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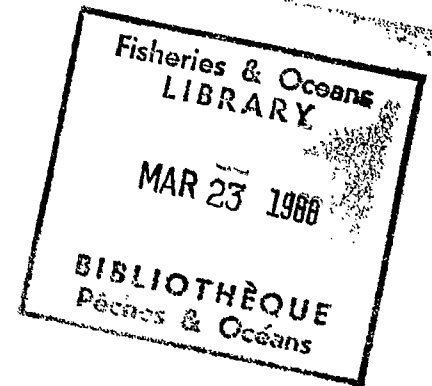
On the development of Pecten albicans Schroter

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TRANSLATOR'S NOTE

- \* I have translated this report as faithfully as possible.
  
- \* Title, Fig., Table and References(English section-- page 15 and 16 in my copy) are the original caption from Japanese text.

Many bivalves belonging to the Pectinidae family are dioecious. However, many Pecten species, such as Pecten maximus(L) are oviparous, self-fertilizing and hermaphroditic. (Coe 1943, Comely 1972, Fretter and Graham 1964, Gruffydd and Beaumont 1972, Tanaka 1971)

Until now, only a handful of studies on the development of Pecten species have been reported. Among the notable studies submitted on the foreign species, there are Comely's report(1972), Gruffydd & Beaumont(1970, 1972) and Dix & Sjardin(1975) on the larval development of Pecten maximus and Pecten meridionalis TATE.

Other studies of the Japanese species have been conducted by Horita(1977) who succeeded in developing the larvae of Pecten albicans SCHRÖTER; Tanaka & Del Rio(1978) who reported about the "i-chi-bu yo-se-i"(part-larvae) hatched from self-fertilized eggs and Semura, Takeuchi & Ōno (1983) who studied the outer-morphology of larvae based upon research studies "On the development of cultural system using coastal fishery resources".

In sandy areas off the coast, Pecten albicans SCHRÖTER has become a propagation target. Rapid increases in productivity are expected with the future advancement of technological development.

In anticipation, it is strongly desirable to gather information on the ecology, physiology and the morphology of Pecten albicans SCHRÖTER in the development stage.

This time, the early larval development stage of Pecten albicans SCHRÖTER and its morphology, focussed on the provincial aspect of the floating stage larvae, were observed. Some considerations were given to the results of our studies.

We would like to express our gratitude to Mr. H. Mizuno, Aichi Prefecture Fisheries Experimental Station, Obari Branch, who assisted in obtaining the stock specimen and Mr. Shigeya Yamamoto who assisted in rearing the <sup>ripe adults</sup> ~~mother-shell~~.

## MATERIAL AND METHOD

Firstly, spawning was induced. Normal Pecten albicans SCHRÖTER eggs, which attained about 100% fertilization, were used to obtain the larvae. The mature ~~mother shells~~<sup>adults</sup> (length 8cm - 10cm) were obtained from Ago Bay and were originally transplanted from Atsumi Sea, Aichi Prefecture for culture purposes.

On April 15(1980), <sup>artificial spawning was</sup> (eggs were) induced by using a thermal-stimulation method. The temperature of the sea water(20 l), containing the 6 ~~mother shells~~<sup>ripe adults</sup>, was raised a total of 5°C from the original 15°C during a period of 30 minutes; the process of raising the temperature was repeated 3 times in 30 minutes.

The larvae were kept in sea water(60 l) and a plankton "Bavlova lutheri" was fed to the larvae. The water temperature was kept between 18°C and 20°C, the salinity 33.5% - 34% and p.h. 8.3.

The provincular structures of the larvae were examined, applying treatment with hypochloric acid-liquid soda and glycerine jelly method(Tanaka, 1979).

## RESULTS OF THE OBSERVATIONS

Egg Development

Spawning of Pecten albicans SCHRÖTER, by thermal-stimulation method, occurred at 20°C. Half of the <sup>ripe adults</sup> ~~mother shells~~ used for the experiment produced large quantities of mature eggs which were fertilized. The egg shape was round and the colouration-orange. The yolk was distributed evenly inside the egg. A loss of the blastula was recognized. The diameter of the egg was approximately 70  $\mu$ m.

The developmental stages of the egg in relation to time after fertilization is shown in Table 1. It took approximately 40 hours for the fertilized eggs to develop into the D-type larvae of the early straight-hinge veliger stage in water temperatures of 15°C.

Table 1. Developmental stages of *P. albicans* at 15°C.

Time after fertilization (Approximately hours)	Stage attained	Time after fertilization (Approximately hours)	Stage attained
0.8	First polar body extruded	5	Sixteen-celled stage
1.0	Second polar body extruded	7	Morula stage
1.5	Formation of first polar lobe	9	Blastula stage
2	Two-celled stage	10	Gastrula stage
3	Four-celled stage	20	Trochophore stage
4	Eight-celled stage	40	Early straight-hinge veliger stage

Fig. 1(A-F) shows the cell division of Pecten albicans SCHRÖTER before the development reached the 4th cleavage stage. A polar lobe forms in the fertilized egg after the release of the 1st and 2nd polar bodies(B. pb) at the animal pole of the egg. This developmental process is embryologically identified among attachment-shell species as Crassostrea gigas; ma-ga-ki(Fujita, 1929), Mytilus edulis; mu-ra-sa-ki ga-i(Miyazaki, 1935) and Pinctada maxima; shi-ro-chō ga-i(Wada, 1942).

The egg reaches the 2-cell stage C (blastomere AB < CD) after the 1st polar body appears. Then, the 2nd polar body appears. The egg proceeds into the 4-cell stage(D) where the blastomere B and the blastomere D are connected at the side of the animal pole. The blastomeres are not equally divided (A < B < C < D).

The cell-cleavage at the 3rd and 4th cleave stage is shown in the Fig. 1(E and F). At the 3rd cleavage stage, the blastomere A at the 2nd cleavage stage divides into 1A and 1a revolving toward the right. At the 4th cleavage stage, 1A at the 3rd cleavage stage divides into 2A and 2a. Then, blastomere 1a divides into 1a<sup>1</sup> and 1a<sup>2</sup>, rotating left and reaches the 8-cell stage(E) and 16-cell stage(F).

When the fertilized eggs develop into the 4-cell stage,

"the decision of the normalcy" of Pecten albicans SCHRÖTER can be made. At this stage, the cleavage can be clearly observed and the length of the blastomeres can be measured.

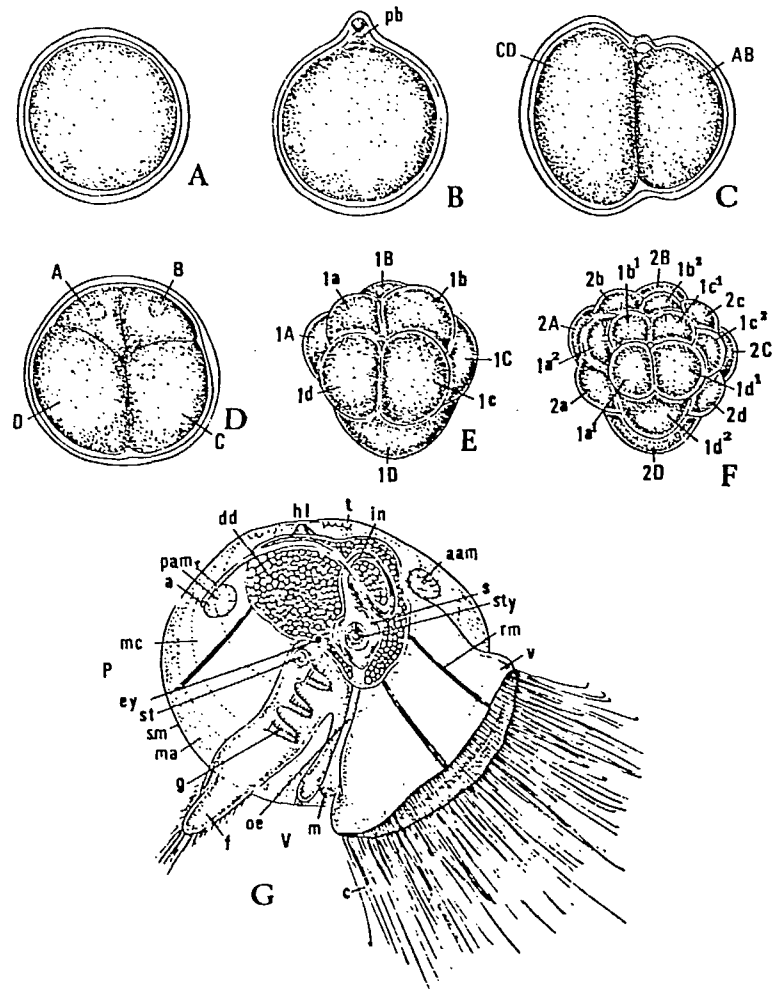


Fig. 1. Development of *Pecten albicans*. A, fertilized egg (70  $\mu$ m in diameter); B, same, protruded 2nd polar body; C, 2-celled stage; D, 4-celled stage; E, 8-celled stage; F, 16-celled stage; G, pediveliger larva with foot and velum extended, showing anatomical relationship, seen from right side (220  $\mu$ m in shell-length). a, anus; aam, anterior adductor muscle; c, cilia; dd, digestive diverticula; ey, eye; f, foot; g, gill; hl, hinge line; in, intestine; m, mouth; ma, mantle; mc, mantle cavity; oe, oesophagus; P, posterior; pam, posterior adductor muscle; pb, polar body; r, rectum; rm, retractor muscle; s, stomach; sm, shell margin; st, statocyst; sty, style sac; t, tooth; v, velum; V, ventral.

## Larvae Development

The shell length of Pecten albicans SCHRÖTER (larva of the D-type) is approximately 105  $\mu\text{m}$  and it is about 1.5 times larger than the egg diameter. The central hinge line of the larvae shell is slightly indented towards the stomach. In the central part of the velum, one flagellum is detected.

After being fed "P. lutheri" for a period of one or two days, the D-type larvae, in all instances, showed a stomach colouration of light orange. By observing the evidence of the colouration, future growth of the larvae was encouraging.

The growth curve of the larvae, which were reared at water temperature  $18^{\circ}\text{C}$  -  $20^{\circ}\text{C}$ , is shown in Fig. 2.

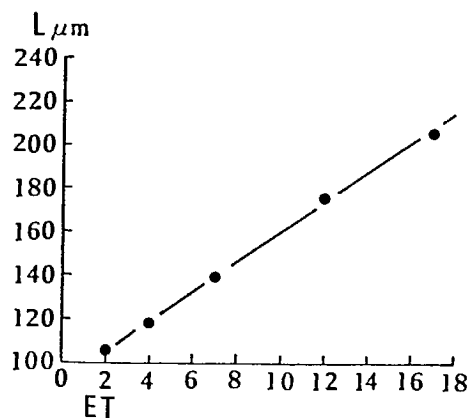


Fig. 2. Normal growth curve of the larvae of *P. albicans* at seawater temperature at 18 to  $20^{\circ}\text{C}$ . L, shell-length ET, elapsed time in days after fertilization.

Individual fast growing larvae, showed abnormal shell length attaining 200  $\mu\text{m}$ , 2 weeks after fertilization. The shell length of the abnormal larvae ranged between 210  $\mu\text{m}$  and 240  $\mu\text{m}$ ; they were smaller than the size of the *P. maximus* (250  $\mu\text{m}$ ) -- Gruffydd and Beaumont(1972).

The anatomical structure of Pecten albicans SCHRÖTER during the developmental stage (Fig. 1, G) shows the intestine(in) -- extension of the stomach(s) and oesophagus(oe) that began from the mouth(mo); which winds and twists once around the orange colored digestive-diverticula(dd) before eventually passing through the rectum(r) to end at the anus(a).

The pediveliger larvae possesses a pair of small eye-spots in the body that show a peculiar movement of repetitive turns towards the right side of the shell.

### Morphology

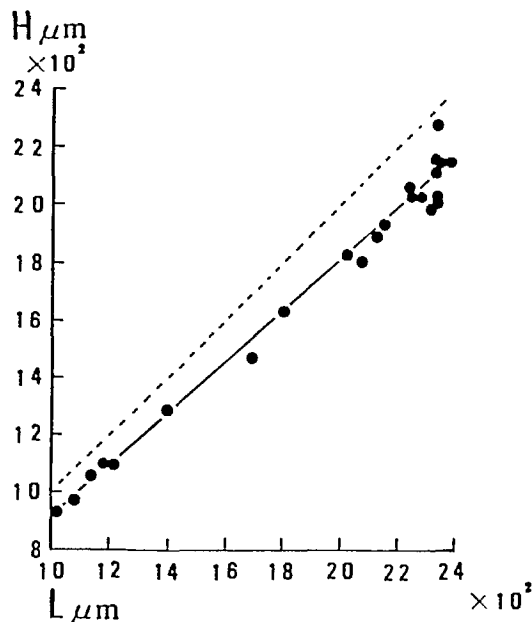


Fig. 3. Length-height relationship of the larvae of *P. albicans*. L, shell-length; H, shell-height.

The size of the larvae is comparatively small. The right hand side and the left hand side of the shell are equally shaped. The regression of the shell height(h) against the shell length is shown in Fig. 3.

$$H = 0.9L + 2(\mu\text{m})$$

The contour of the shell is clearly seen in Fig. 4. The front(anterior)-edge becomes narrower, the back(posterior)-edge shapes up round, the back-stomach-edge protrudes and the umbonal shape remains low even with growth. The growth ring is detailed and it is quite distinctive. The shell does not disclose any particular color.

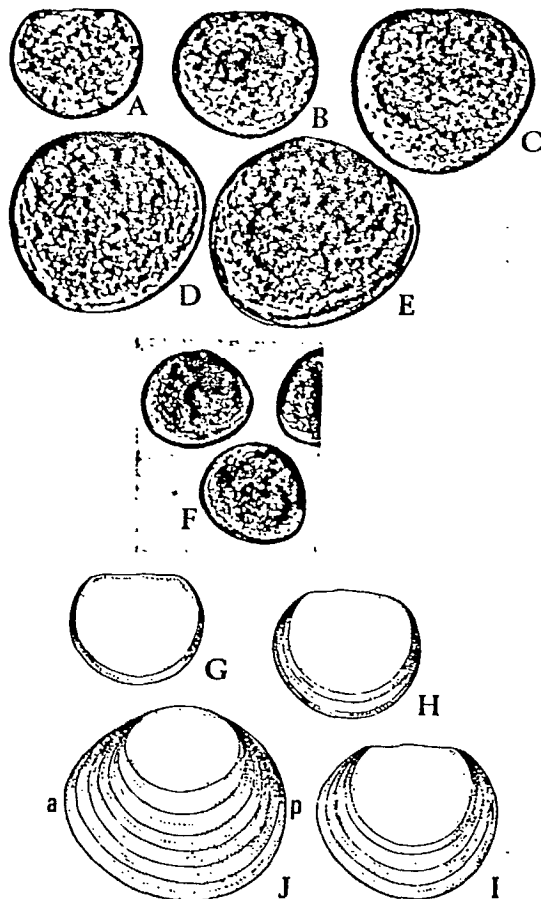


Fig. 4. Larval shells during growth in *P. albicans*. A, B, G, H, I and J, left valve; C, D, E and F, right valve. A and G, D-shaped stage, 110  $\mu\text{m}$ ; B and H, post D-shaped stage, 125  $\mu\text{m}$ ; C and I, early umbo stage, 145  $\mu\text{m}$ ; D, umbo stage, 180  $\mu\text{m}$ ; E, F, and J, metamorphosing stage, 215~225  $\mu\text{m}$ , in each shell-length; a, anterior; p, posterior.

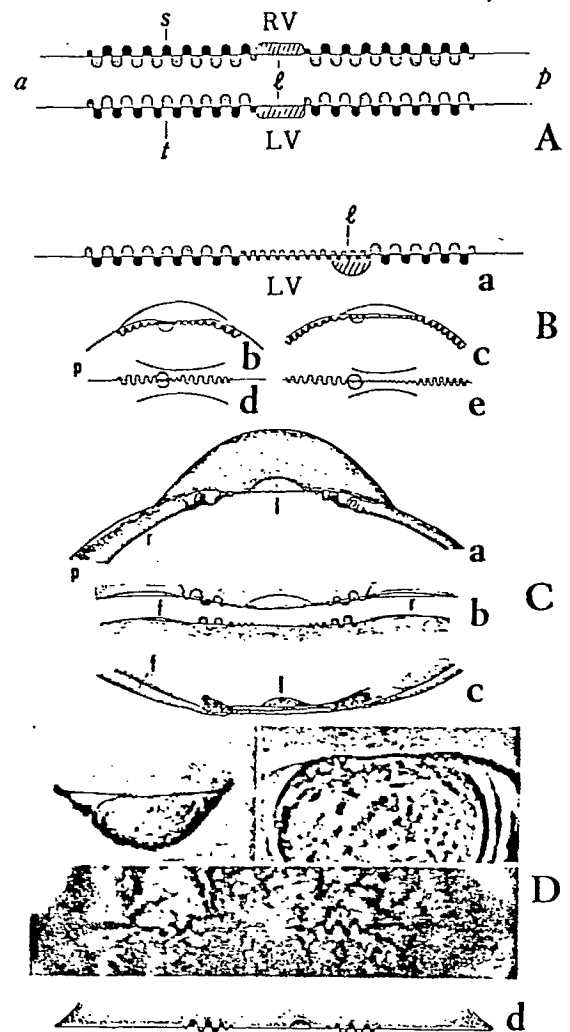


Fig. 5. Four types of the provincular structure of larval shells belonging to the Subclass Pteriomorpha. A, Arcidae (*Anadara*) — Basic type including Pinnidae, Pteriidae, Limiidae and Ostreidae; B, Mytilidae (b, d-*Modiolus*; c, e-*Mytilus*); C, Anomiidae (*Anomia*); D, Pectinidae (*Pecten albicans*). a, anterior; f, flange; l, ligament; LV, left valve; p, posterior; r, ridge; RV, right valve; s, socket; t, teeth.

Fig. 5 shows the provincular structure of a shell. The provinculum is rather thin, round and small. The ligament is situated in the centre between the central part and back part of the provinculum. The provinculum has many teeth and 3-4 equal sized micro-teeth can be observed in front and in the rear of the provinculum.

#### CONSIDERATION

##### Time requirement to reach the D-type stage.

It is believed that the most exhaustive life phase of Pecten albicans SCHRÖTER takes place between the beginning of fertilization and the D-type stage.

According to Gruffydd & Beaumont(1972), it takes approximately 50 hours at water temperature(16°C) for Pecten maximus to reach the D-type stage. However, Dix & Sjardin (1975) claims that it takes 3 days at water temperature(13°C - 15°C) for Pecten meridionalis, and Culling(1974) indicates that it takes a longer period of 4 days at water temperature

(12°C - 18°C) for Placopecten to develop into the D-type stage.

Our experiment took approximately 40 hours at water temperature (15°C), so the time consumption for Pecten albicans SCHRÖTER is shorter than in other species. However, because the spawning of Pecten albicans SCHRÖTER takes place during the winter and early spring (water temperature 10°C - 12°C), it seems that the time required for larval development is longer in a natural environment. The relationships between water-temperature, development time and larvae growth is discussed in the next section.

#### Morphology of the Larvae

According to Tanaka (1982), the provincial structure of the bivalve shell larvae is an important genetic key in classification. In regards to the provincial structure of Pecten albicans family larvae, Dix (1976) stated that Equipecten bifrons is simply a species that possesses typical teeth.

Chanley & Andrew (1971) claimed that they recognized the teeth among Aequipecten irradians larvae during metamorphosis, but they reported that it is difficult to photograph these teeth under an optical-microscope.

Culliney(1974) has also reported that he was unable to take good photographs of the provincular structure among Placopecten magellanicus larvae.

As we observed, Pecten albicans(Fig. 5, D) differ from the other pectenidae family larvae(Fig. 5, A-C), by possessing different characteristics of the provincular structure; ligament position and shape, teeth numbers and configuration, and also evidence of a side-provinculum.

Our findings do not agree with the opinion offered by Rees(1950), who stated that the provincular structure of Pecten albicans superfamily larvae and Anomia lischikei superfamily larvae; na-mi-ma-ga-shi-wa ga-i cho-ka, are the same and that their provincular structure resembles the provincular structure of Mytilus crassitesta superfamily; i ga-i cho-ka.

For the purpose of larvae identification, comparisons in larvae shapes were made between Pecten albicans SCHRÖTER and Chlamys farreri; a-zu-ma-ni-shi-ki ga-i, Jones & Preston) because they are analogous species; they develop during the same time of the year.

Fig. 6 shows the larvae of Chlamys farreri; the <sup>adults</sup> mother shells were transported from Yokohama Chisen. Eggs were obtained using a thermal-stimulation method. The D-type larvae were fed "Chaetoceros" plankton.

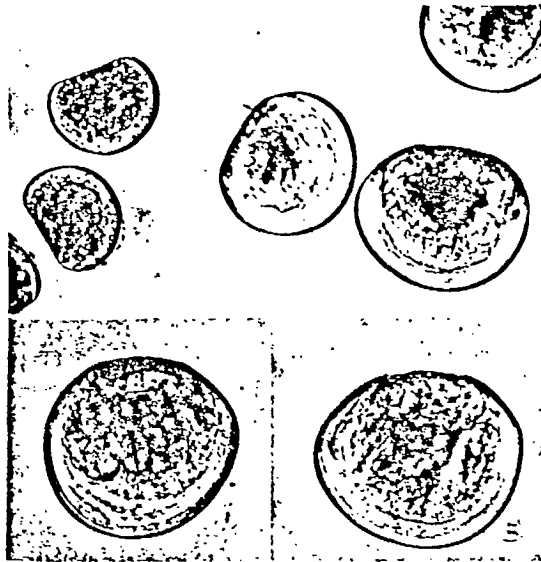


Fig. 6. Photomicrograph of the larvae of *Chlamys farreri* in comparison with *P. albicans* shown in Fig. 4. A, D-shaped stage, 105  $\mu\text{m}$ ; B, 125  $\mu\text{m}$ ; C, 140  $\mu\text{m}$ ; D, 165  $\mu\text{m}$ ; E, metamorphosing stage, 170  $\mu\text{m}$ , in each shell-length. a, anterior; p, posterior.

As indicated in Fig. 6, the shell contour (including the protruding umbo) of *Chlamys farreri* larvae resembles the shape of *Pecten albicans* SCHRÖTER. The back-stomach-edge of *Chlamys farreri* larvae is somewhat round and the left and right sides are not equal. The left shell is larger than the right shell, while colouration is light orange.

The left and right side shell of *Pecten albicans* SCHRÖTER larvae are equal. However, the right side shell is swollen at the development-throat. In order to gain knowledge on the morphological changes, it would be interesting to find out whether the left side shell becomes flat or not.

## SUMMARY

Embryological studies were conducted in April, 1980, in order to investigate the developmental stages and morphological characteristics of hermaphroditic Pecten albicans SCHRÖTER larvae with emphasis on the provincular structure.

By using a thermal-stimulation method, the fertilized eggs of Pecten albicans SCHRÖTER were raised to early D-type (shell length 105 $\mu$ m) larvae at water temperature 15 $^{\circ}$ C, 40 hours after fertilization took place. "Žavlova" were fed to the D-type larvae during the development. After 3 weeks rearing at 18 $^{\circ}$ C - 20 $^{\circ}$ C water temperature, Pecten albicans SCHRÖTER larvae reached metamorphosing stage. The shell length of the larvae at this time measured 210  $\mu$ m - 240  $\mu$ m.

Left and right side shell of the larvae are equal, the umbo is low and the front-edge is narrow and sharp.

The provincular structure of Pecten albicans SCHRÖTER larvae, is different from other species and therefore, is an important factor in larvae identification.

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