OBSERVATIONS ON THE BREEDING ACTIVITY OF THE SHORE BARNACLE CHTHAMALUS MALAYENSIS PILSBRY IN BOMBAY HARBOUR

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ABSTRACT

A prerequisite for rearing larval stages of barnacles which form an important group amongst the fouling organisms is adequate knowledge of the breeding habits and the time and duration of their reproductive phase. Such knowledge is also useful for assessing the results of the field toxicity tests. The present paper gives an account of the breeding behaviour of the shore barnacle, *Chthamalus malayensis*, which is found in great abundance along the Bombay coast. If the breeding behaviour of *Ch. malayensis* reported in the paper represents that of other barnacles in Bombay harbour, the seasonal variations in barnacle settlement reported by other workers can be understood.

INTRODUCTION

FOR laboratory observations on development and toxicity tests of the fouling organism a liberal supply of the fertilized egg masses and larvae of the relevant organisms is necessary. It has been emphasised by Crisp[1], Crisp and Spencer[2], Becker[3], Bookhout and Crisp[4] and Patel and Crisp[5], that the liberation of larvae in the Laboratory would be possible if the breeding cycle of the animal is adequately understood. In the case of barnacle which form a major group of fouling organisms, if the breeding period is known, it may be possible to effect liberation of larvae in the laboratory tank as and when required, by suitably adjusting factors like food supply, oxygen concentration, salinity and temperature. In many western countries, liberation of the larvæ under laboratory conditions has been brought about by stimulating the mature individuals with a hatching substance which is a metabolic product of the barnacle tissues[2].

It has been held by Orton[6], Thorson[7] and Runnstrom[8] that the breeding of the majority of marine organisms is controlled by temperature of sea. Thorson[7] referred to this as Orton's rule. Recently Crisp[13] considered this view with respect to barnacles and explained how in tropical species breeding activities are controlled by temperature. Thus in India, where little is known about the life-cycles of fouling organisms, studies of their breeding behaviour assume special importance.

The object of the work presented in this paper is to ascertain the breeding behaviour and also the time and duration of the breeding phase of *Ch. malayensis* so as to facilitate laboratory breeding of the barnacle. Some preliminary observations on the breeding of *Ch. withersi* PILS., a species often closely associated with *Ch. malayensis* were also made for the purpose of comparison.

MATERIAL AND METHODS

Specimens of *Ch. malayensis* were collected from the wall of the boat-shed at the Naval Dockyard, Bombay. Collections were made twice or thrice monthly from April 1961 to June, 1962, about 25 specimens being generally examined at a time. During this period a total of 38 collections were made and approximately 950 specimens were examined. The observations on the breeding of *Ch. withersi* are based on 30 collections made during June, 1961 to May, 1962.

There are various methods by which the breeding periods of marine organisms could be established. As for barnacles, this could be done by regularly examining planktonic samples for nauplius larvae or else by studying their intensity of settlement on non-toxic panels immersed in the sea at regular intervals[17]. One more convincing and suitable method consists in making actual observations of the gonads for their stages of maturity over a period of one full year so as to bring out the seasonal sequence in gonad activity.

In the present investigation the condition of the ovary of *Ch. malayensis* was determined by microscopic examination, as against the visual estimation (with the naked eyes) adopted by Patel and Crisp[5]. On the basis of the development of ova the unfertilized specimens were conveniently divided into three categories and the fertilized ones into four categories. The percentage occurrence of individuals in each stage was determined for each month.

Stages of Ovarian Development

Condition of the ovary	Stage
Unfertilized specimens—	
(*i*) Ovarian organ not seen	0
(*ii*) Slightly developed ovary with immature ova	I
(*iii*) Ovary with clearly differentiated ova	II
Fertilized specimens—	
(*iv*) Fertilized eggs	III
(*v*) Eggs with embryos (nauplii) inside	IV
(*vi*) Free nauplii in the mantle cavity	V
(*vii*) Spent ovary	VI

In order to determine the stage of male genital organs, one of the vas-deferens together with its testicular follicles was removed from the body cavity of each individual and examined under the microscope,

RESULTS

Growth of the Ovary of Ch. malayensis

The paired ovaries of this barnacle can be easily located after tearing off the membranous base of the shell. The ovaries are situated on either side of the gut and appear whitish to creamy-orange in colour.

Stage: *0*

It appears that this stage when no ovarian tissue is noticeable, is of a very short duration and the individuals with this stage are observed only during the quiescent phase of breeding.

Stage: *I*—(Figure 1)

This stage is of very common occurrence being present during all the months of the year. It makes its appearance for the first time amongst individuals measuring 3 to 4 mm. in basal diameter. The ovary now exhibits a lamellar appearance, each lamella being studed with immature ova. This stage is also noticeable amongst individuals which may still possess the fertilized eggs or the free larvae of the previous broods.

Stage: *II*—(Figure 2)

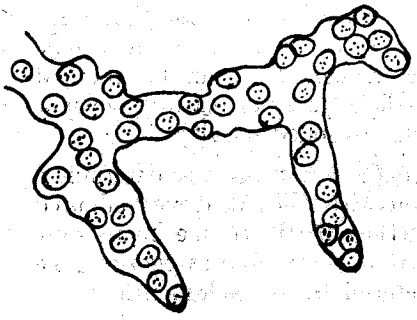

Fig. 1 Stage I
A lamella of the ovary with minute ova.

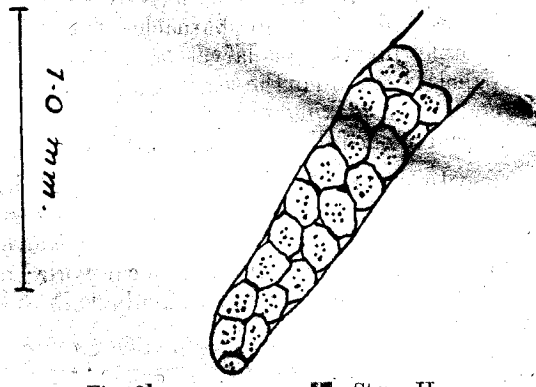

Fig. 2 Stage II
Ovary with clearly differentiated ova.

At this stage the lamellar appearance of the ovary still persists. The ova though clearly differentiated are not separated from one another and display polygonal shapes, as a result of mutual pressure. This stage, unlike stage I, lasts for a very short duration.

Stage: *III*—(Figure 3)

At this stage the egg mass is fertilized and transferred to the base of the mantle cavity where eggs are retained throughout the period of embryonic development. The time of retention of eggs is termed as incubation period by Barnes[14] and Barnes and Barnes[15]. The eggs are oval in shape and are enclosed in a common jelly-like matrix secreted by the glandular cells lining the paired oviducal openings. A careful observation shows the eggs undergoing cleavage activities similar to those described by Groom[9] in *Ch. stellatus* (POLI)

Stage: *IV*—(Figure 4)

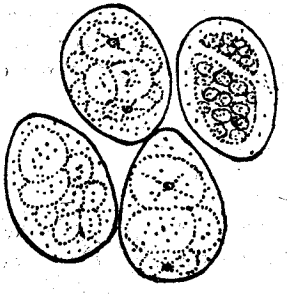

Fig. 3 Stage III
Fertilized eggs,

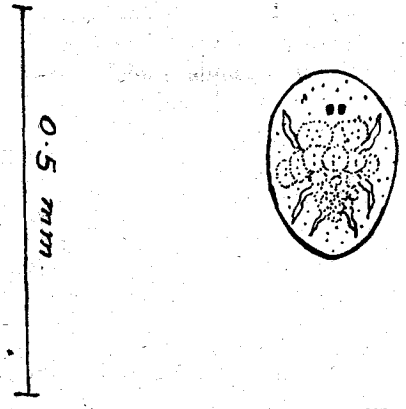

Fig. 4 Stage IV
Egg with nauplius inside (Semi-diagramatic)

It is the last phase of incubation of the eggs in the mantle cavity. This stage is characterised by the presence of embryonic larvae exhibiting naupliar eyes and the appendages—still in the egg cases. The eggs are enveloped in two common jelly like masses, one on either side of the gut.

Stage V—(Figure 5)

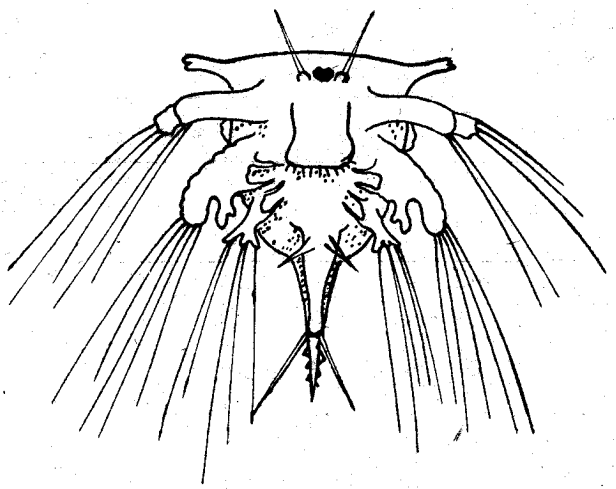

Fig. 5 Stage V

Free nauplius stage. (Semi-diagramatic).

At this stage the eggs hatch-out and a large number of nauplii is seen in the mantle cavity. The nauplii remain here until they are liberated. It has been shown by various workers like Crisp[16], Hutchins [20] and Barnes [18], [19] that the liberation of larvae in barnacles generally depends upon any one or more factors such as atmospheric and sea-water temperatures, salinity, oxygen concentration and also on the amount of available nutrients.

Stage : VI—

This stage is of a very brief duration and can be detected only if a few nauplius larvae fail to leave the mantle cavity of the parent.

It may be observed here that many individuals during active breeding show two or more of these seven stages simultaneously developing in the mantle cavity. It has been noticed that *Ch. malayensis* like some other barnacles produces more than one brood at a time and the incidence of individuals showing two broods at a time are not uncommon. In an individual measuring 9 mm. in a basal diameter as many as 1,890 larvæ have been counted and in one measuring 4 mm. 980 larvæ were encountered.

Condition of the Ovary of Ch. malayensis during different months of the year 1961-1962.

Table 1 shows percentage incidence of *Ch. malayensis* in relation to different stages of ovarian development over a period of 15 months. In many individuals, the ovarian tissu becomes absorbed, and is found to be absent during certain periods of the year. During July—August and December—January in particular, this is conspicuous. The incidence of individuals with Stage—0 is found to be minimum during February to May and again in September—October, when there is active breeding.

The individuals in Stage I, with immature ova are of common occurrence throughout This may be due to two factors. Firstly, *Ch. malayensis* continues to produce fresh masses of ova at all times of the year. Secondly, during the monsoon and winter, majority of the individuals is engaged in a rebuilding of the spent ovarian tissue, resulting in relative abundance of individuals in Stage I. This is evident from Table 1 where over 60 per cent of the individuals are in stage I during August-September period. Similar rise occurs during November and February.

TABLE 1.

SHOWING THE INCIDENCE OF CH. MALAYENSIS (EXPRESSED AS PERCENTAGE) IN RELATION TO DIFFERENT STAGES OF THE OVARIAN DEVELOPMENT DURING THE PERIOD APRIL 1961 TO JUNE, 1962

Period of examination	Stages of ovarian development							Total fertilized specimens (Stages III to VI)
	0	I	II	III	IV	V	VI	
*April (1961)	0	30	0	25	20	52	0	70
May	4	29	0	29	18	16	4	67
June	15	44	3	20	9	5	4	38
July	35	51	11	1	1	0	0	2
August	29	63	8	0	0	0	0	0
September	9	63	9	11	7	1	0	19
October	9	59	0	20	11	7	0	38
November	19	70	5	4	1	0	0	5
December	27	48	18	3	3	0	0	6
January (1962)	44	52	4	0	0	0	0	0
February	3	71	13	11	2	0	0	13
March	0	32	12	35	13	8	0	56
April	0	56	4	22	8	8	2	40
May	4	27	5	30	17	13	4	64
June	12	24	4	16	24	20	0	60

*This work started on 20th April, 1961 and only one collection could be made in this month.

Individuals in Stage II with clearly differentiated ova, are generally few, during April, May, June and again in October, when many of them breed actively. Absence of this stage during this period presumably due to rapid growth of ova, and a conspicuous rise during monsoon months and in December indicates the brief duration of this stage at the time of active breeding.

Table 1 will reveal that a fairly large number of individuals bear fertilized eggs undergoing cleavage (Stage III) during March, April, May, June and again during September-October, thereby indicating that the fertilization in this species occurs during summer. During January, July and August, on the other hand, there is practically no fertilization. The fertilization which starts again during the last week of September, reaches its maximum activity during October and dwindles during November-December.

Stage III is followed by Stage IV and V in quick succession. During March to June a large number of individuals show either developing larvæ or free larvæ in their mantle cavities. During monsoon period, July, August and September, the incidence of individuals with larvæ has been almost negligible but recurrence of larvæ during October is conspicuous. From November to February again, the occurrence of eggs with nauplli is insignificant.

Individuals in the 'spent' condition (Stage VI) could be noticed during May-June. Their absence during monsoon confirms the cessation of breeding during this period.

Breeding in Ch. malayensis

It is evident from the foregoing account that the period from March to June marks speedy maturation of the ova of *Ch. malayensis*. The results in Table 1 suggests a much faster rate of growth of ova during this period, especially as the incidence of individuals with Stage II could be rarely traced in the specimens observed.

The meagre incidence of 'spent' individuals during June and high incidence of maturing Stage I and II, immediately during July indicate that 'spent' condition of the ovary lasts for a brief duration. The majority of the individuals remains in Stage I during July and September. This period thus appears to denote the resting phase with reference to maturation. This condition recurrs during November—February period.

The occurrence of fertilized eggs and larvæ in the mon h of October suggests recurrence of conditions noted during March to June. The following phases in breeding of *Ch. malayensis* thus become evident.

(*a*) Active maturation and breeding during March to June.

(*b*) Lack of breeding during July—September.

(*c*) Recurrence of maturation and breeding during late September and October and

(*d*) Lack of breeding (quiescent phase) during November—February.

As has been mentioned before the alternation in the breeding and non-breeding phases observed amongst barnacles has been attributed to the effects of specific environmental conditions. However, this aspect has not been investigated in the present paper. The breeding activities observed after the monsoon in the month of October are of a very short duration. It is also seen that from November to February, specimens with fertilized eggs are negligible, suggesting the cessation of the breeding activity during this period. (Table 1).

Observations on male genital organs of Ch. malayensis.

In this species the vas-deferens is found to be distinctly present during all the phases of the life cycle. The vas deferens is generally full of sperms, only two individuals out of 950 being without sperms.

As a rule spermatogenic activity of this species has been found to continue uninterruptedly for the entire year (Table II). The activity was, however, found to be considerably high (over 60%) during the period, February to June, with a peak in March when nearly 86% individuals showed matured sperm follicles. In the month of October again, the incidence of individuals with matured sperm follicles was fairly high (55·2%).

Taking into consideration the maturation behaviour of the ovaries and testes in *Ch. malayensis*, it is evident that in a large number of individuals, the eggs and the sperms attain complete maturity during the same months of the year, thus facilitating large scale fertilization in the species.

TABLE II

SHOWING THE PERCENTAGE OCCURRENCE OF CH. MALAYENSIS IN RELATION TO THE CONDITION OF TESTES DURING THE PERIOD, JULY 1961 TO JUNE, 1962

Period of Examination	Mature testes	Immature testes	Testes absent
	(Number of individuals as expressed in percentage)		
July (1961)	27	4	69
August	40	15	45
September	50	21	29
October	55	14	31
November	44	24	32
December	40	20	40
January (1962)	34	34	32
February	66	3	31
March	86	0	14
April	68	0	32
May	70	1	29
June	60	0	40

Comparison of breeding habits of Ch. malayensis with those of other barnacles:

Excepting the preliminary observations reported by Paul[11] and Pillai[17], no systematic account of the breeding habits of Indian species of barnacles is available. Observations on breeding habits of *Ch. withersi* were therefore taken up during June 1961 to May, 1962 along with those on *Ch. malayensis*. In the course of this study it has been noted that a large number of individuals of *Ch. withersi* liberate nauplii during July, September to November and again in February-March. *Ch. withersi* therefore exhibits an ill-defined breeding season. Evidently this is in contrast with *Ch. malayensis* which shows breeding during the summer months. These two species of *Chthamalus* thrive under similar physical, chemical and nutritional conditions of the environment. It will be therefore interesting to ascertain whether these changes are of seasonal or purely phyiological nature. The breeding behaviour of *Ch. malayensis* also differs from that of *Balanus amphitrite variegatus* Darwin. It has been noted from the occasional observations made on *B. amphitrite variegatus* that this species bears fertilized eggs and larvæ during the monsoon months also. Pillai[17] has reported that *B. amphitrite communis* Darwin, from Kerala back waters breeds throughout the year with the maximum intensity during December to April and the lowest during the rainy months, May to August.

The breeding behaviour of *Ch. malayensis* nearly resembles that of temperate species *Elminius modestus* Darwin, *Balanus improvisus* Darwin, *B. amphitrite* Darwin and *B. crenatus* Brug., all of which produce several broods during summer but remains dormant during winter. This type of breeding differs from that of *B. balanoides* (L) and *B. porcatus* (da Costa) recorded in Great Briton[12], both of which give only one brood in a year. In Bombay, *Ch. malayensis* breeds continuously during the summer, but subsequently the breeding activity diminishes considerably and is almost negligible during the winter months.

Assuming that the breeding behaviour of *Ch. malayensis* reported in this paper represents the breeding behaviour of many other species of barnacles in Bombay harbour, the seasonal variations and particularly the high incidence of barnacle settlement during March to May and again in October reported by Iyengar *at al*[10] could be understood. It may also be brought out here that the breeding of *Ch. malayensis* during March to June and again in October and that of *Ch. withersi* during March, July and again from September to November indicate the possibility of obtaining larvæ over a major part of the year. This long range breeding behaviour of the two species put together will eventually facilitate the laboratory toxicity studies on the larvæ in view of which the present work was undertaken.

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