest monsoon, large scale natural mortality of oysters has been noticed in the feral population. This probably is the result of the unfavourable environmental conditions especially very low salinity and high turbidity existing in the estuary.

Conclusion

One of the essential prerequisites for successful culture of oysters is a thorough knowledge of aspects of biology such as breeding season, time and area of spatfall, substrate preferences of spat, rate of growth and eco-physiological requirements of oysters for survival, growth and fattening. The present study has conclusively proved that the Mulki Estuary is an ideal locality for spat collection and growth of *C. madrasensis*. The spat is plentiful in the estuary all through the breeding season of the oyster. The rate of growth of spat and juveniles reported in the present study is perhaps the highest reported from the Indian coasts.

Considering the very poor demand for oysters in the Indian market, it is necessary to popularise the use of oyster meat as an item of protein and carbohydrate-rich sea food. Further, exploring the possibilities of preparation of frozen or canned oyster meat on a large scale may prove to be highly rewarding. The market value of whole oysters along the Dakshina Kannada Coast ranges from Rs. 7 to Rs. 8 per hundred. Small quantities of shucked oysters are bought by some local freezing factories at Rs. 10 per hundred. Large scale farming might prove to be a lucrative business as the demand for raw or processed oyster meat increases.

A scheme of farming operations suitable for the Dakshina Kannada region is given below. The farming operations are divided into three phases of 15 days duration each. The first phase is spat collection. Oyster shells (upper valves of *C. madrasensis*) are suspended at intervals of < 2 cm on wire rope in areas where the current velocity is moderate. This operation must be completed in October. The second phase involves relaying of the settled spat. Here the oyster shells are removed from the strings and the oyster spat adhering to the shell cultch are scraped and reared in trays provided with nylon netting kept on racks set up in the shallow parts of the estuary. Low current velocities are conducive to fast growth of oysters and hence care should be taken to avoid areas of rapid currents while erecting the stakes. During December-January, fouling organisms have to be removed and if the oysters are over-crowded, they

should be separated. The third phase is harvesting. This should be commenced by the end of May and completed before the onset of heavy rains. The farming technique outlined here is quite simple so that even an unskilled fisherman will be able to follow. This simple and less expensive method might be the most suitable one for Indian conditions.

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CULTURE EXPERIMENTS ON THE EDIBLE OYSTER *CRASSOSTERA MADRASENSIS* IN THE BHEEMUNIPATNAM BACKWATER

S. REUBEN*, T. APPA RAO AND P. E. S. MANICKAM
Central Marine Fisheries Research Institute, Cochin-682 018, India

ABSTRACT

Studies on the possibilities of culture of edible oyster *Crassostrea madrasensis* in the Bheemunipatnam Backwater were conducted during 1977-79. Spat collection experiments using different types of spat collectors showed that empty oyster shells and close meshed plastic baskets were most efficient. Setting of spat on the spat collectors kept near the bottom was more than on those kept suspended off bottom. Spatfall was observed throughout the year with peaks in March and October. The oyster attained a size of about 8 cm during the first year. Low salinity during monsoon months appeared to retard growth.

INTRODUCTION

THE CULTURE of edible oyster *Crassostrea madrasensis* has assumed importance in India in the recent years. Hornell (1910) and Paul (1942) gave preliminary accounts of the growth of spat of *Ostrea* (*Crassostrea*) *madrasensis* Preston, while Rao and Nayar (1956) dealt in detail with the growth of the same from the Madras Coast. However, no work has been done on this oyster from the north-east coast of India. Observations on the spat collection and growth of this species in the Bheemuni patnam Backwater are presented in this paper.

The authors are grateful to Dr. E. G. Silas, Director, Central Marine Fisheries Research Institute, Cochin for his constant encouragement during the course of this work.

MATERIAL AND METHODS

The settlement and rate of growth of the spat of *C. madrasensis* were studied from bimonthly samples of spat collected from an experimental rack, set up near the natural oyster bed in Bheemunipatnam Backwater at Nagamayya-palem during 1977-79. The bed is shallow and is situated 3 km north of the bar mouth and has good tidal flow. Different spat collectors, *viz.*, tiles, asbestos sheets, coir ropes, wooden poles, iron meshed cages and oyster shells were used. Plastic baskets with close mesh were also used during 1979. The spat were removed from the collectors when they attained a size of 30-40 mm and reared in iron meshed cages. In order to eliminate any bias in the estimation of growth by using monthly modal values, mean height for every month were also calculated and the mean growth-curve drawn. Data were collected on temperature, salinity and pH to see if they have any bearing on the growth and spawning of the oysters.

SETTING OF OYSTER SPAT

The results of the present study reveal that although the setting of spat has been observed almost throughout the year, two well marked spawning periods, *i.e.*, February-June and September-December are present. March and October appeared to be months of peak setting.

* Present address : Waltair Research Centre, Central Marine Fisheries Research Institute, Visakhapatnam-530 003.

Of all the spat collectors set up near the experimental rack, oyster shells, iron cages with close mesh had good setting while close meshed plastic baskets showed very intensive setting. Further, the spat collectors kept on or near the bottom had more spat than those kept off bottom. During the successful seasons, spat setting on oyster shells at the rate of 4 to 20 numbers per 10 cm² in 1977 and 1978 and at the rate of 15 to 50 numbers per 10 cm² in 1979 was observed. Spat setting on the close meshed plastic baskets during October-December 1979 season was unprecedented and so intensive (100-120 numbers per 10 cm²) that the entire area of the basket, especially the inside, was filled with spat; sometimes a second layer of setting over the first was seen.

in 22 months if length increments in terms of modal values and mean monthly values respectively are taken into consideration. In October 1979 a modal size was seen at 12 mm which shifted to 27 mm in November and 47 mm in December 1979.

The minimum, maximum, mean and modal heights observed during 1978-79 given in Fig. 1. The growth values at the end of February, March, April, May and June 1978 as read from the mean curve (Fig. 1) are 12.7 mm, 23.9 mm 32.0 mm, 38.5 mm and 44.0 mm. The monthly rate of increase in the size of the juvenile oysters was highest (12.7 mm) in February and later values gradually declined to 5.5 mm in June 1978. Apart from slight increase in July

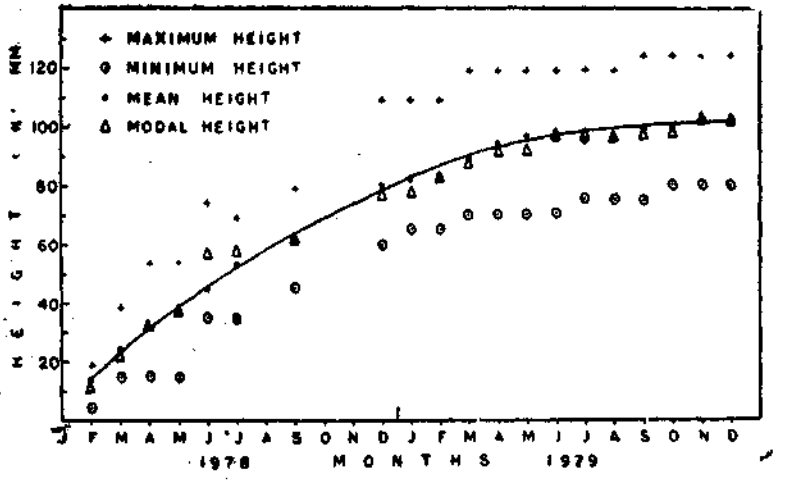

Fig. 1. The growth curve of *Crassostrea madrasensis* of 1978 year class.

Growth of spat

Spat, with a modal size of 12 mm were seen in February 1977. This mode could be traced to the mode at 62 mm in October 1977.

The growth of the spat of modal size of 12 mm in February 1978 could be traced till December 1979. The brood would have originated in January 1978 and a growth of 77.0 mm and 81.8 mm in 12 months and 102 mm and 101.5 mm

1978, the values remained either almost steady or declined until December 1979.

Temperature, salinity and pH of backwater

The temperature of backwater fluctuated from 25.0°C in January to 32.2°C in May during 1977 while it ranged from 22.0°C in October to 33.0°C in May during 1978 (Fig. 2). Water temperature was low from October to December 1978, and high in other months.

However, in 1979 temperature varied between 24.0°C and 28.0°C.

There were wide fluctuations in salinity due to incursion of flood waters in 1977 (8.8‰—32.8‰) and 1978 (7.4‰-34.4‰) with higher values during the first six or seven months followed by lower values during the rest of the year. However, in 1979 salinity values did not show much fluctuation and they ranged from 25.5‰ to 34.9‰.

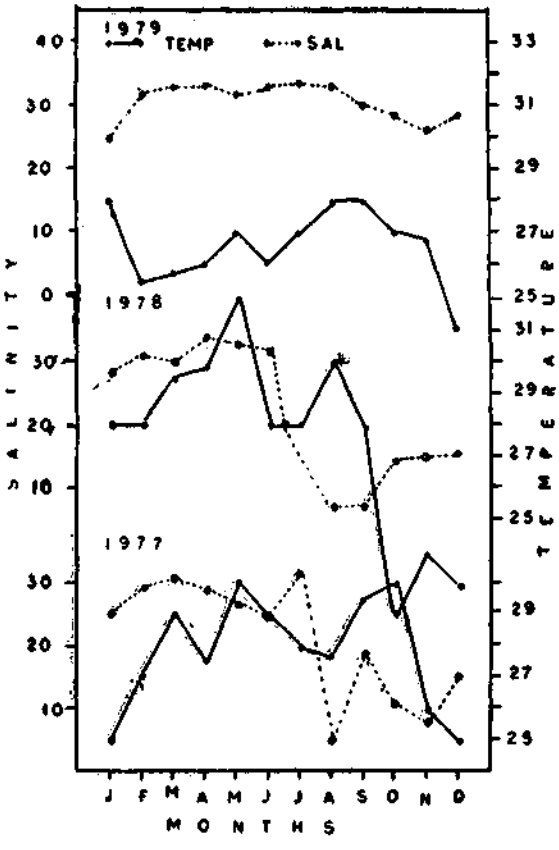

Fig. 2. Monthly variations in the mean values of temperature and salinity in the Bheemunipatnam Backwater during 1977-79.

The range in variation in pH values of the waters near the experimental rack was 8.0—8.2 in 1977 and 8.0—8.1 in 1978. The pH values ranged from 7.6 to 8.2 during 1979.

Age at which the Oysters Reach Marketable Size

It is known, from earlier observations (Rao and Nayar, 1956) that *C. madrasensis* in Madras Backwater, attained a marketable size of 84 mm (maximum size) in one year. The present study reveals that *C. madrasensis* in Bheemunipatnam Backwaters reaches maximum and mean sizes of 110 mm and 81.8 mm at the end of twelve months.

Discussion

Hornell (1910) showed that in about 1½ months the spat of the *O.*(=*C.*) *madrasensis* attain a size of 27 mm. During the present culture experiments the spat of early 1978 brood exhibit a maximum height of 34-39 mm and a mean height of 21.6-23.9 mm in two months. The October 1979 brood however, registered a maximum growth of 47 mm and a mean height of 30.7 mm in two months. The significantly better growth of the October brood coincided with the presence of comparatively clean water devoid of barnacles and other encrustations and better salinity conditions ranging from 26.6 - 29.0‰ as opposed to 8.8 - 15.6‰ in 1977 and 14.7 - 15.8‰ in 1978. Rao and Nayar 1956 observed maximum height of 61 mm in 6 months and 84 mm in 12 months and 109 mm in 17 months. The present investigations showed maximum height of 70 mm, 110 mm and 119 mm at the end of 6, 12 and 17 months respectively, showing higher growth rate.

Rao and Nayar (1956) observed that the growth of the oyster is retarded to some extent under constant high salinity conditions and is slackened by constant low salinity. Durve and Bal (1962) stated that in *C. gryphoides* of the west coast there is rapid rate of growth when the salinity is moderate, retardation of growth during the period of constant high salinity, cessation of growth during the period

of low salinity. During the present investigations it was noticed that the growth were usually affected whenever the salinity values were high *i.e.*, in April-June 1978 and June-August 1979.

The present investigations also show that the adult oyster exhibits remarkable tolerance to lower values of salinity. However, mortality of oysters was noted whenever muddy water flows persistently in the bed area for a week or more. This may be due to clogging of the gills. Further it was noted that oyster spat of less than 20 mm are highly susceptible to lower values of salinity. There were instances of total mortality of the spat when values of salinity come down usually during October and November.

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EXPERIMENTS ON EDIBLE OYSTER SPAT COLLECTION AT TUTICORIN

R. THANGAVELU* AND N. SUNDARAM
Central Marine Fisheries Research Institute, Cochin-682 018

ABSTRACT

Standardizing the technique of spat collection is an important aspect in oyster farming. Although several established methods are in vogue in different countries, appropriate method of spat collection has to be developed to suit the local conditions taking into consideration the availability and cost of the materials employed.

This paper gives details and results of experiments conducted at Tuticorin with different types of spat collectors. Lime-coated tiles proved to be the most effective for spat collection while the use of corrugated asbestos sheets, oyster and mussel shells also gave satisfactory results.

INTRODUCTION

EXPERIMENTS were conducted for procurement of oyster spat at Tuticorin Bay, wherein a natural bed is also found nearby. Various type of spat collectors such as lime-coated tiles, cement-coated tiles, edible oyster shells, mussel shells, corrugated asbestos sheets, coconut shells, bamboo mats and palmyra lattices were used during 1977 to 1979. Among hese, lime-coated tiles, oyster and mussel shell, and corrugated asbestos sheets gave very satisfactory results thereby favouring employment of the same for large scale collection of oyster spat. On the basis of the above experiments, seasonal variation of spatfall and its intensity, appropriate period for laying the tiles to achieve maximum results were determined, apart from standardizing an effective system for the large scale collection of spat.

The authors are thankful to Dr. E. G. Silas, Director of CMFRI for his kind encouragement. Thanks are also due to S/Shri. K. Nagappan Nayar and S. Mahadevan for going through the manuscript and giving valuable suggestions for improvement.

EXPERIMENTS

The results of the experiments in terms of intensity of spat settlement in various months in different types of collectors, season of maximum spat attachment and the comparative efficiency of spat collectors are detailed below.

Experiments with lime-coated tiles

In order to find out the most efficient method of spat collection, several methods were tried. Although the method of collection of oyster spat by using lime-coated tiles had been successfully followed in some of the oyster farming European countries, it was felt that other methods also might yield equally good results. Therefore during these three years attempts were made to test the usefulness of spat collection by employing oyster shells, mussel shells, corrugated asbestos sheets, cement-coated tiles, coconut shells wooden lattices and bamboo mats in addition to lime-coated tiles (Table 1). Whereas the lime-coated tiles were set up during all the three years ; the other spat collectors were tried in different periods.

* Present address : Tuticorin Research Centre of the Central Marine Fisheries Research Institute, Tuticorin, Tamil Nadu.

During 1977 and upto the end of September 1978, all the experiments were confined to the area of natural bed inside the Karapad tidal inlet. The experiments had to be shifted to the open sea coast after September 1978 due to blockage of the tidal creek done by the interested salt manufacturers which hampered the free flow of tidal water in and out of the tidal creek.

TABLE 1. *Total number of spat collectors used*

Name of collector	1977	1978	1979
Lime-coated tiles ..	6,000	61,000	67,000
Oyster shell ..	1,300	3,000	5,400
Asbestos sheet ..	—	—	165
Mussel shell ..	—	70	4,200
Coconut shell ..	450	—	—
Cement-coated tiles	2,000	—	—
Palmyra lattices ..	—	4	—
Bamboo mat ..	6	..	—

Preparation of lime-coated tiles for spat collection

Earthenware semi-cylindrical country roofing tiles were used as spat collectors in experiments conducted from August '77 to October '79. These tiles were procured locally from demolished tiled houses. The size of each tile was 20 cm long and 12 cm wide, weighing 450 gms, curved in a 'U' shape. Lime-coating is an important aspect which is to be done before laying them. The tiles were dipped in lime mortar a few weeks before use. For initiating lime-coating work a lime hod or vat, stirrer, tongs, rubber gloves, sand sieve, balance, bamboo mats, lime and fine sand are necessary. The hod was filled with 50 litres of sea water and mixed with 30 kg of lime gradually. Immediately after adding the lime, stirring the lime was done regularly by means of a specially made oar-shaped wooden stirrer. Preparatory coating of lime was initiated only after 10-15 minutes. Before this a thorough washing of tiles was done so as to remove dirt on the tiles to facilitate efficient attachment of lime to the tiles. A team of 5 persons is necessary for liming the tiles. A small bench was used near the hod to keep the tiles being limed. While one person stirred the hod, two others performed the dipping of tiles by using tongs (Pl. I A). Two more persons arranged the tiles to dry on bamboo mats which were used to avoid contact with the dust and sand from the ground. During the entire operations rubber gloves were used to protect the hands as otherwise contact with lime would cause blisters on the fingers. Five persons can finish 10,000 tiles within 8 hours duration. Drying of tiles was done in a shaded place avoiding direct sunlight. To avoid the same, lime-coating work was invariably done during evening hours and dried during night till the next day morning and stacked and kept in a shady place. The preparatory or undercoat of lime was only an initial process after which a further coating of mortar is necessary. A second coating of lime and fine sand mixture is to be given to the tiles before they are employed for spat collection.

For this second coating a suitable mixture was prepared by mixing 30 kg slacked lime with 40 kg fine sand in 50 litres of sea water. The hod was stirred well to give the consistency of thin oil and while doing so, scum if any formed was collected and scooped out. Tiles with preparatory coating are treated with the above mixture to achieve a thickness of 2 to 3 mm coating. The above cited quantity of mixture would suffice for 500 tiles only, since the thickness of coating is triple the first coating. Sufficient quantity of water was added to the mixture, when coating becomes too thick and stirred well so as not to allow the sand settle to the bottom of the hod. The process of drying was done as in the case of first coating.

This type of lime-coating aids easy removal without causing injury to the spat when they reach 20 to 35 mm in size. The base had therefore to be sufficiently strong to stand for a long period and yet considerably be friable

enough to allow the removal of the spat without any damage while scraping.

Selection of site for laying spat collectors

Karapad Creek was selected for laying tiles since mother oysters were also there, which will ensure success in spat procurement. The nature of bottom of the selected site was sandy mud, with a depth varying from 1.0 to 2.2 metres. An additional site in the Tuticorin Bay was also selected to augment the collection. This area was of peaty mud free of predators, enemies and competing organisms. At the proper spawning time breeding stock of oysters in sufficient quantities were transferred from the creek to the bay. The area was protected from wind and wave action by natural formation of sandy shore arm projecting into the sea on the north-eastern side.

Laying of tiles

Special type of rectangular iron frames of size 100×75×15 cm was made using 5 mm M.S. rod. Each frame was coated twice with lacoloid black and dried for a day to avoid corrosion in the sea water. The frame was knitted with 2 mm synthetic twine to get a mesh size of 2.0 to 2.5 cm. Tiles were arranged side by side in each cage with the concave side facing down. A second layer of tiles was arranged perpendicular to the first one, in such a way that a minimum of 45 to 50 tiles could be kept in a single cage in two layers. Racks of size 14×2 m were constructed by driving 12 poles of 2.5 m length. Cross pole of 2 m long were tied to the poles just 30 cm below the water level. To these longitudinal poles were tied to serve as platform for the tiles. The cage with the arranged tiles (Pl. I B) were transported to the spat collecting ground by means of a dinghi and placed on the rack in such a way that the tiles were always submerged. Each month 1,000 tiles were laid for observing the spat intensity from August 1977 to July 1978 and thereafter till 1979 maximum number of tiles used during the season of maximum spatfall.

Results

Tiles were laid in July 1977 and spatfall was noticed during the month of August with an average of 15 spat per tile. The intensity was high in September, reaching 34 spat per tile (Pl. I C, Table 2). It was possible to observe spat numbering 17 per tile to a maximum of even 84 per tile. In October the average settlement came down to 7 per tile. From November '77 to February '78 no settlement was noticed.

During '78, spatfall was noticed during March and April with an average of 8 and 26 spat per tile which progressively diminished till August and in September the spat was 2 per tile. Practically there was no settlement from June to August. Surprisingly the spatfall was poor in September, 1978. Compared to the spat settlement of the previous year, this failure of spat settlement was disappointing, but could be explained because of some man-made changes brought about in the creek where a stone barricade was put up preventing the free flow of sea water in and out of the creek. The stagnant water in the creek with the abnormal physico-chemical conditions of the water perhaps acted adversely on the spawning of oysters thereby preventing normal spawning activities. However, experiments were continued outside the creek in the open shallow areas of the bar where during the month of April '79 spat settlement of 38 numbers per tile was observed. On several tiles intense settlement was noticed numbering 105 per tile. The settlement was irregular till August '79 and in September and October '79 again spat settlement was noticed with an average of 7 and 4 per tile respectively.

From the above experiments it was possible to deduce two seasons of fairly intense spat

PLATE I A. Application of lime coating to tiles, B. Lime coated tiles kept in trays, C. Lime coated tiles with oyster spat attached and D. Oyster shells strung on galvanized wires for collection of oyster spat.

PLATE II A. Oyster shells kept in a tray for collection of oyster spat, B. Oyster spat which have set on oyster shell cultch, C. Oyster spat which have set on mussel shell cultch and D. Corrugated asbestos sheet cultch with oyster spat attached.

TABLE 2. *Monthly average rate of spat attachment per unit*

Name of spat Collector	1977					1978									1979						
	August	September	October	November	December	January	February	March	April	May	June	July	August	September	April	May	June	July	August	September	October
Lime-coated tile ..	15	34	7	1	—	—	—	8	26	4	—	—	—	2	38	—	—	—	—	7	4
Oyster shell ..	1	9	2	—	—	—	—	3	5	1	—	—	—	—	9	—	—	—	—	2	2
Mussel shell ..	—	—	—	—	—	—	—	—	4	—	—	—	—	—	6	4	—	—	—	1	2
Corrugated asbestos sheet ..	—	—	—	—	—	—	—	—	—	—	—	—	—	—	393	—	—	10	—	36	38

settlement namely March/April (maximum spatfall) and September/October (subsidiary peak). It may be mentioned in this context that Hornell (1921) observed similar feature in respect of the oysters spawning in Pulicat Lake area. Table 1 shows the monthly average rate of spatfall per unit while Table 2 shows the total number spat collectors used during '77 to '79. Since spat attachment on tiles was successful, as many as 61,000 tiles were employed during '78 and produced 3 lakhs oyster spat. During the year 1979, the spat collectors were increased to 67,000 which enabled procurement of 5.82 lakhs of spat.

Features of edible oyster spat

As soon as the spat settles down it looks like a red triangular spat. On the fifth day longitudinal lines make their appearance from the umbo region to the posterior side of the shell. The settled spat can be categorised into three kinds red, black and pale white or ash coloured with red tinge. The red coloured ones are common. At this stage the umbo region is slightly elevated and lamellar process not clear. As the spat grows, the colour appears to fade away. Uniform growth of the spat seems to be influenced by (a) the nature of the surface of the tile, (b) overcrowding of spat and (c) settlement of fouling organism especially barnacles. When the spat is not influenced by any of the above factors and settles on an even surface of the tile, growth is uniform on all directions except the umbo, and the shell assumes a circular shape *i.e.*, growth occurs both lengthwise and breadthwise. The above observation seems to be normal growth pattern of a spat.

Scraping of spat

The spat grown to 20-35 mm size were preferred for scraping. Spat was detached from tiles by using a specially designed chisel or spatula. In the process of scraping great attention was paid for the complete casting of mortar from the tiles. Gentle hits with chisel in different angles of the tiles were sufficient for the detachment of fragments or flakes along with spat.

Oyster shell as cultch

(*i*) *Stringed shells :* Experiments were carried out with oyster shells both by hanging and broad-casting methods. Clean, weathered and aged shells were used for preparation of

strings. The shells were pierced by means of a special hammer and strung to a galvanized wire of 16 size or synthetic rope of 4 mm thickness with one metre length (Pl. I D), each string containing 30 to 35 shells. The strings thus prepared were suspended from casuarina pole in the seed collection areas in such a way as to be completely immersed in water. This method of spat collection was carried out in 1977 but was not giving good results. However, strings used in April '79 in the open sea gave satisfactory resuls by getting on an average of 9 spat per shell.

(*il*) *Broad-casting method:* A different method of experiment was carried out by spreading the old oyster shells in rectangular iron cages during the above period (Pl. II A). Iron cages of size $90\times60\times15$ cm were used and shells were compactly arranged at an average of 100 shells per cage. The cages with the shells were laid on submerged racks. The incidence of spatfall was noted regularly for a period of one year and thereafter during the period of peak season alone. Spatfall was noticed during September '77 and April '78 with an average of 9 and 5 spat respectively and the number of spat being 2-26 and 1-16 respectively (Plate II B).

Mussel shell

Mussel shells were used as spat collectors during April '78, suspending them in a box-type cage from the rack to a depth of 30 cm below the low tide level. Spatfall was noticed on these shells (Plate II C) with an average of 4 spat per shell and the range was 2 to 11 spat per shell. Strings made out of mussel shells were tried from April to October '79. Spatfall was noticed during the month of April and May at the rate of 6 and 4 respectively. No spatfall noticed during the period between June and August '79 and in September again an average of 2 spat per shell was seen. The spat collection was poor as in the case of oyster shells.

Cement-coated tiles

Experiments were conducted by using the common country tiles coated with cement also. 45 kg of cement in 30 litres of sea water would suffice for coating 500 tiles. The coating and drying of tiles were done as in the case of lime-coating work. The tiles were staked in cages and laid on the rack in the creek. On the basis of the results obtained for the month of August and September '77, cement-coated tiles proved to be a good collector with an average rate of spat attachment of 19 and 34 respectively. While comparing with the lime-coated tiles, the attachment of spat was 33% higher in the case of cement-coated tiles. The attachment of barnacles on these tiles was considerably high. Although attachment was heavy, scraping of spat faced a great problem resulting in damage of spat at the time of removal. Hence this method was considered not convenient.

Strings of coconut shells

One metre long strings of 10 coconut shells with a gap of 8 to 10 cm between the shells were used during August to October '77. Lime-coating was also carried out and suspended from the pole alongwith oyster string shells. The experiment with coconut shells proved to be of no use, since the average attachment was one spat per shell even during the peak period of spatfall in September.

Wooden lattices

Palmyra sticks nailed together in the form of lattices, which after coating with tar were used during the spawning period of April '78, but this method also showed very poor attachment of spat.

Bamboo mats

Six numbers of bamboo mats 2 m long and 1.5 m broad, were used during September '77. The mats were examined next month and the

spat attachment was very poor. Heavy accumulation of silt and formation of a thin algal growth in it probably does not permit settlement.

Corrugated asbestos sheet

Corrugated asbestos sheet bits of 30×30 cm size were used as spat collectors in April '79. Five sheets were tied into a 'bouquet' with synthetic or coir rope and placed on the rack. Three such stacks containing 15 sheets were laid on the rack to remain submerged in water to a depth of 15 cm below the water level. The sheets were examined periodically. After 20 days it was observed the heavy settlement of spat with a minimum of 254 and maximum of 517 spat/sheet which gave an average of 393 spat per sheet. Good spat settlement was noticed (Pl. II D) on all sheets except the uppermost one of a bouquet in which the lowerside had got heavy spat settlement but the upper side was deposited with heavy silt.

Detaching the spat was found difficult from the sheets. Hence lime-coated asbestos sheets were used from July to October '79 and the average rate of spat attachment was 10, nil 36 and 38 respectively. The removal of spat was easy as in the case of lime-coated tiles.

Discussion

The Romans cultured oysters as early as the first century B.C. though they did not have scientific knowledge. The practices of the Romans have helped in the revival of oyster culture in France in the nineteenth century.

The available information of the methods of oyster spat collection is scanty in India. However, Hornell (1916, 1921) has reported on the use of roofing tiles at Pulicat Lake. Devanesan and Chacko (1955) tried casuarina twigs, oyster and cockle shells, but did not get encouraging results. Nair (1975) has reported the suitability of using cement-coated oyster shells at Athankarai Estuary. According to Sundaram and Ramadoss (1978) lime-coated flat tiles gave good results at Tuticorin and on an average of 10 to 15 spat settled on either side.

Considering the easy availability and the cheapness of the materials to be used for spat collection from nature and also suitability to collect very huge quantities of spat it is clear that at present the spat collection and supply problem do not pose any difficulties. By the present method, large scale seed demand from the growers market can be effectively met. But this by itself does not assure satisfying the future demands and while planning for future the sure way to cope up with the requirements would be to develop a secure method of hatchery production of oyster spat as has been done in major oyster growing countries like Japan and U.S.A. To this end our experiments are already progressing. But it should be conceded that the maximum use of natural resources must be made for which the lime-coated tiles seems to be the best answer for spat procurement.

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SETTLEMENT OF OYSTER LARVAE IN PAKISTANI WATERS AND ITS POSSIBLE IMPLICATION FOR SETTING UP OYSTER CULTURE

M. MOAZZAM AND S. H. NIAZ RIZVI
Institute of Marine Biology, University of Karachi, Karachi-32, Pakistan

ABSTRACT

The paper deals with the larval settlement of oysters *Crassostrea rivularis* and *Ostrea folium* in the coastal waters of Pakistan. The settlement of oyster larvae was observed to be continuous round the year except for the monsoon months of July and August. The peak of larval settlement of *O. folium* in coastal waters was found to be between October and February. The peak settlement of larvae of *C. rivularis* was found to be during June and in September-October in the salt water creeks near Karachi. The larval settlement of this species was also observed during July-August in considerable numbers in the creeks. In order to determine the choice of substrata exercised by the settling larvae, various materials were used for spat collection. Asbestos and wood appear to be the most preferred of all the substrata exposed in the sea during the present study. The feasibility for setting up oyster culture in the coastal waters of Pakistan based on the results obtained during the present investigation is discussed.

INTRODUCTION

EDIBLE OYSTERS are no longer found in commercially exploitable stocks on the coast of Karachi and Sind, although once they were very abundant. Hornell (1910 a, b) had pointed out that siltation, pollution (sulphurated water) and overfishing were the probable causes of their depletion along this coast. Hasan (1960) examined some of the oyster beds of the Sind Coast which had been surveyed earlier by Hornell (1910 a) and recommended that oyster culture was the answer to increase production in this area. Hasan (1960, 1963, 1964) himself undertook some preliminary experimentation with local oyster with the aim of developing oyster culture, studied their biology, planktonic larvae and spat fall, without having much knowledge of the taxonomy of local oyster. These were subsequently identified by Ahmed (1971) as *Crassostrea rivularis*, *C. virginica*, *C. glomerata*, *C. tuberculata*, *C. gryphoides*, *C. quercina* and *Ostrea folium*. Some other studies on the biology of the local oysters were also published subsequently (Ansari and Ahmed, 1972; Asif, in press; Kazim, 1953; Nooruddin, 1960; Qadri, 1951).

The present paper reports observations on larval settlement and growth of spat of *O. folium* and *C. rivularis* with the hope that if the two species were to be cultivated, abundant seed would be available. The data form part of a study on the biology and larval settlement of fouling organisms which occur in the cooling system of power plant located at Paradise Point, Karachi. Oysters were one of the major foulers in different parts of the cooling system of the power plant (Ahmed *et al.*, 1978). The species *O. folium* was removed in tons from the intake channel, intake bays and other parts of the plant during cleaning operations. *Crassostrea rivularis* which is the principal edible species on this coast (Ahmed, 1971) was also present with *O. folium* in fair numbers. The preferred habitats of *C. rivularis* are the backwaters and creeks along the coast where *O. folium* may also occur occasionally. The occurrence of these two species on the open coast at Paradise Point may be attributed to

the fact that the power plant site has been made artificially ' protected ' and simulates conditions of backwater environment.

We are grateful to Prof. (Dr.) Muzammil Ahmed, Director, Institute of Marine Biology for criticism and suggestions for the improvement of this manuscript. We extend our gratitude to the authorities of the power plant (KANUPP) for providing us facilities to carry out the present investigation. The present work constituted part of a research project, P.S.F. Res. Proj. SU-Ocean-2, awarded to the Institute of Marine Biology, University of Karachi by Pakistan Science Foundation; this financial support is gratefully acknowledged.

Materials and Methods

The biofouling study consisted, in part, of an investigation of the settlement of larvae of different marine organisms on exposure panels (measuring $7.5 \times 10.0 \times 1.25$ cm made of different materials suspended from a wooden raft which was kept afloat at all times in the intake channel of the power plant. Data collected from wooden and asbestos panels only are considered here. Physical parameters such as temperature, salinity, content of suspended matter and transparency of seawater were also determined one to four times per week during the study period 1974-1977 using appropriate methods (Ahmed *et al.*, 1978).

Oyster spat was counted from both sides of the exposure panels under a stereomicroscope. The panels were usually exposed for two weeks at a time but some were left suspended for periods of upto 90 days. Growth of spat was measured from those settled on panels exposed for longer periods of 28, 35, 42, 56, 80 and 90 days. Identification of *O. folium* and *C. rivularis* was difficult to begin with (in 1974) but soon sufficient mastery was attained for reliable distinction. The spat of *O. folium* bears an unmistakably greenish tinge on its left valve and it is absent in *C. rivularies*. Identification of the spats was further confirmed after they had grown to a size of 10 to 15 mm.

Observations

Study area

The main study on settlement of oyster spat was carried out at Paradise Point situated on the west coast of Karachi (Fig. 1). This is basically a rocky shore having frequent stretches of boulders and sand. Physical features of this shore have been described by Heq *et al.* (1978) and Siddiqui (1959). The subtidal area along this shore is generally more deeply inclined than the rest of the coast. Wave action is intense all along the shore except in the protected intake channel of the power plant.

Environmental factors

A graph of seawater surface temperature recorded at the study site is shown in Fig. 2. Minimum temperatures of 20°-22° C were recorded during December-January and maximum temperatures of 28° to 30° C during June-July. Seasonal fluctuations in salinity are illustrated in Fig. 3. In general, salinity remained fairly constant (35 - 36‰) except during the short spell of rains during July to August when salinity as low as 28‰ was recorded. The contents of suspended matter (Fig. 4) of seawater at the study site fluctuated from 0.003 mg/L in November to 0.116 mg/L in June. Maximum values of suspended matter coincided with the Southwest monsoon period (June through September) whereas the minimum values coincided with the clam periods (October - November). Transparency of seawater at the site was found to be less than 1 m during June - July.

Larval settlement

Observations made on larval settlement of oysters on wooden and asbestos panels are presented in Figs. 5 and 6. It is evident that

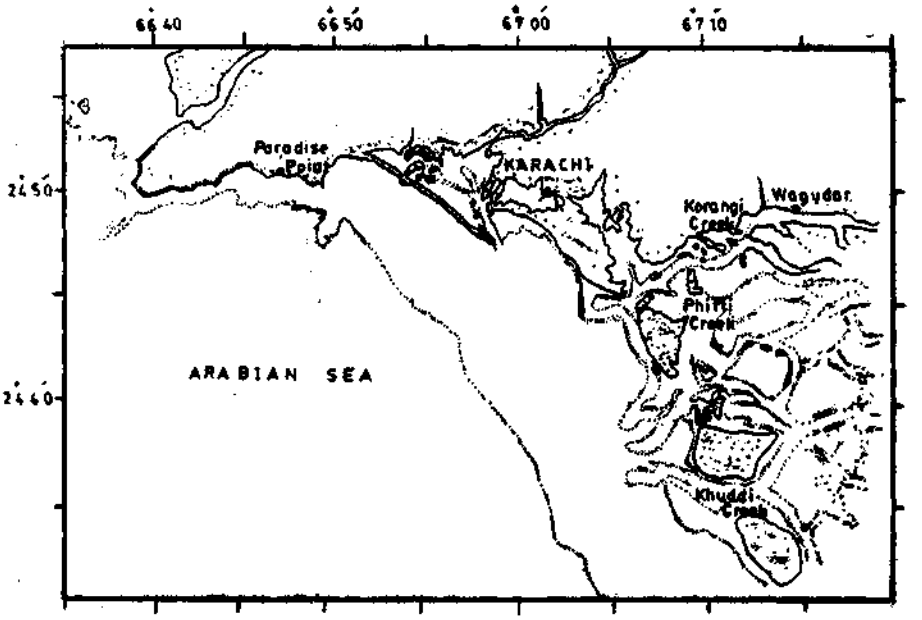

FIG. 1. Map of Karachi showing the study area.

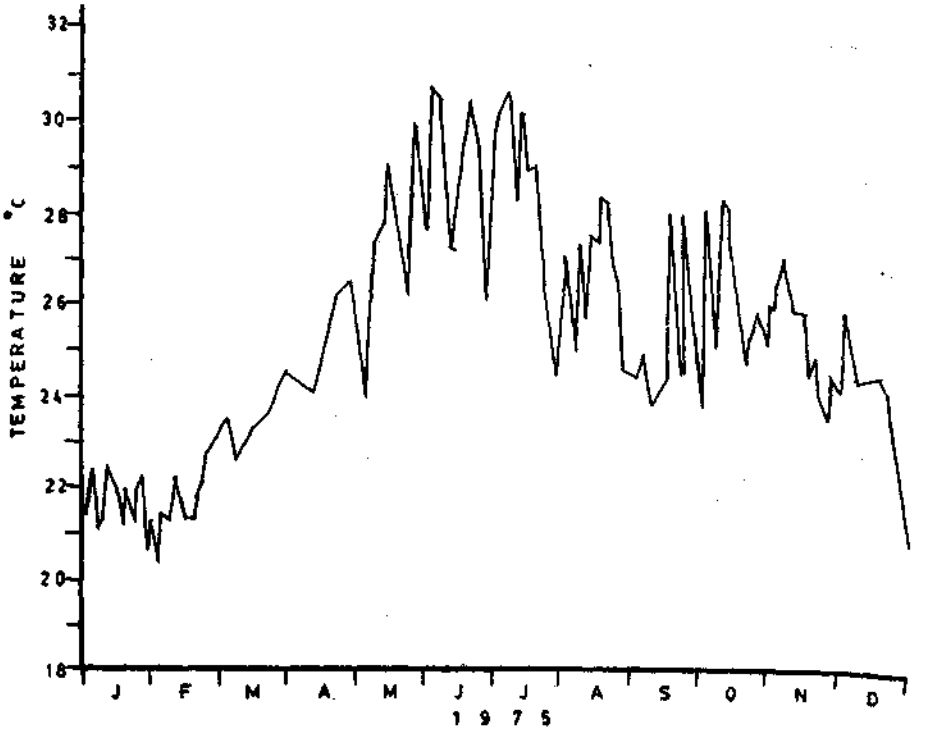

FIG. 2. Seasonal changes in the seawater temperature at Paradise Point, Karachi.

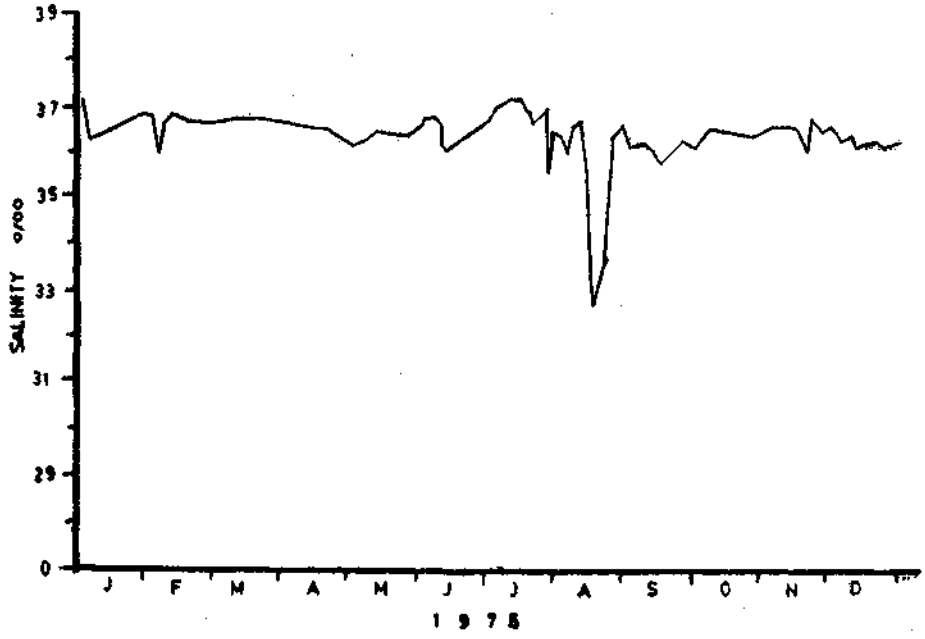

FIG. 3. Seasonal changes in the seawater salinity at Paradise Point, Karachi.

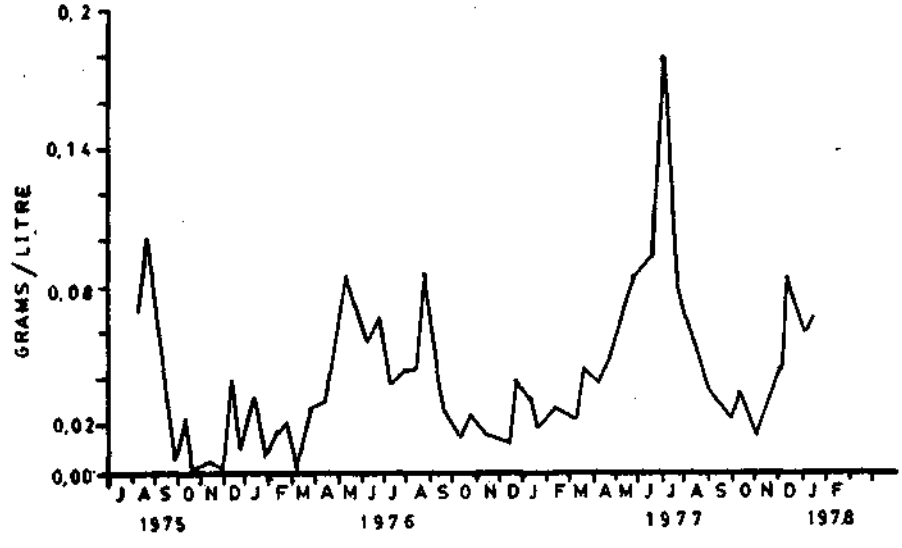

FIG. 4. Seasonal changes in the content of suspended matter in seawater at Paradise Point, Karachi.

maximum spat settlement occurred from October to February and in June with peak settlement during November to February. The larvae which settled during October-February belonged to *O. folium* and those in June to *C. rivularis*. Settlement was also observed during occasional

Larval settlement in creeks

The pattern of larval settlement of *C. rivularis* (identified as *Ostrea discoidea*) recorded by Hasan (1960) from the Wagudar Creek and that recorded from the sea coast at Paradise Point in the present study is almost similar.

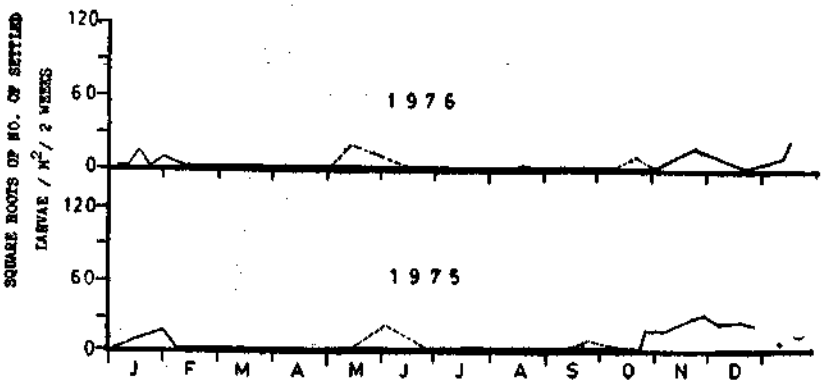

FIG. 5. Seasonal changes in the settlement of oyster larvae at Paradise Point, Karachi on asbestos panels. Dotted line represents spat of *Crassostrea rivularis* and solidline *Ostrea folium*.

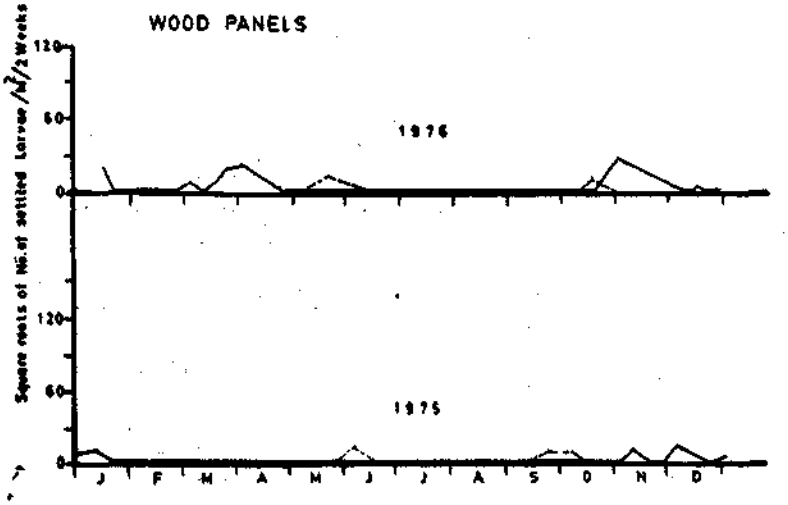

FIG. 6. Seasonal changes in the settlement of oyster larvae at Paradise Point, Karachi on wood panels. Dotted line represents spat of *C. rivularis* and solid line *O. folium*.

clam periods in the turbulent months of July and August. *Crassostrea rivularis* constituted only a small portion of the total larvae settled during September through October; the major larval settlement belonged to *O. folium* during this period.

Settlement at Wagudar creek occurred during July to December whereas at Paradise Point in June and September to October. We tend to attribute this difference mostly to the presence of high contents of suspended matter in seawater (Fig. 4) during the south west monsoon

period. This high content of suspended matter is believed to interfere with larval settlement of many invertebrates in this area (Ahmed *et al.*, 1978). Since the preferred habitats of *C. rivularis* are the protected backwater and creeks the settlement of its larvae on the open coast must have occurred only in the periods of minimum wave action.

Planktonic larvae

Counts of bivalve larvae recorded from plankton samples collected during 1975 are shown in Fig. 7. No attempt has been made to distinguish oyster larvae from those of other bivalves. A maximum number of 8000 bivalve larvae/m^3 occurred in February while they were either negligible or absent in the samples collected during April to June.

Early growth of spat

The pattern of early growth of *O. folium* spat is presented in Fig. 8. Observations on the growth of *C. rivularis* spat were relatively few. It is evident from the figure that growth was slow during the first 5-6 weeks, but increased thereafter. A shell length of 4 mm was attained after five weeks and 15 mm after 85 to 90 days of initial settlement. Growth rate was higher during October-November than during the rest of the year. Growth of oyster spat was inhibited due to fouling by barnacles and other organisms on the panels. Growth as well as fresh settlement of oyster larvae were affected particularly by barnacles during the first six weeks of spat settlement in the period August to November.

Discussion and Recommendations

During the present investigation it was interesting to note that while six species of oysters have been shown to occur along the coast of Karachi (Ahmed, 1971), the exposure panels could only catch spats of two species namely *O. folium* and *C. rivularis*. It may perhaps be attributable to the subtidal heights on which the panels were suspended on the floating raft. It is surprising that spat of *C. madrasensis* (Ahmed, 1971 had originally identified this species as *C. virginica* but now favours its redesignation as *C. madrasensis*; personal communication) which occurs on the same stones and pilings as *C. rivularis*, have not been found on the panels at all.

In the present study spat of *O. folium* attained a size of 15 mm in 85 to 90 days whereas Hasan (1960) reported that *C. rivularis* spat reached the same size in 90 days in Waugudar Creek. It would seem, therefore, that growth rate of *O. folium* spat on the open coast approximates to that of *C. rivularis* in the creeks. Both localities, therefore, may be suitable for spat collection. There does also exist the possibility of relaying in the creeks the spat of *O. folium* caught on the open coast at Paradise Point.

Plankton samples which were analysed during the present study have provided data on the abundance of bivalve larvae on the open coast. Since these could not be identified into species such data cannot be made use of in determining the breeding periods of the two oysters. Hasan (1960) has, however, reported *O. discoidea* (= C. *rivularis*) to spawn during July to November at Waugudar Creek whereas Ansari and Ahmed (1972) mentioned the period April-May to November in Karachi area. The extent of the spawning season of *O. folium* has not been documented so far but their mantle cavities have been seen to contain larvae during May to October (Ahmed, per. comm.).

Data regarding the biology of local oysters spat settlement and growth as well as their spawning, obtained by Hasan (1960), Ansari and Ahmed (1972), Asif (in press) and in the present study could now be utilized for setting up oyster culture along the coast of Karachi. Although various methods of oyster culture are known (Bardach *et al.*, 1972) the following procedures may be adopted in Pakistan in the light of the observations made earlier in this study.

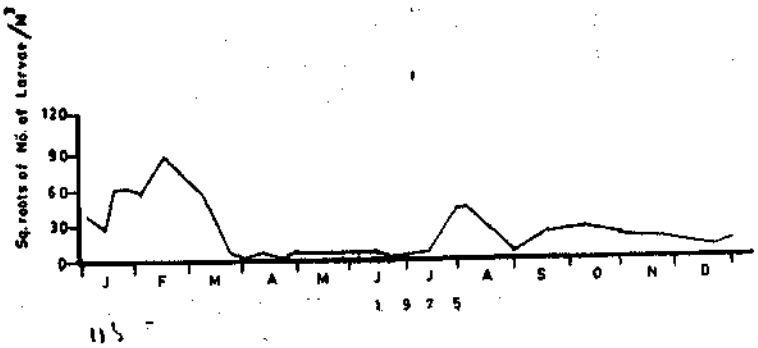

FIG. 7. Seasonal changes in the numbers of bivalve larvae in the plankton collected at Paradise Point, Karachi.

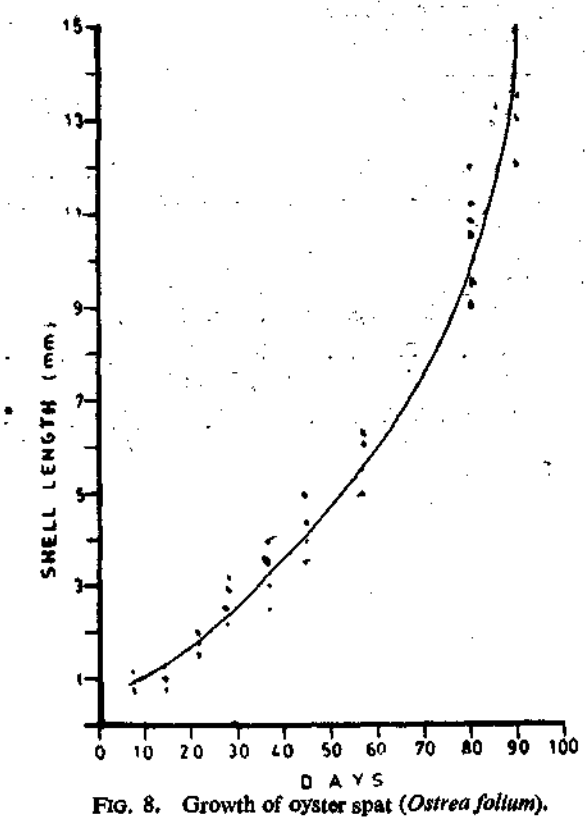

FIG. 8. Growth of oyster spat (*Ostrea follium*).

1. The collection of spat may be made either on wooden panels of the size 30 × 10 × 5 cm or on bamboo sticks measuring 60 cm in length which could be vertically suspended from a wooden raft. Earthenware tiles coated with lime, sand and cement could also be used for spat collection considering their low cost of production as suggested by Hasan (1963). Panels of wood or bamboo would be useful in facilitating the easy detachment of the settled spat.

2. Spat collectors could be placed along the open coast during September to April and in the creeks in July to December. Since variations in periods of larval settlement occur it would be desirable to register the precise start of larva settlement so as to minimize the fouling of spat collectors by other settlers. The precise time of larval settlement during each season could, however, be determined by exposing experimental spat collectors in selected areas at intervals of at least three days.

3. Oyster spat may be allowed to grow on spat collectors in a state of suspension from the

raft for a period of 12 to 13 weeks till a size of about 15 mm is attained before their transfer to the creeks or protected grounds. Care should be taken to rid the spat collectors of fouling organisms.

4. Spat of about 12 to 15 mm size may be taken to the creeks for further growth.

It is hoped that the above mentioned information would be useful in the establishment of oyster culture in the country.

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STUDIES ON MATURITY STAGES AND SPAWNING PERIODICITY OF *CRASSOSTREA MADRASENSIS* (PRESTON) AT TUTICORIN BAY

M. E. RAJAPANDIAN* AND C. T. RAJAN*

Central Marine Fisheries Research Institute, Cochin 682 018

ABSTRACT

Studies undertaken during 1976 on the gonadal maturity of the backwater oyster *Crassostrea madrasensis* at Tuticorin revealed a biannual spawning periodicity. March-April and August-September were found to be the peak periods of spawning. A broad correlation has been observed between diurnal temperature difference and spawning of oyster.

INTRODUCTION

THE PRESENCE of a sizeable population of edible oyster *Crassostrea madrasensis* in the natural bed at Karapad tidal creek opening into Tuticorin Bay of the Gulf of Mannar and the suitable environmental factors in the area favoured the establishment of an experimental oyster farm. Monitoring of the course of reproductive activity of the oyster in the natural bed was taken up as to determine the right time when spat collectors are to be set up. Since oyster reproduction is considerably influenced by environmental factors, observations on temperature, salinity, dissolved oxygen and pH were made. The results of this study were immensely useful in organising oyster spat collection on a large-scale.

The authors express their gratitude to Dr. E. G. Silas, Director of this Institute for his encouragement and they offer their sincere thanks to Shri K. Nagappan Nayar and Shri. S. Mahadevan who supervised the work and made valuable suggestions.

* Present address : Tuticorin Research Centre of the Central Marine Fisheries Research Institute, Tuticorin 628 001.

MATERIAL AND METHODS

A random sample of 50 oysters was collected once in every fortnight from January to December 1976. The stage of maturity was determined by examination of fresh smears of gonad. Histological sections of gonad tissue were prepared and examined.

Water temperature was recorded twice a day at 6 A.M. and 3 P.M. which invariably represented the minimum and maximum temperature of the day. Salinity, dissolved oxygen and pH of the water were recorded once in a week.

OBSERVATIONS

The percentage composition of oysters in different maturity stages (females), indeterminates and males is shown in Fig. 1. For this study the maturing and ripe stages are treated together under 'ripening stage' and partially spent and nearly spent stages as 'spent stage'. The indeterminables and males have been considered as such.

Indeterminable stage

Oysters with gonads in this stage were dominant during July, registering 67% (Fig. 1).

Fairly high percentages were also observed during May-June. In August this condition dropped to 34% and rose only during November and continued to increase till February. A decline in the percentage was noticed from February and the lowest was recorded during April (23%).

Female

(*a*) *Ripening stage :* Although the female oysters showed maturing and ripe stages of gonads almost throughout the year, a biannual maximum was quite apparent during February-April (34-40%) and August-September (29-35%). In the first phase highest percentage was registered during March and in the second during September. During May it decreased to 26% and in the months of June and July it got further reduced to 6% and 7% respectively. From October onwards the percentage of ripening females showed a progressive decline till January (16%).

(*b*) *Spent stage :* Oysters in spent stage were at an extremely low level in the month of July (2%). The poor occurrence of spawners during the preceding month might be the probable cause for this feature. Ensuing the second spawning during October and November, the percentage of this stage increased to 28 in December.

Males

Relatively high percentages of males were observed during the months of April, June, August, September and October (30-38% of samples). The males were relatively more in number in the second half of the year than in the first.

Environmental conditions

Data on temperature, salinity, dissolved oxygen and pH are given in Table 1. There were no marked fluctuations in salinity, dissolved oxygen or in pH. While observing the diurnal

TABLE 1. *Hydrological data of the study area during* 1976. *Monthly averages of daily temperatures (min. at* 6 *a.m. and max. at* 3 *p.m.) and differences are given*

Temp. °C Minimum	Temp. °C Maximum	Variation in daily temp. °C	Salinity ‰	Oxygen ml/litre	pH
23.8	28.2	4.4	35.09	3.92	8.06
23.5	28.4	4.9	35.17	3.75	8.22
26.2	30.8	4.6	36.00	4.14	8.02
27.9	32.6	4.7	36.40	3.61	8.12
27.2	31.7	4.5	36.60	3.78	8.08
27.4	30.5	3.1	35.80	3.96	8.05
25.3	28.2	2.9	37.48	4.05	8.18
25.2	28.5	3.3	36.93	4.09	8.23
25.9	30.1	4.2	36.95	3.97	8.40
27.6	30.4	3.5	36.80	3.98	8.39
27.6	30.4	2.8	34.07	4.14	7.98
25.8	29.3	3.8	31.70	5.04	8.01

variations of the water temperature it was found that during certain months the mean values of the diurnal variations remained high (Fig. 1).

Discussion

Many workers are of the opinion that under natural conditions, gonad development and spawning of oysters are well defined seasonal phenomena (Korringa, 1941 ; Loosanoff and Davis, 1952 ; Butler, 1958 ; Rao, 1956 ; Durve, 1965). Loosanoff and Davis (1952) observed that the period required for maturation of gametes in oyster is a function of time and temperature. He had demonstrated that 50% of the oysters developed mature gametes at temperaturs of 15.0°C, 20.0°C, and 25.0°C in 26.5 days, 7.9 days and 5.4 days respectively. Butler (1955) observed that the spawning and setting of oyster larvae in the natural conditions in all years, first occurred only after there had been a minimum temperature increase of 5°C in the proceeding 30 days period.

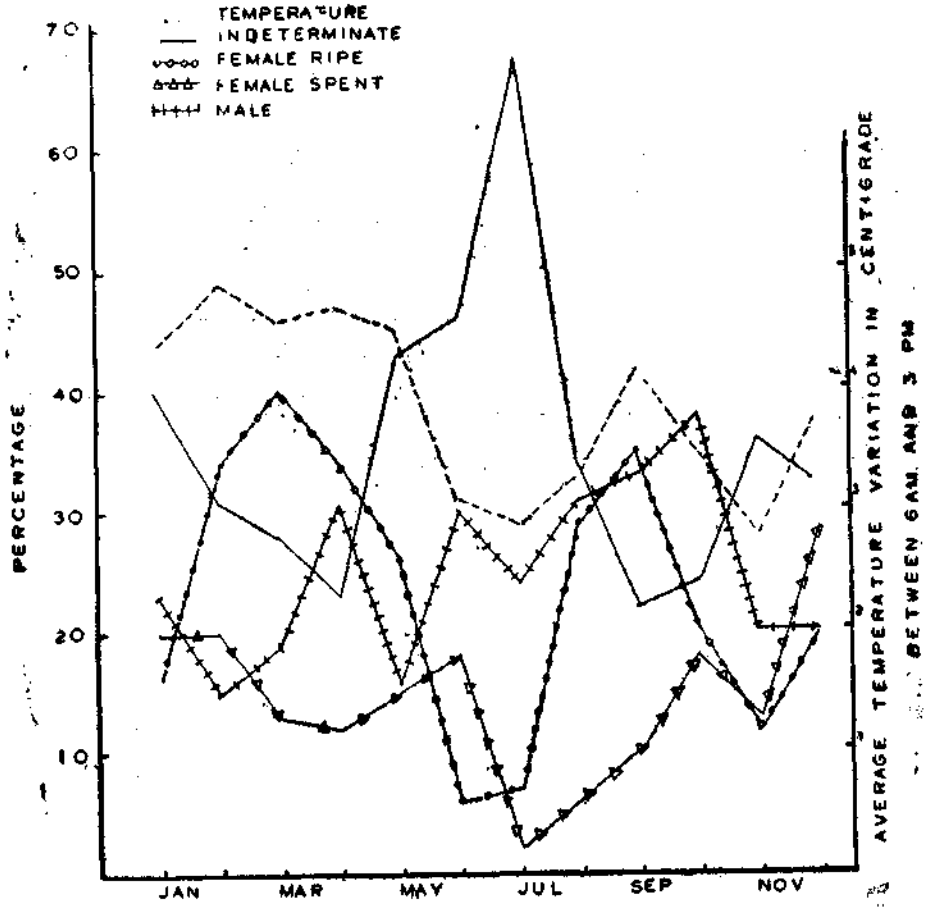

FIG. 1. Percentage composition of *Crassostrea madrasensis* in different maturity stages (females) indeterminates and males. Monthly averages of daily temperature difference between maximum and minimum (data from Table 1) are given.

The monthly averages of the diurnal variations in the minimum and maximum temperature were observed to have well defined relation to the development of gonads, spawning and

the setting of spat. The highest value observed during February initiates the maturation of the oysters and the spawning continues till the month of May. The drop in temperature values during the months of June and July showed a corresponding decline in the spawning activity of the oysters. During the month of September this value of temperature increased showing a rise in the spawning population (Fig. 1).

Experiments on oyster spat collection in Karapad Creek (Thangavelu and Sundaram, 1983) confirm the observations. They observed that the spat collection tiles laid from July 1977 to June 1978 have been shown two distinct periods of spat settlement. The rate of spat settlement during August, September and October 1977 were 15, 34 and 7 spat per tile respectively and from November to February 1978 the settlement was almost nil. During March, April and May of 1978 the rate of settlement were found to be 8, 26 and 4 tiles respectively.

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Proc. Symp. Coastal Aquaculture, 1983, 2 : 479-482

INDUCED SPAWNING OF THE ADULTS AND LABORATORY REARING OF THE LARVAE OF THE EDIBLE OYSTER *CRASSOSTREA MADRASENSIS* (PRESTON)

K. VIRABHADRA RAO

Retired Fishery Scientist, Plot No. 1309, Anna Nagar West, Madras-600 040

ABSTRACT

The paper describes the results of an early experiment on spawning of *Crassostrea madrasensis* and larval rearing in the laboratory. When the gonad was ripe the female readily spawned in the presence of milt from the male oyster in the surrounding water. The results of stimulation by the use of aqueous solutions of chemical substances showed erratic trends.

The fertilised eggs developed into the straight hinge veliger in 12 to 16 hours in a salinity medium of 28 to 30‰ in sea water at a temperature range of 22 to 25°C in the culture bowls. Phytoplankton isolated from the plankton and cultured separately in fingerbowls formed the food of the larvae. The larvae did not show uniform growth. When some of them were well grown to pediveliger stage at the end of 19 days after fertilization, the majority of them remained small and did not grow beyond the early unbonal stage. The fully formed pediveliger after a free swimming existence and occasional creeping at the bottom of the fingerbowl finally settled with the left valve down, cementing it to the glass bowl by a secretion excluding from the foot glands to lead a sedentary life as spat.

INTRODUCTION

INDUCING the adult oyster *Crassostrea madrasensis* to spawn, raising the fertilized eggs to larval stages and rearing the latter to set as spat involves standardisation of techniques which when perfected will ensure increased production to levels to which adequate facilities are built up. At an experimental level, the present writer succeeded in raising fertilized eggs through larval stages to settlement of spat in *Crassostrea madrasensis* for the first time while he was in service at the Central Marine Fisheries Research Institute and this report deals with the methods adopted with brief descriptions of the developmental stages.

MATERIAL AND METHODS

Crassostrea madrasensis were obtained from the Adyar Estuary and Ennur Backwater near Madras during the breeding season (November-December) which has been previously ascertained in the course of separate investigations (Rao, 1951, 1956). Oysters, exceeding 5 cm in long axis and in apparent healthy condition, were cleaned and kept in glass troughs in sea water with salinity adjusted to that of the environment from which they had been collected, by adding required amounts of distilled water. The oysters were fed on an abundance of algal cultures prepared in pastuarised sea water to which soil extract, sodium nitrate and sodium phosphate were added. In the absence of mono-algal stock cultures a few phytoplankters microscopically examined and isolated from inshore plankton were introduced to the culture medium in which they thrived and multiplied in profusion.

Chemical stimulation was tried by injecting the edge of the mantle of the oyster with extremely weak aqueous solutions of potassium chloride, ammonium chloride, ammonium hydroxide and calcium chloride ; of them calcium chloride failed to induce spawning in all cases and the other three solutions produced erratic reactions with majority of the oysters not responding.

Male oysters readily spawned with the increase in water temperature upto 35°C and females only occasionally. It has been observed that the best inducement for the female oyster was afforded by the presence of sperm in the surrounding water. Male oysters have also reacted in a similar manner to the eggs of the oyster introduced into the water. Spontaneous spawning was observed on occasions, soon after the water in the jars was changed.

Spawned out eggs and milt were mixed in fingerbowls in sea water with salinity adjusted to 28 to 30‰ at ambient temperature. It is considered that this is about the optimal level of salinity for this species for larval development (Rao, 1951). Water was changed once a day, filtering it through sintered glass funnels to prevent escape of the larvae. Fresh sea water of adjusted salinity filtered through millipore filter replaced the water removed from the fingerbowls. The developing eggs and larvae were removed at frequent intervals for microscopic examination in cavity slides. When larvae started feeding, about 10 ml of algal culture was added to each fingerbowl every day. Minute microflora like *Chlorella* spp. present in the culture formed the food of the larvae. When the larvae had grown in size, changing the water in the fingerbowls presented no difficulty as they could be spotted under the magnification of a hand lens, picked up with a pipette and transferred to fresh bowls. Settlement of spat took place in the fingerbowls in which they were reared.

Developmental Stages of Eggs and Larvae

The just spawned out eggs measuring 60 to 63 μm in diameter enclosed in a thin vitelline membrane was spherical, colourless and translucent with granular yolk in the cytoplasm and a large hyaline nucleus at the centre. In some the nucleolus was also clearly seen. Within 20 to 30 minutes after fertilisation the first polar body appeared at the apical pole and it was followed by the extrusion of the second polar body. Segmentation of the egg commenced resulting in two unequal cell stage, a pseudo three cell stage, four cell stage (Pl. I A, B) and an eight cell stage in quick succession within about 45 minutes. At eight cell stage, seven cells were small and one was large. The smaller cells the micromeres dividing had spread over the large cell giving rise to the morula or blastula which developed cilia and began rotatory movements in the water at about two and half hours. This was followed by gastrulation at three to three and half hours, partly by epiboly and partly by invagination, the blastopore appearing initially at the vegetative pole but subsequently shifting ventralwards (Pl. I D). The next stage was the trochophore formed at about five and half hours after fertilisation and it measured about 75 μm with a large preoral lobe destined to develop into the velum and a primordium of a shell in a glandular invagination of the ectoderm. The preoral lobe revealed larger cilia which enabled a more rapid movement than in the earlier stage (Pl. I E).

Between twelve and sixteen hours after fertilisation an equivalve early veliger larva in straight hinge stage was formed with a well developed velum protruding out between gaping valves for rapid movement. An archenteric space was visible without the oesophagus, intestine and digestive gland. The larva was thus still apparently incapable of ingesting food and when disturbed it was withdrawing the velum and settling down at the bottom of the culture bowl (Pl. I F). It measured about 85 μm in length at the hinge. With the formation of the alimentary canal, the veliger larvae started feeding on the organisms contained in the algal cultures introduced into the finger bowls. However, after the early veliger stage, the larvae suffered heavy mortalities.

The growth of the veligers was accompanied by the development of the anterior and posterior

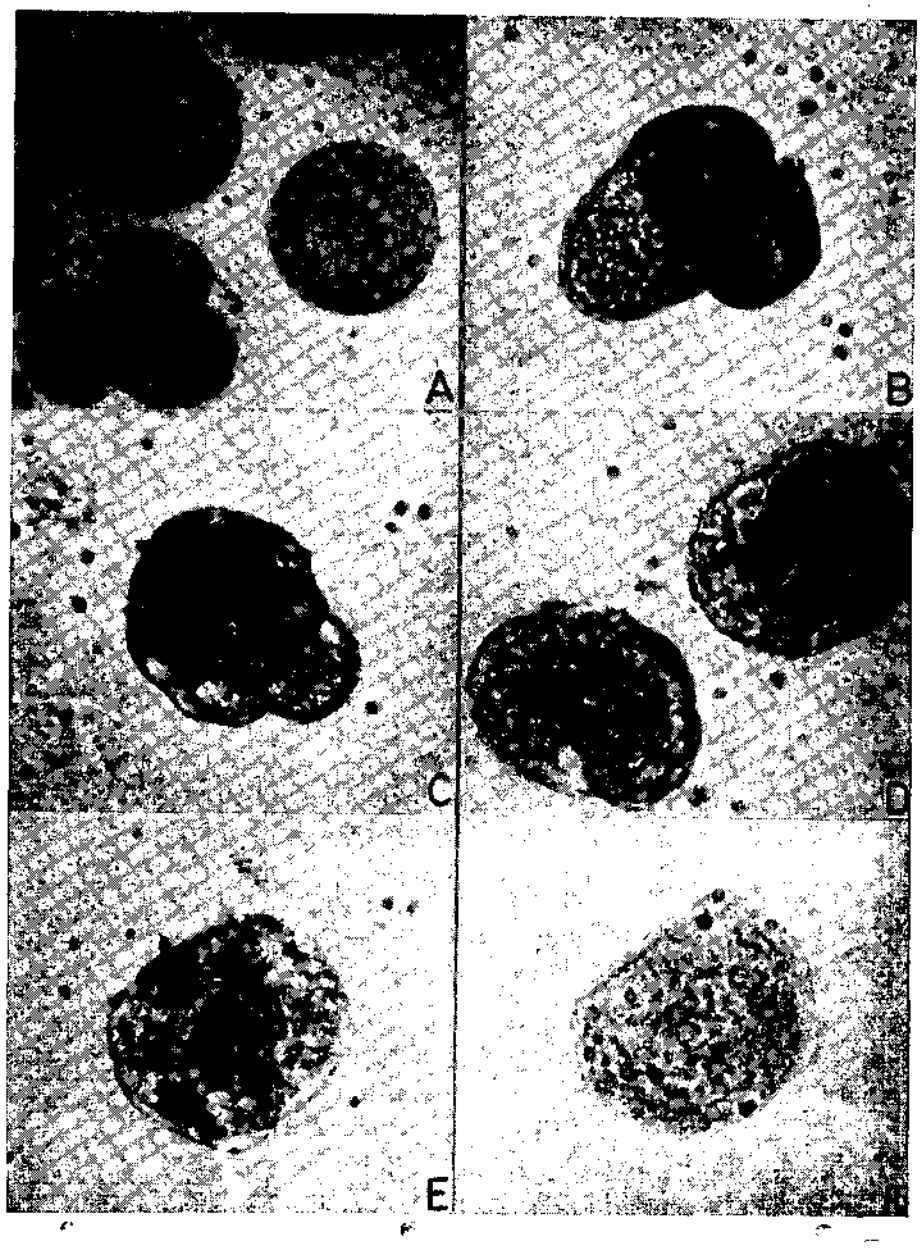

PLATE I. Developmental stages of *Crassostrea madrasensis* : A. Dividing and undivided eggs. Actual size of undivided eggs 60-63 μm, B. 4 Cell stage. Actual size 65-68 μm, C. The smaller cells spreading over the larger cells giving rise to morula stage. Actual size about 70 μm, D. Gastrula stages. Actual size about 70 μm, E. Trochophore stage. Actual size about 75-80 μm and F. Straight-hinge stage of veliger larva. Actual size about 85 μm.

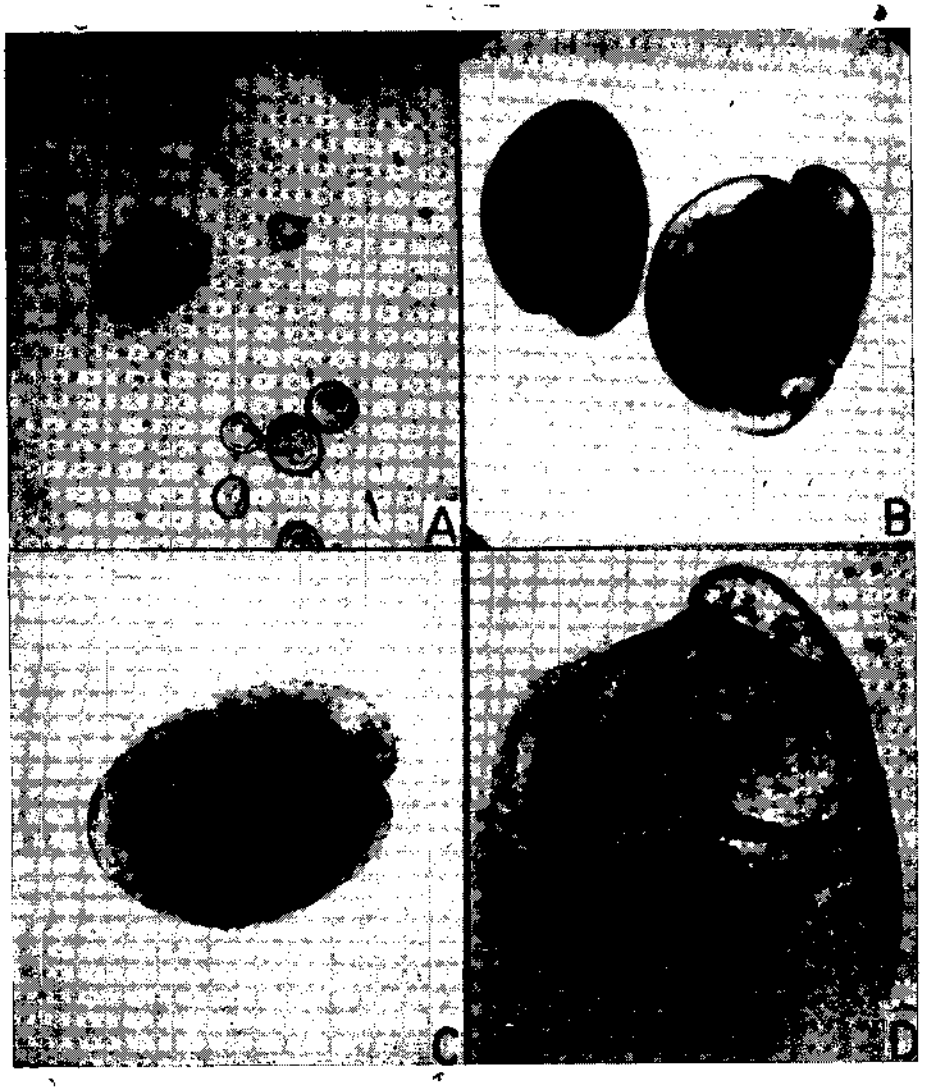

PLATE II. Developmental stages of *Crassostrea madrasensis*: A. Unequal development of the larvae of the same brood, B. Pediveliger stages of larvae. Actual size about 300μm, C. Pediveliger stage about to set. Actual size over 311 μm and D. Early spat, one day after setting. Actual size about 1 mm in long axis.

adductor muscles, the formation of the umbo at the anterior middle region of the hinge, the inequivalve development of the shell, the origin of a rudiment of a gill, the appearance of the statocyst and the pigmented eye spot. The velum had developed into a fairly large structure for locomotion and food capture and a small foot was in the process of formation. By the nineteenth day after fertilization some of the larvae had nearly grown upto 298 μm with a distinct inequivalve shell, broad at the base and narrow and bluntly pointed at umbo acquiring a brownish colouration (Pl. I E, Pl. II C). The foot had grown into a finger shaped or strap-like structure with the help of which the larva now known as the pediveliger was occasionally creeping at the side or at the bottom of the finger bowls. There was no uniformity in the growth of the larvae. While some had grown to pediveliger stage others remained smaller even at the early umbonal stage (Pl. II A).

The largest of the pediveligers observed was 302 μm in its long axis. After occasional creeping with the foot, the pediveliger had settled at the bottom with the left valve down. With the exudation from the foot glands, the shell valve was cemented to the surface of settlement. This brought about a shift from the free-living habit to one of sedimentary life as spat (Pl. II D) which involved rapid structural changes, they being absorption of the velum and the foot, the disintegration of the eye spot, elaboration of the gills, development of the labial palps and a change in position of the adductor muscle very nearly to a central place between the two valves. The anterior adductor muscle disappeared but the statocyst remained. The original larval shell was retained as prodissoconch in the spat and at the edges of the valves fresh shell material was continuously laid down by the secretion from the mantle edges.

Remarks

Some of the techniques developed for induced breeding in large number of molluscs have been reviewed by Ino (1973). According to him they involved physical, electrical, mechanical, chemical and biological stimulations. In general they are not universal in their application, the responses varying with the species concerned. The present writer has tried some of the methods in inducing the green mussel *Mytilus viridis* to spawn with varying degrees of success (Rao *et al.*, 1976). Perhaps, the oysters can more readily be induced to spawn than most other groups of molluscs. In the American oyster *Crassostrea virginica* Galtsoff (1938) reported spawning reaction of female in the presence of sperm in the water. Loosanoff and Davis (1963) reported that the suspension of sex products along with increase in water temperature helped *Crassostrea virginica* and *Ostrea edulis* to initiate spawning. In the present observations *C. madrasensis* could be induced to spawn with the suspension of sperm in the water without increasing the temperature as the prevailing water temperature are always generally high.

C. madrasensis is a dioecious species in which the sexes are separate with hermaphrodite forms occurring occasionally (Rao, 1956). In the open tanks the volume of water required for successful larval culture is to be ascertained by experimentation. The present paper reports only the possibility of initiating tank culture experiments and the details have to be worked out. Mono-algal cultures used for feeding are bound to result in more rapid growth of the late veliger larvae for setting of the spat to take place in a shorter period of time.

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EARLY LARVAL DEVELOPMENT OF EDIBLE OYSTER *CRASSOSTREA MADRASENSIS* (PERSTON)

D. SAMUEL*
Central Marine Fisheries Research Institute, Cochin-682 018

ABSTRACT

Mature male and female edible oyster *Crassostrea madrasensis* selected from the Tuticorin oyster farm were stripped and the eggs were artificially fertilized in glass containers. 91.5 to 92.2 percentage of eggs underwent successful fertilization and the larvae were reared upto umbo stage in one experiment. The size of the mature egg varied from 49 to 59 μ. A method of filtering and washing the eggs using centifugation was devised as the eggs could not be retained even by the finest sieve (Nylobolt bolting cloth 30 HD) available. The fertilized eggs were cultured in different vessels of size ranging from 1 litre to 80 litres containing filtered ultraviolet treated sea water. 85% of the fertilized eggs successfully reached the straight hinge-stage in 18 to 24½ hours. The umbo stage was reached in 11 days, provided *Chlorella salina* was supplied as food. Antibiotics were used to control other organisms developing in the culture and multivitamins as a source of nutritional supplement.

INTRODUCTION

THE DEVELOPMENT of a suitable hatchery for the artificial propagation of the edible oyster *Crassostrea madrasensis* has been necessitated in recent years. Loosanoff and Davis (1963) in mid 1940s modified Well's techniques and standardised a method to culture the American oyster larvae to metamorphosis and later developed methods for to a full fledged oyster hatchery including induced breeding. No successful work on the hatchery production of Indian edible oyster *C. madrasensis* has so far been done. The only available literature in this field is that of Devanesen (1955) who has made certain observations on the early developmental stages of *Ostrea madrasensis* (= *C. madrasensis*.) The present work is an attempt to induce the oyster *C. madrasensis* to breed in the laboratory and to rear the larvae.

* Present address : Department of Zoology, Christian College, Kattakada, Trivandrum-695 572.

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MATERIAL AND METHODS

Adult oysters were selected from the Tuticorin oyster farm, cleaned and placed in 3 litre glass troughs. Temperature and salinity manipulation to induce the oysters to breed in the laboratory proved ineffective. As an alternative stripping of oysters was tried with success. The right valve of the oyster was removed and the content of the gonad was taken out using a fine pipette. The gonadal content was examined under microscope to determine the sex as well as maturity. Round spherical eggs and actively moving sperms alone

were chosen. The gametes were separately diluted in filtered sea water and allowed to stand undisturbed for 20 minutes. By this time the eggs settled down and that helped to eliminate to a great extent the tissue fluids and tissue pieces which form a source of contamination in the culture. The sedimented eggs were separated and mixed with sperms in a petri dish of 15 cm diameter kept shaking to keep the eggs in suspension so as to give good percentage of fertilization and uniform development. Immediately after fertilization but before the commencement of the cleavage, the eggs were centrifuged twice at 1000 rpm, for one minute and the supernatant containing excess sperms and tissue fluids were eliminated.

Vessels of different sizes ranging from 1 litre conical flasks to large cylindrical plastic bins of 80 litres capacity were used for larval culture. For experimental work transparent wide-mouthed plastic bottles of 1 litre capacity were found to be the best suited. Sea water was filtered through cotton wool and subjected to ultraviolet treatment. The required quantities of antibiotics such as penicillin and streptomycin were added.

The fertilized eggs were transferred to the plastic bottles at a density of 1,80,000 eggs/500 ml of sea water. The first change of water was done 25 hours after fertilization and subsequently once in two days. Water was aerated each day for 10 minutes.

Unicellular algae were cultured in conical flasks of size ranging from ¼ litre to 5 litres using Miquel's medium (modified by Ketchum and Redfield) and Erdscriber medium (Gross, 1937). The prevent contamination, the algal food was subjected to ultraviolet treatment prior to being supplied as food.

Narcotization and preservation of the larvae

To study the development of the zygote, cleavage pattern, larval movement and larval structure, samples were taken from the culture and observed under microscope continuously for the first 25 hours. For examination, the larvae were narcotized with menthol and made transparent with glycerol. Neutral red and Rose Bengal were used as stains. Whole mounts were used for photographing.

Observations

Mature female oysters are characterised by the presence of round spherical eggs whereas the immature ones by oval or flask shaped eggs (Pl. I A). The ripe spherical eggs are characterised by a round nuclear region (29 to 33 μ diameter) in the centre surrounded by granular cytoplasm (Pl. I B). The size of the mature eggs range from 49 to 59 μ in diameter; the average being 54 μ. The average dimension of the immature eggs is 43 μ broad and 79 μ long. The fertilized egg contracted and assumed a globular shape if it was not round before. The lighter nuclear region disappeared (Pl. I C).

In 30 minutes after fertilization (Table 1) the polar body is formed (Pl. I D). Then the first cleavage divides the zygote meridianally into two unequal cells representing the anterior and posterior ends of the embryo (Pl. II A). The plane of the second division is at right angles to the first. Both the blastomeres divide synchronously and separate into four quadrates (Pl. II B, C). Further ensuring cleavages result in the multicellular stage (Pl. II D). The embryo later reaches the blastula stage and starts rotating and swimming by means of cilia developed on its surface. In 5½ to 6½ hours after fertilization the gastrula stage is reached. This is followed by the trochophore stage. The shell gland begins to secrete the shell and by 18 to 24½ hours 90% of the fertilized eggs reach the straight hinge or the D-type larvae (Fig. 1 a, b). The average dimension of the larvae is 63 × 49 μ. The colour of the larvae in the area of the digestive

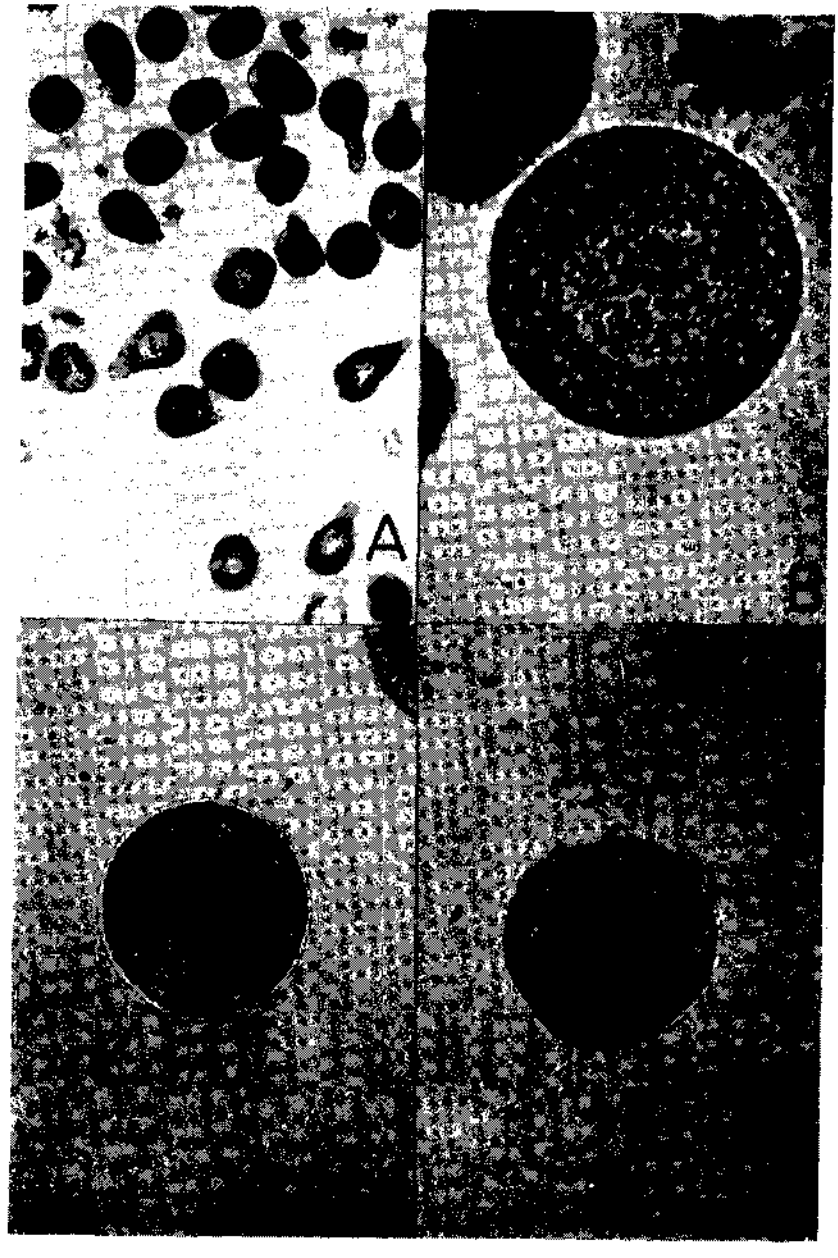

PLATE I. A. Mixture of mature and immature eggs stripped from a female oyster, B. Mature unfertilized egg enlarged, C. Egg immediately after fertilization and D. Formation of polar body.

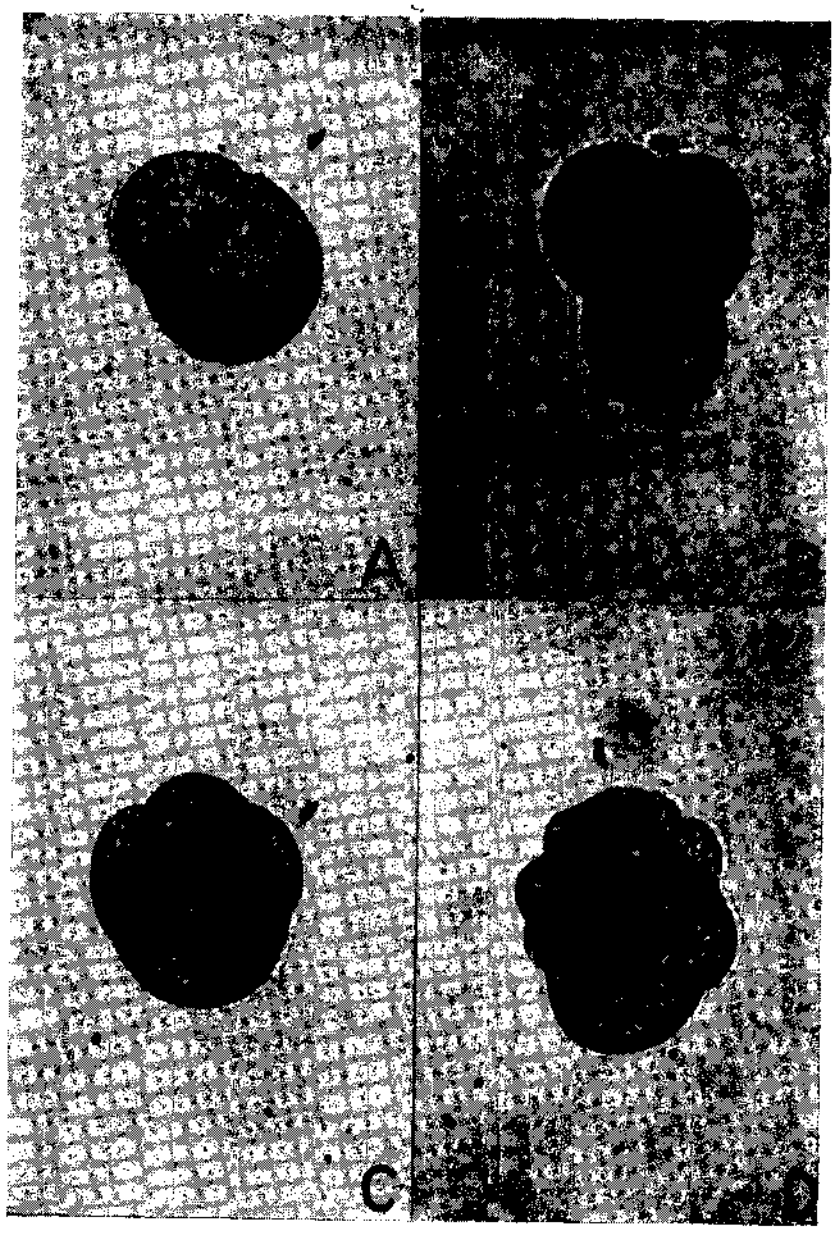

PLATE II. Different cleavage stages of the oyster egg : A. 1st cleavage, B. Commencement of 2nd cleavage, C. 2nd cleavage completed and D. multicelled stage.

TABLE 1. *Course of development and the time required for the artificially fertilized eggs of* C. madrasensis *to reach the different developmental stages*

Course of development	Time after fertilization
Release of polar body	30 m
Ist cleavage	40 ,,
2nd cleavage	50 ,,
3rd cleavage	60 ,,
Multicelled stage	1 h 10 m to 2 h
Blastula stage	1 ,, 50 ,, to 3 ,,
Rotating & Swimming stage	3 ,, 50 ,, to 5 ,,
Gastrula	5 ,, 50 ,, to 6 ,, 30 m
Trochophore	6 ,, 30 ,, to 13 ,, 50 ,,
Straight hinge stage	18 ,, 00 ,, to 24 ,, 30 ,,
Umbo stage	11 days

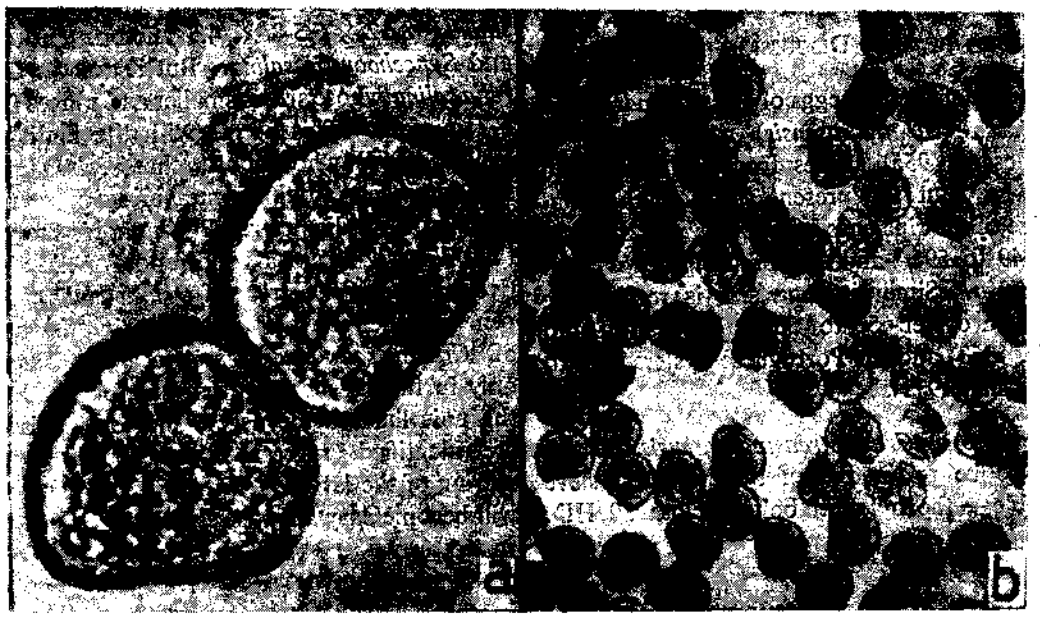

FIG. 1. a. Straight-hinge stage (the D-type larvae) with the velum expanded and b. Straight hinge larvae as seen at the upper layers of the culture medium.

diverticulum is golden brown. But the colour as a whole is usually pinkish orange.

The rate of fertilization was 91.5 to 95.2% and 85% ultimately reached the straight hinge state. *Tetraselmis gracilis* and *Synechocystis salina* proved to be too big for the larvae to feed. In one experiment when *Chlorella* was supplied as food the larvae developed to umbo stage in 11 days.

During the initial experiments the larvae did not survive beyond 24 hours. This was due to multiplication of protozoans. Series of experiments conducted to ascertain the optimum concentration of antibiotics required to effectively control the unwanted organisms revealed that 37.5 ppm of penicillin or 50 ppm of streptomycin or 75 ppm of chloromphenicol is effective. Of these the use of chloromphenicol and streptomycin separately or in combination gave better result than the use of penicillin. However, the larvae did not develop beyond straight hinge stage in most of the experiments.

Discussion

The size of the ripe eggs of *Ostrea madrasensis* (= *Crassostrea madrasensis*) as reported by Devanesen (1955) ranges from 51 to 85 μ in diameter. But the present study reveals that the size of the ripe eggs of *C. madrasensis* ranges from 49 to 59 μ and that of unripe eggs from 43 to 79 μ. Similarity egg size is reported in American oyster - 50 to 55 μ (Loosanoff and Davis, 1963) and in Portuguese oysters - 50 to 58 μ (Imai, 1971).

The eggs of *Crassostrea madrasensis* are too small to be retained even by the finest sieve available — the nylobolt bolting cloth 30 HD. Loosanoff and Davis (1963) experienced the same difficulty with the eggs of *Crassostrea virginica*. They devised the method of 'sedimentation' to partially free the eggs from body fluids, sperms and other contamination. In the present work it is found that centrifuging the eggs is the best alternative to wash and filter the eggs in the absence of a suitable sieve.

Larval development of *Crassostrea madrasensis* follows the usual pattern observed in other oysters. The larvae reached the shelled stage as early as 18 to 24½ hours with a shell length of 63 μ. This more or less agrees with Loosanoff and Davis (1963) who reported that the larvae of *Crassostrea virginica* reached the shelled stage in 24 hours with a shell length ranging from 68 to 75 μ. Imai (1971) record that in portuguese oysters the larvae reach the shelled stage with a shell length of 75 μ in 24 hours

The acceptance of the algae as food depends mainly on their cell size. The larvae of *Crassostrea madrasensis* do not accept *Tetraselmis salina* (8-12 × 6.5-8 × 4.5 microns in size) and *Synechocystis salina*. But it accepts *Chlorella salina* whose size is smaller and ranges from 5-7 microns. This agrees with the finding of Walne (1974) who reports that *Ostrea edulis* fails to ingest anything measuring more than 10 microns in diameter. But he holds the view that *Chlorella* is not a good food, for the digestive enzymes of the larvae of *Ostrea edulis* cannot penetrate the cell wall. It is seen that the larvae of *Crassostrea madrasensis* accept *Chlorella* only in the absence of other suitable food. If the unicellular algae acceptable for the larvae are identified, segregated and cultured, the rearing of the larvae of *Crassostrea madrasensis* will become easy and successful.

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PREDATION OF OYSTER *CRASSOSTREA MADRASENSIS* BY GASTROPOD *CYMATIUM CINGULATUM* (LAMARCK) IN THE OYSTER FARM AT TUTICORIN

R. THANGAVELU AND P. MUTHIAH

Central Marine Fisheries Research Institute, Cochin-682 018

ABSTRACT

One of the essential problems in perfecting the technology of oyster farming is to identify and eliminate the predators causing periodical mortality of oyster in the culture system and to suggest adequate control measures. At Tuticorin it has been observed that the gastropod belonging to the species *Cymatium cingulatum* causes considerable damage to the stock in the oyster farm especially when the oysters are 3 to 4 months old.

Observations on the feeding method and behaviour of the predator are given. Control measures for saving the farm stock are also discussed.

INTRODUCTION

IN AQUACULTURE practices all over the world, culturists are vary often confronted with large-scale mortality of the tended stock, because of predation. This results in economic loss to the farmer. Therefore, while perfecting the technology of growing it is absolutely essential to identify the agents causing such destruction, document them and develop suitable measures to control them effectively.

The known principal predators of oysters are flatworms, drills, starfishes, crabs, fishes and also few birds (Galtsoff, 1964). Since bottom culture of oysters as practised in America, Canada and France suffers from frequent attacks by predators in the farm, the off-bottom culture has the distinct advantage of eliminating predatory animals to a very great extent. Depending on the nature of predators and seasons of occurrence several control measures like periodical mopping and removal of predators, treatment of oyster farm area with chemicals and manual removal of predators have been developed for bottom culture. This point was kept in mind, while attempting experimental oyster culture in the tidal creek and shallow open sea coast at Tuticorin, adopting the rack and tray method. During the course of the experiments during 1977 to 1979 predation occurred only during the year 1978, while attempting large scale culture of oysters along coast. During July to November, large scale invasion of gastropods belonging to the triton group was noticed in the box-type cages where oyster seeds were initially reared. Several oyster seeds ranging from 25 to 85 mm were found dead, their flesh having been totally eaten away. Many live gastropods were found inside the cage suggesting that the mortality would have been caused by these gastropods. Detailed investigation was undertaken to study the cause, the nature and extent of predation. Effective means of combating the menace were also tried. This paper presents the results of the above observations.

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Farm Site

The nature of bottom in farm area is muddy sand with sparse growth of algae and weeds at the depth varying from 1.5 to 2.7 m. The farm is protected from wind and wave action by a sandy shore arm extending in the sea on the North-eastern region of the farm. The salinity in the area ranges from 31.7‰ to 35.6‰. The surface temperature varies from 25.4°C to 30.4°C. Moderately turbid conditions prevail during spring tides.

General Characters of *Cymatium cingulatum* (Lamarck)

All gastropods collected belonged to the species *Cymatium cingulatum* (Lamarck). (Tentative identification of this shell made earlier as *C. pileare* was incorrect).

Description

Commonly known as Poulson's triton, the shells (Plate I A) varied in size from 10 to 76 mm, light weight with 18 flattened spiral cords. In some, the spiral cord at the angular shoulder is knobbed. Former varices are rare ; aperture large ; columella smooth ; outerlip crenulate. The protoconch is persistent as a small smooth spiral structure. Colour yellowish to brown ; periostracum thin, hairy and flakes off when dried.

Mode of feeding

Feeding mechanism involved the muscular action of the foot and the tube shaped extensible proboscis which is an extension of the head region lying just above the foot. Fully extended proboscis is about 3/4th of the height of the shell and it can be pulled back and ensheathed. The radula is attached to the floor of the buccal cavity at the distal end of the proboscis which consists of a ribbon-shaped chitinous band covered with regular rows of many small, fairly hard teeth. The radula is of 'taeno glossate' type. In normal adult *C. cingulatum* ranging from 21.1 mm to 72.5 mm in shell length, the radula varies approximately 2.07 to 5.35 mm in length and from 247 μ to 599.9 μ in width. Regular dimensions vary widely among different individuals of the same shell height.

While feeding the gastropod climbs upon the oyster's upper shell or flat valve, manoeuvers into position with its broad foot and rests upon the posterior margin of the shell with its foot firmly pressed on the edge of the shell. The proboscis extremely pliant and flexible, is inserted in the gape between valves while the oyster starts filtering water. The radula and its associated musculature ensheathed in the buccal floor of the proboscis is extended forward and slowly rasps the mantle portion of the oyster. The acidic fluid which is secreted by the proboscis gland appears to help in paralysing the animal as a result the shell valves remain agape to enable the whole fleshy portion to be destroyed by the predator. This method of feeding is identical to that reported in *Busycon* (Carriker, 1951 ; Magalhaes, 1948) and *Murex fulvescens* (Wells, 1958).

Incidence of predation and mortality rate

Oyster spat settled on lime-coated tiles kept during April-May 1978 for spat collection were taken out and scraped during June. About 3 lakh spat of size 20 to 35 mm were kept in box-type cages and suspended from the racks for initial rearing. In August 1978, oysters above 50 mm size were sorted out and put in rectangular cages for further rearing. During this thinning operation, large number of dead oysters was detected inside the cages. Several live *Cymatium cingulatum* were also found crawling amongst dead shells. The season of

occurrence of this extended upto November 1978 and in all 206 numbers were collected and removed. A total of 39,450 dead oysters were counted. The total mortality was found to be 13% of which 8.91% was during September 1978 and 4.09% in October 1978.

Period and size of occurrence

From Table 1, it may be seen that a total of 206 gastropods were collected in majority of them occurring in September 1978 with a model size group of 55 to 60 mm. The incidence was considerably less in November 1978. Although the animal of size 17 mm in height was recorded during July, the incidence was high in August and September 1979 diminishing thereafter.

A search for simultaneous occurrence of this gastropod in the sea bed surrounding the farm area and the nearby Hare Island area was carried out both by diving and dredging but no live animal was noticed or collected. A single specimen was collected in the far off Tuticorin Harbour break water wall area indicating that they may be expected to occur in the hard rocky bottom area in deeper off-shore regions.

Rate of feeding and size of oysters preyed

Experiments were conducted by keeping 100, 90 and 75 oysters separately in three box-type cages along with two *Cymatium* of size 60 to 67 mm in each cage. Regular observations were made and after an interval of 15 days the number of oysters with empty shells removed were 60, 57 and 53 in cages I, II and III respectively (Table 2). The average rate of feeding was calculated as 1.88 oysters/conch/day. This rate of feeding is more when compared to *Thais* 0.5 oyster/month ; *Urosalpinx* 0.332 to 0.481 oyster/day (Cole, 1942, 1951) and *Murex fulvescens* 3.5 oysters/week (Wells, 1958). *Marula marginella* is known to feed at the rate of 3 spat oysters/12 hours (Thomson, 1968).

In order to find out the size of oysters attacked by *C. cingulatum* a sample of 156, among the dead oyster shells was removed from the cages and measured. The dead shells ranged from 25 to 85 mm. The modal size of oysters killed was 53.3 mm. Nearly 75% of mortality was caused in the size group of 40-65 mm (Table 3).

TABLE 2. *Oysters kept in* 3 *cages with* 2 C. cingulatum *in each at the start of the observation and their fate at the end of* 18 *days*

Cage No.	Oysters quantity	Number of oysters preyed at the end	Rate of feeding
1 ..	100	60	2 oysters/conch/day
2 ..	90	57	1.9 ,,
3 ..	75	53	1.76 ,,
Average rate of feeding			1.88 ,,

TABLE 3. *Showing details of size of oysters killed by* C. cingulatum

Size of oysters	Frequency	Percentage
25-30 ..	2	1.28
30-35 ..	7	4.49
35-40 ..	12	7.69
40-45 ..	21	13.46
45-50 ..	20	12.82
50-55 ..	32	20.52
55-60 ..	26	16.67
60-65 ..	17	10.90
65-70 ..	12	7.69
70-75 ..	4	2.56
75-80 ..	1	2.64
80-85 ..	2	1.28

Preventive measures

Loosanoff *et al.* (1960 a, b) reported that chlorinated benzene is toxic to drills. Mackenzie (1970) standardized the polystream (a mixture of polychlorinated benzenes) treatment at the rate of 9.5 kl/ha. Loosanoff (1957) indi-

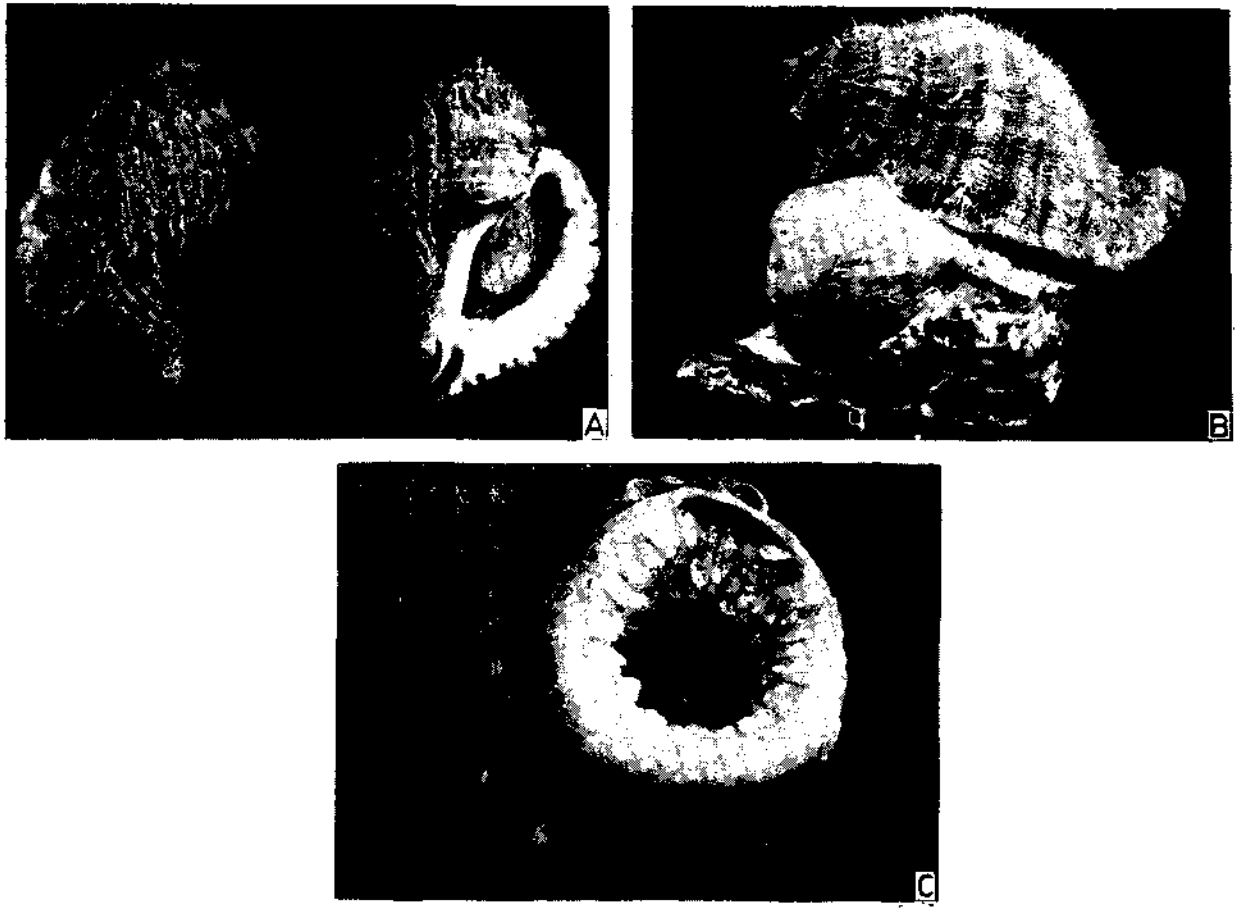

PLATE I A. Dorsal and ventral view of *Cymatium cingulatum*. B. Egg case and gastropod on the shell of oyster and C. Gelatinous egg capsule of gastropod.

TABLE 1. *Size distribution of* C. cingulatum *collected during* 1978 *and* 1979

	1978							1979						
Size group	July	Aug.	Sept.	Oct.	Nov.	Dec.	Total	July	Aug.	Sept.	Oct.	Nov.	Dec.	Total
10-15	—	—	—	—	—	—	—	—	2	—	—	—	—	2
15-20	—	—	1	1	—	—	2	1	9	—	—	—	—	10
20-25	—	—	—	—	—	—	—	—	8	7	1	2	—	18
25-30	—	—	1	—	—	—	1	—	9	11	1	2	—	23
30-35	—	—	4	—	—	—	4	—	10	12	1	1	—	24
35-40	—	—	3	—	1	—	4	—	15	8	1	2	—	26
40-45	—	—	6	4	—	—	10	—	7	12	2	1	—	22
45-50	—	—	9	—	2	—	11	—	5	12	6	—	—	23
50-55	—	—	21	8	—	—	29	—	4	14	2	1	—	21
55-60	—	—	26	14	2	—	42	—	—	5	2	—	—	7
60-65	—	—	24	20	6	—	50	—	1	4	1	2	—	8
65-70	—	—	13	22	2	—	37	—	—	—	—	1	—	1
70-75	—	—	7	6	2	—	15	—	—	—	2	—	—	2
75-80	—	—	1	—	—	—	1	—	—	—	—	—	—	—
Total	—	—	116	75	15	—	206	1	70	85	19	12	—	187

cated that a saturated salt solution may be helpful, in controlling drill population, by killing their eggs and embryos, while still in egg cases. Hand picking, though laborious appeared to be more efficient especially when carried through before egg laying begins. Treatment method by employing chemical does not appear to be practical in an extensive farm area also.

Fecundity and development of Cymatium cingulatum

Sexes are separate. Eight individuals of size above 62 mm in shell length, with egg cases were collected during September 1978. Each egg case with the follicles was found attached to dead oyster shell (Plate I B). The average height of the egg case was 24.9 mm with a diameter of 47.2 mm. Each egg case (Plate I C) contained 271 follicles covered by a gelatinous sheath of light brown colour. The follicles were arranged in rosette and with the average length and width of the follicle was 10.8 mm and 2.9 mm respectively. Each follicle contained, on an average 1278 eggs.

With a view to make some observations on the development of the egg capsules, one of them was removed and kept in an aquarium tank containing filtered sea water taken from the same locality. Soon after they were brought to the aquarium tank a few follicles were cut open carefully, eggs were removed and examined. It was observed that cleavage had already taken place. After one hour, formation of vertical cleavage was observed. Micromeres and macromeres appeared after four hours and gastrulation occurred at the end of 24 hours. Hatching of early veliger stage took place at the end of 44 hours with rudiments of the velar lobes and foot (Fig. 1 a to e). Similar pattern of development was noticed for *Thais haemastoma* by Burkenroad (1931).

Fully developed veliger was found after 72 hours with an egg shaped shell. At this stage liver lobes and velum with distinct cilia were well developed, whereas sub-velum was poorly developed. Foot was found ventral to the velar lobes, beneath which operculum was noticed. At this stage mouth, oesophagus, stomach, intestine and anus could be recognised (Fig. 1 f to h). At the end of the 4th day of hatching, the larvae were found to settle at the bottom and the velum was found very much reduced. Further developmental studies were not possible. The early development of this gastropod is similar to that of the nudibranch *Cuthona adayarensis* described by Rao (1961).

Discussion

Of the principal predators of oysters, flatworms *Stylochus* and *Pseudostylochus* commonly known as ' oyster leeches ' inflict serious damage to oyster population. *Pseudostylochus ostreophagus* was reported to cause mortality from 6 to 42% among the imported Japanese seed oysters (Hyman, 1955). Lunz (1947) reported that blue crabs *Callinectes spadius* destroyed more than 80% of the young oysters by cracking their shells at Wadmalaw Island, United States. The mud and ghost shrimps cause serious harm to the ground by making it too soft. *Cliona celata* and *Polydora ciliata* which bore the oyster shell are quite serious pests. The predation of starfishes *Asterias forbesi*, *Piaster* sp. *Evasterias* sp. and *Pycnopodia* sp. on oysters is considerable (Galtsoff and Loosanoff, 1939). Large rays *Trygon pastinaea* and *Myliobates aquila* are known for their predation on oysters. In Stephen's Bay, it was reported that porcupinefish *Dictylichthys mysersi* and bream *Sphaeriodes* are the predatory fishes (Korringa, 1976). Incidence of predation by the above enemies was not noticed in the oyster farm at Tuticorin.

Prosobranchiate gastropods appear to be the principal enemies of oysters. General information about oyster drill *Urosalpinx cinerea* has

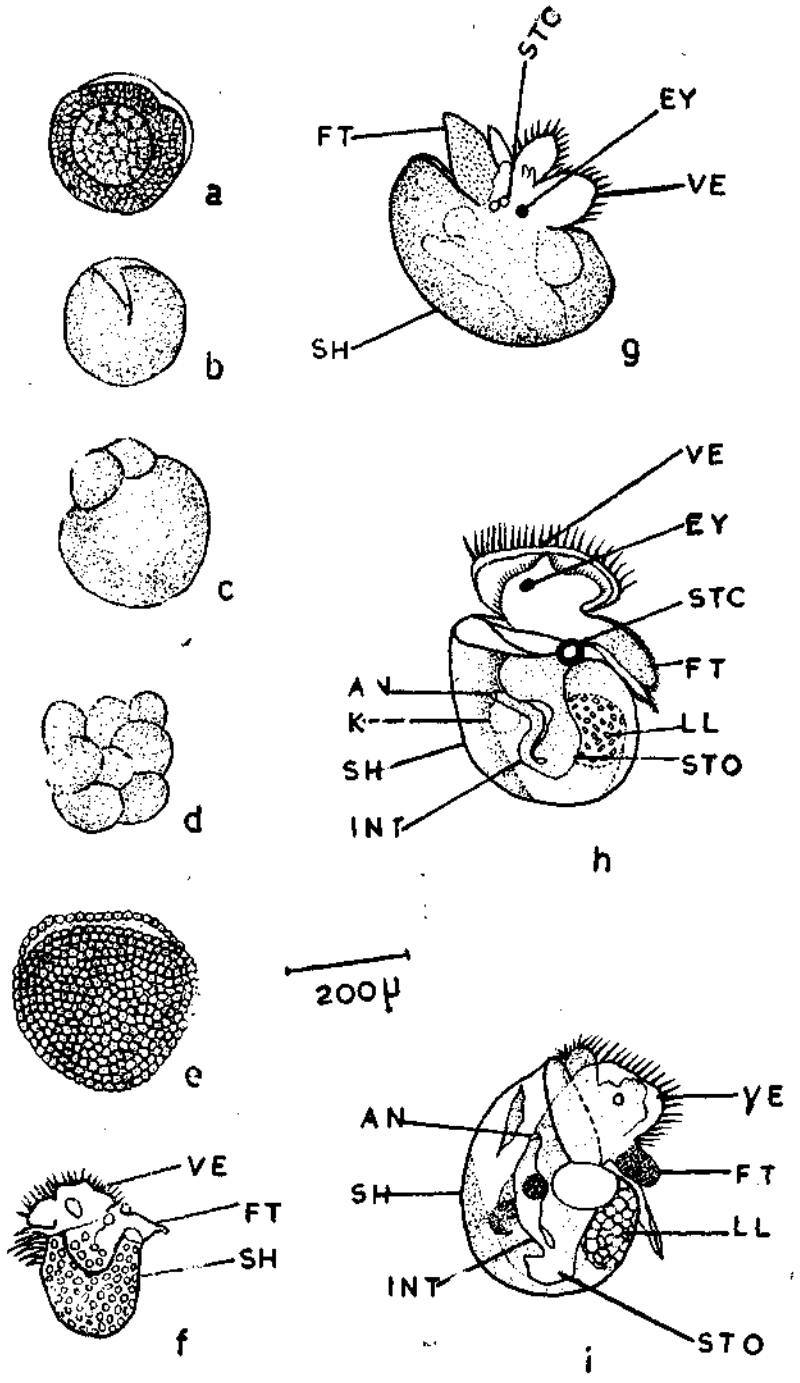

FIG. 1. Developmental stages of *Cymatium cingulatum* (Lamarck) : a. Fertilised egg ; b. Two-cell stage ; c. Four cell stage ; d. Eight cell stage ; e. Gastrula with blasto pore ; f & g. Early veliger ; h. Fully formed veliger and i. Larva at the time of creeping (Sh.—Shell ; Stc—Statocyst ; Ey—eye ; Ve—velum ; Ft.—Foot ; Il.—Liver lobe ; Sto—Stomach ; An—Anus ; Int—intestine ; K—excretory organ).

been given by Carriker (1955) for North American populations and by Hancock (1959) for those of the channel coast of England. *Thais haemastoma* in South Atlantic and Gulf States ; *Ocenebra japonica, Rapana thomasiana* and *Thais fumulosa elavigera* in Japanese waters are the common predators. The common whelk *Buccinium undatum* and dogwhelk *Nucella lapillus* are also known to attack oysters occasionally (Korringa, 1952, 1976).

Apart from the instances of boring on oysters by *Thais rudolphi* near the mouth of Athankarai Estuary (CMFRI, 1974) no major predation of oysters by the prosobranchiates has been so far reported in India. Thus predatory mortality of oysters caused by *C. cingulatum* has been reported for the first time in east coast of India.

In the present studies the percentage of mortality caused by *C. cingulatum* appeared to be less when compared to 30% death caused by drills in Atlantic Coast (Adams, 1947), 75% in Essex oyster beds (Cole, 1962) and 15.6 to 22.6% by Galtsoff (1964). It is however to be admitted that the entire studies now were confined to laboratory conditions and need more confirmation.

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NEUROSECRETORY CONTROL OF REPRODUCTION IN THE OYSTER *CRASSOSTREA CUCULLATA*

R. NAGABHUSHANAM

Department of Zoology, Marathwada University, Aurangabad-431 004

ABSTRACT

Morphology of the neurosecretory cells in the rock oyster *Crassostrea cucullata* was studied in detail. On the basis of size, shape and staining properties two types of neurosecretory cells were distinguished from the dorsal surface of the cerebral and visceral ganglia. *Cell Type I* is pyriform in shape and about 15 to 22 μ in length. The nucleus is oval or round and its diameter varies from 3 to 5 μ. Vacuoles are absent. The neurosecretory material stained red with mallory's and blue-black with CHP stain. *Cell Type II* is oval or round in shape and about 6 to 8 μ in diameter. The nucleus is round and its diameter varies from 4 to 5 μ. Vacuoles are absent. The secretory material stained faint-red with Mallory's but negative with CHP stain. Type I cells showed a distinct annual cycle of secretory activity which has been correlated with reproductive cycle. Thus Type I cells may exert an influence on the gonads. The ablation of cerebral ganglia hastened the spawning reaction.

INTRODUCTION

AMONG the lamellibranchs, neurosecretory phenomena have been studied by a few investigators. Gabe (1955) reported the presence of neurosecretory cells in twenty species of lamellibranchs. Lubet (1955, 1956) found a definite correlation between neurosecretory activity and reproductive cycle in the mussel *Mytilus edulis* and in the clam *Chlamys varia*. Baranyi (1964) observed seasonal changes in the neurosecretory cycle of the mussel *Anodonta cygnea*. The occurrence of neurosecretory cells among the Indian bivalves was reported by Nagabhushanam (1968 a, b) in *Martesia striata*, Nagabhushanam and Lomte (1972) in *Parreysia corrugata*, Deshmuk in (1972) in *Meretrix meretrix* and Nagabhushanam and Mane (1973) in *Katelysia opima*. The present study was undertaken with the view to know the role of neurosecretion in reproduction in *Crassostrea cucllata*.

MATERIAL AND METHODS

The specimens of *Crassostrea cucullata* used in the present study were obtained from the Shirgaon creek near Ratnagiri. Collections were made at monthly intervals from January to December, 1973. Oysters between the size group 35-40 mm in shell length were used for the study. Immediately after collection they were brought to the laboratory, the shells were opened and the soft parts were fixed in Bouin's fluid for 24 hours. The cerebral and visceral ganglia were removed, dehydrated in alcohol, cleaned in xylol and then fixed in paraffin wax. Serial sections were cut at 6 to 8 μ in thickness and stained with Mallory's triple stain and Gomori's chrome-haematoxylin-phloxime stain (CHP).

The extirpation experiments on cerebral and visceral ganglia were carried out in adult and normal sized oysters. To study the effect of ablation of ganglia on reproduction the gonads of the operated oysters were fixed in Bouin's fluid and the slides were prepared for the histological studies.

RESULTS

Neurosecretory cells

The histological study on cerebral and visceral ganglia stained with Mallory's triple stain and

CHP revealed the presence of groups of cells which are comparatively large than and cytologically different from the ordinary ganglia cells. Most of these large cells contain large nuclei and dense cytoplasm; their perikarya and axos are filled with fine secretory droplets. Mallory's stain was found to be the best for staining the neurosecretory material of the neurosecretory cells. The neurosecretory cells possess the characteristic staining properties (Scharrer and Scharrer, 1945).

Two types of neurosecretory cells have been identified on the dorsal surface of the cerebral and visceral ganglia of the oyster *Crassostrea cucullata* and are designated as Cell Type I and Cell Type II. The principal criteria in distinguishing the cell types used were size, general shape of the cell body, presence or absence of vacuoles in the cytoplasm and staining property of the secretory material.

CELL TYPE I: These cell contain dense cytoplasm which stain heavily with Mallory's triple stain. The cells are pyriform in shape but are sometimes irregular. They measure about 15 to 22 μ in length and 6 to 9 μ in width. The nucleus is round/or oval with a diameter of 3 to 5 μ; it may be either central or eccentric in position. The nucleus generally contains one large nucleolus which conspicuously stained with orange G and the diameter varies from 0.5 to 1.0 μ. The secretory material in the cytoplasm stained red with Mallory's and blue-black with CHP. In some sections secretory material appeared as extremely fine granules. Furthermore, all the cells did not show the presence of larger amount of secretory material at any one time. Apparently some cells are at the peak of their secretory process while others are devoid of secretory granule. Vacuoles are generally absent (Table 1).

In some neurosecretory cells, secretory material could be seen in the cell body and the axon. The secretory material in the part of the axon near the cell body appeared as small granules. The secretory material, however, lost its special character when the axon reached the central neuropile of the ganglion.

TABLE 1. *Characteristics of neurosecretory Cell Types I and II present in the central nervous system of* Crassostrea cucullata

Description	Cell Type I	Cell Type II
Shape of cell body	Pyriform or irregular	Oval
Size of cell body	15 to 22 μ (length) 6 to 9 μ (width),	6 to 8 μ (diameter)
Staining reaction :		
Mallory's stain	Red	Faint red
Gomori's stain	Blue-black	Negative
Nucleus	Round or oval (3 to 5 μ diameter)	Round (4 to 5 μ diameter)
Vacuoles	Absent	Present

CELL TYPE II: The Type II cells are rather smaller than Type I cells and oval in shape with a diameter of 6 to 8 μ. Their nuclei are similar to those of Type I cells and measure about 4 to 5 μ in diameter, generaly there is one large nucleolous but occasionally two nucleoli appeared. The secretory material appeared in the form of small granules, stained faint red with Mallory's stain and negative with CHP. The vacuolisation of these cell is very striking and they do not possess a characteristic shape. Occasionally very fine particles are observed in the vacuole.

A comparison is presented in Table 1 of the significant dimensions and tinctorial properties of the two cell types.

Role of neurosecretion in reproduction

To find out the role of neurosecretion in reproduction, the number of Type I and II

neurosecretory cells containing secretory materials were counted every month throughout the period of study and the average results obtained from the ten oysters per month are given in Table 2. The number of Type I cells that contained droplets of secretory material showed a distinct seasonal fluctuation. However, no appreciable change was noticed in the number of active Cell Type II throughout the year.

It can be evident from Table 2 that from February to June, the number of Type I cell containing secretory materials has slowly increased and from July to September maximum number of cells were observed to contain heavily stained cytoplasm, thus showing a great accumulation of secretory product throughout the mature period of the oyster. It has been observed that gametogenetic and maturation periods in *Crassostrea cucullata* were from June and July to September. The spawning period was observed from October to January where the number of Type I cells were found to be reduced at a fairly low level. Thus, the exists a close relationship between the hormonal regulation of cerebral ganglia with the reproductive activity of the oyster.

TABLE 2. *Variation in the number of the Cell Types I and II containing secretory material in cerebral and visceral ganglia of* Crassostrea cucullata *(Figures in the Table represent average of ten oysters). Besides, condition of gonad is also shown*

Year and month	Number of Cell Type I	Number of Cell Type II	Condition of gonad
1973			
January	9	21	Spawning period
February	14	29	Recovery period
March	13	32	Recovery period
April	19	25	Gametogenesis period
May	16	33	Gametogenesis period
June	20	31	Gametogenesis period
July	27	30	Mature period
August	36	28	Mature period
September	31	29	Mature period
October	16	31	Spawning period
November	5	24	Spawning period
December	3	27	Spawning period

Effect of ganglia ablation on spawning behaviour

To see the effect of cerebral as well as visceralectomy on the spawning behaviour of the oyster *Crassostrea cucullata*, experiments were conducted using fully ripe males and females, during August 1973. After ablation and acclimation of such oysters, the gonads were removed and fixed in Bouin's fluid. These gonads were studied histologically to see whether there was any partial spawning of gonads. The results are presented in Table 3.

TABLE 3. *Effect of cerebral and visceral-electomy on spawning reactions in* Crassostrea cucullata

Treatment	Sex of oysters	No. of oysters tested	No. of oysters partially spawned	No. of oysters not responded
Ablation of cerebral ganglia	Female	20	14	6
	Male	20	12	8
Ablation of visceral ganglia	Female	20	3	17
	Male	20	2	18
Control	Female	20	1	19
	Male	20	—	20

Thus, from the Table, it can be revealed that out of 20 cerebralectomized female oysters, 14 have partially discharged the eggs while 12 out of 200 cerebralectomised male oysters, partially discharged the gametes. However, it has been observed that the gonads in visceralectomized male and female oysters did not show much spawning reactions compared to the controls. Apart from this it has been further observed that the female oysters were slightly more responsive towards the cerebralectomy than the male oysters.

Discussion

The nervous system and hormonal apparatus are not as sharply separated in invertebrates as they are in vertebrates and no endocrine glands have so far been encountered in bivalves. The presence of neurosecretory cells in the cerebral and visceral ganglia of many lamellibranchs was recognised by several investigators. It appeared from their studies that the number and location of the neurosecretory cells differed from species to species. Gabe (1955) suggested that the neurosecretory cells were numerous and scattered in the primitive species than in the more specialized ones where they showed a tendency to form groups. Further he observed a dense cap of neurosecretory cells in the cerebral and visceral ganglia of *Nucula nucleus*, whereas they were scattered in the corresponding ganglia of *Teredo* (Gabe and Rancurel, 1958). The neurosecretory cells were aggregated at a laterodorsal point of anterior face of the cerebral ganglion and dorsal distribution of these cells in the visceral ganglion in *Mytilus edulis* (Lubet, 1955). Anthemnusse (1963) observed that the neurosecretory cells in *Dreissena polymorpha* were concentrated in four distinct masses in the cerebral ganglion and five masses in the visceral ganglion. Even distribution of neurosecretory cells in all the three ganglia (cerebral, visceral and pedal) of *Anodonta cygnea* was observed by Baranyi and Salanki (1963). The dorsal surface location of neurosecretory cells in *Crassostrea cucullata* of the present investigation corresponds with the situation of the neurosecretory cells in *Crassostrea virginica*, *Modiolus demissus* and *Meretrix casta* (Nagabhushanam, 1963, 1965, 1968 a), *Meretrix meretrix* (Deshmukh, 1972), *Crassostrea gryphoides* (Nagabhushanam and Mantale, 1972) and *katelysia opima* (Nagabhushanam and Mane, 1973).

Several types of neurosecretory cells may occur in the nervous system of lamellibranchs (Martoja, 1972). Two types of neurosecretory cells were found in many bivalves (Lubet, 1955, 1956; Fahrmann, 1961; Nagabhushanam, 1970; Deshmukh, 1972; Nagabhushanam and Mantale, 1972; Nagabhushanam and Mane, 1973). However, three types of neurosecretory cells were identified by Baranyi and Salanki (1963) in *Anodonta cygnea* whereas one type of neurosecretory cell was observed by Nagabhushanam (1964) in *Modiolus demissus*. In accordance with the findings of Lubet (1955) and later workers, two types of neurosecretory cells were recognised in the cerebral and visceral ganglia of the oyster *Crassostrea cucullata*.

On the analogy of the endocrine regulations known to exist in the groups of animals, it could be inferred from the results of present study that the neurosecretory cells in the ganglia most probably control the reproductive process by means of hormonal factors. Lubet (1955, 1956) in *Mytilus edulis* and *Chlamys varia* found that the accumulation of secretory material in the pear-shaped cells of the cerebral ganglia was accomplished during the phase of gametogenesis. He further stated that the secretion was produced just before gametogenesis and was maximal at the time of gamete maturation. Just before each release of gametes (spawning) some of the neurosecretory cells emptied their secretions. Later, Lubet (1959) stated that maturation of gonads was only influenced by the pear-shaped cells and was independent of small multipolar cells and the cells of visceral ganglion. These facts have been further confirmed by Nagabhushanam (1963, 1964) in *Crassostrea virginica* and *modiolus demissus*, Deshmukh (1972) in *Meretrix meretrix* and Nagabhushanam and Mane (1973) in *katelysia opima*, where Type I cells were found to influence the gonad maturation and spawning while 'A' cells were found to stimulate the gonad maturation and spawning in *Anodanta cygnea* (Baranyi, 1964). However, it has been observed in the present study that on the onset of spawning, pyriform cells from the cerebral ganglia released their secre-

tory product which is a responsible factor for the spawning reaction in oysters. This discharge of secretory product was found to be at greater extent as the spawning reached its peak. After spawning, during gametogenesis and maturation period again the secretory products started accumulating in the pyriform cells.

In general, it was observed that the extirpation of cerebral ganglia resulted in the hastening of spawning in the bivalves. Lubet (1955, 1956) removed the cerebral ganglia in *Mytilus* and found that spawning was hastened. He, therefore, concluded that the neurosecretory material has an inhibitory effect on spawning and that only after the secretions are emptied does the bivalve become sensitive to the environmental factors that provoke spawning. In the present study, it was observed that the removal of cerebral ganglia in *Crassostrea cucullata* hastened thes pawning reactions.

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NEUROENDROCRINE CONTROL OF REPRODUCTION IN SOME EDIBLE OYSTERS (GENUS *CRASSOSTREA*)

KIRAN M. DESAI AND DHIRAJ NIMAVAT *
Department of Biosciences, South Gujarat University, Surat-395 007

ABSTRACT

Secretory activity of neurosecretory cells (NS cells) of cerebral and visceral ganglia using Aldehyde-Fuchsin (AF) and chrome-Alum-Heamatoxylin-phloxine (CAHP) staining techniques were studied. The index of the activity was found to be high at time of the gametogenesis and spawning of *C. gryphoides*, *C. belcheri*, *C. rivularis* and *Crassostrea* sp. of the Gulf of Kutch. Role of the NS cells in triggering the gametogenesis and spawning are discussed. Environmental parameters such as salinity and temperature are also found to influence the spawning of the oysters.

INTRODUCTION

IN BIVALVE molluscs the presence of neurosecretory activity in cerebral and visceral ganglia (Gabe, 1955; Lubet, 1955 a) and also in pedal ganglia (Fahrmann, 1961) has been demonstrated. Many authors have presented evidences of cyclic changes in neurosecretory activity in molluscs (Lubet, 1959, 1966 ; Nagabhushanam, 1963 ; Antheunisse, 1963 a, b; Badino and Marchionni, 1972 ; Nagabhushanam, 1964 b, c ; Desai *et al.*, 1971). Several electron microscopic studies on molluscs have reaffirmed the secretory activity of the neurosecretory cells (Boer *et al.*, 1968 ; Bonga, 1972 ; Gerschenfeld, 1963). Studies made on neurosecretory cells of gastropods have indicated influence of neurosecretory cells over physiological activity like spawning, gonadial maturation (Gabe, 1955 ; Lubet, 1955 b, 1959 ; Nagabhushanam, 1963 ; Desai *et al.*, 1971 ; Badino and Marchionni, 1972), osmoregulation (Hekstra and Lever, 1960 ; Lever *et al.*, 1961 a ; Nagabhushanam, 1964 a ; Bonga, 1972). With a view to study the influence of neurosecretory cells over various phases of reproductive cycle of the edible oysters of Saurashtra Coast, the present investigation was undertaken.

MATERIAL AND METHODS

Oysters of species *Crassostrea rivularis*, *C. belcheri*, *C. gryphoides* and *Crassostrea* sp. maintained at Sikka in the Gulf of Kutch were studied. Six to eight specimens of each species were sacrificed by the first week of every month (January through December) of 1974. The cerebral and visceral ganglia along with surrounding tissue mass (1 mm × 1 mm) were dissected out and fixed quickly in Bouin's fluid. Paraffin sections of 5 microns were taken and stained by Bargmann's Chrome-Alum-Hematoxylin-phloxine (CAHP) technique (Pearse, 1968). The neurosecretory cells of both cerebral and visceral ganglia were classified into two categories, granulated and completely degranulated, based on the histological preparations. About 200 to 300 neurosecretory

* Present address: Department of Fisheries, Government of Gujarat, Jodiya, Gujarat.

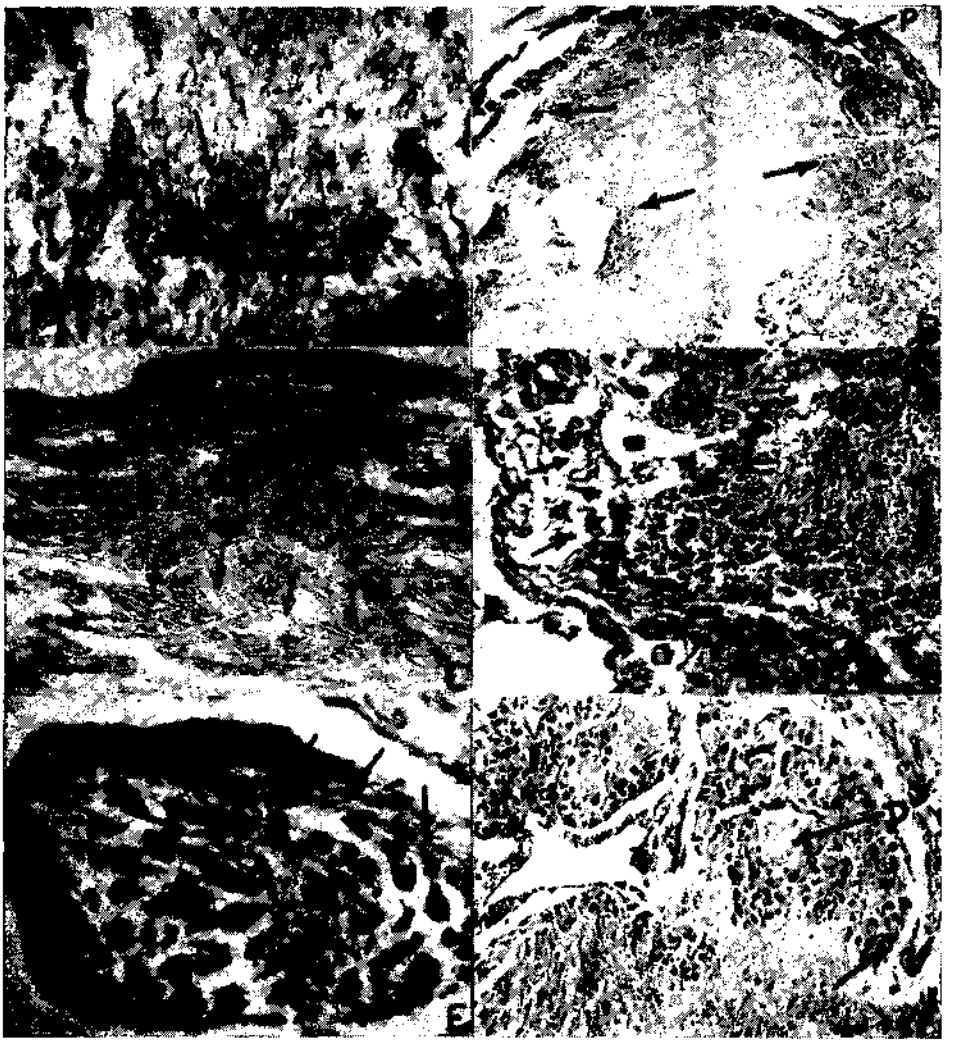

PLATE I. *Crassostrea rivularis*: A. Parasagittal section of cerebral ganglia showing peripheral distribution of the neurosecretory cells (as indicated by arrows, × 450); B and C. L. S. of visceral ganglia showing neurosecretory cells in the peripheral region (as indicated by arrow) and pathway (P) (B : × 120; C : × 450) Note: Kidney tubules in the vicinity of ganglia (as shown by arrows); D. L.S. of visceral gangli, (magnified view showing release of the neurosecretory material on the wall of kidney cells (as indicated by arrow) (× 450); E. Parasagittal section of cerebral ganglia showing neurosecretory cells and pathway full of neurosecretory material (as indicated by arrows) (× 1700) and F. Magnified view of L.S. of visceral ganglia showing neurosecretory cells with enlarged nuclei and nucleoli (as indicated by arrow) (× 1700) Note : Pathway full of neurosecretory granules degenerated and vacuolated neurosecretory cells (D). Compare with Plate II A.

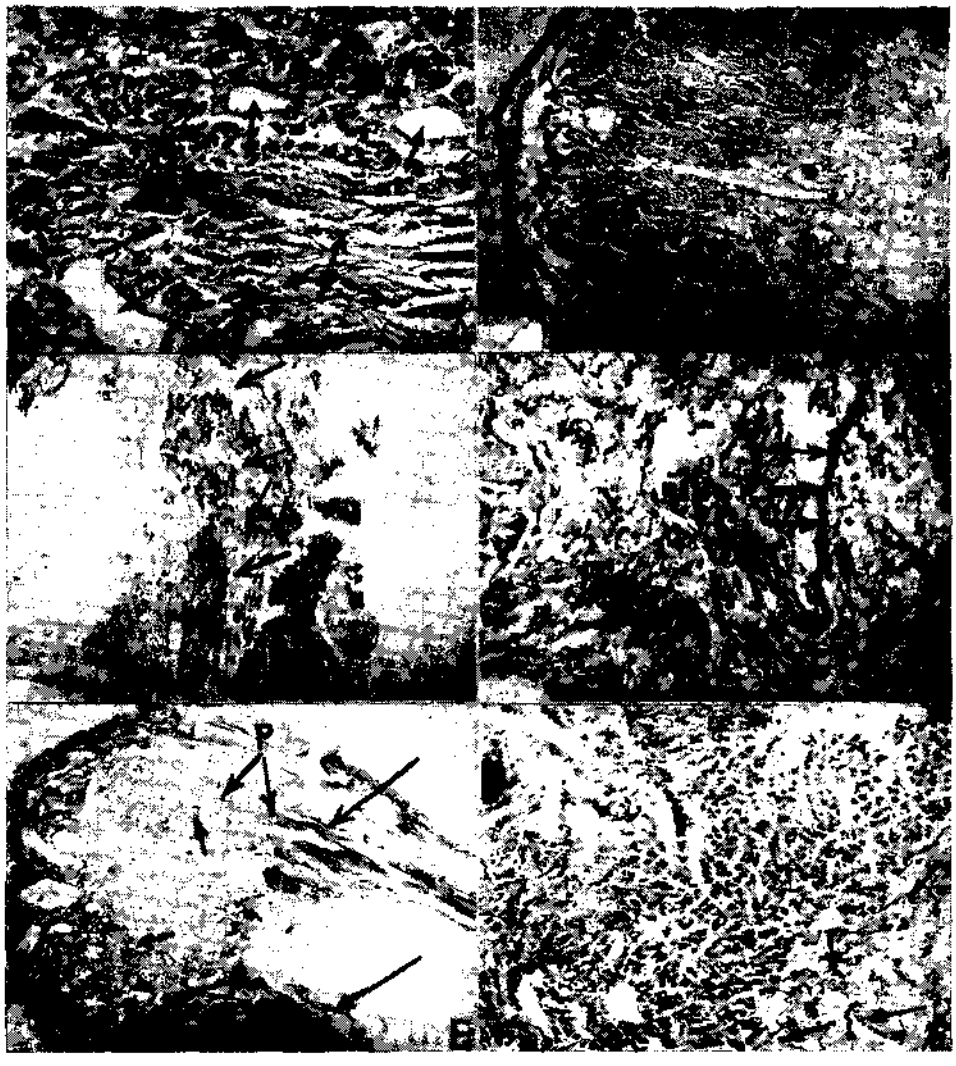

PLATE II A. L.S. of the visceral ganglia of *C. rivularis* showing lacunae in the ganglionic mass (as indicated by arrows) and neurosecretory cells with large nuclei (× 450) Note : CAHP positive granules in the pathway (P) ; B. L.S. of the visceral ganglia of *C. rivularis* showing extreme degeneration and partial loss of organization ; C. Parasagittal section of cerebral ganglia of *Crassostrea* sp. showing degeneration in the neurosecretory cells and in the pathway with NSM (as indicated by arrow) (× 120) Note : Conspicuous lacunae in the ganglionic mass ; D. Parasagittal section of cerebral ganglia of *Crassostrea* sp. showing active, granulated neurosecretory cells and neurosecretory material in the pathway (as indicated by arrows) (× 450) ; E. L.S. of visceral ganglia of *C. rivularis* showing granulated neurosecretory cells indicating resumption of granulation in neurosecretory cells (as indicated by arrow) (× 120) Note : Densely granulated pathway (as indicated by long arrow and P) and F. L.S. of visceral ganglia of *C. rivularis* showing granulated neurosecretory cells indicating resumption of neurosecretory activity (as indicated by arrows) (× 450) Note : Densely granulated pathway (as indicated by long arrow).

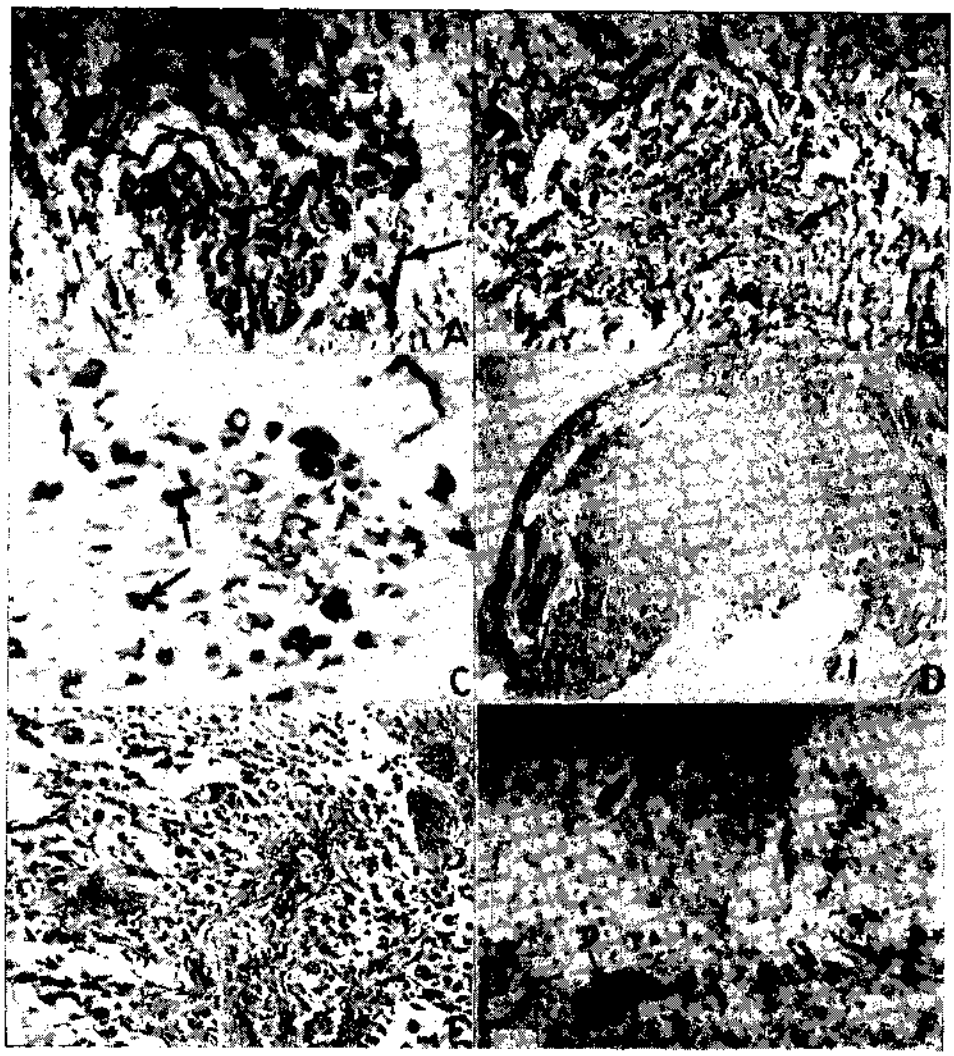

PLATE III. A. L.S. of visceral ganglia of *C. belcheri* showing termination of pathway releasing neurosecretory granules over capillary walls (as indicated by arrow) (×450) ; B. L.S. of visceral ganglia of *C. belcheri* showing shrunk degenerative change (asindicated by arrows) and neurosecretory material in the pathway (× 450) ; C. Parasagittal section of cerebral ganglia of *C. rivularis* showing shrunk neurosecretory cells (as indicated by arrow) granulated neurosecretory cells (× 1700) Note : Intense lacunae formation ; D. L.S. of visceral ganglia of *C. belcheri* (× 120) Note : lacunae formation, loss of nervous tissue, fragmentation of nerve fibres reaching a new height ; E. Magnified view of L.S. of visceral ganglia of *C. belcheri* (×450) and F. Parasagittal section of cerebral ganglia of *Crassostrea* sp. showing intense degenerative changes (× 1700) Note : loss of NS cells, dissolution of NS cells (D) and lacunae formation at climax.

cells were calculated and monthly record of this is presented in Fig. 1.

Several degenerative changes recorded in the ganglionic tissues and neurosecretory cells *viz.* pronounced vacuolization of the cells, shrinkage of nerve cells as well as the neurosecretory cells, formation for lacunae in the ganglia, disintegration of nerve fibres, nervous tissues, etc. were classified as follows:

1. Mild (+) — Very few region of the ganglia showed changes
2. Moderate (++) — About 25% of ganglionic tissue showed changes
3. High (+++) — More than 60% to 70% of the ganglionic tissue showed changes
4. Very high (++++) — Almost all the cells and almost entire ganglionic tissue showed changes.

Observations

The neurosecretory cells were located in peripheral region of both cerebral (Pl. I A) and visceral (Pl. I B, C) ganglia of the oysters. Around the visceral ganglia kidney tubules were located (Pl. I B) and release of neurosecretory material via nerve on cells of kidney tubules was observed (Pl. I D).

Monthly cyclic activity of the neurosecretory cells of cerebral and visceral ganglia of C. rivularis, Crassostrea *sp.*, C. belcheri *and* C. gryphoides

January : The neurosecretory cells of both cerebral (Pl. I E) and visceral (Pl. I F) ganglia were loosely arranged and most of them showed large vacuoles. The nuclei were enlarged with nucleoli, degeneration and loss of some neurosecretory cells was evident and most of the neurosecretory cells were degranulated. The pathway was full of CAHP positive granules. Release of these granules on cells of the kidney was observed from the visceral ganglia (Pl. I D), through nerve arising from the visceral ganglia.

February — May : The neurosecretory cells continued to show degenerative changes as observed in January. Many neurosecretory cells were shrunk and pathway continued to show CAHP positive material. The ganglionic mass of both the ganglia showed conspicuous lacunae (Pl. II A, B, C).

June, July : The compactly arranged neurosecretory cells of the cerebral (Pl. II D) and visceral (Pl. II E, F) ganglia showed intense granulation and conspicuous nuclei with nucleoli. The pathway showed densely stained large granules. The germination of axons over wall of capillaries of the ganglionic mass was evident (Pl. III A).

August, September : The neurosecretory cells were shrunk and few were degenerated. However, some neurosecretory cells showed intense granulation and possess large nuclei with nucleoli. CAHP positive granules were observed in the pathway (Pl. III B, C).

October - December : Most of the neurosecretory cells of both the ganglia were shrunk. The ganglionic mass showed prominent lacunae, loss of nervous tissue and fragmentation of nerve fibres. In short, the degenerative changes reached a peak in the postspawning period (Pl. III D, E, F).

Discussion

It is evident from Fig. 1 that a large number of neurosecretory cells of both the cerebral and visceral ganglia show more degranulation during spawning season in the four species of the edible oyster. The degranulated neurosecretory cells are on increase from May, reaching a high during the spawning period, in particular in June, July and August. The number of degranulated neurosecretory cells decreases considerably during post-spawning, *i.e.* in September, October and November in the four species. However, the number of degranulated neurosecretory cells again increases

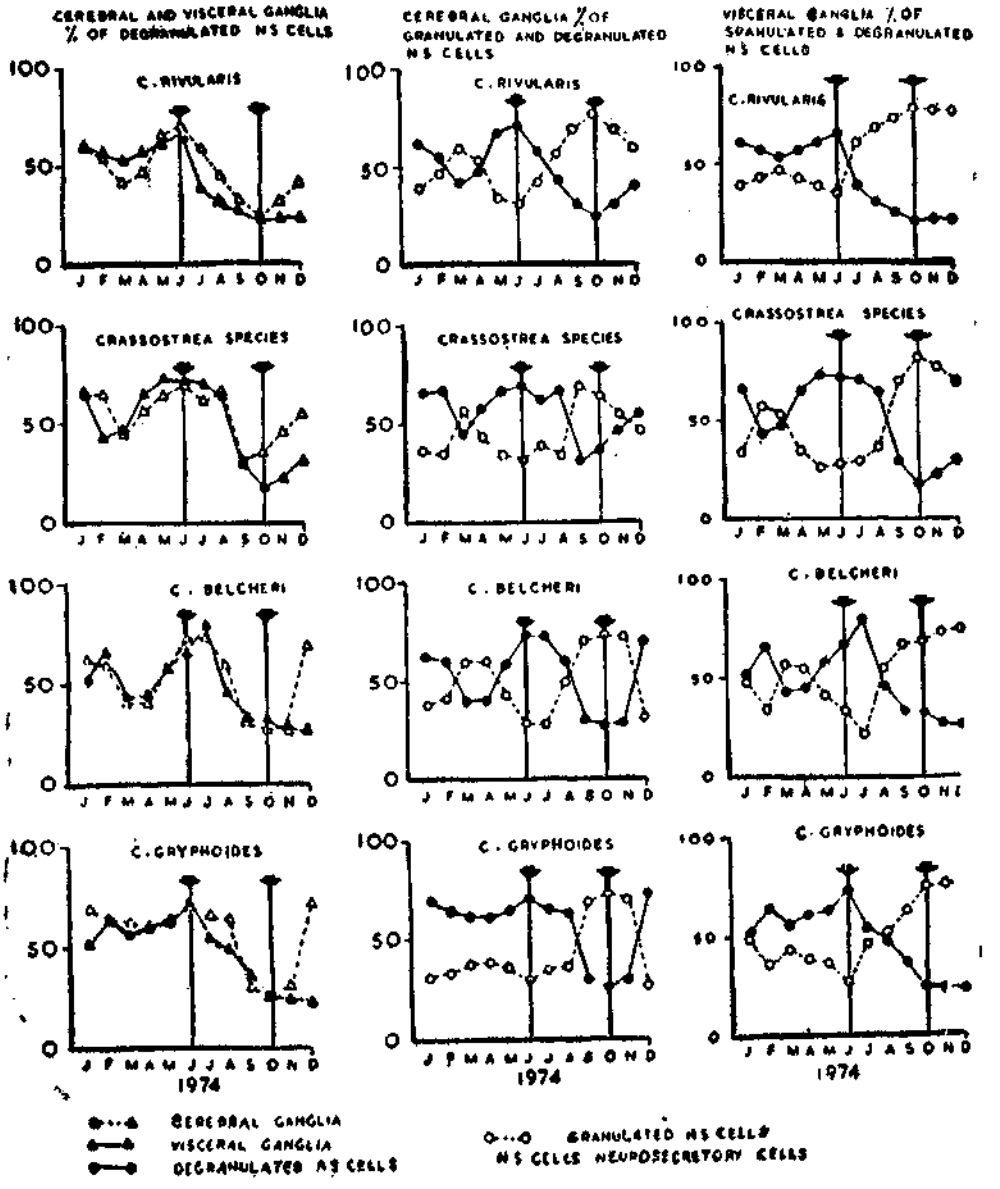

FIG. 1. Monthly percentage composition of granulated and degranulated neurosecretory cells of the cerebral and visceral ganglia of four species of the edible oysters.

from December onwards to February, with the beginning of gametogenesis. It can be said now that more number of degranulated neurosecretory cells in the cerebral ganglia are observed during spawning and onset of gametogenesis in the edible oysters. This is suggestive of cyclic changes in the neurosecretory cells of the cerebral and visceral ganglia of the oysters in relation to the above two phases of reproductive cycle. The neurosecretory cells of cerebral ganglia of pond animal *Lymnaea stagnalis* (Joosse, 1964) have been reported to undergo a seasonal cycle which could be correlated with spermatogenesis and possible caudo-dorsal cells of the snail control oviposition. Lubet *et al.* (1973) have suggested that the cerebral ganglia contain factors responsible for oogenesis completion. Experimental studies involving ablation of the cerebral ganglia in the oyster *C. virginica* resulted in the spawning in female oysters (Nagabhushanam, 1964 a). Histological studies on neurosecretory cells of *C. virginica* (Nagabhushanam, 1963) and in slug *Laevicaulis alte* (Nagabhushanam and Kulkarni, 1971) are suggestive of a secretory cycle of neurosecretory cells of cerebral ganglia in relation to spawning. The results presented here suggest that both the cerebral and visceral ganglia influence gametogenesis in both male and female oysters and also act as trigger for spawning in the edible oysters.

The visceral ganglia (Fig. 1) also show maximum number of degranulated neurosecretory cells during spawning season of the oysters and during onset of the gametogenesis. Thus it is possible to state that the neurosecretory cells of the visceral ganglia influence spawning and gametogenesis in the edible oysters like neurosecretory cells of the cerebral ganglia. In *Mytilus edulis* (Lubet, 1955 a), the secretory cycle of neurosecretory cells of cerebropleural and visceral ganglia show degranulation during the spawning. In Zebra Mussel *Dreissena polymorpha* (Antheunisse, 1963 a, b) the neurosecretory cells of cerebropleural and visceral ganglia show the discharge of the neurosecretory material during spawning. The release of neurosecretory product from the cerebral and visceral ganglia in the clam *Donax truneulus* stimulate gametogenesis in both male and female (Badino and Marchionni, 1972). The neurosecretory activity as evident from maximum degranulation has been correlated gametogenetic process and spawning in *Pinna atropurpurea* and pearl oyster *Pinctada fucata* (Desai *et al.*, unpublished). The degenerative changes, as evidenced by lacunae in the ganglia, dissolution of nerve fibres, degeneration of neurosecretory cells, etc, of high intensity, appear during postspawning of the oysters as reported in slug *Laevicaulis alte* (Desai *et al.*, 1971), *P. autropurpurea* and *P. fucata* (Desai *et al.*, unpublished) and in barnacles (Desai and Senthikumar, 1975).

The neurosecretory material via nerve of the visceral ganglia has been found to deposit the neurosecretory granules over the cells of kidney tubules of the edible oysters. This may be for osmoregulatory process. It has been reported by Nagabhushanam (1964 a) that the neurosecretory cells of visceral ganglia play a role in osmoregulation of *C. virginica* and are influenced by the changes in salinity of medium. The observed release of the neurosecretory material on wall cells of kidney tubules of the oysters studied can be considered as a probable pathway for the neurosecretory cells of visceral ganglia for osmoregulatory function. However, further work in this direction is necessary.

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PURIFICATION OF FARM GROWN OYSTERS

K. NAGAPPAN NAYAR*, M. E. RAJAPANDIAN* AND D. C. V. EASTERSON

Central Marine Fisheries Research Institute, Cochin-682 018

ABSTRACT

The purification of farm grown oysters by scientific methods is essential before they are marketed. Oyster farming is a new venture in India and the technique has recently been perfected by the Central Marine Fisheries Research Institute. The most important task is to perfect the depuration process for which there is no standard method. The present paper gives a satisfactory plan for small scale purification of harvested oysters before they are marketed for safe human consumption.

INTRODUCTION

OYSTERS, mussels and clams are filter feeders and they accumulate pathogenic bacteria such as *Salmonella* spp., *Straphylococcus* spp. and *Clostridium* spp. (FAO and WHO, 1974). Under certain conditions they accumulate biotoxins, pesticides and also heavy metals (Wood, 1972). The only viral disease transmitted appears to be infectious ' viral hepatitis ' (Mason and Mc Lean, 1962). Hence purity of these shellfishes is vital before they are marketed. The awareness that the shellfishes could be purified and thus rendered harmless, goes back in history much farther than the medieval times. The Romans during the first century B.C. consumed cockles and oysters after treating them in tanks which are the earliest known examples of ' Cockle washery ' (Yonge, 1962).

Commercial producers of shellfish in many countries practise purification methods, though this differs from country to country. There are diverse methods of purification, from simple washing in chlorinated waters to exposure to ultra-violet treatment and ozonisation.

Cleaning of bacteria-contaminated oysters using their own physiological filtration mechanism was developed at the Fisheries Experimental Station, Conway, U.K. (Dodgson, 1928). Wells (1923) described a purification plant in New York, U.S.A. using chlorinated sea water. This method of chlorine sterilisation is still in vogue in many of the developed countries. Recently this process of purification has been somewhat superceded by ultraviolet sterilisation (Quayle, 1969) and ozonisation. Using sophisticated ultraviolet sterilisers reduction in coliforms, as high as 99.9% could be attained (Wood, 1961 ; Kelly, 1961).

This paper presents a plan that has been evolved for the purification of oysters farmed at Tuticorin by the Research Centre of the Central Marine Fisheries Research Institute. The water of Gulf of Mannar, particularly that in the Tuticorin Bay where the farming is centered, is to a considerable extent free from sewage and industrial pollution. But to avoid the potential rise of infection which may discourage the consumption of oysters, a depuration system has been set up. Presently about

* Present address : Tuticorin Reasearch Centre of Central Marine Fisheries Research Institute, Tuticorin 628 001, Tamil Nadu.

4800 oysters *i.e.*, nearly 600 kg can be purified per day.

Source and Quality of the Water

Sea-water required for the tanks is drawn from the Tuticorin Bay and is moderately turbid. The annual mean temperature variation of the water is 25.4°C to 30.4°C ; salinity from 31.7‰ to 37.48‰ dissolved oxygen content from 3.18 ml/l to 5.04 ml/l and pH varies from 7.68 to 8.43. The proximate bacterial count of seawater is 200-300 MPNS/ml and the coliform bacteria count 50-100/100 ml (Velankar, 1955). As the seawater required for the plant should be essentially free from turbidity, successive units for sedimentation and sand filtration have been designed for the water system to reduce turbidity which reduces the bacterial count also to a certain extent.

Water Treatment

Seawater is drawn from a distance of 80 m away from shore to an underground sump through a P.V.C. pipe line of 10 cm diameter. The pipeline has been laid at a gradient of 60 cm and water flows into the sump at the rate of 130 litres per minute. At the lowest tide the sump maintains 1,425 litres of water and during high tide the sump collects 2,500 litres of water.

Sedimentation and Filtration

From the sump the water is pumped to the sedimentation tank which consists of 4 chambers, each measuring 1.0 × 1.5 × 1.2 m. The water passes through these chambers before reaching the filter bed and turbidity of the water is very much reduced by this process.

All suspended particles in the water are effectively removed by the sand filters in the filtration tank (1.8 × 1.8 × 1.2 m). The filtered water is collected in a ground level storage tank measuring 3 m × 3 m × 1.2 m with a capacity to hold 10,000 litres of water. By employing a rotary pump driven by a 1 H.P. motor, the storage tank can be filled in an hour. All these tanks are constructed with brick and cement mortar (Fig. 1).

Oyster Cleaning Tanks

Three concrete oyster cleaning tanks each measuring 2.5 × 2.5 × 1.0 m have been constructed. For each tank separate channels of supply and drainage have been provided. The drain valve in each tank is provided in a corner fitted with a galvanised iron 'T'. The vertical limb of the 'T' is raised at a height of 50 cm so that the same height of water column can be maintained inside the tank (Pl. I A). The horizontal limb is plugged when the tank is engaged in cleaning operation. This plug is removed while draining the tank. Operated by a rotary two stage pump the water supply is effected from the top of each tank through a P.V.C. tube with jet arrangements. An independent tap is fixed to the pump with control valve for the operation of a hose.

Operation of Cleaning Tanks

Four wooden grids of the size 42 × 120 cm with a height of 15 cm are placed on the floor of one of these tanks. Oysters are arranged 2 deep on 26 nylon knitted trays each measuring 60 × 60 × 15 cm and placed on the wooden grids (Pl. I C). The oysters are thoroughly hosed by a strong jet of water to remove external mud and dirt (Pl. I B) which are sent out through the outlet. The oysters are then inspected and the damaged and broken ones are removed. By this time the second tank is made ready for cleaning the oysters. Filtered seawater is filled in the tank for cleaning the oysters. The filtered seawater is filled in the tank to a height of 50 cm and the trays with oysters numbering 2,400 are transferred into it.

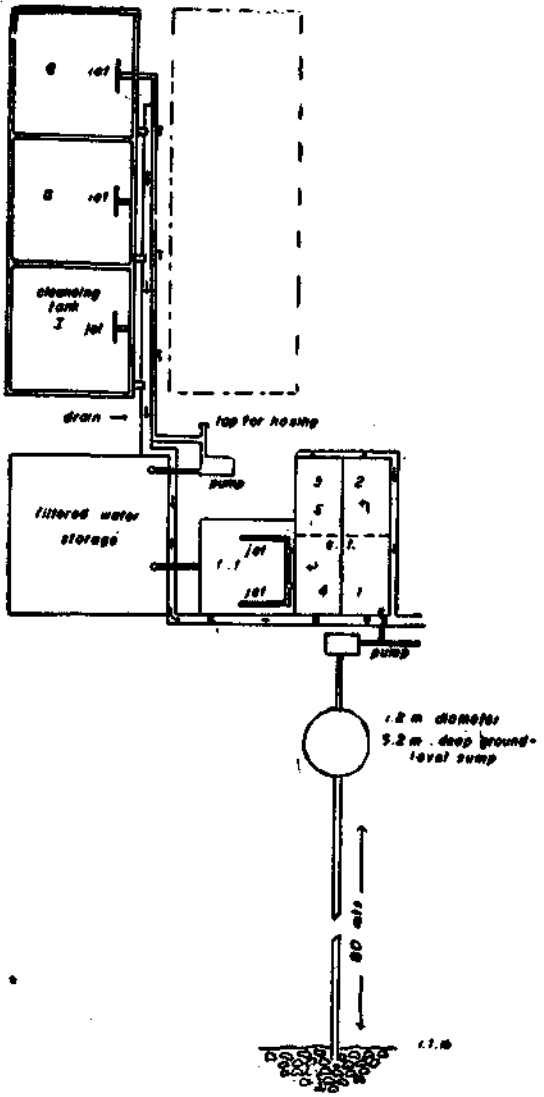

FIG. 1. Layout for purification tanks for edible oyster (f.t.—filtration tank, m.l.t.m. — maximum low tide mark and s.t.—sedimentation tank).

Through small holes a slow flow of water is maintained. The drain valve is adjusted to maintain a water column of 50 cm inside the tank. The oysters are allowed to remain for 12 hrs in this medium to rid them of bacteria by natural physiological process.

Later water in the tank is drained and the oysters are once again hosed by a strong jet of water. By this flushing the accumulated faeces and pseudo-faeces are removed from the tank and also the faeces that might have lodged between the oysters are eliminated. This reduces the chance of oysters repolluting one another. The tank is again filled with water and the oysters are immersed in it for another 12 hrs. At the end of this cleaning period the oysters are kept for one hour in freshly chlorinated seawater with 3 ppm chlorine. Immediately after this the oysters are dechlorinated by hosing filtered seawater and placing them in the holding tank. The object of dechlorination is to allow the oysters to function normally before sale and to remove the smell of chlorine. Although the chlorination method of depuration of shellfishes is followed at present, it is intended to use a suitable ultraviolet steriliser to achieve maximum hygienic standard.

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SOCIO-ECONOMIC PERCEPTIVES OF OYSTER CULTURE IN INDIA

K. NAGAPPAN NAYAR* AND S. MAHADEVAN
Central Marine Fisheries Research Institute, Cochin-682 018

ABSTRACT

The potential for introducing any labour intensive, income generating marine farming programme aimed at uplifting the socio-economic conditions of the artisanal fishermen has to be properly assessed by the farm scientists before embarking on large-scale propagation of the venture in question over wider areas. In a vast country like India this aspect has to be studied region by region since conditions differ from the coast of one State to another. Proper orientation given based on such a careful analysis will help to ensure effective functioning of the scheme, particularly when it happens to be a new venture. 'Transfer of Technology' scheme initiated in 1979 provided an opportunity to test the feasibility of introducing oyster culture technology developed by Central Marine Fisheries Research Institute amongst a group of small-scale fishermen at Tuticorin.

INTRODUCTION

IT has been stated by fishery experts all over the world that capture fisheries are very likely nearing the limits of practical production capabili ies in spite of the fact that the potential resources of deeper sea areas of many underdeveloped and developing countries ate yet to be fully utilised. (Hanson, 1974) Fishery economists hold the view that the cost of seafood production by means other than maricu ture are likely to continue to rise because of capital expenditure involved. It is in this context that maricu ture, particularly molluscan cul ure assumes greater significance. Greater predictability of production and absence of entry limitations create confidence in those who prac ise culture operations. Added to these, the more pervasive the practice of mariculture becomes the more routine its procedures and the production. This is again strengthened by cost reductions effected through simplification, improvements and refinements in techniques. The economics of production are affected only by local environmental advantages and disadvantages as manifested in topographical, physical, chemical and biological properties of farm site. These can be easily overcome by selecting the right area for culture and identifying similar watermasses for further expansion. Considering this, comparison of the economics of production in one country, of any particular culturable species, with another country may not help to evaluate their relative efficiency; more so because of the disparity in labour cost, cost of farm materials, market disposal rates and intensity of consumer demands. But the very fact that culture practices are prevalent and expanding in several countries show that the income generated by such means leads to profit to those engaged in the ventures.

Prawn farming in brackishwater areas has been in vogue in some parts of our coast, mainly Kerala State and has served as a forerunner of similar enterprises elsewhere in very recent years. The same cannot be said about any other culturable species including molluscs. Molluscan culture for producing sea food is something to which the fishermen are not mentally adjusted to eventhough subsistence fishing for clams in estuarine regions is prevalent. Kerala, Karnataka and Maharashtra fisherfolk regularly exploit natural avai ability of clams

* Present address : Tuticorin Research Centre of the Central Marine Fisheries Research Institute, Tuticorin-628 001, Tamil Nadu.

and mussels during certain seasons for consumption and sale in the local market.

Oyster culture

Notwithstanding the natural occurrence of beds of edible varieties like *Crassostrea madrasensis, C. gryphoides* and *C. discoidea,* oysters do not appear to have been exploited except for meagre exploitation in a strip of Tamil Nadu Coast and Maharashtra. Numerically and distribution wise *Crassostrea madrasensis,* called 'Madras oyster' enjoys a better status and is therefore a candidate for culture attempts on scientific lines. The need for culture of oysters in India even when natural stock is left unutilised is best understood only when the difficulties involved in collecting them from natural habitats are explained. The natural beds are rugged areas and because of the habit of oysters to settle down and get cemented to the substratum and grow as clumps, removal and separation of oysters is a time consuming process. The places where they settle down and grow are shallow, muddy locations, at times inaccessible without injuring the feet or getting the body bruised. Hours of work in the natural bed may but yield scanty oyster flesh not commensurate with efforts spent which the fisherfolk otherwise feel can be expended by engaging themselves in other easy money earning efforts. This has resulted in their total disregard for oyster exploi ation from time immemorial. The present day techniques of culture of oysters had put oysters on a par with other edible seafood products with very little difficulties posed in harvesting the tended stock.

Main considerations and criteria in oyster culture

(*a*) *Area availability and resources*

Tamil Nadu coastal area has been identified as rich in the natural resources of the edible oyster *Crassostrea madrasensis* and environmental characteristics of the water mass are also suitable for attempting oyster culture. It has been estimated that an area of 0.145 million hectares of brackishwater area are available for utilization (Silas, 1977) and an equal area of shallow coastal belt can also be utilised for establishing farms wherever conditions permit.

(*b*) *Availability of culture technique*

Initial breakthrough in oyster culture has been made by C.M.F.R.I. at Tuticorin. The feasibility of culturing oysters by following 'Rack' system has been shown whereby a production estimate of 140-150 tonnes per hectare is within easy reach. The initial cost involved in the process does not entail huge capital investments. The period needed for growing oysters to marketable size is inside of 12 months from the date of spat collection and only marginal attention and labour is needed for farm management. Women and children can easily carry out the works, connected with routine farming operations. The entire process is ideally suited for marginal fishermen for taking up the venture as a spare time avocation.

(*c*) *Cost of production*

Cost of production involved, worked out for a set of 1 rack, is as follows :

Cost

		Rs.
Teakwood (17 poles)	..	170.00
Ropes etc.	..	30.00
		200.00
Cost of 20 cages per rack	..	600.00
Total	..	800.00

Production

No. of oysters at 250 per cage	..	5000
Mortality 8%	..	400
		4600

At the end of 12 months wt. of oysters at the rate of 8 oysters/kg. $\frac{4600}{8} = 57.5$ Kg

Total wt. of flesh at 10% of weight 57.5 Kg

The oysters (with shell) can be marketed to internal agencies at the rate of Rs. 20/100 which will yield Rs. 920 per rack, thus yielding a net profit of Rs. 120 per rack. The area occupied by a single rack is 25.5 sq. m and in 1 hectare it is possible to erect at least 250 racks which will yield a production of not less than 140 tonnes (shell on weight) and a minimum of 14 tonnes meat weight. In terms of income per hectare, yield will be Rs. 30,000 giving an average of 2500 per month. For managing a hectare farm collective labour needed will amount to 10 members although it might be less depending on the ability of fishermen to manage. Each rack together with the cages is good for a period of 3 years and so capital investment is ' zero ' in second and third year. Maintenance expenditure will amount to Rs. 350 per year by way of mending the cages and renewing and replacing the snapped ropes. Thus the venture is not only financially sound but also productive and employment oriented. (The figures are based on actuals arrived at by operating 100 racks during 1976-77 and 1977-78).

(*d*) *Economic considerations*

The earning capacity of an active fisherman is considered fairly high, especially so when he is a mechanised boat owner. Naturally the appeal to him will be less. On the other hand an artisanal fisherman who ekes out a living of bare means will be benefited by the culture practice. In any effort to make the fisherman get involved a daily income appears to hold the key, since they are poor and cannot wait till the year to market their products and get the proceeds as in the case of agriculturists. This is because of their poverty and habit of purchasing their domestic food and ration requirements on a day-to-day basis. Therefore any effort to involve them in farm practice should be without affecting their normal fishing routines whereby they get daily returns.

(*e*) *Mobility of labour*

Labour in fishing industry has not exhibited signs of mobility so far. But in recent years the outlook has changed when it comes to the question of earning more money by exploiting resources of the sea. Such works involve shifting of operations from one place to another, total abandonment of traditional fishing for the sake of new enterprise, a sort of 'pescatorization' (FAO, 1957). This change in attitude augurs well for introducing new systems of fishing like ' oyster culture '.

Experimental transfer of technology

At Tuticorin Research Centre where oyster farming methods have been developed, it was planned to introduce the programme of transfer of technology on oyster culture to a selected group of small-scale fishermen. Many fishermen who were controlled did not come forward since no money is to be given to them to start with either as grant or subsidy. It was explained to them that all they need to do is to spend a few hours of their leisure which would eventually bring them a steady additional income without hampering their normal fishing activities. But many could not comprehend the significance of this programme probably because the very idea was new to them. Those who were convinced, expressed willingness to take up oyster farming as an additional occupation. A benchmark survey was conducted to get information on family details especially their income, etc. of 15 families selected for this programme. There is not much social disparity amongst them although a group of four fishermen owning boats seemed to enjoy a slight edge over the rest. All the fifteen were classified as belonging to economically backward fisherfolk.

The level of education attained by them is deplorably low. For a well developed urban

centre like Tuticorin with plenty of facilities and opportunities for getting atleast a fairly moderate level of education with all incentives offered by Government, the youth are least attracted and continue their blissful oblivion forsaking their educational pursuits in favour of the call of the sea.

The annual income of a labourer is approximately Rs. 3600. Of this major slice goes for domestic expenditure. The boat owner's annual net income is about Rs. 5400 while his budget for domestic side is a bit high, a considerable sum is earmarked and spent for the maintenance of craft and gear. Ultimately the boat owners as well as labourers draw blank in their credit since the habit of 'savings' is unknown to them. They not only spend what they earn everyday, but also borrow regularly.

During the initial period, considerable pains were taken to explain to the fishermen about the necessity of constant farm attendance. Several goodwill visits to the fishermen colonies by the connected staff were made. This exercise had the desired effect and now, the fishermen visit the farm during their leisure time after fishing even without our initiative. At present the womenfolk are not in a position to get involved in the farming system, apart from help rendered in fabricating the oyster rearing trays in their huts. Although the children in the family are not at present fully involved in any of the activities of the parents, this younger generation may by virtue of seeing the new type of activity engaged of the elders, take to oyster farming in full measure in the years to come.

The programme is being implemented with certain amount of optimism that this initial experiment an extension education would form the basis for an oyster culture industry in the country. Perhaps a clearer picture might emerge at the end of the first harvest based on which future course of action can be planned.

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A PLAN FOR MOLLUSCAN EXPERIMENTAL HATCHERY AT TUTICORIN, INDIA

K. NAGAPPAN NAYAR* AND D. C. V. EASTERSON

Central Marine Fisheries Research Institute, Cochin-682 108

ABSTRACT

The Central Marine Fisheries Research Institute has developed techniques of oyster culture aimed at production for marketing. At present it is a semiculture depending on nature for collection of spat. Reliance on wild spat has to be reduced since it does not guarantee seed supply at all times. A plan has been evolved for hatchery production of oyster seeds. The paper describes the hatchery system being developed at Tuticorin. The layout with green house, sea water filtration, purification and temperature control systems has been so designed as to achieve tangible results.

INTRODUCTION

THE CENTRAL MARINE FISHERIES RESEARCH INSTITUTE has been actively engaged in researches on pearl culture with the species *Pinctada fucata* (Gould) from 1972 at Tuticorin situated on the Gulf of Mannar along the southeast coast of India. In 1976 the culture of edible oyster *Crassostrea madrasensis* (Preston) was taken up at the centre. The pearl culture technology has been fairly well established, so also the techniques of oyster culture. The two systems rely on spat or juveniles collected from nature by diving with SCUBA for pearl oyster and setting spat collectors for edible oyster. Natural spatfalls are seasonal and erratic and the areas of spat collection also vary. An organism cannot be considered a serious candidate for large-scale culture unless the juveniles are made available economically in large numbers. This implies human control over spawning and the ability to bring high percentage of healthy larvae through the post-larval stages. A hatchery can supply healthy spat uniformly round the year. To meet this demand, a shellfish hatchery was designed at Tuticorin.

The authors express their deep gratitude to Dr. E. G. Silas, Director, Central Marine Fisheries Research Institute for providing the necessary facilities and encouragement.

Layout Plan

The hatchery comprises of two green houses, two nurseries, a breeding laboratory, a nutrition laboratory, a phytoplankton culture room, a general laboratory and other infrastructures such as stores, air compressor room and a standby power generator plant (Fig. 1).

The green houses are of 20 × 10.8 m each with translucent roofing of corrugated fibreglass material with a maximum transmittance of 70% light fixed on tubular truss work. To have proper ventilation heavy duty exhaust fans have been installed. The phytoplankton culture room and the breeding-cum-larval rearing laboratory have been air-conditioned.

Sea water supply

Sea water is pumped from the adjoining bay. The process of collection, sedimentation, filtration, etc, has been described by Nayar *et al*,

* Present address : Tuticorin Research Centre of Central Marine Fisheries Research Institute, Tuticorin, Tamil Nadu.

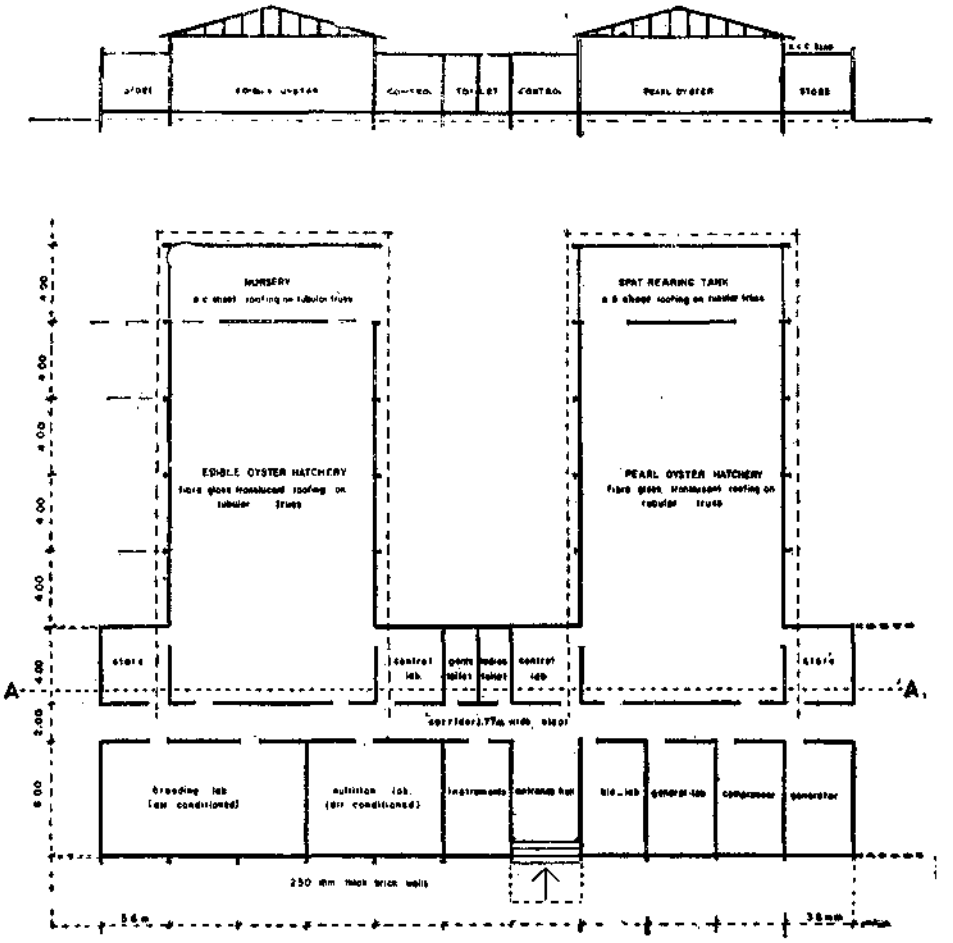
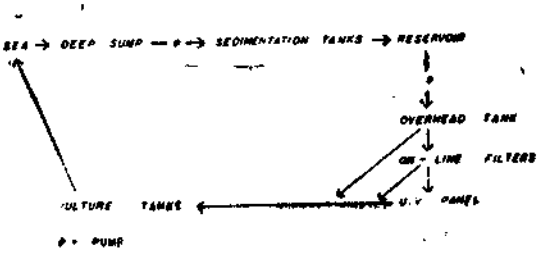


FIG. 1. Hatchery building layout plan (cross section through A-A at the top).

(1980). The hydrological factors of the Gulf of Mannar show a yearly bimodal cycle, due to the influence of southwest (May-June) and northeast (October-December) monsoons. The monthly mean salinity varies from 35.5‰ in March to 31.0‰ in July. Dissolved oxygen ranges from 4.4 ml/l in July to 3.1 ml/l in March. The water is rich in oxygen during January to July and May to September (Marichamy and Siraimeetan, 1979 ; Easterson and Mahadevan, 1980).

The sea water circulation is as given below. Only heavy duty P.V.C. conduits are being used throughout.



In all the pumps used the impeller is of polypropylene with no metals coming into contact with sea water. For the maintenance of breeders and to the nursery tanks, sea water is supplied straight from the overhead tank. To the breeding and larval tanks the water is passed through on-line sintered glass filters of numbers 1 (90-150 μm), 2 (40-90 μm) and 4 (5-15 μm) porosity, and afterwards through a series of cartridge filters in the order of 5, 3, 1, 0.4 and

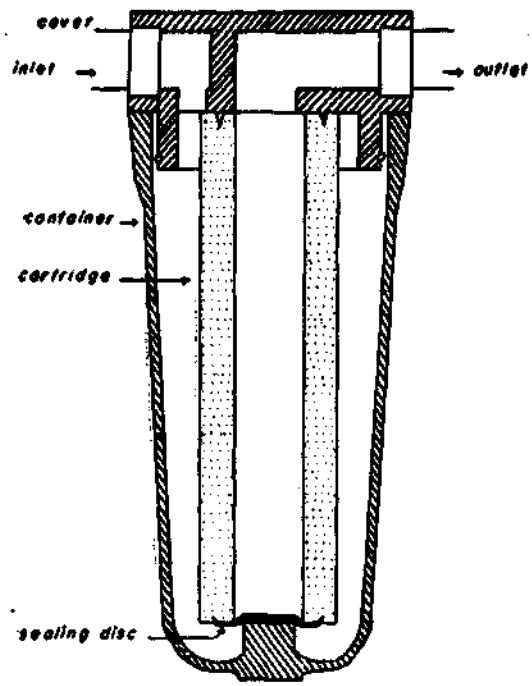


FIG. 2. The details of the cartridge filter used.

0.2 μm effective size retention (Fig. 2). Periodically filtered water is back pumped and the glass filters auto-claved. U. V. panel of similar design as stated by Shelbourne (1964) is used. The ultra-violet tubes are 30 W each and water passes through a distance of 2.5 m in each panel through a series of baffles.

Aeration

An air compressor of 5 H.P. with a provision for a storage pressure tank is used. The air is passed through John-Fowler type of air filters and dry, dust-free air is passed through G.I. pipes with a series of control valves. The air before being used in the larval rearing tanks is passed through on-line sintered glass filter of bacteriological grade. In addition an air pump coupled with oxygen analyser is also available to maintain dissolved oxygen at any desired level above ambient.

Temperature control system

Two types of temperature control systems are available. The first type can be used only to maintain temperature above the ambient. This is done using a silica-cased immersion heater in conjunction with a JUMO contact mercury thermometer and an electrical relay. Here an air pump also serves as a stirrer. This system is simple and can be installed in any type of aquarium tank.

The second type has provision for both cooling and heating (Fig. 3). This embodies two units. The culture tank unit is comprised of three chambers one within the other. The outer and the middle chambers are of anodised aluminium while the innermost is of stainless steel. As required, hot or cold freshwater is circulated in between the innermost and the middle tank. The space between the middle and outer compartments, is stuffed with glass wool for insulation. The JUMO contact thermometer is positioned in a corner of the stainless steel tank wherein oysters are kept. This is covered with a transparent lid to aid visual observation and the water is stirred by means of an aerator. The necessary electrical instruments are in the second unit. It has a two-layered aluminium tank of which the inner sheet is perforated throughout and the copper cooling coil is fixed in-between. A 1000 W heating element has been fixed projecting inside the aluminium tank to avoid cooling coils coming into contact with the heater. Through an opening, freshwater can be poured inside upto 2/3 level, so that copper coils and the heating element are always kept immersed. The aluminium tank is insulated. A 1/3 HP air cooled compressor similar

to the one used in refrigerators is kept below. All these are enclosed within a steel framed anodised aluminium cabinet. At the top towards one side a 1/20 HP water pump, relay and control switches with pilot lamps have been fixed within a metal case. The temperature is set by the JUMO contact thermometer. The pump is connected with the aluminium tank

Operational plan

The programme is as follows :

Quarantine tanks :

Sexually ripe oyster-Brushed, washed, cleaned in 3 ppm chlorine for 10 minutes-Washed out and kept in growing tanks for conditioning.

Breeding tanks :

Inducement of spawning by thermal stimulation-

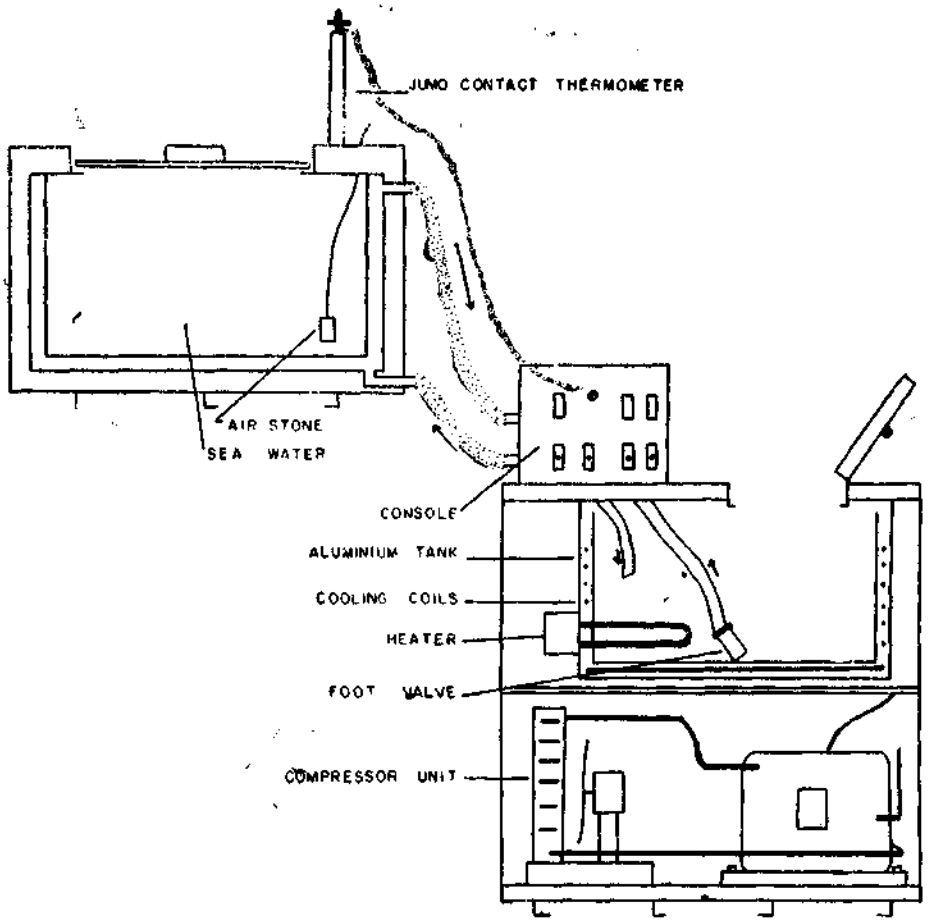


FIG. 3. Details of the temperature control system with provision for both cooling and heating.

through a P.V.C. pipe and foot-valve, and with the culture tank through asbestos insulated P.V.C. tubes, and operates in synchronisation with the heating and cooling elements. The culture tank is kept a few centimetres above the instrument unit.

Transferred to filtered 'bacteria-free' sea water-Releases spawn and eggs fertilized-Oysters transferred to sea.

—Larvae grown in 'bacteria-free' sea water upto eye-spot stage.

Settling tanks :

Kept upto thumb nail stage

Nursery :

Grown— Spat collected—Transplanted to farm.

The tanks used are of fibreglass. Powdered oyster shell and black coloured high density polyethylene sheets are used as spat collectors. Below the thumb-nail stage the spat are removed and grown in tanks wherein seawater along with micro-algae are passed so as to upwell from the bottom. From these nursery tanks the spat are transplanted to the sea farm.

Food

The phytoplankters — *Isochrysis galbana, Pavlova lutheri, Synechocystis salina, Chlorella ovalis, Tetraselmis gracilis* and *T. chui* — are cultured in an airconditioned laboratory at 23° ± 2°C. Clear glass jars of 25 litre capacity are used for mass culture. Widemouth Erlenmeyer flasks of 250 ml capacity and 4 lit Haffkine culture flasks are used for subcultures. The culture flasks are illuminated with day light fluorescent strip lamps. The photoperiod is controlled by means of an automatic switch clock. Enriched Miquel's medium is used. The pH is adjusted to 7.5 — 8.0 using 1 N HCl and 1 N NaOH and the salinity is maintained at 30-32‰ by adding boiled and cooled distilled water. The composition of modified Miquel's medium is as follows :

i. *Miquel's fluid-A*

Potassium nitrate	20.2 g
Distilled water	100 ml

ii. *Fluid-B*

Di-sodium monophosphate	4.0 g
Calcium chloride	4.0 g
Hydrochloric acid	2 ml
Distilled water	80 ml

(slight warming is required to dissolve the salts)

iii. *Mineral mix*

Sodium salt of EDTA	300 mg
Ferric chloride	8 mg
Manganese chloride	12 mg
Zinc chloride	1.5 mg
Cobalt chloride	0.3 mg
Copper sulphate	0.12 mg
Boric acid	60 mg
Distilled water	100 ml

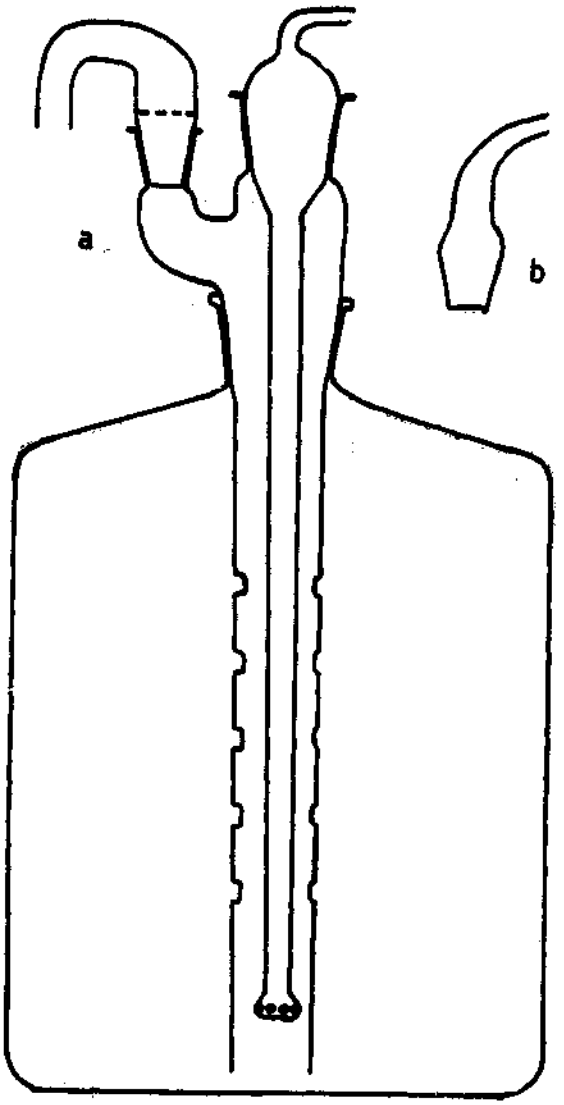


Fig. 4. Bacteria-free algal culture jar.

iv. *Vitamin mix*

Cyanocobalamin	0.1 mg
Thiamine hydrochloride	10.0 mg
Biotin	1.0 mg
Distilled water	100 ml

To one litre of filtered and autoclaved sea water 2 ml and 1 ml of Miquel's fluids A and B respectively are added and to this, 2 ml of mineral mix, is added and checked for pH and salinity. Later 1 ml of vitamin mix is added. Walne's medium (Walne, 1974) is also used in the hatchery.

Large scale bacteria free continuous cultures are grown in a series of 25 lit glass jars designed for the purpose (Fig. 4 a). The air filtered through a on-line sinted filter of 0.1 μm pore size is passed through the vertical tube. The air thus passed is also used to stir the culture continuously following the principle of air-lift. The side arm with a sintered filter serves as air outlet while the filter prevents any possible entry of particles. Nutrients and sea water are added through this side arm after removing the filter assembly. While removing the culture the air supply is disconnected from the vertical tube and coupled through the side arm using the unit shown in *Fig.* 4 b. A siphon is connected to the vertical arm. The air pressure built-up inside the flask and the siphon facilitate the removal of culture. At a time only 3/4 of the culture is removed and the remainder is used to maintain the culture. After removing the culture fresh enriched sea water is poured and the system is operated in its grow out mode. Periodically the system needs to be sterilized.

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PROGRESS AND PROBLEMS OF MUSSEL CULTURE DEVELOPMENT IN SINGAPORE

L. J. CHEONG AND F. Y. CHEN

Primary Production Department, Aquaculture Unit,
Changi Fisheries Research Complex, Changi Point, Singapore 1749,
Republic of Singapore

ABSTRACT

The development of mussel culture in Sinapore is aimed at mass producing a source of rich, but inexpensive protein for the populace. Studies have been conducted on the various methods of culture, including the raft, long-line, pole and bouchot methods, and of these, the raft and long-line have been shown to be the most promising for conditions in the Republic. Thinning of spat-laden ropes to production ropes has been found to be both laborious and time-consuming ; a new type of rope incorporating both spat-catching and grow-out phases without the process of thinning has been effective in reducing the cost of labour and time. Yield is also considerably higher in the non-thinned rope, *i.e.*, 52 kg as against 46 kg shell-on mussels per 4 m length rope per 6 months culture.

The major constraint to large-scale mussel culture development is the handling of large quantities of fresh shell-on mussels harvested by any one time. Mechanisation of bulk handling and deputation of these fresh mussels are concurrently being investigated.

INTRODUCTION

THE GREEN MUSSEL *Mytilus viridus* Linnaeus (synonym *Mytilus smaragdinus* L.) occurs in varying concentrations in the coastal waters of the Republic of Singapore. It is found in considerable abundance, attached in clusters to rocks and fishing stakes at the intertidal and sub-tidal zones of the Johore Straits, a waterway separating the island Republic from peninsular Malaysia (Fig. 1).

Mussel production in Singapore, estimated at 250 tonnes, is derived from artisanal fishermen who harvest from the fishing stakes during low tide. The mussels are cooked in a metal drum, shucked by hand and sauteed on a metal plate. They are then sun-dried and sold as dried mussel meat. Being filter feeders of planktonic organisms in the sea, mussels are efficient converters of primary organic matter to animal protein which compares well in terms of dry weight with other common food items like beef, pork, mutton, chicken and egg (FAO, 1972).

In an attempt therefore to mass-produce inexpensive animal protein for the populace, the Primary Production Department initiated studies in late 1975 on the intensive culture of mussels in Singapore's coastal waters. Under the raft method of culture, mussels were found to attain a market-size of 6-7 cm within 6 months from time of spat settlement, at a production rate of 120 kg/m^2, *i.e.*, an extrapolated production of 5,200 tonnes per hectare of effective area per growing period of 6 months (Chen, 1977).

Mussel as a protein-rich resource has therefore a great development potential in the South-east Asian region. This paper summarises Singapore's experience in the development of mussel culture.

The various aspects of the work could not be conducted without the assistance and cooperation of our colleagues at the Aquaculture Unit. Our special gratitude goes to Messrs. H. B. Lee, E. S. Lee and L. C. Lim and their field staff who carried out the major part of the experiments: Acknowledgements are also due to the Primary Production Department of the Ministry of National Development and the International Development Research Centre of Canada for their continuous support.

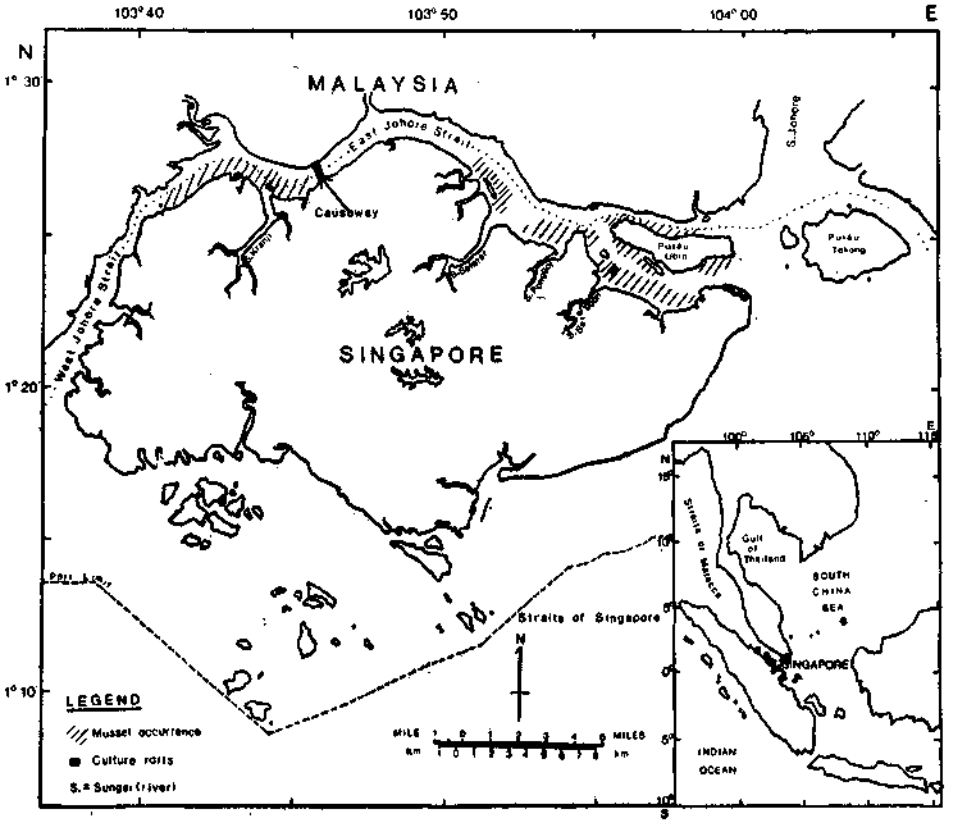

FIG. 1. Mussel distribution in Singapore (Inset: Geographical location of Singapore).

SELECTION CRITERIA

Site selection

Grounds that are suitable for spat collecting as well as production are typified by waters having high phytoplankton concentrations averaging 30.6 μg Chlorophyll *a* /litre seawater (range: 23-40 μg/l) and slow currents of 0.17 (flood tide) to 2.25 (ebb tide) m/sec. In areas where only production is possible, that is where spat are lacking, the current is faster, about 0.25-0.35 m/sec, with phytoplankton averaging 24.5 μg Chlorophyll *a* /litre seawater (range: 17-33 μg/l). In the former area net primary productivity is about 100 mgC/m^3/hr, while that of the latter is about 73 mg C/m^3/hr.

Generally, shel ered estuarine areas with mussel stocks are suitable locations for spat collection. Three areas in Johore Straits, namely Sungei (river) Shelter, Sungei Serangoon and Sungei Kranji Estuaries (Fig. 1)

have been so identified with S. Kranji being most consistent in spat output.

Culture methods

The raft and long-line methods of suspended rope system have been found to be most suitable for Singapore. Due to the lack of extensive mud flats, the pole method is unsuitable.

Raft specifications

Rafts used are basically wooden pontoons with cross-beams for suspending the culture ropes. Wood are of the mixed medium and heavy hardwood variety for the main frame and mixed light hardwood for the cross-beams. Plastic drums, each of 200 litre capacity, are used for floating the raft. Metal oil-drums can also be used but they have a short life-span of about five months. Raft size is based on unit modules of 5 × 5 m (25 m²) productive area, and a raft size of 150 m² has been found to be suitable for production operation (Fig. 2). Shades made from the fronds of attap (*Nipa frutescens*) attached to wooden frames and placed on top of the raft have been found to improve mussel growth.

Concrete anchors are used for anchoring the rafts. Depending on the current prevailing and the type of sea-bed, the total weight of anchors used is usually twice that of the combined weights of raft and full load of mussels, the latter being the weight in water (20% of weight in air). Length of anchor rope is approximately 3-5 times that of Low Water Spring Tide (LWST) depth so as to allow for raft movements arising from tidal fluctuations. Rafts are also placed length-wise,

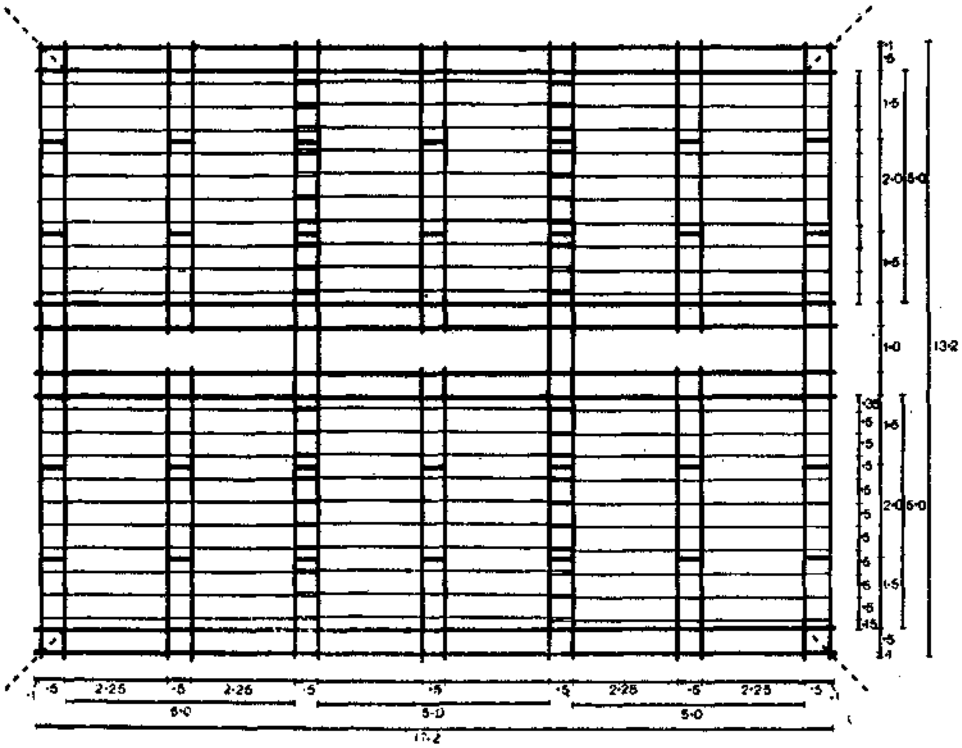

FIG. 2. Plan view of a 150 m² mussel culture raft. — Main frame, — Cross-beam and ....Anchor line) (Specification in metres).

parallel to the flow of flood and ebb tides to minimise wave resistance, and held in a row to maximise utilisation of water space and economise on cost of anchoring individual rafts (Pl A). Such arrangement also reduces the problem of entanglement of the suspended culture rope.

Longline specifications

A longline consists of a single main submerged line or rope held by plastic buoys placed along it at 4.5 m intervals and having culture ropes suspended from the main line at 5.0 m intervals. The depth of submersion of the main line depends on the current at site, the stronger the current the deeper being the submersion. The main line may be 50 to 100 m long and anchoring is at both ends of the line in a manner similar to that of the raft. Details are shown in Fig. 3. Design of the long-line is adopted from that used in the Republic of Korea (Pyen, per. comm.).

nursery rope used for spat settlement is made of coconut coir fibres and is 4 m in length and 40 mm in diameter with a total surface area of 0.503 m^2. It is tied to the cross-beams of the raft by a short piece of polyethylene rope of 14 mm diameter attached to its upper portion. A brick attached to its lower portion serves as weight. The nursery ropes are thus suspended from the raft at a rate of 4 ropes/m^2.

After about 2 months of culture the spat are transplanted to production ropes made of polyethylene material. Each rope is 5.5 m long and 14 mm in diameter. The first 1.5 m is used for tying and the remaining 4 m, which is always immersed in water, for culture purposes. Surface area of the culture portion is 0.1760 m^2. Wooden pegs or chopsticks of approximately 20 cm length each are inserted at 0.5 m intervals to prevent mussel slippage after thinning.

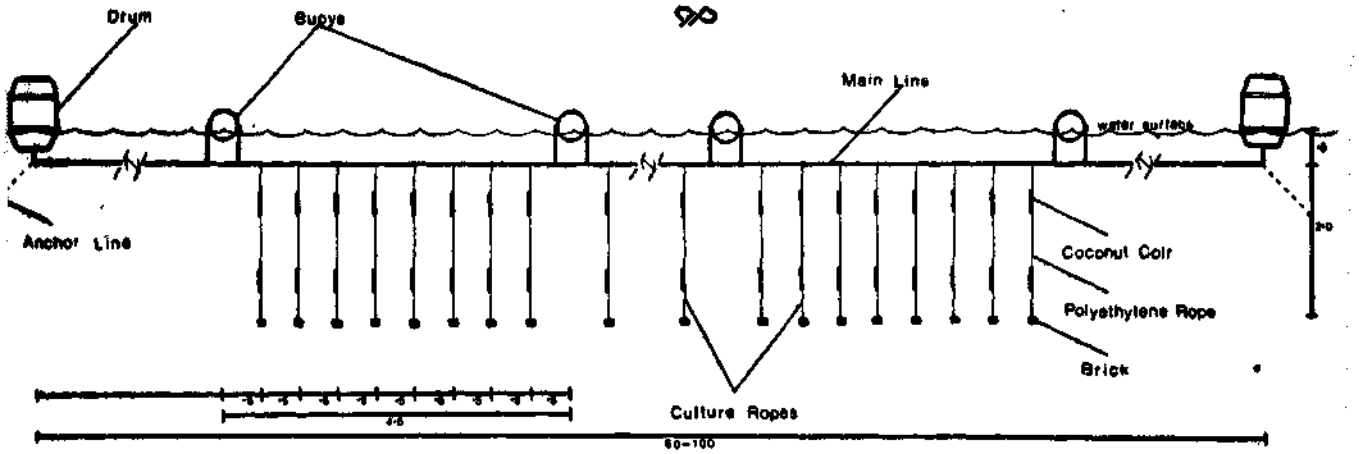

FIG. 3. Schematic representation of a long-line layout.

CULTURE TECHNIQUE

Rope culture with thinning

The method which has been used previously requires the process of thinning by transferring mussel spat from nursery to production ropes. These ropes are illustrated in Plate I B. The

During transplantation or thinning a narrow strip of cotton netting of 13 mm mesh-size and about 25 cm wide is placed beneath the entire culture length of the production rope. Small clumps of mussel spats, plucked from the nursery rope, are then placed on top of the net and around the rope at approximately

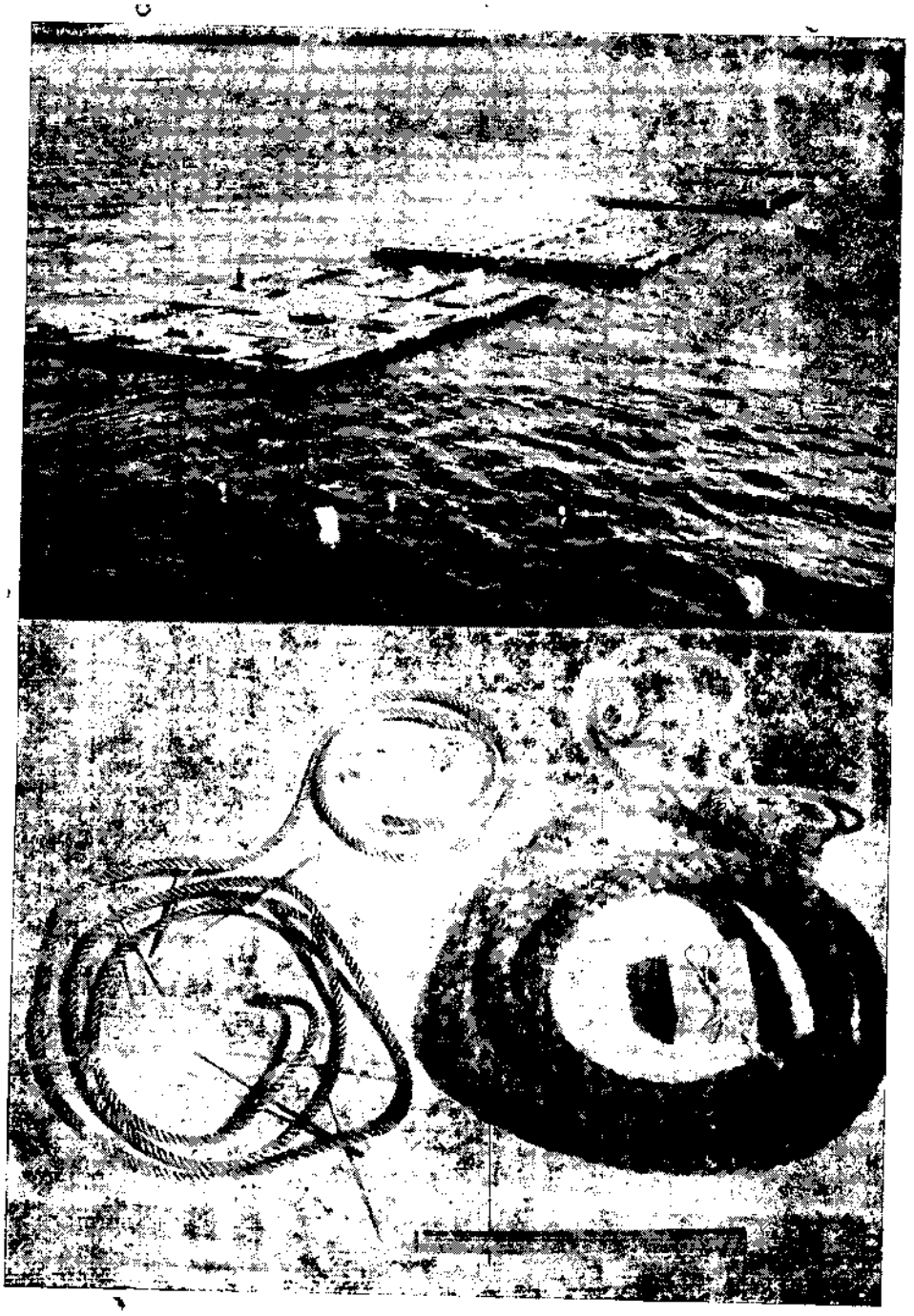

PLATE I A. Aerial view of an experimental mussel culture farm comprising 4 adjoining rafts of 150 m² each and B. Ropes used in culture method employing thinning.
(Right: spat-collecting rope made of coconut coir material; Specifications: 40 mm diameter and 4 m length. Top portion consisting of 1½ m polyethylene rope for tying raft and *Left:* production rope made of polyethylene material; 14 mm diameter, 4 m length for culture and 1½ m length for tying. Chopsticks or wooden pegs at ½m intervals to prevent slipping of mussel clums).

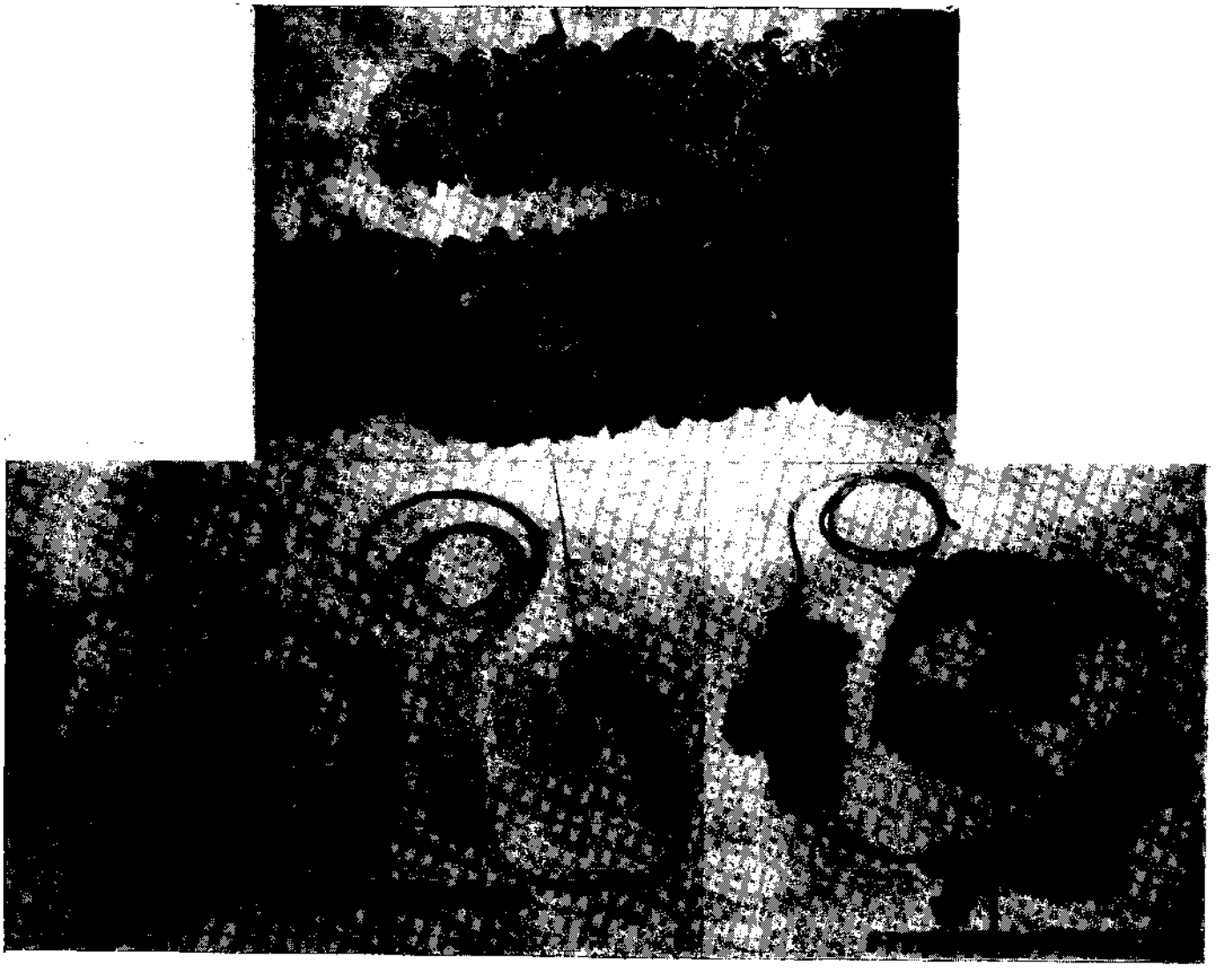

PLATE II A. A thinned-on production rope after 10 days immersion the sea. (*Note :* Rotting of cotton netting and firm attachment of mussel spat to new rope). B. Polyethylene-coconut ropes used in culture method without thinning. *Right :* a 2 m polyethylene rope (14 mm diameter) with 2 pieces of coconut ropes (40 mm diameter) of 30 cm in length each. *Left :* 4 m polyethylene rope 4 pieces of coconut rope and C. A 4 m polyethylene-coconut rope with mussel spat. Note that spat settle mainly on the coconut ropes and later move outwards along the polyethylene rope (arrow mark).

3,000 spats per rope. This density has been found to be optimal in Singapore waters. The cotton netting is then sewed up so as to enclose the spat and rope within. The rope is then suspended from the raft also at 4 ropes/m^2. After about 10-14 days immersion the netting binding the nets would disintegrate leaving behind the mussels which would have by then re-attached themselves firmly to the new rope (Pl. II A). The mussels are cultured for a further 4 months before harvesting. By that time, *i.e.*, after 6 months culture, the mussels would have reached market-size of 6-7 cm shell-length. Yield of each rope averaged 46 kg with a range of 36-53 shell-on (Table 1).

TABLE 1. *Comparison of yields from floating suspended and fixed culture systems after 6 months culture*

	Floating suspended culture				Fixed culture			
	Raft			Long-line	Pole (Refer to text)		Bouchot (Refer to text)	
	polyethylene	poly-coco coconut rope (without thinning)		poly-coco (without thinning)	Bamboo	Bakau	Bamboo	Bakau
	4 m	2 m	4 m		2 m	2 m	2 m	2 m
Total yield incld. mussels and fouling (kg)	50.93	38.77	59.63	28.22	29.33	11.29	37.55	28.27
Yield of market-size mussels, 6-7 cm (kg)	45.93 (r : 36.15 —53.32)	32.53 (r : 25.86 —42.33)	52.06 (r : 28.30 —79.81)	23.57 (r : 15.75 —33.16)	17.16 (r : 7.23 —26.65)	6.08 (r : 4.43 —9.90)	27.35 (r : 21.35 —35.22)	20.62 (r : 17.00 —25.48)
Yield of market-size mussels/metre (kg/m)	11.48	16.27	13.02	11.79	8.58	3.04	13.68	10.31
wt. of market-size mussels to total yield (%)	90.2	84.4	85.9	83.5	58.5	53.9	72.8	72.9
Std. deviation of yield market size mussels (kg.)	5.76	6.87	17.59	5.82	7.74	2.10	5.89	3.57
Coefficient of variabiltiy (C.V.)	12.5	21.1	33.8	24.7	43.4	34.5	21.5	17.3

r : range

Rope culture without thinning

The thinning process, as elaborated above, is laborious and im -consuming. One worker requires about 15 minutes to prepare a single thinned-on production rope. Moreover 2 types of rope, the nursery and production rope, are required. The coconut coir is usable only for about 6 months before it is weakened by *Teredo* borers. There is also the added cost of cotton netting material used for thinning and the loss of spat resulting from handling.

A new method has now been established which eliminates this operation. This method uses a rope which combines the spat-collecting and grow-out phases. This rope termed as polyethylene-coconut coir rope (poly-coco) consists of a main polyethylene rope of 14 mm diameter with pieces of 30 cm coconut coir ropes attached in the centre of each metre of the culture portion of the main rope. The first 1.5 m of the main rope is used for tying, and the remaining portion for culture. This culture portion which is immersed in water

can either be 2 or 4 m in length (Pl. II B). Choice of rope length is determined by handling convenience and depth of water at LWST at culture site. Combined surface area of the culture portion is 0.164 m^2 for 2 m length rope and 0.328 m^2 for 4 m length.

The ropes are similarly suspended at 4 ropes/m^2 and shaded. Spats settle more densely on the open ends of the coconut fibres and along the adjunction between the coconut and

Harvesting is done after 6 months culture as in earlier instance. Yield is considerably higher, averaging 52 kg (range : 28-80 kg) for 4 m rope, and 33 kg (range : 26-42 kg) for 2 m rope. Yield per metre of rope is highest in the 2 m non-thinned poly-coco rope *i.e.*, 16 kg/m. However, as expected, thinned on ropes have higher percentage of market-size mussels and more uniform yields as indicated by the lower standard deviation and coefficient of variability.

TABLE 2. *Comparison of spat-catching efficiency and mean spat settlement distribution in coconut coir ropes and poly-coco ropes after 2 months raft culture*

Data	Coconut rope		poly-coco rope	
	2 m	4 m	2 m	4 m
Surface area of rope ..	0.251m^2	0.503m^2	0.163m^2	0.327m^2
Spat distribution (spat/m^2)				
First metre ..	25,361	26,757	31,073	40,258
Second metre ..	24,851	19,353	21,401	33,982
Third metre ..	..	19,870	..	35,347
Fourth metre ..	..	15,939	..	35,464
Mean spat settlement (spat/m^2) ..	24,607	20,480	26,237	36,263

polyethylene ropes. The coconut pieces thus act as media for spat collection and, by more or less limiting settlement to these areas, effectively distribute settlement uniformly over he ntire rope. The spats densely attached to the pieces of coconut coir rope require more space for further growth, resulting in their migration to the polyethylene portion of the poly-coco rope (Pl. II C). This re-distribution effectively thins out the spat.

The spat-catching efficiency of the poly-coco rope is also much improved over the earlier described nursery rope consisting only of coconut coir fibre. Uniformity in spat-settlement is also more evident in the former as shown in Table 2.

OTHER METHODS

The method of longline culture (Fig. 3) is basically similar to that of the raft, except that the culture ropes are suspended at 0.5 m below water surface instead of from the surface. Due to entanglement of the longer 4m poly-coco ropes, shorter 2 m ones have been found more convenient to use. Yields average 26 kg with a range of 16-33 kg.

Poor yield is recorded for the pole method using bamboo (*Bambosa* spp.) and bakau (*Rhizophora* spp.) poles. Yields from bamboo average 17 kg and range from 7 to 27 kg, while those of bakau average 6 kg with a range of 4 to 10 kg. Fouling mainly by barnacles

is heavy and much time is expended on cleaning the mussel shells. Wrapping spat-laden poly-coco ropes on poles, as in the bouchot method (Bardach, 1972; Mason, 1972), could be practised. Fouling is reduced considerably and yield improved. The average yield of one bamboo pole wrapped with two pieces of 2 m poly-coco spat-laden rope is 27 kg, ranging from 21 to 35 kg. Yield from bakau pole averages 21 kg with a range of 17 to 25 kg.

Post-harvest Operations

Depuration

Preliminary studies on post-harvest operations of mussels have been initiated recently. Depuration or the process of self-purification by the shellfish in 'clean' water, is important in the sale of fresh shell-on mussels which re eaten raw or semi-cooked. Since most of the mussels are sold or served in the cooked or dried state, such studies are aimed more for future app'ication. Investigations are being conducted on the effectiveness of chlorine and ultra violet radiation as sterilising agents. Preliminary results show that UV is a more convenient and practical method and it leaves no residue in the seawater after treatment. Complete bacteria-kill of seawater inoculated with *Escherichia coli* at a concentration of 5.5×10^4 organisms/ml is 2½ hours by UV treatment under recirculating water system. Facilities for depurating mussels on a pilot scale are presently being established.

Pre-processing

A major constraint to large-scale mussel development lies in the handl ng of large quantities of fresh shell-on mussels harvested at any one time. The problem can however be resolved through mechanisation as is practiced in some parts of Europe (Westbroek, 1978). The Department is looking into the establishment of such facilities for various pre-processing treatment including declusterer for separating mussel clumps into individuals, debearder for the removal of the byssal threads and desheller for the shucking or dislodging of the meat from the shell.

Conclusion

The level of mussel culture research in Singapore is sufficiently advanced for large-scale development to be implemented in the near future. The immediate task would be to resolve handling of large quantities of fresh shell-on mussels through mechanisation. Research efforts are directed at determining the carrying capacities of the grounds identified so that guidelines can be formulated on the size and distribution of mussel farms.

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CULTURE OF THE BROWN MUSSEL *PERNA INDICA* AT VIZHINJAM, SOUTHWEST COAST OF INDIA

K. K. APPUKUTTAN* AND T. PRABHAKARAN NAIR

Central Marine Fisheries Research Institute, Cochin-682 018

ABSTRACT

The brown mussel *Perna indica* is found to be abundant in the intertidal rocky area from Vizhinjam to Cape Comorin on the southwest coast of India. Results of culture experiments conducted at Vizhinjam during 1976-1979 period is summarised with details on environmental features of mussel farm, method of brown mussel culture on rope, seeding seasons, growth rate, spawning periodicity, production rate and animal association. The average growth rate of cultured mussels in the bay was 2.92 mm/month. Experiments on culture in the open sea have shown a faster growth rate of 5 mm/month. Spawning of *P. indica* begins by May and lasts till September with peak during July to August. Natural settlement of spat was noticed from July and the peak period of settlement was September-October. Production rate was estimated as 150 tonnes per hectare area for brown mussel. The average yield per metre length of rope was 12 kg.

INTRODUCTION

IN INDIA the green mussel *Perna viridis* and brown mussel *Perna indica* are found in abundance in the intertidal zone of the coastal areas attached to rock, pilings and other hard stratas. *P. indica* has a very restricted distribution from Pondicherry to cape comorin on the east coast and from Quilon to cape comorin on the west coast. From Vizhinjam to Muttom this species is found in greater abundance. Jones and Alagarswami (1969) while describing the molluscan fishery resources of India have pointed out the possibilities of mussel cultivation in India. Davies (1970) mentioned mussels as a potential resource which can be cultivated along Indian Coast. In India experiments on rope culture of mussel were initiated at Vizhinjam in 1971 (Anon., 1978) with a view to developing techniques suitable for the Indian Coast. Important works on mussel culture in India are by Achari (1975), Rao *et al.* (1960), Qasim *et al.* (1977), Appukuttan *et al.* (1980), Kuriakose (1980), Kuriakose and Appukuttan (1980), Rangarajan and Narasimham (1980) and Rajan (1980). In the present communication the results of experiments on brown mussel culture at Vizhinjam from 1976 to 1979 are given.

* Present address : Vizhinjam Research Centre of Central Marine Fisheries Research Institute, Vizhinjam.

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RESULTS

Rope culture experiments during 1976 to 1979 at Vizhinjam

The environmental features of mussel culture site in Vizhinjam Bay where the present experiments were conducted have been described in

detail by Appukuttan *et al.* (1980). The bay is protected and has a depth range of 10-15 m. There is good settlement of *Perna indica* seed in the natural bed in and around Vizhinjam in all the years. The site selected for open sea mussel culture experiments was 1-2 km away from the shore. The depth here ranged from 15 to 25 m. The sea is calm from December to the end of May but is subject to severe monsoon conditions there after making it impossible to keep floating rafts in the sea. The environmental features of the open sea farming site were similar to those of the bay.

The data on salinity and temperature from 1976 to 1979 are given in Fig. 1. Water temperature ranged between 20.75° C and 30.05° C, the lowest during monsoon period and highest during January/February. The minimum salinity observed was during May-June and maximum during summer (March). Salinity was about 33‰ in most of the months in all the three years at Vizhinjam. The decline in salinity and temperature coincided with the monsoon season. Average percentage of silt observed at different depths for three years (Table 1) showed that at 0-1 m silting was insignificant. At 2-4 m silting was more and water was highly turbid during monsoon months. The percentage of silt was calculated from the volume of silt bound in fixed quantity of water collected from different depths at equal intervals.

TABLE 1. *The percentage of silt by volume observed at different depths in Vizhinjam Bay during 1976-79 (Average of three year data)*

Months	0-1 m	1-2 m	2-3 m	3-4 m
July ..	1.00	28.20	42.33	57.33
August ..	1.58	21.88	22.40	34.33
September ..	1.17	5.75	14.45	16.18
October ..	1.50	6.27	8.03	15.10
November ..	1.42	3.02	5.87	10.47
December ..	1.78	2.63	2.83	4.56
January ..	2.05	2.71	4.25	7.17
February ..	1.70	3.48	3.87	6.17
March ..	1.75	3.07	2.75	5.45
April ..	1.47	3.30	4.19	7.52
May ..	0.66	3.95	10.70	11.28
June ..	1.68	14.38	13.80	32.63

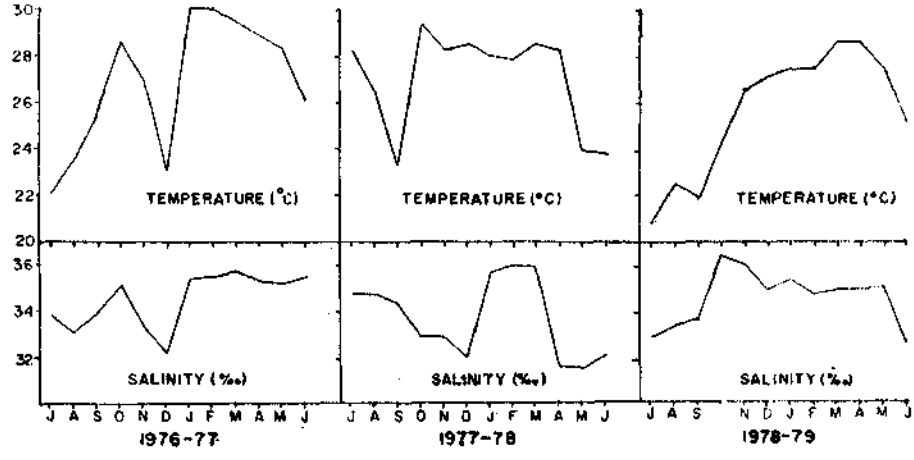

FIG. 1. Seasonal variation in salinity and temperature in Vizhinjam Bay during 1976-1979.

Method of rope culture of brown mussel

Floating rafts of area range 30-100 m^2 were used in all the experiments at Vizhinjam (Pl. I A). Achari (1975), Appukuttan (1980 a) and Appukuttan *et al.* (1980) have described

the methods of rope culture for various experiments at Vizhinjam. Kuriakose and Appukuttan (1980) have given the details of rope culture techniques adopted at Calicut and Vizhinjam.

Seeds for mussel culture were available along the intertidal rocky area in the Vizhinjam Coast and good settlement of mussel seed is observed all along the coast (Pl. I B). Using iron chisel the mussel seed are collected from the rocks. The seed are cleaned and spread over a piece of old cotton net or mosquito net and the nylon rope is kept over the net. The net is wrapped over the rope securing the seed intact and both the ends of net were stitched using cotton twine (Pl. I C). Apart from the seed collected from the natural bed, artificial spat settlers such as roofing tiles, split nylon rope, strings of coconut shells, split bamboo poles and 1 × 1 m iron cages covered with nylon screen were used in the farm to collect mussel seed. Split nylon rope, roofing tiles (Pl. I D) and iron cages gave encouraging results. During 1978, 402 kg of mussel seed were collected from spat settlers released in the bay. This was used for seeding the ropes in December.

In 1976, seeding work commenced by June but could not be continued in August due to rough weather ; it could be resumed only during September-December. In 1977, seeding was done during September-December. In 1978 seeding could commence only by November. Generally September to December period was found ideal for seed collection and seeding. In 1976, four rafts with 13 coir ropes and 110 nylon ropes of 1 to 6 m long were set inside the bay. In 1977, two rafts were launched in the open sea and maintained for 3 months with 36 seeded nylon ropes of 10 m length ; 81 seeded nylon ropes were set in the bay in the corresponding period. In 1978, three rafts with 114 seeded nylon ropes with average length of 5.5 m were kept in the open sea for 5 months. 144 seeded nylon ropes of average length of 6 m were suspended in 6 rafts inside the bay.

Growth rate

Samples were taken every 15th day from numbered ropes which had been seeded and suspended in the same period. The mean length of mussels of each sample was taken into consideration for calculating the growth rate. Fig. 2 depicts the growth of *P. indica* under culture conditions in the bay and Fig. 3 gives its growth in the open sea farm. Analysis of samples show that the average growth rate for all the three years inside the bay have shown similar trends. In 1976-77 the growth increment per year was 35 mm, 35.6 mm in 1977-78 and 36.3 mm in 1978-79. The average monthly growth rate was 2.92 mm. The growth rate of open sea mussels in 5 months was 25 mm *i.e.*, 5 mm/month. In open sea conditions the mussels grow faster and attain marketable size.

Increase in total weight and meat weight of cultured mussels

Fortnightly samples were used for finding out increase in total weight and meat weight from bay and open sea. In 1976-77 the increase in total weight observed was 10.75 g for 12 months, 9.24 g in 1977-78 and 8.9 g in 1978-79 in the bay (Fig. 2). The average total weight increase was 0.77 g per month. The increase in meat weight for 1976-77 was 4.9 g, in 1977-78 3.49 g and 3.52 g in 1978-79. The calculated meat weight increase per month was 0.32 g. In the open sea samples the increment in total weight and meat weight was greater than that of bay (Fig. 3). The increase in total weight in 5 months was 9.72 g with 1.94 g/month and that of meat weight for the same period observed was 4.5 g with 0.9 g/month. The meat weight — total weight relationship is given in Table 2.

Spawning periodicity

The maturity stages of gonad were classified as follows :

Stage I — Ova have not attained regular shape, sperms non-motile; Stage II — Granulation in the ovary observed, ovary with-

PLATE I A. Floating rafts for rope culture of mussel, B. Natural mussel bed at Vizhinjam, C. Seeding of mussels on rope, D. Spat settlers with mussel spat and E. Brown mussels grown to harvestable size on rope.

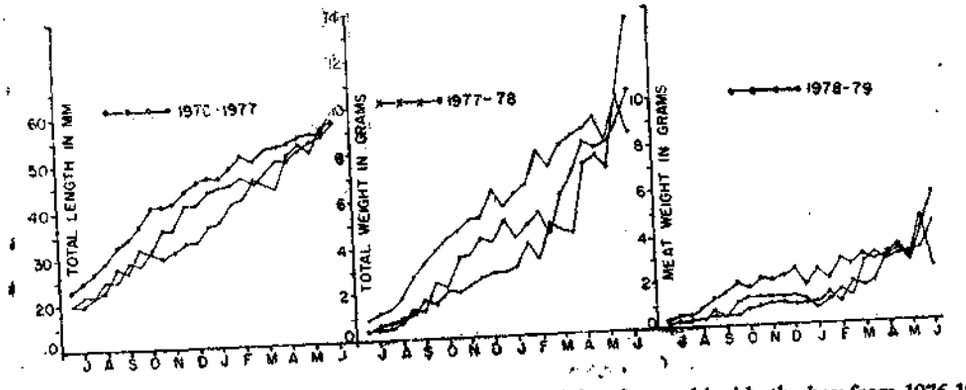

FIG. 2. Growth rate and increase in total weight and meat weight of mussel inside the bay from 1976-1979.

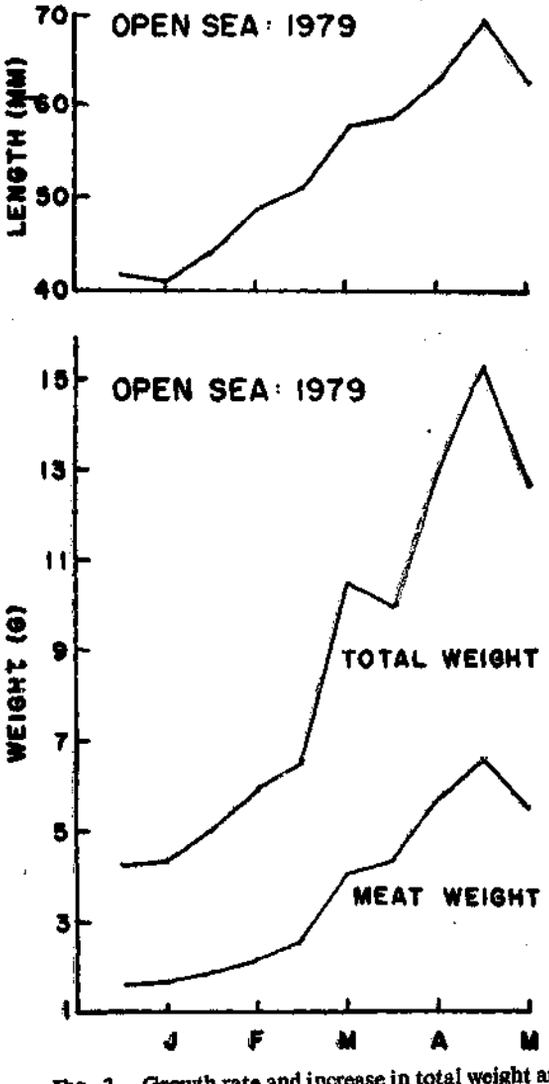

FIG. 3. Growth rate and increase in total weight and meat weight of mussel in the open sea in 1979.

out regular shape, sperms non-motile; Stage III — Ova spherical, ovary of brick red colour, sperms motile and Stage IV — Spent. For determining the stages of maturity in each month 50-300 mussels were examined from the ropes. Maturity stages were observed in different months in the bay for 3 years (Fig. 4). Since mussels above 20 mm in size were examined in the present study, indeterminate stages were not found in the samples. In 1976-77, Stage I was observed from July to March, Stage II from December to April and Stage III from April to June and Stage IV in July and August. In 1977-78 Stage I was observed from September to March, Stage II from November to March, Stage III in July, November, December and from March to June and Stage IV from August to November. In 1978-79 Stage I was present from October to March, Stage II from November to December and in March, Stage III in July to August and April to May period and Stage IV in September and in June. In the open sea samples Stage I to III alone were observed from January to March in 1978 and from January to May in 1979. Stage I was present in January to February and from March onwards Stage II and III were present. In April and May all specimens were in Stage III.

Spawning of *P. indica* commences by May and lasts till September with peak during July-August. Natural settlement of spat in mussel beds was noticed from July and peak period of

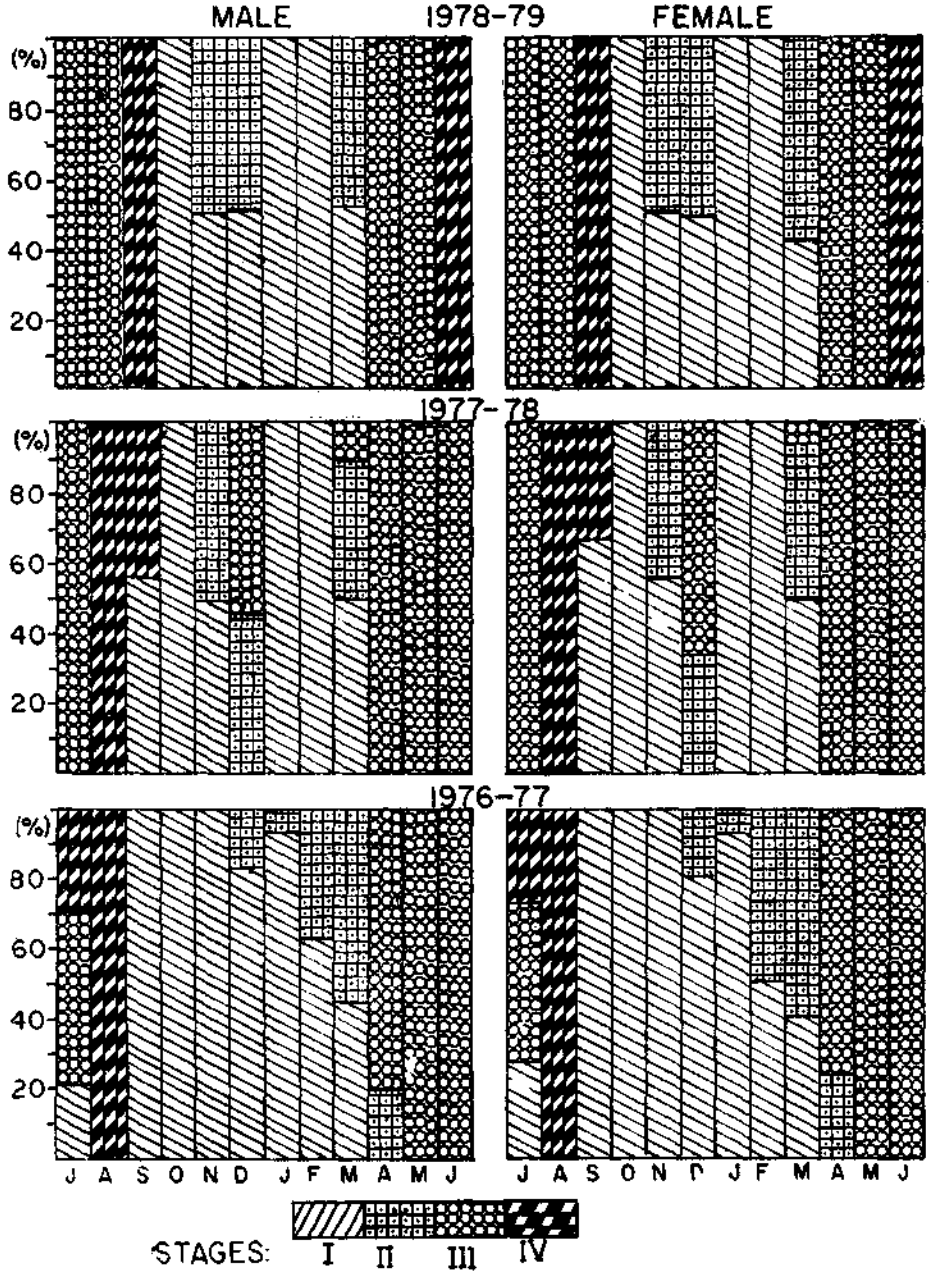

FIG. 4. Maturity stages of *P. indica* in different months in the bay from 1976 to 1979.

TABLE 2. *The average values of meat weight expressed as percentage of total weight of mussels of Vizhinjam Bay and open sea in successive months*

Month	Bay 1976-77	Bay 1977-78	Bay 1978-79	Open sea 1978	Open sea 1979
July ..	38.20	47.58	38.24	..	..
August ..	38.40	31.45	37.04	..	..
September ..	38.30	37.90	37.85	..	..
October ..	37.95	38.78	41.73	..	..
November ..	39.60	35.92	40.97	..	..
December ..	37.95	36.72	42.26	..	..
January ..	34.80	21.31	35.38	27.31	36.37
February ..	33.58	26.17	35.29	28.18	36.31
March ..	34.94	26.24	32.88	30.98	38.44
April ..	33.15	37.44	40.69	..	43.58
May ..	32.62	37.28	41.31	..	43.33
June ..	40.53	33.78	39.19	..	..

settlement was in September and October. Clutches were kept in the farm and settlement of mussel spat was observed from June to September.

Production rate

In Vizhinjam Bay *P. indica* reaches marketable size (55-59 mm) in 8 months from October to May (Pl. I E). In the open sea marketable size of 60-65 mm is reached in five months. Harvesting of mussel could be done in May Experiments in the bay have shown that the average production per metre length of rope is 10-12 kg. In a single raft of 6 × 5 m size, 30 ropes of 5 m length could be accommodated. The production per raft is 3 tonnes and in a hectare area 50 such rafts can be launched giving an estimated production rate of 150 tonnes/ha. Open sea experiments showed that within 5 months (January to May) the mussel production per metre length of rope is 15 kg and the estimated production rate per hectare is between 180-200 tonnes. The production rate is high in the upper two metres of ropes, moderate between 2-4 metres and below that there is a sharp decline in production.

Animal associations

Among the fouling organisms noticed in the mussel farm, the barnacle *Balanus amphitrite* was the most important and next to that was *Crassostrea cucullata*. An interesting observation found during 1977 onwards in the farm area and in the natural bed was the unusual heavy spatfall of *Modiolus* sp. The settlement of *Modiolus* starts by May onwards, which almost coincides with the settlement of brown mussel in natural beds and on spat collectors kept inside the bay. One of the reasons for poor settlement of *P. indica* in natural bed during 1977-78 and 1978-79 period was the heavy spatfall of *Modiolus* sp. before the settlement of brown mussels commenced. During March-April *Avicula vexillum* was found attached to rope and spat collectors in large quantities. Simple ascidians *Ascidia* sp. were commonly found on mussel ropes throughout the year. Crabs of the genera *Thalamita*, *Porcellana* and *Pinnotheres* were found on the ropes. *Pinnotheres* sp. was found inside the mussels but no incidence of damage was noticed. Encrusting tubicolous polychaetes, nematodes, crinoids, flat worms and algae were also found over the mussel

ropes. The species of sponges found encrusting over the mussel shells were *Haliclona tenuiramosa* (Burton), *Callyspongia diffusa* (Ridley), *C. fibrosa* (Ridley and Dendy), *Mycala mytilorum* (Annandale). Occasionally coral growths over the mussel shells was observed. Appukuttan (1980 b) has described the details of production of mussels by fish *Rhabdosargus sarba* in the Vizhinjam Bay.

Remarks

Various experiments conducted at Vizhinjam have shown that rope culture of *P. indica* in protected areas and also in open sea yields high production and this technique can be utilised for commercial scale production of mussels along this coast. The present investigation at Vizhinjam has shown that salinity and temperature, which are most important factors determining the spawning activities of mussels have not shown much differences in the bay and open sea. During monsoon period there was change in salinity and temperature. Qasim *et al.* (1977) have made similar observations in the green mussel culture in Goa Coast. Silting percentage was found higher in depths beyond 2 metres and this was high from May to October. The main reason for poor settlement of mussel seed at Vizhinjam was the indiscriminate exploitation of adult mussels from the mussel beds by the fishermen and also due to heavy settlement of *Modiolus* sp. in this area. Experiments have shown that the ideal length of mussel seed for seeding work is between 20 and 35 mm. Among the spat collectors used, roofing tiles, iron cages and split nylon ropes give good results. The peak season for mussel seeding is September to December. Open sea culture experiments have shown that rafts can be positioned only in December and will have to be taken back to the bay by the end of May, since mussel rafts cannot be kept in permanently due to heavy wave action in the sea. The growth per year in bay was 35 mm and that per month 2.92 mm, whereas in the open sea growth in 5 months was observed to be 25 mm with 5 mm growth per month, thus showing a faster growth in open sea conditions. Qasim *et al.* (1977) have recorded 8 mm/month growth on ropes at Goa in *P. viridis*. Kuriakose (1980) recorded 10.6 mm to 13.5 mm growth per month for *P. viridis* from Calicut Coast. The average increment of total weight of *P. indica* inside the bay was 0.77 g per month and meat weight increment was 0.32 g per month. In the open sea the increase of total weight per month was 1.94 g and that of meat weight 0.9 g/month. Meat weight ratio was high from May to December and hence it is felt that cultured mussels could be harvested from May onwards. Spawning of *P. indica* commences by May and lasts till September with peak during July to August. Rao *et al.* (1976) have reported that *P. viridis* breeds along the Goa coast almost throughout the year with two peaks of spawning — one from September to November and another during February-March.

The estimated production per hectare in the bay is 150 tonnes and in the open sea it is between 180 and 200 tonnes. An annual yield of 480 tonnes per hectare of green mussel has been reported by Qasim *et al.* (1977). The present study has shown that settlement of *modiolus* sp. spat from May onwards has an adverse effect on the natural settlement of brown mussel at Vizhinjam. The effect of other fouling organisms on cultured mussels is negligible.

Davies (1970) has pointed out mussels as a potential food resources and Jones and Alagarswami (1969), Achari (1975), Rao *et al.* (1976), Qasim *et al.* (1977) and Silas (1980) have emphasised the need for culture of mussels to increase the production. Present observations show that mariculture of *P. indica* on commercial scale operation is feasible along our coasts.

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MASS PRODUCTION OF SPAT OF GREEN MUSSEL *MYTILUS VIRIDIS* LINNAEUS IN FRENCH POLYNESIA

AQUACOP*

Centre Océanologique du Pacifique CNEXO-COP B. P. 7004 Taravao. Tahiti (French Polynesia)

ABSTRACT

To develop mussel culture in tropical environment, trials for mass production of spat of *Mytilus viridis* were conducted at the Centre Océanologique du Pacifique (COP) of the Centre National pour l'Exploitation des Océans (CNEXO) in Tahiti.

This paper presents the results obtained in 800 l and 10 m³ larval rearing tank with an initial density of 2,500—3,000 larvae per litre.

Metamorphosis and settlement on nylon meshes are achieved between the 15th and the 20th days. The settlement percentage is between 30 and 60%.

INTRODUCTION

SINCE 1975, the Centre Océanologique du Pacifique (COP), a laboratory of the Centre National pour l'Exploitation des Océans (CNEXO) has developed a programme of studies on the bivalve molluscs culture in tropical area. First experiments were on larval rearing and spat production of the Pacific oyster *Crassostrea gigas*. They allowed to define a larval rearing method (AQUACOP, 1977), adapted from those described by Loosanoff and Davis (1963) and Walne (1966).

Since 1978, studies were conducted on the tropical species *Mytilus viridis*, using the larval rearing method defined for *C. gigas* and the first results on larval development obtained by Tan (1975) and Rao *et al.* (1976). Ten larval rearings were carried out in 800 l fiberglass tanks (AQUACOP, 1979) and one in a 10 m³ fiberglass tank. From February 1978 to September 1979, 8.5 10^6 commercial size spat have been produced.

MATERIAL AND METHODS

The bivalve molluscs hatchery is presently composed of two parts : (1) a wholly closed area, with a semi-transparent roof and fiberglass-sheet walls, containing eight double-shell 800 l cylindro-conical fiberglass tanks, thermoregulated by circulating seawater inside the double wall. These tanks have already been described in a previous paper (AQUACOP, 1979). (2) an open area, only sheltered by a semi-

* Aquaculture team of the 'Centre Océanologique du Pacifique'

— *Algae and mollusc cultures :* J. L. Martin D. Coatanea, O. Millous, Y. Normant, O. Le Moine, G. Oudin, T. Belle.

— *Nutrition :* A. Febvre, J. J. Lainé, J. M. Peignon, P. Maréchal.

— *Water quality control and treatment :* J. Calvas, V. Vonau.

— *Pathology :* J. F. Le Bitoux, G. Breuil, S. Robert.

— *Crustacean and fish cultures :* P. J. Hatt, M. Jarillo, J. P. Landret, J. Goguenheim F. Fallourd, O. Avalle, J. Moriceau, S. Brouillet, H. Crieloue, H. Pont, D. Amaru, V. Vanaa, A. Bennett, D. Sanford.

— *Technology :* J. F. Virmaux.

— *Aquaculture program coordinator in tropical area :* A. Michel.

transparent fiberglass roof, containing a 10 m³ U-shaped fiberglass tank. This tank is made of a wood-frame, which subtends a transparent fiberglass sheet (Fig. 1). In this tank, diurnal

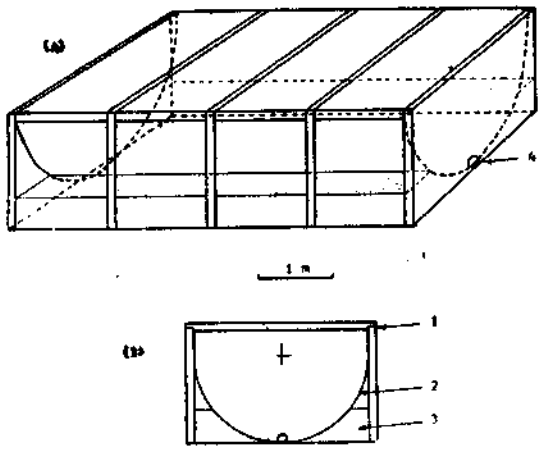

FIG. 1. 10 m³ U-shaped tank—A. General view and B. Cross section.
1. Wooden frame, 2. 1 mm fiberglass sheet, 3. Bed of sand and 4. Draining pipe.

temperature variation may reach 3.5°C particularly after cold nights. Aeration is realized with a perforated pipe running on the bottom on the whole length of the tank.

The hatchery is supplied with filtered sea water (Philippe filters 5 μ and 0.5 μ).

According to the season, the mean larval rearing temperature varies from 15°C to 28°C.

Spawning is induced by thermal stimulation (19°C-35°C), after having evaluated the maturity stage of mussels. Males and females are put apart after spawning starts. Fertilisation is realized artificially (10 spermatozoids for one ovule). The fertilised eggs are put for 24 hours in a 800 l tank until the D-larvae appear.

D-larvae are then shared out among the larval rearing tanks at the density of 3,000 larvae per litre. Water is changed every two days in the 800 l tanks. The 10 m³ U-shaped tank is filled up progressively for the first five days and the first draining takes place at day 6 and thereafter every 3 days. Larvae are first recovered on 48 μ screens. At day 6, they are screened on 65 μ, at day 8 on 58 μ and after day 10 on 100 μ. Countings and measurements are realized on samples taken at the moment of screenings.

Larval rearings 1 to 8 were fed on *Isochrysis* sp. locally isolated and *Monochrysis lutheri* (25,000 cells of each species per ml). After metamorphosis, *Skeletonema costatum* was added (52,000 cells per ml).

Larval rearing 9 was fed on *Isochrysis* sp. and *Monochrysis lutheri*, and after metamorphosis on *Chaetoceros* sp., a strain coming from Oshima, Japan. Larval rearings 10 and 11 were fed on *Isochrysis* sp. and *Chaetoceros gracilis* a strain coming from Costa-Rica (25,000 cells per ml of each species). These last two tropical strains are cultured in the COP algae room in 200 l and 300 l tanks at 25°C *Monochrysis lutheri* and *Skeletonema costatum* are cultured at 20°C. Algae are distributed after the filling up of the tanks. Every day the remaining algal density is controlled and if necessary larval tanks are complemented in algae up to the required density.

Treatment with 12.5 mg/m³/day of Treflan, an herbicid used in agriculture containing 50% Trifluralin was experimented in first three rearings. In rearing 6, an attempt to treat rearing water with one part per million of total chlorine was carried out. All other rearings were treated with 14 mg/l of sulfadimerazine every two days.

Settlement is realized on 5 mm mesh anti-hail nylon nets which are hanging in larval tanks since pediveliger stage. In rearing 11, an experiment to estimate the settlement rate was carried out, by weighing 10 of the 80 nylon net collectors before and after settlement. About ten days after the end of settlement, nets are removed from larval tanks to a 15 m³

pregrowth concrete tank. After larval rearing 11, some of the young spat were transferred to a pregrowth fiberglass raceway 18 m long, 0.5 m wide and 0.15 m deep. The pregrowth tanks are supplied with water from a penaeid shrimp growing pond effluent and receive each day 7 m³ of outdoor cultured *Chaetoceros gracilis* at a density of 1.2 10^6 cells per ml.

Results

Spawning : As shown in Table 1, spawning is obtained all year long. When the gonads are well ripe, only one jump of temperature is needed. Fertilization rate is high (more than 90%) and straight-hinged larvae appear within 24 hours.

Larval rearing : Mean survival rate between day 2 and 10 is 70%. The best survival rate (83%) between day 2 and 10 was obtained in the 10 m³ U-shaped rearing tank (rearing 11). Density before settlement is adjusted to 2,000 larvae per litre, as it was shown by AQUACOP (1979) that higher density at that stage induced a delay at settlement.

Treatment with Treflan for preventing fungal attack was abandoned after rearing 3, as it proved to be useless when compared with untreated tanks. Treatment with chlorine involved a complete mortality at day 6 in rearing 5 and was used no longer. Sulfadimerazine action in bacteria control gives good results (AQUACOP, 1979) and this antibiotic is used routinely.

The algae mixture *Isochrysis* sp.—*Monochrysis lutheri* has been abandoned after rearing 10. In this rearing, the mixture *Isochrysis* sp.—*Monochrysis lutheri* was replaced by the mixture *Isochrysis* sp.—*Chaetoceros gracilis*. Results are given in Table 2. The use of the two tropical strains mixture *Isochrysis* sp.—*Chaetoceros gracilis* leads to a faster growth and a better survival rate at day 10. In the well-lighted 10 m³ U-shaped tank (rearing 11), algal multiplication was higher than their consumption by mussel larvae and this tank did not require to be complemented in algae each day.

Settlement : In rearing 11, the settlement rate was estimated at 45%. This rate takes into consideration the spat settled on the collectors and on the walls of the 10 m³ tank (Table 3).

Pregrowth : The commercial spat size of 6-7 mm was reached in one month after the end of

TABLE 1. *Larval rearing of* Mytilus viridis *in the COP*

Larval rearing No.	1	2	3	4	5	6	7	8	9	10	11
				1978					1979		
Date of spawning	Feb.	Apr.	June	June	July	Oct.	Nov.	Jan.	Jan.	May	July
Number of D-larvae at day 2 (x10^6)	7.4	10.6	1.6	7	3.9	26.4	18.2	16.7	29.3	11.5	30
Mean temperature (°C)	27.5	28.1	27	26.5	*a*	26	25.7	27.1	28	26.7	24.3
Survival rate (%) at day 10	81	76	63	59		56	74	*b*	78	65	83
Day of beginning of settlement	19	15	15			16	18		18	15	16
Day of end of settlement	26	22				26					26
Commercial size spat (x 10^4)	0.35	1.03		0.43		1.04	1.98		1.6	0.31	1.8
% Spat/D-larvae at day 2	5	10		5		4	11		5	3	6

(*a-Mortality after treatment with* 1 *ppm total chlorine and b-Mortality by bacterial disease*)

TABLE 2. *Influence of algae mixture* Isochrysis *sp.* — *Monochrysis lutheri and* Isochrysis *sp.* — Chaetoceros gracilis *on the larval growth of* Mytilus viridis

Day	Rearing 10*				Rearing 11**	
	Isochrysis sp. *Monochrysis lutheri*		*Isochrysis* sp. *Chaetoceros gracilis*		*Isochrysis* sp. *Chaetoceros gracilis*	
	Size (μ)	Survival rate (%) from day 2	Size (μ)	Survival rate (%) from day 2	Size (μ)	Survival rate (%) from day 2
3					76	
4					81	
5	90.6	95.4			85	
6	100.8		116.6	89.5	93	
7	109.7	81.8	134.3		115	
8	119		144.4	81.7		
9	129.7	72.3	171.4		154.3	
10	151		192.2	80 (c)		
11	164	66 (c)	206.3			
12			237.1		220	83 (c)
13	206		252			
14			280		255	
15	233		settlement		286	
16					settlement	
17	settlement					

* Rearing 10 in 800 l tanks.
** Rearing 11 in 10 m³ tank.
(c) Screening on 100 μ mesh.

TABLE 3. *Estimation of the settlement rate in rearing* 11

Number of pediveliger larvae at day 15	$20\ 10^6$
Mean number of spat per collector	$72{,}000 \pm 18{,}700$ ($p=0.05$)
Total number of spat on collectors (day 34)	$5.8\ 10^6 + 1.5\ 10^6$
Number of spat settled on walls (day 34)	$3.1\ 10^6$
Settlement rate (between 34 and day 15)	45%

settlement in rearing 1, 2, 3, 4. In rearing 11, two months were needed to reach the same size of 6-7 mm, because of the overload of pregrowth tanks (Table 4). The best growth was obtained in the raceway supplied with a flow of 7 m³ per hour of a shrimp growing pond effluent. In concrete 15 m³ pregrowth tank, a better growth was achieved with spat lying directly on the bottom of the tank, than with spat settled on net collectors.

tors in the rearing tanks. In rearing 11, the great part of spat settled on the walls of the tank indicates that the eighty collectors were insufficient to ensure whole settlement.

The use of the effluent of shrimp growing ponds as water supply for pregrowth tanks induces inequal pregrowth results, according to the phytoplanktonic quality of the ponds

TABLE 4. *Growth in weight* (W mg) *and size* (L mm) *of spat of* Mytilus viridis *in reearing* 11

Days after settlement	Pregrowth					
	net collectors		tank bottom		the raceway	
	W (mg)	L (mm)	W (mg)	L (mm)	W (mg)	L (mm)
24	4.1	2.5				
30 ..	4.2					
36* ..	5.9	3.3	5.9	3.3	5.9	3.3
43 ..	7.8	3.5			9.7	4.2
50 ..	11.7		18.3	4.6	22.2	
57 ..	13.9	4.4	25.6	5.8	60	6.6
64 ..	25.8	4.6	44.7	6.3		

* Partial removal of spat from collectors.

DISCUSSION

The use of tropical strains of *Isochrysis* sp. and *Chaetoceros gracilis* improved the quality of food. As a matter of fact, these two strains stay alive and keep on dividing in larval rearing tanks. Using these two species as food, the 11th rearing which was conducted in the 10 m³ tank at the lowest mean temperature of 24.3°C with important daily variations (3.5°C) gave any way good results.

Compared to the eight 800 l tanks, operating the 10 m³ tank is easier for an equivalent production and consumes less water. The use of such rearing tanks, cheap and easy to build increases the efficiency of the method.

Improvement in the settlement rate could be obtained by hanging up more nylon net collec-

water. This cause of variations is partially reduced by the daily supply of 7 m³ outdoor cultured *Chaetoceros gracilis* in pregrowth tanks. The present pregrowth equipment set limits the potential marketable spat production at the COP at about 2.10^6 spat per month.

These first results achieved by the COP agree with the high interest which has been brought on mussel culture in Indo-Pacific region since a few years. In Tahiti where no species of mussels live in natural environment, a large-scale culture operation had to rely on a hatchery spat production. Since 1978, maturations and induced spawnings from imported broodstock are routinely achieved throughout the year and the larval rearing method proves to be reliable.

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SEASONAL CHANGES IN THE GONAD AND ASSOCIATED NEUROSECRETION IN THE GREEN MUSSEL *PERNA VIRIDIS*

U. H. MANE AND R. NAGABHUSHANAM*

Marine Research laboratory, Ratnagiri-415 612

ABSTRACT

The green mussel *Perna viridis* spawns in Bhatia Creek at Ratnagiri from July end to early September and again in February-March. The analyses of organic composition of gonad showed a close relationship with the reproductive cycle ; the fat content reached its peak in mature gonads (in July and January) whereas glycogen in immature ones (in May-June and November-December). High protein content in mature gonads declined after the release of sex products. The neurosecretory cycle studied with the help of histological details of the cerebral ganglia during different seasons showed a close relation to the histological stages obtained in gonads. The release of secretory material was found to be one of the factors, besides environmental factors like salinity and temperature, responsible for spawning of the green mussel.

INTRODUCTION

RECENTLY some attention has been given to the culture of green mussel *Perna viridis* along the west coast of India (Qasim and Achari, 1972 ; Rao *et al.*, 1975, 1976 ; Qasim *et al.*, 1977). Considering the interest generated in mussel culture, there is not much information available on the biological aspects including growth, reproduction and biochemical composition of the green mussel except for the studies of Rao *et al.* (1975, 1976), Wafar *et al.* (1976) and Qasim *et al.* (1977) from Goa Coast. The present investigation on aspects of annual reproduction, associated neurosecretion and the seasonal biochemical composition was undertaken during 1971-72 and 1974-75 with the view of understanding the physiology of *Perna viridis*.

MATERIAL AND METHODS

The mussels were collected at low tide from Bhatia Creek near Ratnagiri once in a fortnight during March 1971 to March 1972 and again once in a month during March 1974 to March 1975. Mussels ranging 70-80 mm in shell length were shucked and the gonads were fixed in aqueous Bouin's fixative prepared in sea water. Mussels ranging from 45-55 mm in shell length were dissected for cerebral ganglia and these were fixed in above fixative. Totally 390 mussels were shucked for gonads and 276 for cerebral ganglia. The gonad and ganglia were then dehydrated, paraffinized and sectioned at 8-9μ. The gonad sections during each month were stained with Ehrlich's haematoxylin and eosin whereas the ganglia sections were stained with Mallory's triple and Gomori's chrome alum haematoxylin.

REPRODUCTIVE CYCLE

Perna viridis in spawning condition shows pronounced differentiation in gonad colour ; a female is bright orange red and a male whitish yellow. Gonad tubules extend into the mantle region during maturation. After spawning the gonad and the mantle collapse in thickness. The gonad histological study through different seasons showed following changes and figured

* Department of Zoology, Marathwada University, Aurangabad-431 004, India.

as percentage frequency of mussels in each month in Fig. 1. The spawned gonad had the reproductive tubules largely emptied but a few residual eggs or sperms remained. Samples in this condition occurred from second fortnight of September. The disintegration of residual products began due to considerable accumulation of phagocytic cells in the tubules. Through second fortnight of September and October the vesicular connective tissue between the tubules began occupying greater portion between the tubules and later entire gonad passed into neutral stage and the sex could not

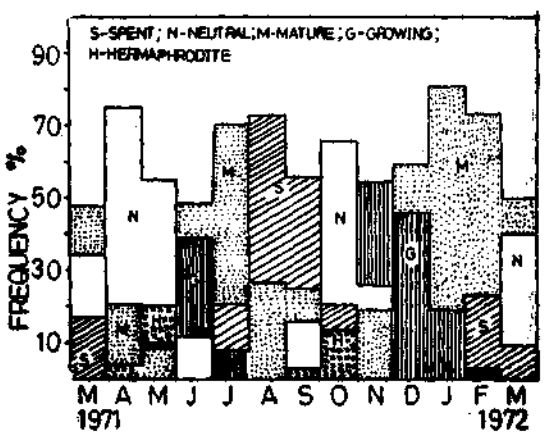

FIG. 1. The gonad of *Perna viridis* in different histological stages represented as mussels in the respective stages during 1971-72.

be distinguished. As gonad development started from November the follicle streaks became wide and were lined by primary sex cells. During the process of maturation in January the sex cells occupied much of the gonad tubules which had expanded considerably. At this stage the colour differentiation over gonad and mantle became apparent. In February few mussels spawned which resulted in the occurrence of partially spent males and females in the samples collected during the month. Those spawned in February ultimately passed into neutral condition after disintegration of unspawned sex products left in the tubules but those which did not spawn still remained in mature condition. The samples collected in April showed subsequent increase in the neutral gonads and less mature mussels without any partially spawned ones. The mature mussels showed accumulation of phagocytic cells in and outside of the gonad tubules probably indicating the process of disintegration of the sex products which failed to spawn. From June onwards once again the gonad maturation began continuing till second fortnight of July to repeat the cycle again.

Few mussels in April and May showed ova along the periphery and sperms in the central region of the tubules. Again in September and October few mussels showed large unspawned eggs in the central region and spermmatogonia along the periphery of the tubules. This rare occurrence of individuals with reproductive elements of both sexes is an indication that hermaphroditism is not a regular feature. As the germ cells have been observed to stop proliferating components of one sex and giving rise to those of the other sex, hermaphroditism is said to be a purely transitional phase. Coe (1943) observed that the majority of pelecypods have separate sexes with an occasional hermaphrodite making its appearance. In *Modiolus demisus*, *Mytilus edulis* and *M. californianus* he did not observe any evidence of ambisexuality except undifferentiated gonads and only occasional hermaphroditism. Wiborg (1946) found that sexes of *Modiolus modiolus* are usually separate with hermaphrodites occurring in 2 to 8 per cent of individuals.

NEUROSECRETORY CYCLE

The morphological characteristics of the neurosecretory cells at the various stages of their secretory activity (Fig. 2) have been mentioned earlier in *Perna viridis* (*Mytilus viridis*) by Nagabhushanam *et al.* (1971). The average frequency of the neurosecretory cells in 4 successive stages of the secretory activity

during 1974-75 was determined by counting the number of cells under every stage (Fig. 3).

Stage I

This initial phase was distinguished by the presence of scattered neurosecretion granules in the cytoplasm when compared with other phases. Moreover, the maximum number of cells in this stage were found during post-monsoon and early part of summer and especially during October and April.

Stage II

In this intermediate stage the neurosecretion droplets were concentrated around the nucleus. This stage was particularly seen during late summer and late postmonsoon.

Stage III

The accumulation of neurosecretion granules in the axon hillock is the main characteristic feature in this stage. The number of cells in this stage gradually increased during early monsoon and early winter.

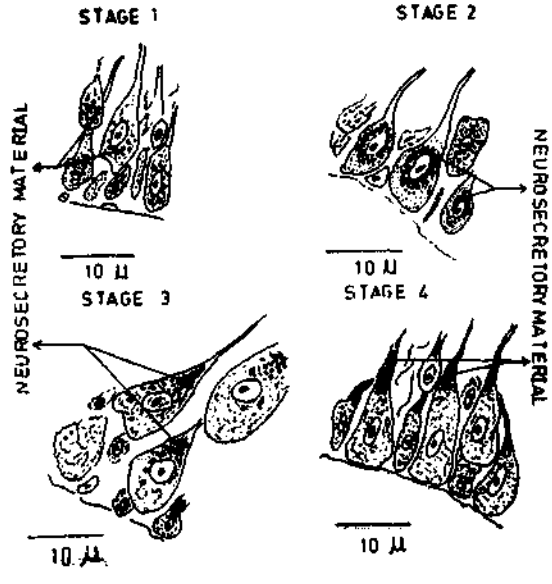

FIG. 2. Neurosecretory stages in the neurosecretory cells from cerebral ganglia in *Perna viridis*.

Stage IV

During this end phase the neurosecretion granules were found not only in the axon hillock but also in the proximal part of the axon. The maximum number of cells in this stage were found during late monsoon and late winter.

It appears that a seasonal cycle exists in the neurosecretory activity since the maximum of the various stages demonstrate a distinct sequence. The cycle begins immediately after spawning with maximum of stage I in April and

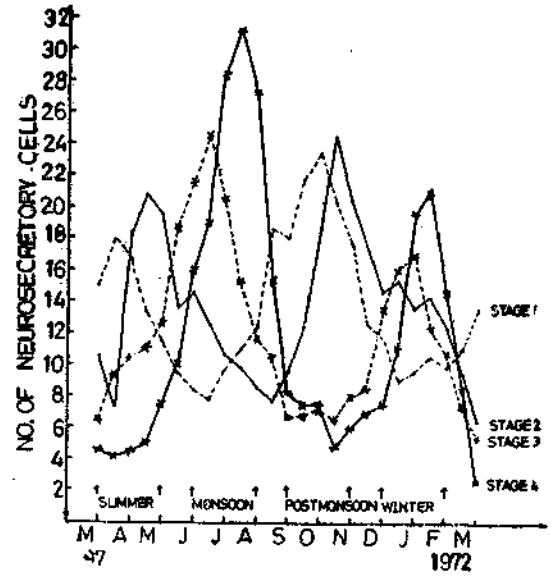

FIG. 3. The average number of neurosecretory cells from cerebral ganglia in *Perna viridis* through neurosecretory stages during 1971-72.

October. Thereafter Stage II follows which shows peak in May and November. During July and January a clear maximum in Stage III is seen followed by Stage IV with maximum in August and February. This means that the discharge of neurosecretory substance is considerably increased just before and during spawning. During the decline and termination of spawning activity in August and February a few number of cells in Stage I make their appearance.

Discussion

The complex of physical variables in the environment influence the sequence and the timing of events in the reproductive cycle of marine invertebrates (Giese, 1959). In temperate region gonad development and reproduction in marine bivalves have been correlated with the wide range of temperature fluctuations (Posgay, 1950 ; Loosanoff and Nomejko, 1951 ; Loosanoff and Davis, 1952 ; Dickie, 1953). *Mytilus edulis* at Conway rapidly spawns with rapid rise in temperature (Chipperfield, 1953). Studies on five different Mytilidae species (including *M. edulis*) indicated that factors behind optimal temperature play an important role in determining the potential reproductive season in each species (Wilson and Hodgkin, 1967). On the other hand, in tropical region, the reproductive cycles of many marine bivalves have been correlated with the salinity fluctuations (Rao, 1951 ; Durve, 1965 ; Alagarswami, 1966 ; Nagabhushanam and Mane, 1976).

Recent studies on *Perna viridis* from Goa Coast by Rao *et al.* (1975) have shown that in the green mussel breeding takes place throughout the year with maximum larval abundance in October-November and with little less abundance again in March. According to our study from Bhatia Creek at Ratnagiri the mussel show indication of spawning from July to early September. The mussels along Goa Coast also spawn during this time. Those mussels reaching full maturity in January respond to high salinity and temperature showing evidence of spawning for a short period from February to beginning of March. But many failed to spawn perhaps because of much increase in salinity and temperature which might not have been favourable to them to continue spawning. In Goa Coast the temperature of water is somewhat at low level in summer than in Ratnagiri (Bhatia Creek) though the salinities in both areas remain high. The studies by Rao *et al.* (1975) from Goa Coast are based on older and younger mussel populations. They found a restricted breeding in older ones taking place from July to December whereas in small ones, from January to April. Thus the population as a whole is said to have round the year spawning. In our study from Bhatia Creek, only the adult mussels belonging to 70-80 mm in shell length were studied.

Neurosecretion affecting reproduction in bivalve molluscs is known through the studies of Lubet (1955, 1959), Nagabhushnam (1963), Antheunisse (1963) and Nagabhushanam and Mane (1976). In *Perna viridis* a parallel relationship exists between neurosecretion and reproductive cycle. Both the cycles start in the beginning of post-monsoon and summer. With the maturation of sex cells in gonad tubules the secretory material starts accumulating in the cytoplasm of the neurosecretory cells and the number of such cells considerably increase in mature mussels in early monsoon and early winter. The massive release of cerebral neurosecretion occurs just before the mussels spawn. Further studies are needed to know the nature and localisation of the specificity of the neurosecretion material which will be of considerable importance in aquaculture of shellfishes.

The supply of nutrient reserves from the ingested food to gonads for growth and their utilisation in the biochemical synthesis of the developing gametes was reported in case of marine invertebrates by Giese (1959). In present study, as reported by Mane and Nagabhushanam (1975), the glycogen content in the gonad of *Perna viridis* increased from March to June and in October-November with the gametogenetic activity and ultimately decreased in mature ones. In turn, the fat content from April to July and from November to January increased. The fat content sharply dropped in the spawned gonad, whereas the protein content remained fairly high throughout all seasons and decreased only in the spawned gonad.

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PRELIMINARY STUDIES ON PRESERVATION AND TRANSPORTATION OF GREEN MUSSEL *PERNA VIRIDIS*

K. K. BALACHANDRAN AND P. V. PRABHU

Central Institute of Fisheries Technology, Cochin-682 029

ABSTRACT

Studies were undertaken on preservation in ice of Green mussel *Perna viridis* and meat shucked from fresh and boiled mussels. Biochemical and organoleptic changes taking place in the meat during the storage were followed to assess the shelf life inasmuch as the material is suitable for further preservation by canning. Similar studies were carried out on transportation of whole mussel and the meat in various conditions. The results of the above studies are presented in this paper. On further processing into canned product it has been observed that meat shucked from fresh mussel and preserved in ice yielded comparatively better product when canned in oil.

INTRODUCTION

THE OVER-DEPENDENCE on prawns as the principal raw material, the rapid growth in the number of sea food processing plants and the relatively stagnant position in the landings of prawns have forced the Indian sea food processing industry in general and the sea food canning industry in particular, to look for materials other than prawns for processing for their sustenance and any possible expansion. One such raw material of great promise and which the industry can easily obtain and which also commands a good export potential is mussel. Green mussel *Perna viridis* is available in substantial quantities from their natural beds along the west coast particularly from Calicut towards north. The recent successful development of culture technique for mussels has added a new dimension in its potential availability as well as its importance. Mussel is collected for its meat which is consumed in the nearby areas. The fact that mussel meat finds favour mostly with people of low economic status and that too when fish is scarce or costly, results in poor return to the producer. In the absence of a system of conservation by processing and preservation any poor response from the market will result in the wastage of this protein rich food. All these problems can be solved if it is utilised for processing by the seafood processing establishments.

Cochin has a concentration of seafood processing establishments, both freezing and canning, whereas mussel is not available in this area. Therefore the industry intending to process mussel meat will have to transport the raw material from the areas of its production. Transportation of live mussel has been found to be ideal, but involve the transportation of the bulk of shell which is a waste material. Data is scarce on the ice storage characteristics as well as transportation for subsequent use for processing. Therefore studies were undertaken on the shelf life of mussel, whole or meat in ice, transportation of mussel and meat in ice under different conditions and their preservation by canning, the results of which are presented in this paper.

Material and Methods

Green mussel *Perna viridis* collected from Calicut in the west coast was used in this study. They were allowed to starve for 24 hours before used for experiments. A set of experiments on transportation and the use of transported material for canning was carried out using mussel cultured at Calicut by the Research Centre of Central Marine Fisheries Research Institute. The following sets of experiments were carried out on iced storage *viz.*, iced storage of whole mussel and that of meat shucked from fresh as well as boiled mussel, the iced material being utilised for canning during the progress of storage. Whole mussel, meat shucked from fresh mussel, fresh shucked meat cooked in water and meat shucked from boiled mussel were transported separately in ice by road from Calicut and used for canning employing the method reported by Balachandran and Nair (1975). When iced stored whole mussel was used, the meat was shucked from it in fresh condition and then used for canning. Moisture, fat, ash and total nitrogen (TN) were determined according to the AOAC (1975) methods. Glycogen was estimated by the method of Vande Kliey (1951). Non-protein nitrogen (NPN) was estimated on the trichloracetic acid extract of the meat by the Kjeldahl's method.

Results and Discussion

Proximate composition of the meat of green mussel is given in Table 1.

Table 1. *Proximate composition (%) of meat of the green mussel* Perna viridis

Moisture	..	78.24 — 80.28
Protein	..	11.08 — 12.61
Fat	..	2.38 — 3.02
Glycogen	..	5.36 — 7.91
Ash	..	3.60 — 4.21

Changes taking place in the various fractions of whole mussel, meat shucked from fresh mussel and that from boiled mussel during iced storage are presented in Table 2.

Table 2. *Changes taking place in the mussel composition during iced storage*

Nature of material	Days of storage	Moisture (%)	TN (%)	NPN (%)	Glycogen (%)
Stored in ice	0	78.36	1.93	0.352	5.43
Whole mussel	2	81.26	1.87	0.361	5.06
	3	82.34	1.81	0.314	3.04
	4	82.40	1.82	0.272	3.01
Fresh shucked meat	0	78.36	1.98	0.352	5.43
	2	81.49	1.82	0.324	4.92
	3	82.64	1.80	0.302	3.16
	4	82.91	1.76	0.264	2.95
Boiled and shucked meat	0	74.27	2.03	0.382	3.94
	2	76.59	1.98	0.317	3.03
	3	76.43	1.96	0.310	2.76
	4	76.86	1.89	0.312	2.58

Progressively iced stored material when canned in refined ground nut oil and examined gave the organoleptic rating as given in Table 3.

TABLE 3. *Organoleptic rating of canned mussel meat processed out of correspondingly iced stored material*

Days of storage	Material used	Overall organoleptic rating
Initial	A	Very good
	B	Very good
	C	Good
2	A	Good
	B	Good
	C	Fair
3	A	Good—Fair
	B	Good—Fair
	C	Fair
4	A	Fair
	B	Fair
	C	Fair—Poor

A — Whole mussel.
B — meat shucked from fresh mussel.
C — meat shucked from boiled mussel

Chinnamma *et al.* (1970) have reported that fresh mussel preserved in ice remained in organoleptically acceptable condition upto 9 days. Chinnamma George (1974) reported that frozen mussel meat prepared out of whole mussel iced stored for 8 days remained in acceptable condition only for 15 weeks whereas the fresh frozen mussel meat remained in acceptable condition for 40 weeks. In the present study it has been observed that canned product prepared out of iced stored whole mussel or fresh shucked meat yielded a product good in organoleptic characteristics upto 2 days storage. The colour of the meat, its flavour and juiciness scored always better than the boiled and shucked meat. Meat shucked from boiled mussel lacked in colour, flavour and juiciness compared to the other two products and scored only fair even on the second day of storage. Therefore it follows that the ideal period of iced storage for mussel to be used for subsequent canning is 2 days either as whole mussel or meat shucked from fresh mussel.

Similar results were obtained when the cultured mussel transported in different forms were canned. Whole mussel as well as meat shucked from fresh mussel yielded better products as far as the colour, flavour, juiciness and general acceptability of the products are considered which observation is in conformity with those made on the iced mussels grown in the wild. However, the only defect encountered with the fresh shucked meat is that on subsequent canning there takes place minor distortion in the shape in certain samples. Therefore it is recommended to the industry that to make the transportation operations economic it is better to shuck the meat from fresh mussel and transport in ice.

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EXPERIENCES IN THE TRANSFER OF TECHNOLOGY OF OPEN SEA MUSSEL CULTURE UNDER THE OPERATIONAL RESEARCH PROJECT

S. J. RAJAN AND V. SELVARAJ*

Central Marine Fisheries Research Institute, Cochin-682 018

ABSTRACT

The paper deals with the problems encountered while implementing a programme on the transfer of technology of open sea mussel culture to the fishermen at Kovalam, near Madras under the Operational Research Project on 'Blending Sea Farming with Traditional Capture Fisheries'.

INTRODUCTION

MARINE fisheries development in the country has been impressive during the past three decades. But the benefits accrued have not helped the poor fishermen engaged in artisanal fisheries and their per capita income has hardly improved. In order to benefit the fishermen and their family members, it was felt that blending of culture fisheries with traditional capture fisheries would help to enhance production and the earning capacity of the rural community.

The authors wish to thank Dr. E.G. Silas, Director, Central Marine Fisheries Research Institute for having suggested the problem and for the guidance given. Our thanks are also due to Dr. P. V. Rao, Central Marine Fisheries Research Institute for his valuable suggestions in the preparation of this paper.

OPERATIONAL RESEARCH PROJECT

An Operational Research Project on ' Blending Sea Farming with Traditional Capture Fisheries ' was started in April, 1978 at Kovalam, a fishing village, 35 km south of Madras. This fishing village has 175 families comprising a total of 975 fishermen. The per capita income is Rs. 369 per annum. The main aim of the project is to train fishermen in the methods of mariculture of fishes, prawns and molluscs so that these could be undertaken along with capture fisheries.

The Institute's field laboratory established at Kovalam in 1976 has found the feasibility of culturing mussels, lobsters and prawns. In phase I of the programme it was decided to introduce open sea mussel culture. One hundred youth of the village in the age group of 15 to 25 years were enrolled and grouped in batches of ten with an elected leader for each group. These people were trained for taking up mussel culture in the open sea off Kovalam.

Selecting a suitable area was a problem as traditional fishing areas and chartered navigational routes had to be avoided, which left only a part of the open coastal waters available for taking up the mariculture programme.

Kovalam has a small bay with a maximum depth of 8 m. There is a chain of huge submerged rocks in the 8 m depth range at a distance of one and a half kilometres from the shore. These rocks locally called as ' pavai ' form the eastern boundary of the bay. Within

* Present address : Kovalam Field Centre, Central Marine Fisheries Research Institute, Kovalam near Madras.

the bay area the sea bottom is mostly sandy with a few submerged patches of rocks.

At Kovalam the sea is calm from December to April *i.e.* post north-east monsoon period. From May to June which is the transition period from the north-east to the south-west monsoon, the sea becomes rough with high swells. These swells are more marked as the sea breeze starts blowing in the afternoon. From July to September there is a change in the direction of the wind and current. At this time the sea is calm whenever the current and wind are in the same direction and rough when they are variable. With the onset of the north-east monsoon in October there is a reversal in the direction of the current and wind. Cyclonic conditions with very rough sea and huge swells prevail in this area.

In 1976 when the experiments on open sea mussel culture were started at Kovalam, both seed and adult mussels *Perna viridis* were transplanted from Ennore a place 20 km north of Madras. The mussels established themselves well at Kovalam. There was spatfall from January to September at regular intervals and mussel spat settled profusely on the interdidal rocks.

A floating raft of size 4 metre square (16 sq. m area) was made out of Casuarina poles tied with coir and nylon ropes and mounted on four sealed 200 litre barrels which acted as floats. The raft was anchored in 8 metres depth with 4 grapnel type anchors of 20 to 25 kg weight each. Subsequently the raft size and anchoring arrangements were changed to reduce costs.

The minimum time required for seed mussels of size 15 to 20 mm to gorw to marketable size of 75 to 80 mm is 5 months. The high swells of May/June and cyclonic conditions of October/November, however, did not permit the rafts to be kept at sea. The mussels seeded in January/February grew well and reached 45 to 50 mm by May. Due to the roughness of the sea in May/June the raft got washed ashore. The second seeding done in July/August also had the same problem and the mussel could not reach marketable size before the north-east monsoon.

Earlier, in 1978, it was proposed to go in for pole culture in the shallower regions on the northern side of the bay. Teak wood poles of 9 m length and 50 to 60 cm girth at the bottom were pile driven to a depth of 2 metres below the sea bed using water jet. The area selected was in the 4 to 6 m depth range just outside the breakers.

The first seeding on poles was done in July/August. There was also profuse natural seed settlement on some poles, but soon the bark peeled off resulting in the loss of mussels attached to the poles. Fresh seeding was done after removing the bark, In November/December, 1978 and also in May, 1979 most of the poles were dislodged due to cyclonic conditions.

The results of these experiments at Kovalam showed that for a surf beaten coast, exposed to variable currents and winds and prone to cyclonic storms, the rafts must be more rugged and streamlined. It was then decided to try a submerged raft and the first such raft was designed and anchored in June, 1979. While the raft design was the same as for surface rafts, the float design was changed to keep the raft in the column water.

A new type of conical float was fabricated by cutting and shaping empty oil drums. The apex of these conical floats were weighted inside and to this was fixed a ring. To this ring a swivel was also provided. One end of a two metre long chain was shackled to the swivel. Six such floats were fixed to the raft, *i.e.* three on two opposite sides by shackling the free end of the two metre chain around the teak poles. The raft now slowly got submerged to

1½ m below the surface of the sea, *i.e.* the length of free chain between the conical float and raft. The buoys themselves now floated with the broadside up and their apex in the water partially submerged. The streamlined shape of the floats allowed waves to pass over without resistance. The swivel used in the float was to counteract the current action which tended to twist the chain. As the mussel ropes grew in weight the buoys got half submerged in water. The number of buoys to be used depends on their buoyancy in relation to the weight and number of ropes put on the raft which could easily be adjusted by experience. With the development of the submerged raft it was possible to keep the raft in position long enough for a harvest. The first harvest of mussels from such a raft was taken on 26th October, 1979 at Kovalam. A submerged raft is therefore considered better suited for this area at places where the sea bottom permits pile driving of the poles to half their length and where the tidal range is also high permitting easy handling. Submerged raft will perhaps be the only solution in places where there are strong winds and currents.

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EXPERIMENTAL CULTURE OF THE BLOOD CLAM *ANADARA GRANOSA* (LINNAEUAS) IN KAKINADA BAY

K. A. NARASIMHAM*
Central Marine Fisheries Research Institute, Cochin-682 018

ABSTRACT

On an experimental basis the culture of blood clam *Anadara granosa* was taken up in the Kakinada Bay. Three pens made of split bamboo screens, each measuring 100 m², were stocked with clams of 25.1, 23.4 and 24.3 mm mean size at the rate of 3,000, 7,000 and 14,000 per pen respectively in May 1979. In order to determine the optimum stocking density 6 dealwood cases were filled with mud upto 10 cm depth and were stocked with clams at the rate of 40 to 240/m². The results of these studies together with the environmental data are given in this paper.

INTRODUCTION

MEMBERS of *Anadara* Gray (Fam : Arcidae) are popularly called blood clams, ark shells and cockles—the last name because of their superficial resemblance to the European cockle *Cardium*. In a number of Southeast Asian countries *A. granosa* (Linnaeus) is fished and utilised as food. It is cultured in China, Japan, the Philippines, Thailand, Indonesia and Malaysia (Pathansali and Soong, 1958, Bardach *et al.*, 1972; Chen, 1976). Along the Indian Coast, *A. granosa* forms a fishery of some magnitude in the Kakinada Bay where an estimated 1,000 tonnes are landed annually (Narasimham, 1973). *A. granosa* was experimentally cultured in the Kakinada Bay and the results are given in this paper.

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* Present address : Kakinada Research Centre of C.M.F.R.I., Kakinada 533 002.

PHYSIOGRAPHY OF THE FARM SITE

The site selected for the farm is in the Kakinada Bay and is contiguous with the natural clam bed. It is about 300 m from the shore, north of Yetimoga and is sparsely populated by the clam. The bottom is muddy with 64% clay, 25% silt and 8% made of sand and dead shells. The farm site is in the subtidal region where the minimum depth of water at the low tide is 25 cm.

Monthly average values of temperature, salinity and dissolved oxygen from May to December 1979 are given in Fig. 1. In May 1979 the temperature was 33.5°C and after a gradual fall to 28.9°C in August there was a slight increase to 30.4°C in October and it touched the lowest value, 27.8°C in December. The salinity was high in May (33.66‰) and June (34.40‰). With the onset of southwest monsoon there was gradual fall in salinity until October when it reached (22.29‰). During 1979 southwest monsoon failed and hence the high salinity in June-September. In November it went down to 13.69‰. There was a slight increase to 15.30‰ in December. The

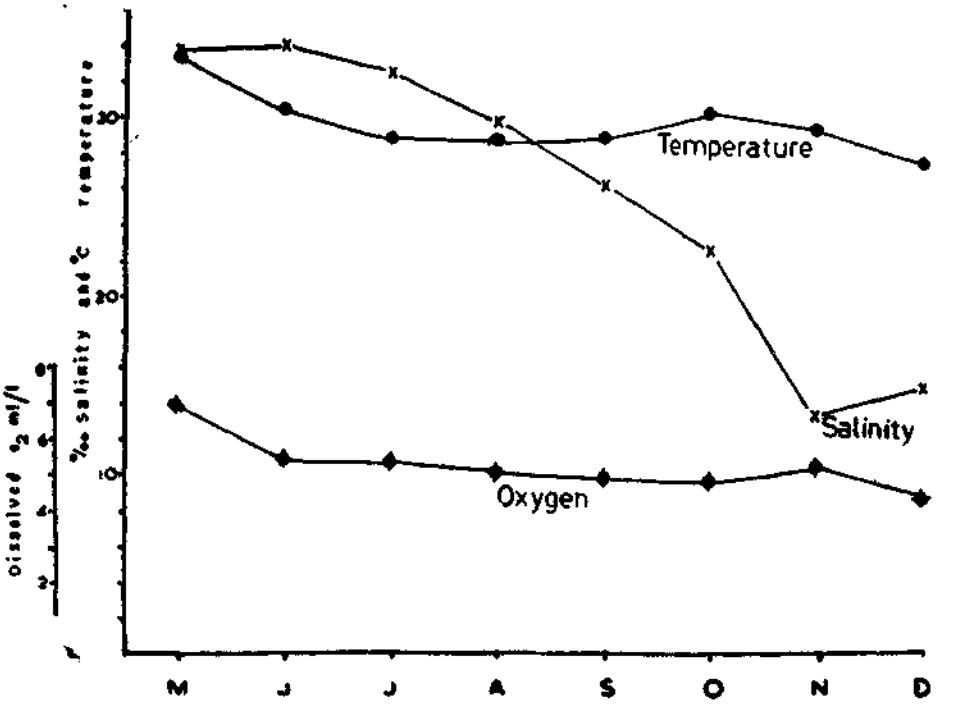

FIG. 1. Average monthly temperature, salinity and dissolved oxygen at the farm site.

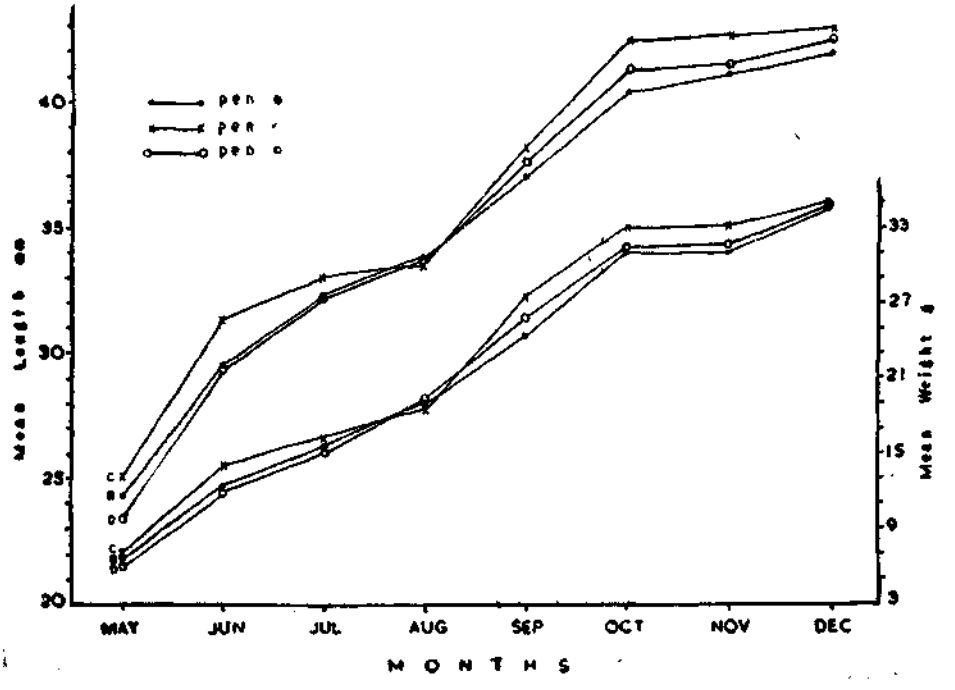

FIG. 2. Growth of *A. granosa* in length and weight in pens.

low salinity values in November and December were due to the northeast monsoon when a cyclonic storm hit the coast in the last week of November 1979. Generally the values of dissolved oxygen were high and from a high value of 7.0 ml/l in May1979 it showed a fall upto October (4.98 ml/l). In November it increased to 5.45 ml/l and touched 4.45 ml/l in December. The primary productivity was estimated by the dark and light bottle experiments conducted at fortnightly intervals during July-December (Table 1).

TABLE 1. *Average primary productivity of the waters at farm site*

Month	Gross production mg C/litre/day	Net production mg C/litre/day
July '79	0.2916	0.2144
August '79	0.3602	0.1672
September '79	0.5275	0.1544
October '79	0.3109	0.1201
November '79	0.2413	0.0886
December '79	0.4202	0.2401

CULTURE EXPERIMENTS

Clams were cultured in pens and in dealwood boxes during May 1979 to December 1979. Split bamboo screens, interlaced with hemp twine were used in constructing pens. Each screen measured 5 m long, 0.3 m high and interspaced with 6 numbers of 1 m long bamboo sticks which are driven upto 0.75 m into the mud to hold the screen vertically. Three pens, each measuring 100 m, were erected. Also an experiment on the off bottom culture in nylon mesh cages, suspended from a raft off Vakalapudi was conducted during May 1978 to October 1978.

Pen culture

The details of stocking of *A. granosa* in pens designated C, D and B are given in Table 2. The growth in mean length and mean weight of the clams in the three pens showed the same pattern (Fig. 2). The clams grew to 40.6 to 42.7 mm mean length and 31.1 to 33.0 g mean weight by October (Table 2). By December, they attained a mean length of 42.3 to 43.4 mm and a mean weight of 34.6 to 35.8 g. In spite of varying stocking densities, the growth in different pens was similar. The average growth during the culture experiment was 2.6 to 2.8 mm/month. It was fast in June-July and September-October and slow in August and November-December periods (Fig. 2). While the slow growth in August could not be related to the environmental parameters studied, that in November and December coincided with reduced salinity (Fig. 1). Similar retardation of growth during the period of low salinity was observed by Rao (1952) and Rao *et al.* (1962) in the bivalves studied by them.

Production

The clams were harvested by hand-picking at low tide on 23rd and 24th October 1979 (harvest 1, Table 2). Pen C with a stocking density of 3,000 clams, gave a production of 89.35 kg/100 m^2 with a survival rate of 90.3%. A production of 183.2 kg/100 m^2 was obtained in pen D, stocked with 7,000 clams and the survival was 84.0%. Pen B was stocked with 14,000 clams and the yield obtained was 385.3 kg/100 m^2 with a survival of 88.6%. Obviously the disparity in the survival rate in the pens is narrow and pen B with maximum stocking density gave the highest yield.

All clams harvested in October were restocked on the same dates in the respective pens to study their further survival and production at the end of the North-east monsoon. The clams were finally harvested on 20-21 December 1979 (harvest 2, Table 2). For the 7 month period the production in pen C was 84 kg with a survival of 79.5/, in pen D 167 kg with a survival of 66.7% and in pen B 357 kg against a survival of 73.8%. It may be seen that the extension of the culture period by 2 months

resulted in lower survival and production. In the last week of November 1979 a severe cyclonic storm hit the Andhra Coast and the bamboo screens of the farm, already weakened due to submersion for 6 months were damaged and had to be replaced.

The results show that culture of *A. granosa* for a 5 month period during May-October is advantageous when compared with the extended culture upto December.

Biology of cultured clams

The index of condition (% meat weight in total weight) was studied in the cultured clams and also from the natural bed for comparison. In September the average index of condition of clams in the pens varied from 22.2 to 24.0 and in the natural bed a comparable value of 23.1 was noticed (Table 3). In October-December the index for the cultured clams was invariably higher when compared with the condition obtained in the wild population.

TABLE 2. *Stocking and harvesting particulars of* A. granosa

Stocking	Pen C	Pen D	Pen B
Area of pen (sq. m)	.. 100	100	100
Date	.. 24.5.79	21 & 24.5.79	23 & 24.5.79
Numbers stocked	.. 3000	7000	14000
Size range in mm (mean)	.. 19-29 (25.1)	19-29 (23.4)	19-29 (24.3)
Total weight kg	.. 21	43.5	94
Mean weight (gm)	.. 7.0	6.2	6.7
Harvest 1			
Date	.. 24.10.79	24.10.79	23.10.79
Size range in mm (mean)	.. 36-52 (42.7)	34-50 (41.5)	34-49 (40.6)
Mean weight (gm)	.. 32.97	31.14	31.06
Estimated numbers	.. 2710	5883	12406
Survival rate (%)	.. 90.3	84.0	88.6
Production for 5 months (kg/100 m^2)	.. 89.35	183.2	385.3
Harvest II			
Date	.. 21.10.79	21.10.79	20.12.79
Size range in mm (mean)	.. 35-53 (43.4)	35-51 (42.8)	33-52 (42.3)
Mean weight (gm)	.. 35.20	35.79	34.57
Estimated numbers	.. 2386	4666	10327
Survival rate (%)	.. 79.5	66.7	73.8
Production for 7 months (kg/100 m^2)	.. 84	167	357

TABLE 3. *Average index of condition (meat wt. as % of total weight) in clams from the farm area and natural bed*

Month	Pen C	Pen D	Pen B	Natural bed
September '79	22.2	24.0	22.5	23.1
October '79	21.1	20.6	20.1	17.5
November '79	19.0	18.7	19.7	16.0
December '79	16.6	17.2	16.6	15.0

During September-October over 88% of clams from pens were in active stage of gametogenesis and none in spawning condition. By November spawning commenced in 30% of clams and by December, 65% were in spawning condition. In September a vast majority (98%) of clams from the natural bed were in resting stage with considerable phagocytic activity. In October 80% were in active stage. In November 18% and in December 80% were in spawning condition. The reproductive cycle in both the cultured and wild clams showed the same general sequence of events with considerable spawning activity in November-December period.

Box culture

Six wooden boxes, each measuring 0.25 m^2, were filled with mud obtained from the clam bed, upto 10 cm depth and stocked with clams on 23.5.1979. The clams were stocked in multiples of 10, the first box having 10 and the last box 60 clams. The clams varied in size from 19-25 mm with the mean length in different boxes varying from 21.6 to 22.4 mm and mean weight 5.0 to 6.5 g. Each box was covered with nylon netting (1 cm mesh), nailed with 1 m long bamboo sticks on 4 sides and these were driven into the mud close to the pens. Every month, during the last week, all the clams were measured and total weight obtained from each box; also the mud in the box was changed. In boxes 2, 3 and 6 a total of 34 clams died during the 7 month experiment period and on a number of occasions crabs were found in these boxes.

By October the clams had grown to a mean length of 39.5 to 40.7 mm (mean weight 25.5 to 28.7 g) and by December they attained a mean length of 41.0 to 42.4 mm (mean weight 27.7 to 30.2 g) in different boxes. The mean length of the clams from all boxes was within 1 standard deviation on either side of the mean of any particular box in a given month indicating that the growth of the clams in different boxes, stocked with varying densities, is not significantly different. The average growth for the experiment period worked to 2.7 to 3.0 mm/month which compares favourably with the 2.6 to 2.8 mm/month observed in the pens. The clams had grown fast during June-July and September-October period and growth was slow in August and November-December period, a condition identical to the one observed in the pens.

Off bottom culture

A total of 56 cages with nylon yarn meshes around 0.5 m diameter metal rings were suspended in tiers from a raft moored in 5 m depth off Vakalapudi light house (10 km north of the pen site). Stocking was done during March-May 1978 and the density per cage (area 0.2 m^2) varied from 9-396 clams. Observations showed near cessation of growth and very heavy mortality, varying from 70.7 to 100% and the experiment was abandoned in October 1978. The condition of the clams was also poor compared to the condition of natural bed population. However, 19 clams of mean length 25.5 mm stocked in a metal box filled with mud upto 10 cm depth and suspended in a cage from the same raft showed a growth of 2 mm/month. This experiment indicated that *A. granosa* does not thrive when suspended in the water column in metal ring cages without muddy substratum.

Remarks

There is scope for blood clam culture in the Kakinada Bay where there are extensive mud flats spread over several hundred hectares. *A. granosa* thrives and grows rapidly in intertidal flats of soft mud containing upto 90% silt and very few shellfish grow in such an environment. The present study has clearly demonstrated that a very high production of the blood clams per unit area can be obtained by simple transplantation of seed clams to sparsely populated areas which are suitable for growth. As *A. granosa* is cultured elsewhere without any enclosures, the pens can be dispensed, once the culture is undertaken in large areas. The growth rate in the pen is faster, compared to that obtained in Malaysia (Narasimham, 1968) or Taiwan. This would help to harvest the clams much earlier, thus reducing the culture period and production cost. Growth of *A. granosa* in dealwood boxes at densities of 40 to 240/m^2 did not reveal any significant difference in the growth. In Malayasia a final density of 300 to 600/m^2 is achieved after thinning. There is need to determine the stocking density for obtaining optimum growth and production in the Kakinada Bay.

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EXPERIMENTAL CLAM CULTURE AT MULKI, DAKSHINA KANNADA

K. SATYANARAYANA RAO* AND G. SYDA RAO
Central Marine Fisheries Research Institute, Cochin-682 018

ABSTRACT

The clams *Meretrix casta* var. *ovum* and *M. meretrix* were cultured in pens in fish farm at Mulki. *M. c.* var. *ovum* of the size range 16 - 28 mm stocked in February, 1979 showed a growth of 9 mm between February and June. The average weight of the clams increased from 5.3 gm to 12.6 gm in four months. *M. meretrix* 16 - 28 mm in size stocked in January showed a growth of 22 mm in four months at the end of which a maximum size of 43 mm was reached. In the four months period, the average weight of the clams increased from 5.3 gm to 17.6 gm. The scope for culturing clams of *Meretrix* spp. in the estuarine systems of Karnataka Coast has been pointed out.

INTRODUCTION

THE ESTUARIES along the west coast of India, particularly those of Karnataka are rich in clam resources (Jones, 1970; Alagarswami and Narasimham, 1973). In Karnataka estuaries clam beds comprising of a number of edible species like *Meretrix casta* var. *ovum*, *M. meretrix*, *Villorita cyprinoides*, *Paphia malabarica* and *Katelysia opima* are exploited regularly on a large scale for food and for the manufacture of lime. The present paper apprises the efforts undertaken at Mulki, Dakshina Kannada (South Kanara) in culturing clams of *Meretrix* spp. in pens constructed in brackish water area.

We express our sincere thanks to Dr. E. G. Silas, Director, Central Marine Fisheries Research Institute for his encouragement and to Shri K. Nagappan Nayar, Shri S. Mahadevan and Shri K. A. Narasimham, C.M.F.R. Institute for critically reading the paper and giving helpful suggestions. We express our gratitude to Shri M. H. Dhulkhed for offering facilities and to the Department of fisheries, Government of Karnataka for permitting us to make use of their fish pond at the Government Fish Farmat Mulki.

* Present address : Tuticorin Research Centre of C. M. F. R. I., Tuticorin, Tamil Nadu.

MATERIAL AND METHODS

Clam culture was carried out in four rectangular pens (Fig. 1) constructed in a fish pond of the Karnataka State Fisheries Department Fish Farm at Mulki. The fish pond is on the eastern side of the creek branching off from

FIG. 1. Four pens constructed in a pond of Karnataka State Government Fish Farm at Mulki where clam culture was carried out.

Pavanji Estuary at Mulki. The depth of the pond was 0.5 m at low tide and 1.5 m at high tide and the bottom was sandy with some

amount of silt and clay. The size of the pens were (1) 7.6 m × 3.65 m (area 27.74 sq. m), (2) 11.5 m × 3.65 m (area 41.97 sq. m), (3) 6.5 m × 3.65 m (area 23.72 sq. m) and (4) 11.9 m × 3.65 m (area 43.43 sq. m). The pens were constructed with bamboo strip frames 1.5 m × 1.5 m in size. The bamboo frames were driven 20 cm into the bottom of the pond and they were tied together vertically with coir ropes and to the supporting vertical casurina poles fixed on the outer side of the pens to prevent the entry of predators. Small sized clams of *Meretrix casta* var. *ovum* and *M. meretrix* were collected from Sambhavi, Gurpur and Coondapur Estuaries in January, February and March 1979 and stocked in the pens at different densities. Data were collected on their growth in length and weight at monthly intervals. Data on salinity and temperature were recorded weekly once.

Results

Stocking

Three pens were stocked with *Meretrix casta* var. *ovum* and one with *M. meretrix*. In pen one, 7,500 seed clams of *M.c.* var. *ovum* 4-8 mm in size (length) range collected from Gurpur Estuary were stocked in March 1979 at a density of 250 per sq. m. The total weight of the seed clams was 1.35 kg. In pen two, 2,620 clams of *M. c.* var. *ovum* 16-28 mm in size and 2.5-9.0 gm in weight (average weight 5.3 gm) were stocked at a density of 60 per sq. m. In pen three, 4,200 clams of *M. meretrix* 16-28 mm in size and 3.0-9.0 gm in weight (average weight 5.3 gm) were stocked in January 1979 at a density of 177 per sq. m. In pen four, 3,000 seed clams of *M. c.* var. *ovum* of the size range 4-8 mm weighing totally 0.54 kg were stocked in March 1979 at a density of 68 per sq. m.

Growth of Clams

The size of *Meretrix casta* var. *ovum* in pen two showed an increase as reflected in modal size shifting from 24 mm in February to 26 mm in March, 28 mm in April, 31 mm in May and 33 mm in June 1979 thus showing growth rate of 2 mm per month. In May, however it was higher being 3 mm. The total growth as indicated by the shifting of the modal size in the four months period was 9 mm and the increase in average size was 6.5 mm. The maximum size of clams increased from 28 mm in February to 37 mm in June. The average weight of the clams increased from 5.29 gm to 12.58 gm between February and June showing an increase of 7.29 gm. The average meat weight of the clams increased from 0.53 gm to 2.32 gm in the four months.

Meretrix meretrix exhibited an increase in size of 22 mm as shown by shifting of modes between January and May. A mode at 20 mm in January could be traced to 34 mm in March, 38 mm in April and 43 mm in May. The rate of growth was fast, 7 mm per month between January and March and later it was slow being 4 mm per month. The average size showed a steep increase from 23.6 mm to 34.0 mm between January and March and increased to 35.6 mm and 37.5 mm only in April and May respectively. The average weight of the clams increased from 5.3 gm to 17.6 gm. This shows an increment of 12.3 gm. The meat weight of the clams increased from 0.91 gm to 2.27 gm in the period showing a gain in weight of 1.36 gm.

Survival

A total of 2,110 individuals of *M. c.* var. *ovum* out of 2,620 stocked in pen two in February 1979 survived at the end of four months in June, '79 indicating a survival rate of 80.5%. Of 4,200 clams of *M. meretrix* stocked in pen Three in January, '79, 3,047 survived in May, 1979 showing a survival rate of 75.5%. 10,500 seed clams of *M. c.* var. *ovum* stocked in pens One and Four in March 1979 did not survive. These seed clams were of very small size being 4-8 mm in length. The bottom of the pens

consisted of a good percentage (63.5%) of fine sand with grain size of 63 μ to 250 μ and 4.7% of silt and clay (Table 1). The mortality of the young clams could be due to their being smothered by fine sand, silt and clay, especially the first component which was predominant.

TABLE 1. *Grain size analysis of the bottom sediment of pens in which clams were cultured*

Size of grains (in μ)		Percentages
>1000	..	1.1
500 — 1000	..	5.8
250 — 500	..	24.9
125 — 250	..	44.2
63 — 125	..	19.3
<63	..	4.7

A yield of 31.79 kg of *M. casta* var. *ovum* was obtained from pen Two 41.97 sq. m. in area. From pen Three with an area of 23.72 sq. m, a yield of 53.62 kg of *M. meretrix* was got.

Hydrological conditions

Salinity of the water in the pens increased slowly over a limited range of 32.16‰ in January to 34.87‰ in April and decreased to 33.50‰ in May when there was rain on some days and influx of freshwater from the upper reaches of Pavanji river. The salinity decreased further to 32.38‰ in the first half of June and fresh water conditions prevailed during the second half of the month following heavy monsoon rains.

The water temperature in the pens also increased gradually from 30.3° C in January to 33.3° C in April and decreased to 32.8° C in May. The temperature showed an increase again in the first half of June and dropped to 27.3°C in the latter half of the month with the commencement of rains.

TABLE 2. *Salinity and temperature of the water in pens in which clam culture was carried out at mulki*

Months	Salinity ‰	Temperature °C
January '79	32.16	30.3
February	32.65	30.5
March	33.96	31.4
April	34.87	33.3
May	33.50	32.8
June First half	32.38	34.2
June Second half	0.00	27.3

The slow rate of growth of *Meretrix meretrix* in April and May may be due to the water temperature being higher with a range of 32.8° to 33.3° C compared to 31.0° to 31.4° C in previous months. In the afternoons the temperature was often 34 to 36°C in the summer months of April and May which may be responsible for marked retardation in growth. *M. c.* var. *ovum* appears to be a hardy form as the rate of growth of this species was not affected.

REMARKS

The results obtained in the present work indicate that there is good growth of the clams *Meretrix casta* var. *ovum* and *M. meretrix* reared in pens in estuarine environment. The survival rate has been quite high being 80.5% and 72.5% in *M. c.* var. *ovum* and *M. meretrix* respectively. This may be due to slightly larger initial size of the clams chosen for stocking taking into consideration the bottom of the pens which was containing some silt and clay apart from sand. In clam culture, survival rate is known to vary widely over a range of 30 to 75% and 50% is considered quite satisfactory (Chen, 1976). The yield of *Meretrix* spp. obtained in the present work is also quite good and it could be stated the two species

M. c. var. *ovum* and *M. meretrix* would be suitable for culture in pens in shallow creeks and estuarine systems and adjoining low-lying areas along Karnataka Coast.

Resources survey carried out earlier by the authors has revealed the existence of extensive seed beds of *M. c.* var. *ovum* and *M. meretrix* particularly the former species in six estuaries of Dakshina Kannada District, namely Gurpur, Pavanji, Sambhavi, Swarnanadhi, Sitanadhi and Coondapur Estuaries. Therefore there is no likelihood of dearth of seed clams which would be needed for starting culture operations.

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DISTRIBUTION OF SEED CLAMS OF *MERETRIX CASTA* (CHEMNITZ) IN VELLAR ESTUARY

P. V. SREENIVASAN
Central Marine Fisheries Research Institute, Cochin-682 018

ABSTRACT

A survey conducted in Vellar Estuary during April and May 1978 on the abundance of the seed of *Meretrix casta* (Chemnitz) indicated that the densest population occurred in the mid-portion of the estuary. The population was from scarce to absent near river mouth and moderate in the upper reaches. From the environmental characteristics associated with such distribution, it is evident that *M. casta* prefers a salinity range of 30 - 32‰, a depth of 50 - 100 cm and a sandy muddy substratum.

INTRODUCTION

THE BACKWATER clam *Meretrix casta* (Chemnitz) occurs commonly in estuaries along both the coasts of India. Observations on the ecology of this species are available in the works of Parulekar *et al.* (1973) from Mandovi, Cumburjua and Zuari Estuaries and Harkantra (1975) from Kali Estuary. Though extensive beds of this species occur in Vellar Estuary (Lat. 11° 29' N and Long. 799 49' E), so far no information is available on its pattern of distribution. Therefore a survey was undertaken in April 1978 and was repeated in May 1978 to find out the areas of higher and lower densities of the seed and the associated environmental parameters. Results of this survey are presented in this account.

I am grateful to Dr. E. G. Silas, Director, Central Marine Fisheries Research Institute, Cochin for suggestion of the work and for his constant encouragement. My thanks are also due to Shri K. Nagappan Nair and Shri S. Mahadevan for their comments and improvements of the paper. I also thank Dr. N. Jayabalan for his help in the grade analyses of the sediment.

MATERIAL AND METHODS

During the investigation, it was observed that the tidal influence in Vellar Estuary extended from the river mouth to 10 km interior and *M. casta* was available even at that distance. This 10 km stretch was sampled by fixing 30 stations, with 3 stations in each km range, numbered 1 to 30 commencing from the upper reaches of the estuary to river mouth. The samples were taken from 1 m^2 area using a frame. Specimens inside the frame were collected with a hand showel, washed and weighed. The total number of live specimens was estimated by direct counting or by sub-sampling. A maximum of 100 specimens were measured for length. Mud samples were taken using an iron core sampler.

For grade analysis of sediment, the class units 0.5 mm, 0.25 mm, 0.125 mm and 0.025 mm based on Wintworth System were used. Twenty gram of the sediment was treated with 20 ml of H_2O_2, heated for 30 minutes in order to oxidise the organic matter and then dried. The residue was sieved through a set of sieves of apertures 0.5 mm, 0.42 mm, 0.25 mm, 0.125 mm, 0.063 mm and 0.025 mm. Following Wells

TABLE 1. *Distribution of the seed of* M. casta *in Vellar Estuary (in order of abundance) and associated environmental characteristics*

Density by weight (in g)	Density by numbers	Length range (in mm)	Station No.	Depth (in cm)	Salinity (‰)	Temperature (°C)	Substratum	Sediment particle size (in mm)
7015	108355	1-12	20	100	32.90	29.4	Sand mud	0.063-0.125
4384	11159	4-27	16	50	31.50	29.3	Sand mud	0.063-0.125
3590	8608	4-21	15	75	31.05	29.5	Fine sand	0.125-0.250
3151	7544	1-25	14	100	30.89	29.0	Sand mud	0.063-0.125
1977	11367	1-12	23	50	33.07	30.0	Sand mud	0.063-0.125
1746	4689	1-21	12	100	29.09	29.0	Fine sand	0.125-0.250
1713	15900	1-12	22	100	33.45	30.0	Sand mud	0.063-0.250
1367	571	16-30	1	200	14.61	28.2	Medium sand	0.125-0.420
1287	6935	1-18	11	25	29.08	29.4	Mud	0.025-0.063
1134	10095	1-12	9	100	27.84	28.7	Medium sand	0.125-0.500
942	23495	1-12	10	25	25.52	29.2	Sand mud	0.025-0.125
910	3291	1-33	4	50	18.82	29.5	Fine sand	0.125-0.250
893	1219	1-42	8	200	25.66	28.7	Fine sand	0.125-0.420
736	110	22-39	2	100	14.74	28.3	Sand mud	0.063-0.250
728	9257	1-12	24	100	32.98	28.9	Sand mud	0.063-0.125
720	12145	1-21	7	75	27.04	29.4	Sand mud	0.063-0.125
683	112	16-42	3	25	15.34	29.2	Medium sand	0.125-0.420
262	16650	1-12	6	100	25.44	29.0	Sand mud	0.063-0.125
68	1533	1-18	13	50	30.88	28.7	Sand mud	0.063-0.250
3	212	1- 9	5	100	24.53	28.8	Fine sand	0.125-0.250
2	112	1-12	26	50	32.80	29.8	Fine sand	0.125-0.250
1	75	1- 9	30 & 27	25	33.04	29.4	Fine sand	0.125-0.250
0	—	—	17, 18, 19, 21, 25, 28 & 29	50-150	31.60-33.60	29.1-30.1	Clay or Fine sand	0.025-0.063 0.125-0.250

(1957), broader grouping of the substratum was made as coarse sand : 0.5 mm, medium sand : 0.250 — 0.5 mm, fine sand : 0.125 — 0.25 mm, mud : 0.063 — 0.125 mm and clay : 0.025 — 0.063 mm. Combination of these groups such as sandy—muddy was used when any of the combinations exceeded 90%.

Results

Abundance of *M. casta* in different stations (average of the two sets) and the associated environmental characteristics are presented in Table 1. Most dense populations were observed at stations 20, 16, 15 and 14 which are located in the middle portion of the estuary. Populations were moderate in the upper reaches where only large individuals were present. Near the river mouth the population was from scarce to absent. According to Durve (1963), both high and low salinities affect the physiological activities of *M. casta* and this explains low abundance in these two areas.

According to Wells (1957), at a given location the abundance of clam depends upon the number of larvae set and survived at the spot since there is little movement among larger individuals. Therefore, the distribution of seed and adults of *M. casta* would depend upon the factors which influenced the larvae to settle.

Very high concentration of *M. casta* was observed in areas where the depth was 50 cm to 100 cm, salinity between 30 — 32‰ and the substratum sandy muddy. Moderate population was recorded in areas both with low and higher salinities, shallow and deeper areas and with sandy and sandy muddy bottom. The population was totally absent in clayish substratum as well as in high saline areas. Preference of *M. casta* towards fine sandy to sandy muddy areas may be due to its burrowing habit as well as to the fact that the clam larvae prefer to settle on solid substratum covered by a thin layer of sediment (Carricker, 1952).

Based on the above observations, it can be suggested that to achieve better results of clam culture with *Meretrix casta*, the site should preferably be of sandy muddy substratum at a depth of 50 to 100 cm and in salinity of about 30‰.

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GROWTH OF THE CLAM *MERETRIX CASTA* (CHEMNITZ) TRANSPLANTED IN THE VELLAR ESTUARY

P. V. SREENIVASAN

Central Marine Fisheries Research Institute, Cochin-682 018

ABSTRACT

The backwater clam *Meretrix casta* on transplantation in the Vellar Estuary, was observed to grow from 7.5 mm in length in September 1976 to 41.5 mm by October 1977 attaining 34 mm in 13 months. Corresponding increase in breadth, depth and weight was 29.9 mm, 24.8 mm and 31.07 gm respectively. Growth was retarded in October-December when low salinity prevailed in the estuary. Growth was fast in January-March when salinity and temperature began to rise. Growth was moderate to low from April to September which was the spawning period for the species in Vellar Estuary. Growth of the transplanted individuals was much faster than those in the natural bed. Dimensional relationships of breadth, depth and weight on length of the cultured clams were also estimated.

INTRODUCTION

AMONG the commercially important clams, the backwater clam *Meretrix casta* (Chemnitz) is one of the most important along the coasts of India. Large quantities of this clam are collected for food and for its use as bait. Shells are utilized for making lime. Studies on the growth of this species were made by Abraham (1953) from Adyar Estuary, Durve (1970, 1973) from Marine Fish Farm at Mandapam, Seshappa (1967) from Beypore and Korapuzha Estuaries, Salih (1973) from Cochin area, Parulekar *et al.* (1973) from Goa region and by Harkantra (1975) from Kali Estuary. Though rich beds of this species with an estimated production of 3300 tonnes was observed in Vellar Estuary (Anonymous, 1978), so far no information is available on the growth of this species from this area. An attempt was made to study the growth of *M. casta* on transplantation as well as in the natural bed during the period from September, 1976 to October, 1977 and the results are presented in this account.

I gratefully acknowledge Dr. E. G. Silas, Director, Central Marine Fisheries Research Institute, Cochin for suggesting the problem and for his encouragement throughout the period of investigation. My thanks are due to Shri K. A. Narasimham of this Institute for critically going through the manuscript and suggesting improvements.

MATERIAL AND METHODS

Seed of *M. casta* numbering 2,000 were collected from a clam bed in Vellar Estuary near railway bridge (6 km from river mouth) in September, 1976 and were transplanted into a dealwood box of the size 75 × 50 × 15 cm, containing sand upto 8 cm depth. The box was covered with a nylon fishing net and kept in the estuary near the Marine Biological Station (2 km from river mouth) at ½ metre depth below low tide level on the ground. Possibly due to persistent low salinity of the waters during the time of transplantation and also due to predation by crabs in spite of netting, heavy mortality was observed in September-December and by January, 1977 only 420 clams were living. Later on by strengthening the netting further mortality

due to predation was prevented. Sampling was done monthly once and each time 100 specimens were studied for length and a subsample of 30 specimens for breadth, depth and weight. The linear measurements were taken upto 0.1 mm and the weight upto 0.01 gm precision and were done as per the method followed by Abraham (1953). For the length frequency studies, 1 mm groupings were made. In November, 1977 the experiment was abandoned as the box was washed away due to unprecedented flood in Vellar. For comparative study, samples were also collected from the natural bed except in the monsoon season when floods in the estuary made the collections difficult. Data on salinity and temperature were also collected.

Observations

Length frequency of *M. casta* from the natural bed is given in Fig. 1. Due to fishing, settlement and growth of new broods and

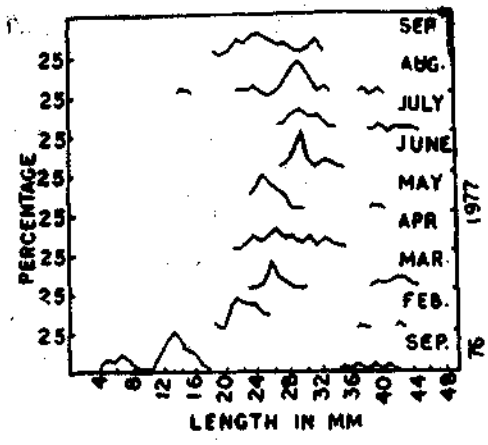

Fig. 1. Length frequency of *M. casta* in natural bed.

also possibly due to migration of adult clams (Parulekar *et al.*, 1973; Harkantra, 1975) considerable fluctuations were observed in the length frequency distribution. A prominent size group with mode at 14 mm in September 1976 can be traced to 22 mm in February 77, 26 mm in March, 27 mm in April, 30 mm in June and 32 mm in September thus showing a growth of 18 mm in 12 months.

Progression of modal groups in the transplanted sample is given in Fig. 2 from which it can be seen that the seed of *M. casta* transplanted at a length range of 4.0—8.0 mm in

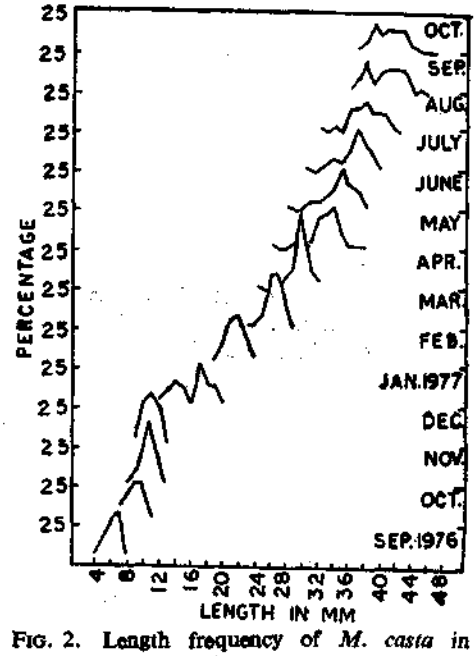

Fig. 2. Length frequency of *M. casta* in the transplanted sample.

September 1976 reached a length of 37-47 mm by October 1977 showing a minimum growth of 33 mm and a maximum growth of 39 mm. In Fig. 3 are plotted the monthly values of length and the corresponding values of depth, breadth and weight. It is evident that the clams measuring 7.5 mm in September 1976 have grown to 41.5 mm in October 1977. Corresponding increase in breadth was from 5.4 mm to 35.3 mm, in depth it was from 3.4 mm to 26.2 mm and in weight the increase was from 0.27 gm to 31.34 gm. Net growth observed in 13 months was 34 mm in length, 29.9 mm in breadth, 22.8 mm in depth and 31.07 gm in weight.

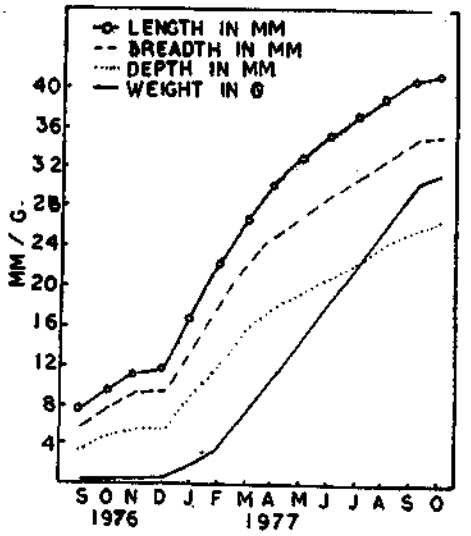

FIG. 3. Monthwise mean values of length, breadth, depth and weight of *M. casta* in the transplanted sample.

density varied from 2,500 to 5,000 live clams per square metre depending on the factors such as seed settlement, mortality, fishing and movement.

The rate of growth in length, breadth, depth and weight in the transplanted clams in each month is given in Table 1 from which it can be seen that the rate of growth was not uniform throughout the period of observation. From September to December the growth in length was only 3.9 mm, from January to March 15.4 mm, April to June 8.5 mm, July to September 5.3 mm and from September to October 0.9 mm. When studied in relation to the environmental parameters like salinity and temperature (given in the same Table) the slow growth period from September to December coincides with low salinity and temperature values due to monsoon. Fast growth was

TABLE 1. *Monthly rate of growth of* M. Casta *in length, breadth, depth and weight in relation to salinity and temperature*

Month	Growth in length (mm)	Growth in breadth (mm)	Growth in depth (mm)	Growth in weight (gm)	Temperature (°C)	Salinity (‰)
September 1976 ..	..	..	..	..	29.0	19.10
October ..	2.0	2.0	1.5	0.09	29.5	20.50
November ..	1.7	1.8	0.8	0.07	28.0	20.50
December ..	0.2	0.1	0.2	0.04	25.0	23.10
January 1977 ..	5.4	4.4	3.1	1.03	27.3	25.61
February ..	5.8	4.7	2.6	2.05	28.2	25.61
March ..	4.2	4.2	3.9	3.63	29.0	24.16
April ..	3.4	2.5	2.3	3.65	29.4	32.60
May ..	3.1	2.3	1.4	4.02	30.2	33.58
June ..	2.0	2.0	1.8	3.67	31.0	33.82
July ..	1.9	1.4	1.5	3.38	30.2	34.65
August ..	1.6	2.3	1.8	3.54	28.4	32.43
September ..	1.8	1.5	1.4	4.25	29.0	31.42
October ..	0.9	0.7	0.5	1.65	28.5	8.09

The growth observed in the transplanted clams was very fast when compared to the growth in the natural bed and such instance of high growth was earlier observed by Durve (1973) in *M. casta*. This may probably be due to less competition for space and food in the transplanted area than in natural bed where the observed in January to March when the temperature and salinity are rising. From April onwards, the growth was observed to be slow. During these months, settlement of seed clams was observed in the Vellar Estuary indicating that these months may be the active spawning period of *M. casta*. Abraham (1953) observed

that *M. casta* matures at a very small size of 12 mm and all the specimens among the transplanted ones are potential spawners and therefore a slower growth rate is probable. The retarded growth in October may be due to lowering of salinity and temperature due to the onset of monsoon.

A comparison of salinity and temperature and rate of growth, indicates that active growth was noticed when the salinity was 25‰-30‰ and the temperature 27-29°C, moderate to slow growth was observed when the salinity was 33‰-36‰ and a temperature of 30-31°C and the growth was retarded when the salinity was below 20‰ and the temperature 25°C. Slower growth rate observed by Durve (1970) in *M. casta* in the Marine Fish Farm may be due to high salinity and temperature in the farm.

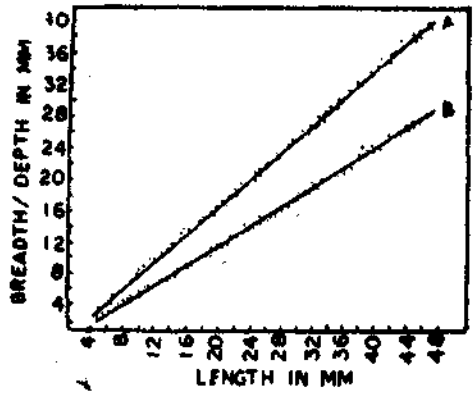

FIG. 4. Relationships of breadth (A) and depth (B) on length. (Dark line denotes the calculated values.)

Dimensional relationship

Relationships of breadth and depth on length (Fig. 4) and weight on length (Fig. 5) are expressed by simple allometric equation $y=a+bx$ where x is the length and y the variable and a and b are constants. The values obtained are :

Breadth :	- 0.1845 + 0.8324 x
Depth :	- 1.3195 + 0.6309 x
Weight :	- 0.000 4425 + 3.1476 x

When the correlation coefficient was calculated, it was observed that all of them are highly significant (r=0.995 and P=0.001). The

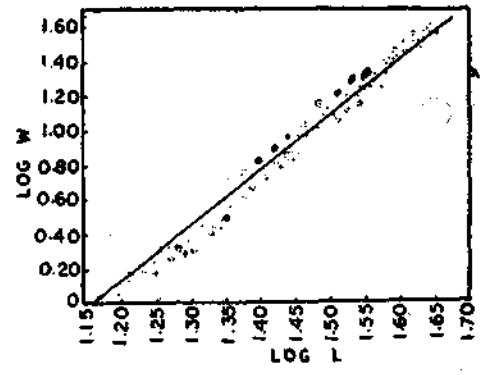

FIG. 5. Relationship of log W on log l. (Dark line denotes the calculated values.)

calculated values for depth, breadth and weight, when compared to the mean values of each month show a striking similarity except in higher size groups where the deviation was observed to be high.

DISCUSSION

Abraham (1953) concluded that *M. casta* spawned in August can attain 29.5 mm length by next April in Adyar Estuary. Durve (1970) observed a growth of 11.74 mm in length in 19 months for the same species in a pond at Marine Fish Farm at Mandapam. Later in another experiment, he (Durve, 1973) estimated a growth of 14.4 mm in length from November to April on transplantation. Salih (1973) observed a growth of 33.5 mm in 9 months and 35.4 mm in 11 months in different broods. In Goa region, Parulekar *et al.* (1973) calculated that an individual of *M. casta* of 5.9 mm in length recruited in October 1971 reached

38.9 mm by the end of 1972 showing 32 mm growth. Harkantra (1975) observed that *M. casta* of Kali Estuary 15 mm in length reaches 40 mm by June and then the growth was found to be static. Except for the observations of Durve (1970), in all other cases the growth of *M. casta* was not uniform but showed variations with changes in the environmental conditions. Growth was found to be retarded or slowed down twice in a year first due to low salinity caused by monsoon floods and secondly due to spawning activity which was always observed to be in the months of high salinity and temperature. Such a trend is noted in the present observation also.

There is much similarity in the growth in *M. casta* reported by Abraham (1953) from Adayar Estuary and that of the same species from Vellar Estuary. Probably this may be due to their close location and also were affected by similar factors especially hydrological conditions.

Present investigation is significant for aquaculture of clams since it indicates the suitable period of seeding when the growth is fast (January to February) and harvesting when the growth is retarded (earlier to October) so as to get maximum return.

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SOME ASPECTS OF REPRODUCTIVE BIOLOGY OF THE CLAM *KATELYSIA OPIMA* GMELIN FROM RATNAGIR

R. NAGABHUSHANAM AND U. H. MANE*
Department of Zoology, Marathwada University, Aurangabad-431 004

ABSTRACT

The reproductive study on *Katelysia opima* of Ratnagiri Coast showed two spawning periods — a major one occurring in October-November and a minor one in March-April. An attempt to find out correlation of the neurosecretory changes in pyriform cells of the cerebral ganglia with that of gonad index during different seasons showed a close relationship with the reproductive cycle. Laboratory experiments showed that the secretory material from pryriform neurosecretory cells of the cerebral ganglia was responsible for the release of sex products in the clam.

INTRODUCTION

THE SOFT CLAM *Katelysia opima* is one of the few clams collected for eating purpose by the coastal populations, especially the fishermen community. The investigation on various aspects of the biology of the clam was undertaken at Marine Research Laboratory, Ratnagiri on the west coast of Maharashtra State during the years 1970-74. The clam fishing in this area is mainly dependent upon 3 species - *Meretrix meretrix*, *Katelysia opima* and *Paphia laterisulca* locally called Mula, Wati and Tasari respectively.

ENVIRONMENT

Katelysia opima is primarily marine and is found burrowing in sand and mud in quite shallow waters but secondarily invaded backwaters and estuaries in which its distribution is confined to regions in the closest vicinity of the sea (Rao, 1952). It is never found far up the river mouth where salinities always are too low. In Kalbadevi Estuary the clam bed of *K. opima* is situated at the seaward region of the estuary in muddy intertidal zone. The clams face exposure to low tide and to salinity fluctuations in all seasons and could tolerate different salinities in summer and monsoon (Mane, 1974 a, b). In summer, because of considerable solar radiation, water temperature increases but in monsoon and winter it decreases. The annual salinity distribution pattern can be divided into two major periods. From June/July to September, the amount of influx of freshwater from Kalbadevi River is considerable. The saline water in the estuary is brought through tidal action. The high salinity in rest of the seasons is a common phenomenon due to combined effects of evaporation and low influx of river water. Each tidal cycle brings seawater in all seasons. Based on monthly

* Present address: Marine Research Laboratory of Marathwada University, Ratnagiri-415 612.

records (once a week with average) the temperature and salinity during the years 1970-71 and 1972-73 are plotted in Fig. 1.

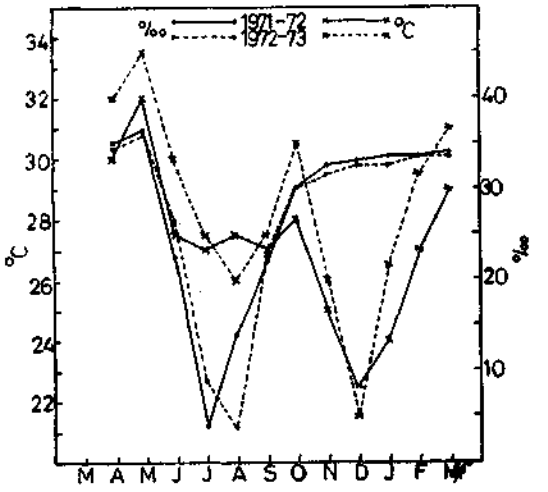

FIG. 1. Salinity and temperature variations over the clam bed in Kalbadevi during 1970-71 and 1972-73.

Reproduction

The gonad index, representing the percentage of the body weight attributed to that body component (Giese, 1969), was studied during different season in 1970-71. The observations indicate that maximum increase in the index occurs immediately after monsoon and again at the close of the winter and decreases before beginning of the winter and in middle of summer (Fig. 2). This increase and decrease indicates the plump and flaccid condition respectively, of the gonad.

The gonads of about 1200 adult clams (above 2 years old) were fixed in Bouin's fixative during different seasons of 1970-71 and were subsequently stained with Delafield's haematoxylin and eosin after paraffinizing and sectioning at 8-10μ.

The spawned gonad has the reproductive tubules largely emptied of the gonad products but a few residual eggs or sperms remain. The wall of the tubule does not retain the original round or oval shape. The vesicular tissue between the tubules expand. Later, usually be middle of winter, developing sex cells are seen round the edges of the tubules. This proceeds till winter ends. In beginning of summer both males and females are nearly ripe with tubules filled with ripe eggs or sperms and virtually no vesicular tissue remains. The clams at this stage are ready to spawn. Spawning takes place in first half of summer. Through

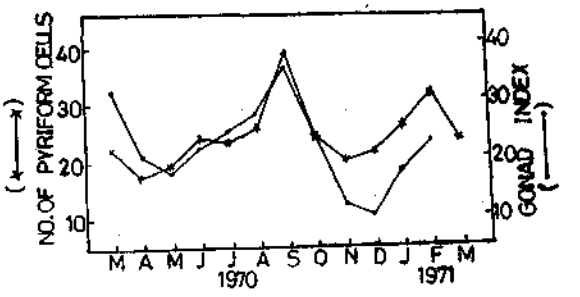

FIG. 2. Correlation of the gonad index (during 1970-71) and the number of pyriform neurosecretory cells with secretory material in cytoplasm from cerebral ganglia (during 1971-72) of *Katelysia opima*.

the second half of summer and beginning of monsoon most of the residual sex products are resorbed. The development then proceeds in gonad and slowly the males and females reach ripe condition through remaining part of monsoon. Soon after the end of monsoon spawning takes place which continues till beginning of winter. The clams in different maturity stages during different seasons show that much of the ripe sex products are released in outside environment after monsoon than in beginning of summer.

From Indian waters both continuous and discontinuous spawning seasons in bivalve molluscs have been reported and regarding the nature of their reproductive cycles even in the same species, clams from one locality are known

to differ from those of other. *Meretrix casta* on the east coast spawns twice in a year during April-May and again in September, whereas the same species spawns several times in a year in Adyar Estuary (Abraham, 1953) and Mandapam Camp (Durve, 1964). *Donax cuneatus* from Madras Coast and those of Palk Bay has only single reproductive cycle although in the former locality the breeding is relatively much longer (Nayar, 1955). In contrast to this, Rao (1968) in the same wedge clam reported prolonged spawning from January to June along Madras Coast. In *Katelysia opima* only one spawning season occurs in Adyar Estuary at Madras (Rao, 1952) but the same species in Kalbadevi Estuary at Ratnagiri spawns twice in a year according to our study.

One of the most important questions in the problem of reproductive synchrony is the mechanism which regulates gametogenesis and spawning in tropical marine bivalve molluscs. The water temperature does not fall as in temperate waters and it remains comparatively high throughout the year except for a few degrees drop in winter. The low salinity in monsoon in Kalbadevi Estuary affects the activity of the clams. Filtration and respiration slow down because the clams for majority of period remain with shell valves closed (Mane, 1975 a, b). The clams could tolerate considerable low salinity in monsoon than in summer (Mane, 1974 b). Due to unfavourable conditions gametogenesis and maturation take place slowly ; through monsoon gonads of the clams reach plump condition. As the salinity of the estuary increases along with temperature after the close of monsoon, the plump clams receive favourable conditions and spawning begins which continue till mature sex products are released out. With further increase in temperature and salinity along with the rematuration process in following seasons the gonads of the clams reach plump condition in early summer — the period when the clams receive optimum temperature and salinity. Thus the plump clams begin to spawn due to the pressure changes in the estuary.

Neurosecretory cycle

Loosanoff and Davis (1952) stated that control of gametogenesis and spawning in bivalve molluscs may be either exogenous or endogenous. Exogenous factors (like temperature and salinity) continually influence the gonad through all stages of development. Conversely, endogenous control (by an endocrine system) could be more or less independent of changes in environmental factors and gametogenesis would be affected only indirectly by environmental parameters which would act on endogenous regulating system. Endogenous system affecting gametogenesis in bivalve molluscs does exist in the cerebral ganglia (Lubet, 1955 ; Nagabhushanam, 1963 ; Nagabhushanam and Mane, 1976). Cerebral ganglia of 120 numbers of *Katelysia opima* (above 2 years old) were serially sectioned after fixing in Bouin's fixative and paraffinizing and were stained with Mallory's triple and Gomori's chrome alum haematoxylin during different seasons of 1971-72. The microscopic observations show 2 types of neurosecretory cells—type I having pyriform and type II oval in shape. These two types further differ from each other in cell and nucleus measurements, situation of nucleus and nucleoli and conspicuous staining reactions to the cytoplasmic neurosecretory . aterial. The observations of neurosecretory material and counting of the two types of cells with the secretory material in cytoplasm in different seasons have shown that the number of pyriform cells varies seasonally than the oval cells. This seasonal variation in release of secretory material from pyriform cells shows close relationship with the plump and flaccid condition of the gonad. The clams with increased gonad index show more pyriform cells with secretory material in cytoplasm whereas those with decreased gonad index show less such pyriform cells (Fig. 2).

Lubet (1955) observed active phase of neurosecretion during gametogenesis in *Mytilus edulis* and *Chlamys varia*. A copious release of cerebral neurosecretion occurs a short time before the gametes are released. During the period of restocking of the gonads that follows gamete release, there is a renewed repletion of neurosecretory cells of the cerebral ganglia. The decreased neurosecretory activity appears to coincide with the sexual resting phase during which the gonads undergo considerable involution. A hypothesis that postulates the existence of a link between the neurosecretory activity of the cerebropleural ganglia and the morphogenesis or activity of the gonad could be formulated. Information on such activity of secretory material from cerebropleural ganglia and its relation to reproduction exists through studies of Antheunisse (1963) in zebra mussel *Dreissena polymorpha*, of Nagabhushanam (1963) in *Crassostrea virginica*, and of Nagabhushanam and Mane (1976) in *C. gryphoides*. Such activity also exists in *Katelysia opima* according to our present study.

The neurosecretion and reproduction run parallel, but it is essential to find whether this endogenous factor has something to do with the spawning of the clams. For this, laboratory experiments were conducted to remove cerebral ganglia from *Katelysia opima* during the plump condition of gonad at the close of monsoon and again at the beginning of summer in 1972-73. The gonads of such clams after treating for histological studies have shown that the females are more sensitive than males after cerebralectomy in emptying the gonad tubules (Table 1). It is possible that the secretory material from the cytoplasm of pyriform cells of cerebral ganglia is responsible for emptying of the mature sex products from gonad. Further study is needed to find out the nature and composition of the secretory material which will be very useful in use of hormones for induction of sexual maturation and spawning as is being used in finfish controlled reproduction.

TABLE 1. *Response of cerebralectomized* Katelysia opima *in relation to gonad products release*

Treatment	Sex	Number of clams tested	Number of clams Responded
Monsoon—1972			
Ablation of cerebral ganglia	Female	20	16
	Male	10	7
Control	Female	10	2
	Male	10	1
Beginning of Summer 1973			
Ablation of cerebral ganglia	Female	24	22
	Male	17	13
Control	Female	15	1
	Male	12	—

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A CRITICAL REVIEW OF PROGRESS AND PROBLEMS OF PEARL CULTURE IN INDIA

K. ALAGARSWAMI

Central Marine Fisheries Research Institute, Cochin 682 018

ABSTRACT

A major breakthrough was achieved when techniques for the production of cultured pearls in the pearl oyster *Pinctada fucata* were successfully developed for the first time in 1973 at the Central Marine Fisheries Research Institute. Since then progress has been made in several biological and technical areas of pearl culture. Training programmes have been conducted to extend the know-how to the maritime States and Union Territories.

The paper recapitulates the recent achievements in pearl culture in India and identifies the areas which require a major thrust to strengthen the technological base. It also outlines the immediate prospects for the development of a pearl culture industry in the country.

INTRODUCTION

THE PEARL culture industry of the world owes its origin and development to Japan where it started with the initial success of late Kokichi Mikimoto in the production of a few half-pearls in 1893. The industry in Japan, based on the species *Pinctada martensii* (= *P. fucata*), became firmly established by 1926 with the production and marketing of spherical pearls and a pre-war peak production of 4 tons was achieved in 1938. The production suffered during the war only to prosper again after 1948 and an all-time record production of 130 tons of cultured pearls was achieved in 1966. Since then the production has declined due to several factors. Cahn (1949), Alagarswami (1970), Shirai (1970), Furukawa (1972), Wada (1973) and Mizumoto (1979), among others, have dealt with the story of pearl culture in Japan.

The first pearl culture farm outside Japan was established in Australia in 1956 and it has become the second important country for the production of cultured pearls. The Australian production, which is based on the silverlip pearl oyster *Pinctada maxima*, was 107,777 numbers of round pearls, 62,179 baroques, 413,964 half-rounds and 161.7 tons of pearl-shells in the year 1971. Franklin (1973) and Hancock (1973) have briefly described the pearl culture industry of Australia. Papua New Guinea has a few pearl culture farms in Fairfax Harbour (DEA, Canberra, 1970). A moderate pearl culture industry, using *P. maxima*, *P. margaritifera* and *Pteria macroptera*, exists in the Philippines (Blanco, 1972). Malaysia, where pearl culture began in Sabah in 1963, has a small-scale industry producing about 50,000 half-round pearls in *P. margaritifera* (Fisheries Division, Malaysia, 1972). Thailand has a very moderate pearl culture industry along the Andaman Sea Coast based on *P. maxima* and *Pteria macroptera*. Burma's pearl culture operations using *P. maxima* are located in the Mergui Archipelago. Indonesia has had mixed success with *P. maxima* in Butung island at the southern end of Celebes (Shirai, 1970).

Pearl culture has also spread to Hong Kong, Korea and Fiji Islands. A project was established in Sudan a few years ago for production of cultured pearls in *P. margaritifera* (Reed, 1965). It is seen from the above account that pearl culture industry is confined to the Indo-Pacific region.

Interest in pearl culture in India dates back to 1913-14 when late James Hornell drew up a scheme for the establishment of a pearl oyster farm at Krusadai at the head of the Gulf of Mannar. The experiments which were actually started there in 1933 and continued intermittently over a period of about three decades, yielded some basic data on the rearing of pearl oysters under captivity but failed to produce the techniques for the production of cultured pearls (Devanesan and Chidambaram,1956; Devanesan and Chacko, 1958). Experiments taken up later from 1956 at Sikka in the Gulf of Kutch also were not successful (Pandya, 1974). A breakthrough was achieved in 1973 in pearl oyster farming and production of culturedpearls in the Indian pearl oyster *Pinctada fucata* (Gould) at the Central Marine Fisheries Research Institute which started the experiments at Tuticorin in 1972 (Alagarswami, 1974 a). Alagarswami (1974 b) and Alagarswami and Qasim (1973) have indicated the prospects of pearl culture in India.

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Recent Developments in Pearl Culture in India

Pearl oyster resources

The species of pearl oyster which is of primary importance is *Pinctada fucata* (Gould), formerly referred to as *P. vulgaris*. The species contributes to the natural pearl fisheries of the Gulf of Mannar and Gulf of Kutch. Hornell (1922) produced a treatise on the Indian pearl fisheries of the Gulf of Mannar and Mahadevan and Nayar (1968, 1973) have added more recent information on the ecology of pearl banks and pearl fisheries. Easwaran *et al.* (1969) have described the pearl fisheries of the Gulf of Kutch. The pearl oyster resources of both the regions are well known for their wide fluctuations. After a successful series of pearl fishery from 1955 to 1961 yielding an estimated 86 million oysters, the Gulf of Mannar beds have entered the unproductive cycle. The Gulf of Kutch fishery which is very subdued in magnitude producing an average of about 17,000 oysters per fishery conducted every three or four years, has also not revived after the fishery of 1966.

Alagarswami and Qasim (1973) have indicated the presence of pearl oysters in the beds of Gulf of Mannar which provided the initial material of 2,742 oysters during 1972-73 for the pearl oyster farm at Veppalodai near Tuticorin. From this scarce situation it was possible to step up the collection from the natural beds in the gulf to a total of 49,028 pearl oysters, of which 27,730 were *P. fucata*, during the years 1975-1978 (CMFRI, 1979). Signs of slow but steady revival of *P. fucata* populations have been observed during the above period at least in some of the beds. Alagarswami (1977) has discussed the problem of larval transport and settlement of pearl oyster in the Gulf of Mannar. It is probable that the revival noticed in some of the shallower beds such as Devi paar is due to the farming of pearl oysters in the nearby Veppalodai area (CMFRI, 1979).

An interesting phenomenon noticed with regard to the pearl oyster resource is the settlement of pearl oysters in the inshore waters including harbours. The breakwaters of Tuticorin Harbour constructed in 1972-73 were found colonised by the pearl oysters in 1974 (Alagarswami, 1977). Spatfall was observed in the fishing harbour under construction at

Vizhinjam near Trivandrum which has led to the collection of spat on a regular basis although natural beds of pearl oysters have not so far been located in this region (CMFRI, 1974).

However, in both the harbour areas the pearl oyster populations consist of a number of *Pinctada* species including *P. fucata*, *P. sugillata*, *P. chemnitzii*, *P. anomioides* and *P. margaritifera* (Alagarswami, 1977). Incursions of *Pinctada* species, other than *P. fucata*, on the natural beds of Gulf of Mannar were also noticed for the first time in 1975. Constituting 34.3% of the total collections in 1975, the non-*fucata* component increased to 67.5% in 1976 and declined to 50.6% in 1977 and to 15.3% in 1978 implying the transient nature of this population on the natural beds (CMFRI, 1979).

A preliminary resources survey carried out by the Central Marine Fisheries Research Institute in 1978 in the Andaman and Nicobar Islands has shown the occurrence of the blacklip pearl oyster *P. margaritifera* in moderate quantities in the islands. This is a potential resource for pearl culture which needs development.

Attempts have been made for the collection of spat in the farm areas at Veppalodai and Tuticorin harbour basin. Although spat of the non-*fucata* species could be collected in the inshore farms, *P. fucata* spat are very rare. It is likely that the larvae of *P. fucata* are more sensitive to the turbid conditions of the inshore waters and show preference to clear waters for settlement. *P. fucata* component is getting gradually reduced in the Vizhinjam Bay as the silt load is increasing.

Hatchery production of seed, in the light of the above situation, is of great importance. The initial experiments in this direction have provided the techniques for the controlled spawning of pearl oyster and rearing of larvae upto early veliger stage.

Pearl culture technology

The techniques for the production of free, spherical cultured pearls in the Indian pearl oyster *P. fucata* were developed for the first time in India in 1973 (Alagarswami, 1974 a). In the first series of experiments, a rate of 55.8% production was achieved. Subsequently, it has been possible to raise the production rate to 62.8% in single implantations (Alagarswami, 1974 a). Multiple implantation of nuclei has further enhanced the pearl production rate to 180.6% in respect of the number of oysters used (Alagarswami, 1974 c). The possibilities of re-use of oysters for a second crop of pearls have also been indicated.

It has been stated earlier that the inshore waters have a good potential for production of species such as *P. chemnitzii*, *P. sugillata* and other flat oysters in large quantities. It was found that although they produce pearls, they are not of good quality in terms of lustre (CMFRI, 1979). Using the mantle grafts of *P. fucata* on these species it has been possible to improve the quality slightly but the pearls are still inferior to those produced by *P. fucata*. Some of the factors which are responsible for the poor production rate and quality in these species are that they have a thick byssal mass which hinders insertion of nuclei; the epithelium of the gonad is very thin and hence the tear of tissue is greater; the concavity of the left valve is very shallow and therefore the oysters cannot take in large nuclei; and the lustre of the nacre of the shell itself is watery and not bright. It has to be noted that these species are not used in any part of the world for pearl production.

Preliminary data have been obtained on the growth of cultured pearls in *P. fucata* in the Indian waters. The nacreous deposit on the radius of the pearls has been found to be 0.22 mm, 0.26 mm and 0.32 mm on a 3 mm diameter nucleus in 94-108, 161 and 191 days respectively. On the 4 mm diameter nucleus,

the deposition of nacre was 0.31 mm in 161 days. On the 5.81 mm diameter nucleus, the growth was 0.26 mm in 159 days (Alagarswami, 1975). Against the 0.3 mm average annual increase in diameter reported by Cahn (1949) for the growth of cultured pearls in the Japanese waters, the rate of growth of nacre has been found to be much faster in the Indian waters. This enables considerable reduction in the post-operative culture duration.

The Japanese pearl culturists use methods of physical exhaustion through under nourishment and forced ovulation or egg extraction to condition *P. fucata* for the surgery, taking advantage of the thermal stratification of the bays (Alagarswami, 1970; Wada, 1973). In the inshore waters of 5-8 m depth where the pearl oyster farm has been located at Tuticorin the above methods cannot be applied. Menthol has been found to be an efficient chemical for narcotising the oysters for surgery. Experiments have been carried out on the economic use of menthol and minimum doses required have been ascertained. The recovery of oysters on reimmersion in fresh sea water is fast.

Some success has been achieved in developing techniques for the production of shell-bead nuclei (Velu *et al.*, 1973). Conch (*Xancus pyrum*) shell bits have been processed to round beads of different diameters. These have been successfully used in experimental production of cultured pearls. Surgical tools required for the nucleus insertion in the oyster have been produced indigenously (Alagarswami and Sivarajan, 1975).

Farm management

There is very little choice of sites along the coast of the mainland of India to establish pearl culture farms. Elsewhere in the world, the farms are located in the protected bays surrounded by chains of islands. These bays have deep and calm waters with moderate currents to bring in food and flush the droppings from the farm. Such areas are very rare to come across along the coast of the mainland of India. The coastal areas are rather shallow and are subject to rough sea conditions during the monsoons and are also prone to cyclones. The Gulf of Mannar, to some extent, offers a compromise with regard to availability of oysters and workable farming conditions.

The experimental pearl oyster farm could be maintained round the year at Veppalodai off Tuticorin in the Gulf of Mannar. The group of small islands at the head of the gulf provided some protection to the farm at Krusadai Island. The experimental farms in the harbour basins both at Tuticorin and Vizhinjam are only temporary measures and these areas cannot be considered for any long-range programmes. At present farming is done in depths of 4-8 m at these sites.

Raft culture method was introduced in the mariculture activities of India beginning with the establishment of the pearl oyster farm at Veppalodai. In general, the rafts are made of teak poles, lashed with coir ropes and buoyed up with metal barrels. Unit rafts, measuring about 6 × 5 m, are moored independently at two ends using iron anchors and chains. The rafts are clear off the sea surface by about 50 cm. For holding the pearl oysters, frame nets and baskets made with an iron-rod frame and nylon webbing are used. Protective coats of paints are given to all the iron materials. Rafts made of timber sizes and secured with bolts and nuts have also been used. The rigid rafts have to be sturdy to withstand the rough sea conditions.

Aspects of environment for promoting the growth of the oysters and production of good quality cultured pearls have been given some attention. The data have shown that the shallow waters (depth 4-5 m) off Veppalodai do not promote proper growth of oysters (Chellam, 1978). The probable reasons for this are that the turbidity is very high, the coastal

currents are swift and biofouling and boring is heavy. In spite of these handicaps of the environment the rate of pearl production and quality of cultured pearls have been found to be good at Veppalodai.

Fouling and boring have been found to be the two major factors causing deterioration in the health of oysters at Veppalodai. Barnacles, bryozoans and ascidians are the dominant fouling organisms present almost throughout the year and occurrence of spat of edible oysters and *Avicula* is seasonal. The sponge *Cliona vastifica* and the polychaete *Polydora ciliata* contribute to heavy boring on the shells. Alagarswami and Chellam (1976) dealt with these problems in detail and related mortality of oysters in the farm to the heavy biofouling and boring of shells. Frequent shell cleaning operations to scrape the fouling organisms disturb the metabolism of the oysters and the tender growth shoots on the shell margin are eroded at each operation affecting the growth of the oysters. Experiments on the control of the boring organisms have shown that the polychaete borers can be controlled by immersion of the oysters in fresh water and the boring sponge can be killed by brushing the external surface of the shells with 1% formalin (Velayudhan, 1983).

Pearl oysters maintained in the Tuticorin harbour basin show better growth and higher survival rate. The basin is calm, protected against waves by the breakwaters and oyster cages are suspended at a depth of 6-8 m. The water is relatively clear and fouling and boring of the oysters is less compared to the conditions at Veppalodai. Data on farming of pearl oysters in the harbour basin would indicate the minimum environmental conditions required for successful farming of oysters in the inshore waters.

Farming of pearl oysters at Krusadai Island which was revived by the Department of Fisheries of Tamil Nadu in 1978 is reported to have produced good results with regard to the growth of oysters.

At Sikka on the Gulf of Kutch, pearl oysters have been farmed in a pool in the intertidal area constructed with stone embankments (Pandya, 1974). Oysters have also been maintained in shallow tanks on the shore. *P. fucata* of the Gulf of Kutch is an intertidal population unlike in the Gulf of Mannar and hence it could be successfully farmed in above such areas.

Training in pearl culture

The Central Marine Fisheries Research Institute offers training courses in pearl culture to aid further development in this field (CMFRI, 1977). The programme is conducted at two levels: a long term course (six months) at the managerial level and a short term course (six weeks) at the technician level. The former is a package programme of training to include all aspects of pearl culture and the latter is a specific programme on farm maintenance and oyster surgery only. During the years 1976-1979 one long-term course and two short-term courses have been completed. The courses are open to candidates sponsored by the governments of maritime States and Union Territories and Universities. Nationals from other countries, sponsored by the respective governments are also admitted into the courses.

Pilot project on pearl culture

On the industrial side, the only development so far has been the setting up of a pilot project for the production of cultured pearls at Vizhinjam by the Government of Kerala. The resource base for this project is the spatfall in the Vizhinjam Bay. The Central Marine Fisheries Research Institute has established that good quality cultured pearls could be produced from *P. fucata* occurring in the bay (CMFRI, 1974) which has led to the starting of the pilot project.

Research Thrusts for Future

The above review on the recent advances in pearl culture in India would show that the basic technology for commercial production of cultured pearls has been developed during the last seven years. However, several areas of research remain open for future investigations which are essential for strengthening the technology for improvements in resource availability, farming conditions, and production rate and quality of cultured pearls.

Production of additional resource

In view of the fact that production of pearl oysters in the natural beds is not steady and therefore undependable, the first priority is given to develop suitable technologies for assured supply of oysters. The Japanese pearl culture industry is not dependent on the wild populations of *P. fucata* and almost the entire supply of oysters comes from collection of spat using cedar sprigs as collectors (Alagarswami, 1970). The pearl-shell industry of Sudan was revived through the method of large-scale collection of *P. margaritifera* spat using split bamboo shelves in the Dongonab Bay in the Red Sea (Reed, 1962, 1965). Spat collection has not made any headway for *P. maxima* in the Australian waters and in the adjacent areas and only natural populations of the species support the pearl culture industry.

It has been seen earlier that spat settlement in the inshore waters, including the harbour basins of Tuticorin and Vizhinjam, is of a multi-species character and the component of *P. fucata* is very low. Techniques should be developed for the collection of *P. fucata* spat in the natural beds, particularly in beds such as Devi paar where oyster collections have been good in the recent years. In the Gulf of Kutch, the pearl oyster population is widely scattered on an extensive intertidal zone and spat collection should be attempted in the deeper areas adjacent to the flat. The occurrence of *P. margaritifera* in the Andaman and Nicobar Islands has been mentioned earlier. The oysters have been found to occur in the tidal flats and on the pillars of piers. Attempts will have to be made to find out if the split bamboo shelf method as practised for the species in the Dongonab Bay will prove successful in the Andaman Sea.

Hatchery production of *P. fucata* contributes to a small percentage of oysters used in pearl culture in Japan (Wada, 1975). Hancock (1973) has reported attempts at Port Moresby to develop hatchery techniques for *P. maxima*. Priority is being given at the Central Marine Fisheries Research Institute to develop a hatchery for pearl oyster at Tuticorin. Success has already been achieved in controlled spawning of *P. fucata* in alkaline sea water medium (Alagarswami *et al.*, 1983 a). The larvae have been reared in the laboratory upto the straight-hinge stage (Alagarswami *et al.*, 1983 b). Further development in this direction awaits identification of suitable micro-algae and mass culture of the same for feeding the larvae.

Work on the genetics of pearl oysters is very recent (Wada, 1975 a, b). *P. fucata* occurs in two distinct ecological habitats in the Indian waters, namely the deep beds (15-25 m) in the Gulf of Mannar and the intertidal flats of the Gulf of Kutch. It would be of interest to study the genetic resources of these two populations. The hatchery when it is developed, should attempt genetic improvement of the stocks through a breeding programme using the above two populations.

Improvement in farming technology

The technology of pearl oyster farming has evolved in Japan from the initial scattering of oysters on the sea floor through the pearl-string method to the raft culture of oysters which at present is the standard practice (Alagarswami, 1970). It is still the ideal system for rearing such large numbers of oysters under manageable

conditions in protected bays. Although the raft culture technique has been adopted in India, there is considerable scope for improving upon this system to develop methods with reference to the environmental conditions of each farming area. Flexible rafts made of ropes and floats are extensively used in the Seto Inland Sea of Japan where there is wave action.

Interest of scientists seems to have reverted back to the merits of culturing the oysters on the natural beds themselves. Yamaguchi and Hasuo (1978) have taken up comparative studies on the activity of pearl oysters between suspended and bed culture methods in Japan. A pearl culture farm located in Broome in Australia was to try a deep water technique where pearl oysters would be cultured on equipment anchored to the sea bed (Anon., 1977). If a manageable and economically viable technique could be developed in India for culturing the pearl oysters on the natural beds themselves in the Gulf of Mannar, it would substantially enhance the quality of oysters and the quality of the cultured pearls.

Transplantation of pearl oysters to the island ecosystems of the Andaman and Nicobar Islands and Lakshadweep will have considerable importance for commercial pearl culture operations. These provide conditions resembling closely those of the Japanese and Australian farms. *P. fucata* could be transported from the mainland to these islands. Similarly transplantation of *P. maxima* from the eastern bounds of the Andaman Sea to the Andaman and Nicobar Islands could be taken up. If these become successful, the islands could form one of the important pearl culture centres of the world for pearls from the three major species *P. fucata*, *P. maxima* and *P. margaritifera*.

Improvement in pearl production

Pearl production depends on three aspects, namely the physiological condition of the oyster, the techniques of surgery and the environment. No work has been done on the physiology of the Indian pearl oyster so far. Basically, the biochemical composition of the pearl oyster should be investigated in detail. The physiology of nutrition and reproduction should be studied. This would lead to an understanding of the physiological changes that take place due to insertion of nucleus and manipulating such changes to the advantage of production of quality pearls. The histochemistry of mantle tissue and pearl-sac formation also need detailed studies. Calcium metabolism of the pearl oysters and physiological regulation of secretion and deposition of the different layers of shells should be understood. Relation between the maturity stages of the gonad and pearl-sac formation and also quality and shape of the cultured pearls in relation to site of nucleus implantation should be studied. Nutritional requirements in tank culture of oysters should be evaluated for controlled production of pearls. The Japanese workers have recently taken up studies on tissue culture of the outer epithelial cells of the mantle to be able to produce aragonite pearls of good quality (Wada, 1973; Machii, 1974). Work in this direction will be very important for the future of the pearl culture industry.

Improvements in surgical techniques would also be necessary. Satisfactory pearl production rates have already been obtained in single and multiple implantations. There is still scope for a marginal increase of these rates. A careful analysis is needed to reduce the slipping rate of nuclei from the implanted oysters. Pearls with protuberances, dimples and dirt appear in the collection and these may be due to faulty techniques. Graft tissue plays a critical role in pearl production and a detailed study is needed to understand the changes that take place in the mantle tissue from the time it is excised from the oyster till the pieces are inserted. Detailed studies on the convalescence of seeded oysters and post-operative care would also be required.

Development of pearl culture based on *P. margaritifera* in the Andaman waters would need a strategy different from that for *P. fucata*. The blacklip pearl oyster is capable of producing half-round pearls and rarely round ones. Appropriate techniques of pearl production in this species must be developed.

An aspect of immediate concern to pearl culture in India is the production of shell-beads for use as nuclei. The pig-toe (*Tritogonia*), three-ridge (*Pleurobema*) and washboard (*Megalonais*) shells of the Tennessee River of the United States of America are used in production of nuclei in Japan. It would appear that no fresh water mussels similar to the above are available in the Indian rivers. The shells of *Lamellidens* are too thin to be used for the purpose. Similarly the shells of *Parreysia*, though stouter than those of *Lamellidens*, are not thick enough to render them suitable for producing beads of more than 3 or 4 mm diameter. The black clam *Villorita* has a stout shell but is too small to be economic and it may not give larger than 3 or 4 mm diameter beads. Some experimental work has been done with the marine shells of the conch (*Xancus pyrum*), the turban shell (*Turbo niloticus*) and the giant clam (*Tridacna* spp.), the last two from the Andaman waters. It was found that the conch shell would be a better material than the other two in terms of cost and also specific gravity. The conch shells form the raw material for the shell-bangle industry in India and the wastes alone could be used in the production of shell beads for pearl culture (Velu *et al.*, 1973). More detailed evaluation is required in material selection. The techniques of producing shell-beads developed as a preliminary step by Velu *et al.* (1973) need improvements for large-scale production and refinements for achieving dimensional accuracy and finish.

Some basic studies on the hydrological conditions of the pearl culture farm at Veppalodai have been made. There is need for a more critical investigation on the factors that influence the biology and physiology of the pearl oysters and production of pearls. Matsui (1960) has discussed the aspects of the environment of pearl culture grounds in Japan in relation to pearls. According to him, the growth of oysters is strongly influenced by the chemical substances contained in sea water and plankton and the metal ions might affect the quality of cultured pearls. Studies on the nutritional environment, that is the quality and quantity of food available, are important.

Growth of pearls is very much related to the temperature of the environment (Wada, 1969). It has been seen that in the Gulf of Mannar the rate of deposition of nacre is very high (Alagarswami, 1978). However, faster growth is not always linked with quality of the nacre. Thinner layers of nacre are said to improve the quality of the pearls and, therefore, the Japanese farmers resort to 'make-up' culture in areas which are suitable for producing finer quality of nacre before harvest (Alagarswami, 1970). The hydrogen-ion concentration is also an important factor and a lower pH range of 7.3-7.5 appears to improve the lustre of the pearl. These factors are important in deciding the areas for the final stages of culture and seasons for pearl harvests.

Mortality of oysters has been found to be related to heavy load of fouling in some months (Alagarswami and Chellam, 1976). Oysters heavily infested by these pests produce pearls of poor quality. The environmental study of pearl culture farms should pay attention to this aspect.

Tremendous advances have been made by the Japanese scientists on studies on the composition, structure, colour and growth of cultured pearls (Sawada, 1954; Wada, 1976; Wada and Suga, 1977). Besides using the traditional methods of bleaching and dyeing for colour adjustment, colour changes in pearls

have been effected by radiation of gamma rays and neutron bombardment (Sawada, 1959; Shirai, 1970). It is necessary that basic studies are taken up on some of the above aspects in cultured pearls produced in the Indian waters.

Immediate Prospects for Commercial Production

Although pearl culture technology was developed in India in 1973 and attempts have been made since then to propagate the potential for commercial production of cultured pearls, an industry is to yet a make a start. The only programme which has a commercial touch is the pilot project at Vizhinjam. Several entrepreneurs are aware of the potential and have obtained detailed information from the Central Marine Fisheries Research Institute. As pearl culture is a new venture for the country there is some hesitation in the minds of the people to take up this challenge. Perhaps the government sector should give a lead and encourage development of an industry.

The present state of technology is considered adequate to support a moderate pearl culture industry. The *P. fucata* resource position at this time appears satisfactory in some of the natural pearl oyster beds of the Gulf of Mannar. Rate of production and quality of pearls are close to what is obtained in Japan. The necessary infrastructure for training the manpower for the industry already exists in the country. The shell-beads could be imported from Japan for the present until indigenous production is achieved. The industry could be located in the Gulf of Mannar in view of the proximity of the natural resources and advantages of suitable sites. India has a well-established trade for pearls which depends entirely on imports for use within the country and for re-exports. Advantages of pearl culture over pearl fishing have been well established in many countries. Wada (1973) pointed out that the major factor responsible for the remarkable growth of production is that pearls give the highest profit return of all marine products cultivated in the coastal waters of Japan. India which is poised for taking up major programmes in mariculture should take a step forward in the direction of establishing a pearl culture industry without any further loss of time.

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PEARL OYSTER CULTURE IN SUDANESE RED SEA

Y. B. ABU GIDEIRI
Department of Zoology, University of Khartoum, Khartoum, Sudan

ABSTRACT

The techniques of culture of the Mother-of-Pearl Oyster *Pinctada margaritifera* var. *erythraensis* (Jameson) followed in Sudan are briefly described. Natural stock of the pearl oyster occurring in shallow waters within the 7.5 m depth zone are found to be healthier and more in number than those in the deeper waters. Spat are obtained by the collecting rafts set in the sea in July soon after the spawning of the oyster. After 10 or 12 weeks when the spat settle densely on the collectors, they are transferred to nursery trays erected in the sea. Young oysters are grown for one year in the trays, when the shells measure 5 to 7 cm in diameter. At this stage, they are transferred to growing traps. Oysters reach marketable size in about 3 years. Harvesting of the oysters is generally carried out in April-May. Average annual production of oysters realised by this culture operations has been about 330 tonnes. However, in 1969 and later in 1973 and 1975, the oysters in the farm sustained heavy mortality. Concerted efforts are being made to revive the oyster stock in the farm.

INTRODUCTION

IN SUDAN, the fishery for the mother-of-pearl oyster *Pinctada margaritifera* var. *erythraensis* (Jameson) and trochus shell *Trochus dentatus* (Forsskal) is far more important than that for fresh fish and the future prospects of the former seem much higher. For this reason, shell fisheries has received a disproportionate amount of attention during the past years. Ancient heaps of fossilised pearl shells on ' Um el Sheikh Is. ' in Dongonab Bay were recently investigated and found to be approximately 1500 years old. One heap was estimated to contain 3000 tons of shell. This proves that pearl oysters have been fished since very early times, solely for value of the few pearls they may contain. Attention of the outside world was first drawn to Dongonab Bay by the writings of the famous Swiss Explorer Burkhardt who called there in 1814. The first study of pearl fisheries of the Sudanese Red Sea was started in 1040. During the period 1905 to 1922 methods for the cultivation of the pearl oysters solely for the value of its nacre were attempted.

Dongonab Bay is different from any other part of the Sudanese Coast and has far greater potential than other areas. It is the only place in Sudan where pearl oysters may be cultivated. The bay is a large, shallow, protected inlet of almost 100 square miles area with practically no tides and very gentle currents. Because it is relatively shallow and has no great interchange of water with the main body of the Red Sea, the water cools and warms more readily and thus the annual variation in sea temperature in the bay is greater than in other parts of the Red Sea. This is a very important factor, as the spawning of pearl oysters is influenced by temperature.

The stocks of pearl oysters in deeper water are not as rich as reputed to be. The healthiest pearl oysters are found in less than 5 fathoms

depth and very few are found beyond a depth of 15 fathoms, because the natural substrata in the deeper water is not suitable for their settlement and growth. Shells from deeper than 7 fathoms are usually infested with such parasites as mud worms (*Polydora*) and boring sponges (*Clione* spp.) and more than 50% of these are unsuitable for market. By not allowing the introduction of underwaters breathing apparatus in the fishery of natural stocks, the pearl oysters in these areas beyond the reach of skin divers, will be left as a breeding reserve and there will be no danger of overfishing, even if all the oysters are removed from the shallow banks each year.

Trochus shells are not found in depths of more than 9 fathoms and are most common in depths from 2 to 5 fathoms.

Mother-of-pearl oyster culture

After devising methods of cultivating pearl oysters, Crossland (1905-1922) put his findings into practice on a very large scale and during his latter years he was producing a regular annual crop of more than 300 tons of first quality Mother-of-Pearl shells.

After the original survey of the natural stocks in 1958 it was realized that the solution to increasing production lay, not in the more skillful exploitation of the natural stocks which may eventually result in overfishing and depopulation, but in cultivation. The techniques of pearl oyster cultivation which have since been adopted are described here.

Spat collection

The crucial part of pearl oyster cultivation is the collection of 'Spat'. Pearl oysters produce millions of eggs, from which, under natural conditions, only an infinitesimal percentage survives. Basically, the idea of spat collection is to place specially designed collecting apparatus, in strategic places in the sea which affords favourable conditions for the settlement of the planktonic larvae, during the spawning season.

Spawning seems to be influenced mainly by sea temperature. When compared to most other tropical areas where pearl oysters occur, Dongonab Bay has a relatively high annual variation in sea temperature. Here the temperature varies from 34°C during August to 21°C during December and January, whereas in most tropical areas the variation ranges between 26°C and 31°C. This appears to be the reason for the brief and well defined two or three month spawning season at Dongonab, while in some other areas, this species spawns intermittently throughout the year. At Dongonab the spawning season for *P. margaritifera* fortunately does not coincide with the spawning of any other common marine organism.

Under natural conditions, after a few weeks of drifting planktonic life the young larvae are forced, by the weight of their rapidly forming shells, to settle on the sea bed. In most cases the substrata of sand, mud or live coral are unsuitable for settlement and growth.

The collecting rafts are set in the sea after spawning begins, *i.e.* in early July, soon after the regular, fierce north-east wind which blows at almost gale force for about 5 days, usually in late June. They are suspended about two feet beneath the surface, using plastic floats and are anchored by chain to a mooring block. The lowest part of the collector is set at least 3 fathoms above the sea bed.

About 50 days after the spat collectors are laid, spat settlement is seen. Before this the collectors are sometimes fouled with a dense growth of hydroids, but these usually disappear after a month or so. Within 10 to 12 weeks the settlement of spat is so dense that if left longer under such crowded conditions, the spat would become stunted because of lack of food and oxygen. By this time, most of the

spat are from 1 to 5 cm diameter and are ready for transferring to the next stage of cultivation.

Nursery stage

After experiments and experience it was found to be preferable to keep the young oysters for one year on nursery trays, until the shells reach a size when they are no longer vulnerable to attacks from predatory fish, crabs and molluscs.

The nursery is constructed *in situ* by driving two rows of teak poles in the sea bed and attaching to them, one above the other about 40 cm apart, five rows of 6 × 6 inch mesh, welded galvanized wire of No. 4 gauge. The bottom shell is covered by a layer of $\frac{3}{4}$ inch chicken mesh wire of 19 gauge. The large mesh of the upper layers allows a better flow of water and hence more food for the oysters, while the small mesh of the lower layer collects any small oysters which fall off the bamboos during planting and in the subsequent year.

During the first year the young oysters are very vulnerable to attacks from fish, molluscs and crabs. The side and top of the nursery must therefore be very well covered with 2 inch mesh, 19 or 20 gauge chicken mesh. The nursery is sometimes infested with a dense growth of stinging anemones. It is difficult to calculate the mortality during the nursery stage but if the spat are carefully handled it should be less than 10 per cent.

Growing stage

By June, July and August of the following year the shells measure from 5 to 7 cm diameter and are sufficiently strong to withstand attacks from predators.

The growing traps are constructed by supporting sheets of rigid No. 4 gauge, 6 × 6 inch mesh wire weld, 40 cm above the seabed on iron posts. This base is covered with a layer of 1 inch mesh 16 gauge, galvanized wire netting.

To ensure rapid growth and even size it is essential to have even distribution of the oysters on the growing trays. The optimum spacing has proved to be approximately 50 per square yard. The oysters need almost three years on growing trays to reach optimum marketable size, which is about 200 grams per pair of valves, dry chipped weight.

Harvesting

The oysters are ready for harvesting when they are between 3 and 4 years of age. To spread the work out evenly throughout the year, they are harvested in April and May. During mid-summer the shells are inclined to crack around the edges. Skin divers go down and 10 men can easily harvest one ton per day. Shells below the optimum marketable size are replanted on growing trays and allowed to grow for one more year.

In 1969 and later in 1973 and 1978 the oysters in the farm experienced massive mortality. Since then local and foreign efforts have been made to bring about a recovery of the farm. The assistance of the International Development Research Centre (IDRC) in this respect is highly appreciated. It is expected to determine the causative factors for the occasional mortality and evolve methods to safeguard the stock. It is hoped that the farm will shortly resume its activity as a commercial venture.

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ON THE GROWTH OF THE PEARL OYSTER *PINCTADA FUCATA* (GOULD) UNDER FARM CONDITIONS AT TUTICORIN, GULF OF MANNAR

Y. JEYABASKARAN, DANIEL SUDHANDRA DEV, I. NALLUCHINNAPPAN AND N. RADHAKRISHNAN

Pearl Culture Scheme, Tamil Nadu Fisheries Department, Tuticorin-628 001

ABSTRACT

The growth of *Pinctada fucata* under farm conditions has been studied. The rate of growth is high during the first three months. The growth rate shows an inverse relationship to temperature, salinity and quantity of foulers settled on the oysters.

INTRODUCTION

THE FIRST attempt to culture pearl oysters in captivity was made at Tuticorin as early as in 1864 by Phipps as is evident from Markham's (1866) letter written to the British Government in India. A pearl oyster nursery at Tuticorin near Hare Island was established in November, 1864.

An attempt to rear pearl oyster spat in the farm at Krusadai Island was carried out by Devanesan and Chidambaram (1956). The results of the studies on the growth of pearl oyster spat in cages in the farm at Tuticorin during 1978-79 are presented in this paper.

MATERIAL AND METHODS

A specially designed cage called ' pigeon-hole cage ' made of iron frames and nylon rope was used. Iron rod of 6 mm size and nylon rope of 2 mm thickness were used for fabrication of the cage. The size of the cage is 60 × 45 × 9 cm which is compartmentalised into 20 cubicles of 12 × 11.2 × 9 cm size. The cage was suspended at a depth of 3 to 4 m from a floating raft. Twenty spat collected from a pearl bank in the Gulf of Mannar on 24th January 1978 were put one each in the cubicles of the cage and observations on growth were made for one year. The water temperature and salinity were recorded. Data on the average weight of foulers barnacles and bryozoans per oyster was also noted.

RESULTS

Oysters ranging from 21.0 to 29.0 mm (DVM) were put in the cubicles, the average size being 21.6 mm DVM in January 1978. The oysters had grown to the size range of 52.8 to 62.1 mm DVM, the average size being 58.6 mm DVM, by January 1979 at the end of one year (Table 1). The minimum and

TABLE 1. *Monthly average size of pearl oysters reared in ' Pigeon-Hole ' cages at Tuticorin*

Months	DVM (mm)	HL (mm)	T (mm)
January '78	21.6	27.9	8.3
February	32.3	33.4	9.7
March	36.5	36.0	11.4
April	39.3	37.4	12.9
May	40.4	38.3	14.1
June	42.8	39.3	14.6
July	44.8	40.8	16.4
August	47.8	43.3	17.3
September	49.5	44.4	17.8
October	51.9	45.6	19.4
November	53.2	46.5	20.4
December	56.3	48.0	20.9
January '79	58.6	50.1	21.4

DVM—Dorso-ventral measurement
HL—Hinge length
T—Thickness

maximum bottom temperatures were 25.5°C (January) and 31.3°C (April) respectively. The salinity ranged between 29.06‰ (December) and 33.85‰ October) as given in Fig. 1. The weight of the foulers per oyster varied between 3.5 g in January and 32.0 g in May (Fig. 1).

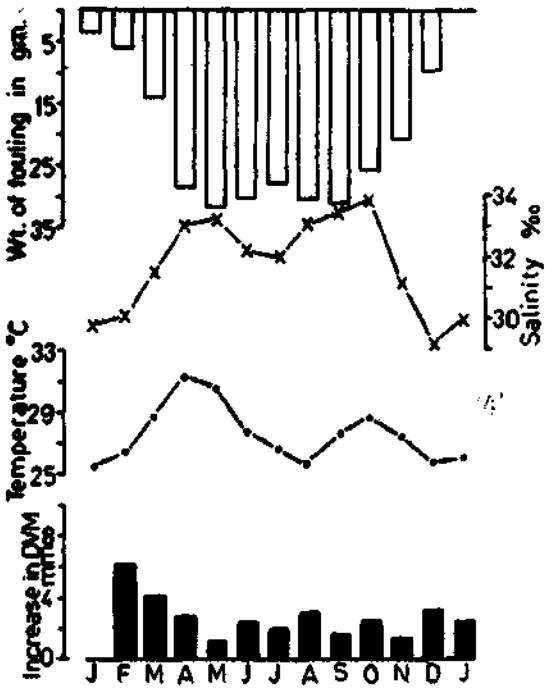

Fig. 1. Growth of pearl oyster in relation to temperature, salinity and weight of fouling.

Discussion

The increase in average size of pearl oyster was 37 mm at the end of one year. It is evident that the growth of *Pinctada fucata* is very fast during the first three months under farm conditions when the oysters are small in size (Fig. 1). Such a fast growth of pearl oyster under farm condition was also reported by Devanesan and Chidambaram (1956) at Krusadai Island, Gulf of Mannar. Their observations showed more rapid growth of the oysters than that recorded during the present study.

The rate of growth of oysters was high during winter months when the temperature was low; another period of fast growth rate was seen during August when the temperature was low (Fig. 1). Similar findings have been reported by Gokhale *et al.* (1954) and Pandya (1976) in the Gulf of Kutch. The growth rate showed inverse relationship with salinity values. The salinity was less during January-March and November-December (Fig. 1). The growth of the oysters was high during these periods.

The weight of foulers per oyster was very high during summer months and very low during winter months (Fig. 1). The growth rate of pearl oyster exhibits an inverse relationship with the quantity of foulers in farm conditions. The salinity values during Southwest monsoon period (April-October) are high in the Gulf of Mannar area (Prasad, 1958; Chandrasekaran and Sudhakar, 1967) due to the influence of the drift current flowing northward. The higher salinity was favourable for the growth of foulers on the oysters and during the same period the growth was less. Similar relationship of salinity and settlement of foulers was observed by Ganapati *et al.* (1958), Antony Raja (1959) and Jeyabaskaran *et al.* (in Press).

The observations reveal that the young oysters of size 20-30 mm DVM can be raised to the required size for nucleus implantation work within 4 months period at Tuticorin under farm conditions.

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ON CONTROLLED SPAWNING OF INDIAN PEARL OYSTER *PINCTADA FUCATA* (GOULD)

K. ALAGARSWAMI, S. DHARMARAJ*,
T. S. VELAYUDHAN*, A. CHELLAM* AND A. C. C. VICTOR*

Central Marine Fisheries Research Institute, Cochin-682 018

ABSTRACT

Induced spawning of pearl oysters serves a dual purpose in pearl culture. Besides providing the gametes for the hatchery production of seed, it conditions the oyster for nucleus insertion for the production of cultured pearls. Experiments were conducted on induction of spawning in *Pinctada fucata* using hydrogen peroxide, Tris buffer, sodium hydroxide and combinations of hydrogen peroxide+Tris/NaOH. In other experiments ammonium hydroxide (N/10) was injected into the adductor muscle or foot of the oyster. Thermal stimulation was also attempted.

Spawning response to H_2O_2 treatment was not quite satisfactory. Concentrations of 3-6 mM peroxide was found to evoke some response. Hydrogen peroxide in alkaline medium using Tris gave slightly better results. Tris-buffered sea water with a pH of 9.0 by itself was found to induce 78.6% of the pearl oysters to spawn. Similarly, the alkaline sea water medium with NaOH stimulated spawning in 68.4% of the oysters at pH 9.5. Injections of 0.2 ml of N/10 NH_4OH resulted in the spawning of 48.1% of the treated oysters. Thermal stimulation by raising the sea water temperature from 28.5°C to 35.0°C, gave good results on the occasion when 87.5% of the oysters spawned. But in several other experiments the response was either nil or poor. The present study has indicated that an alkaline sea water medium (pH 9.0-9.5) would be useful for the controlled spawning of the Indian pearl oyster.

INTRODUCTION

MANAGEMENT of reproduction is one of the facets of aquaculture and it serves the primary need of providing seed for stocking the farms. Great strides have been made in controlled spawning of molluscs in shellfish hatcheries. Loosanoff and Davis (1963), Loosanoff (1971) and Ino (1972) have reviewed the methods employed for out-of-season spawning of molluscs. The commonest technique used for inducing spawning is conditioning the molluscs for accelerated development of gonad through thermal stimulation and spawning them by a quick rise in temperature to the optimum level and adding egg or sperm suspension (Loosanoff and Davis, 1963). This method has been particularly successful for species in the subtropical and temperate regions.

The Japanese workers have mostly relied on chemical stimulation for spawning the molluscs. The methods include spawning the animals in ammoniated sea water (Wada, 1942 ; Sagara, 1958 a) and injection of neutral potassium salts (Iwata, 1948 a, b) or ammonium hydroxide (Sagara, 1958 b). Stripping the gonad and treating the eggs with a weak solution of ammonium hydroxide has also given good results in many cases (Loosanoff and Davis, 1963).

* Present address : Tuticorin Research centre of CMFRI, 93 North Beach Road, Tuticorin-1.

Methods such as giving a mild electric shock (Iwata, 1950) and pricking or severing the adductor muscle (Loosanoff and Davis, 1963) have proved useful for spawning the mussels. A more recent technique which has been successful in the case of a variety of molluscs is the addition of hydrogen peroxide to alkaline sea water (Morse *et al.*, 1976, 1977, 1978).

Induced spawning of pearl oyster serves a dual purpose in pearl culture. Besides providing the gametes for the hatchery production of seed, it conditions the oysters for nucleus insertion for the production of cultured pearls. The best site in the body of the pearl insertion of nucleus is the gonad and when a gonad contains eggs or sperms it is difficult to obtain good results (Mizumoto, 1979). Therefore, the pearl culturists have invented the method of forced ovulation called 'egg extraction' in order to produce large sized good quality pearls (Alagarswami, 1970, Wada, 1973). Wada (1947), Kobayashi and Yuki (1952) and Yamaguchi (1957) have worked on artificial breeding of *Pinctada martensii*, Wada (1942) on *P. maxima* and Setoguchi (1959) on *P. margaritifera*. In view of this importance, experiments were carried out on controlled spawning of the Indian pearl oyster *Pinctada fucata*, using some of the established techniques during August-September 1979 and the results are presented in the paper.

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Material and Methods

Pearl oysters brought from the pearl culture farm were examined for the maturity condition of the gonad. Oysters with fully ripe gonads were selected for the experiments on induction of spawning. They were kept in glass vessels containing 6 or 3 litres of the experimental solutions. Aeration was given only after the oysters were transferred from the experimental solutions to fresh sea water. All the experiments were conducted under ambient temperature conditions.

Experiments were conducted on induction of spawning using hydrogen peroxide, Tris-buffer, sodium hydroxide and combinations of hydrogen peroxide+Tris-buffer/sodium hydroxide. Ammonium hydroxide was injected into the adductor musle and foot of the oysters. The experimental procedures are given under the respective sections.

Results

Spontaneous spawning of pearl oysters

Pinctada fucata has been observed to spawn spontaneously in the laboratory. Usually it happened with a change of sea water in the vessels containing oysters for some time. In some instances, when oysters brought from the farm were cleaned and placed in troughs and sea water which had been kept standing in carboys was added, spawning occurred. On a few occasions, during November-December, pearl oysters collected from the natural beds (depth 15m), when brought up and immediately transferred to a vessel containing sea water drawn from the surface of the sea, spawned vigorously. In all such cases of spontaneous spawning, change of fresh sea water drawn from a source which is different from the one the oysters were living prior to the change, has brought about spawning. Although the factors responsible for inducing the oysters to spawn have not yet been identified, it appears that differences in density and temperature, or physiological stress might trigger the spawning act.

During the period 1973-79, spontaneous spawning was observed on 23 occasions: January-3, February-1, June-1, July-3, August-4, September-2, October-1, November-3 and December-5. Even with these limited data,

it is found that the frequency of spawning is more during November-January, which is about the Northeast monsoon period, and July-August, the period of southwest monsoon.

Experiments on induced spawning

Effect of hydrogen peroxide : Morse *et al.* (1976, 1977, 1978) have originally developed the technique of inducing spawning in abalone, mussel, scallop and the mangrove oyster by the addition of hydrogen peroxide to sea water, which is normal or alkaline. The technique was applied on the pearl oyster.

From 30% (weight) stock solution of hydrogen peroxide (guaranteed reagent grade ; stored at 0-4°C), a 6% solution was prepared. The experiments were carried out at concentrations of 1.532, 3.064 and 6.128 milliomolars (mM) prepared by adding 6.25 ml, 12.5 ml and 25.0 ml of 6% H_2O_2 solution respectively to 6 litres of fresh sea water. Selected oysters were kept in the laboratory for accilimatisation for at least 12 hours before using them in the experiments. Since it was difficult to distinguish males and females based on external appearance of the gonad, no attempt was made to sex them prior to experiments.

Sea water of identical quality was used both for acclimatisation and for the experiments so that the probability of factors such as temperature, salinity and pH influencing the animals was ruled out. The oysters were kept in the experimental medium for pre-determined durations at the end of which the solution was siphoned out and fresh sea water was added without disturbing the animals. The above procedure was common for all the experiments described in the paper except for the one on injection of NH_4OH.

Five experiments were conducted on different dates involving a total number of 111 oysters in the H_2O_2 medium and 37 oysters in the controls. The pooled results are given in Table 1.

TABLE 1. *H_2O_2 induction of spawning in the pearl oyster* Pinctada fucata

Treatment	No. of oysters		
	Tested	Spawned	
		Male	Female
H_2O, pH 8.1-8.2 (control)	37	0	0
H_2O_2, 1.532 mM ..	37	0	0
H_2O_2, 3.064 mM ..	37	3	1
H_2O_2, 6.128 mM ..	37	2	2

The temperature of sea water ranged 26.5°-29.0°C for all the five experiments and the difference within each experiment varied from 0.4° to 1.1°C. The immersion time in H_2O_2 was 2½ hr in two experiments 4 hr in one and 8 hr in two. Spawning took place from 2 hr 40 min to 3 hr 03 min after immersion in H_2O_2 in the treatments of 2½ hr and from 5 hr 52 min to 6 hr 05 min in the treatment of 5 hr duration. Spawning was observed always after the change to fresh sea water.

Effect of Tris buffer

Tris (Hydroxymethyl)-Aminomethane was used to increase the pH of sea water. The buffer was slowly added to fresh sea water reading the pH. Sea water with pH values of 8.5, 9.0, 9.5 and 10.0 was used as experimental solution. Normal sea water with a pH of 8.1-8.2 was used for control. A total of 94 oysters were used in Tris solution and 27 as controls in three sets of experiments. The pooled data are given in Table 2.

TABLE 2. *Tris induction of spawning in pearl oyster* Pinctada fucata

Treatment	No. of oysters		
	Tested	Spawned	
		Male	Female
H_2O, pH 8.10-8.25 (control)	27	0	0
Tris, pH 8.5 ..	28	0	0
Tris, pH 9.0 ..	28	4	18
Tris, pH 9.5 ..	28	9	2
Tris, pH 10.0 ..	10	2	0

The duration of immersion in Tris solution was 3 hr in one experiment and 4 hr in the other two. The temperature for all the experiments ranged 26.9°-28.9°C, but within each experiment the difference was between 0.4°-1.2°C. There was no spawning in the controls. Experiment with pH 10 was conducted only once and 2 males spawned very mildly. Satisfactory results were obtained in pH 9.0 and 9.5. In most cases (22 oysters out of 35 spawned) profuse spawning took place in the Tris medium itself and in some animals, at pH 9, spawning occurred after changing the medium to fresh sea water. The spawning response was observed within one hour after immersion in 15 oysters, between 1-2 hr in 5 oysters and after 2 hr 10 min in the remaining 2 oysters. Thirteen oysters spawned after changing to fresh sea water and the response came from 3 hr 15 min to 4 hr 44 min after the time of the immersion in Tris.

Effect of alkali (NaOH)

By dissolving pure pellets of sodium hydroxide in sea water, solutions having a pH of 8.5, 9.0, 9.5 and 10.0 were prepared. The solution turned rather opaque at the two higher concentrations. Controls, using normal sea water, had a pH of 8.0 and 8.1. A total of 67 oysters were used in the alkaline medium and 20 as controls in two experiments. The combined data of the two experiments are given in Table 3.

TABLE 3. *NaOH induction of spawning in pearl oyster* Pinctada fucata

Treatment	No. of oysters		
	Tested	Spawned	
		Male	Female
H_2O, pH 8.0-8.1 (Control)	20	0	0
NaOH, pH 8.5 ..	19	0	0
NaOH, pH 9.0 ..	19	0	0
NaOH, pH 9.5 ..	19	6	7
NaOH, pH 10.0 ..	10	0	0

The immersion time was 3 hr in one experiment and 4 hr in the other. The temperature range was from 26.9° to 27.8°C. In both the experiments spawning occurred only at pH 9.5. Of the 13 oysters, 9 spawned in the NaOH medium itself, 8 of them between 1-2 hr and one 2 hr 58 min after immersion. The 4 oysters which spawned in fresh sea water after the change responded from 3 hr 27 min to 5 hr 03 min after introduction to the alkaline medium.

Effect of H_2O_2 in alkaline medium (H_2O_2 + Tris)

Morse *et al.* (1978) reported that addition of Tris buffer, though not essential for induction of spawning, acts to increase the proportion of animals that will spawn in response to a given concentration of peroxide. They found that Tris at pH 9.1 was effective in the case of abalones. Two experiments were conducted on the pearl oyster to ascertain the spawning response in H_2O_2 buffered by Tris to pH 9.1. The experiments were carried out concurrent to those on H_2O_2 induction. Animals in Tris solution at pH 9.1 formed controls. The results are presented in Table 4.

TABLE 4. *Induction of spawning in pearl oyster* Pinctada fucata *by H_2O_2 in alkaline medium of Tris*

Treatment	No. of oysters		
	Tested	Spawned	
		Male	Female
Tris, pH 9.1 (control)	16	0	1
Tris, pH 9.1 + H_2O_2, 1.532 mM	16	0	0
Tris, pH 9.1 + H_2O_2, 3.064 mM	16	3	7
Tris, pH 9.1 + H_2O_2, 6.128 mM	16	0	0

Spawning occurred only at 3.064 mM concentration of peroxide in both the experiments, after the oysters were changed to fresh sea water at the end of 4 hr of treatment. Spawning response came actually 6½ to 8 hr after the

immersion in the experimental medium. One of the oysters in the control spawned mildly.

Effect of H_2O_2 in alkaline medium ($H_2O_2 + NaOH$)

Morse *et al.* (1978) found that use of sodium hydroxide to adjust the sea water to pH 9.1 was apparently as effective as Tris in facilitating induction of spawning in the abalones by hydrogen peroxide. This was tested on the pearl oyster in three sets of experiments carried out concurrently with those on H_2O_2 induction. The pH was adjusted to 9.0 by the addition of NaOH pellets. The results are presented in Table 5.

TABLE 5. *Induction of spawning in pearl oyster* Pinctada fucata *by H_2O_2 in alkaline medium of NaOH*

Treatment	No. of oysters		
	Tested	Spawned	
		Male	Female
NaOH, pH 9.0 (control) ..	21	5	5
NaOH, pH 9.0+H_2O_2, 1.532 mM	21	0	0
NaOH, pH 9.0+H_2O_2, 3.064 mM	21	0	0
NaOH, pH 9.0+H_2O_2, 6.128 mM	21	2	0

The duration of immersion was 2 hr 30 min in two experiments and 5 hr in the third. At 6.128 mM concentration mild spawning occurred in two males on changing to fresh sea water, 2 hr 35 min after immersion in the solution. The oysters did not spawn in other concentrations. On the other hand, spawning was observed in the controls in all the three experiments.

Effect of injection of ammonium hydroxide

A dilute solution of 0.1 normal NH_4OH was prepared from a stock solution and a total of 47 pearl oysters were treated with injections of 0.1, 0.2 or 0.3 ml of the dilute solution. Controls were kept without injection. Ten oysters each which were given 0.1 ml and 0.3 ml of ammoniated sea water did not show any spawning response. Among 27 oysters which were injected with 0.2 ml solution, 13 spawned profusely. In one experiment, when the injection was given at the base at the foot, all the seven oysters spawned profusely. In another, injection administered in the adductor muscle stimulated all the five oysters to spawn. Oysters of the control did not spawn on either occasion.

DISCUSSION

Morse *et al.* (1978) indicated that spawning in molluscs may result from a peroxide-induced stimulation of the endogenous enzymatic synthesis of potent hormone-like prostaglandin molecules. They found that the alkaline medium, though not essential for the induction of spawning, increases the proportion of animals that will spawn in response to a given concentration of hydrogen peroxide. Whereas they succeeded in spawning molluscs with hydrogen peroxide, they did not with several other oxidising agents (Morse *et al.*, 1976). The results on H_2O_2 induction presented in Table 1 indicate poor response of *Pinctada fucata* to this treatment. While Morse *et al.* (1976) have obtained spawning in 30 abalones out of 31 and in all the 6 groups of mussels in the concentration of 5 mM of H_2O_2, in *P. fucata* spawning was observed only in 4 specimens each, out of 37, in the concentrations of 3.064 mM and 6.128 mM. Spawning took place, as observed by Morse *et al.*, after replacing the medium with fresh sea water and within 3 hr ± 30 min after the first addition of H_2O_2 in the experiments where the oysters were treated for 2½ hr.

Testing in the alkaline medium (pH 9.1) of H_2O_2, using Tris (Hydroxymethyl)—Aminomethane, the result was better at 3.064 mM

concentration with 10 out of 16 oysters responding (Table 4). In other two concentrations (1.532 mM, 6.128 mM) there was no spawning. Spawning response in the alkaline medium (pH 9.0) of H_2O_2, using NaOH, was not satisfactory as only 2 oysters out of 21 spawned in 6.128 mM solutions, with no induction in the other two concentrations (Table 5). On the other hand, 10 oysters from among 21 spawned in the controls. Although Morse *et al.* (1978) have not given the percentage of animals spawning in the alkaline medium (using Tris as well as NaOH), it has been stated that alkalinity promotes both the peroxide activation and the induction of spawning.

Morse *et al.* (1978) found 2-4 mM peroxide optimal for spawning abalones, mussels, scallop and the mangrove oyster. The present study would show that concentrations between 3-6 mM can induce spawning in *Pinctada fucata*. Although spawning induction has been observed the percentage of response is relatively low. Perhaps some more experiments could decide the usefulness of this technique for large-scale spawning of pearl oysters. Peroxide induction has been found to be better in the alkaline medium of Tris. However, Tris by itself has been found to give superior results.

Alkaline sea water has been widely used by the Japanese workers for inducing spawning and artificial fertilisation in pearl oysters. Kobayashi (1948) found a pH of 8.6 optimum for the artificial fertilisation of the eggs of the Japanese pearl oyster *Pinctada martensii* (= *P. fucata*) and he was unable to obtain fertilisation under lower pH conditions. Kobayashi and Yuki (1960) found that the rate of fertilisation reached almost 100 per cent by increasing the pH from 8.3 to 8.6 by adding ammoniated sea water. Wada (1942) induced spawning in *P. maxima* by half per cent 0.1 normal NH_4OH sea water. Wada (1947) obtained artificial fertilisation of *P. martensii* by application of 1/10,000 to 1/1000 normal ammoniated sea water. Setoguchi (1959) got good results in *P. margaritifera* using 1.2-1.5% 0.1 normal NH_4OH sea water. Setoguchi (1957, 1958) reported that a pH of 8.7-8.9 was effective in inducing spawning of *Pteria macroptera* which is another species used in pearl culture. In the present study, alkaline sea water was prepared with Tris-buffer and sodium hydroxide and the spawning trials were carried out in pH 8.5, 9.0, 9.5 and 10.0. With Tris, 78.6% of the oysters spawned at pH 9.0, 39.3% at pH 9.5 and 20.0% at pH 10.0 and none in controls with pH 8.10-8.25 (Table 2). With NaOH, 68.4% of the oysters spawned at pH 9.5 (Table 3) and 47.6% at pH 9.0 (Table 5). Unlike with H_2O_2, profuse spawning takes place in the alkaline sea water itself in the case of Tris as well as NaOH. The spawning response in Tris is relatively higher than in NaOH. In both cases, pH 9.0-9.5 appears optimal for induced spawning. While pH 8.5 evokes no response, pH 10.0 with Tris has triggered spawning in 20% of the oysters. The results obtained in the present experiments would show that higher alkaline condition (pH 9.0-9.5) is even a better medium than hydrogen peroxide for inducing the pearl oysters to spawn.

Sagara (1958 b) has induced spawning in the clam *Meretrix* by injection of NH_4OH 1/20 N in the gonad. Iwata (1948 a, b) has succeeded in spawning several species of clams by injection of 2 ml of neutral potassium salt solutions into the visceral cavity. In the present study injection of 0.2 ml of N/10 NH_4OH administered in the foot or the adductor muscle has resulted in the spawning of 48.1% of the oysters treated. The spawning response of pearl oysters for NH_4OH injection has been proved. Further experiments are needed to determine the optimum concentration of the ammoniated sea water required for achieving higher spawning rates.

As in the case of edible oysters and clams, some success has been achieved in thermal

stimulation of spawning in the pearl oyster. Kuwatani *et al.* (1974) observed mature and spawned individuals in tanks with circulating sea water of higher temperature (26° and 30°C) and suggested that spawning might be induced by this method in winter. Wada (1976) induced spawning in the pearl oyster by raising the temperature from 25°C to 30°C and found that more than 700-800 degree-days would be required for the gonads to mature in the Ago Bay. During the course of the experiments reported here, six trials were carried out to spawn the pearl oyster under thermal stimulation. While the spawning response was nil in three and poor in two experiments, good results were obtained in 14 among 16 pearl oysters by raising the temperature from 28.5° to 35.0°C in the sixth experiment. Rao *et al.* (1976) obtained spawning in the mussel *Mytilus viridis* by increasing the temperature from 26.5°-28.0°C to 32.0°-35.0°C. These two results coming from the Indian waters might suggest, although the data are admittedly inadequate for attempting any generalisation, that in the tropical species of molluscs thermal stimulation for artificial spawning may require raising the temperature to a higher level such as 35°C or about 7°C above the ambient temperature.

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EMBRYONIC AND EARLY LARVAL DEVELOPMENT OF PEARL OYSTER *PINCTADA FUCATA* (GOULD)

K. ALAGARSWAMI, S. DHARMARAJ*, T. S. VELAYUDHAN*, A. CHELLAM AND A. C. C. VICTOR*
Central Marine Fisheries Research Institute, Cochin-628 018

ABSTRACT

Hatchery production of seed is of great importance in aquaculture, particularly in situations where availability of natural seed is undependable. Besides, the system provides advantages of selective breeding keeping in view the genetic factors. Commercial hatcheries already sustain large-scale production of edible oysters in several countries.

Since the natural production of pearl oysters in the pearl banks of the Gulf of Mannar is characterised by very wide fluctuations, ability to produce pearl oyster seed through hatcheries is of great practical importance for the development of pearl culture industry in India. A major research effort is, therefore, being devoted to this problem. A number of experiments on spawning and rearing of *Pinctada fucata* larvae were conducted during 1978-79. The eggs, measuring 47.5μ develop into straight-hinge veligers of 67.5 x 52.5μ size in 20 hours 40 minutes after fertilisation. The paper describes the different stages of the development of *P. fucata* from fertilisation to the straight-hinge larvae as obtained during the period of study.

INTRODUCTION

RECENT advances in the rearing of marine bivalve larvae have largely been due to the successful work carried out at three centres, namely Milford in U.S.A. (Loosanoff and Davis, 1963), Conway in U.K. (Walne, 1964) and Sendai in Japan (Imai and Sakai, 1961). Loosanoff (1971) has reviewed the development of shellfish culture techniques. These researches have led to the establishment of commercial hatcheries for the edible oysters and clams (Davis, 1969). Compared to these developments on the larval rearing of oysters and clams (Davis, 1969). Compared to these developments on the larval rearing of oysters. clams, mussels (Bayne, 1976) and abalones, pearl oysters of the genus *Pinctada* have received limited attention. Recently, hatchery production of seed of *P. fucata* (Gould) has developed in the pearl culture industry mainly in the southern part of Japan (Wada, 1975). For two other important species employed in pearl culture, namely *P. maxima* and *P. margaritifera* artificial seed production technique is yet to be established (Mizumoto, 1979). In this light, the initial success achieved in India during 1978-79 in rearing *P. fucata* larvae upto the straight-hinge stage is considered important and reported here.

The authors are grateful to Dr. E.G. Silas, Director, Central Marine Fisheries Research Institute for the encouragement given to this work.

MATERIAL AND METHODS

Pinctada fucata collected from the natural beds (depth 15-20 m) in the Gulf of Mannar or taken from the farm (depth 4-8 m) at Tuticorin were examined for sexual maturity and, for each experiment, about 15 oysters with ripe gonad were introduced in a glass trough (10 l) containing filtered sea water (Pl. I A). In

* *Present address :* Tuticorin Research Centre of CMFRI, Tuticorin 628 001

several experiments the males and females spawned spontaneously and fertilisation, embryonic and early larval-development proceeded normally.

Fifteen minutes after each successful spawning, when the medium had turned 'milky' (Pl. I A), the gamete suspension from the trough was transferred into several one-litre glass beakers containing filtered sea water. After making sure that the fertilised eggs had settled down, the water was partially siphoned out and fresh sea water was added. By repeating this process, the excess sperms and broken tissue material were removed from the beakers. All the culture vessels were immersed in a common water bath to minimise temperature variations. No aeration or mechanical agitation was employed.

The cluster forming behaviour of the embryos at the blastula stage was taken advantage of for picking the embryos and transferring into a fresh set of beakers. After the larvae reached the straight-hinge stage, it was attempted to feed them by introducing small quantities of micro-algae such as *Tetraselmis gracilis, Synechocystis salina* and *Chlorella marina.*

Larval rearing was carried out under ambient temperature conditions during December, 1978, and January, March, July and August, 1979. The temperature range in the rearing medium during the period of experiments was 27.0°-28.9°C; salinity ranged 34.40 - 35-67 ppm and pH 7.55 - 8.25.

Observations

Spawning behaviour

In natural spawning, the male was always found to be the first to spawn. Under the stimulus of the sperm suspension the females spawned later. In successful spawning experiments the time lag between the spawning of the first male and the female oyster was less than 30 minutes. The male discharges the spermal fluid in several spells of continuous streams (Pl. I B) through a temporary oval opening formed by the mantle opposite the urogenital opening. The female spawns the eggs both as discontinuous ribbon-like structures (Pl I C) when the eggs are entangled in broken gonadal tissue and as free ova in continuous stream. The entangled eggs always settle to the bottom and the free eggs float. The sperms get dispersed immediately in the water column (Pl I D). In advanced stage of the spawning act the valves of the oyster remain wide open whether there is discharge of gametes or not. The oyster becomes exhausted and do not respond to disturbance, and the valves would close only when pressed tight. It takes considerable time for the oysters to recover from the spawning fatigue.

Mature eggs

The just spawned eggs have an irregular shape and most of them are pyriform. The average dimensions of the irregular eggs are 73.9μ along the longest axis and 45.2μ in breadth (Pl. I E). A large clear germinal vesicle (nucleus), measuring an average 24.7μ in diameter, is distinctly seen. In some instances the nucleolus can also be seen in the germinal vesicle. The yolk cytoplasm is heavily granulated and is opaque. The egg is enclosed in a vitelline membrane which persists on the developing embryo till it reaches the early trochophore stage.

Soon after discharge, the eggs become activated. They assume a spherical shape and the germinal vesicle breaks down either before or after the assumption of spherical shape (Pl. I F). The spherical egg measures an average 47.5μ in diameter. The breakdown of germinal vesicle marks the resumption of meiosis.

Fertilisation and cleavage

During the process of fertilisation, the first and second polar bodies are emitted. The

vitelline membrane is lifted off from the cytoplasm to form a conical protuberance on the spherical egg. The polar bodies persist on the embryo even in the blastula stage.

The first cleavage which takes place 45 minutes after fertilisation is along the polar axis so that the polar bodies come to lie on the cleavage furrow (Pl. II A). It results in two unequal cells, a micromere and a macromere. The macromere is formed by the fusion of one of the daughter cells of the first cleavage and the polar lobe formed at the vegetal pole prior to the onset of cleavage. However, the trefoil aspect reported for *Pinctada margaritifera* (Tranter, 1958), *Mytilus* (Reverberi, 1971) and *Mytilus edulis* (Rao *et al.*, 1976) is not distinct in *Pinctada fucata*. At the second cleavage, the micromere divides normally into two daughter micromeres but the macromere divides unequally resulting in a micromere and a macromere which is again formed by the fusion of the second polar lobe with the cell. Thus the four-cell stage consists of three micromeres and a macromere which is almost as large as the macromere formed at the first cleavage (Pl. II A). At all stages of further cleavage to the eight-cell (Pl. II B), sixteen-cell (Pl. II C) and beyond (Pl. II D) a large macromere is always present. Soon the micromeres almost envelop the macromere resulting in the morula stage of embryo.

Blastula and gastrulation

The blastula is reached with the formation of the blastocoel which is a hollow space within the embryo. The cells develop minute cilia and rotation of the embryo starts. At this stage the embryo lifts itself up and swim in the water column. Gastrulation takes place by epiboly. The blastopore which was originally situated in the centre of the vegetal hemisphere of the embryo later shifts to the ventral side. The archenteron communicates with the exterior through the blastopore.

At the blastula stage the embryo exhibits phototropism. They are attracted towards concentrated artificial light and form aggregations. They ascend and descend the water column close to the side of the beaker. The embryos are visible to the naked eye at this stage. In diffuse light the embryos get scattered and swim individually.

Trochophore larva

With the reorientation of the blastopore to the ventral side, the dorso-posterior region of the embryo is marked by a straight thickening of the ectoderm at the animal pole. A single apical flagellum, 15μ long, develops at the anterior end and a band of strong cilia appears in the pretrochal part (Pl. II E). A tuft of cilia also develops at the opposite end. The minute cilia noticed earlier in the blastula stage have disappeared. A shell gland forms from the thickened dorsal ectoderm and secretes the first transparent larval shell, the prodissoconch I. The trochophore stage (Pl. II E) thus formed settle to the bottom of the beaker. The apical flagellum in some of the larvae grow to 47μ and in such cases the movement of the larvae is impeded by entangling with the flagella of others.

Veliger

Transformation of the trochophore to veliger stage is brought about by the following prominent changes : deposition of the larval bivalve shell (prodissoconch II) by the developing mantle ; the rearrangement of preoral cells into a powerful velum with well-developed cilia ; disappearance of the apical flagellum and preoral and posterior ciliary bands and elongation of the larva along the antero-posterior axis with the formation of the larval organs (Pl. III A, B).

The shell of the veliger is transparent with conspicuous granules obscuring observation of the developing organ systems. The margin of the shell is darker. The shell assumes

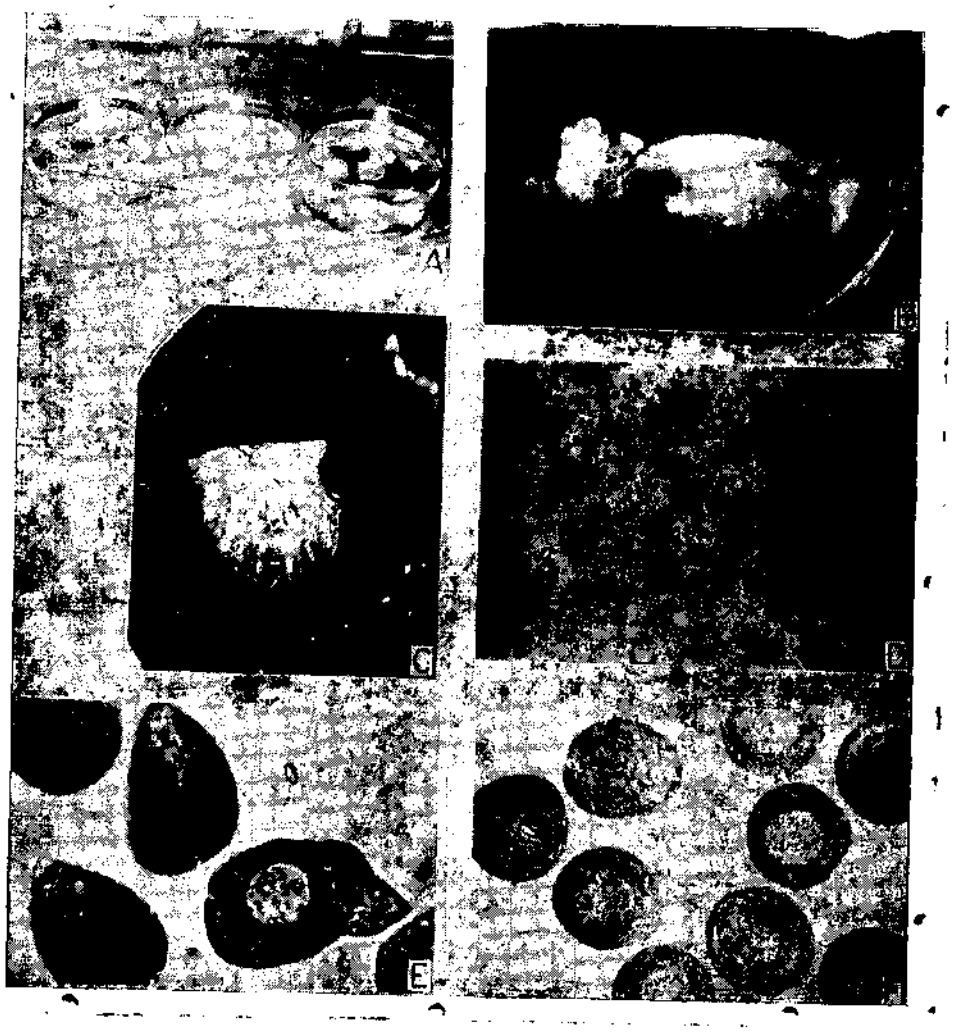

PLATE I A. Spontaneous spawning of the Indian pearl oyster *Pinctada fucata* (in the middle trough); B. Male spawning in a continuous stream; C. Female spawning eggs in discontinuous spurts, eggs entangled in broken gonadal tissue seen at the right top; D. Sperms; E. Just spawned eggs, large clear germinal vesicle is seen in one of the pyriform eggs and F. Eggs assume spherical shape, germinal vesicle is clearly seen in some while in others it is breaking down.

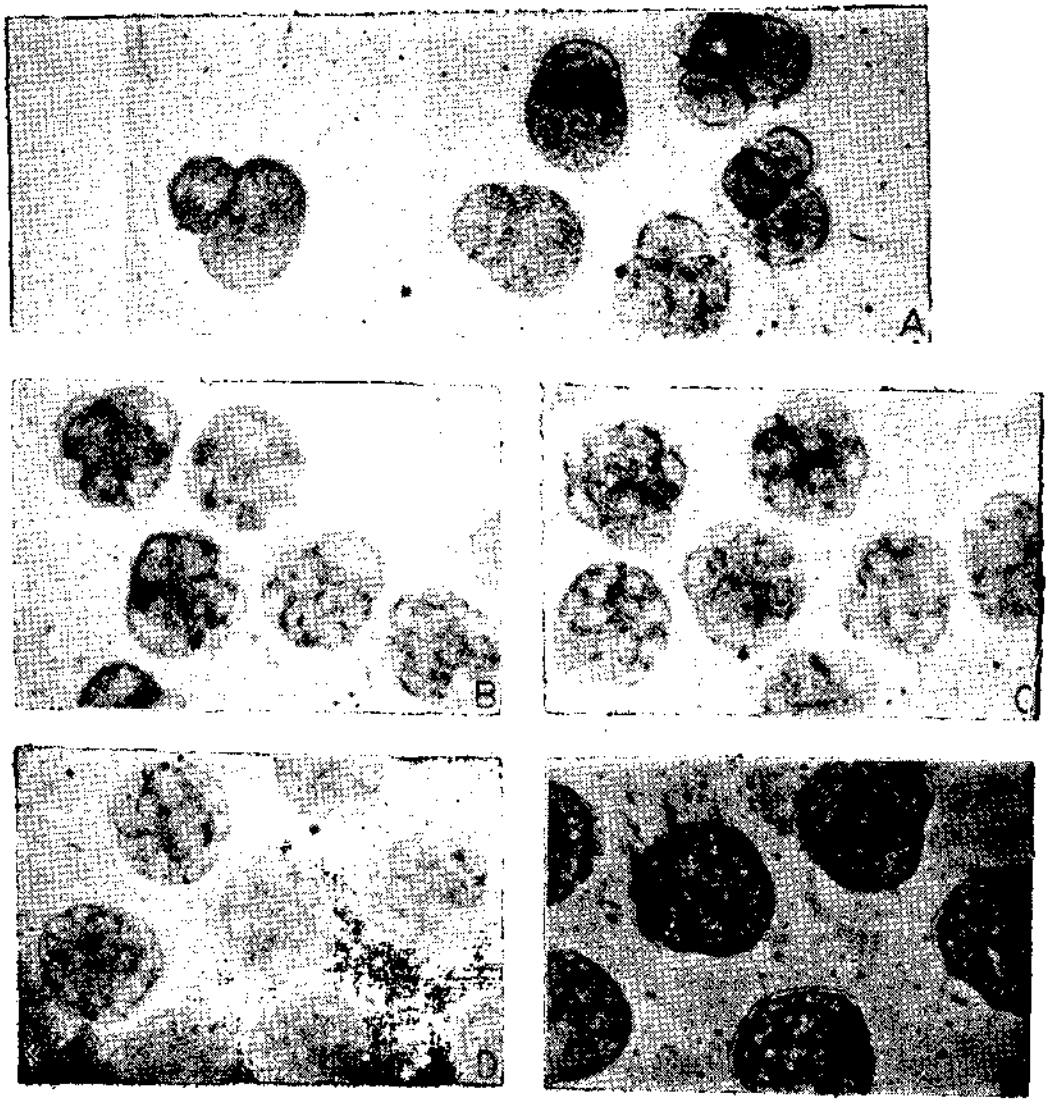

PLATE II A. Early stages of cleavage of *Pinctada fucata* eggs, at the extreme left is seen the first cleavage stage ; one of the micromeres is seen fusing with the polar lobe to form the macromere ; other figures show the 2-cell and 4-cell stages, the polar body is seen at the furrow of cleavage ; B. 8-cell stage ; C. 16-cell stage ; D. Advanced stage of cleavage, one macromere is still seen distinct from the micromeres which are spreading over the former to form the morula and E. Trochophore larvae, a single long apical flagellum and the ciliary band can be seen anteriorly and in the dorso-posterior region a straight ectoderm thickening has been formed.

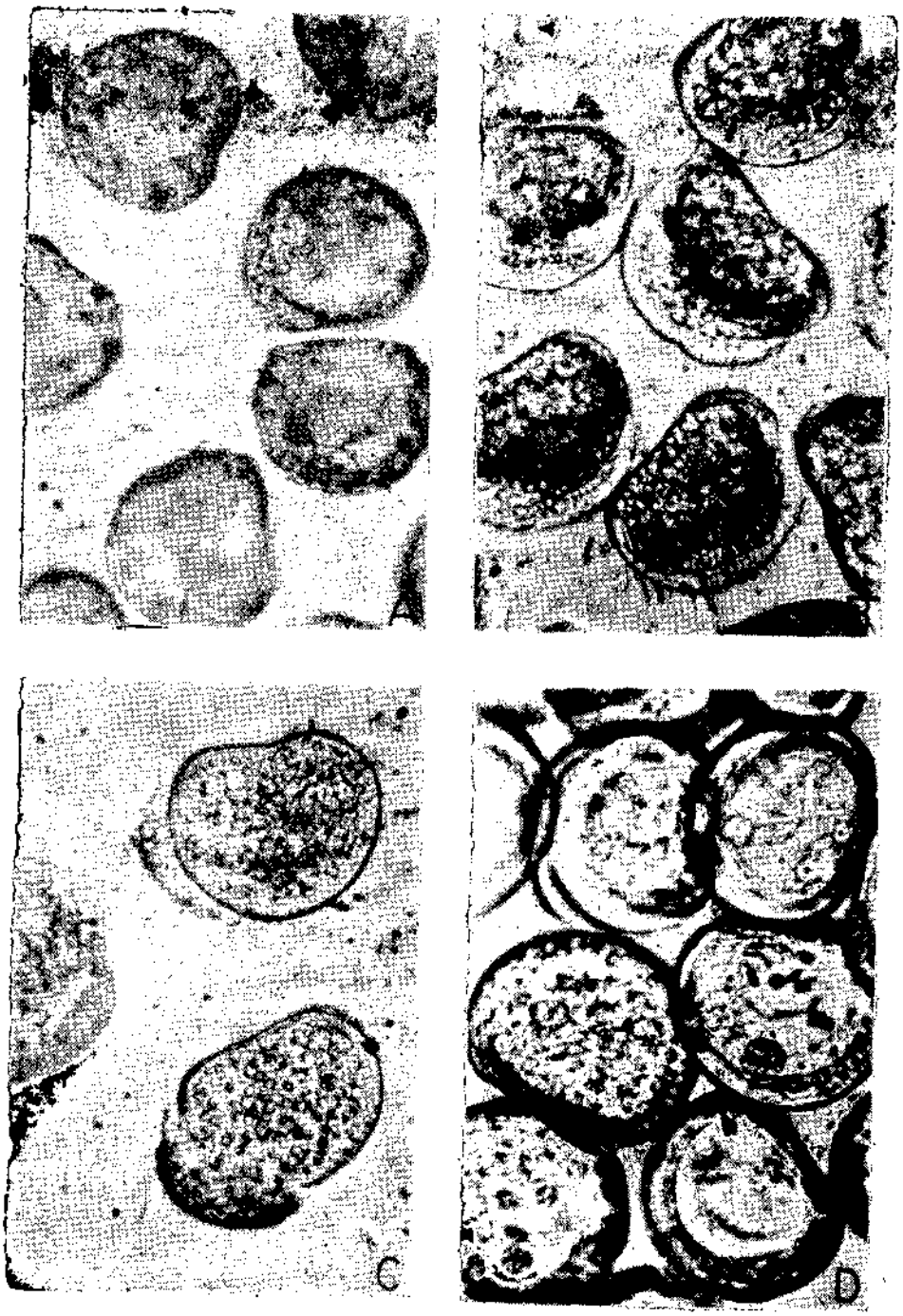

PLATE III A. Early veligar larvae of *Pinctada fucata* ; B. Straight-hinge larvae; C. The straight-hinge larvae are heavily granulated, velum is seen protruding and the ciliary action is clear, and D. Mortality of straight-hinge larvae, the larvae die due to lack of appropriate food, the inactive larvae are attacked by ciliates which take a heavy toll.

typical straight-hinge on the dorsal aspect and a curved lower margin. The velum protrudes outside creating a strong ciliary current which wafts the minute particles into the stomodaeum (Pl. III C). The ciliary current also assists in the movement of the larvae. Ciliary movement can be seen in the digestive diverticula. The velum and other organs are heavily globulated (Pl. III C). The straight-hinge larva measures an average 67.5μ along the antero-posterior and 52.5 μ along the dorso-ventral axis. Some of the largest larvae measured 72.5μ × 57.5μ. The straight-hinge stage was reached in 20 hr 40 min after fertilisation.

TABLE 1. *Sequence of development in the pearl oyster* Pinctada fucata *from fertilization to straight-hinge larva*

Developmental Stage	Time sequence hr. min.	Size (μ)
Pyriform egg	—	73.9 × 45.2
Spherical egg	0-15	47.5
2-cell stage	0-45	60.0
4-cell stage	1-00	60.0
8-cell stage	1-18	62.5
Blastula	5-54	57.5
Beginning of rotation	6-45	57.5
Apical flagellum develops	7-30	57.5
Trochophore	10-54	57.5
Early veliger	18-54	65.0 × 52.5
Straight-hinge larva	20-40	67.5 × 52.5

Attempts on larval feeding

The straight-hinge larva marks the end of one phase in the development of *Pinctada fucata* obtained without much difficulty and also the beginning of a critical phase where further development would depend on the management of the environment of culture and supply of food. Attempts were made to feed the larvae with the microalgae *Tetraselmis gracilis*, *Synechocystis salina* and *Chlorella marina* and also yeast patricles. Although the larvae survived for a maximum period of 30 days no further growth was observed. Ciliates which start appearing about 12 hours after fert lisation start taking an increasing toll of the pearl oyster larvae (Pl. III D). Experiments on feeding and control of the culture medium are in progress for taking the straight-hinge larvae to the full-grown stage and obtaining spatfall in culture tanks.

Time sequence of development

The time taken for reaching different stages of development is given in Table 1. Variations in time and size of embryos and larvae have been noticed within and between broods. The data given in the Table relate to one series.

DISCUSSION

The development of the Indian pearl oyster upto the straight-hinge larva shows a striking similarity to that of the Japanese pearl oyster. The stages of development illustrated by Ota (1964) are identical to those observed in the present study. Kobayashi (1948), as cited by Cahn (1949), has observed that, in the Japanese pearl oyster, the unfertilised egg measuring 48μ reaches the veliger stage (66 μ × 50 μ) in 24 hours. In the Indian pearl oyster, the unfertilised egg measuring 47.5 μ reaches the straight-hinge veliger stage (67.5 μ × 52.5 μ) in 20 hours 40 minutes. Wada (1942), as cited again by Cahn (1949), has reported the same-type of early development for *Pinctada maxima* in the tropical Palau region. In *P. maxima* the mature egg measuring 59μ reaches the veliger stage (77μ × 62μ) in 18½ to 19 hours. Kobayashi (1948) found that the optimum temperature for fertilisation was 28°-30°C Wada's (1942) experiments were conducted at 26°-31°C. In the present study, fertilisation and development took place in equally higher temperatures of 27.0°-28.9°C. It is seen that within the higher temperature range, development proceeds with close similarity in *P. fucata* of the Japanese and Indian waters and *P. maxima* of the tropical Palau region.

The Japanese pearl oyster larva reaches the umbo stage (217μ × 189μ) in 10 days and full-grown stage (304μ × 269μ) in 20 days as cited by Cahn (1949) and spat attachment (443μ × 391μ) takes place 25 days after fertilisation Ota (1957) reported that the free-swimming planktonic larva measures 90-100μ at the D-shape or straight-hinge stage, 120-130μ at the umbo stage, 170-200μ when it has the pigment spot and 200-230μ at full-grown stage. For the Indian pearl oyster these advanced stages of larva remain yet to be studied. However, Herdman (1903) and Hornell (1922) have attempted to bridge the gap between the veliger larvae of *Margaritifera vulgaris* (=*Pinctada fucata*) reared in the laboratory and the earliest attached spat seen on the pearl banks of the Gulf of Mannar off Sr. Lanka Coast by the forms that they collected by two-net on the pearl banks. Later, based upon the descriptions of these forms, Sudhakar and Chandrasekharan (1968) ascribed certain larvae measuring from 243μ × 217μ to 365μ × 278μ collected on the Tholayiram paar in the Gulf of Mannar off the Indian Coast to *P. fucata*. However, positive identification of the pearl oyster larvae in the plankton would be possible only through the direct method of rearing the larvae to the spat stage.

Tranter (1958) observed that in *Pinctada margaritifera*, activation of eggs takes place immediately before spawning. His observations are based on sections cut through spawning ovaries and ingested eggs and not on spawned eggs. In the present study on *P. fucata* it was observed that the spawned eggs are mostly irregular and they assume spherical shape within 15 minutes after spawning. Germinal vesicle persists even in spherical eggs for some time before it breaks down. Kuwatani (1965) experimentally found that the lowest concentrations of ammoniated sea water needed for nuclear breakdown in more than 80% of eggs of the excised gonads of *P. fucata* was $0 - 5.70 \times 10^{-3}$ *N* NH_3. Wada (1961, 1963) has carried out similar experiments on the disruption of germinal vesicle using ammoniated water on the eggs of *Pinctada*, *Crassostrea* and other bivalves. In the present study, normal development of eggs was observed from natural spawning and disruption of germinal vesicle took place in a majority of eggs without the ammoniated sea water medium.

Ukeles (1971) has made and extensive review of the nutritional requirements in shellfish culture. Kobayashi (1948) reared pearl oyster larvae on *Monas* culture. Wada (1973) found that *Monochrysis lutheri* (7×2.5μ) and *Chaetoceros calcitrans* (4 × 2μ) proved to be good single foods for the D-shape larvae (straight-hinge stage) of *P. fucata* and that a mixture of the above two provided slightly better growth rate. He also observed that larvae fed with *Chlorella* sp. (2 - 4μ) showed the poorest growth rate and remarked that although the young larvae cannot utilise algae such as *Chlorella* with a cell wall, the older ones can feed and digest these. Attempts by us to feed the pearl oyster larvae with *Tetraselmis*, *Synechocystis* and *Chlorella* which are some of the common forms in the nannoplankton have not so far given any success. Rao *et al.* (1976) have reared the larvae of *Mytilus viridis* upto the pediveliger stage feeding them with *Chlorella*, *Tetraselmis gracilis*, *T. chui* and *Synechocystis* sp. Although the mussel larvae were maintained by them upto 56 days, spat settlement did not take place. Thus the nutritional aspect remains a major constraint in the development of hatchery technology for the marine bivalves in India.

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STUDY ON THE STOMACH CONTENTS OF PEARL OYSTER *PINCTADA FUCATA* (GOULD) WITH REFERENCE TO THE INCLUSION OF BIVALVE EGGS AND LARVAE

A. CHELLAM*

Central Marine Fisheries Research Institute, Cochin-682 018

ABSTRACT

The pearl oyster which is a filter-feeder as any other bivalve mollusc, has been considered to feed on phytoplankton including detritus. A detailed study made on the feeding of pearl oyster *Pinctada fucata* collected from the pearl culture farm as well as natural beds showed the presence of bivalve eggs and larvae along with copepods and crustacean larvae, spicules of sponges, etc. Several samples of oysters collected at different periods of 1977 and 1978 gave the same results indicating that this is a regular feature.

Experiments were conducted to elucidate the role of bivalve larvae in the feeding of pearl oysters. Trochophore larvae of *P. fucata* obtained in rearing experiments were fed to starved oysters. These were readily ingested by the pearl oysters. The larvae were passed out in the faecal matter alive and these, when reared, developed into straight-hinge stage.

INTRODUCTION

OUR knowledge of the food and feeding habits of the pearl oysters in the Indian waters is very meagre apart from the passing reference made by Herdman (1903). Ota (1959) has studied the feeding habits of the Japanese pearl oyster and the production of faeces in relation to seasonal changes, nuclear insertion and culture conditions. He had observed that the pearl oysters feed on the swimming bivalve larvae. Upon frequently noticing the presence of bivalve eggs and larvae in the stomach of the Indian pearl oyster, a detailed study was carried out on the significance of their presence in the feeding regime of the species and the results are reported.

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MATERIAL AND METHODS

During the course of a study on the biology of pearl oyster *Pinctada fucata* in the Veppalodai pearl culture farm and the Gulf of Mannar pearl banks, the stomach contents of pearl oyster were examined based on regular samples. A total of 426 and 154 oysters for the years 1977 and 1978 respectively from the farm and 150 and 70 oysters during the same period from the pearl banks were examined for qualitative assessment of the stomach contents.

Starved oysters were fed with the laboratory reared pearl oyster larvae. The stomach and intestinal contents of these oysters were examined

* Present address : Tuticorin Research Centre of the Central Marine Fisheries Research Institute, Tuticorin.

periodically to find out the fate of the larvae ingested. The pseudo-faeces which were discharged by the oysters were examined and the larvae thrown out were separated and reared. The faecal matter was also examined and the larvae present were separated and reared.

Observations

Stomach contents of oysters from the farm

The stomach and intestinal analysis of oysters from the farm showed that 59.5 and 66.3 per cent of the stomachs contained eggs and shelled larvae of bivalves respectively, almost throughout the year. The size range of the straight-hinge larvae was from 27.5 to 115 μ in the dorsoventral axis and 37.5 to 125 μ in the anteroposterior axis. They were of different shapes suggesting that they belong to more than one species. Larvae with umbone ranged in size from 162.5 to 232.5 μ in dorsoventral axis and 200 to 275 μ in the anteroposterior axis. Other larvae found in the stomach were gastropods in March, April, August and September, heteropods in April, June, August and October, and nauplii in July, August, October and November. Copepods and their appendages, spicules of sponges and some unidentified spores were the other constituents. *Euglena* and *Ceratium* along with some other flagellates were also present. The diatoms were represented by *Navicula, Nitzschia, Oscillatoria, Coscinodiscus, Gyrosigma, Pleurosigma, Asterionella, Thallasiothrix, Diploneis, Amphora, Rhizosolenia* and *Chaetoceros*. The organisms were found in a matrix of mucus and organic detritus. Some were in semidigested condition and occasionally frustules of copepods too were observed. A few minute sand particles also were present in the stomach content of oysters.

Stomach contents of oysters from the pearl banks

Oysters from Vaipar Peria paar, Devi paar and Tholayiram paar collected from January to April and November to December were analysed for the study. The depth at which the oysters were collected ranged from 12-21 m.

Of the stomachs examined, 71.4 per cent contained eggs of pearl oysters throughout the course of observation and in 4.3 per cent cases shelled bivalve larvae were present during April and November of both the years. The larvae were at different stages of development in straight-hinge and umbonal stages. Copepods and their appendages, nauplii, spicules of sponges, *Euglena* and flagellates were the other inclusions in the stomach. The diatoms enumerated earlier were also present. These organisms were found mixed in a good amount of organic detritus along with few sand particles.

Experimental Feeding of Oysters

As the presence of bivalve larvae was a regular feature in the stomachs of pearl oysters especially from the pearl culture farm, some experiments were conducted in the laboratory to find out whether the larvae are ingested as food and if so whether they are digested.

Twenty healthy oysters from the farm were brought to the laboratory and five were examined immediately for their stomach and intestinal contents. All the oysters were in spent condition. Eggs of different shapes, bivalve larvae of straight-hinge and umbo and diatoms were present in them. Detritus was in fairly good quantity with few sand particles. The remaining oysters were kept in clean, filtered sea water. Next day, the examination of the faecal matter showed the presence of bivalve larvae, pteropods, appendages of copepods, detritus and sand particles. Examination of the contents of stomachs and intestines of three oysters on opening showed that they were empty except for the presence of few sand particles. Ten such oysters were fed with straight-hinge larvae in filtered sea water medium. Within 45 minutes all the larvae in the water had been filtered and ingested. The pseudo-faeces ejected as puffs of loose mucus was found to contain large number of larvae of pearl oysters entangled in mucus.

The faecal matter as ribbon-like extrusions began to appear within 2 hours of feeding. A large number of pearl oyster larvae wrapped in mucus like material were present in it. The faecal matter extruded next day was found to contain only mucus-like matter and sand particles. The stomach and intestinal contents of three oysters cut open showed that they were empty except for mucus and a few sand grains.

In another set of experiment, ten oysters in healthy condition, kept starving over night, were fed with laboratory reared trochophore larvae, 5 hours old. The purpose of the experiment was to find out whether the larvae passed out in the faecal matter were alive and if so were capable of further development. The pseudofaeces was collected and the larvae embedded in the loose mucus were separated and reared. The larvae developed further into straight-hinge stage. The larvae which were separated immediately from the faecal matter also developed to straight-hinge stage. It was observed further that the larvae were able to pass through the alimentary system within 2 hours of feeding. Most of the larvae retained in the stomach for longer time were dead. A few larvae passing out alive after a prolonged period were feeble and inactive.

Discussion

Herdman (1903) noted that the unicellular organisms such as spores of algae, diatoms, infusorians and foraminifers formed largely the food of pearl oysters and that, radiolarians, minute embryos and larvae of various animals, algal filaments, spicules of Alcyonarians, sponges and small numbers of minute sand grains were also found in the stomach occasionally in large quantities. Ota (1959) has observed the swimming bivalve larvae also in the stomach contents of the Japanese pearl oyster during summer. Kuwatani (1965), feeding the oysters with charcoal particles, noticed particles upto 30 × 17.5 μ size in the oesophagus and only 17.5 × 10 μ size and less had passed through the digestive diverticula from the stomach. Cerruti (1941), quoted by Korringa (1952), has recorded as stomach contents of *Ostrea edulis* large quantities of organic detritus, diatoms, flagellates, larvae of molluacs, eggs and gastrula of a variety of marine invertebrates, plant fibres and pollen grains and he found that only very small diatoms were reduced to empty frustules in the intestinal tract.

According to Korringa (1952) the feeding method of lamellibranchs certainly is very wasteful. Only part of the material ingested can be digested. No evidence of selection of food is found and the actively feeding oyster appears to ingest anything that it can ingest. Some microorganisms found in the stomach are not digested and they emerge from the oyster unchanged, often alive. Lamellibranch larvae are often ingested in great numbers. They always leave the oyster alive. These larvae very rarely succeed in freeing themselves from the mucus and organic detritus they are wrapped in, and therefore are doomed to perish. The experimental feeding of pearl oysters with the laboratory, reared pearl oyster larvae shows that most of the larvae ingested pass through the alimentary tract often alive. If they pass out within two hours, they could develop further if separated from the faeces immediately.

The present observations on the feeding of pearl oyster *P. fucata* are in agreement with the previous works on oyster feeding. The presence of diatoms, dinoflagellates, larvae of various animals, plants and other animal remains, detritus and sand particles in the stomach of pearl oyster shows that the pearl oyster, like any other bivalve mollusc, ingestes all the materials that it can. The plankton investigations in the Veppalodai pearl culture farm area during the years 1975 to 1977 (to be reported elsewhere) showed that the lamelli-

branch larvae were present throughout the year with a major peak during July to November and a secondary one from March to May. The measurements of the planktonic lamellibranch larvae of straight-hinge and umbonal stages were close to those of the larvae found in the stomach of pearl oysters from the farm at Veppalodai.

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SETTLEMENT AND GROWTH OF BARNACLE AND ASSOCIATED FOULING ORGANISMS IN PEARL CULTURE FARM IN THE GULF OF MANNAR

S. DHARMARAJ* AND A. CHELLAM*

Central Marine Fisheries Research Institute, Cochin-682 018

ABSTRACT

The settlement of fouling organisms on the pearl oysters, pearl oyster shells and wooden test panels was compared. The barnacle *Balanus amphitrite variegatus* was the major fouling organism observed and the polychaete *Polydora ciliata* and the sponge (*Cliona vastifica*) were the main boring organisms. The intensity of settlement at different depths and the period of abundance of these organisms were studied. The successive settlement and fast growth of barnacles resulted in heavy loading on the oysters within a short period. The fouling load and the rate of growth of barnacles were higher on the shells than on live oysters. The settlement was more at the top rows of the sandwich-type frame net.

INTRODUCTION

A DETAILED study on the fouling and boring organisms in the pearl culture farm at Veppalodai has been made by Alagarswami and Chellam (1976). Kuriyan (1950) and Ananthanarayanan (1967) have given a list of fouling organisms on the pearl oyster cages at Krusadai Island, Gulf of Mannar. The pearl oysters reared in the farm at Veppalodai are affected by fouling throughout the year. Besides elucidating the problems of biofouling with emphasis on barnacles, the study, reported here, aimed at comparing settlement and growth of barnacles on wooden panels and pearl oysters on one hand and on pearl oyster shells and live oysters on the other.

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Present address: Tuticorin Research Centre of Central Marine Fisheries Research Institute, Tuticorin-628 001, Tamil Nadu.

MATERIAL AND METHODS

Three series of experiments were conducted to study the fouling and boring aspects in the pearl culture farm at Veppalodai using the following material as the substratum: (i) wooden test panels, (ii) live oysters and (iii) shells of pearl oysters. Each wooden panel unit consisted of three square planks, each of 20 cm × 20 cm × 2.5 cm, which were suspended vertically from the raft in a manner that the top-most one was at 0.5 m, the middle one at 1.4 m and the bottom one at 2.0 m from the surface of the sea water and the depth of water below raft was 2.0 m. The panel of three planks was terminated at the end of days and a fresh one was introduced. The barnacles were counted irrespective of their size. Height of the barnacles was taken as the standard measurement as Crisp and Patel (1965) have done. The study covered a period of 12 months from January to December, 1975. Another set of wooden panels suspended simultaneously gave

the data on cumulative fouling load and growth of barnacles for a period of 1, 2, 3, 4, 5, and 6 months. Beyond that period the panels could not be kept in the sea due to heavy damage by the pholad mollusc *Martesia* sp. Hence a fresh set was introduced for the second 6 months of the year.

In the second and third series of experiments, commencing March 1977, live pearl oysters and shells (both valves) were arranged in separate sandwich-type frame nets and first, third and fifth rows of the net were recorded.

Results

Monthly panels

Balanus amphitrite variegatus was dominant fouling organism on the panels throughout the year. *B. a. communis* and *B. a. venustus* were few. The settlement of barnacles on the panels at different depths was found to differ from month to month. From January to middle of June the settlement was less. During

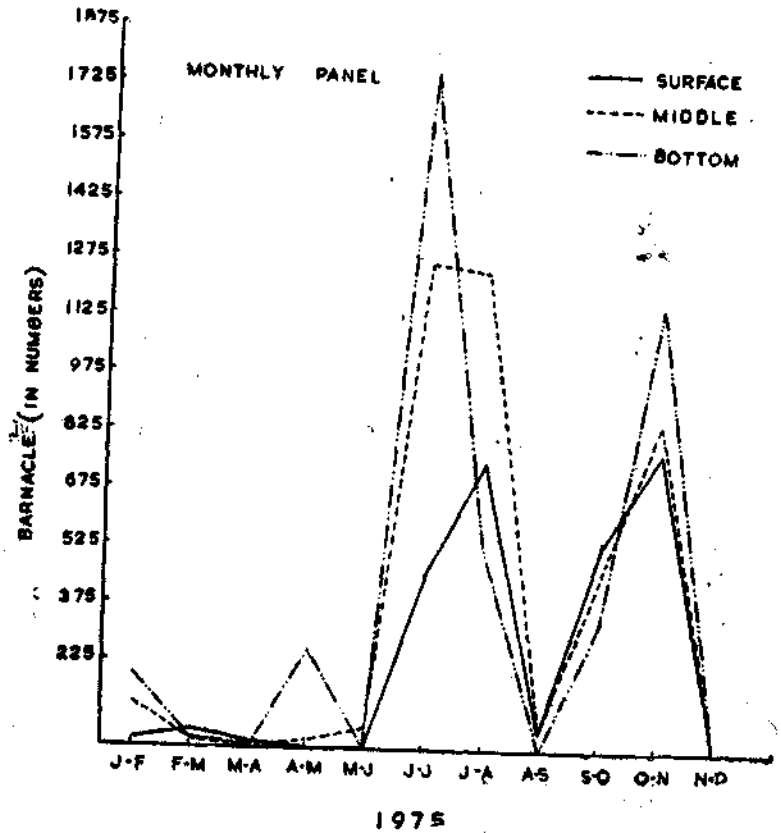

Fig. 1. Settlement of barnacles on the wooden panels in the pearl culture farm at Veppalodai during 1975.

suspended simultaneously from the rafts after recording the measurements and weight of individual oyster and shell. These were examined on completion of one month. The number and size of barnacles which have settled on both valves of oyster and shell in the this period the total number of barnacles settled on all the three panels (at different depths) ranged from 26 (in March) to 268 (in April). Two peaks of heavy settlement were recorded, one was fron middle of June to August and the other from September to November (Fig. 1),

A minimum of 2,500 barnacles (July) to a maximum 3,460 barnacles (June) were noted during the first peak and in the second peak it ranged from 1,290 (September) to 2,710 (November). When the total number of barnacles settled was taken into account for the whole period, the bottom panels showed higher settlement rate (39.3%) and the surface panels had less (23.8%).

For the period from January to December the size of the barnacles settled ranged from 0.1 to 9.0 mm in height. In the first half the maximum size of barnacle recorded was 6.0 mm and it was 9.0 mm in the second half. The size 0.1-1.5 mm dominated on the panels during February to June and December. From July onwards the dominant size group was found to be 1.6-3.0 mm. It was 4.6-6.0 mm in November and 3.1-4.5 mm in January.

Cumulative fouling

Fluctuation in settlement of barnacles on the longterm panels at different depths was noted In the first half of the year, the settlement was more on the bottom panels in the first 2 months but it was not the case in the 3rd, 4th, 5th and 6th months in which the high settlement was on the surface panels. The total number of barnacles on the three panels at different depths for 1, 2, 3, 4, 5 and 6 months was 313, 99, 65, 6280, 5140 and 2730 respectively. When the total settlement of barnacles for the 6 months was considered, it was 47.0% on the surface panel, 35.6% on the middle panel and 17.3% on the bottom panel. Barnacles of size 1.5mm and less in height were present in all months. The barnacles which settled on panels kept in January 1975 grew to 6.2 mm in July 1975 and 26.9 mm by January 1976.

The weight of fouling on the surface panels had a uniform increase from 1 to 6 months. In the first month, it was merely 8 g but in the sixth month it had increased to 268.5 g. On the middle panels the increase in weight of fouling was from 15 to 89.5 g. On the bottom panels the weight of fouling ranged from 20.5 to 103 g. For the first 6 months the surface panel had 45.5% of fouling weight, the middle one 28.3% and the bottom ones 26.2%. It was also noticed that the volume of total fouling on the surface panels increased uniformly from 3.5 ml to 179 ml for the first to sixth month. In the case of middle and bottom panels, the total fouling volume was not uniform and was less compared to the surface panels.

During the second half, the total number of barnacles settled on all the panels was 2500, 5540, 2760, 2720, and 1680 for the 1, 2, 3, 4, and 5 months respectively. More number of barnacles settled on the middle panels (36.7%) followed by the bottom panels (33.4%). The surface panels had less barnacles (29.8%). In this half year the maximum size ranged from 7.4 mm to 26.9 mm.

There was no definite pattern in the settlement of other fouling organisms at different depths on monthly and long-term panels. Of the five foraminiferans observed on these panels, *Discorbis* sp., *Quinqueloculina* sp. and *Orbulina* sp. occurred in the three levels throughout the year 1975. The presence of amphipods (from May to December) bivalve molluscs (from March to November) and nematode worms (from June to October) was recorded. Other organisms such as *Nymphon* sp., *Cryptophallus* sp., *Sphaeroma* sp., Gastropods (*Oliva* sp.), *Modiolus* sp., *Avicula* sp. and simple ascidians were found in lesser numbers during certain months of the year.

Fouling on oysters

The average surface area of both the valves of the oyster used for the experiment ranged from 42.6 to 59.1 sq.cm. The number of barnacles per unit area of 10 sq. cm on an oyster in the first, third and fifth row of the frame net is given in Table 1. The settlement per unit area per oyster ranged from 3.4 (in May) to 62.2

TABLE 1. *Settlement of barnacles and fouling load on pearl oysters and shells in sandwich-type nets in the pearl culture farm at Veppalodai in 1977*

Period	No. of days	Materials	No. of barnacles per 10 sq. cm			Fouling load per oyster/ shell (pair) (g)			Fouling load on net (gm)
			I row	III row	V row	I row	III row	V row	
March	30	O	11.6	5.7	5.8	..	..	..	..
		S	14.5	13.4	10.2	..	..	..	..
Mar.-Apr.	29	O	25.2	7.5	8.2	..	..	..	..
		S	62.9	39.6	35.7	..	..	..	..
April-June	39	O	0.4	3.0	3.2	..	..	..	..
		S	3.4	2.6	1.8	..	..	..	..
June	21	O	15.3	12.4	14.0	..	..	..	..
		S	18.0	20.6	16.4	..	..	..	..
June-July	31	O	21.6	9.2	9.9	9.1	7.4	7.5	400
		S	28.7	23.9	21.2	17.5	12.9	13.3	500
July-Aug.	32	O	21.8	15.3	11.8	6.8	10.1	11.7	500
		S	24.9	25.2	35.7	25.5	16.8	13.6	750
Aug.-Sept.	32	O	29.4	29.4	20.6	2.4	4.3	1.7	300
		S	53.2	32.5	25.1	6.5	5.3	6.6	910
Oct.-Nov.	34	O	24.5	19.4	15.0	14.0	11.2	7.4	1300
		S	16.3	..	37.0	19.4	32.2	40.0	1500
Nov.-Dec.	35	O	65.5	..	57.4	34.3	32.0	..	2240
		S	107.2	..	120.7	48.0	63.9	..	2490

O = Oyster ; S = shell (pair).

barnacles (in November). The settlement of barnacles on the unit area was 8.2 in March to May ; 14.0 in June to August and 36.1 in September to November. The maximum size of barnacles on oysters was found to be 8.9 mm in August. The total number of barnacles on oysters of each row of the frame net showed variation. In the first row it was high and the intensity decreased towards the bottom rows. The average number of barnacles on unit area for the period from March to November was 23.9 in the first row and 16.2 in the fifth row. In November a maximum fouling load of 33.1g/ oyster was recorded. The accumulation of fouling organisms and other suspended detritus on the frame net holding oysters increased in weight from 400 g in July to 2,240 g in November.

Bryozoans and hydrozoans occurred on all the oysters throughout the period of observations. Other organisms observed in good quantity were amphipods and *Hydroides* sp.

Fouling on shells

The shells used in the experiments had an average surface area of 54.7 to 63.6 sq.cm. The settlement of barnacles per unit area of

10 sq. cm of shell was 2.6 barnacles in May and 113.9 in November. The number of barnacles in the unit area of the shells was 19.6 (March to May); 23.8 (June to August) and 59.1 (September to November). The maximum size of barnacle recorded on the shells was 15.7 mm (August). The variation in settlement on different rows of shells in frame net also existed in each month (Table 1). The average number of barnacles per unit area for the whole period of observations was 36.7 on the first row and 33.6 on the fifth row. The weight of fouling organisms and detritus on the net holding the shells ranged from 500 g (March) to 2490 g (November). A maximum fouling load of 55.9 g per shell was recorded in November.

The settlement of bryozoans on the shells was rare compared to oysters where each oyster was fouled by it. A sparse settlement of hydrozoans was noted. Amphipods were also observed in lesser numbers on the shells.

Boring organisms

The intensity of boring by the polychaete *Polydora ciliata* and by the sponge *Cliona vastifica* was found to differ from place to place. *P. ciliata* was more in the sheltered bay farm at Tuticorin Harbour than at the shallow farm at Veppalodai. Oysters from the natural pearl banks showed very less boring. Blisters caused by the boring polychaetes were practically few in the oysters of 40 mm in DVM and less. It was also noticed that farm oysters were subject to boring, whereas oysters collected from the south breakwater of the Tuticorin Harbour (depth 0.5 - 1.0 m) showed negligible instances. The maximum infection by *C. vastifica* was on the Vappalodai farm oysters and the intensity was less in harbour farm and negligible in pearl banks.

The pholad mollusc *Martesia* sp. was not a common borer on the oysters of the farms. There were a few instances of boring by the mollusc on oysters kept for a prolonged period in the farm. Its destructive nature was noted on the test panels which could not be retained in the sea for more than six months. The intensity of attack was high on the surface panels and less on the bottom panels.

REMARKS

The settlement of barnacles on the panels at different depths varied from month to month. The bottom panels showed a higher settlement (39.3%) during the year 1975. Depth may influence the settlement of barnacles and *B. a. variegatus* was found to prefer 5 feet level for maximum settlement in Madras Harbour (Antony Raja, 1959). Barnes (1956) found that most liberations of cirripede larvae occurred when temperature was high and minimum plankton and less silt were observed. However, the turbid nature of water in the shallow farm at Veppalodai probably afforded ideal conditions for the settlement and growth of numerous fouling organisms (Alagarswami and Chellam, 1976). Skerman (1956) found in Auckland Harbour that silt or other suspended detritus in sheltered regions can get incorporated on the slime films that develop on panels and these granular films can promote barnacle settlement on these surfaces.

Disparities in the occurrence of barnacles on the long-term panels are not uncommon. Nair (1965) observed that the number of barnacles remaining over the blocks after prolonged periods of exposure (1 to 6 months) need not give correct indication of the number that actually settled over the blocks. From January to June the maximum growth of barnacles on monthly panels was found to be 6.0 mm in height and from July to November it was 9.0 mm. On long-term panels the best grown barnacles were 6.4 mm in height in the first half and 26.9 mm in the second half. This clearly indicates that the period from July to November was better for good growth.

The intensity of boring was high in oysters of size 40 mm and above. Herdman (1905) also had reported the destructive nature of boring by *Cliona margaritiferae* on the Cheval paar oysters of Sri Lanka pearl banks. A high intensity of boring by the sponge *Cliona vastifica* has been observed on the oysters at Veppalodai farm. The infestation of *Martesia* sp. was mostly confined to shallow inshore waters of the seas and estuaries and at depths ranging from 0.5 to 6.0 m attack of the borer is quite predominant (Balasubramanyan, 1968). Of the test panels suspended at depths ranging from 0.5-2.0 m, the surface ones were damaged more and the intensity decreased towards bottom.

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ON THE OCCURRENCE OF SHELL BORING POLYCHAETES AND SPONGES ON PEARL OYSTER *PINCTADA FUCATA* AND CONTROL OF BORING ORGANISMS

T. S. VELAYUDHAN*

Central Marine Fisheries Research Institute, Cochin-682 018

ABSTRACT

Boring organisms, particularly polychaetes and sponges cause considerable damage to the pearl oyster *Pinctada fucata* reared in the farm. The most serious pests are the polychaete *Polydora ciliata* and the boring sponge *Cliona vastifica*. The incidence of attacks by the boring organisms on oysters from the pearl culture farm at Veppalodai and Tuticorin Harbour and from the natural beds have been studied and the oysters from Veppalodai were found to be the most affected.

Of the several methods tried to control boring organisms, immersion of the oysters in fresh water was found to be most effective in controlling the polychaetes. Brushing the external surface of the shells with 1% formalin was found to be effective in controlling the boring sponge.

INTRODUCTION

THE FOULING and boring organisms in the pearl culture farm always pose problems. Nishii (1961), Mizumoto (1964, 1966, 1968) and Yamamura *et al.* (1969) have reported in detail the fouling and boring organisms encountered in the pearl culture farms in Japan. Alagarswami and Chellam (1976) have made a comprehensive study on the fouling and boring organisms in the pearl culture farm at Veppalodai. Shirai (1970) discusses the use of saturated salt solution for the removal of the polychaetes from the pearl oyster shells. Korringa (1952) reports some of the parasites and diseases of the oysters and the control measures. De Laubenfel (1930) has reported the use of fresh water for killing the boring sponges from the oysters. In Indian waters, no work appears to have been done on the control of boring organisms on the oysters. The experiments carried out during 1978-79 to control boring organisms on pearl oysters and the possibilities for making use of the results of the work on the farm oysters are discussed in the present paper.

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MATERIAL AND METHODS

In the Veppalodai pearl culture farm, pearl oysters were reared at a depth of 3 to 4 metres by

* Present address: Tuticorin Research Centre of Central Marine Fisheries Research Institute, Tuticorin-628 001, Tamil Nadu.

raft culture method. In the Tuticorin Harbour basin rafts were moored at a depth of 8 to 10 metres. The farm oysters as well as freshly collected oysters from the pearl banks in the Gulf of Mannar were examined for the presence of shell boring organisms from January 1978 to September 1979.

Treatment with 1% formalin on the shells

After noting the presence of boring polychaetes, sponges and *Martesia* sp. on 225 oysters, experiments were conducted in five batches. The oysters were arranged in a frame net of 45 × 65 cm with equally divided horizontal compartments. Dilute 1% formalin was brushed on the right and left valves of the oysters. The oysters were kept in air for 15 minutes to 2 hours and, after washing in fresh water, transferred to normal sea water. The effect of the formalin on *Martesia* sp. and sponge was noted after keeping them in the farm for one week.

Immersion in brine solution

The oysters affected by boring polychaetes were selected and were immersed in brine solution ranging in salinity from 42.3 to 78‰ in containers of 5 litres capacity. The time of immersion was from 4 hours to 21 hours. A total of eleven experiments was conducted with 160 oysters. On immersion in a particular concentration, the tentacular movements of the polychaetes were observed. After counting the number of polychaetes in each oyster, they were transferred to normal sea water and kept in the laboratory. The oysters were examined for results after two days.

Immersion in fresh water

Two hundred oysters affected by the borers were selected and were immersed in fresh water. The immersion time ranged from 1 hour 15 minutes to 10 hours. Later they were transferred to normal sea water. The oysters were kept in the laboratory for a week for obervations.

Observations

The spionid polychaete worms *Polydora ciliata* Johnston and *Polydora* sp. and the clionid sponge *Cliona celata* and *Cliona vastifica* were the main borers on the pearl oyster *P. fucata* from the farm as well as the natural beds. Other boring polychaetes recorded were of the families Sylidae, Nereidae, Terebellidae and Cirratulidae. The pholad mollusc *Martesia* sp. made large holes in the oyster shells. The isopod *Sphaeroma* made shallow grooves superficially on the oyster shells.

In 1978, 205 oysters from the pearl banks Devi Paar, Kodamuthu Paar, Vaipar Periya Paar and Tholayiram Paar were examined. *Polydora* infestation ranged from 1-16 worms per 10 oysters. Of the 290 oysters examined during January to April, 1979, 64 (22.1%) were affected with an average of 4 worms per 10 oysters for the whole period.

In 1978, a total of 606 oysters were examined from the natural beds for sponge 6 oysters (0.99%) showed sponge boring. In the year 1979, out of 630 oysters, 0.95% were infested by sponge.

The data on the percentage of oysters affected by boring by *Polydora* and *Cliona* and the worm load on oysters from the farms at Veppalodai and Tuticorin Harbour are presented in Fig. 1. *P. ciliata* and *Cirratulus cirratus* caused considerable damage to the pearl oyster shells (Plate I).

Experimental Results on Control of Boring Organisms

Treatment with 1% formalin

The exposure for 15 minutes after brushing with 1% formalin was found effective in killing the sponges and *Martesia* sp. completely and *Polydora* suffered a mortality of 87.7% (Table 1).

Immersion in brine solution

In a salt solution of 78‰ all the polychaete worms were dead and the minimum time required was 7 hours and 40 minutes. In the lower concentrations the mortality of the oysters increased when the duration of immersion of oysters was prolonged (Table 1).

size range of 40 to 55 mm in DVM during August, 1979. 573 oysters were treated with fresh water and 148 oysters were brushed with 1% formalin. In the next month when examined 0.1% of the oysters were dead. In October and November 2.3% and 0.8% mortality was noted. The empty oyster shells were

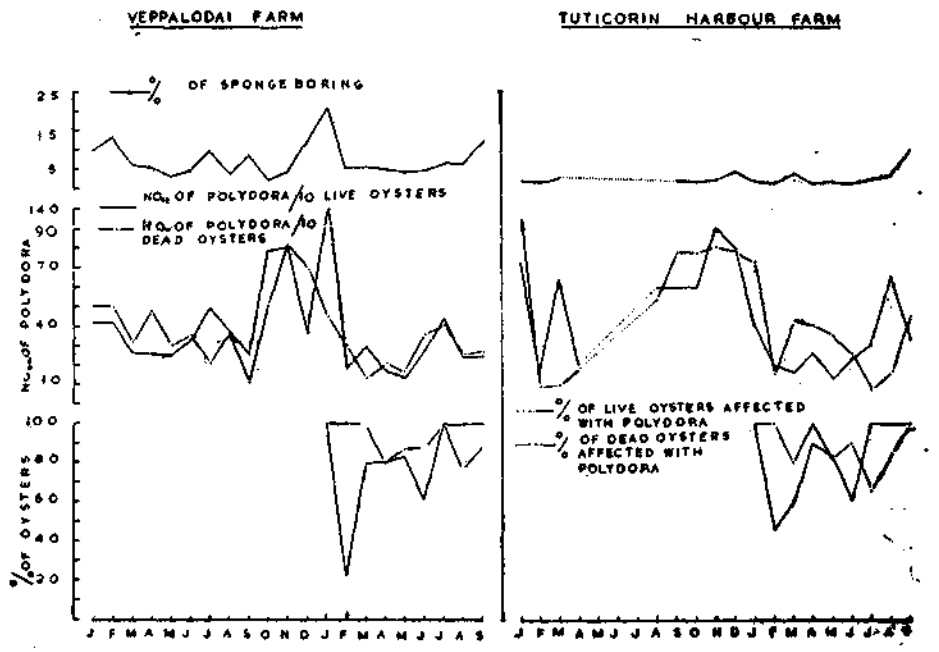

Fig. 1. Polychaete (*Polydora* sp.) and sponge boring on pearl oysters of Veppalodai and Harbour farms.

Immersion in fresh water

The oysters immersed in tap water for 1 hour and 15 minutes to 10 hours were all in good condition (Table 1). In about 6 to 10 hours all the *Polydora* as well as *Cirratulus* were killed. Feeble and decaying worms could be seen in between the shell layers and inside the blisters after four hours of immersion. Worms found on the peripheral sides of the shells and in small blisters were affected more than others by this treatment.

Application of control measures on farm oysters

A total of 721 oysters were selected for the control of borers from Harbour farm with the

examined and were without worms in the case of fresh water treated oysters. The incidence of boring by sponges ranged from 1.4 to 4.3% in the fresh water treated oysters. In the formalin treated oysters no fresh sponge attack was noticed. In both cases, the oysters were found in good condition as other healthy oysters in the farm.

Discussion

Nelson and Stauber (1940) recorded that *Polydora lingi* copiously secreted strands of thick mucus in the form of net work, which caught sediment, detritus, oyster faeces and

TABLE 1. *Experiments on the control of boring organisms on pearl oysters*

No. of oysters used	Duration Hrs	Duration Min	Treated with	Mortality of oysters (percentage)	Percentage of mortality/removal of *Polydora*	*Syllis*	*Terebellid*	*Martesia*	Cirratulid	Sponge
4	2	0	1% Formalin	100	100	X	X	X	X	X
10	1	0	Do.	100	100	X	100	X	X	100
10	0	30	Do.	10	100	X	X	100	X	100
85	0	30	Do.	12.5	46.42	X	X	100	X	X
30	0	30	Do.	10	X	X	X	X	X	100
56	0	15	Do.	Nil	87.71	X	X	X	X	X
30	0	15	Do.	Nil	X	X	X	100	X	100
225										
			Brine Solution							
18	19	0	42.3‰	Nil	100	X	X	X	100	X
25	21	0	42.3 ,,	Nil	100	100	X	X	X	X
10	9	0	55.0 ,,	Nil	100	X	X	X	X	X
10	9	30	55.0 ,,	Nil	60	X	X	X	X	X
37	4	0	60.0 ,,	Nil	90.6	X	X	X	X	X
10	8	30	60.0 ,,	Nil	100	X	X	X	X	X
10	8	15	60.0 ,,	Nil	100	X	X	X	X	X
10	8	0	71.6 ,,	Nil	50	X	X	X	X	X
10	8	0	71.6 ,,	Nil	100	X	X	X	X	X
10	7	40	78.0 ,,	Nil	100	X	X	X	X	X
10	7	50	78.0 ,,	Nil	100	X	X	X	X	X
160										
			Fresh water							
86	1	15	,,	Nil	34	X	X	X	X	X
97	3	30	,,	Nil	57.07	X	X	X	X	X
4	4	00	,,	Nil	75	X	X	X	100	X
4	6	00	,,	Nil	100	X	X	X	100	X
5	8	00	,,	Nil	100	X	X	X	100	X
4	10	00	,,	Nil	100	X	X	X	100	X
200										

X — Not present before and after the experiment.

rejected material. The decomposition of the mass covering the oysters was said to have produced so much hydrogen sulphide that it finally led in early spring to an oyster mortality sweeping over acres of Delaware beds. Korringa (1952) found in the South Carolina waters that about 54% of the edible oysters below the low water mark had mud blisters, while only 20% of the oysters above the low water mark were infected. The percentage of blisters change with the size of the oysters. It was noticed that the pearl oysters of 40 mm in DVM and less in size from the farms and natural beds had few instances of boring by *Polydora* and sponge. Mohammad (1972) indicated that a total of 4.68% of *Pinctada margaritifera* were infected with *Polydora vulgaris* in Kuwait, Arabian Gulf. The infestation was higher in older oysters (14.77%), in those with pearls (19.43%) and highest in old pearl carriers (41.2%), Mizumoto (1964) noted 7 species of polychaetes, of which *Polydora ciliata* formed 88% single and 11.4% double wormed blisters in the pearl oyster (*P. martensii*). Herdman (1905) found from the South Cheval Paar (Sri Lanka Pearl Banks) that more than 75% of the oysters were affected by *Cliona margaritiferae*. Alagarswami and Chellam (1976) observed from Veppalodai farm that a total of 93 shells (20.07%), out of 450 shells examined, were infected with the boring sponge *Cliona celata*. In January 1979, 21.05% of oysters were affected by boring sponge in the Veppalodai farm.

In Holland, a satisfactory treatment against *Polydora* was to expose the oysters either for 16 hours to fresh water or 3 hours to 0.5‰ solution of ammonium salt of dinitro orthocresol in sea water (Korringa, 1951). In the Australian waters, the *Polydora* infection in the edible oysters was checked by exposing the oysters to air and sun when the worms were quickly killed (Yonge, 1960). It was noticed in the present experiment that fresh water treatment for about 6 to 8 hours killed all the polychaete worms, *Martesia* sp. and other organisms found on pearl oysters. Funakoshi (1964) and Shirai (1970) reported the use of saturated salt solution to kill polychaetes from the pearl oysters. Immersion of the affected oysters in a saturated solution of ordinary table salt for 40 minutes was hundred per cent effective in destroying the worms. The experiments using different salinities revealed that when the concentration of the brine was lowered the time required for killing the worms was more and mortality of the oysters also set in. Since the usage of fresh water to kill the borers was cheaper and safe this process was successfully applied to the affected oysters of the harbour farm. Brushing the affected parts of the shells with 1% formalin exposing for 15 minutes to air, washing in fresh water and returning to the farm was found to be very effective.

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ECOLOGICAL CONDITIONS OF THE PEARL CULTURE FARM AT VEPPALODAI IN THE GULF OF MANNAR

A. C. C. VICTOR*

Central Marine Fisheries Research Institute, Cochin-682 018

ABSTRACT

The seasonal changes in atmospheric temperature, surface water temperature, salinity, dissolved oxygen, pH, turbidity and silt deposition at and in the vicinity of the pearl culture farm located off Veppalodai in the Gulf of Mannar were studied during the period from January, 1974 to December 1978. The surface temperature exhibited a clear double oscillation every year and the atmospheric temperature which was invariably higher, also registered two maxima and two minima. The salinity was high during the period of the south-west monsoon and low during the north-east monsoon. There was not much variation in pH values and dissolved oxygen content. The water was turbid during most part of the year, with higher dissolved oxygen content. Silt deposition was observed to be high during December. The values of primary production were high in September.

INTRODUCTION

FOR the survival and optimum growth of pearl oysters, the ecological conditions should be most conducive, inasmuch as growth is strongly influenced by the chemical substances and trace elements present in the sea water and the quality and quantity of the plankton. Matsui (1958) emphasized this fact and pointed out that the resultant variations in the width of pearl oyster shells influence the size, colour and lustre of the cultured pearl. Besides, spawning of the oyster and settlement of spat depend greatly on the quality of the water and the currents prevailing from time to time. Alagarswami (1970) has drawn attention to the necessity of hanging pearl oyster cages at such depths where optimum temperature conditions prevail and also the desirability of shifting the depth of suspension when the hydrographical conditions change from season to season. Also a moderate current is found necessary not only as a source of oxygen but also to bring in planktonic organisms on which the pearl oyster feeds.

Observations on the physico-chemical conditions of the pearl and chank beds in the Gulf of Mannar were studied earlier by Pillai (1962 a, b) and Chandraseharan *et al.* (1967). Prasad and Nair (1963) have reported that the waters off Tuticorin at 10 metres was found to be very productive. As an integral part of the pearl culture experiments off Veppalodai in the Gulf of Mannar, the basic ecological conditions prevailing in the locality were studied. Since pearl production has been successful at Veppalodai (Alagarswami, 1974), a knowledge of the environmental conditions of the area would be useful in extending the operations to other areas.

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* Present adress: Tuticorin Research Centre, Central Marine Fisheries Research Institute, Tuticorin, Tamil Nadu.

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Material and Methods

The pearl culture farm is located in the Gulf of Mannar off Veppalodai (08°57' N and 78°14'E) about 25 km north of Tuticorin. Observations were made at three stations — the farm (station 1), near the shore (station 2) and near Karaichalli Island (station 3) as shown in Fig. 1. The depth range at these stations are 3.75-4.25 m, 2.50-3.00 m and 4.25-4.75 m respectively. Sea water samples were collected once a week from each station between 0830 and 0930 hours. The atmospheric and surface water temperatures were recorded. Water samples were collected for the estimation of salinity and dissolved oxygen by standard titration methods. Hydrogen-ion concentration was determined with a pH meter. The transparency of the water was measured using a secchi disc. Silt deposition was estimated by suspending a 270 ml capacity glass bottle with a diameter of 2.54 cm at the mouth, at a depth of two meters. Gaarder and Gran (1927) light and dark bottle oxygen technique was employed to estimate the primary productivity. Sets of light and dark glass bottles of 125 ml capacity were filled with sea water collected from the Veppalodai pier and suspended in the same area. The incubation period lasted for 12 hours from 0600 hours to 1800 hours. The initial and final oxygen content of both the bottles were determined by Winkler method. It was observed that the temperature, salinity, dissolved oxygen and pH of the surface waters of all the three stations did not show any marked variations and hence, the data collected from the stations were pooled together. Data on rainfall were obtained from the records of the Port Trust, Tuticorin. The observations relate to a period of five years from 1974 to 1978.

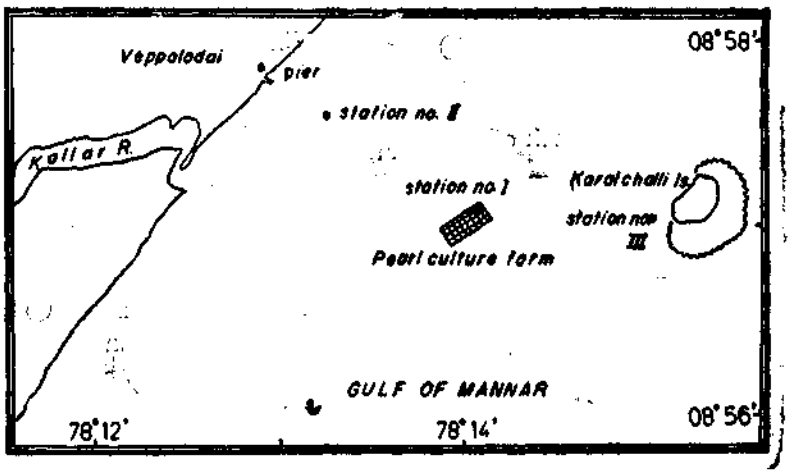

Fig. 1. Veppalodai pearl culture farm in the Gulf of Mannar and sampling stations.

Observations

Atmospheric temperature

The monthly mean atmospheric temperature varied between 25.2°C (January) and 31.6°C (March) in 1974, 26.1°C (December) and 30.9°C

(May) in 1975, 27.2°C (December) and 31.6°C (May) in 1976, 25.8°C (January) and 31.4°C (April-May) in 1977 and 26.2°C (December) and 31.3°C (May) in 1978 (Fig. 2). Except in 1978, two distinct temperature maxima were observed in each year — March (31.6°C) and July (30.9°C) in 1974, May (30.9°C) and October (30.3°C) in 1975, May (31.6°C) and August-September (31.6°C) in 1976 and April-May (31.4°C) and September (30.5°C) in 1977. In 1978 only one temperature maximum was observed in May (31.3°C). In contrast, only one temperature minimum was observed in each year, January (25.2°C) in 1974, December (26.1°C) in 1975, December (27.2°C) in 1976, January (25.8°C) in 1977 and December (26.2°C) in 1978. Judging from the general trend of atmospheric temperature, there appears to be a strong maximum during April-May, a weak minimum during June-July, a weak maximum during September-October and a strong minimum during December-January. In 1974, the cyclic variations are advanced by one or two months. In 1978, the weak maximum and minimum were not observed.

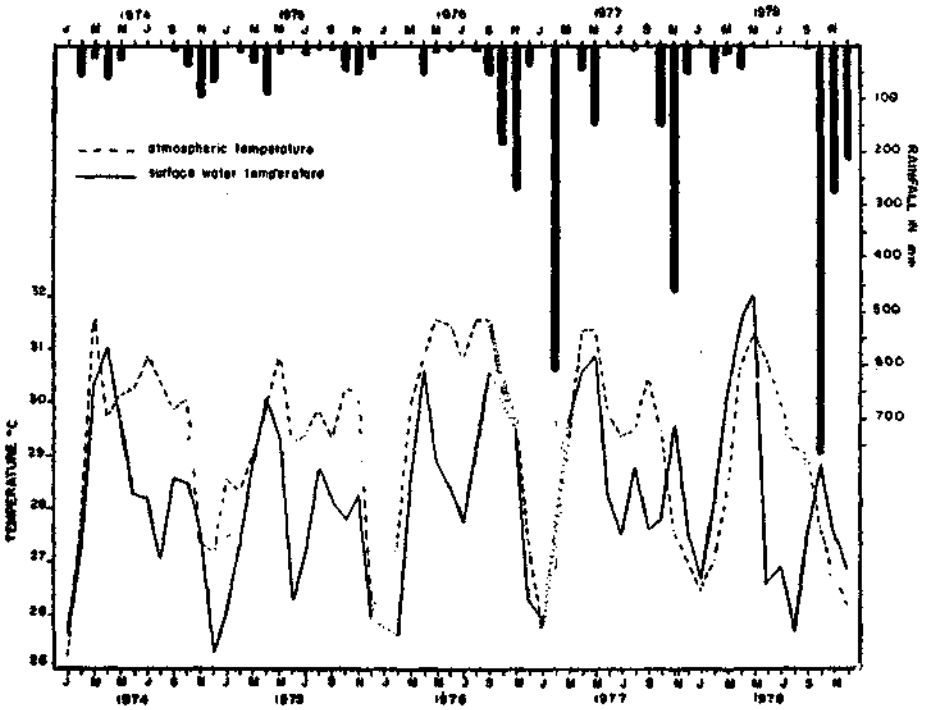

FIG. 2. Seasonal variations in atmospheric temperature, surface water temperature and rainfall (Months in which no observations were made are shown in dotted lines).

Surface water temperature

The monthly mean surface water temperature varied between 25.5°C (December) and 31.1°C (April) in 1974 ; 26.0°C (December) and 30.1°C (April) in 1975, 25.6°C (February) and 30.6°C (April) in 1976 ; 25.9°C (January) and 30.9°C

(May) in 1977 and 25.7°C (August) and 32.1°C (May) in 1978 (Fig. 2). Two temperature maxima were observed in each year — April (31.1°C) and September (28.6°C) in 1974, April (30.1°C) and November (28.3°C) in 1975, April (30.6°C) and September (30.6°C) in 1976. May (30.9°C) and November (29.6°C) in 1977 and May (32.1°C) and October (28.8°C) in 1978. The two minima were observed in August (27.1°C) and December (25.5°C) in 1974, June (26.3°C) and December (26.0°C) in 1975, July (27.7°C) and December (26.3°C) in 1976, July (27.5°C) and December (27.6°C) in 1977 and August (25.7°C) and December (26.9°C) in 1978. Data are not available for the month of January 1976. However, February 1976, recorded lower temperature as compared to the corresponding month of preceding years.

Rainfall

The annual rainfall was 368 mm, 282 mm, 615 mm, 1489 mm and 1370 mm during 1974 through 1978 respectively. The maximum monthly rainfall was 95.6 mm in November 1974, 50.6 mm in November 1975, 269.9 mm in November 1976, 616.4 mm in February 1977 and 770.0 mm in October 1978 (Fig. 2). The period June-August is the period of least or no rainfall. September-May period is the rainy period with main peak in October-November. A secondary peak of rainfall is noticeable during February-April also. The air and sea temperatures appear to be correlated with rainfall. The air temperature is as low as that of the surface temperature during peak rainfall periods. During non-rainy or least rainy periods in June-July the air temperature is much higher than that of sea surface.

Salinity

The monthly mean salinity values varied from 32.15 ‰ (January) to 35.26 ‰ (August) in 1974, 32.38 ‰ (December) to 35.13 ‰ (August) in 1975, 32.79 ‰ (February) to 35.58 ‰ (July) in 1976, 28.48 ‰ (November) to 34.78 ‰ (July) in 1977 and 28.95 ‰ (January) to 34.28 ‰ (July) in 1978 (Fig. 3). In contrast with the temperature, the salinity showed a single peak in July/August preceded by a gradual rise from January and followed by a gradual decline to December. The peak salinity period corresponds with the no rainfall period. Salinity becomes the lowest during January/February, followed by peak rainfall in October-November. But such a decline in salinity is not noticeable after the secondary rainfall peak in February-April.

Dissolved oxygen

The monthly mean dissolved oxygen values ranged from 4.6 ml/l (November) to 5.6 ml/l (January) in 1974, 4.8 ml/l (December) to 5.9 ml/l (April) in 1975, 4.4 ml/l (April) to 6.0 ml/l (March) in 1976, 4.4 ml/l (September) to 6.0 ml/l (May) in 1977 and 4.4 ml/l (November to 6.4 ml/l (August) in 1978 (Fig. 3).

pH

The pH values were recorded from January 1974 to November 1975. The monthly mean pH values varied from 7.96 (February) to 8.42 (November) in 1974 and 8.03 (June) to 8.29 (February) in 1975 (Fig. 3).

Water transparency

The monthly mean transparency of water varied from 0.69 m (May) to 1.73 m (September) during 1974, 1.12 m (September) to 2.60 m (November) during 1975 and 1.44 m (August) to 2.29 m (July) during 1976 (Fig. 3).

Silt

Data were collected from October 1975 to April 1976 only. The average rate of silt deposition per day was calculated. The maximum of 3.13 ml/day was recorded in the month of December 1975 and the minimum 0.87 ml/day during March 1976.

Primary production

Table 1 illustrates the mean monthly values of gross and net productivity expressed as mg C/m³/12 hours day for the year 1978. The mean monthly values of gross production varied between 195.8 mg C/m³/12 hours day (March) and 377.0 mg C/m³/12 hours day (September) in 1978. There are two peaks of production one in May (345.2 mg C/m³/12 hours day) and another in September (377.0 mg C/m³/12 hours day). The two depressions are one in March (195.8 mg C/m³/12 hours day) and the other in October (211.9 mg C/m³/12 hours day).

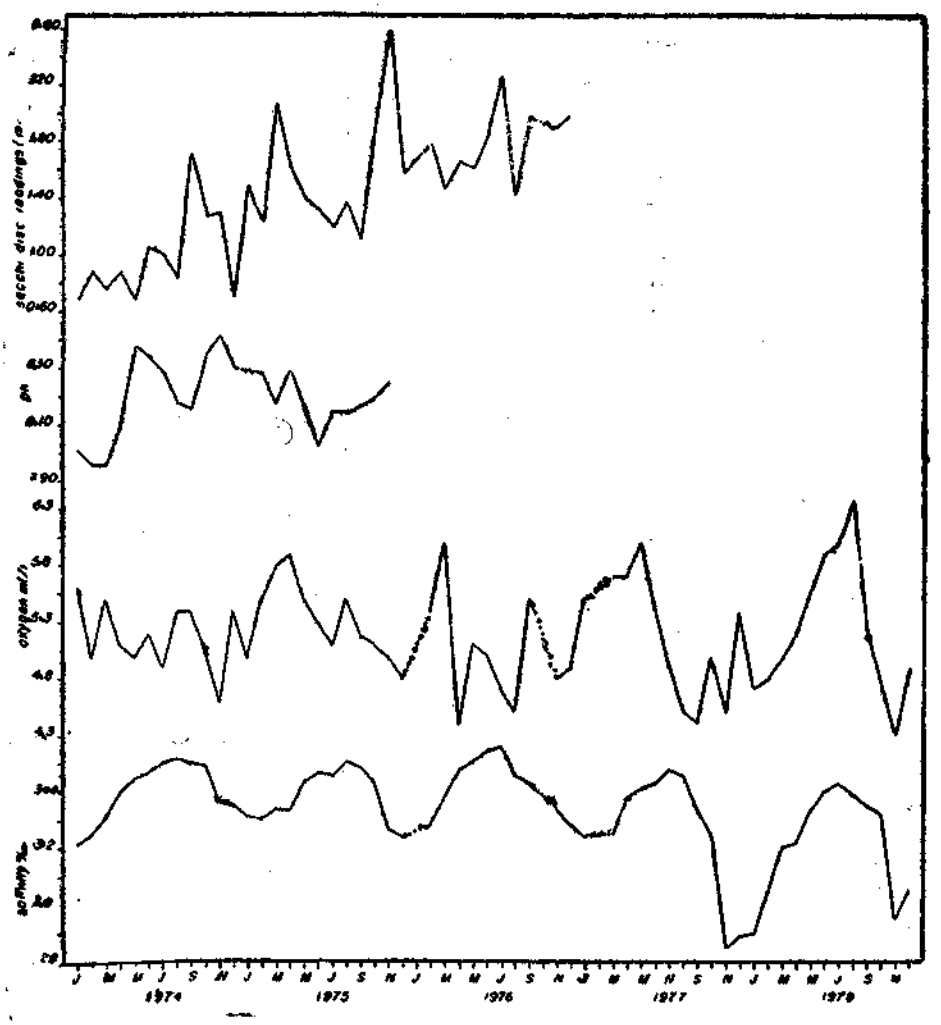

FIG. 3. Seasonal variations of salinity, dissolved oxygen, pH and clarity (Months in which no observations were made are shown in dotted lines).

TABLE 1. *Values of mean monthly primary productivity and Respiration at Veppalodai Pearl Culture Farm during 1978*

Month	Primary productivity (mg C/m³/12 h day)		Respiration (mg C/m³ 12 hrs)	% of gross
	Gross	Net		
January ..	278.5	181.6	96.9	34.8
February ..	202.8	96.9	105.9	52.2
March ..	195.8	109.0	85.9	44.3
April ..	278.6	181.7	96.9	34.8
May ..	345.2	212.0	133.2	38.6
June ..	295.6	167.2	128.4	43.4
July ..	369.5	243.8	125.7	34.0
August ..	355.8	286.2	69.6	19.6
September ..	377.0	255.6	121.4	32.2
October ..	211.9	149.9	62.1	29.3
November ..	294.8	244.3	50.5	17.1
December ..	339.2	269.9	70.3	20.7

DISCUSSION

The fluctuations in the monthly mean atmospheric temperature followed the same pattern during the five years of study from 1974 to 1978. In general the atmospheric temperature exhibited three distinct periods of change; rising temperature from January to May, high temperature from June to October and decreasing temperature from early November to late December. The decrease in temperature to the lowest levels in December is due to the effect of an active north-east monsoon. The monthly mean surface temperature also exhibited a similar trend; temperature rise from January to April, temperature fluctuations during June-October with minimum during July-August and temperature decrease from early November to late December. During July-August the strong winds associated with the south-west monsoon caused a high rate of evaporation resulting in the lowering of temperature (Prasad, 1957). The steep fall in the surface temperature during November-December was primarily due to the onset of the rain-bearing north-east monsoon. In all the five years of study the coincidence between atmospheric and the surface water temperature was not continuous. Except in January and April 1974, January, March, November and December 1977, January to May and October to December 1978, the atmospheric temperature was found to be higher than the surface water temperature. Prasad (1957) observed that the average atmospheric temperatures were above the mean surface temperatures mostly in the months of May, June and July at Mandapam and that the atmospheric temperature was never above the surface temperature continuously for more than four months.

The variations in salinity followed the same pattern during the five consecutive years of study. Unlike temperature, salinity appears to be monocyclic in its annual variation. High salinity values prevailed over a period of about six months from May to October and the low salinity values from November to April in 1970. The period of very high values and very low values coincide with south-west monsoon and north-east monsoon respectively. Jayaraman (1954) recorded high salinity values from

November to April in the Gulf of Mannar and Palk Bay which is in agreement with the present observations. During the present study, the highest individual salinity value of 36.68‰ was recorded on June 29, 1976. The highest salinity value recorded by Chandraseharan *et al.* (1967) in the pearl and chank beds off Tuticorin was 36.12‰ on September 1, 1961. The lowest individual salinity value obtained during the present study was 9.27‰ on November 25, 1978. Chandraseharan *et al.* (1967) recorded a low salinity of 29.13‰ on December 5, 1962 and Chacko and Sambandamurthy (1969) recorded the lowest value of 11.2‰ during 1962-63 on the pearl banks of the Gulf of Mannar. The effects of heavy rainfall in the coastal and interior areas and the influx of fresh water into the sea from the numerous rivers and rivulets including the major river Tambraparni and the two small rivers Kallar and Vaippar in the vicinity of Veppalodai considerably lowers the salinity. Alagarswami and Victor (1977) have shown that salinity conditions reached the low level of 15.59‰ on November 8, 1977 in the Veppalodai pearl culture farm. However, there was no mortality of pearl oysters mainly because the low saline conditions prevailed only for a short while.

There is no perceptible seasonal variation in dissolved oxygen. However, during certain months the values were pronouncedly high as well as low. The range of 4.4 - 6.4 ml/l observed in the present study area agrees with the observations of Jayaraman (1954) on the surface waters off Mandapam. The hydrogen ion concentration ranged from 7.96 to 8.42 and there was no marked seasonal variation.

The south-west and north-east monsoons cause the presence of large quantities of suspended matter in the water. The monthly mean transparency of water was never high, and the values ranged from 0.69 m to 2.60 m in the depth range of 2.50 - 4.75 m. Turbidity and dissolved oxygen showed good correspondency; more turbid the water, more was the oxygen content. The rate of silt deposition per day varied from 0.87 ml (March 1976) to 3.13 ml (December 1975). The highest silt deposition observed in the month of December 1975 coincides with low salinity and temperature conditions.

There were two peaks of primary production one in May (345.2 mg C/m^3/12 hours day) and another in September (377.0 mg C/m^3/12 hour day). Prasad and Nair (1963) reported that the waters off Tuticorin were remarkably productive and showed a rate of 252.6 mg C/m^3/day at 10 metres. The range of 195.8 - 377.0 mg C/m^3/ 12 hour day observed in the present study area is relatively higher than the above values.

The present study deals with only a few ecological parameters of the pearl culture farm. Investigations on other factors such as nutrients and energy content of suspended organic matter which are relevant have been started only recently. Compared to the environmental condirions of pearl culture farms in Japan (Matsui, t1958; Alagarswami, 1970) and Australia (Hancock, 1973) those at the Veppalodai farm are far different in regard to at least three factors — protection from winds and waves, depth and turbidity — and the latter would seem apparently unfavourable. The difference is so with regard to the conditions of the natural pearl oyster beds in the Gulf of Mannar. However, the initial successful development of pearl culture technology at Veppalodai for producing high percentage of good quality cultured pearls would show that shallow open coastal areas may be considered for establishing pearl culture farms in the absence of more ideal sites.

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OXYGEN CONSUMPTION IN PEARL OYSTER *PINCTADA FUCATA* (GOULD) AND *PINCTADA SUGILLATA* (REEVE)

S. DHARMARAJ*

Central Marine Fisheries Research Institute, Cochin-682 018

ABSTRACT

The rate of oxygen consumption of pearl oysters *Pinctada fucata* and *P. sugillata* from different localities namely pearl culture farm, pearl banks and near-shore waters of Tuticorin in the Gulf of Mannar has been studied. In *P. fucata* oxygen consumption was 1339 µl/hr. for oyster size range 40-50 mm; 1650 µl/hr for 50-60 mm and 1810 µl/hr for 60-70 mm. The rate of oxygen consumption of *P. sugillata* from the pearl banks as well as from the near-shore waters showed a linear relation with size of oyster. The rate of oxygen consumption of *P. sugillata* from pearl banks (depth 12-21 m) was less when compared to that in the oysters of the near-shore waters (depth 0.5-1.5 m). *P. fucata* from pearl culture farm survived after 21 hours of exposure to air. *P. fucata* from pearl culture farm could survive upto 19 hours, upto 24 hours in the case of *P. sugillata* from near-shore waters and upto 27 hours in *P. sugillata* from pearl banks, in anaerobic conditions. In all the oysters there was remarkable increase in shell activity and shell gape when there was a decline in the oxygen level.

INTRODUCTION

PEARL OYSTERS occur in the intertidal region of Gulf of Kutch and at a depth of 12-21 m in the pearl banks of Gulf of Mannar. The settlement of pearl oysters on the granite stones at Tuticorin Harbour at a depth of 0.5-1.5 m was first recorded by Nayar *et al.* (1978). The physiology of respiration of bivalves such as *Cerastoderma edule*, *C. glaucum* (Boyden, 1972), *Arctica islandica* (Taylor, 1976), *Donax cuneatus*, *D. faba* (Rao and Kutty, 1968), *Gryphaea virginica* (Korringa, 1952) and the pearl oyster *Pinctada fucata* (Uemoto, 1968; Itoh, 1976) have been studied in detail. Cahn (1949) reported the role of oxygen during conditioning of the pearl oyster *P. martensii*. Okawa (1959) studied the oxygen consumption in relation to the feeding of *P. martensii*. The present paper deals with the oxygen consumption of the Indian pearl oysters *Pinctada fucata* and *P. sugillata*, their survival in anaerobic conditions and exposure to air. These findings have practical utility in conditioning and transporting of oysters.

I express my deep sense of gratitude to Dr. E. G. Silas, Director, Central Marine Fisheries Research Institute, Cochin for his encouragement and to Dr. K. Alagarswami of the Institute for his guidance and offering valuable suggestions. My thanks are also due to Shri K. Nagappan Nayar for extending all facilities and to Dr. K. Alagaraja, Central Marine Fisheries Research Institute, Cochin, Dr. M. Narayanan Kutty and Dr. N. Sukumaran, Fisheries College, Tuticorin for their helpful suggestions in the analysis of the data.

MATERIAL AND METHODS

Pinctada fucata were collected from the pear banks at a depth of 12 m and reared in the pear culture farm in Tuticorin Harbour. *Pinctada sugillata* were obtained from the same area and also from the near-shore waters of Tuticorin Harbour at a depth of 1 m.

* Present address: Tuticorin Research Centre of the Central Marine Fisheries Research Institute, Tuticorin, Tamil Nadu.

Twenty five oysters of *P.fucata* and 35 oysters of *P. sugillata* were used in this study. *P. fucata* from the farm ranged from 47 to 60 mm in dorsoventral measurement (DVM) (2.3-4.4 g in wet weight), *P. sugillata* from pearl banks varied between 10.0 and 66.3 mm in DVM (0.01-5.5 g in wet weight) and *P. sugillata* from near-shore waters ranged from 22.5 to 57.4 mm in DVM (0.09-3.9 g in wet weight). The rate of oxygen consumption was estimated by Winkler's method and expressed in terms of μl of O_2/hour. The 'b' values were determined by using the equation,

$$Y = a + bX.$$

During the experiment, initial conditioning time, shell activity, shell gape and behaviour of oysters were also recorded.

P.sugillata from pearl banks (21.8-54.0 mm DVM) and from near-shore waters (36.0 — 71.4 mm DVM), and *P.fucata* from pearl culture farm (30.0 — 54.5 mm DVM) were used to study the survival rate in anaerobiosis. The oysters were kept in anaerobic medium for different durations of 6, 9, 12, ... and 33 hours. Instead of releasing the oysters directly to oxygen-free water, they were allowed to respire in a medium with known oxygen level. The level of oxygen in the medium was estimated at intervals and the time at which the medium contains zero oxygen was noted. The survival rate was then calculated from this level.

RESULTS

Rate of oxygen consumption

Pinctada sugillata of similar size groups collected from the two ecologically varying

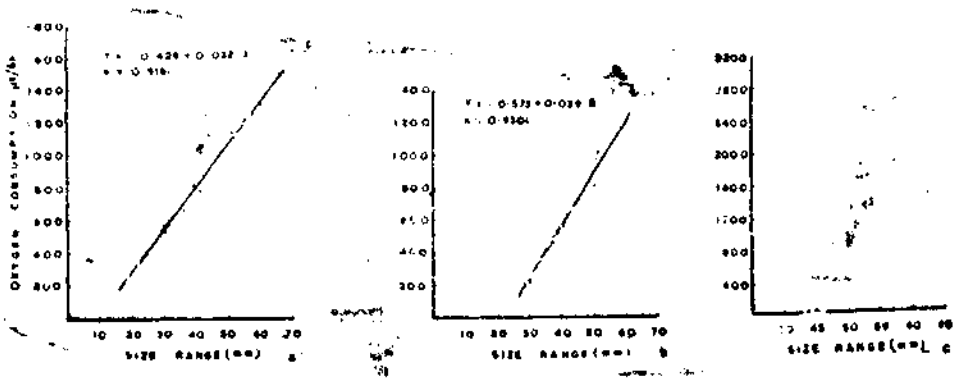

Fig. 1. Oxygen consumption in : a. *Pinctaaa sugillata* from near shore waters (depth 1.0 m) at Tuticorin Harbour basin, b. *Pinctada sugillata* from pearl banks in the Gulf of Mannar (depth 12 m) and c. *Pinctada fucata* from the pearl culture farm at Tuticorin Harbour (depth 3 m).

P. fucata tested for oxygen consumption were kept out of water for different durations of 6, 9, 12, 15, 18, 21, 24 and 30 hours and the post-exposure rate of oxygen consumption was estimated based on wet tissue weight in varying temperature from 26.0°C to 29.4°C (Mean 27.9°C).

conditions held their characteristic metabolic rate and indicated linear trend in both the cases with high positive correlation (Fig. 1 a, b). The rates of oxygen consumption of different size groups of *P.sugillata* from pearl banks and from near-shore waters and *P. fucata* from pearl culture farm are given in Table 1.

TABLE 1. *Oxygen consumption (µl/hour) of* Pinctada sugillata *and* Pinctada fucata

Species & locality	Size Groups (mm)					
	10-20	20-30	30-40	40-50	50-60	60-70
Pinctada sugillata						
From pearl banks	..	255	345	588	1045	..
From nears hore waters	618	828	510	879	1170	1361
Pinctada fucata						
From pearl culture farm	..	..	..	1339	1650	1810

In the case of *P. fucata* linear trend cannot be seen because of the smaller size groups which were experimented (Fig. 1 c).

Rate of oxygen consumption prior to and after aerial exposure

P. fucata exposed to 9, 12 and 18 hours showed higher rate of oxygen consumption during the first hour on reimmersion and became normal from second hour (Fig. 2 b, c, d). *P. fucata* exposed to 6 and 21 hours showed lesser rate of consumption during the first hour on reimmersion (Fig. 2 a, e).

Survival rate after exposure to air

Pinctada fucata kept out of water for 21 hours were experimented for determining the post-exposure rate of oxygen consumption. 100% survival of these oysters could be recorded after this treatment. The oysters exposed to 24 hours were also treated in the same manner. They exhibited a few shell activities on reimmersion to sea water and then died.

Tolerance limit of pearl oysters in anaerobiosis

P. fucata was found to be less tolerant to anaerobic conditions than *P. sugillata*. Mortality of *P. fucata* in anaerobic medium set early from 19th hour. In the case of *P. sugillata* from near-shore waters the mortality began from 24th hour. *P. sugillata* from the pearl banks were found to tolerate upto 27 hours in anaerobiosis.

Behaviour in air

During exposure to air *Pinctada fucata* were kept in a tray with right valve down. After 6 hours of exposure, though the valves were closed, shell activity was noted intermittently. After 9 hours the valves showed partial gaping and after 12 hours the valves widely separated and the mantle edges withdrew from the shell edges. Voluntary shell movement was observed upto 18 hours of exposure and it ceased on further exposure. The measure of shell gape was found proportional to the duration of exposure.

Behaviour on return to sea water

The oysters exposed for 6 hours showed normal closing and opening of shell valves on return to sea water. Few oysters, after 18 hours of exposure, exhibited partial closure of valves for 1 to 3 hours and normal shell closure commenced afterwards. The oysters after 21 hours of exposure closed their valves on immersion and opened in a few minutes. In this case partial closure of valves persisted throughout the experiment. Such response ceased beyond 21 hours of exposure.

The mantle which was withdrawn from the shell valve edges after 12 hours of exposure

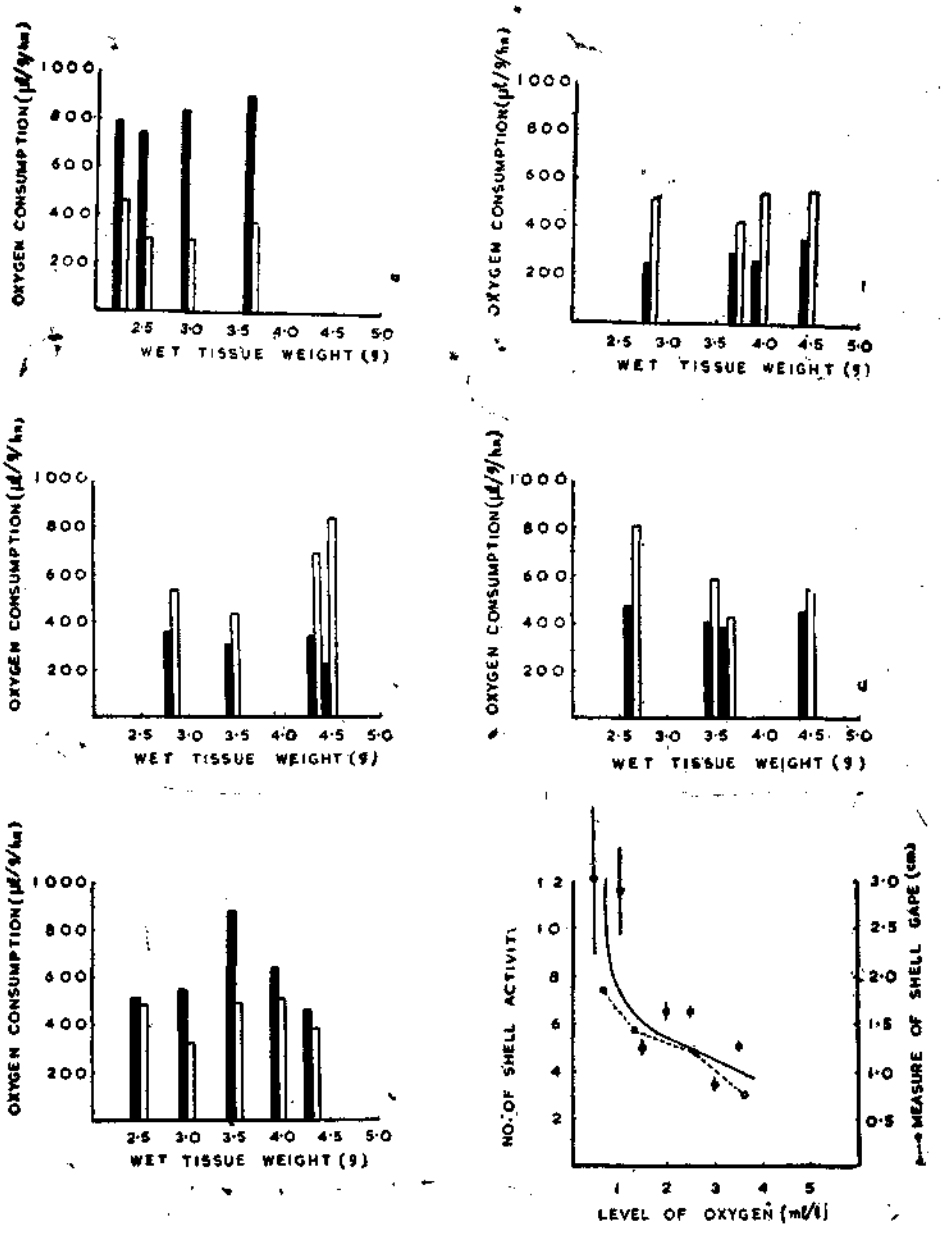

Fig. 2. Pre-(solid block) and post-exposure (hallow block) rate of oxygen consumption in *Pinctada fucata* for the first hour on immers on: a. after 6 hours of exposure, b. after 9 hours of exposure, c. after 12 hours of exposure, d. after 18 hours of exposure, e. after 21 hours of exposure and f. The measure of shell gape (open circles) and shell activity (closed circles) in relation to various level of oxygen concentrations. Each point in shell activity is a mean of 3-8 values (± S.E.).

took 2.45 to 3.30 hours to attain normal position. The same process required 5 hours in the case of oysters exposed to 21 hours. On the contrary the gill on reimmersion came to normal functioning within two hours and the movement of particles were clearly seen over the gills.

Initial conditioning time and shell activity

Initial conditioning time is referred to as the duration between the time of immersion and the first opening shell valves. The conditioning time of the oysters freshly collected from the farm ranged from 2 to 65 minutes whereas the oysters exposed for 9 to 21 hours took hardly 0 to 15 minutes.

The amplitude of shell activity of unexposed oysters was accelerated below ambient oxygen concentration and was maximum at 1 ml/litre of oxygen (Fig. 2 f). Further retention of oysters in hypoxic medium resulted in the complete closure of valves. A minimum shell activity was recorded in normoxic conditions.

Shell gaping

The measure of shell gape was found to vary inversely to the amount of oxygen in the medium. The shell gape over various levels of oxygen is given in Fig. 2 f. At lower oxygen level the shell opened to the maximum extent, thus exposing the gills fully to the medium. The oysters under well-aerated conditions kept their valves open only narrowly.

Discussion

The average dissolved oxygen content of the surface and bottom waters of pearl banks have been recorded as 4.22 and 4.37 ml/l respectively and that of the pearl culture farm in Tuticorin Harbour were 5.05 and 4.77 ml/l respectively. In the present study *P. sugillata* from the pearl banks showed lesser rate of oxygen consumption than those from the nearshore waters. The data reflect on the characteristic low metabolism of the benthic forms.

In *Pinctada fucata* the rate of oxygen consumption of the size groups 40-50 mm was 1339 µl/hour, 50-60 mm 1650 µl/hour and 60-70 mm 1810 µl/hour. Oxygen availability at these rates should be taken into account while conditioning the oysters before nucleus implantation for pearl production.

A batch of 25 *P. fucata* and 24 *P. sugillata* were taken from Tuticorin to Dhauli and brought back without any mortality. During the above transportation by train over a distance of 1896 km and time of 43.25 hours the pearl oysters were intermittently immersed in seawater for a while and covered with wet gunny bag. Korringa (1952) indicated that at low temperatures *Ostrea edulis* can withstand upto 24 days of exposure and complete recovery was possible after 18 days of exposure. The survival time of *Donax cuneatus* exposed to air at room temperature (30°C) was found to be 69 hours and *D. faba* in the same condition survived for 94 hours (Rao and Kutty, 1968). *P. fucata* in the present study, withstood exposure upto 21 hours.

The onset of mortality in *Pinctada fucata* started from 19th hour in anaerobic medium. It is probable that in aqueous medium the metabolic end products might readily be released which in turn caused deleterious effect on the oysters. Korringa (1952) stated that the edible oyster *Gryphaea virginica* excretes measurable quantities of organic acids such as lactic acid which would release CO_2 from the oyster shell thus increasing the level of CO_2 in the medium.

The post-exposure rate of oxygen consumption in *P. fucata* after 6 and 21 hours of exposure was less than the pre-exposure period. During the period of 6 hours of exposure the oysters closed their valves tightly and hence 'Oxygen debt' might not have been incurred. After 21 hours of exposure the indebtedness might have crossed the safe level and, after 3 more hours (24 hours of exposure), cent per cent mortality

sets in. An increase in cardiac and respiratory activity of the bivalve *Arctica islandica* following the periods of shell closure has frequently been interpreted as representing the repayment of 'Oxygen debt' incurred during anaerobiosis (Taylor, 1976).

In *Pinctada fucata* a higher rate of oxygen consumption was recorded during the first hour on reimmersion and normal rate of consumption resumed afterwards. Taylor (1976) reported a similar trend in *Arctica islandica* in which the rate of oxygen consumption on reimmersion was about three times faster than normal and declined to say more or less constant for about 20 hours. Boyden (1972) has opined that the increase in oxygen uptake after exposure reflects increased activity to flush nitrogenous excretory products from the tissues. The initial conditioning time of the exposed oysters revealed an urgency to open their shell valves for the purpose of respiration.

Under hypoxic conditions the oyster tries to obtain the same amount of oxygen normally required. Hence it has been forced to exhibit more shell activity either to replenish the medium or to ventilate the respiratory organs for the supply of adequate oxygen. Similar effect was reported by Taylor (1976) in *Arctica islandica*. He also suggested that the change over to anaerobic metabolism may take place at low oxygen tension but before the medium is completely devoid of oxygen. Various workers have established that bivalves can metabolise anaerobically during the periods of aerial exposure or hypoxia.

Information on the relationship between the shell gaping in pearl oysters and the oxygen tension in the medium has much value in the controlled culture of oysters in the laboratory. The measure of shell gape has been recognised as an indicator of the level of oxygen in the medium and it is inversely proportional to oxygen level.

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PRELIMINARY EXPERIMENTS ON BREEDING OF CEPHALOPODS

D. SIVALINGAM AND S. KRISHNA PILLAI*
Central Marine Fisheries Research Institute, Cochin-682 018

ABSTRACT

The results of preliminary experiments on hatching and rearing of a squid *Sepioteuthis lessoniana* and a cuttle fish *Sepia aculeata* are described. The hatchling survived for a maximum of 10 days.

INTRODUCTION

BIDDER (1950), Wells (1962), Boycott (1965), Fields (1965) and Schroder (1969 a, b) expressed difficulties in maintaining squids alive for appreciably long durations. Attempts to rear young squids captured from sea by trawlers and small set nets in Japan have resulted in high rate of mortality and this method of collection and culture was considered impractical (Bardach *et al.*, 1972). Egg clusters were collected from spawning areas and hatched and reared in laboratories in Japan. This approach also posed problems such as providing right type of food for young ones. Choe and Oshima (1963) reared three species of cuttle-fish *Sepia esculenta*, *S. subaculeata* and *Sepiella maindroni* and two species of squids *Sepioteuthis lessoniana* and *Euprymna beryii* in Japan. La Roe (1971) cultured and maintained *Sepioteuthis sepioidea* and *Doryteuthis plei* from egg stage to sexually mature condition. In India, no work has been done on rearing cephalopods except on the embryonic development of *Sepioteuthis arctipinnis* (= *S. lessoniana*) (Alagarswami, 1966). The present authors made attempts to culture the squid *Sepioteuthis lessoniana* and cuttle fish *Sepia aculeata* at Mandapam along the south-east coast of India based on collections made from coastal areas during March-June, 1979.

The authors are grateful to Dr. E.G. Silas, Director, Central Marine Fisheries Research Institute for his keen interest and encouragement and to Dr. P.S.B.R. James for facilities provided and to Shri P. Bensam for critically going through the paper and for giving valuable suggestions for improvement. The authors are also thankful to Mr. P. Raghavan for taking photographs.

COLLECTION OF EGG CLUSTERS

Rao (1954) has stated that squids begin to spawn from January in offshore waters and migrate inshore where they continue to spawn till the end of July. In the Palk Bay, fishermen capture spawning squids by encircling them by '*Chippi valai*' after placing branches of *Cassia* plant on which the species lay their eggs. Often egg clusters are found washed ashore in Palk Bay and Gulf of Mannar.

Dhargavalasai on Palk Bay appeared to be the best place for collection of squid eggs. Egg clusters of *Sepia aculeata* which are relatively rare were found only on one occasion in the shore-seine attached together with those of the squid.

* *Present address:* Regional Centre of CMFRI, Mandapam Camp, Tamil Nadu.

Description of Egg Capsules

The egg capsules of *S. lessoniana* contained a central gelatinous matrix by which they were attached to marine algae, sea grass or any other submerged objects. From the base, the egg capsules were arranged in radiating pattern. Squids are known to prefer their own egg masses already present in the environment for depositing their subsequent egg capsules (Plate I). The weight of the egg clusters collected in the present investigations varied from 122 g to 1,500 g and the number of egg capsules in a cluster ranged from 26 to 545 (Table 1).

Table 1. *Weight of egg clusters, the number of capsules in each and mean weight of capsules in the case of* Sepioteuthis lessoniana

Weight in g	No. of egg capsules	Mean weight of a capsule (in g)
122	32	3.81
157	32	4.91
200	52	3.85
162	26	6.23
146	66	2.21
880	300	2.93
1500	545	2.75

The number of eggs in a capsule and their range and mean length are listed in Table 2. In the fully developed condition, the movement of embryos were visible through the translucent gelatinous capsule.

Table 2. *Length of egg capsules and number of eggs in* Sepioteuthis lessoniana

Number of egg capsules studied	Number of eggs present	Egg capsule length in mm	
		Range	Mean
13	1	25-37	30
66	2	35-50	38
249	3	35-60	48
131	4	50-73	58
52	5	60-78	66
14	6	79-82	75

The egg capsules of *S. aculeata* were pear shaped (Plate I) and each was found attached to a central gelatinous matrix and contained only one embryo. The embryos were visible through the translucent gelatinous capsule. The number of eggs in a few clusters collected and their length and width are given in Table 3.

Table 3. *Number of eggs, their mean length and width in the case of* Sepia aculeata

Number of capsules in a cluster	Length of capsule in mm		Width of capsule in mm		Average weight of the capsule in g
	Range	Mean	Range	Mean	
13	12-22	19.5	8-16	13.6	1.4
15	16-21	19	11-20	14	1.5
16	17-22	19	12-17	14.3	1.5

Hatching Experiments

The egg clusters of *Sepioteuthis lessoniana* and *Sepia aculeata* collected on 27-3-'79 from the shore seines operated in Dhargavalasai were reared in an aquarium tank. The water temperature ranged from 29°C to 30°C and salinity from 31.05‰ to 32.87‰. The distal eggs were found to develop faster than the basal ones in the strand and hatch out earlier. Hatching was observed on 3rd and 4th April. The newly hatched squids were active and agile and measured 7 to 8 mm. Fresh plankton consisting of copepods and decapod larvae were introduced in the tank as food ; but it was observed that they were not feeding on plankton and the hatchlings survived only for 5 days.

In the case of egg clusters collected on 19-4-'79 most eggs hatched out from 24th to 29th April, 1979. A total of 384 young squids hatched out and all the young ones were reared in a shaded concrete tank. The young ones survived for a total duration of ten days. Later collections of egg capsules on 17-5-'79, 16-6-'79 and 27-6-'79 also gave a hatching percentage of 80 to 100.

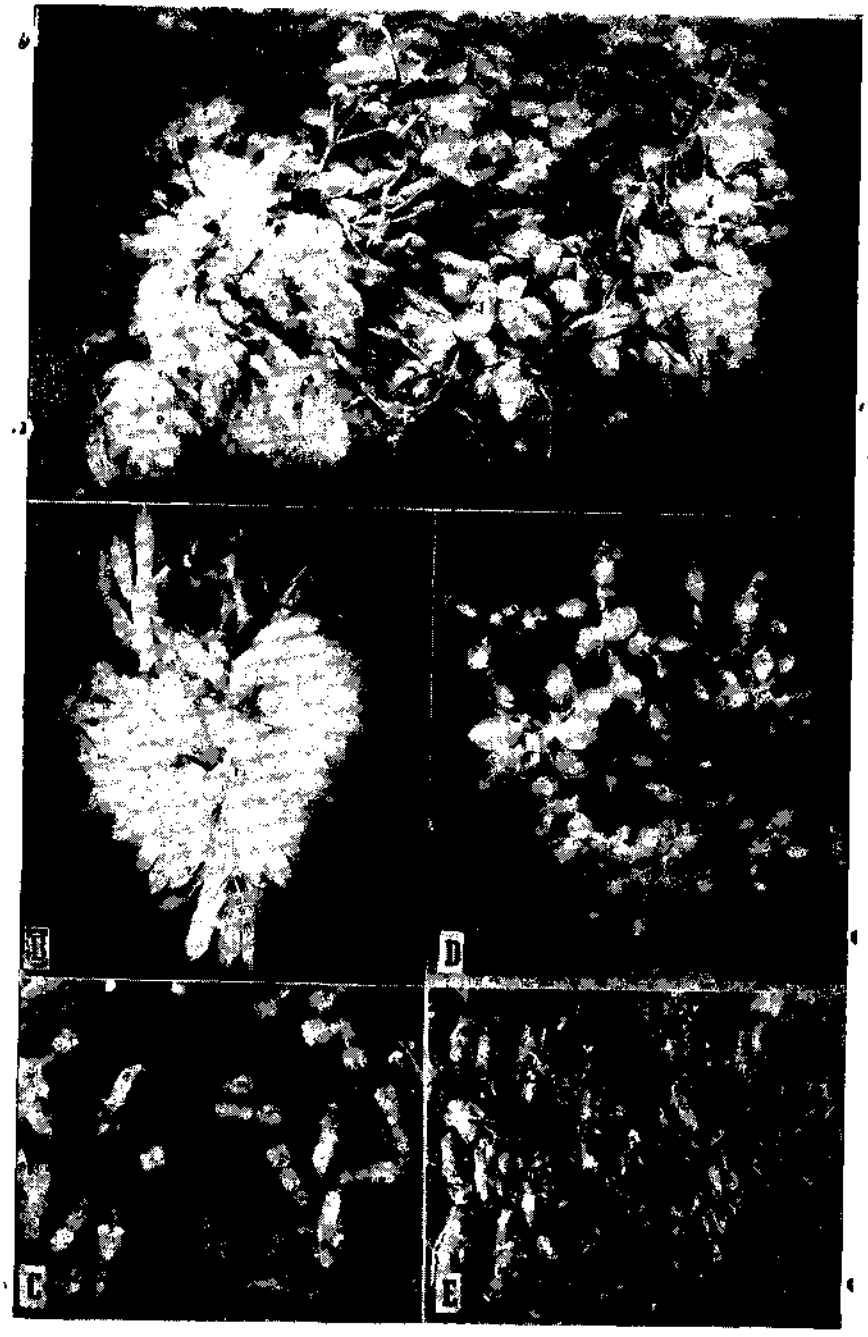

PLATE I A. Egg clusters of squid and cuttlefish found laid up together, B. Egg clusters of the squid *Sepioteuthis lessoniana*, C. Hatchlings of *Sepioteuthis lessoniana*, D. Egg clusters of the cuttlefish *Sepia aculeata* and E, Hatchling of *Sepia aculeata*.

Egg capsules of *Sepia aculeata* introduced into tanks on 27-3-'79 hatched out on the night of 31-3-'79 and also subsequently. The young cuttlefish measured 10 to 11 mm (Plate I). They were always observed to hatch out of the capsules near the base of their attachment. All the young cuttlefish died within 4-5 days.

Remarks

La Roe (1971) recommends opaque tanks especially of wood or cement as superior to glass tanks for hatching and survival of young ones of squids. Seminatural substrata including gravel, rock pieces or sea grasses were found to provide a calming and beneficial effect on the squids. But Choe and Oshina (1963) prefer glass tanks indoors for rearing experiments. The present studies on the survival of newly hatched squids show that in glass tanks with running sea water inside the aquarium survival was for 5 days only, while in concrete tanks with artificial sandy bottom, granite, coral stone and seaweeds, survival was for ten days.

La Roe (1971) succeeded in rearing newly hatched squid *S. sepioidea* by giving proper choice and quantity of food upto sexually mature condition for a period of 146 days. *S. sepioidea* was found to thrive well on the mysid *Musidium columbia* and older ones were found to feed primarily on *Gambusia*, as well as on penaeid prawns and fish of 3.7 cm in length. In the present study, newly hatched squids and cuttle fishes *S. lessoniana* and *S. aculeata* were provided with fresh plankton containing copepods and decapod larvae twice a day but the hatchlings did not prefer this food.

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PCBs AND PESTICIDES CONTENT IN CULTURED COCKLES FROM THE STATE OF PENANG MALAYSIA

P. M. SIVALINGAM, I. ALLAPITCHAY*, H. KOJIMA* AND T. YOSHIDA*
School of Biological Sciences, Universiti Sains Malaysia, Pulau Pinang, Malaysia

ABSTRACT

A survey on the PCBs and persistent pesticides content in cultured cockles obtained from 10 different farms in the State of Penang, *viz.* Bukit Dumbar, Pantai Acheh, Kuala Sungei Pinang 1, Kuala Singei Pinang 2, Kuala Jalan Bharu, Batu Maung 1, Batu Maung 2, Batu Maung 3, Sungei Nibong and Kuala Juru, demonstrated values ranging between the region of 160.29-335.31 ppbs for PCBs (Kanechlor 400) and non-detectable levels of p,p'DDE in most samples except for cockles obtained from the Batu Maung 1 site which demonstrated a value of 9.24 ppb.

The values for PCBs content in cultured cockles on comparison with shellfish of *Barbatia bicolorata*, *Atrina vexillum*, *Pinctada vulgaris* and *Saccostrea cucullata* from the Marine Depot, which had values of 436.55, 519.79, 467.25 and 461.57 ppbs respectively, were relatively low.

Based on these results it is concluded that cultured cockles in the State of Penang are acceptable for consumption from the viewpoint of their contamination.

INTRODUCTION

IN THE Malaysian context, the extent of heavy metal contamination has already been reported by Lee and Low (1976) and Sivalingam *et al.* (1979). Similar pollution evaluation studies on tropical benthic algae have also been carried out by Sivalingam (1978). However, the evaluation of pollutants such as PCBs, persistent pesticides and oil are yet to be reported.

The aquaculture of cockles *Anadara granosa*, in the world is well known to be regionalised within the west coast of Peninsular Malaysia (Table 1) with an annual production of 46,423.19 matric tons in 1977 with the State of Perak being the largest producer followed by Penang, Selangor, Kedah and West Johore.

TABLE 1. *Production of cockles* Anadara granosa *in Malaysia during the year* 1977**

States in west coast		Production (Ton)
Perlis	..	—
Kedah	..	233.82
Penang	..	4143.92
Perak	..	39,230.6
Selangor	..	2803.9
Negri Sembilan	..	—
Malacca	..	—
West Johore	..	10.95
Kelantan	..	—
Trengganu	..	—
Pahang	..	—
East Johore	..	—
Total (Tons)	..	46,423.19

** Data from annual fisheries statistics, 1977.

* Present address : Department of Marine Environmental Technology, Tokyo University of Fisheries, 4-5-7, Konan Minato-ku, Tokyo, Japan.

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P. M. SIVALINGAM, I. ALLAPITCHAY*, H. KOJIMA* AND T. YOSHIDA*
School of Biological Sciences, Universiti Sains Malaysia, Pulau Pinang, Malaysia

ABSTRACT

A survey on the PCBs and persistent pesticides content in cultured cockles obtained from 10 different farms in the State of Penang, *viz.* Bukit Dumbar, Pantai Acheh, Kuala Sungei Pinang 1, Kuala Singei Pinang 2, Kuala Jalan Bharu, Batu Maung 1, Batu Maung 2, Batu Maung 3, Sungei Nibong and Kuala Juru, demonstrated values ranging between the region of 160.29-335.31 ppbs for PCBs (Kanechlor 400) and non-detectable levels of p,p'DDE in most samples except for cockles obtained from the Batu Maung 1 site which demonstrated a value of 9.24 ppb.

The values for PCBs content in cultured cockles on comparison with shellfish of *Barbatia bicolorata*, *Atrina vexillum*, *Pinctada vulgaris* and *Saccostrea cucullata* from the Marine Depot, which had values of 436.55, 519.79, 467.25 and 461.57 ppbs respectively, were relatively low.

Based on these results it is concluded that cultured cockles in the State of Penang are acceptable for consumption from the viewpoint of their contamination.

INTRODUCTION

IN THE Malaysian context, the extent of heavy metal contamination has already been reported by Lee and Low (1976) and Sivalingam *et al.* (1979). Similar pollution evaluation studies on tropical benthic algae have also been carried out by Sivalingam (1978). However, the evaluation of pollutants such as PCBs, persistent pesticides and oil are yet to be reported.

The aquaculture of cockles *Anadara granosa*, in the world is well known to be regionalised within the west coast of Peninsular Malaysia (Table 1) with an annual production of 46,423.19 matric tons in 1977 with the State of Perak being the largest producer followed by Penang, Selangor, Kedah and West Johore.

TABLE 1. *Production of cockles* Anadara granosa *in Malaysia during the year* 1977**

States in west coast		Production (Ton)
Perlis	..	—
Kedah	..	233.82
Penang	..	4143.92
Perak	..	39,230.6
Selangor	..	2803.9
Negri Sembilan	..	—
Malacca	..	—
West Johore	..	10.95
Kelantan	..	—
Trengganu	..	—
Pahang	..	—
East Johore	..	—
Total (Tons)	..	46,423.19

** Data from annual fisheries statistics, 1977.

* Present address: Department of Marine Environmental Technology, Tokyo University of Fisheries, 4-5-7, Konan Minato-ku, Tokyo, Japan.

As yet, though endeavours are made to introduce farming of cockles in East Malaysia *i.e.*, Sabah and Sarawak, they have always turned out to be unfavourable.

Previous studies on trace metals contamination had thrown some light on this aspect of aquatic pollution and hence to envisage in depth on (environmental contamination with regards to this important marine protein source, the authors have endeavoured to make a general survey of PCBs and persistent pesticides content in cultured cockles from ten different farms in the State of Penang. These pollutants were then compared and evaluated with some commonly found shellfish at the Marine Depot, a station located near the university. This paper will discuss the level of PCBs and persistent pesticides contamination in cultured cockles which is of paramount importance in Malaysia and establish a baseline level of these contaminants for this species.

The authors would like to express their gratitude to the School of Biological Sciences, Universiti Sains Malaysia, Malaysia and Tokyo University of Fisheries, Tokyo, Japan for all the aid rendered during the course of this study.

Materials and Methods

Matured cockles were obtained from ten different culture farms (Fig. 1) in the State of Penang in July 1978 and the other shellfish species from the Marine Depot at the same time. The samples were brought to the laboratory, washed completely of contaminants and opened to obtain the meat. Normally 50 shellfish at a time were obtained from each culture farm and on pooling the meat they were completely dried in an air-oven at 95°C for *ca.* 72 hours before being powdered in a mortar. Triplicate lots from each culture farm were prepared to perform the analyses for PCBs and persistent pesticides content.

The dried powdered shellfish samples were extracted for their PCBs and pesticide contaminants as indicated in the flow-sheet on the following page.

After extraction of the PCBs, DDE and other pesticides in fractions 1 and 2 they were subjected to gas chromatographic analysis A.G.C.—4BM Shimadzu gas chromatograph under the following operating conditions was employed.

Column packing	: OV 17/1.5% Chromosorb W
Detector temp.	: 210°C
Column temp.	: 190°C
N_2 flow	: 30 ml/minute
Chart speed	: 10 mm/minute
Range	: $10^2 \times 8$
ECD	: 63_{Ni}

The gas chromatogram of the two fractions was then compared with authentic samples of PCBs and pesticides (Fig. 2) and the amount of both contaminants was calculated using the following formula :

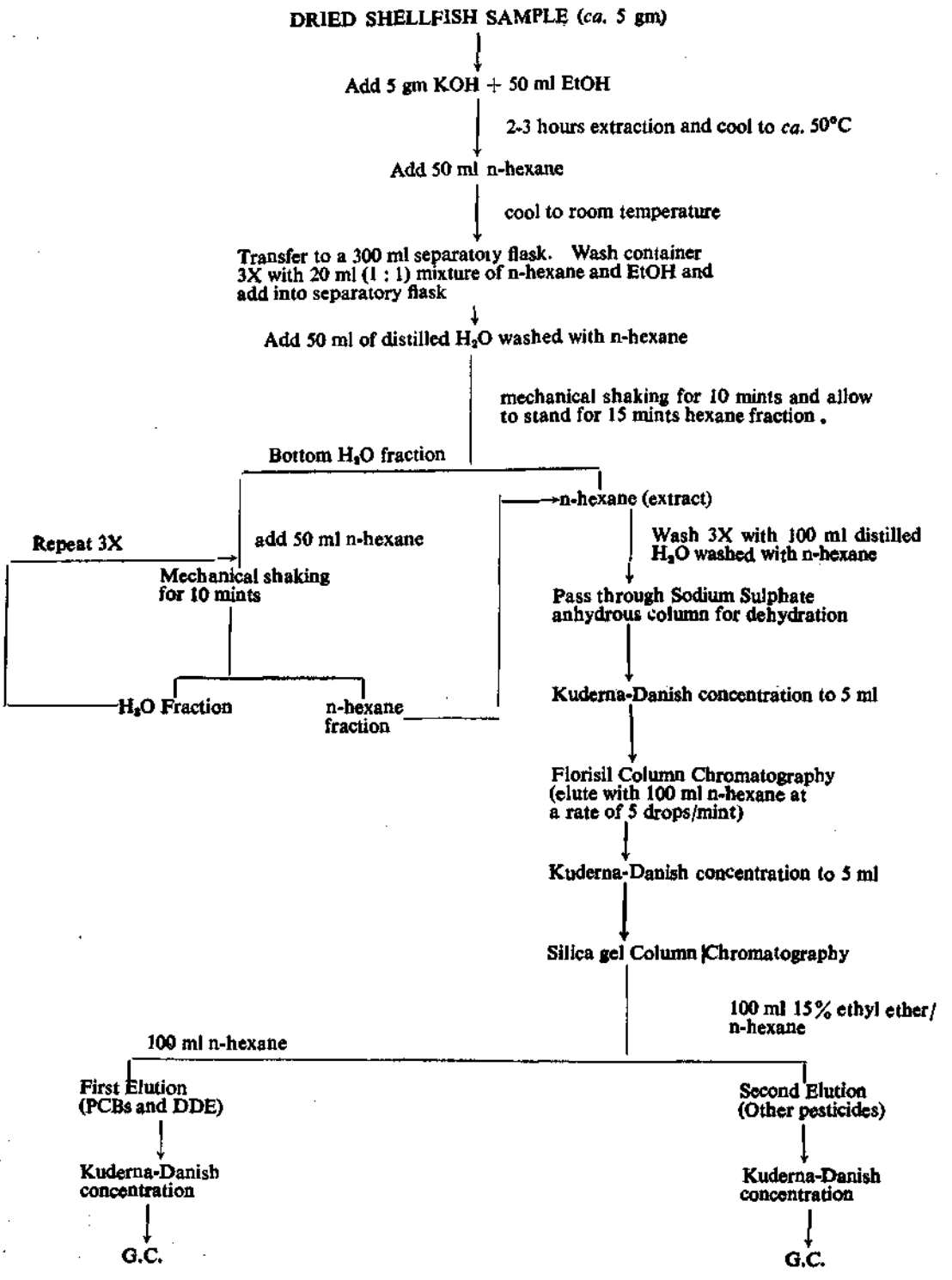
$$\frac{V \times h_{\text{PCB or DDE}} \times V^1}{m \times v \times h} \times C = \text{result in ppm}$$

where

V = total volume of n-hexane extract (ml)
m = weight of sample (gm)
v = volume of sample injected (μl)
V^1 = volume of standard injected (μl)
h^1 = peak height of PCB or DDE in the standard (mm)
C = concentration of standard (μg/μl)

Results

Table 2 shows the contents of PCBs and persistent pesticides in cultured cockles *Anadara granosa*, *Barbatia bicolorata*, *Atrina vexillum*, *Pinctada vulgaris* and *Saccostrea cucullata*. It is noticeable that the PCBs (Kanechlor 400) content in cultured cockles along the east


DRIED SHELLFISH SAMPLE (ca. 5 gm)
Add 5 gm KOH + 50 ml EtOH
2-3 hours extraction and cool to ca. 50°C
Add 50 ml n-hexane
cool to room temperature
Transfer to a 300 ml separatory flask. Wash container 3X with 20 ml (1 : 1) mixture of n-hexane and EtOH and add into separatory flask
Add 50 ml of distilled H2O washed with n-hexane
mechanical shaking for 10 mints and allow to stand for 15 mints hexane fraction.
Bottom H2O fraction
n-hexane (extract)
Repeat 3X
add 50 ml n-hexane
Mechanical shaking for 10 mints
Wash 3X with 100 ml distilled H2O washed with n-hexane
Pass through Sodium Sulphate anhydrous column for dehydration
H2O Fraction
n-hexane fraction
Kuderna-Danish concentration to 5 ml
Florisil Column Chromatography (elute with 100 ml n-hexane at a rate of 5 drops/mint)
Kuderna-Danish concentration to 5 ml
Silica gel Column Chromatography
100 ml 15% ethyl ether/ n-hexane
100 ml n-hexane
First Elution (PCBs and DDE)
Second Elution (Other pesticides)
Kuderna-Danish concentration
Kuderna-Danish concentration
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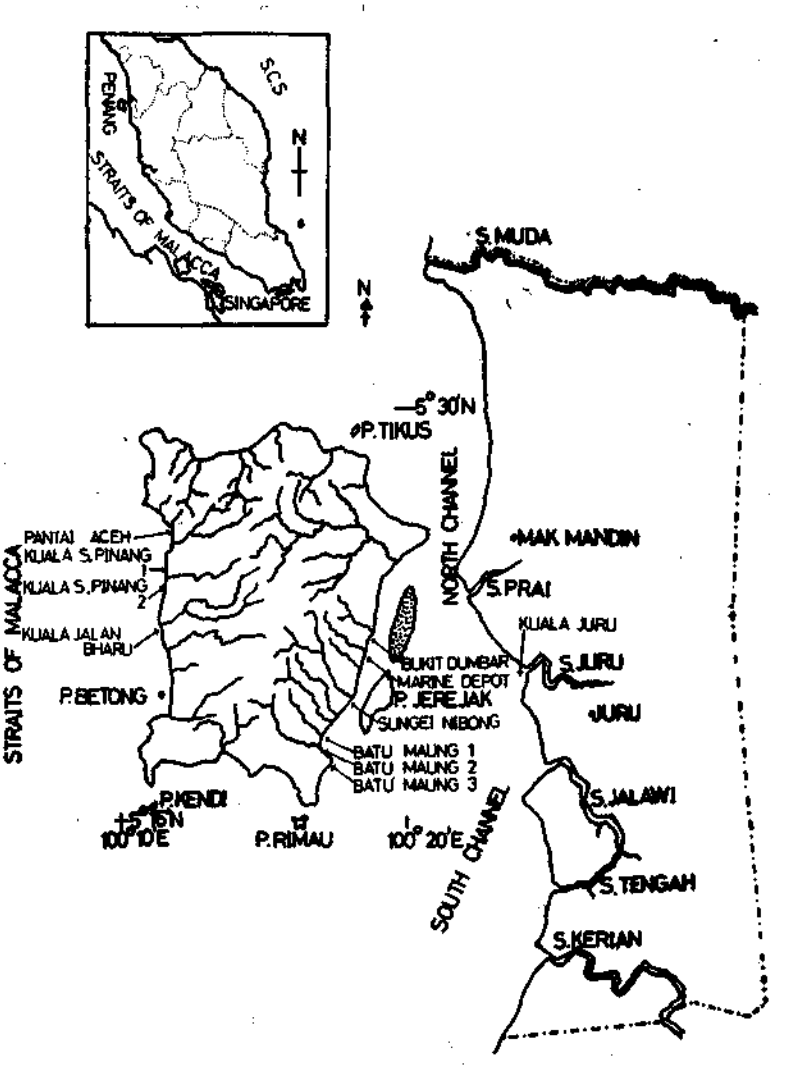

Fig. 1. Sampling sites of cultured cockles and some common shellfish of Penang Island.

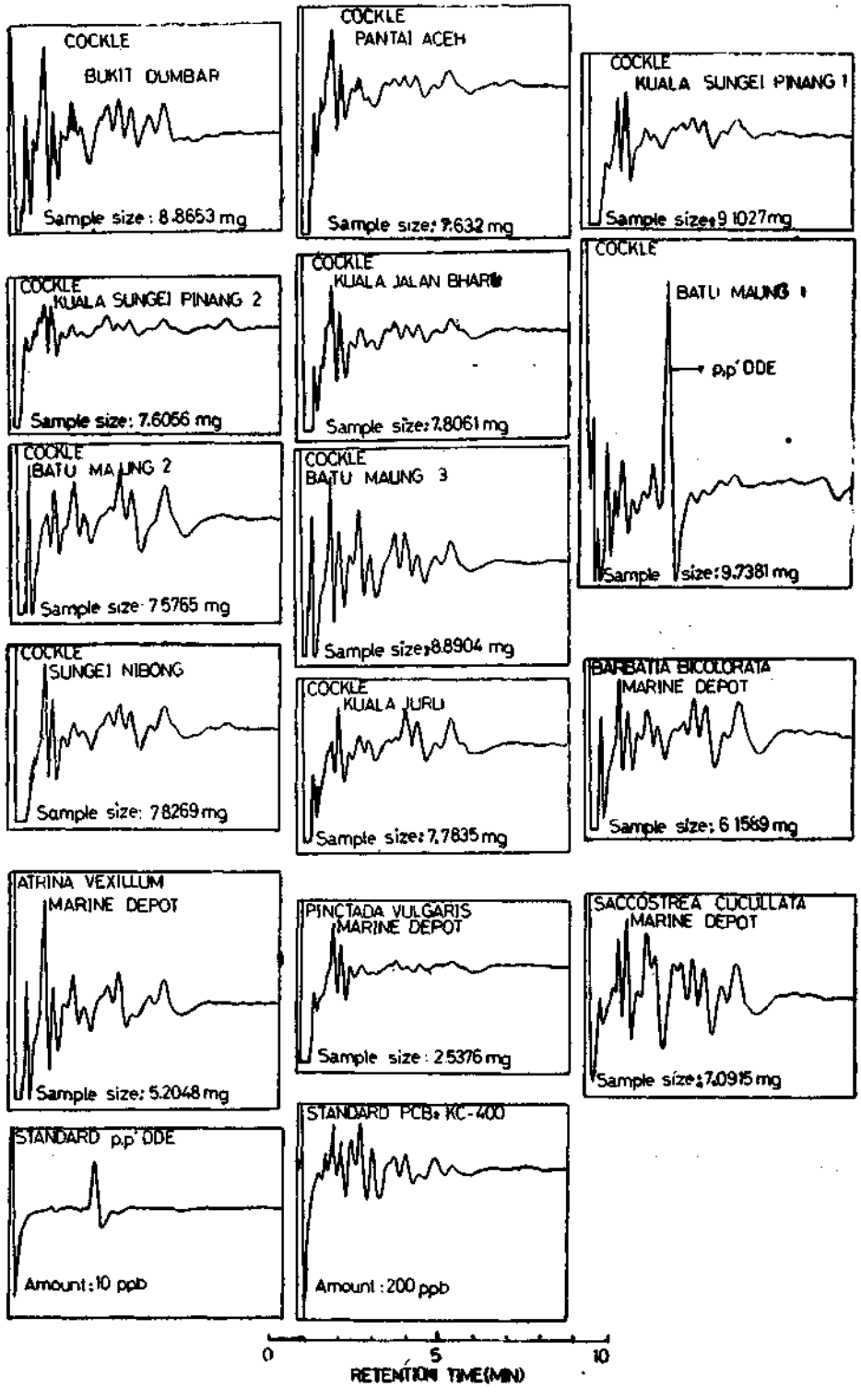

Fig. 2. Gaschromatograms of PCBs and Pesticides extracted from cultured cockles and some common shellfish of the island of Penang.

TABLE 2. *PCBs and pesticides content in cultured cockles and some common shellfish of the island of Penang, Malaysia*

Sampling Site	Species	Date of Sampling	PCBs (KC 400) (ppb)	p, p'DDE+ (ppb)
Bukit Dumbar	*Anadara granosa*	12 Sept. 1978	335.31	N.D.*
Pantai Aceh	*Anadara granosa*	14 Sept. 1978	249.45	N.D.
Kuala Sungei Pinang 1	*Anadara granosa*	14 Sept. 1978	174.45	N.D.
Kuala Sungei Pinang 2	*Anadara granosa*	14 Sept. 1978	160.29	N.D.
Kuala Jalan Bharu	*Anadara granosa*	12 Sept. 1978	256.72	N.D.
Batu Maung 1	*Anadara granosa*	14 Sept. 1978	198.93	9.24
Batu Maung 2	*Anadara granosa*	14 Sept. 1978	306.38	N.D.
Batu Maung 3	*Anadara granosa*	14 Sept. 1978	309.97	N.D.
Sungei Nibong	*Anadara granosa*	14 Sept. 1978	279.5	N.D.
Kuala Juru	*Anadara granosa*	18 Sept. 1978	193.10	N.D.
Marine Depot	*Barbatia bicolorata*	18 Sept. 1978	436.55	N.D.
	Atrina vexillum	18 Sept. 1978	519.79	N.D.
	Pinctada vulgaris	18 Sept. 1978	467.25	N.D.
	Saccostrea cucullata	18 Sept. 1978	461.57	N.D.

* not detectable.

coast of Penang Island ranges between 198.93 and 335.31 ppbs while those on the west coast are between 160 and 256.72 ppbs. This reflects the possibility of a lower level of contamination on the west coast where there is little industrialisation. It should also be noted that the contents of PCBs in cultured cockles in the Kuala Juru area showed only a value of 193.10 ppb. These values on comparison with *Barbatia bicolorata, Atrina vexillum, Pinctada vulgaris* and *Saccostrea cucullata* from the Marine Depot are relatively low.

As for pesticides content, it is obvious that only p, p'DDE is defected in the Batu Maung site 1 sample with a value of 9.24 ppb.

DISCUSSION

Surveys of PCBs content in wet tissues of *Crassostrea* from Nagasaki, Hiroshima and Osaka, *Tapes* from Ise, Mie and Matsusaka, *Corbicula* from Shimane, *Meretrix* from Korea and *Mytilus californianus* from CA, U.S.A., indicated values of 0.014, 0.031, 0.27, 0.006, 0.002, 0.087, 0.015 and 0.004 ppms respectively (Akio Nakamura and Takashi Kashimoto, 1978). Similar investigations by Sims *et al.* (1977) in clams, mussels, oysters and scallops from the Northwest Atlantic during 1971-72 indicated values of 0.016, 0.023, 0.005 and 0.018 ppms respectively. Samples of mussel from Puget Sound, Washington had values between 0.010-0.210 ppm and those in fisheries product of mussel from the Dutch Coast *ca.* 0.265 ppm and 42.5 ppm in the fat of the product at one station while *ca.* 0.335 ppm in the product and 51 ppm in the fat in another station in 1976 (Hagel and Tuinstra, 1978) were recorded. Based on this information and since it is a well-established fact that the normal moisture content of shellfish varies between 70% and 80%, the values of PCBs content indicated for cockles as dry weight of samples are not in the least overwhelming.

With regard to p,p'DDE content, it has been reported by Sims *et al.* (1977) that bivalves

from the Northwest Atlantic have 0.51 ppm for wet weight samples which again warrants acceptability of the cultured cockles in the state of Penang.

Based on the information presented above and the comparative values, it can be concluded that the PCBs and persistent pesticides content in cultured cockles from the state of Penang are fairly below the acceptable value. It is also intriguing that the PCBs content in cultured cockles from the west coast of the island of Penang as compared to those of the east coast are smaller. This point requires further investigation and clarification.

Finally, the authors also feel that probably the present data could act as a baseline study for future problems of PCBs and persistent pesticides contamination in cultured cockles in Malaysia.

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TOXICOLOGICAL STUDIES ON THE GREEN MUSSEL *PERNA VIRIDIS*

N. R. MENON[1], N. A. REDDY[2] AND A. E. EKNATH[3]
College of Fisheries, University of Agricultural Sciences, Mangalore

ABSTRACT

The toxicology of *Perna viridis* was studied employing a few important toxicants, such as NH_3, NH_3^+, LDO, HSD and Heptachlor. Lethal concentrations leading to 50% mortality of test population at 96-h (LC 50-96 h) were delineated. Effective time (ET50) was defined for the set reaction like mortality or cessation of activity. The observations gave the following results. LC50 values for NH_3 is 7.6 mg l^{-1} at 96 h, and that for NH_4^+ 13.0 mg l^{-1}. However, in concentrations relatively lesser than the above total cessation of byssus thread secretion occurred. The pedal gland activity has been tampered with, in concentrations at lower threshold levels. The byssus threads produced showed retrogression in development under ammonia stress.

The 96 h-LC50 for the LDO-WSF was 13% and HSD-WSF 8.5%. An assymptotic pattern of tolerance reaction was observed for LDO-WSF at lower concentrations employed. The rate of byssus thread secretion was considerably reduced in oil polluted test media.

It is proved that Heptachlor, an organochlorine is highly poisonous to *Perna viridis* even at very low concentration. The 96 h-LC50 falls at 3.3 μl l^{-1}. Even the presence of 0.1 μl l^{-1} reduced byssus secretion by 50%, while at 1.0 μl l^{-1} the secretion was totally suspended. At concentrations above 0.0 μl l^{-1} the animals were in a state of suspended animation. Considerable quantities of mucus secreted on the gill surface impaired gill irrigation.

INTRODUCTION

DURING the present investigation, bioassay studies were conducted in the laboratory under 'static' conditions. The experiments conducted have been programmed to find out the lethal and sub-lethal toxicity, the former explained with the help of mortality tests and the latter based on the assessment of an important rate function byssus secretion. Lethal effects refer to the death of the organisms. There is a general feeling that the study of acute toxicity is not sufficient and that there must be more concern with sub-lethal effects. It is relatively easy to document small changes within an animal, but there is always a doubt whether the changes are deleterious or merely within the normal range of adaptation of the animal. It is necessary that studies on sub-lethal changes be carried far enough to show the ecological relevancy of the results.

Byssus thread formation the important rate function employed during the present investigation to assess the sub-lethal effects of NH_3, NH_4^+, LDO, HSD and Heptachlor on *Perna viridis* is a very useful tool (Roberts, 1975; Reddy and Menon, 1978, 1979a; Eknath and Menon, 1979).

Information on the effects of NH_3 and NH_4^+ in the environment on the life processes of marine organisms is scarce. Working on the effects of ammonium nitrate on fertilization and early development of *Choromytilus meridionalis*, Brown (1974) found that concentrations as

[1] Present address: Department of Marine Sciences, University of Cochin, Cochin-682 016.
[2] Present address: Central Inland Fisheries Research Institute, Barrackpore, West Bengal.
[3] Present address: Department of Oceanography, Dalhousie University, Canada.

low as 0.5 ppm retard early development. Lot of information is available on the effects of petroleum products on the life and activity of commercially important bivalves. Blumer *et al.* (1970), Lee *et al.* (1972), Gilfillan (1973) and Reddy and Menon (1978) have worked on the effects of petroleum hydrocarbons on marine bivalves. Trying to evaluate stress profiles induced by pesticides, Eisler (1972) has given an account of Methylparathion, Endrin and Methoxychlor on the various organs of fishes and bivalves. Schimmel *et al.* (1976) studied the effects of Heptachlor on several estuarine organisms. Papers of topical importance are those of Armstrong *et al.* (1976) and Dethlefsen (1977).

Materials and Methods

Individuals of *Perna viridis* Kuriakose and Nair were collected from the rocky shores of Someswara Beach (12°47′ N and 74°51′ E) near Mangalore and transported to the laboratory 24 hours before the experiments. The unfed animals were kept in the laboratory under aerated condition. Young animals of the size range 20 to 24 mm were used for the experiments.

Liquor ammonia containing 25% of free ammonia was the source of molecular ammonia and ammonium sulphate was the source of ammonium (NH_4^+). Extract containing water soluble fraction of High Speed Diesel (HSD) and Light Diesel Oil (LDO) were prepared following the procedure of Dunning and Major (1974). The aqueous fraction was considered as 100% sea water-oil extract. The different concentrations employed are expressed in terms of percentage of WSF. Commercial Heptachlor was used for studies on Heptachlor toxicity. Usually a carrier solution (acetone) is used to prepare Heptachlor suspension ; however, during the present study only simple suspensions were prepared without a carrier solution. The sea water used for all the experiments was collected from Someswara Beach and had a salinity of 32.0 to 32.8‰ and pH 8.10 to 8.18. The sea water was filtered with Whatman filter paper. All experiments were conducted under room temperature. The experimental vessels were cylindrical glass troughs of 5 litre capacity containing 4 litre of test solution.

Since the purpose of the investigation was to delineate both acute and sub-acute toxicity levels, rate of mortality and rate of activity by way of byssogenesis (Roberts, 1975) were estimated. Mortality tests were conducted for 96 h. Inability to close the valve on mechanical stimulus and valve gaping of 5 mm were the criteria used to determine death. To check the possibility of revival, those mussels considered dead by the above criteria were exposed to normal control conditions for 24 h. Dead individuals were removed from the experimental media at 12 h intervals. Usually 10 mussels were used at each concentration. Byssus thread formation was calculated as the number of threads secreted by one mussel. Readings were taken at 12 h intervals (at 0600 and 1800 hrs). Since the majority of the threads formed developed adhesive discs, these discs were counted and then the whole byssal mass (stem and threads) was cut off and the mussels were left in the test solution. It was found that cutting off the threads does not affect further byssogenesis.

Results

Mortality

Ammonia

The concentration of ammonia used was 1 mg l^{-1} to 33 mg l^{-1}. Death of test individuals occurred in 5.5 and 8.25 mg l^{-1} after 48 h exposure. One hundred per cent mortality occurred before 96 h at 16.5 and 22.0 mg l^{-1}. The ET50 values worked out from the results obtained on mortality are presented in Fig. 1. It is evident from the illustration that a cons-

picuous difference in time exists for the incidence of 50% mortality between concentrations of 16.5 and 20.0 mg l^{-1}. The 48h-LC50 was 14.0 mg l^{-1} (pH 8.775) and 96h-LC50 7.6 mg l^{-1} (pH 8.520) (Fig. 1).

Ammonium

The ammonium concentration employed for the tests varied between 5.0 to 75.0 mg l^{-1}. Hundred per cent mortality occurred in 36 h at 75.0 mg l^{-1} and in 48 h at 50.0 mg l^{-1}. The ET50 values, computed from the mortality rates show that 50% of the test organisms died within 49.8 h at 15.0 mg l^{-1}. Curiously enough, the difference between the ET50 values for 50.0 and for 75.0 mg l^{-1} is only 1.8 h. The 48h-LC50 was 15.5 mg l^{-1} (pH 8.075) and 96h-LC50 13.0 mg l^{-1} (pH 8.125) (Fig. 2).

Light Diesel Oil (LDO)

Affecting survival rate at 48 h, 60% of the animals were killed at 15% concentration after 72 h. While a few numbers of the test individuals survived even after 60 h in 20% all the organisms died at 36 h in 30%. It took 67.8 h at 15% of WSF to kill half of the test animals while the duration was only 16.2 h in 30% WSF (Fig. 3). 48h-LC50 falls at 18.5% and 96 h-LC50 at 13%.

High Speed Diesel (HSD)

Hundred per cent mortality occurred at around 96 h in 20%-WSF, whereas, this limit was found to reach in 30% around 48 h. The ET50 values showed gradual decrease from lower to higher concentrations. 48h-LC50 was 14% and 96h-LC50 8.5% (Fig.4).

Heptachlor

Among the seven concentrations employed ranging from 1 μl to 10 μl l^{-1}, death occurred after 84 h of exposure at 3 μl l^{-1} and 4 μl l^{-1} whereas, from 60 h onwards *P. viridis* started dying at 5, 7.5 and 10 μl l^{-1}. A conspicuous feature of the results obtained is that death takes place only at a later stage and once the animals start dying, the increase in mortality rate is sudden. The ET50 ranged between 85.8 h at 4 μl l^{-1} and 58.8h at 10 μl l^{-1}. 96h-LC50 was 3.3 μl l^{-1} (Fig. 5).

Byssogenesis

The process of byssus-thread secretion by some bivalves is referred to as byssogenesis. The rate of byssus production, the nature of secretion and the morphology of the thread are useful criteria employed to assess activity.

Ammonia and ammonium

Continuous production of byssus threads was evident only at 2.75 and 5.50 mg l^{-1} of ammonia whereas the individuals exposed to 16.5 and 22.0 mg l^{-1} did not produce any threads. The total number of byssus threads produced by all the mussels exposed to different concentrations showed sharp decline between 5.5 and 8.25 mg l^{-1} (Fig. 6).

The intensity of secretion of byssus thread production dropped significantly above the concentration of 10.0 mg l^{-1} of ammonium. Those animals kept at 50.0 mg and 75.0 mg l^{-1} did not produce any threads (Fig. 7).

Light Diesel Oil (LDO)

Retardation or cessation of byssus secretion happened only at higher concentration, *viz.*, 15 to 20% of light diesel oil (LDO) water soluble fraction. Although those mussels exposed to 5 to 10% of LDO-WSF were active at the early phase of the test, the animals became less active after 36 hours of exposure. The amount of threads produced after this time interval was negligible. No animal exposed at 20% and 30% LDO-WSF produced byssus threads (Fig. 8). The analysis of variance indicated that the difference in byssogenesis between concentrations is insignificant at 1% level.

High Speed Diesel (HSD)

In the case of *Perna viridis* exposed to the various concentrations of the water soluble

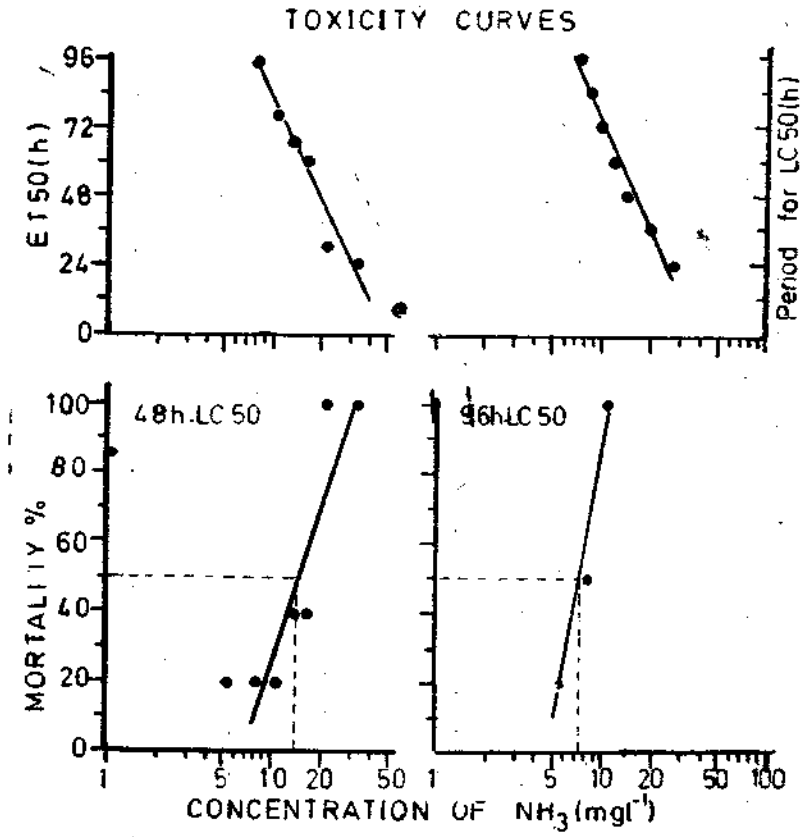

Fig. 1. Toxicity curves showing the different lethal concentration (LC) and the effective times (ET) under varying NH_3 concentrations.

fig 2

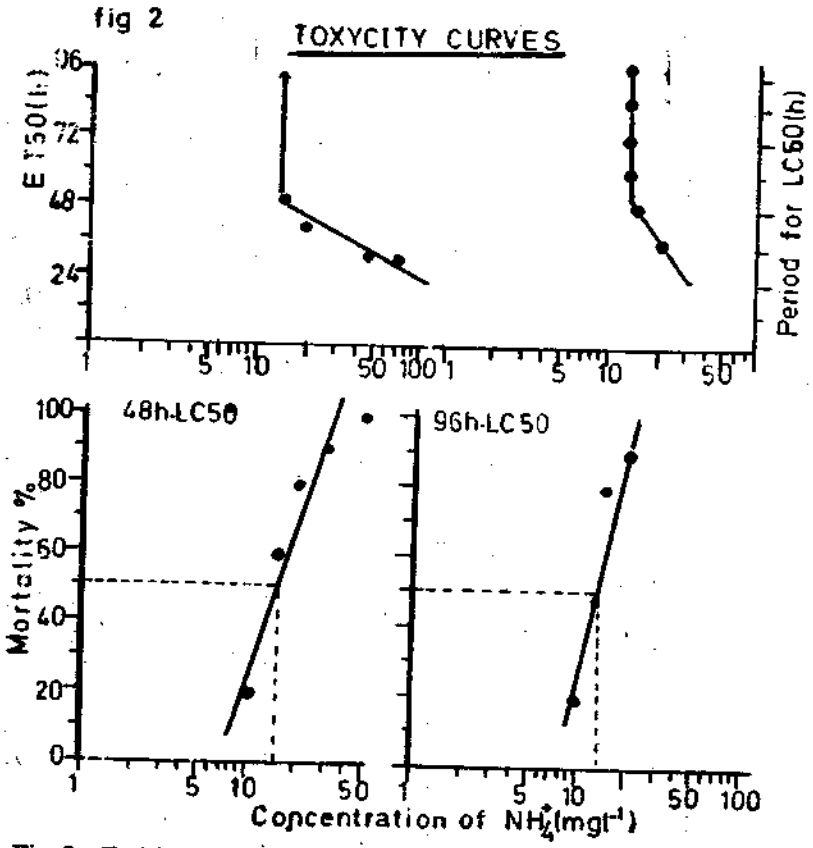

Fig. 2. Toxicity curves showing the different letahl concentrations (LC) and the effective times (ET) under varying NH_4 concentrations.

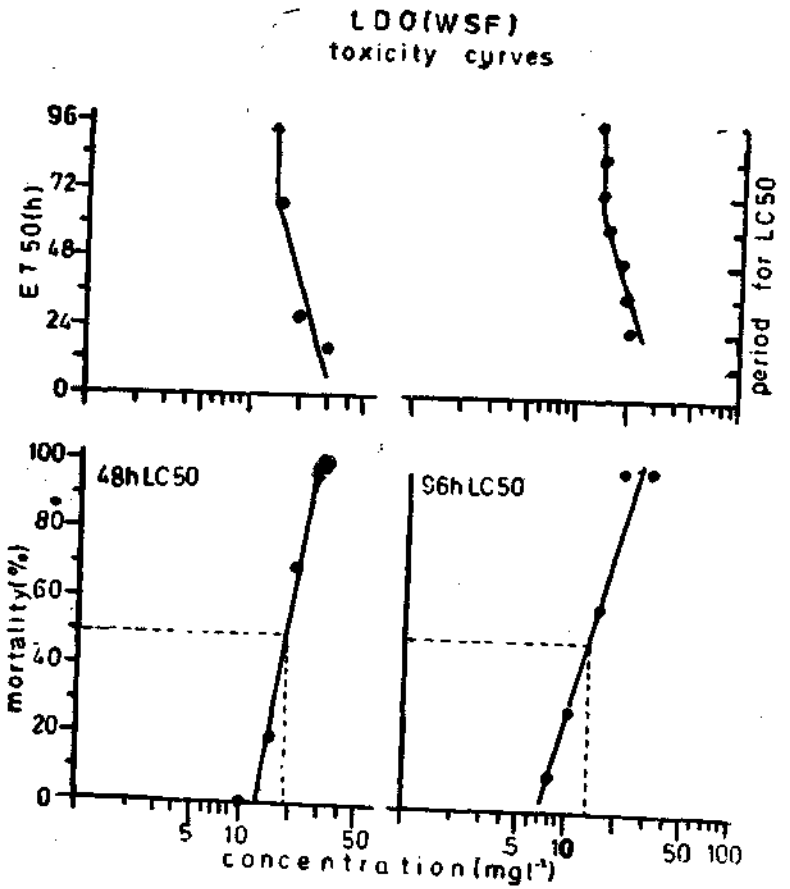

Fig. 3. Toxicity curves showing the different lethal concentrations (LC) and the effective times (ET) under varying concentrations of Light Diesel Oil—Water Soluble Fractions (LDS-WSF).

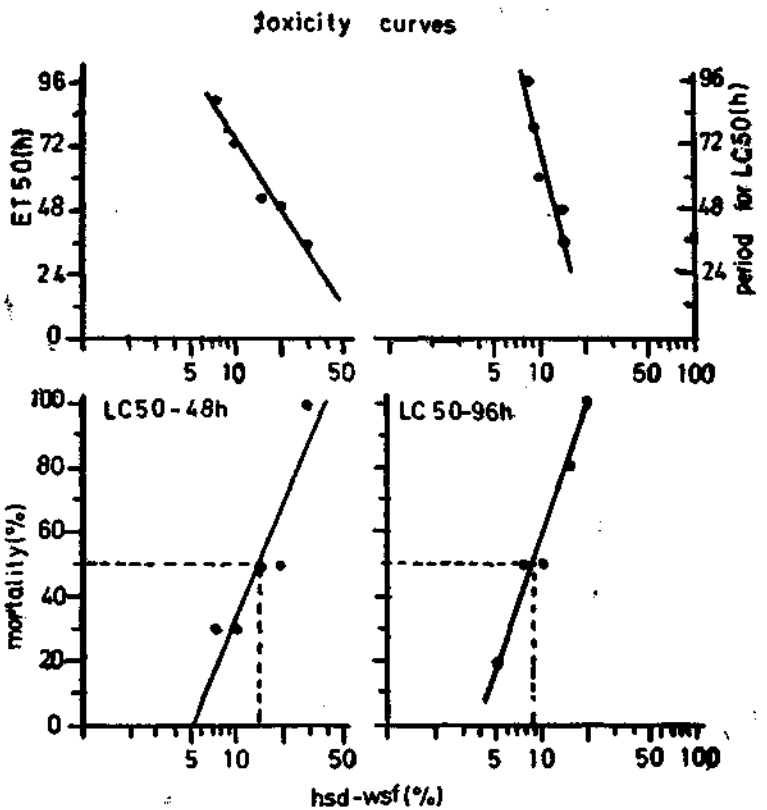

Fig. 4. Toxicity curves showing the different lethal concentrations and effective times (ET) under varying conconcentrations of High Speed Diesel-Water Soluble Fractions (SD-WSF).

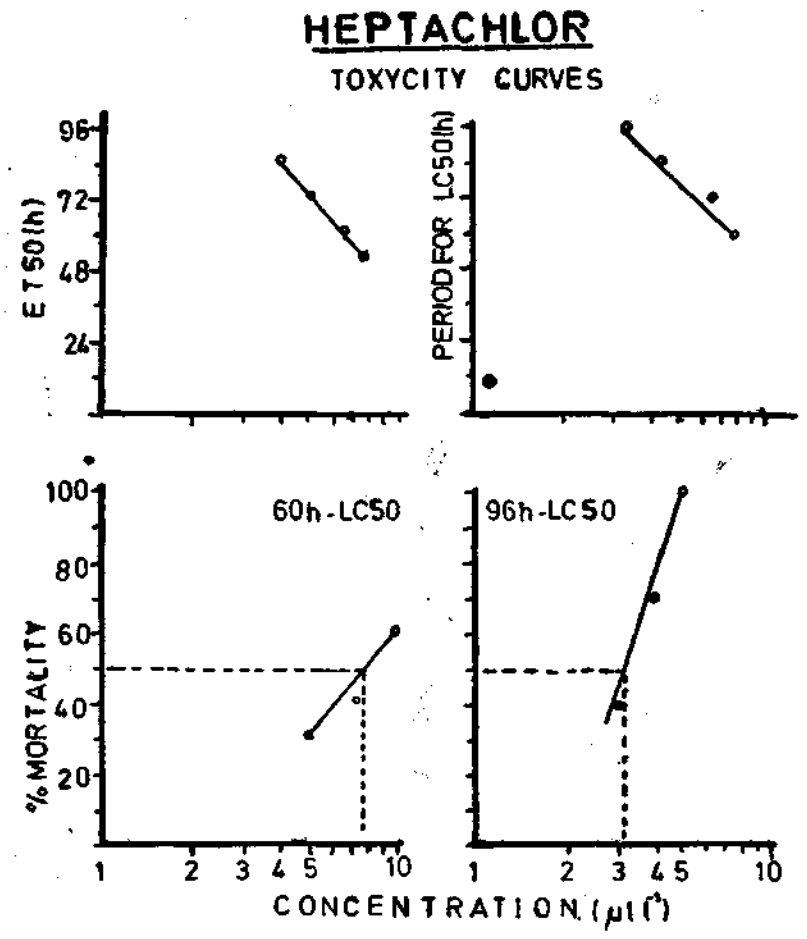

Fig. 5. Toxicity curves showing different lethal concentrations and effective times (ET) under varying concentrations of heptachlor.

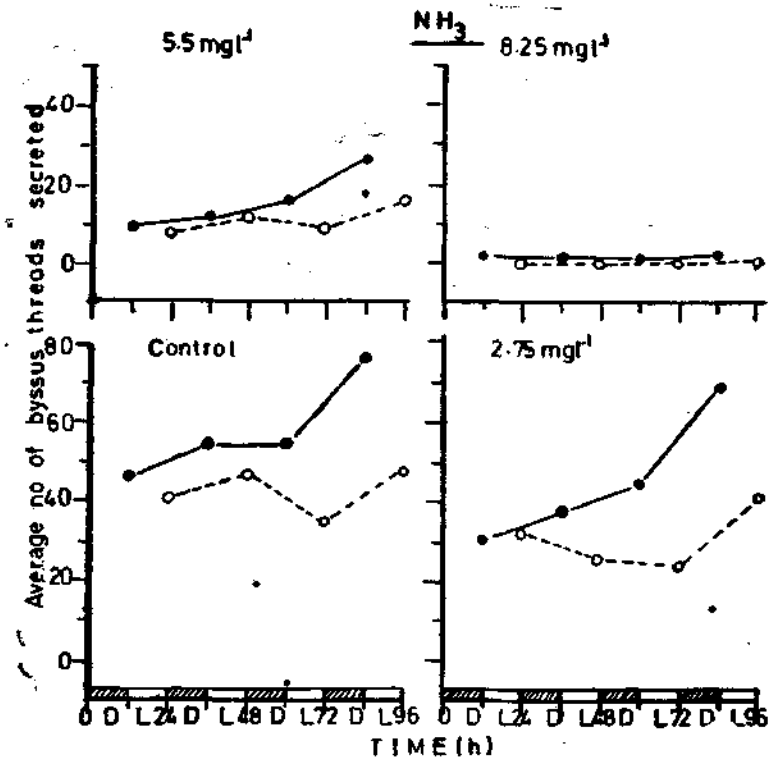

Fig. 6. Byssogenesis—as a function of NH_3 exposure (●—Dark regime, ○ Light regime).

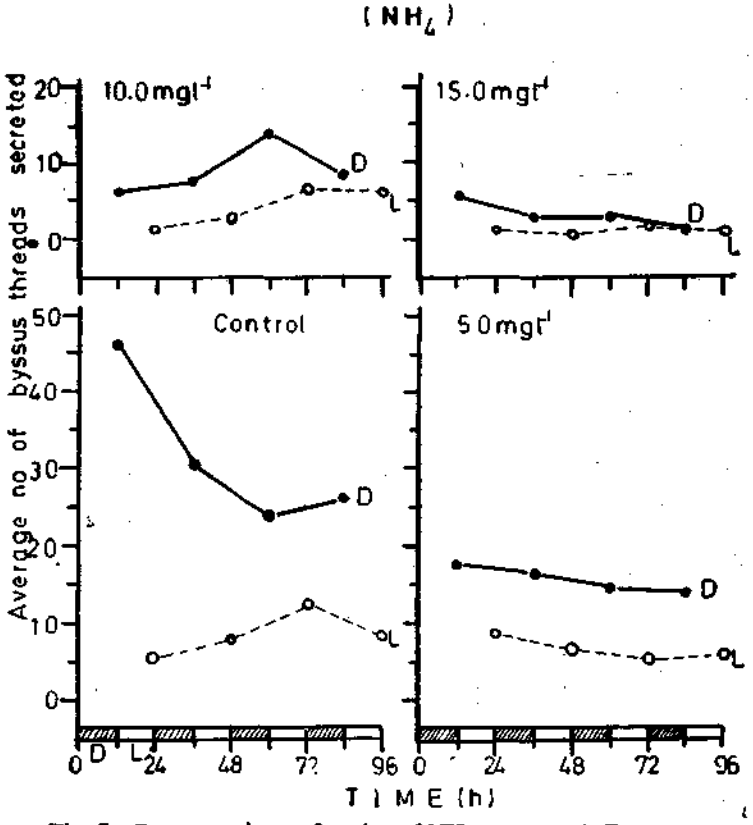

Fig. 7. Byssogenesis as a function of NH_4 exposure (●D—Dark regime, ○L—Light regime).

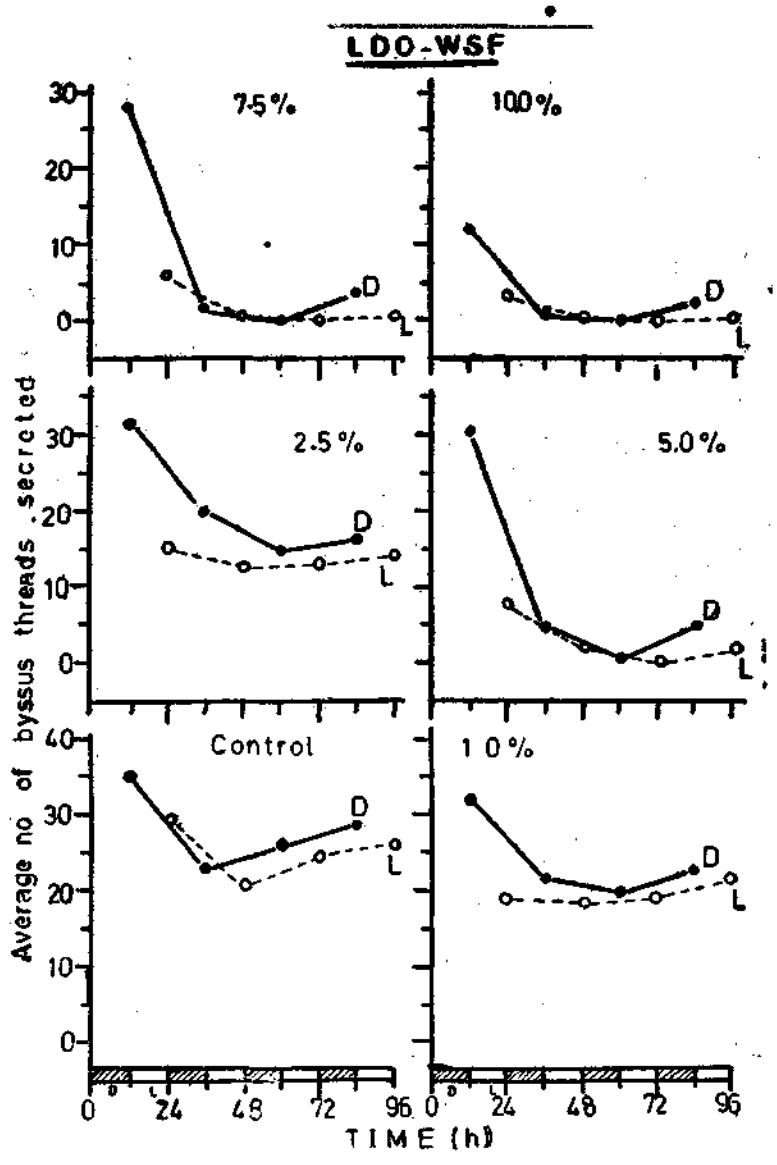

Fig. 8. Byssogenesis as a function of exposure of Light Diesel Oil —Water Soluble Fraction (LDO-WSF) (●D = Dark regime, ○L = Light regime).

fraction of high speed diesel (HSD-WSF) the reactions were different. Rate of byssus thread production was very low beyond 7.5% whereas, those test individuals maintained at 5% depicted only a restricted rate of activity. Pedal gland activity was totally suspended in 30% after 24 h. The reduction in the total number of threads produced was drastic from 10% and above (Fig. 9). The ANOVA shows that the observed variations between intervals and concentrations are significant at 1% level.

Heptachlor

The capacity of *P. viridis* to produce byssus threads when exposed to Heptachlor concentrations ranging from 0.1 to 1.0 μl l^{-1} has been worked out. Even in the presence of 0.1 μl l^{-1} resulted in 50% reduction of byssus production (Fig. 10).

Discussion

Considerable attention has been paid in recent years to study the toxicology of culturable marine bivalves. Information gathered from such studies will be of immense practical importance for any project on culture of these bivalves. The toxicants employed in the present investigation are those which are found as pollutants in the coastal waters of India.

Detailed studies on the tolerance of acute toxicity and sub-acute toxicity to the various important pollutants, such as ammonia, ammonium, oil, pesticides and heavy metals, by *Perna viridis* have been conducted by the authors and the results are under publication (Reddy and Menon, 1978, 1979 a, b ; Eknath and Menon, 1978, 1979, 1980).

Becker and Thatcher (1973) found that both nitrite ion (NO_2^-) and ammonia are highly toxic to a wide variety of marine organisms. Epifanio and Srna (1975), discussing the effects of different concentrations of ammonia, report that adults of *Crassostrea virginica* have a median tolerance limit (96h Tlm) of 0.082 m l^{-1}. But in the case of *Mercenaria mercenaria*, these values showed a downward trend. The present study clearly indicates that the tolerance limits of *Perna viridis* to NH_3 and NH_4^+ are different. During the course of the experiments, it was observed that individuals of *P. viridis* exposed to higher concentrations of NH_3 and NH_4^+: secreted considerable quantities of mucus. Enlightening on the biological consequence of ammonia accumulation, Kinne (1976) remarked that continuous exposure could bring about hyperplasia of the gill epithelium in the case of freshwater fishes.

The water soluble fractions of the oil can contain different percentages of soluble hydrocarbons. Discussing on the effects of exposure of crustaceans and molluscs to various concentrations of oil, Anderson *et al.* (1974) found that there is an accentuation of respiratory rate at WSF of No. 2 Fuel oil between 15 and 20%. Since it is not possible to quantify exactly the amount of oil present in the WSF at the present instance, LC50-48 and 96 hours are described based on percentage of WSF. Discussing on the relative merits of WSF of refined and crude oil, Anderson *et al.* (1974) found that the soluble fractions of refined oil are much more toxic than crude. Molluscs have been found to have variable responses to oil. Successive immersion caused little mortalities in oysters (Chipman and Galtsoff, 1949). However, some bivalves and gastropods have been found to be very sensitive to oil (North *et al.*, 1964 ; Miranov, 1967). *Donax spiculum* was found to be less tolerant to light diesel oil-WSF when compared with *Perna viridis* (Reddy and Menon, 1978).

Heptachlor and its metabolite Heptachlor epoxide (Davidow and Radomsky, 1953) have been found in estuarine and marine systems (Schimmel *et al.*, 1976). This organochlorine has been proved to be highly toxic to nontarget marine organisms. Static bioassay studies with

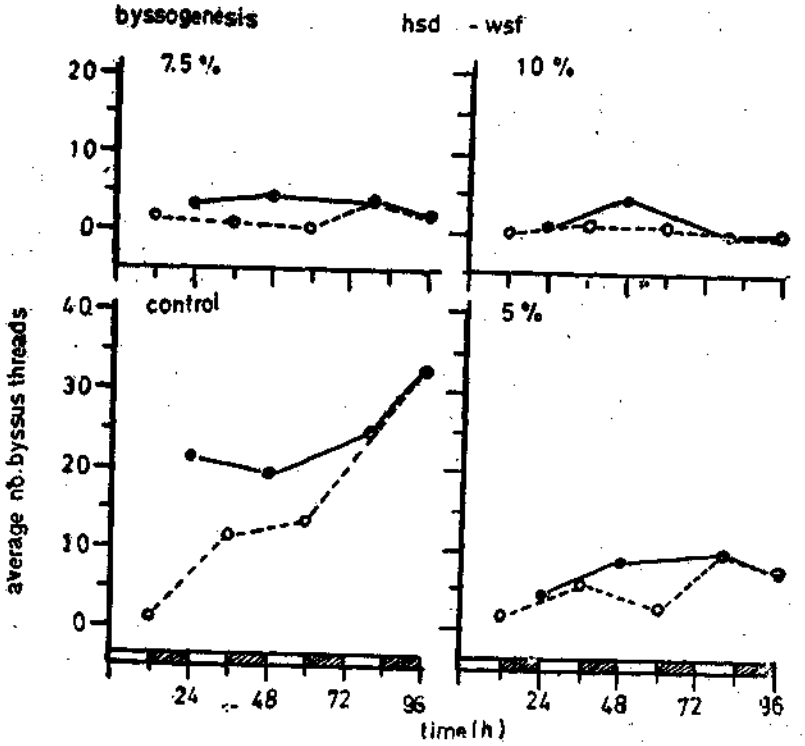

Fig. 9. Byssogenesis as a function of exposure to High Speed Diesel—Water Soluble Fraction (HSD-WSF) (● Dark regime, ○ Light regime).

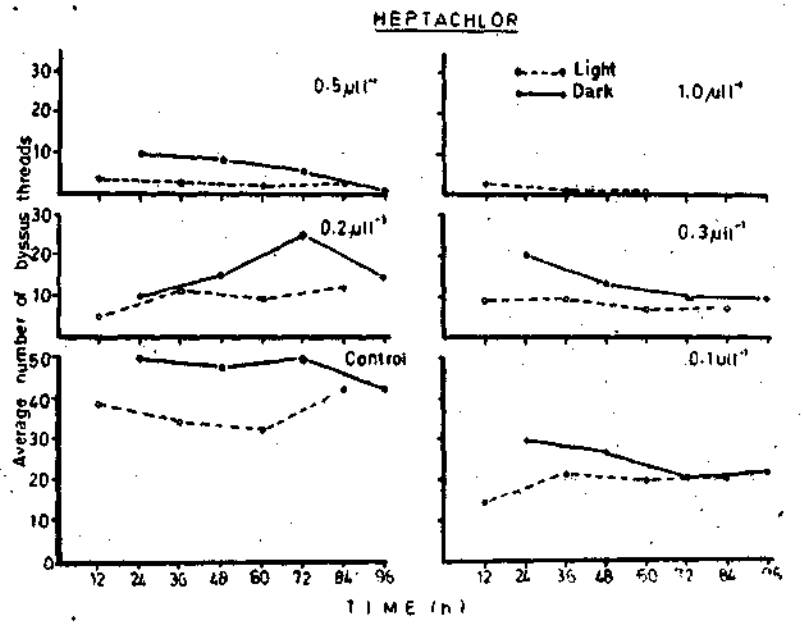

Fig. 10. Byssogenesis as a function of exposure to Heptachlor.

estuarine species and organochlorine conducted by Eisler (1969, 1970a) showed that in the case of *Crangon septumspinosa*, 96h-LC50 was 8 $\mu g\ l^{-1}$ whereas in the case of a fish *Thellasoma bifaciata* it was 0.8 $\mu g\ l^{-1}$. Discussing on the results obtained from his studies, Schimmel *et al.* (1976) opined that the extreme toxicity of Heptachlor represents a potential dangerous situation. Oysters have been found to have the capacity to accumulate very high quantities of Heptachlor, the concentration factor could be as high as 21,300 times. Since the organochlorine pesticides are lipophilic, it is very likely that tissues which contain more of fat accumulate them. Discussing on the effects of Endosulfan on *Mytilus edulis*, Roberts (1975) remarked that assimilation of Endosulfan could result from direct absorption across exposed tissue surfaces or when the pesticide is ingested through particulate matter. It is possible that in the case of bivalves, the path of entry is the gill as a consequence of absorption on the gill surface and subsequent absorption. Although, the addition of low quantities of Heptachlor did *not* colour the test medium, the brownish hue of the mucus and pseudofaeces indicates high rate of adsorption of this pesticide.

The rate of byssus-thread production is influenced by various environmental factors. These include water movement, salinity, position occupied by the mussels in the intertidal region, dissolved oxygen concentration, temperature, oil, detergents and mercury salts (Roberts, 1975). Under ammonia and ammonium stress the byssus threads produced lacked hardening of the surface and resulted in the inability of the discs to attach firmly. Continuous production of byssus threads was stopped well below the LD-50 96h values obtained for ammonia and ammonium. This indicates that although the mussels continue to survive in sub-lethal concentration the byssus production might either be reduced or suspended which will have important ecological significance on the mussel population.

Although *Perna viridis* could tolerate higher concentrations of LDO and HSD-WSF, they could not produce byssus threads at such concentrations. Dunning and Major (1974) found that *M. edulis* did not produce threads in 12% ESSO-extract, although the mussels survived. These authors maintain that prevention of byssus secretion is obviously detrimental to the survival of intertidal organisms exposed to tidal action. The discs of attachment were also not properly developed in concentrations where the mussels secreted byssus in the present instance.

Roberts (1975) found that 50% reduction in byssal attachment occurs in the presence of 0.45 mg l^{-1} of Endosulfan. According to him, the reduction in byssal attachment is owing to the impairment of pedal gland activity. In the present instance even the presence of 0.1 $\mu l\ l^{-1}$ of Heptachlor resulted in reducing byssus secretion by 50%. The byssus secretion is controlled by at least four factors, such as collection of secretions from the pedal gland, rapid molting of threads, subsequent tanning and mechanical action of the foot (Bairati and Vitellaro, 1974). Any pollutant that could influence anyone of the above reactions is bound to influence the rate of byssogenesis and the structure of the byssus threads.

The present studies have indicated that while defining safe concentrations of pollutants with reference to *Perna viridis*, detailed studies on acute and sub-acute toxicity should be conducted. Only when information on these aspects is available, a meaningful standard could be fixed for any pollutant that is likely to be discharged into or near an area where *P. viridis* is inhabiting or is being cultured.

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HEAVY METAL CONCENTRATION IN THE LAMELLIBRANCH *PERNA VIRIDIS*

GODWIN WESLEY AND P. J. SANJEEVA RAJ
Department of Zoology, Madras Christian College, Tambaram, Madras-600 059, India

ABSTRACT

Concentrations of heavy metals like Cu, Co, Fe, Mn, N, Pb and Cr in the various tissues like the gills, foot, gonad, adductor muscle, liver and shell of the green mussel *Perna viridis*, around the Madras Atomic Power Plant, Kalpakkam have been bioassayed. The increasing or decreasing order of concentrations of these heavy metals in the individual tissues and their significance in human health physics as well as in coastal aquaculture of mussels are discussed.

INTRODUCTION

IT has been known for many years that concentration of heavy metals is significantly higher in the marine biosphere than in the hydrosphere (Noddack and Noddack, 1939; Clarke and Wheeler, 1922). Heavy metals in trace amounts are normal constituents of marine organisms and some of them such as zinc, copper and cobalt are essential for animals for their normal growth and development. However, at sufficiently high concentrations heavy metals are toxic to organisms and their consumers at higher trophic levels including man and hence it is important to know the cumulative nature of their concentration above the normal range in the environment and the biosphere.

Several species of bivalves have been used in attempts to assess or bioassay the concentration of stable elements as well as unstable elements, as reviewed by Phillips (1977). In spite of the several recent studies, the mechanisms whereby trace elements are incorporated into the marine organisms are still not fully understood. Earlier work has been summarised by Vinogradov (1953), but his review covers only the gross analysis of a few individual organisms and also some of the work referred to was based on questionable experimental techniques. Later work was concerned with the role of these organisms, as indicators for the concentration of heavy metals (Folson *et al.*, 1963; Mauchline *et al.*, 1964, Bryan *et al.*, 1966, Templeton and Preston, 1966; Young and Folsom, 1976). Bivalves accumulate both radioactive and non-radioactive trace metals with the concentration factor of 10^3 to more than 10^6, depending on the tissue of the species and the metal concerned. These organisms are filter-feeders and thus obtain trace metals not only through their food and the surrounding solution, but also through incorporation of inorganic particulate material by osmosis (Moore, 1971). However, the exact proportion of the gross trace elements of the whole body derived from each of these three routes in bivalves is still uncertain. Bioassay of mussels (*Mytilus edulis*) for the uptake of both radionuclids and stable metals from the fluid medium without any feeding in the laboratory, reveals rather lower concentrations than those observed in mussels from the natural

environment (Pentreath, 1973; Phillips, 1976), thereby suggesting the uptake through food to be the most important route.

Our thanks are due to Mr. M.A.R. Iyengar and to Mr. K.V.K. Nair, Environmental Survey Laboratory, Kalpakkam for providing the necessary facilities and for loaning the relevant literature. One of us (G.W.) is grateful to the Indian Council of Medical Research for the award of a Junior Research Fellowship during the tenure of which this work has been carried out.

Material and Methods

Specimens of *Perna viridis*, 7.0-12.0 cm long, were collected from the intertidal area of the coast of the Bay of Bengal at Kalpakkam, Tamil Nadu (12°33° N and 80°11° E). The animals were washed in sea water and the epiphytes and epizoic forms were scraped off. The shell was opened by gentle heating under an infrared lamp. About 25 specimens were dissected and each type of tissue was pooled to get about 3-5 g by dry weight. The tissues — gill, foot, gonad, adductor muscle and liver — were washed with demineralised water and dried in a hot-air oven at 120°C for about 24 hours. The dry materials were digested in concentrated nitric acid to avoid possible trace element evaporation (Doshi *et al.*, 1969). The digested material of the desired tissue was diluted at known concentrations and analysed by atomic absorption spectrometry.

Results and Discussion

The concentrations of different elements in the five different soft tissues and the shell are expressed as μg/g of the dry tissue concerned (Table 1).

Copper: Copper is one of the most toxic metals and the ability of some bivalves to concentrate this element to several times the concentration present in the surrounding sea water is of considerable significance. Cu^{++} has an important biochemical role as an enzyme activator, as a constituent of flavoprotein and in combination with protein in the form of the haemocyanin present in the haemolymph of certain molluscs, crustaceans and arthropods. It is not certain whether any bivalve has haemocyanin, because in most species there is no oxygen carrier inside the organisms (Morton, 1958).

TABLE 1. *Concentration of selected trace metals in the green mussel* Perna Viridis. *All values are μg/g (ppm) in dry tissues*

Tissues	Cu	Co	Fe	Mn	Ni	Pb	Cr	Zn
Gill ..	12.15	3.03	416.80	91.14	6.58	10.13	40.55	48.2
Foot ..	5.43	0.90	168.25	19.90	2.26	5.43	BDL	29.49
Gonad ..	7.64	0.99	68.39	25.56	1.66	4.32	BDL	47.33
Adductor muscle ..	2.55	0.86	121.49	7.22	2.55	4.25	BDL	58.45
Liver ..	7.75	1.36	57.48	17.79	2.28	4.56	BDL	33.95
Shell ..	5.09	10.18	18.91	6.91	10.91	53.81	12.36	19.12

BDL: Below detection limit.

The concentration of copper in the five different tissues of the mussels show marked differences. The gills show high amounts of Cu^{++} whereas the other soft tissues show lower concentrations which is consistent with the findings of Korringa (1952) and with those of Nair and Anderson (1977) who have noted the concentration of positive polyvalent ions such

as Al^{+++}, Cu^{++}, Fe^{++}, Zn^{++}, Hg^{++} and Mn^{++} in the mucus sheets of oyster. Next higher concentration of Cu^{++} was seen in the liver and foot and very low concentration was recorded in the adductor muscle. Liver also is known to accumulate Cu^{++} considerably (Vinogradov, 1953).

Zinc : Zinc is widely distributed among different organs and has a great importance in biochemical systems, being an enzyme activator and constituent of several important metalloprotein enzymes such as carbonic anhydrase and carboxypeptide (vallee, 1963; Schelske, 1964). Nair *et al.* (1969) found that bivalves tend to concentrate Zn^{65} to a very great extent. The data presented here shows higher concentration of Zn^{++} in the adductor muscle, followed by the gills and the gonad. Compared to Cu^{++} the concentration of Zn^{++} was very high in *Perna viridis*. Nair and Anderson (1977) observed that the highest concentration was in the visceral mass and then in the gills of *Mytilus edulis* from the Norwegian waters.

Manganese : Studies on manganese in sea water were carried out by Harvey (1949) and it is noted that most of the manganese in sea water exists in particulate form. Goldberg (1957) assumed that particulate manganese dioxide plays its role as a chemical scavenger in the sedimentary processes. Since the bivalves are filter feeders, there is every possibility of manganese incorporated sedimentary particles enteringinto the system and getting concentrated. In the present study very high Mn^{++} concentration was noted in the gills and very low concentration in the adductor muscle. It may be due to the suspended particles getting into the gills during filtration. Next to gills, gonads appear to concentrate more Mn^{++}.

Lead : Lead is a cumulative cellular poison. This trace element has rather an obscure biochemical role. The variable levels of this element in individual organisms is difficult to explain. However Pb^{++} is strongly basic, it will readily coordinate with suitable organic ligants (Brooke and Rumsby, 1965). In the present study shell showed higher concentration of lead than soft tissues. Lead like other metals studied, was concentrated in the gills to a higher extent compared to the other soft tissues, followed in the order of abundance by the foot. liver and gonad. Similar results were reported from the Norwegian waters (Nair and Anderson, 1977).

Iron, Nickel and Chromium : Favoured sites of these elements of the iron-family are the gills, foot and the adductor muscle. It is considered that this is mainly due to ingestion of sedimentary materials of small particle size. Chromium concentration was below detection level in the foot, gonad, adductor muscle and liver. Of these three elements, iron concentration was higher than those of Mn^{++} and Cr^{++}.

The role of Cr^{++} in biochemical systems is ill established but this metal also has been shown to restore activity of the metal-free carboxypeptidase, as seen in the activity of phosphoglucomutase (Vallee *et al.*, 1960; Strickland, 1949). Cr^{++51} is shown to bind to the protein position of the haemoglobin molecule (Chernoff, 1961; Heisterkamp and Ebaugh, 1962). It is likely that the high levels of Cr^{++} in the gills observed in the present work may be due to contamination by sedimentary materials.

Nickel also has been shown to activate certain enzymes such as arginase (Greenberg, 1960), certain carboxylases (Weissbach *et al.*, 1956) and to restore activity to metal-free carboxypeptidase (Vallee *et al.*, 1958). In the present study, high concentration of nickel was seen in the gills, liver, foot and gonad, in order of their abundance.

Cobalt : The uptake of Co^{58} and Co^{60} by aquatic organisms under laboratory conditions have been well studied. Patel and Ganguly (1968) discussing the concept of acute and

chronic tissue concentration of elements, observed that chronic concentration factors (10^3-10^5) were significantly higher for cobalt than acute concentration factor (10^2) as for other transition elements also of metabolic significance. In the present study, Co^{++} concentration was higher in the gills and considerably higher in the shell also, but poor in the adductor muscle.

The following is the order of ion concentrations in the different tissues of *Perna viridis* from the Kalpakkam waters.

Gill	Fe > Mn > Zn > Cr > Cu > Pb > Ni > Co	
Foot	Fe > Zn > Mn > Pb = Cu > Ni > Co > Cr	
Gonad	Fe > Zn > Mn > Cu > Pb > Ni > Co > Cr	
Adductor muscle	Fe > Zn > Mn > Pb = Ni > Cu > Co > Cr	
Liver	Fe > Zn > Mn > Cu > Pb > Ni > Co > Cr	and
Shell	Pb > Zn > Fe > Cr > Ni > Co > Mn > Cu	

Conclusion

In the present study lamellibranch gills seem to concentrate almost all the elements considered here and that too in higher concentration. Liver and gonads concentrate these elements in lesser magnitudes. Similarly foot and adductor muscle also showed similar concentration but only zinc is more concentrated in adductor muscle. Adductor muscle concentrates the other elements at comparatively low levels. Shell concentrates lead to a greater extent but the copper content was poor.

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OVER-VIEW OF FISH AND SHELLFISH DISEASES PROBLEM AND THEIR CONTROL IN MARICULTURE

S. MAHADEVAN

*Central Marine Fisheries Research Institute, Cochin-682 018**

ABSTRACT

Culture practices of brackishwater prawns, marine fishes, mussels, oysters and crabs are rapidly expanding in India. Side by side with the advances in this line it is very essential to bestow our attention to identify and control any organism co-existing and affecting the cultured crop in any given area. In this context we may have to contend with, primarily, direct threats to productivity posed by the detrimental effects of cohabiting organisms belonging to three basic groups (a) competitors (b) predators and (c) disease and parasitism. Problems have manifested themselves in early mariculture ventures providing some insight into the potentially more serious problems that may be expected in large-scale mariculture. The premis that disease and parasite control must be assigned high priority in the development of mariculture is supported by examples all over the world. Documenting symptoms, isolating pathogens, and studying them, developing preventive and curative measures are all some of the works that are to be looked into in addition to studying disease vectors, environmental factors predisposing an organism to vulnerability or favour disease resistance.

Thus there is a strong case for starting fish pathology division to tackle the above problems in India where little work has been so far in progress on marine fish and shell fish diseases.

INTRODUCTION

FISH and shell fish culturists all over the world are quite often confronted with problems of large-scale mortality of the tended stock due to diseases, predation and ecological variations and environmental stress. Considerable work has been done in the developed countries in this realm. Sindermann (1970) has reviewed the work. Considering the voluminous literature on the incidence of fish diseases amongst fresh-water fishes and the marine fishes in the wild, very little information is available on the fishes and shellfishes subjected to marine farming. Southeast and East Asia is the world's main source of aquaculture products both for food and ornamental purposes. Even in this region what little information on fish diseases that is available concerns the freshwater forms and there is a big gap to be filled up in respect of the diseases of marine fishes, molluscs and crustaceans. Due to the lack of knowledge and understanding of the effects of diseases in mariculture production in this region the industry goes on without the benefit of technical assistance in their control and treatment. This situation would continue for some more time. The effects of diseases are likely to be more and more felt due to intensification of aquaculture in many countries, including India where stress has been laid on mariculture. Raising of many species of marine teleosts, molluscs and crustaceans in brackishwater, estuarine and bay-water environment, has been satisfactorily developed and demonstrated. Even as we develop techniques of production it is imperative to evolve methods to solve problems

* Present address : Regional Centre, Central Marine Fisheries Research Institute, Mandapam Camp, Tamil Nadu.

posed by disease out-break, by documentation of different types of diseases, development of meliorative measures and eradication of causative agents by suitable techniques.

It may also be mentioned here that transport of live fish seed and ornamental fishes is also a source of spreading disease from one country to another. The world trade in ornamental fish is worth $ 4000 million a year. Efforts have been made by agencies like International Office of Epizooties (OIE) American Fisheries Society, World Health Organisation (WHO) to prevent spread of communicable disease through fish transport from one geographical area to another. Quarantine measures and regulations have been suggested by FAO (Report of FAO, 1977; Zenny, 1978) to tackle problems arising out of these. In India, except for inland fish seed transport, marine systems have not so far resorted to transport of seeds from one part of the coast to another (In recent years there has been some movement for prawn seed). Adherence to the measures and principles laid down in quarantine regulations would prevent the possibility of disease transmission from one area where particular disease may be prevalent to another area where such a possibility did not exist before.

Intensive and efficient culture of any animal requires crowding or stocking them more densely than in nature. Under these conditions there are more vulnerable to disease and fall prey to many types of diseases, directly and indirectly, reaching catastrophic proportions at times (Bardach *et al.*, 1972; Huet, 1972). This imposes the demand for knowledge about the nature and control of diseases. In an unenclosed environment the problems of culturists are of still greater magnitude.

In order to highlight the extensive damage done by various disease causing agents, the present paper reviews some of the commonly known significant diseases amongst fishes and shellfishes. Broadly, the diseases may be classified as infectious, parasitic, noncommunicable and of unknown etiology. Parasitic infestation may be endoparasitic or ectoparasitic. Infectious diseases are caused by viruses, bacteria and fungi and less frequently by algae ; other causes of debilitation and mortality include deficiencies, wounds, poisons, environmental factors such as temperature, salinity and predation.

In all the above cases the symptoms are documented, causative agents identified, pathogens isolated and studied and curative and preventive measures are sought through experimentation. By developing control in these areas it may be possible to keep down the incidence of disease and parasitism in cultures to levels at or below the rate of occurrence in the natural environment, or more to the point, to levels acceptable to a viable mariculture. The diagnosis and treatment of fish diseases have reached the highest levels of sophistication in the culture of temperate freshwater fishes. Bauer *et al.* (1973) have given an exhaustive list of literature on diseases of pond fishes. Si dermann (1966) has given a very valuable, up odate review of the diseases amongst marine fishes.

Microbial diseases

Microbial diseases are those of viral, bacterial, fungal and protozoal etiology. All these tend to destroy the tissues of the host and multiply within the host. The pathology depends on virulence of dose, resistance of individuals, infective dose, environmental variables and host nutrition. The effect may be from chronic to acute affliction leading tomortality in several cases.

Viruses

Viruses and known etiological agents for neoplastic, hyperplastic and hypertrophic diseases. Lymphocystis diseases and certain Papillemas are known to be of viral origin. Chronic fibro-epithelial tumor or ' Cauliflower disease ',

white, raised patches on skin called ' Fish-pox ' are often caused by viral infection. *Baculovirus* are known to infect the hepatopancreas of blue crabs apparently not causing overt diseases in the host. RLV (Reolike Virus) causes fatal mortality due to neurological damage. HLC (Herpeslike Virus) also causes fatalities after 40 days due to massive destruction of hemocytes. CBV (Picornalike virus) bring about death because of its predilection for epidermal tissues. The above diseases are mostly in crabs in culture system than in the wild state (Johnson, 1978). Farley (1978) reports Virus and Virus like lesions of groups pedoiridae, papoviridae, herpetoviridae, iridoviridae, togaviridae, retroviridae and reovfridae in 13 species of molluscs.

Bacterial diseases

Reports of bacterial epizooties in marine fishes are infrequent according to Sinderman (1966) due to lack of observation or inadequate examination rather than lack of occurrence. ' Red disease ' due to *Vibrio anguillarum* and *Pseudomonas* (*Aeromonas*) *punctata* leads to mortality. Bacterial dermatitis accompanied by ulcerations and fin-rot due to *Pseudomonas* sp. is also common. *Mycobacterium* spp. are known to cause spontaenous tuberculosis. *Myxobacterium* and *Chondrococcus columnaris* are known to infect gills, fins and skin. Many of these bacteria normally present in sea water or on the surface of the fish invade and cause pathological effects if fish are injured or subjected to severe environmental stress.

Gaffkemia is a fatal disease causing heavy mortality among lobsters. The causative agent is *Aeromonas viridans* var homari (Stewart, 1978) An anaerobic bacteria *Eubacterium tarantellus* if present in the C.N.S. of fishes might be a possible disease agent (Udey, 1978).

Fungi

It is stated by Sindermann (1966) that with the single exception of *Ichtyophonus hofei* fungal diseases of marine fishes are almost unknown. The infection often results in necrosis and mortality of the invaded stock.

Protozoa

Sporozoa and cindospora are among the best known serious pathogens of marine fishes. In addition hemo-flagellates and ciliates are also known to affect the fish stock. Coccidia, myxosporidea and microsporidea bring out severe effects on hosts causing nerve and muscle degeneration and Castration. *Eimeria* spp. and haemogregarines have been studied in detail among Coccidia. Diseases caused by myxosporidea are common among marine fishes (also freshwater fishes). The afflicted fishes show wormy or mushy or jellied condition or milkiness. Those afflicted in the gall bladders show enlargement and discolouration of the organ. Myxosporidea infection in somatic muscles cause extensive damage. *Hannenquya salminicola* invading the flesh causes white cysts formation called ' tapioca disease '. Gill invading myxosporidea are not of any serious consequences to marine fishes. *Myxosolus* sp. and *Myxosoma* spp. affliction do not appear to result in mortality although they cause hypertrophy and ' whirling disease ' respectively. Several serious diseases result from microsporidian invasion. *Glugea* spp. are known to produce visceral involvement preventing reproductive activities. Some destroy digestive tract and impair metabolic functions. Occasionally the host dies where infection is serious. Another microsporidea *Nosema* sp. produce tumor like cysts in the C.N.S. in the host. *Pleistiphora* sp. produces intramuscular cysts reducing the marketability the fish because of the bulges noticed on the host. Among microsporidians *Ameson* sp., *Chapmanium* sp., *Agmasoma* sp. and *Indosporus* sp. have all been reported as parasitic on prawns and crabs (Sprague, 1978). Sprague (1978) also reports *Paramoeba perniciosa* causing serious mortality among molluscs.

Parasitic diseases

Diseases considered under this are those caused by helminths (trematodes, cestodes, nematodes, acanthocephalans) and parasitic copepods. These are non-multiplicative in the host after invasion. Helminths, as larval infection, are of great significance. Adults occur in digestive tracts but the larvae are found in the viscera. Growth retardation, tissue disruption, metabolic disturbances and mortality of hosts in serious infestations are characteristic of the helminth invasion of the host.

Digenetic trematodes occur in digestive tracts and the migrating cercariae and metacercariae of these encyst beneath the body of fish hosts as pigmented cysts-as ' black spots '. At times larval fishes infested by these die. Adult digenetic trematodes, do not cause serious disease. Monogenetic trematodes parasitic on gills and body surfaces many become serious depending on optimum conditions. *Benedenia* and *Axine* infestation are also known leading to ematiation and making the fish vulnerable to bacterial dermatitis. *Echinostephanus* sp. and *Tormopsolus* infest intestine. In natural conditions they are less of a serious threat. *Gyrodactylus* spp. cause mortality. Adult cestodes are also not uncommon and are harmful to digestive tract but the larval stages are the most dangerous. Nematode larvae infesting the flesh and viscera reduce the commercial value of the hosts. Effects of *Parrocaecum* spp. and *Contracaecum* sp. *Cucullanellus* sp. and *Eustoma* sp. have been reported. The ' spiny-headed ' acanthocephalans are represented as adults and larvae in marine fishes. *Echinorhynchus* sp. *Telosentis* sp. *Pamphorhynchus* sp. are reported from them.

Parasitic copepods

Several species of ectoparasitic copepods *Caligus* and *Lernaeopoda* on gills, buccal cavity and fins are known. Members of Lernaeoceridae, Penellidae and Sphyriidae families are known to be particularly serious to the host. *Lernaescera* spp. are parasitic on gill region and heart which causes loss in weight, anaemia of fish which kills the host. *Sphyrion* sp. causes ulceration and death. *Lernaecenicus* sp. (Pavillon) is another parasite of less consequence. *Panella* sp. causes cysts in the tissues.

Protozoan, helminth and copepod parasities of fishes although not often direct cause of death, can act to weaken the host and help the entry of secondary invaders leading to mortality on large-scale. The fishes so infested often fall prey to predators because of their inability to escape. Environmental variations also kill them on account of lack of resistance.

Dinoflagellates also cause diseases. The gill parasite *Amylodinium* sp. causes velvet disease leading to mortality. *Haematodinium* spp. is parasitic on crabs (Sawyer and Maclean, 1978).

A ciliate parasite *Cryptocaryon* sp. causes ' white spot disease ' on gills and epidermis. Several ciliates *Anbiphyra*, *Epistylis*, *Chilonodella* and *Trichodina* can cause mortality under certain conditions (Rogers, 1978). The flagellate *Costia* and *Bodomonas* can cause mortality, although easily controllable in early stages.

Two algae *Leucosphaera* and *Thallamobella* are responsible for epizootics and mortalities in many fishes.

Diseases among molluscs, lobsters and prawns

Examples of epizootic disease and parasitism have occurred in cultivated molluscs. An important factor in the diseases of cultured shellfish population is the introduction and spread of pathogens through the transfer of seed stock to growing areas especially in oysters. In Europe the effects and spread of *Mytilicola intestinalis*, a parasite invading

cultured mussel stocks, has been documented from very early years. In Japan, evidence of disease in oysters, both bottom grown and suspended stocks, has been documented. On the west coast of U.S., three diseases are known in oysters — a parasitic copepod (*Mytilicola orientalis*), bacterial and an amoeboid organism. On the east coast of U.S. four diseases predominate — *Dermocystidium marinum*, a fungal attack, Malpeque Bay disease in Canada, Sea-side disease (haplosporidian disease by *Minchinia costalis*). Mass mortality of bivalve larvae by gram negative bacilli *Vibrio* and *Aeromonas* sp. has been recorded. Other epizootics is caused by *Sirolpodium zoopthorum* a fungus killing the larvae and juveniles.

In the case of lobsters pathogens like *Pythium* affect the stock. Mycosis is caused by *Aphanomyces* sp. in European freshwater cray fish (Brown spot disease). Amongst prawns cultured in brackishwater masses chondrococcus infection (*Flexibactor* sp.), Haemorrhagic septicemia by *Pseudomonas*, vibriosis by *Vibrio anguillarum*, enteric bacterial infection by *Escheriella coli* and soft shell infection (unknown reason) have been reported in India (Mahadevan *et al.*, 1978).

Remarks

The review does not detail the disease noticed amongst freshwater fishes cultured in India. Because of the enormity of marine ecosystem, disease problems are also varied and many. Continuity of observations on diseases to document the diseases is essential. Delmendo (1978) gives a list of diseases documented in the Indo-Pacific region. Amlacher (1970) quoted by Delmendo (1978) has given a detailed account of treatment procedures of certain known diseases. A comparative statement of disease agents and prevalence in Southeast Asian countries is found in a recent publication of the International Development Research Centre, Canada, summarising the outcome of a workshop on fish disease held in Indonesia in 1978. Developing countries should strengthen their capabilities in controlling fish diseases. Diagnosis and treatment of fish requires specialized training and equipment. Manpower to provide this should also be developed.

Our present knowledge of marine fish diseases is rudimentary and needs to be strengthened in the years ahead. Already a start has been made by the Central Marine Fisheries Research Institute in 1976 to investigate the diseases common among the prawns, mussels oysters and eels.

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INVESTIGATIONS ON PARASITIC CILIATES (PROTOZOA) OF CERTAIN SHELLFISHES IN THE HOOGHLY-MATLA ESTUARINE COMPLEX

YUSUF ALI JAMADAR AND AMALESH CHOUDHURY*

Susama Devichoudhurani Marine Biological Research Institute, P.O. Baman Khali, Sagar Island, 24 Parganas, West Bengal

ABSTRACT

The present paper deals with the results of six years' investigation on parasitic ciliates in the mollusc population of the mangrove swamps in the deltaic Hooghly-Matla Estuarine complex of West Bengal, particularly around Sagar Island. Many gastropods and pelecypods inhabiting the area are found to be infested with hymenostomatid, thigmotrichid and peritrichid ciliates. The parasitic ciliates belong to the genera *Cristigera, Ancistrocoma, Raabella, Ancistrumina, Fenchella, Protophrya, Boveria, Nucleocorbula, Scyphidia* and *Trichodina* are found to prefer *Crassostrea cucullata, Mactra luzonica, Meretrix meretrix, Donax lubricus, Modiolus (Modiolus) striatulus, Cerithidea obtusa, Littorina melanostoma, L. scabra scabra* and *Glaucomya sculpta* as hosts. The pattern of association between the parasites and their respective molluscan hosts are described and discussed.

INTRODUCTION

MULLER (1788) was the first to describe a ciliate *Tricoda ciliata* in *Mytilus edulis*. Later Ehrenberg (1838) contributed his monumental taxonomic work during the first half of the nineteenth century. He described a big ciliate named *Leucophyrs anodontae* from *Anodonta* of the river Ob-Siberia. Stein (1861) created for it the genus *Conchophthirus* and described the second species of this genus *C. steenstrupi* from the slime of terrestrial *Pulmonata*. Quennerstedt (1867) described a ciliate *Opalina mytili* from *Mytilus edulis* from the west coast of Sweden. Maupas (1883) found this and another species of the same genus in *Mytilus edulis* and *Venus striatula* respectively, from the Mediterranean and erected the genus *Ancistrum* for them.

Issel's memoir (1903) is the first comprehensive systematic investigation on ciliates from lamellibranchs and gastropods. He found and described several species of *Ancistrum* and *Boveria* and a genus *Plagiospira*. Pickard (1927) created for *Boveria* a new family Boveriidae and included it under the order *Heterotricha*.

Chatton and Lwoff (1923, 1949, 1950) published a long series of papers on the ciliate fauna of molluscs which were included in a memoir on the morphology and systematics of the thigmotrich ciliates.

The extensive work of Raabe (1967, 1970), Jarocki (1934, 1935), Jarocki and Raabe (1932), Ridder (1933, 1934), Uyemura (1937), Antipa (1971), Antipa and Small (1971) Kozloff (1946), Kazubski (1958), Corliss (1961, 1977), Khan (1970), Hatzidimitriou and Berger (1977) have enriched our knowledge on thigmotrich and other ciliates considerably.

From the Indian sub-continent, Ghosh (1922), Chakraborty (1936) Ganapati and Nagabhushanam (1955) and Santhakumari and Nair

*Present address: Department of Zoology, University of Calcutta, 35 Ballygunge Circular Road, Calcutta-700019.

(1970, 1973) found a total of 14 determined species. The sole intent of this paper is to elucidate about the ciliate parasites of commercially important shellfishes from the Gangetic deltaic regions of Sunderbans.

According to Sprague (1969), the disease agents are a significant part of the marine environment. It is to be hoped that this fact will soon be given adequate recognition by individuals and agencies who are becoming increasingly concerned with the environment as it relates to human interests.

Materials and Methods

Host materials were collected from the mangrove swamps of deltaic Sunderbans of the Hooghly-Matla Estuarine complex. Living ciliates were examined and studied under microscope in fresh smears from the respective organs diluted with 0.5% saline solution or with body fluid. Sometimes these were also anaesthetised with 0.4% $NiSO_4$ solution (Bovee, 1958) and examined underphase contrast or Carl Zeiss Plankton microscope. Semidried smears containing the ciliates were fixed in Schaudinn's fluid (about 60°C). The ciliates were, in addition to nuclear stain, impregnated by $Ag\ No_3$ (Klein's dry method). Sections of the host tissues, to demonstrate different ciliate's preferred microhabitats, were studied by the application of usual histochemical techniques.

Observations

Order : Hymenostomatida Delage and Herouard, 1896

The ciliates of this order have ventrally located buccal cavity with an undulating membrane on right and three membranelles on left ; body ciliation uniform (Kudo, 1966).

Genus : **Cristigera** Roux, 1901

Small, 15-60 μm long ; ovoid : much compressed with a pos oral depression ; usually with refractile pellicle ; with a caudal cilium ; peristome closer to mid-ventral line ; on its right edge occurs a membrane which forms a pocket around cytostomal groove and on its left edge e ther free cilia or a membrane which unites with that on right ; no semi-circular swelling on the left side or oral region ; round macronucleus with a micronucleus ; contract le vacuole posterior ; fresh or salt water (Kudo, 1966).

But the ciliate *Cristigera susamai* n. sp. communicated in the present paper was recovered from the gills and labial palps of the bivalve host *Crassostrea cucullata* though the infected shells constitute a very insignificant part of the gregareous host population. Surrounding brackish water of the *Crassostrea* habitat has been thoroughly examined during different seasons for the ciliate but with negative results. So the ciliate under consideration is presumed to establish a sort of heterospecific association (commensalsm) with the molluscan host in contrast to the other members of the genus all of which are free living. Specific characterzations, morphometric measurements and the geographic distribution of the ciliate with a bivalve host distinguish it obviously from all the other members of the genus *Cristigera* recorded and described so far from different parts of the globe.

Order : Thigmotrichida Chatton and Lwoff, 1922

These ciliates which, according to Corliss (1961) number nearly 150 species, are almost exclusively found in bivalves. They occur in most, if not all of the more familiar marine bivalves such as species of *Mytilus, Mya, Mercenaria, Macoma, Mactra, Cranostrea*, etc. Perhaps some of them are harmless commensals on the gills or other part of the host body, but others undoubtedly irritate the tissues.

Genus : **Ancistrocoma** Chatton and Lwoff, 1926

The body is elongate, pyriform, with pointed anterior end containing a contractile suctional

tentacle for attachment to the host. Rows of cilia occur on the dorsolateral and ventral sides. They are parasitic on gills, palps and other body surfaces and sometimes invade the tissues.

A. pelseneeri C. and L., *A. thorsoni* Fenchel and *A. dissimilis* Kozloff are found in a single bivalve host *Mactra luzonica* (Deshayes). Out of a total of 33 host materials only 4 were found to be profusely infected. Pauley *et al.* (1967) pointed out, they may contribute significantly to diseased conditions. Lom and Kozloff (1968) have studied ancistrocomid ciliates including *A. myae* from *Mya arenaria*. They found a number of pathological manifestations in the host cells to which the ciliates were attached.

Genus : **Ancistrumina** Raabe, 1959

Relatively scarce ciliature and rather small dimensions of the body (20-40 μm). Number of kineties 12-30. Two adoral kineties begin at small distance from the apical suture and running backwards, make a large loop at a distance of about 1/4 from the kind body pole. The argyronemes of the naked peristomal field are scarce; only two meridional argyronemes present. Parasites of the intestine and the mantle cavity of fresh water and marine Gastropoda and Bivalvia.

This genus includes presently some 22 species to which we add one more new species *A. obtusae* from the gastropod host *Cerithidea obtusa* (Lamarck) an ancistrumid ciliate to be presented for the first time from the Indian sub-continent. The proposed new species *A. obtusae*, having a more or less globular body configuration and with 19-22 general somatic kineties, beg to differ from all other so for described members of the genus *Ancistrumina* not only on the basis of its different host species, microniche and geographically distant locale but also on the basis of various morphometric parameters.

Ancistrumina barbata (Issel, 1903) recorded from the same host from Sagar Island, more or less corroborates with the same species described from the hosts *Fusus syracusanus* L. and *Murex trunculus* L. and the number of kineties falls within the range given by the earlier author.

Genus : **Fenchelia** Raabe, 1970

Elongated, slightly flattened body. Number of kineties, about 26. Two adoral kineties start about 1/3 of the body length from the anterior pole and make a big loop near the hind body pole. On the dorsal side the body continues posteriad in a cone-shaped prolongation. Parasites of the mantle cavity of marine Mollusca.

Raabe was inspired to establish this new genus in order to separate the species *Ancistrum crassum* Fenchel, 1965 which he considered as divergent from other species of the genus *Ancistrum* and *Ancistrumina* in a sufficient degree. In the present paper we add two more species under the genus *Fenchelia* which are claimed to be new to science. These are *F. sagarica* and *F. kapili* inhabiting mantle cavity and ctenidium respectively of the gastropod host *Cerithidea obtusa* (Lamarck). Though they have common host they differ from each other in their number of kineties and preference of microniche. Because *F. kapili* lives in between the ciliated epithelium of the host's ctenidium, its thigmotactic field on the left side bears cilia (8 μm) for attachment in comparison to those (5 μm) of *F. sagarica* which prefers mantle cavity as its microhabitat.

Genus : **Protophrya** Kofoid, 1903 Emend, Chatton and Lwoff, 1949

Strongly flattened body and of oval outline; the left body side concave. The ciliature dense, the number of kineties about 65. Both adoral kineties begin at a level of 1/3 of the body length from the apical end and form

a small arc in the hind part of the body. The naked peristomal field is very narrow.

This genus was first established by C. A. Kofoid in 1903 based on the solitary species *P. ovicola* described by him in the same year from the brood sac of *Littorina saxatilis* from New Port, North America. Later, Cepede (1910), Chatton and Lwoff (1949) and Fenchel (1965) found this species with little variations from several gastropod hosts of the genus *Littorina* from different parts of Europe.

The proposed new protophrian ciliate parasitises the mantle cavity and buccal mass of gastropod molluscs *Littorina* (*Littorinopsis*) *scabra scabra* inhabiting the rocky platforms of Indian coastal waters and *L. melanostoma* living in mangrove swamps of Sunderbans. As it is evident from the mensural data, the ciliate group occurring in *L.* (*L.*) *scabra scabra* is characterized by smaller body dimensions and smaller number of kineties, whereas the other group from *L. melanostoma* displays greater body dimension and greater number of kineties. Comparative morphometric measurements reveal that both these groups of ciliates while maintaining the general body configurations of the genus, as described by Raabe (1970), fall short of *P. ovicola* in size (88-102 μm $\times$ 71-77 μm) and macronucleus size in the Indian species is proportionately bigger than that of *P. ovicola* in relation to their respective body size. We suggest that the two groups of ciliates occurring in two different Indian hosts showing variations in their mensural data, belong to one species, *P. indica* n. sp. and represent merely its ecologic variability.

Genus : **Boveria** Stevens, 1901

Pear-shaped often strongly elongated body. The ciliature not abundant, number of kineties 20 to 31. Long adoral cilia. Two adoral kineties begin in the vicinity of the enlarged, posterior body pole and make around it a large, involutive spiral, lying then perpendicularly to the kineties of the general ciliature. Parasites of the mantle cavity of marine Bivalvia and of the respiratory organ of Holothuroidea.

Issel (1903), Ikeda and Ozaki (1918), Nelson (1923), Pickard (1927), Levinson (1941) and Chatton and Lwoff (1949) have found and described several *Boveria* species from holothurians and lamellibranchs.

Boveria teredinidi Nelson, 1923 has also been examined by the present authors from two different lamellibranch hosts *Mactra luzonica* (Deshayes) and *Donax lubricus* Hanely. A morphometric comparison of the Indian materials with those of the Atlantic Coast reveals that there are some obvious differences in the morphometric measurements and number of kineties but as regards the adoral ciliary lines, making a double dexiotropic spiral about the peristomal field and the distal leiotropic loop of half a spiral, more or less corroborate with those of Pickard's observations. The differences are presumably due to wide geographical isolation and new host and new environmental interactions.

ORDER : PERITRICHIDA Stein, 1859

These ciliates have a conspicuous oval ciliature but usually none on the body of the adult. They are often sessile and often colonial.

Genus : **Scyphidia** Dujardin, 1841

Cylindrical, posterior end attached to submerged objects or aquatic animals ; body usually cross-striated ; fresh or salt water.

Scyphidia (*Gerda*) *ubicuita* Hirshfield, 1949 has been recorded from two new gastropod hosts *L. melanostoma* and *L.* (*Littorinopsis*) *scabra scabra*. The two hosts show peculira nich preference, the former is exclusively foliage living avoiding inundation during high tide and the latter prefers tree-trunks or solid substratum periodically enjoying inundation. The

two populations of *S.* (*Gerda*) *ubiquita* examined from two different gastropod hosts display some variability in their morphometric measurements but they belong to the same species and the variabilities are possibly due to different host and habitat exposures.

Scyphidia (*Gerda*) *bengalensis* n. sp. is unique in its configuration and nuclear pattern and dimension in particular. The scopular base is very small is comparison to the body size and is firmly adhered to the surface of its host. It has been observed by Lom and Corliss (1968) that the permanent fixation to a host is uniformly assured by secreted sticky substances, either in the form of simple agglomerations of a mucus material at the posterior end of the body as in *S.* (*Gerda*) *ubiquita* or by a scopular cilia as in *S. inclinata* Lom and Corliss, 1968.

Genus : **Trichodina** Ehrenberg, 1838

Body barrel-shaped ; with well developed adhesive basal disk and a skeletal ring with radially arranged denticles composed of distally projecting blades and medially extending spines ; adoral ciliary row spirals one to three times ; without cirri ; commensals on, or parasitic in aquatic animals.

T. gangetica n. sp. is a first record in bivalve host *Modiolus* (*Modiolus*) *striatulus* from the Indian subcontinent. The epizoic trichodinid prefer the pulps and gills of this bivalve host to which they remain attached with the help of their adhesive discs. So far fifteen trichodinid species have been reported from various mollusc hosts.

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SOME PATHOLOGICAL ASPECTS AKIN TO SPONGE BORING IN MOLLUSCAN SHELLS

P. A. THOMAS*

Central Marine Fisheries Research Institute, Cochin-682 018

ABSTRACT

Boring sponges are considered to be a menace to the oyster beds and coral reefs in many parts of the world. In India too, where the molluscan population exists in fishable magnitude, the boring sponges pose a serious threat to their fishery.

The main target of the boring sponge is the calcareous shell of the host and the techniques adopted by the sponge to gain entry into the hard parts of the host are the same for all the species of boring sponges. But the shells of the different species react differently to the intruder, namely the sponge. These reactions often produce a wide variety of pathological symptoms in the host. Several live shells infested by boring sponges have been collected during the years 1964-1978 from both natural and artificial beds and the pathological aspects have been investigated, the results of which are presented in this paper.

INTRODUCTION

MANY species of sponges are known to bore into submerged calcareous objects like coral rocks, molluscan shells, calcareous algae, etc. A detailed survey made by the present author (Thomas, 1972, 1975) revealed the presence of 32 species of boring sponges in Indian waters and it was concluded that this is an area which harbours the maximum number of boring sponges in the world. Besides, one species (*Cliona vastifica* Hancock) which is rather common in the marine environment is unique in its distribution since it has succeeded in colonising the estuarine areas posing a serious threat to the gregarious molluscs found in the estuaries (Thomas, 1975).

Boring sponges are not considered as parasites since they obtain their food from extraneous sources (Old, 1941). The calcareous object, whether shell or coral, provides only a shelter and the ramification made by the sponge inside a living shell is liable to produce considerable physical and physiological strain on the host. Some of the common diseases found in the mollusc are reported herein.

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MATERIAL AND METHODS

Bored shells collected from the natural beds and also those found discarded in the various chank godowns were mainly utilised in the present study. Pearl oyster shells cultured both

* Present address : Vizhinjam Research Centre of C.M.F.R.I., Vizhinjam, Kerala State.

at Tuticorin and Vizhinjam were also used in evaluating the incidence and magnitude of damage caused by boring sponges on culture rafts. Spicules of sponges were prepared following standard methods and the various species were identified.

Pathological Aspects

Porosis: The primary system of sponge infection is the formation of minute chambers and interconnecting canals within the shell. and start etching out minute particles from the shell liberally (size, 0.020-0.050 mm) and form an initial chamber (Fig. 1 a). The mechanism of boring, according to Rutzler and Rieger (1973) is by a combination of chemical and enzymatic action and the filopodial basket produced by the archaeocytes at the vicinity of etching play an important role in the cutting as well as removal of these minute particles. Such particles are continuously expelled through the excurrent stream of water in a living sponge (Fig. 1 c, e). The initial chamber, thus formed, may have a

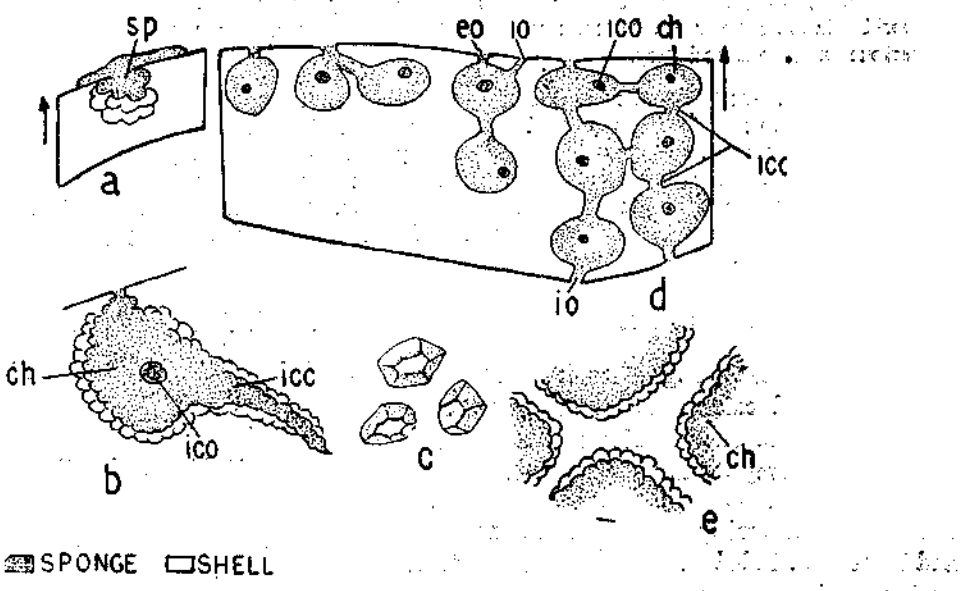

Fig. 1. a. Just attached Clionid larva gains entry into the shell (Sp : Sponge larva), b. An initial chamber (ch) is formed. Inter-chamberal connective (icc) is formed by etching out minute particles from the shell. Inter-chamberal connective opens to the adjacent chamber through inter-chamberal opening (ico), c. Particles etched out from the shell, d. Section of shell showing the arrangement of chambers inside. As growth advances more chambers are formed in different layers. The innermost chambers open to the mantle cavity through incurrent and excurrent papillae. Shell becomes porous on both surfaces and e. Chambers enlarged to show the etched out interior.

French fishermen noted this disease as early as 1823 and called it ' spice bread disease '. The chambers formed inside the shell may in one or several rows according to the thickness of the shell and are formed in the following manner. The free swimming larva, soon after settling on the shell spreads out on the surface diameter of about 1.5 mm. Further growth inside the shell is effected through branches formed from the initial chamber. The branches are formed in the same manner as the initial chamber and each branch after a short distance form a new chamber and thus the spreading of the sponge inside the shell is

effected. Each chamber communicates with the outside through two different types of papillae, the incurrent and excurrent. Both these papillae, at this stage, are directed towards the outer part of the shell and the water drawn in through the former is circulated through the canals inside the sponge and then expelled through the latter.

Monofacial and bifacial porosis: Normally when the shell is in its actively growing phase, both the incurrent and excurrent papillae open out at the outer part of the shell (monofacial, Fig. 2 a, ; Pl. I A, B). This is because every attempt by the sponge to pierce the inner surface of the shell is foiled by the mantle of the mollusc by secreting enough shell material in between. As suggested by de Laubenfels (1947) this results in a heavy drain of the oysters energy. But when the mollusc becomes physiologically weak or old, the sponge succeeds in piercing through the inner surface of the shell. At this stage the shell becomes porous on both surfaces (bifacial, Fig. 2 b, Pl. I F, G, I).

Hinge imbalance: The hinge mechanism of bivalves play an important role in the welfare of the organism. Hence, any cavity formed on the teeth or any outgrowth produced due to any repair, can considerably affect its smooth functioning. Defects of this type are rather common.

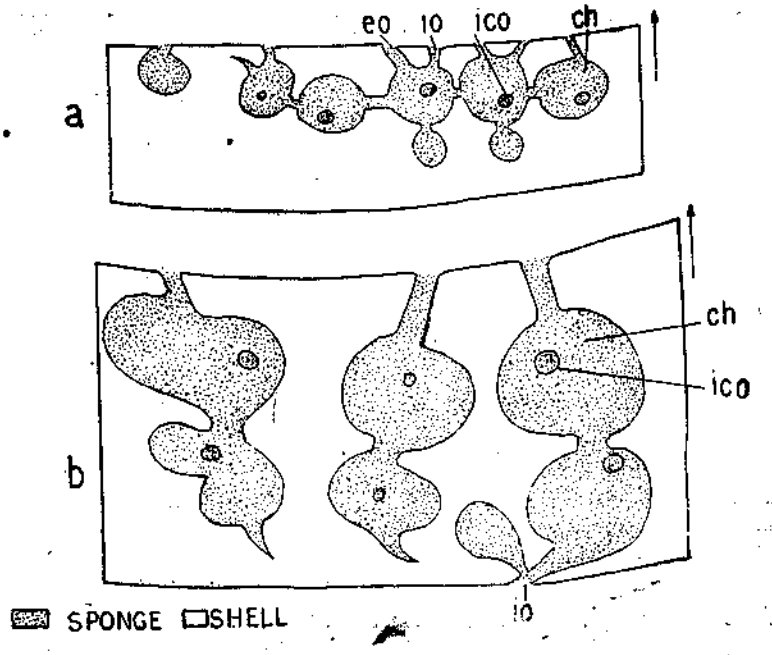

Fig. 2. a. Monofacial porosis. Both incurrent (io) and excurrent (eo) openings open towards the outer surface of the shell and b. Bifacial porosis. The inner surface of the shell becomes porous as the papillae formed from the innermost chambers pierce the inner wall.

Insertion scar imbalance: When the inroads of sponge become extensive at or near the adductor attachment zone, slightest pressure exerted by this muscle may cause complete breakdown of this area. Such a shell, which cannot open and close at will, may fall prey to other animals or may eventually die off (Pl. I G, H.). Rock oysters found in plenty along the estuaries are generally prone to this disease.

The cultured pearl oysters at Vizhinjam, exhibit another type of insertion scar damage. In these, blisters are formed at the adductor attachment zone which weaken the efficiency of adductor muscle (Pl. I E).

Fragility: Although the production of more openings on the surface of the shell is an indication of increased activity of sponge inside, merely by superficial examination alone the extent of damage caused to the shell cannot be assessed. As the inroads of sponge become extensive, more calcareous particles are expelled in the form of chips through the excurrent stream of water and this results in a gradual loss of weight on the part of the host. Chambers and interconnections lie more concentrated in the middle layers of the shell and as their number increases due to growth, the middle layers of the shell become hollow. Certain 'pillar like' structures which are the remnants of the middle layers of the shell, may be seen connecting the inner and outer layers of the shell (Fig. 3 a, b). Such shells, though externally firm and intact, may crumble at the slightest pressure.

Loss of thickness: Certain species of sponge (usually *Spirastrella* sp.) when they infect a shell, the infection may remain localised without spreading much inside the substratum. The other parts of the shell may grow in thickness, but the part infected by sponge will never increase in thickness. Bivalves with thick shells usually exhibit this symptom.

Undulosis: Undulating lines or ridges generally occur in the inner side of the shell when the infection is rather heavy. Polychaetes, mainly of the genus *Polydora*, are capable of producing such thickened ridges inside the shell. But this could be differentiated from sponge infection by its varicose nature and uniformly tubular cavity which it encloses (Pl. I C).

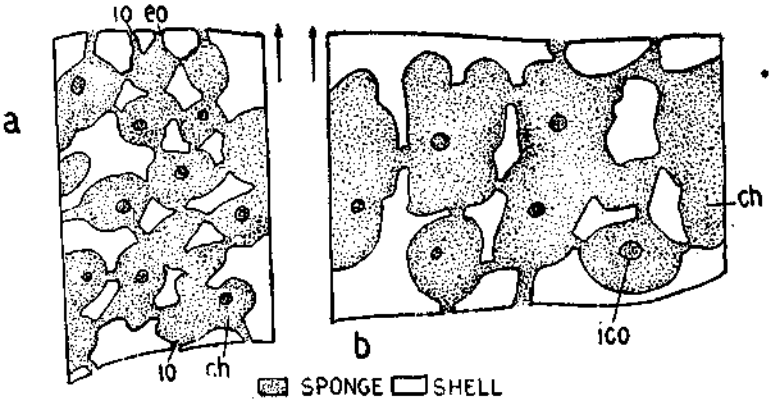

Fig. 3. Advanced stage of sponge infection in : a. Pearl oyster and b. *Xancus pyrum*. Chambers become large in the middle layers of the shell, but the outer and inner surfaces appear rather intact. Such shells get crumbled when pressure is exerted.

Atrophy: Shells possessing lateral projections or spines exhibit different grades of atrophy. Spines and lateral processes, when infected directly at their base, may even show stunted growth.

Blister formation: Blister formation is a closely associated phenomenon of sponge boring only in the case of shells possessing pearly layer. Blisters are formed inside for two purposes *viz.*, (1) to prevent the chamber formed inside the shell (by sponges) from

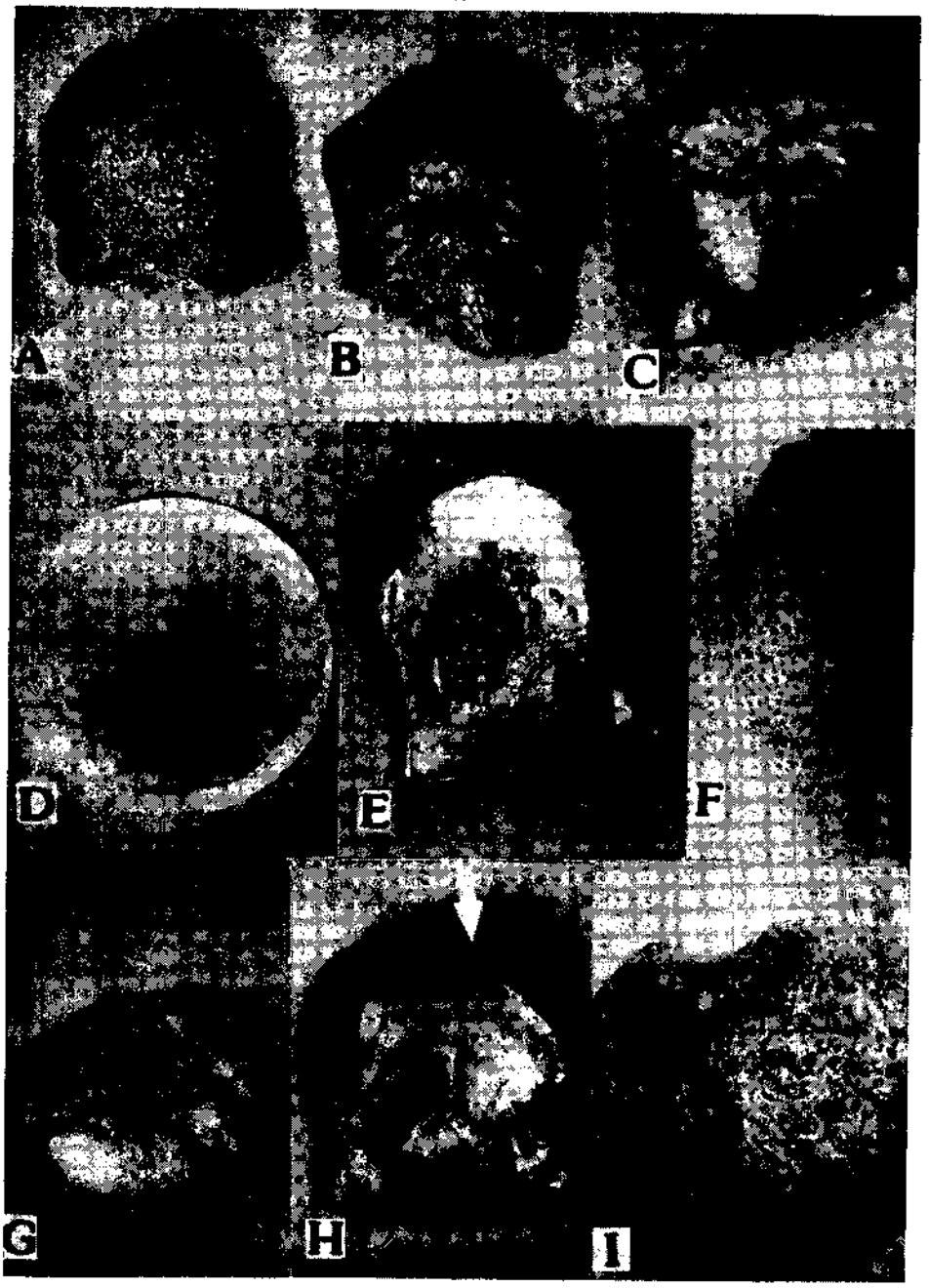

PLATE I. Pearl oyster shell infected by : A. *Cliona margaritifera*. The inroads of the sponge is seen upto the edge of the shell, B. *C. lobata*, C. Varicose nature of *Polydora* infection, D. Linear and reticulate pattern of boring noted in the Window pane oyster from Goa (Zuari Estuary). Sponge is *C. vastifica*, E. Blister formation at the adductor attachment zone (shown with an arrow). Smaller blisters seen around adductor attachment zone are those made by the repair of chambers, F. Shell of *Xancus pyrum* bored by *C. celata*. Both surfaces are uniformly bored (bifacial porosis). Larger openings are those made by *Lithophaga* sp., G. Pearl oyster shell infested by *C. margaritifera*. Inner surface is minutely pierced by sponge, H. Pearl oyster shell infested by *C. margaritifera*. The inroads of the sponge may be seen upto the edge of the shell (marked with an arrow) and I. *Crassostrea* sp. from the Zuari Estuary, Goa. Both surfaces are damaged to the maximum. Larger openings are made by boring mollusc (*Lithophaga* sp.).

contacting the soft parts of the mollusc and (2) to prevent the papillae, formed from the innermost layer of chambers, opening into the inner part of the shell. The etching and removal of calcium carbonate matter, though practically effected all along the zone of contact of sponge with the shell, is more pronounced at or near the actively growing tips only. The papillae, which are directed towards the inner side of the shell, are prevented from opening to the inside by constant secretion of nacreous layer by the mantle. But when conditions become unfavourable the process of repair becomes rather slow and the sponge again penetrates through the shell. When favourable conditions set in, some of the openings thus formed — probably the smaller ones — are again repaired and the others are left permanently open. The constant secretion of shell matter around a specified point of disturbance, thus, result in the formation of blister. Blisters may be classified into simple or compound based on the shape of the apical region (Fig. 4).

Melanosis : When the opening made by sponge at the inner surface of the shell is repaired by the nacreous material, a black patch is often formed at the site of the original pore. Apical region of the blisters usually contain such pigmented patches (melanoid blister, Fig. 4)

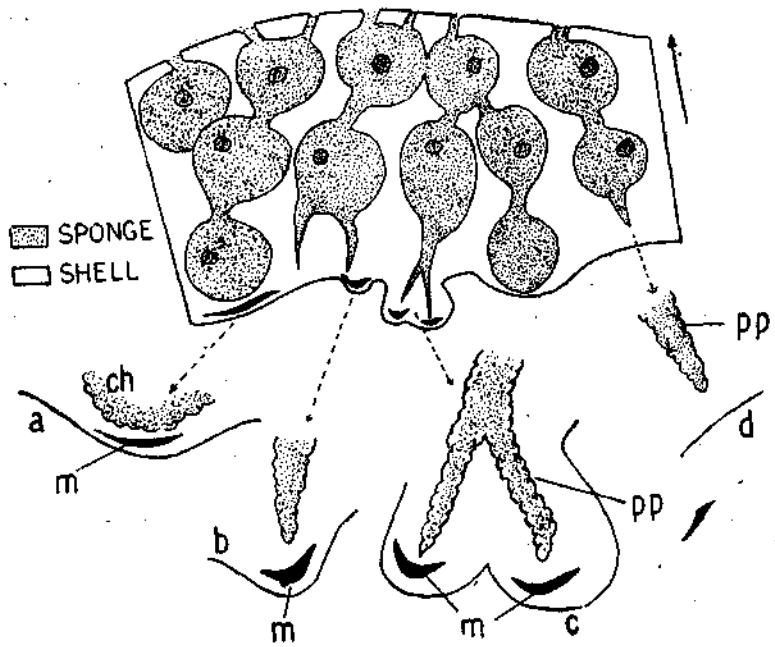

Fig. 4. Section of pearl oyster shell to show the different types of blisters : a. Blister formed by the opening of chamber, b. Blister formed by the papilla, c. Compound blister, d. A papillar projection which has not yet opened into the mantle cavity (m- black pigment ; pp- papillar projection).

Nacreerosis : Nacreous layer erosion in pearl oyster is a rare disease. Normally shells which are heavily infested by *Cliona margaritifera* and *M. lobata* exhibit this disease. As the first step, the nacreous layer becomes less lustrous followed by its erosion either partly or completely. The total damage if the nacre secreting cells on the mantle is the chief cause of this disease.

Lysis and Pustulosis : When sponge comes into contact with the soft parts of the mantle,

certain reactions are noted along the area of contact. The first sign is that the mantle becomes flabby followed by the formation of dark pigmented pustules exactly opposite the holes on the shell. The tissue often gets detached from the shell and presents a diseased look (Hornell, 1904). Warburton (1958) could note such pustules in oyster and considered that these are blood cells.

Discussion

The above mentioned are some of the ailments found in the mollusc as a result of sponge infection. The shells dealt with herein have been collected mostly from natural beds and only a few were obtained from culture rafts. Hence the data available at present are rather insufficient to compare and contrast the various diseases occurring in these two environments, their frequency of occurrence, etc. But as far as the distribution of boring sponge is concerned, these two environments show marked difference ; the salient features of which may be summarised as follows :

1. The percentage of incidence of boring sponges is considerably more on the culture rafts when compared to that in the natural beds. An incidence of 8.5% was noted in the natural beds off Tuticorin (Thomas, 1979) while that in culture rafts at Tuticorin was 20.7% (Alagarswami and Chellam, 1976) and at Vizhinjam, 33.3% (Thomas, MS).
2. The number of boring sponge species occurring in the culture raft is always greater than that in natural beds. This is effected by harbouring certain species from natural beds, and is done in two ways, (a) by harbouring certain species which are dominant in other habitats like coral reefs or even other species of mollusec and (b) by harbouring some species which remain quiescent in nature.
3. Boring sponges inhabiting the culture rafts produce more larvae as against those in natural beds. This makes their dispersal easier in an environment rich in calcium carbonate in the form of shells.

The above facts show that culture rafts provide a condition quite congenial for the boring sponges. The ecological equilibrium found in nature is disturbed in this artificial environment. More stress to the ecological aspects should be given in the culture of marine molluscs which are prone to sponge boring.

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THE RATE OF ENCRUSTATION OF BIOFOULING ORGANISMS ON EXPERIMENTAL FLOATING NET-CAGES IN TROPICAL COASTAL WATERS

S. H. CHEAH AND T. E. CHUA*

Division of Fisheries and Marine Science, University of Agriculture Malaysia, Serdang, Selangor

* *School of Biological Sciences, University of Science Malaysia, Penang*

ABSTRACT

The rate of encrustation of biofouling organisms on experimental net frames in relation to position of the frames and mesh size was studied. From the investigation on the rate of encrustation of biofouling organisms in relation to position of the frames, it was observed that the species composition in the net of both frames were similar. The initial biofoulers on both frames were similar, they being *Crypstosula*, barnacles and tubeworms. At the peak of encrustation, the net of the vertical frame was encrusted with mainly *Symplegma*, whereas the net of the horizontal frame was mainly encrusted with *Crypitosula*. Generally, the net of the vertical frame was more encrusted than that of the horizontal frame. The species composition on the wood of both frames were similar. The initial biofouler on the frames were *Cryptosula* and barnacles. At the peak of encrustation, the wood of both frames were encrusted with mainly *Cryptosula*, barnacles and tubeworms. Generally, the wood of the horizontal frame was more encrusted than that in the vertical frame.

From the investigation on the rate of encrustation of biofouling organisms in relation to mesh size, it was observed that the species composition of Group 1 organisms (*Cryptosula*, *Trididemnum*, *Botryllus*, *Symplegma* and other compound tunicates) on the net of the 3 horizontal frames were different but the species composition of Group 2 organisms (barnacles, oysters, tubeworms and gastropods, etc.) on the 3 frames were similar. It was further noted that increase in mesh size was always accompanied by a decrease in species diversity of Group 1 organisms. On the contrary, smaller mesh net usually had more encrustation of Group 1 biofouling organisms.

INTRODUCTION

MARINE boring and fouling organisms have been known to damage boats, ships, port facilities and other marine structures since the beginning of man's maritime activities. Some of these organisms that occur on floating net-cages burrow into, adhere on or are sedentary on the substrate and in so doing choke up the meshes and damage the cages. Cheah and Chua (in press) identified 2 categories of tropical fouling organisms associated with floating net-cages. The first category is the micro-biofoulers such as algal spores, diatoms and to a lesser extent marine bacteria that forms the primary organic film while the second category are the macro-biofoulers such as 'borers' like the shipworm (*Teredo*), the bivalve (*Martesia*), the gribble (*Limnoria*) and other encrusting organisms namely tubeworms, barnacles, bryozoans, hydroids, mussels, oysters, algae, etc.

Recent development in mariculture throughout the world has intensified interest on fouling organisms associated with floating net-cages although information on this aspect is scanty. Some of the prominent research work along this line are given in Milne (1972, 1975 a, b) on marine fouling of various submerged netting fabrics in Scotland, Koops (1971) on fouling

prevention of net-cages in the Western Baltic Sea, Moring and Moring (1975) on succession of net biofouling material at Pugeat Sound, Washington.

The problem of marine fouling of floating net-cage in tropical Malaysian conditions is serious because frames are rapidly attacked by borers and the floats and nets are soon encrusted with a wide variety of fouling organisms (Chua and Teng, 1977). Reference on marine except the reports by Berry (1966) and Cheah and Chua (in press).

This paper presents the results of a preliminary investigation on the rate of encrustation of biofouling organisms on experimental net-cages.

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Materials and Methods

The present investigation was conducted at a marine cage culture farm located in the Western Channel of the Straits of Penang.

Two experimental net frames, A and B, of size 30.5 cm x 30.5 cm with wood surface area of 1007 cm^2 and stretched mesh of 1.27 cm were suspended in horizontal (Frame A) and vertical (Frame B) positions respectively. Another set of experiment consists of two net frames, C and D, of similar frame size and wood surface area but varied in mesh size, *i.e.*, 2.54 cm mesh for frame C and 3.81 cm mesh for frame D. All the four frames were suspended at the same time and same depth so that variation due to differences in duration and depth was eliminated. The stages of colonization by fouling organisms were then monitored and recorded at weekly intervals for a duration of 4 months.

In order to quantify the extent at which the net frames were colonized by biofoulers, the organisms were divided into 2 groups. Group 1 organisms consisted of colonies where their growth covered a wide area namely *Cryptosula*, *Trididemnum*, *Botryllus*, *Symplegma* and other compound tunicates. Group 2 organisms consisted of individual organisms which could be easily counted such as barnacles, oysters, tubeworms and gastropods, etc.

Results

Rate of Encrustation with respect to position of experimental net frame

The species composition of Group 1 organisms on the net of vertical and horizontal frames were similar. The species recorded were mainly *Cryptosula* sp., *Trididemnum* sp., *Botryllus* sp. B and other compound tunicates which occurred on both frames while *Botryllus* sp. A only occurred on Frame A and *Symplegma* sp. only occurred on Frame B. The initial biofoulers on the net in both cases were *Cryptosula* and other compound tunicates occurring as early as 5th or 6th week after submergence of the frames. The sequence of sucession was not marked. The times at maximum encrustation on the net of both frames were different. Maximum encrustation was observed on horizontal (Frame A) and vertical (Frame B) nets sometime during the 10th-11th week and 8th-9th week respectively. This implies that the maximum encrustation of Group 1 organisms on net-mesh occurred earlier in the vertical position. At the peak of encrustation Group 1 organisms occupied a larger area on the vertical than on the horizontal nets (Fig. 1). The major fouling organism on the net of Frame B was *Symplegma* which occupied 91.9 cm^2 while that on the net of Frame A was *Cryptosula* which occupied 39.0 cm^2. The total area

encrustaed on the nets of Frames B and A were 223.3 cm and 108.3 cm² respectively at the peak of encrutsation (Fig. 1). *Cryptosula* were equally abundant on both frames, but *Trididemnum* became more abundant on horizontal net while *Botryllus* sp. B were plentiful on the vertical net.

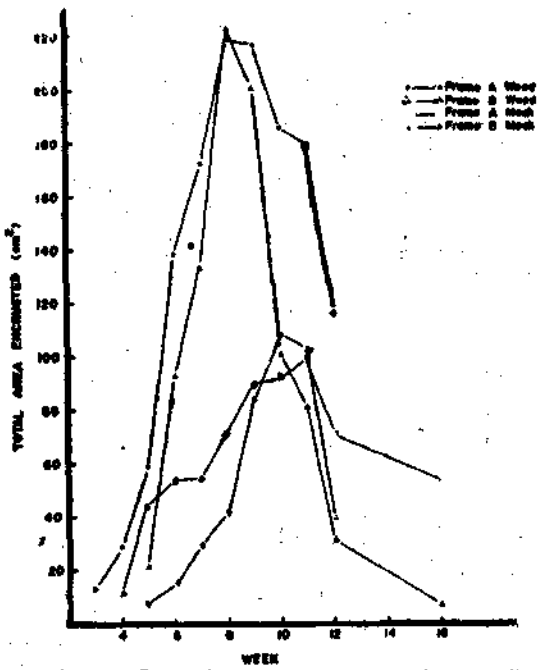

FIG. 1. Rate of encrustations in relation to different position of submerged net-frames : Group 1 biofoulers. Frame A in horizontal position and Frame B in vertical position.

The species composition of Group 2 organisms on the net of both frame were also similar, namely barnacles, oysters, tubeworms and gastropods. The initial biofoulers in both cases were barnacles and tubeworms. The encrustation of Group 2 organisms on the net of both frames continued to increase in number upto the 12th week. There were generally more barnacles and oysters on the horizontal net (Frame A). Tubeworms, however, were more abundant on the vertical net (Frame B).

In the case of Group 1 organisms encrusting the wood, the species composition on both frames were also similar. Four species were recorded on the wood of Frame A but 3 species were recorded on the wood of Frame B. *Cryptosula* and other compound tunicates occurred on both frames whereas *Botryllus* sp. B. and *Symplegma* only occurred on Frame A and *Trididemnum* only occurred on Frame B. The initial and subsequently dominant biofoulers in both cases was *Cryptosula* which occurred in the 3rd and 4th week after submergence of Frames A and B respectively. The times at maximum encrustation of Group 1 organisms on the wood of both frames were different. This occurred on Frames A and B during the 8th and 10th week after submergence of frames respectively.

This implies that maximum encrustation of Group 1 organisms occurred earlier in the horizontal Frame A. At the peak of encrustation, Group 1 organisms encrusted a larger area on the wood of the horizontal frame than the wood of the vertical Frame B (Fig. 1). The total area encrusted at this peak on Frame A and B was 218.8 cm² and 92.2 cm² respectively. *Cryptosula* exhibited a peak of encrustation in the 8th week after submergence of Frame A covering an area of 182.5 cm². This was not exhibited on Frame B where the area covered by *Cryptosula* after the 6th week remained more or less stable at 46.4 cm². The other biofoulers on the wood of Frames A and B reached maximum growth in the 10th week after submergence of the frames, followed by a decrease after the maximum growth of each species.

The Group 2 organisms that occurred on the wood of both frames were similar. 4 species were recorded namely barnacles, tubeworms, oysters and gastropods. The initial biofouler in both cases were barnacles which occurred on the 3rd and 4th week after submergence of Frame A and B respectively. In general, there were more biofoulers on the wood of Frames A compared to Frame B at any one time (Fig. 2). The dominant biofoulers in both cases were barnacles and tube-

worms. It was observed that in both cases, a decrease in the number of barnacles was accompanied by an increase in the number of tubeworms. This suggests some form of succession. There was gradual increase in the encrustation of oysters on both frames reaching a peak in the 9th and 8th week after submergence of Frames A and B respectively. The numbers of gastropods on both frames were approximately the same and they occurred after the 8th week of submergence of the frames.

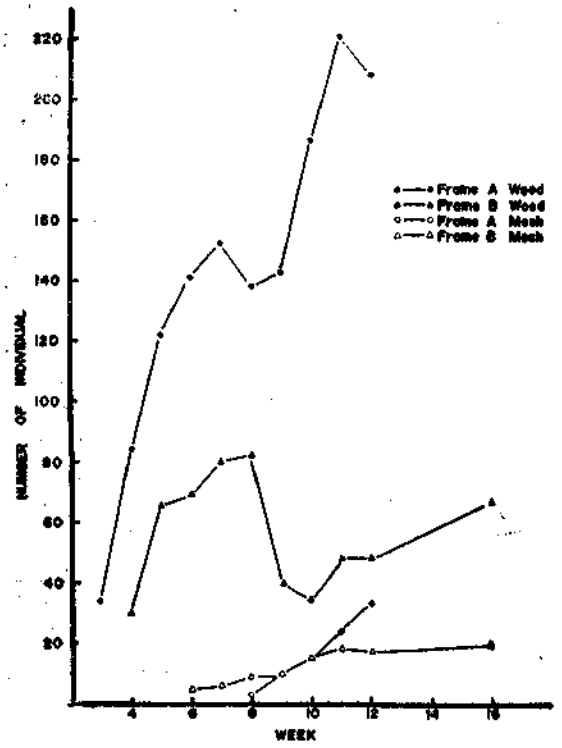

FIG. 2. Rate of encrustations in relation to position of submerged net-frames: Group 2 biofoulers. Frame A in horizontal position and Frame B in vertical position.

Bowerbankia started to encrust both frames in the 6th and 7th week after submergence of Frames A and B respectively. In both Frames the wood boring bivalve *Martesia* occurred after the frames were submerged for more than 11 weeks; however no quantitative results were recorded.

Rate of encrustation with respect to mesh size

The species composition of Group 1 organisms on the three different mesh nets suspended horizontally were different. Frames A (mesh 1.27 cm), C (mesh 2.54 cm) and D (mesh 3.81 cm) were encrusted with 5 species, 3 species and 1 species respectively. This gave an indication that increase in mesh size was accompanied by decrease in species diversity. The rate of encrustation was also slower on the net of bigger mesh. The initial and subsequently dominant fouler on all nets of different mesh sizes was *Cryptosula* which occurred on the 6th, 7th and 8th week after submergence of the Frames A, C, D respectively. The encrustations on the net of Frames A and C showed gradual increase in growth reaching a peak in the 10th and 11th week respectively. *Cryptosula* on Frame D exhibited a more or less constant area of encrustation after the 10th week of submergence of the frame. At the peak of encrustation the Group 1 organisms encrusted the largest area on the net of Frame A, followed by Frame C and then Frame D (Fig. 3). The total area encrusted at this peak on Frames A, C, D were 108.29 cm^2, 25.61 cm^2 and 12.9 cm^2 respectively. *Trididemnum* and other compound tunicates recorded on Frames A and C were absent in Frame D. *Botryllus* sp. A and *Botryllus* sp. B recorded on Frame A were absent on Frames C and D. In comparison, *Cryptosula* encrusted 39.02 cm^2 on Frame A, 16.77 cm^2 on Frame C and 12.90 cm^2 on Frame D. The encrustations on Frames A and C showed decline in area encrusted after the peak but this was not observed on Frame D.

In the case of Group 2 organisms on the net, the species composition was more or less similar on all frames. The initial biofouler on Frames A and C were tubeworms, but that on Frame D was oysters. The encrustations of Group 2 organisms on the net on all frames continued to increase in number upto the 12th week. As expected the smaller mesh net was

heavily encrusted when compared with nets of larger mesh sizes (Fig. 4).

Discussion

The experiments on encrustation of biofouling organisms with respect to different positions of the net frame indicated that the species composition of biofoulers on the nets in both frames were similar. All the organisms had equal potential to foul net irrespective of whether the nets were placed in the vertical or horizontal position. However, the net frame in the vertical position were more encrusted than that on the horizontal frame indicating that some of the biofoulers preferred the former position. *Symplegma*, *Botryllus* sp. B, other compound tunicates and tubeworms occurred more abundantly on the vertical frame. The net of the vertical frame may be acting as a partial barrier to the horizontal movement of the planktonic larval forms and in doing so allowed the free living larvae to settle on the substrates more readily. *Cryptosula*, barnacles and oysters were more abundant in the net of the horizontal frame. The larval forms of these species may have adhered more readily to the horizontal net. The Group 1 organisms showed an increase in growth initially, attaining maximum growth within 8 weeks and declined thereafter. This was the result of natural death or grazing by fauna, *e.g.* fish and crabs in the vicinity. However, the Group 2 organisms progressed in number till the 12th week with no sign of decline in number.

The rate of encrustation was different in both frames where it was observed that the vertical

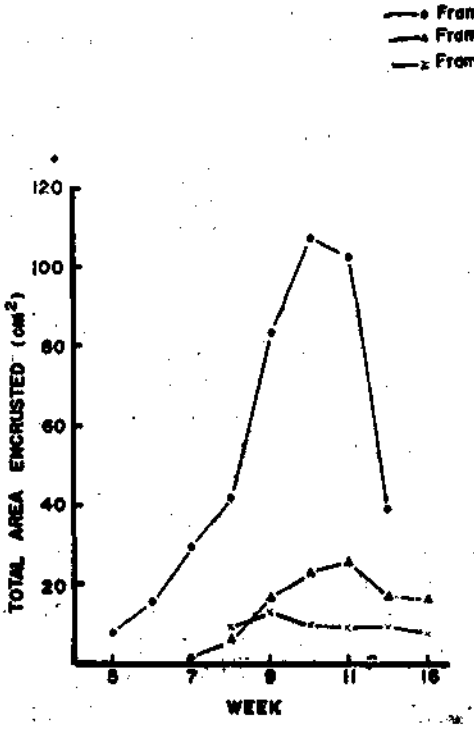

FIG. 3. Encrustation rates in relation to mesh size of submerged net-frames suspended horizontally: Group 1 biofoulers.

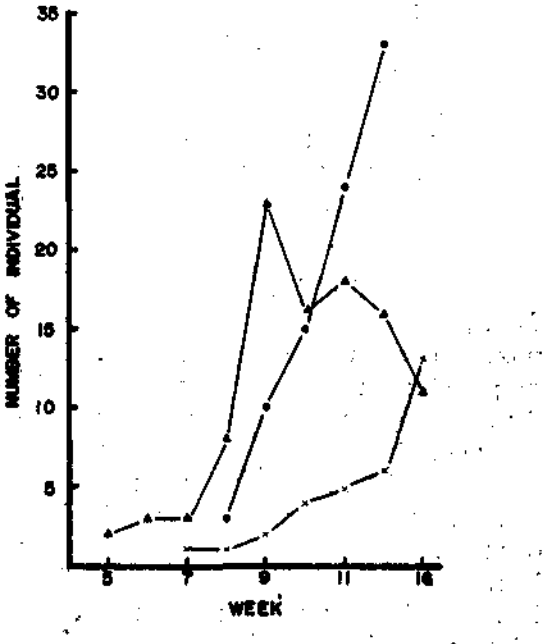

FIG. 4. Encrustation rates in relation to mesh size of submerged net-frames suspended horizontally: Group 2 biofoulers.

frame was more rapidly encrusted than the horizontal frame.

One might argue that since the Group 1 organisms showed a decline in growth after the 8th and 9th week after submergence of the vertical frame, choking of the net should not pose a threat to water movement by biofouling organisms. However, the Group 2 organisms showed a continual growth in number and size especially oysters like *Pinctada* which would eventually obstruct the free passage of water through the mesh of the nets. Koops (1971) working on the Western Baltic Sea reported that *Mytilus edulis* on net-cages prevented sufficient exchange of water so that inside the cage, the dissolved oxygen level dropped. Moring and Moring (1975) also mentioned that heavy clogging of netting by marine algae and invertebrates resulted in less flow through the pens.

In the case of biofouling on the wood of the frame, it was observed that this was equally prone to encrustations as the nets. At all stages of encrustation there were more abundant Group 1 and 2 biofouling organisms on the wood of the horizontal frame than that on the vertical frame. This was in contrast to the situation on the net where Group 1 biofouling organisms predominated. *Cryptosula* and barnacles which were the main wood biofoulers had the habit of encrusting the ventral surface of the wood. As Frame A was in the horizontal position, it allowed maximum area for attachment to the ventral surface. In both frames the decline in growth of *Cryptosula* and barnacles was accompanied by the increase in growth of *Botryllus* sp. B, *Symplegma*, *Tridi-demnum*, other compound tunicates, tube-worms and gastropods. This suggested some form of succession where the initial biofouling organisms, *Cryptosula* were being replaced by *Botryllus* sp. B, *Symplegma*, *Trididemnum* and tubeworms. The initial dominance of barnacles was also reported by Lee and Trott (1973) who worked in Hong Kong waters. They also reported that the dominant barnacles were succeeded by a *Styela-Mytilus* community. However, this has not been observed in the present investigation.

Bowerbankia was observed encrusting the wood from the 6th week after submergence of both frames. It looked glassy and had the potential to cause clogging of the net if it occurred in large quantities. The wood borer *Martesia* began to occur at the 11th week after submergence indicating that damage of wood began approximately 2 months later. The presence of this borer caused a lot of damage to wooden structures which commanded loss of money in replacing the wooden portions of the cage. Boyle and Turner (1976) reported that larval stages of *Martesia* penetrated wood after 48 days.

Insofar as the encrustation of biofouling organisms in relation to mesh size of the net is concerned, it has been shown that the species composition, abundance and rate of encrustation of the biofoulers vary with the size of the mesh used. The results indicated that the smaller the mesh size, the faster the encrustation. This was probably due to the slower speed of the water flushing through the mesh as well as the increase in area for anchorage or attachment of the biofoulers due to smaller mesh size. When an organism settled on the substrate it provided an environment for other biofoulers to settle and as such the small mesh frames usually contained more species and in larger numbers.

The present findings agreed with the general observations that the horizontal portion of floating net cages are usually less fouled by biofoulers than the vertical net walls. The results clearly suggest that in rich tropical waters such as that of Penang Straits, the cages should be changed as frequently as once a week for smaller mesh-sized cages and not more than four weeks for those of larger mesh (about 1.25-2.5 cm). This means that more labour is

needed to clean and change the nets. It is therefore imperative that more research into the types of materials most suitable for cages in tropical marine water so as to avoid or reduce biofouling or other control measures should be initiated in view of the rapid expansion of marine cage culture in many tropical countries.

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WOOD-BORING ORGANISMS IN RELATION TO AQUACULTURE ALONG THE COASTS OF INDIA

K. DHARMARAJ AND N. BALAKRISHNAN NAIR
Dept. of Aquatic Biology & Fisheries, University of Kerala, Trivandrum-695 007

ABSTRACT

Timber is extensively used for the construction of a variety of aquaculture facilities and equipments all along our coasts and this activity is increasing rapidly. One of the serious impediments in the use of timber for sea and/or brackishwater structures is that it is subjected to severe damage, among others, by marine wood-boring organisms. 28 species of shipworms (Teredinidae), 4 species of piddocks (Pholadidae), 5 species and 1 variety of pill-bugs (Sphaeromatidae) and not less than 9 species of gribbles (Limnoriidae) constitute the marine wood-boring community in Indian waters. Their incidence, relative abundance, nature and degree of timber destruction and ecology in some aquafarms situated along the coasts of South India are examined and it was found that they are responsible for huge financial loss all along our sea-front. The nature of attack on different types of wooden equipments used for mariculture like, floating rafts, submerged racks, stakes, piles, etc. showed variation —so also the intensity of destruction at various localities. The occurrence and relative abundance of these pests along the coasts of India, their incidence and activity in different aquatic environments such as the open sea, coastal zones and bays, estuaries, backwaters and mangrove swamps, etc. are presented. Some suggestions for the control of them and the best period for installations of equipments in aqaufarms are also indicated.

INTRODUCTION

THE IMPETUS to study marine wood-boring organisms mainly stems out from the recurring economic loss they cause by destroying the water front timber structures. In India the fishing industry alone has been reported to suffer an annual loss of about ten million rupees owing to the ravage of wooden catamarans, boats, etc. by molluscan and crustacean wood borers along the coasts (Purushothaman and Rao, 1971). Marine wood-borers are ubiquitous pests of all manner of timber in the sea, estuaries, brackishwaters and often in almost fresh water localities also. But, the damage caused by them in aquacultural operations has not been properly assessed. Timber has been extensively used for constructing a variety of aquaculture facilities and equipments like rafts, racks, pens, stakes, etc. and the activity is increasing. Only a very few of the vast literature on marine borers specifically refer to their incidence in aquafarms. Much work has been done in India on the biology (Nair, 1964, 1965, 1967, 1968; Nair and Saraswathy, 1971; John, 1968; Cherian, 1973) and systematics (Nair, 1954, 1955; Pillai, 1961; Sreenivasan, 1959) of these pests, but very little attention has hitherto been paid to correlate their ecology and timber destruction in aquafarms even though the economy of mariculture depends much on the cost of construction and maintenance of wooden equipments.

In many localities along the coasts of India, massive infestation by shipworms, piddocks and/or sphaeromatids destroying anything made of timber has become a serious impediment for mariculture operations (Nair and Dharmaraj, 1979, 1980). Nelson (1922) reported that cypress wooden platforms immersed in Barnegat Bay (New Jersey) to collect oyster

spat collapsed in a short span of time owing to very heavy invasion of *Teredo navalis*. Wooden sticks used in the culture of oysters in New South Wales were also subjected to rapid decay due to the ravages of *Nausitora* that they had broken to pieces before the oysters themselves matured (Roughly, 1925). Roughly (1925 a), also reported that timber borers like *Limnoria*, *Sphaeroma* and *Nausitora* are classed by cultivators amongst the oyster pests since they shorten the life of all sorts of timber used in culture operations. Wooden railings of the oyster plots in Bacoor Bay, Philippine Islands have been destroyed by *Martesia striata* (Villadolid and Villaluz, 1938). Attack by the shipworm *T. navalis* has been a serious menace to wooden equipments used for mariculture in the Gulf of St. Lawrence, Canada (Needler and Needler, 1940).

Part of the work (studies from Vizhinjam and Neendakara harbours) was carried out under the CSIR Project entitled, ' Composition and energy flow... [No. 38 (232)/76 GAU II dated 2.6.76] at the Department of Aquatic Biology and Fisheries of the University of Kerala. Surveys from different localities were financed by the U.G.C. National Fellowship to one of the authors (N.B. Nair). We are thankful to Prof. H. P. C. Shetty and Dr. P. Natarajan and Dr. N. R. Menon of the College of Fisheries, Mangalore, Mr. J. J. Joel of the CMFRI, Mangalore, Prof. Dr. R. Natarajan and Mr. S. Sethuramalingam of the CAS in Marine Biology, Parangipettai, Dr. P. S. B. R. James of the CMFRI, Mandapam camp and Shri K. Nagappan Nair and Shri S. Mahadevan of the CMFRI, Tuticorin for the helps extended then and there during field surveys.

Method of Approach

The incidence of timber boring animals and the nature of destruction in localities like Tuticorin, Mandapam, Krusadai Island, Parangipettai, Vizhinjam, Neendakara and Mangalore were examined by collecting infested samples of wood from aquafarms situated in these localities wherever possible, from other water front timber structures and also from drift wood collected from the shores. The pattern of distribution of these pests along the Indian Coast was compiled from the results of our surveys as well as from earlier reports on the subject. In the section on the period of settlement, for Visakhapatnam, Madras and Cochin Harbours data from earlier works have been adapted and the sources are acknowledged appropriately.

Results and Discussion

Species of borers and their distribution along the coasts of India

The heterogenous assemblage of marine wood boring community in India is derived from four families belonging to two phyla. Molluscs are represented by teredinids (shipworms) and pholadids (Piddocks) and arthropods are represented by sphaeromatids (pill-bugs) and limnorid (gribbles). Twenty-eight species of shipworms (Table 1) have been reported to occur along the coasts of India and of these a few like *Lyrodus pedicellatus*, *Teredo furcifera*, *Bankia carinata*, *Bankia campanellata* and *Nausitora hedleyi* are the dominant and highly destructive species. Four species of piddocks (Table 1) occur along our coasts and among them *Martesia striata* is the most destructive species. About five species and a variety of pill-bugs (Table 2) occur in India of which *Sphaeroma terebrans* and *S. annandalei* are serious pests of wood. About nine species of gribbles (Table 2) have been known from the coasts of India and nearby islands. Limnorids have not been considered as serious pests of wood in India (Pillai, 1961; Becker, 1958) by virtue of their inability to tolerate the high water temperatures of the tropical seas, but recent reports (Karande, 1978; Sreenivasan

TABLE 1. *Distribution of molluscan wood borers along the coasts of the maritime States in India and Union Territories*

	Species	West Bengal	Orissa	Andhra Pradesh	Tamil Nadu	Andaman and Nicobar Islands	Kerala	Karnataka including Goa	Maharashtra	Gujarat
1.	*Bactronophorus thoracites* (Gould)	..	+	+	+	+	..	..	..	+	..
2.	*Dicyathifer manni* (Wright)	..	+	+	+	+	..	+	+	+	+
3.	*Teredothyra smithi* (Bartsch)	..	..	..	..	+	..	..	..	..	+
4.	*Teredothyra excavate* (Jeffreys)	..	..	..	..	+	..	..	..	..	..
5.	*Teredothyra matocotana* (Bartsch)	..		..	..	..	..	..	..	..	+
6.	*Teredora princesae* (Sivickis)	..	..	+	..	+	..	+	..	..	..
7.	*Uperotus clavus* (Gmelin)	..	..	..	+	+	..	..	..	..	..
8.	*U. rehderi* (Nair)	..	..	..	..	+	..	..	..	..	..
9.	*Teredo furcifera* von Martens	..	..	..	+	+	+	+	+	+	+
10.	*T. fulleri* Clapp	..	..	..	..	+	..	..	..	..	..
11.	*T. bartschi* Clapp	..	..	..	+	+	..	..	..	..	..
12.	*T. clappi* Bartsch	..	..	..	+	+	..	+	..	+	+
13.	*T. triangularis* Edmondson	..	..	..	+	+	..	..	..	..	..
14.	*Lyrodus pedicellatus* (Quaterfages)	..	..	+	+	+	..	+	+	+	+
15.	*L. affinis* (Deshayes)	..	..	..	+	+	..	..	..	..	..
16.	*L. massa* (Lamy)	..	..	..	+	+	..	+	..	..	..
17.	*Nototeredo edax* (Hedley)	..	..	..	+	+	+	..	..	+	+
18.	*N. knoxi* (Bartsch)	..	..	+	..	..	..	..	..	..	..
19.	*Nausitora dunlopei* Wright	..	+	+	+	+	..	..	..	..	..
20.	*N. hedleyi* Schepman	..	..	+	..	+	..	+	+	+	+
21.	*N. fustcula* (Jeffreys)	..	..	+	+	..	..	..	..	..	..
22.	*Bankia campenellata* Moll and Roch	..	+	+	+	+	+	+	+	+	+
23.	*B. carinata* (Gray)	..	+	+	+	+	..	+	+	+	..
24.	*B. fimbriatula* Moll and Roch	..	..	+	..	..	..	..	..	..	..
25.	*B. bipalmulata* (Lamarck)	..	..	..	..	+	+	..	..	..	..
26.	*B. bipennata* (Turton)	..	..	..	+	+	..	..	..	..	..
27.	*B. nordi* Moll	..	+	..	..	+	..	..	..	+	..
28.	*B. rochi* Moll	..	+	+	+	..	..	+	+	+	..
29.	*Martesia fragilis* Verrill and Bush	..	..	..	..	+	+	+	..	..	..
30.	*M. striata* Linne	..	+	+	+	+	+	+	+	+	+
31.	*Xylphaga* sp.	..	..	+	..	..	..	..	..	..	..
32.	*Barnea birmaniea* Philippi	..	..	..	..	+	..	..	..	..	..

+ = Present. .. = Not recorded.

TABLE 2. *Distribution of crustacean wood borers along the coasts of the maritime States in India and Union Territories*

Species	West Bengal	Orissa	Andhra Pradesh	Tamil Nadu	Andaman and Nicobar Islands	Kerala	Karnakata including Goa	Maha-rashtra	Gujarat
Sphaeroma terebrans Bate	..	..	..	+	+	..	+	+	+	+
S. annandalei Stebbing	..	..	..	+	+	..	+	+	+	+
S. annandalei travencorensis Pillai	..	..	..	..	..	..	+	+	..	..
S. tuberculatum George	..	..	..	..	+	..	..	..	..	..
S. walkeri Stebbing	..	..	..	+	+	..	+	..	+	..
S. triste Heller	..	..	..	..	+	+	..	..	..	..
Limnoria indica Becker and Kampf	..	..	..	..	+	+	..	..	+	..
L. septima Barnard	..	..	..	..	..	+	..	..	..	..
L. tripunctata Menzies	..	..	..	..	+	..	..	..	..	..
L. bombayensis Pillai	..	..	..	..	..	..	..	..	+	..
L. pfefferi Stebbing	..	..	..	+	+	+	..	..	..	..
L. unicornis Menzies	..	..	..	..	..	+	..	..	..	..
L. platycauda Manzies	..	..	..	..	..	+	..	..	..	..
L. andamanensis Rao and Ganapati	..	..	..	..	..	+	..	..	..	..
L. insulae Menzies	..	..	..	..	..	+	..	..	..	..

+ = Present. .. = Not recorded.

and Chandramohan 1975; Santhakumaran, 1969) and their abundant occurrence observed along the coasts of Krusadai Island indicate that they are also posing increasing threat to the timber structures in our coastal waters.

The pattern of distribution of the twenty-eight species of shipworms and four species of piddocks along the coasts of India is illustrated in Fig. 1. This aspect could not be considered complete because virtually nothing is known regarding the incidence of timber boring animals except from very selected localities where major harbours are situated or from the vicinity of research institutes which carry out studies on marine boring animals. Reports from other localities are mostly based on a few collections which are not expected to give a complete list because these pests are well known for their variability in incidence with space and time (Dharmaraj and Nair, 1979). However, the occurrence of borers marked in various localities would represent the most dominant forms and so shall give an idea of the highly destructive species in the respective localities. It could be observed that some species like *Dicyathifer manni*, *Teredo furcifera*, *Lyrodus pedicellatus*, *Nausitora hedleyi*, *Bankia campanellata*, *B. carinata*, *B. rochi* and *Martesia striata* are widely distributed along our coasts.

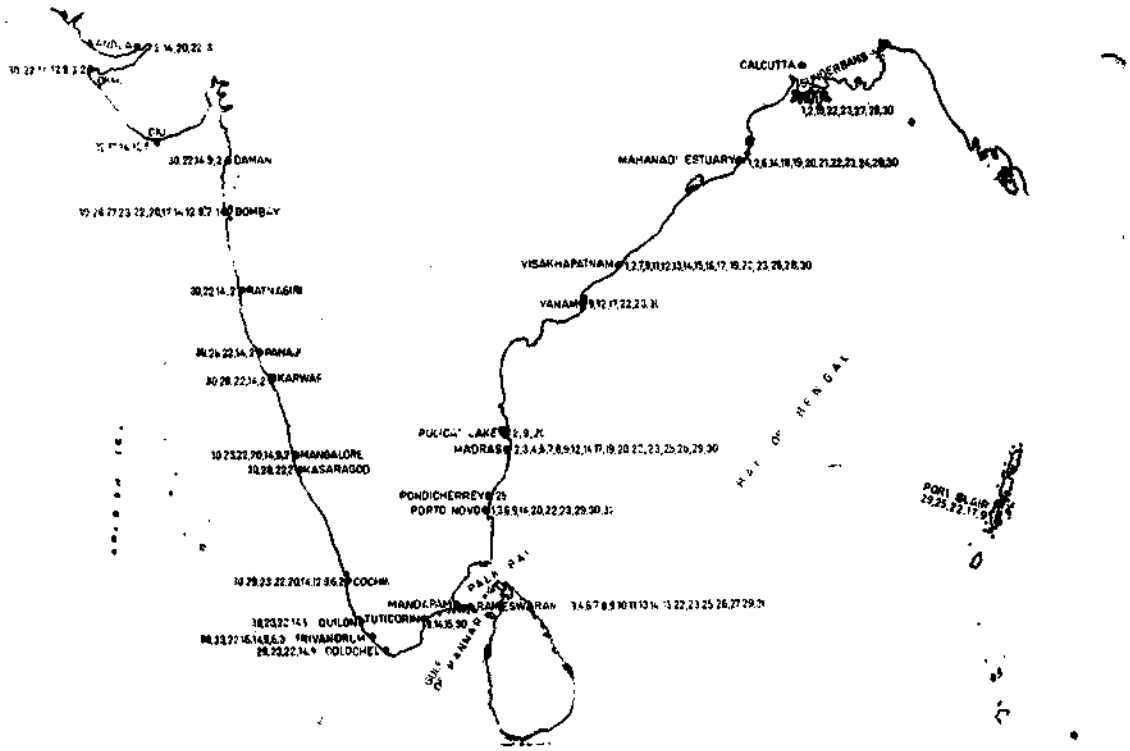

FIG. 1. The pattern of distribution of marine timber boring molluscs along the coasts of India. The numbers refer to the corresponding species listed in Table 1.

Table 1 presents the nature of distribution of molluscan wood borers along the coasts of the various maritime States of India and in the Andaman and Nicobar Islands. Almost all of them except *Bankia rochi*, *B. fimbriatula*, *Nausitora fusticula*, *Nototeredo knoxi* and *Teredothyra matocotana* occur along Tamil Nadu Coast. Next higher species abundance is along

eighteen species of molluscan wood borers have been reported. This is roughly proportional to the amount of work carried out from the coasts of the various states. The number of species occuring along the coasts of other States might also be more than that is realised and it necessitates further detailed surveys along the unexplored coastal zones.

Some interesting features could be observed from the records of occurrence in various localities and from the nature of distribution of the wood boring pests along the coasts of India. Many species show a discontinuous distribution, *e.g.* species of *Teredothyra*, *Uperotus*, etc. *T. smithi* has been collected from Tamil Nadu and Gujarat Coasts, *T. excavata* from Tamil Nadu and *T. matocotana* from Gujarat only. Species of *Uperotus* are characteristic in their occurrence only along the coasts of Andhra Pradesh and Tamil Nadu. *Teredo furcifera* one of the widely distributed species, has not so far been reported from West Bengal and Orissa Coasts. *Teredo fulleri* seems to be a characteristic borer occurring in the Gulf of Mannar and the Palk Bay Coasts of Mandapam and the near by islands. *Teredo bartschi*, *T. triangularis*, *Lyrodus affinis*, *Nototeredo knoxi*, *Nausitora dunlopei*, *N. fusticula*, *Bankia fimbriatula*, *B. bipennata*, *Xylophaga* sp. and *Barnea biramanica* also have been reported from the east coast of India, but so far not recorded from the west coast.

Table 2 presents the nature of distribution of the crustacean wood borers. Throughout the west coast and along the east coast bordering Andhra Pradesh and Tamil Nadu, *Sphaeroma terebrans* has been reported to occur abundantly in brackishwaer and estuarine localities. *S. annandalei* also shows a similar pattern of distribution. Nothing is known of the crustacean wood borers from Orissa and West Bengal. *S. annandalei* var. *travancorensis* occurs along Kerala and Karnataka Coasts and *S. tuberculatum* is known from its type locality (Tuticorin) only. *S. triste* occurs along the coast of Tamil Nadu and in Andaman and Nicobar Islands. *S. walkeri* has been collected from Maharashtra, Kerala, Tamil Nadu and Andhra Coasts. Of the nine species of limnorids, all but *Limnoria tribunctata* and *L. bombayensis* occur in Andaman and Nicobar Islands. Limnorid are not active along the coasts of Kerala, Karnataka, Andhra Pradesh and Gujarat. In some localities like Bombay, Madras and Mandapam, the incidence of limnorids has been steadily increasing and are likely to cause severe damage to the superficial layers of wood. *Limnoria indica* and *L. bombayensis* are the destructive species along the coasts of the Indian main land.

Incidence in aquafarms

Though, the scope, prospects and necessity of coastal aquaculture in India are very great, it has not yet flourished as an industry partly because of many difficulties encountered in the maintenance of aquafarms. Almost all the species of wood-borers except *Uperotus clavus*, *Martesia fragilis*, *Teredothyra* spp. and few others could invade and destroy the timber structures in aquafarms. Different types of wooden equipments used in aquafarms would experience different manners of borer activity depending on the level and locality of exposure. In the Karapad Creek oyster farm at Tuticorin, destructive species of borers were found to be *Teredo furcifera*, *Lyrodus pedicellatus* and *L. affinis*, of which the first one has been the most dominant species (Nair and Dharmaraj, 1979). The pholad *Martesia striata* also has been a serious pest here, along with the teredinids. Observations on the nature of vertical distribution of borer infestation on piles in these aquafarms revealed that teredinids infest the piles from surface to mudline with dense settlement near the bottom and the attack of pholadids is dense a little below the low water mark.

The incidence of wood-borers in the aqua-farms situated along the Gulf of Mannar and the Palk Bay Coasts of Mandapam and the nearby islands was also studied (Nair and Dharmaraj, 1980). The farms situated within short distances between one another showed varied species composition of borers, variations in the intensity of timber destruction and in the relative abundance of different species of borers. In the pearl oyster and sea-weed farms of Krusadai Island, the relative abundance of different species of shipworms has been as follows : *Teredo furcifera*—*T. fulleri*—*Lyrodus pedicellatus*—*Teredo triangularis* and *T. bartschi*. The most affected were the sea weed farms where numerous wooden piles have been installed for the rope culture of *Gracilaria* spp. and other algae. Shipworms eat away the heart of the timber and render it a weak, porous and fragile mass eventually leading to the toppling of piles which normally break at a point a little above the mud-line (Fig. 2). In these farms gribbles (*Limnoria* spp.) also have been active in destroying the superficial layers of the piles. Wood used in the oyster and fish farms situated in the Gulf of Mannar Coast of Mandapam Camp have been riddled mostly by *Teredo furcifera* and *Lyrodus pedicellatus*. Stakes used here were found to be serviceable only for about four months despite the traditional protective coatings applied to them prior to installation. Along the coast of Hare Island, *Teredo furcifera*, *T. bartschi*, *T. fulleri* and *Lyrodus pedicellatus* have been the dominant species of shipworms. Stray occurrence of *Limnoria* spp. also has been noticed here.

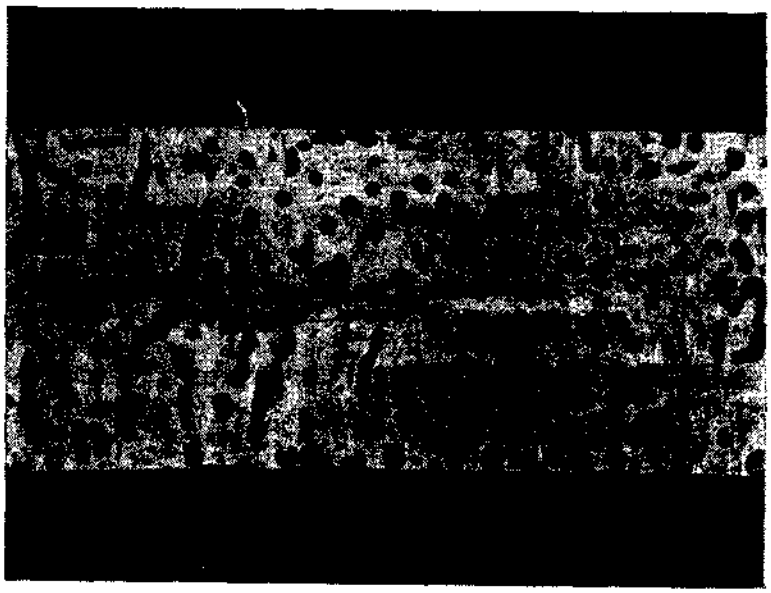

FIG. 2. Vertical section of a *Casuarina* pile used for mariculture operations showing the nature of internal damage caused by shipworms.

Wood used in an experimental aquafarm in Vellar Estuary at Parangipettai was found to harbour *Lyrodus pedicellatus* and *Nausitora hedleyi*. Abundant occurrence of *Sphaeroma*

terebrans and *S. annandalei* also has been observed in the estuary and in Pichavaram mangroves connecting the Vellar-Coleroon estuarine system. The characteristic mangrove shipworm *Bactronophorus thoracites* which has been reported as a serious pest of wood including living mangrove trees of Sunderbans (Roonwal, 1954) also occurs in the Pichavaram mangroves.

In the Nethravathy—Gurupur Estuary and the coastal waters of Mangalore which offer great potentialities for mariculture, teredinids, pholadids as well as sphaeromatids were found to be very destructive. Dominant species included *Lyrodus pedicellatus, Dicyathifer manni, Nausitora hedleyi, Martesia striata, Martesia* sp. *Sphaeroma terebrans* and *S. annandalei*. Almost all these species are capable of tolerating the comparatively wide fluctuations in salinity commonly encountered in estuarine environments and so would impose serious hindrances to the maintenance of timber structures for mariculture operations.

Ashtamudi Lake, situated in the southwest coast of India also offers a vast stretch of water body in which large scale culture of aquatic organisms could be easily carried out. The stakes used for the operation of Chinese nets for fishing have been attacked by various species of borers like *Sphaeroma terebrans, S. annandalei* and *Martesia striata*. The dominant teredinid borer in Neendakara Estuary and Ashtamudi Lake is *Lyrodus pedicellatus*. Near the bar-mouth occasional incidence of species like *Teredo furcifera* and *Bankia carinata* also has been observed. In the interior arms of the lake away from the sea, the destructive activity of sphaeromatids has been very severe. *Nausitora hedleyi* which is one of the dominant molluscan borers in the nearby Vembanad Lake has not been known to occur in Ashtamudi Lake. The wooden rafts used for the culture of edible mussels and pearl oysters in Vizhinjam Bay are attacked by the teredinids *Lyrodus pedicellatus, Bankia carinata, B. campanellata* and *Teredo furcifera*. *Lyrodus massa* also occurs in the area. The pholad *Martesia striata* has been highly destructive in this locality from where sphearomatids and limnorids have not been collected.

Nature of infestation in different types of wooden equipments

Depending on the depth of exposure of the wooden equipments, the intensity of destruction by borers would vary. Piles and stakes are the most affected ones in aquafarms since they run throughout the water column and are subjected to the settlement of all the groups of borers. Shipworms generally infest the wood from surface to bottom of the water column with dense settlement a little above the mud-line both in coastal waters and in estuarine areas (Nair, 1966; Nair and Dharmaraj, 1979), however, *Teredo furcifera* has been found to settle abundantly near the low water mark in Visakhapatnam Harbour (Nagabhushanam, 1960). *Martesia striata* exhibits remarkable adaptations in their vertical ditribution to overcome unfavourable environmental features. In coastal waters it is active throughout the water column as shipworms, but the region of dense settlement is normally a little below the low water mark. The region of dense attack of *M. striata* in estuarine environments is near the bottom (Nair, 1966; Dharmaraj and Nair, 1979). Thus in estuarine areas, attack by both shipworms and piddocks is concentrated towards the bottom and this region becomes weak and breaks off. Sphaeromatdis and limnorids abound the timber structures at the inter tidal zone. In the backwater and estuarine environments, piles and pillars subjected to sphaeromatid attack assume characteristic hour-glass shape in course of time on account of the combined action of borers and the mechanical action of waves. Floating rafts in coastal areas are likely to be invaded by piddocks, limnorids and shipworms in the areas of contact with the water but the

intensity of destruction would be less. Rafts exposed in brackish water and estuarine areas are susceptible to the heavy attack of sphaeromatids. Cages and racks used in mariculture operations would experience varied types of borer activity depending on the depth of exposure and the species of borers present in the area.

Active species of borers in different aquatic biotopes

Despite the ability of many species of timber destroying animals to acclimatize to the changing environmental features and to the different types of aquatic biotopes like coastal waters, estuaries, backwaters and mangroves; and the euryhaline nature of most of them, some species are active only in certain biotopes. In Table 3 is presented the nature of horizontal distribution of dominant species of borers in the different aquatic biotopes of Parangipettai. It could be seen that *Lyrodus pedicellatus* occurs in all these environments and *Bactronophorus thoracites* occurs only in mangrove area. The distributions of the other species are also similar to the generalised account given below.

Wood-boring sphaeromatids are active in brackish water areas where salinity of the ambient water is always lower than that of the normal sea water. High salinity has been considered deterimental to their activities (John, 1971). Both salinity and temperature are important in regulating the incidence of limnorid wood-borers. No species of *Limnoria* has been observed to be destructive in low saline areas and the increasing density of them in certain coastal zones (Karande, 1978) might be due to their gradual acclimatization to higher temperatures. The pholadid wood-borer *Martesia striata*, by virtue of its extreme euryhalinity has been able to survive efficiently in almost all types of aquatic biotopes, whereas the other species of the genus *M. fragilis* occurs mostly in offshore waters. Of the dominant species of teredinids in India, *Lyrodus pedicellatus* has been highly destructive in estuaries, mangroves, backwaters and also in certain coastal localities. *Teredo furcifera* seems to be active mostly in coastal waters, however with limited intrusion into estuaries also. Some earlier reports on the dominance of this species in estuaries and backwaters might be due to the then prevalent confusion on the distinction between *T. furcifera* and *Lyrodus pedicellatus*. *Bankia carinata* and *B. campanellata* have been destructive in coastal waters only. Species of the genus *Nausitora* have mostly been collected from estuaries, backwaters or mangroves. *Bactronophorus thoracites* and *Dicyathifer manni* are also normally confined to brackishwater localities. *Teredo clappi*, *T. bartschi*, *T. fulleri*, *Bankiarochi*, *Teredora princesae* and *Uperotus rehderi* are also destructive in certain coastal and offshore localities and these species generally work along with some other dominant forms.

Notes on the biology of wood boring animals

Sphaeromatids and pholadids probably bore into wood for shelter alone, while teredinids and limnorids use the wood also as food. It has been indicated that Limnoria can digest and subsist on wood (Ray, 1959 a, b). Though, cellulase is present in the digestive tract of some of the sphaeromatids, algae, fungi, etc. are the essential food materials for their healthy growth and activity, and they merely waft off the particles of wood while boring (John, 1968). Various authors have suggested that teredinids use the wood excavated by them as food (Nair and Saraswathy, 1971). Presence of cellulase and the cellulose and hemi cellulose digesting capacity of the digestive extracts of teredinids have been reported (Hashimoto and Onoma, 1949; Miller and Boynten, 1926; Nair, 1955 a, 1956; Dean, 1978). However, controvercial views still exist on whether the cellulases present in the digestive tract of shipworms are produced by the animal

TABLE 3. *Distribution of woodboring animals in the aquatic biotopes of Parangipettai and their incidence in drift wood and catamarans*

Species	Biotope				Drift Wood	Catamaran logs
	Vellar Estuary	Killai Backwater	Pichavaram mangrove	Neritic zone of the Bay of Bengal		
Teredinidae						
Bactronophorus thoracites	..	..	..	+	..	..	..
Teredothyra smithi	..	..	..	..	..	+	..
Teredora princessae	..	..	..	..	..	+	+
Teredo furcifera	..	+	..	..	+	+	+
Lyrodus pedicellatus	..	+	+	+	+	+	+
Nausitora hedleyi	..	+	..	+	..	..	..
Bankia carinata	..	..	..	..	+	+	+
B. campanellata	..	..	..	..	..	+	+
Pholadidae						
Martesia fragilis	..	..	..	..	..	+	..
M. striata	..	+	+	+	..	..	+
Barnea birmanica	..	..	..	+	..	..	..
Sphaeromatidae						
Sphaeroma terebrans	..	+	+	+	..	..	..
S. annandalei	..	..	+	+	..	..	..

+ = Present. .. = Not collected.

itself or by commensal bacteria. Some species of shipworms need plankton for their normal growth and propagation. Species of *Martesia* are known to feed on planktonic food (Sreenivasan, 1960; Balasubramanian, 1970).

Parameters like salinity, temperature, water currents, etc. have been responsible for the regulation of breeding and dispersal of sphaeromatids. Limnorids and sphaeromatids carry their eggs in brood pouch and the juveniles are liberated only at an advanced stage of development when they could burrow by themselves. Adult boring isopods also could excavate new burrows many times and on account of this they are capable of migrating to safer localities when the environmental conditions become adverse in an area. The absence of strictly passive dispersal and capability of the adults to migrate are the reasons for the localised severe attack of boring isopods recurring in some localities. Oviparity and viviparity have been observed among shipworms. Species of *Bankia*, *Nausitora*, etc. have external fertilization while some like *Teredo* and *Lyrodus* have internal fertilization and incubate their eggs in the mantle cavity or gill until they are developed to an advanced stage. Larvae of shipworms and piddocks lead a pelagic free swimming life for durations varying with species and locality. Then they settle and crawl over the surface of wood till finding a suitable spot to bore into (Nair, 1956 a). When no suitable substratum is available, the larval life could even be prolonged for several days in most species and this may account for the wide distribution of wood boring pelecypods. Besides, transport of the adults through drift wood to distant places is yet another means of dispersal for the teredinids and pholadids.

Period of settlement in relation to aquaculture operations

Precise knowledge on the periods of settlement of the various species of wood borers in any locality would help to minimise their attack on water front wooden materials. If repairs, replacements and fresh installation of piles and stakes in aquafarms are carried out at certain specific periods of the year, avoiding the peak periods of invasion by borers, the degree of destruction could be minimised. Biologically, this aspect of study is significant as it is a reflection of the breeding season and a reliable measure of breeding success (Nair and Saraswathy, 1971). Each species of borers active in a locality usually have different breeding periods and seasons of settlement. A species may also have specially varying seasons of settlement governed by local environmental features. The density of distribution of marine borers has fluctuated over long periods and in one and the same period it has differed considerably in various locations along the same stretch of coast line.

In Fig. 3 is illustrated the period of settlement of *Teredo furcifera* and *Lyrodus pedicellatus*

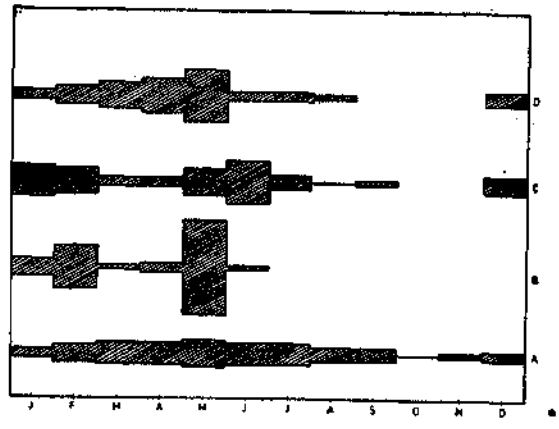

FIG. 3. The period of settlement of teredinine wood borers. A. *Teredo furcifera* in Visakhapatnam (Nagabhushanam, 1962), B. *T. furcifera* in Cochin (Santhakumari and Nair, 1975), C. *Lyrodus pedicellatus* in Neendakara and D. *L. pedicellatus* + few *T. furcifera*) in Vizhinjam.

in certain localities along our coasts. Nagabhushanam (1962) found that in Visakhapatnam harbour, settlement of *Teredo furcifera* is almost throughout the year with a peak during summer and very little invasion during

winter. In Cochin Harbour attack by *T. furcifera* is confined to the high saline summer months only, their activity being checked by the prevailing low saline ambient waters during the rest of the year (Santhakumari and Nair, 1975). *Lyrodus pedicellatus* attacked wooden panels intensively during January, February, May and June in Neendakara Harbour. During August and September some settlement occurred and no evidence of attack was registered during October and November . In Vizhinjam Bay, except during September and October *Lyrodus pedicellatus* settled throughout the year, however with dense invasion from February to May.

Periods of invasion by the dominant species of *Bankia* and *Nausitora* are illustrated in Fig. 4. *Bankia campanellata* in Visakhapatnam Harbour has been found to invade

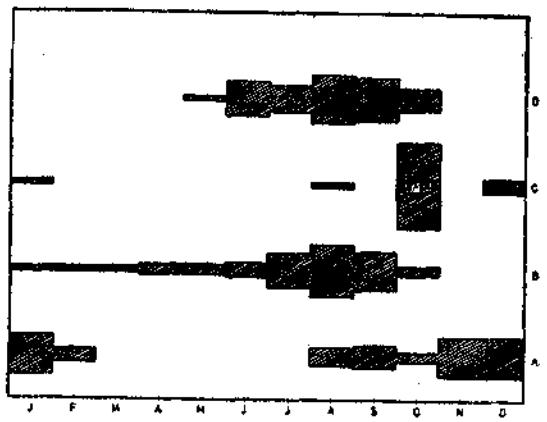

FIG. 4. The period of settlement of bankiine wood borers. A. *Bankia campanellata* in Visakhapatnam (Nagabhushanam, 1962), B. *B. carinata* in Madras (Nair, 1957), C. *Nausitora hedleyi* in Cochin (Santhakumari and Nair, 1975) and D. *Bankia* spp. in Vizhinjam.

timber structures from August to February with no settlement from March to July (Nagabhushanam, 1962). Attack by *B. carinata* in Madras has been heavy from July to September though it occurred almost throughout the year except during November and December (Nair, 1957). Santhakumari and Nair (1975) found that *Nausitora hedleyi* invades wood in Cochin Harbour only during low saline periods. Its attack has been very heavy during October when nearly 85.7% of the total settlement for the year occurred and there was no settlement from February to July. *Bankia* attack in Vizhinjam Bay also is seasonal from May to October with dense settlement during August and September.

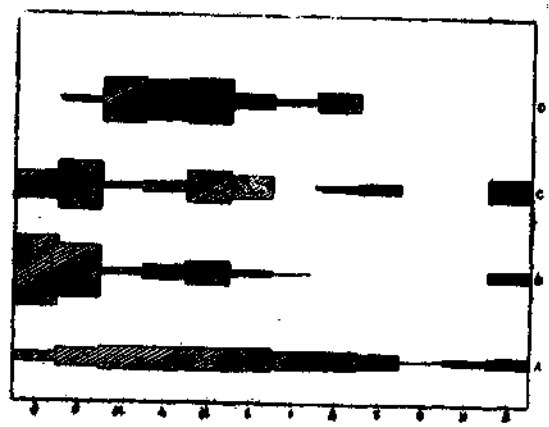

FIG. 5. The period of settlement of *Martesia striata*. A. in Visakhapatnam (Nagabhushanam, 1962), B. in Cochin (Santhakumari and Nair, 1975), C. in Neendakara and D. in Vizhinjam.

The pholad *Martesia triata* was found to settle almost throughout the year in Visakhapatnam Harbour but the settlement has been sparse during October and dense from March to June (Nagabhushanam, 1962). In Cochin Harbour the season of settlement of *M. striata* is from December to July with dense settlement during January and February (Santhakumari and Nair, 1975). There has been no invasion from August to November when the salinity of the area has been very low. In the barmouth of Ashtamudi Lake (Neendakara) wooden panels were attacked by *M. striata* heavily during January, February and May. The settlement has been less during March, April, August and September and no attack was registered during July, October and November (Fig. 5). *M. striata* invaded timber panels

exposed in Vizhinjam Bay from February to August with dense settlement from March to May. The rest of the year showed no evidence of attack by this species.

No information is available on the period of invasion by sphaeromatids along our coasts except from the southwest coast where, particularly in Cochin Backwater it has been intensively studied (John 1968; Nair, 1967; Cherian, 1973). The pattern of seasonal settlement of the sphaeromatids (Fig. 6) indicate that low salinity is favourable for their

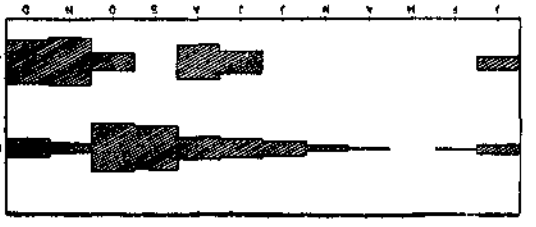

FIG. 6. The period of invasion of wood boring sphaeromatids (*Sphaeroma terebrans*+*S. annadelei*) A. in Cochin (Santhakumari and Nair, 1975) and B. in Neendakara (Dharmaraj and Nair, 1979).

invasion and boring activity. In Cochin Backwater panels immersed during July, August and October to January have been heavily riddled by them (Santhakumari and Nair, 1975). They were found to invade timber structures almost throughout the year in Neendakara Harbour with heavy incidence during October and September. Summer months registered the least attack and in March no invasion occurred (Dharmaraj and Nair, 1979).

The above account indicates that teredinine borers are highly destructive along our coasts almost throughout the year leaving only a small gap during the northeast monsoon period and in all the localities studied, they readily settle during high saline summer months. The bankiine borers are active during winter months. Of these some like *Nausitora hedleyi* invariably prefer low saline ambient waters for their initial settlement. *Martesia striata* generally settles during high saline summer months though it is present in brackishwater areas also. The period of the Northeast monsoon is a comparatively safe one from the attack of *M. striata*. Sphaeromatid invasion in the backwater localities of India has been dense during the monsoon and post-monsoon months and poor during the summer. Virtually nothing is known of the seasonal incidence of *Limnoria* spp. in Indian waters.

Causes for and suggestions to minimise the attack by borers

The incidence of timber boring animals in a locality is a function of the availability of timber structures for their settlement and growth. Transport of the boring molluscs and crustaceans from distant localities are almost always possible by water movements. The presence of already infested timber structures in a locality would accentuate the severity of destruction of freshly immersed timber. Most of the boring pests attain sexual maturity within a short time after settlement in tropical waters and produce many broods during the course of one year resulting in heavy damage. Massive infestation by borers in the oyster farm at Tuticorin has been attributed to the dense population of shipworms and *Martesia* present in the vicinity and the almost uninterrupted breeding habits of these tropical marine wood borers. Removal of all sorts of infested timber structures would greatly help to reduce the ravages. Fresh installations should be carried out during certain specific periods of the year avoiding the peak invasion periods after a scrutiny of the period of settlement of different species of borers. Much of the damage to floating wooden rafts could be avoided if care is taken to see that they do not come into contact with water, by using large buoys or through an effective concrete insulation. When not in use, rafts, cages and racks

should be kept out of water to kill the borers infesting them and to avoid fresh settlement.

Timbers show varying degree of resistance to marine borer attack though none of them is totally immune. *Casuarina equisetifolia* piles are commonly used for the construction of aquafarms along the southeast of India because of their easy availability, comparatively low cost and desired uniform shape. Studies of Nair (1956 b), Nagabhushanam (1960 a) and Edmondson (1955) on the natural durability of various species of timbers have shown that *Casuarina* is easily susoeptible to marine borer attack. In Karapad Creek oyster farm at Tuticorin, along with *Casuarina*, piles of *Tectona* are used, but the latter is also not resistant to the attack of neither teredinids nor pholadids. Along the coast of Krusadai Island three species of timbers (*C. equisetifolia*, *Thespesia populnea* and *Prosopis juliflora*) are used for pilings. Though none of them is totally resistant, among them *Prosopis* piles apparently give longer service than the other two. Considering its local availability and cheapness, it could be conveniently used for pilings in calm shallow coastal waters. Species of timbers belonging to the family Palmae (*Borassus flabellifer*, *Cocos nuelfera*, etc.) have been found to be comparatively resistant to the attack of teredinids and to a lesser extent to the attack of pholadids also (Nair, 1956 b; Santhakumaran, 1970). These timbers are likely to give longer service life in localities where crustacean borers like *Sphaeroma* and *Limnoria* are not active.

Though many types of mechanical, chemical, electrical and biological means to deter and destroy the boring pests are known, the destruction continues. Impregnating the timber with hydrocarbons and then charring them, desiccating the infested timber structures, exposure to fresh water, introduction of poisons to the ambient water, fixing hard particles or completely sheathing the exposed surfaces, exploding dynamite in water, electrolytic protection, electricution, chlorinating the water, etc. are some of the methods used against borers other than chemical preservatives on timber (Nair and Saraswathy, 1971). In aquafarms, chemical preservatives should be tried cautiously without affecting the culturing organisms. Purushothamam and Rao (1971) tentatively classified ascu, creosant and erosote-coal tar as good preservatives, creosote — fuel oil, pure creosote and copper resinate as moderate preservatives and pentachlorophenol as poor preservative, of the various types used. Initial settlement over creosoted piles may be delayed for some more time than in untreated piles. However, in localities like Mandapam, even creosoted piles are destroyed in less than six months duration after installation.

Tentative assessment of the loss due to borer activity in aquafarms

Casuarina has been the widely used timber as stakes and piles for oyster and sea-weed culture, in pens for fish culture, etc. along the southeast coast of India. In shalllow coastal areas, piles measuring 2 to 2.5 m length are preferred for these purposes and each pile costs about Rs. 7.50. For any small oyster farm about 100 such piles are necessary for installing stakes and for horizontal supports. The piles, even after the application of protective creosote coatings, commercially termed kriside, prior to installation have been found serviceable normally for about six months only, in the Gulf of Mannar. If two replacements are carried out annually, the cost of piles in a small farm alone would amount to about Rs. 1,500 per farm/year. Besides a sum of Rs. 1,000 would also be required for transportation, applying protective coatings, labour charges to install, etc. Altogether, in each farm, the destruction by marine borers would result in an annual economic loss of about Rs. 2,500. In our developing country where timber

resources are also becoming scarce, this loss is considerable. The extent of the loss is of special significance since this loss affects one of the most depressed and economically poor sections of the society namely the fihsing community.

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