Reproduction of *Acanthaster planci* in Okinawa[1]

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Abstract

Reproduction of *Acanthaster planci* (L.) was studied using samples collected in Okinawa once or twice a month from February 8 to December 12, 1970. Mean gonad index (gonad weight per cent of body weight) increases steadily from February to early June, then declines rapidly from late June to early August. It remains at low levels from early August to early December or possibly to January except October when there is a secondary peak. Along with this change, there are parallel changes in the mean values and frequency distribution patterns of the size of egg cells and in the relative abundance of mature eggs. These and other evidences lead to a conclusion that in Okinawa *A. planci* breeds mainly during June and July and possibly during October. Nevertheless, the gonads contain mature eggs during other months, except December, though at very low ratios. It is not certain whether actual breeding takes place throughout the year.

INTRODUCTION

Answering the questions whether or not *Acanthaster planci* has a definite breeding season and if it does, when and how long the breeding takes place in a given geographical locality is important not only from the biological point of view but also from the practical viewpoints of deciding the most effective time when the control measures should be taken intensively if they are to be controlled. Although some informations on the subject were available at the time the present study was initiated at the begining of the year of 1970, these informations were obtained in the tropical areas and hence assumed to be not directly applicable to the starfish populations of Okinawa which lies in the subtropical area, namely between the latitudes of 26° and 27° N.

From previous works reported by such authors as Chesher (1969) and Cheney (1972) on the *A. planci* populations of Guam and other Micronesian islands, Pearsons and Endean (1969) on those of the Great Barrier Reef, Branham *et al.* (1971) on the Molokai population in the Hawaiian Islands, and Crump (1971) on the populations of the Port Sudan area of the Red Sea, a general statement can be deduced that *A. planci* has extended period of spawning season at lower latitudes whereas

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at higher latitudes it has a shorter and more clearly defined breeding season.

The present study made it clear that in Okinawa, *A. planci* has a short breeding season of one month or so, from June to July, and supports the last half of the above statement. In addition, some evidences were obtained which indicate that there is a secondary, probably incomplete breeding season in October.

MATERIALS AND METHODS

Samples of *A. planci* were collected once or twice a month during the period between Feb. 8, 1970 and Dec. 12, 1970. Collections were made at the southern and mid-western coasts of Okinawa Island. All the samples were collected on the reef edge within 5 meters in depth. At the time of collection, *A. planci* was not so abundant that considerable searching had to be made on some collecting dates. As a result, only small number of samples were collected on some days, and in almost all the samples it was not possible to keep the size of animals uniform. The collecting dates and number of starfish collected are as follows: Feb. 8, 3 female and 2 male; Feb. 22, 1 each; March 15, 3 female and 2 male; Apr. 24, 4 female and 5 male; May 23, 2 female and 1 male; June 7, 2 female; June 26, 4 female and 10 male; July 19, 3 female and 5 male; Aug. 9, 3 female and 3 male; Sept. 17, 3 female, 4 male, and 1 sterile; Oct. 25, 4 female and 2 male; Dec. 12, 3 each. The total number of samples was 74–34 female, 39 male and 1 sterile starfish. No collection was made in November.

All the animals collected were brought to the laboratory where their size, body weight and gonad weight were determined. The size of the starfish was measured from the tip of one arm to that of the tip of the opposite arm. The animals ranged from 19.0 to 45.0 cm in size and from 320 to 1950 g in wet weight with the mean values of 30.1 cm and 767 g, respectively. The mean values for the groups of animals collected on the same day ranged from 24.5 to 35.9 cm in size and from 600 to 904 g in body weight. Although the size and weight of the animals are more or less similarly distributed in each day-group, the animals collected in the earlier period of the year tended to be smaller than those collected in the later period. The mean size and body weight of the animals collected between Feb. 8 and June 7 are 25.3 cm and 605 g, respectively, while those collected during the period between June 24 and Dec. 12 are 32.1 cm and 822 g, respectively.

The gonads were completely removed from each individual. Care was taken not to damage any part of gonads. After determining the wet weight, all the gonads were preserved in 10 per-cent formalin for later analyses on the size of oocytes and anatomical and histological examinations. The gonad index was calculated by the formula;

$$\text{Gonad Index} = \frac{\text{Wet weight of gonad (g)}}{\text{Wet weight of animal body (g)}} \times 100$$

The size of oocytes was estimated by squeezing out the oocytes from each

ovary on the slide glass and measuring, under a microscope, the size of the longer axis of each oocyte with a micrometer. Measurements were made on 33 oocytes randomly selected from each of the three samples of ovaries taken out of each starfish. This means that altogether 99 measurements were made for each starfish. Unfortunately two females collected on Sept. 17 and all of those collected on Mar. 15 and Aug. 9 were not available for the measurements of the size of oocytes because of the accidental drying of the samples. The gonads of the March and August samples were also not available for the anatomical and histological studies described below. The oocytes were grouped into the size classes of 40 μ each from 40 μ upwards. Those less than 40 μ were included in the 0–40 μ class, although those less than 15 μ in the longer axis were not measured. The frequency histograms of the size classes were prepared for each month except March and August.

The anatomical features of the gonads were examined under a stereoscopic microscope. Histological sections were prepared from the gonads of one female and one male starfish, randomly chosen from each monthly sample. The samples of gonads were embedded in paraffin, sectioned at a thickness of 10 μ, and stained with hematoxylin and eosin.

The data for sea water temperature were obtained from the Ryukyu Weather Bureau (1968) and presented in Fig. 1. The data are arranged as the 10-day means of the daily mean temperature of the surface water at Naha, Okinawa, for 7 years from 1961 to 1967.

RESULTS AND DISCUSSION

Seasonal Changes in the Gonad Index

Seasonal changes in the gonad index are shown in Fig. 1. The mean gonad indices for both female and male steadily increased from February to June when they reached the maximum values, and from then to August the indices decreased rapidly with the lowest values attained in September. From August to December and probably to January these remained at low levels except in October when the graph shows the secondary peak. The patterns of the change are more or less similar for both sexes, although the female gonad index is generally higher than the male one.

Examination of the graph shown in Fig. 1 reveals a good correlation existing between the rising temperature in spring and the increase in the gonad index. It is suggested that temperature is a limiting factor for maturation and spawning of the reproductive cells in spring and that after spawning in summer with temperature maintained at optimum levels, a new process of gametogenesis proceeds at a rate determined physiologically until October when temperature decreases sufficiently to resume the limiting role. It is also suggested that temperatures below 25°C could be limiting to gametogenesis of *A. planci* in Okinawa. The fact that the gonad index of the October samples is considerably lower than those of the samples collected in June may have resulted from a shorter period of time than would be

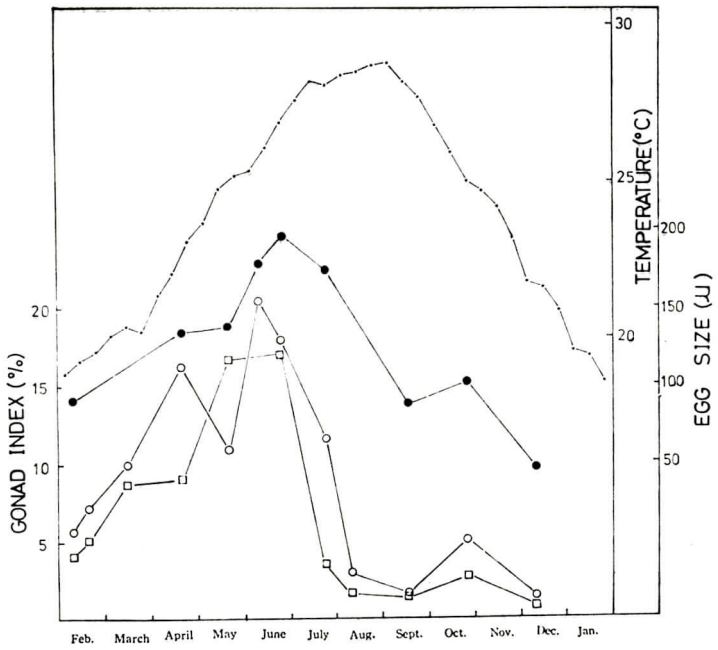


Fig. 1. Seasonal changes in the gonad indices of the female (—○—) and male (—□—) and of the monthly mean size of the oocytes (—●—) of *A. planci*. Ten-day means of the surface sea water temperature for 7 years from 1961 to 1967 are also plotted (-·-·-).

needed for completing a normal process of gametogenesis.

Seasonal Changes in the Size of Oocytes

The mean size of oocytes changed more or less parallel to the changes in the gonad index (Fig. 1). The graph again shows two peaks, one in June and the other in October, though the latter is not conspicuous.

The size frequency histograms shown in Fig. 2 clearly indicate that from December to July the relative abundance of the larger oocytes increases while that of the smaller ones decreases progressively. In both December and February the histograms are unimodal with the mode lying in the 40–80 μ class, though the relative abundance of the larger oocytes are greater in February than in December. The histograms become bimodal in April and remain so through October. In April and May the primary modes lie in the 40–80 μ classes while the secondary in the 200–240 μ class (April) or in the 240–280 μ class (May). The primary mode is shifted to the 200–240 μ class in June, to the 160–200 μ class in July and then to 0–40 μ class in September. In July there is a sudden decrease in the relative abundance of the oocytes larger than 240 μ in size. These changes suggest that the oocytes larger than 240 μ in size are mature and most actively released in July. There is a shift in the secondary mode from September to October, namely from

the 160–200 μ class to the 200–240 μ class. The general shape of the histogram of October closely resembles that of May. These can be taken as the evidences which indicate that some of the oocytes started to mature sometime after the majority had been released probably by August and reached maturity in October. These

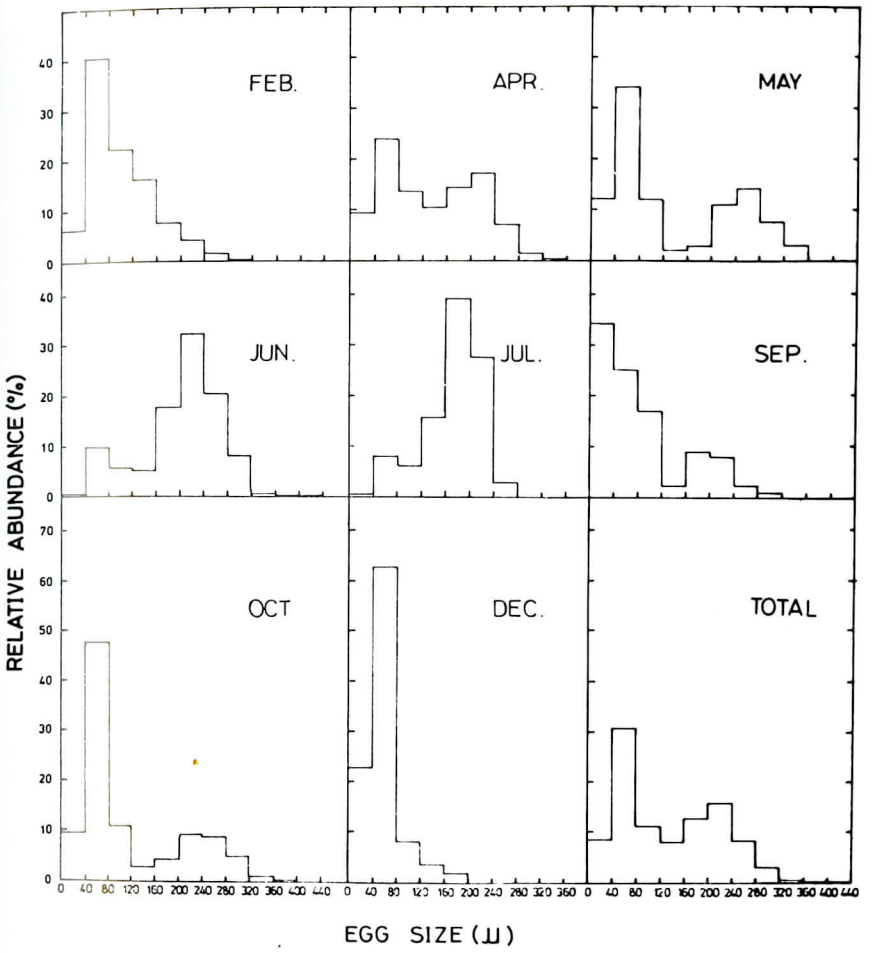


Fig. 2. Size frequency histograms of the oocytes of *A. planci*.

oocytes may be either released or absorbed within the ovary by December when the oocytes larger than 200 μ are not present at all and even those larger than 80 μ are very few.

This sequence of changes can also be shown by the seasonal change in the percentage of the oocytes larger than 240 μ in size, the change which proceeds as follows: 2.1 (Feb.), 9.4 (Apr.), 24.8 (May), 29.3 (June), 2.8 (July), 3.4 (Sept.), 15.3 (Oct.), and 0 (Dec.). The oocytes which can be regarded as mature are present, though with a varying frequency, in all the months studied except December.

Anatomical and Histological Observations

The gonads appear as bunches of grapes, 14 to 16 bunches being attached in a row to each side of the inner wall of the arms. The gonadal bunches contained in one animal are more or less similar in size except those lying at each end of the rows,

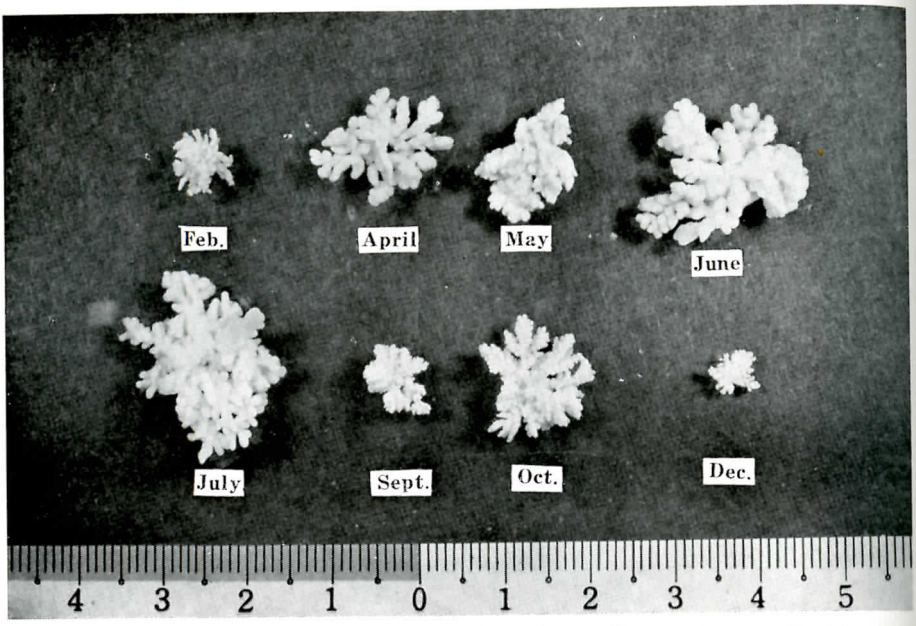
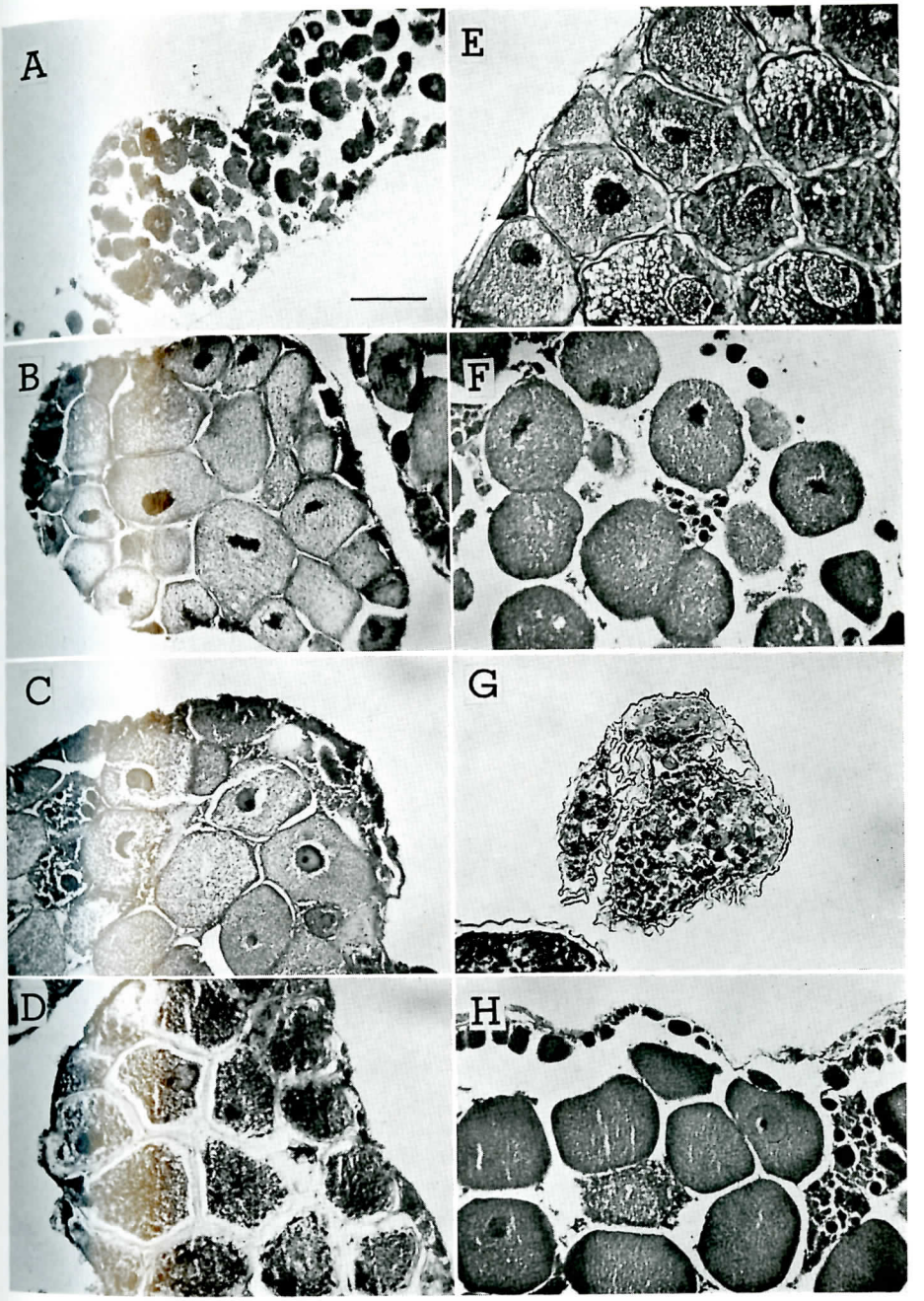


Fig. 3. Representative ovarian bunches of *A. planci*. The samples were fixed in 10% formalin. The May sample has contracted due to drying.

which are somewhat smaller than the rest. The bunches of grapes increase in size from December through February to June (Fig. 3). It can be observed from Fig. 3 that the change in the size of gonads parallels the change in the gonad index and the size of the oocytes (Figs. 1 and 2).

Examination of histological sections reveals that the size of oocytes is smallest in both December and September (Fig. 4, A and G). There is a rapid growth of

Fig. 4. Histological sections of the ovaries of *A. planci*. A: Dec. 12, immature oocytes. B: Feb. 8, some oocytes have reached full size, with the smaller oocytes attached to the ovarian wall. C: Apr. 24, two cells containing basophilic globules seen on the left are found among the growing oocytes. D: May 23, the connective tissue membranes surrounding the grown oocytes with each other are the most conspicuous among all the monthly samples. E: June 26, almost all the oocytes have reached full size. F: July 19, mature oocytes are seen free in the lumen, with some having turned into ova (note nuclei of some cells); a reticulate cell can also be seen in the center. G: Sept. 17, immature oocytes. H: Oct. 25, oocytes have reached full size again but no ova can be found; two reticulate cells with basophilic globules are seen on the right. The scale bar in A corresponds to 100 μ.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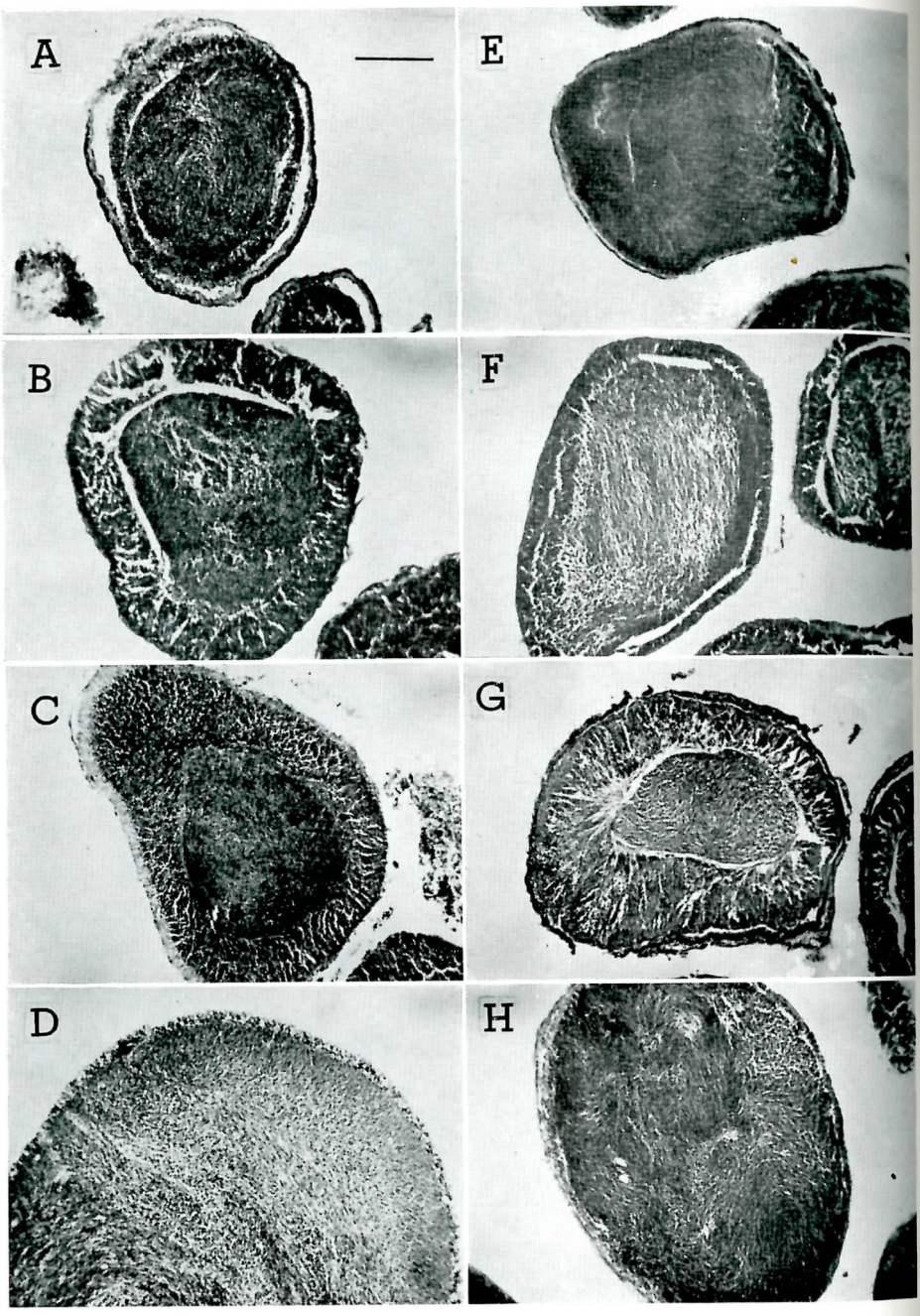
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oocytes from December to February and most of them reach full size in May and June. The larger oocytes tend to be accumulated in the center of the ovarian tubule, while the smaller are closely attached to the ovarian wall. Up to June the oocytes are connected with each other by thin layers of connective tissue, but in July they are free from each other and some of them have matured into ova (Fig. 4, D, E, and F). This state of ovary in July may be related to ovulation. The state of the ovary of the October samples resembles closely those of the July samples, except that no ova are contained in the former. If maturation of the oocytes of the July samples started in December and that of those of the October samples in September, there is a considerable difference in time needed for reaching maturity between the two groups. The shorter period for the October group could be accounted for by the optimum condition of temperature during the period of oogenesis. The ovaries of the April, July and October samples contain many reticulate cells filled with basophilic globules. These may be comparable to the nutritive phagocytic cells reported for various sea urchins (McPherson, 1969).

Histological observations on testes reveal that mature spermatozoa are present throughout the year, including September and December when the gonad indices are lowest (Fig. 5). However as is shown in Fig. 5, there is a cyclic change in the thickness of the germinal layer which contains spermatogonia, spermatocytes, and spermatids. The thickness of this layer is smallest in October (about 5 μ thick) and increases to a maximum value of about 40 to 80 μ in April. It then starts to decrease from May to a low value of about 5 to 15 μ in June, followed by an increase to a high value of 40 to 80 μ in September.

The changes in the thickness of the germinal layer would reflect the spermatogenetic activities of the testes. In October the testes are filled with spermatozoa, which will be released sometime in October or later. With the release of mature spermatozoa, the testes start to build up new germinal layer. The proliferation of spermatogonia and growth of spermatocytes would continue until April, from then on spermatocytes change into spermatozoa through spermatids. From April to June or July this process continues and more and more spermatozoa are produced, eventually leading to the liberation sometime in June or July. From probably August to September there is a rapid process of spermatogenesis which may lead into a second period of liberation of spermatozoa sometime in October or November. Like the female gonad index, the male gonad index of the October samples

Fig. 5. Histological sections of the testes of *A. planci*. A: Dec. 12, three layers can be recognized, the outer layer of the ovarian wall, germinal layer in the middle, and the central mass of spermatozoa. B: Feb. 8, the germinal layer has increased in thickness. C: Apr. 24, the germinal layer has reached a maximum thickness of about 40 to 80 μ. D: May 23, the ovarian tubule is filled with spermatozoa. E: June 26, the germinal layer exhibits the minimum thickness. F: July 19, the germinal layer starts to increase in thickness. G: Sept. 17, the germinal layer has reached the maximum thickness again. H: Oct. 25, the germinal layer has reached the minimum thickness again, with the tubule filled with spermatozoa. The scale bar in A corresponds to 100 μ.

is again much lower than those of the June or July samples. However the histological evidences for testes indicate much more clearly the presence of two periods of gametogenesis within a year than those for ovaries.

CONCLUSIONS

All the available data on the seasonal changes in the gonad index, the mean size of the oocytes, the size frequency histograms of the oocytes and the size of the individual bunches of gonads, and on the histological changes of gonads lead to a conclusion that in Okinawa, there is a definite breeding season for *A. planci* which lasts for about a month from June to July. More extended period of spawning have been reported for the tropical populations (Branham *et al.*, 1971; Cheney, 1972; and Crump, 1971). The latitudinal difference in the reproductive pattern has also been reported for other echinoderms (Pearse, 1969b).

These data also lead to a conclusion that there is a secondary period of potential breeding for the starfish in October. Although the low levels of October peak in the graphs for the gonad index and the mean size of oocytes reduces the significance of breeding in October, it does not necessarily rule out the possibility of actual breeding to take place in October. The presence of the oocytes regarded to be mature in relatively high percentage (15.3 per cent) and the histological evidences that both ovaries and testes are filled with mature reproductive cells suggests rather strongly that the starfish is at least potentially capable of breeding in October. A similar pattern of reproduction with two cycles of gametogenesis within a year has been reported for a zoanthid in Okinawa Island (Yamazato *et al.*, 1973). However there are no evidence which indicate definitely that *A. planci* does actually breed in October.

Although temperature may not be a sole factor involved in controlling the reproductive cycle of *A. planci* as has been suggested for sea urchins by many workers (Pearse, 1969a), it can be regarded as a principal factor for initiating the cycle at such a geographical area as Okinawa where temperature fluctuates considerably. Temperature may also be responsible for maintaining the synchrony of the cycle and for triggering the spawning. A first rise of temperature beyond 25°C or so which is encountered in May or June would be responsible for the spawning taking place in June. Temperature continues to rise until late August and early September (Fig. 1). The high temperature at this time of the year is favorable for rapid growth of the oocytes after the preceding group of oocytes were spawned. However temperature starts to drop below 25°C before gametogenesis is completed. This could explain the fact that the gonad indices in October are not as high as those of the June samples.

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