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Protogonyaulax (Dinophyceae) in the Gulf of Thailand[1)2)]

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Abstract

Four species of *Protogonyaulax*, *P .cohorticula*, *P. fratercula*, *P. leei* (Balech) comb. nov., and *P. tamarensis*, are found in the Gulf of Thailand. *P. cohorticula* and *P. fratercula* make chains of more than eight cells, although *P. tamarensis* and *P. leei* (Balech) comb. nov. less than four cells. Shapes of the four species are so similar that it is difficult to distinguish and identify clearly only from appearance without close observation of thecal plates. They are distinguished by criteria such as shape of an apical pore complex, position of a ventral pore, position of anterior and posterior attachment pores, and shape of a sulcal anterior plate. The most prominent characteristic of *P. cohorticula* is an anteriorly prolonged sulcal anterior plate. *P. fratercula* has an dorso-ventrally pointed apical pore complex. *P. leei* (Balech) comb. nov. is distinguishable from *P. tamarensis* by its ball-shaped outline and shallowly impressed sulcus. Though all the four species have a ventral pore, position of the pore of *P. leei* (Balech) comb. nov. differs from those of others. The pore of *P. leei* (Balech) comb. nov. is located inside the first apical plate, whereas others have the pore on the suture between the first and fourth apical plates.

In Thailand, paralytic shellfish poisoning (PSP) from eating green mussel *Perna viridis* occurred in May of 1983 at Pranburi (Fig. 1) and caused 63 people to become ill, one fatally. This is the first case of PSP recorded in Thailand (TAMIYAVANICH et al. 1985). In order to identify the causative organism of the PSP, we have monitored occurrence of toxic dinoflagellates in the Gulf of Thailand. Species belonging to the genus *Protogonyaulax*, especially, were observed in detail, because the toxin component of the shellfish collected just after the PSP incident indicated that *Protogonyaulax* was responsible (TAMIYAVANICH et al. 1985). The purpose of the present paper is to describe morphological characteristics of four *Protogonyaulax* species, which have been found to be endemic in the Gulf.

Materials and Methods

Phytoplankton samples were collected occasionally from Pranburi, Cha Am, Ban Laem, Maeklong river mouth, Chao Praya river mouth and Ang Sila from 1983 to 1986 (Fig. 1). Preserved plankton samples collected from offshore area in the Gulf of Thailand, in 1985 by one of the authors (P.P.), were also studied.

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[3)] タイ湾に分布する *Protogonyaulax*（渦鞭毛藻）
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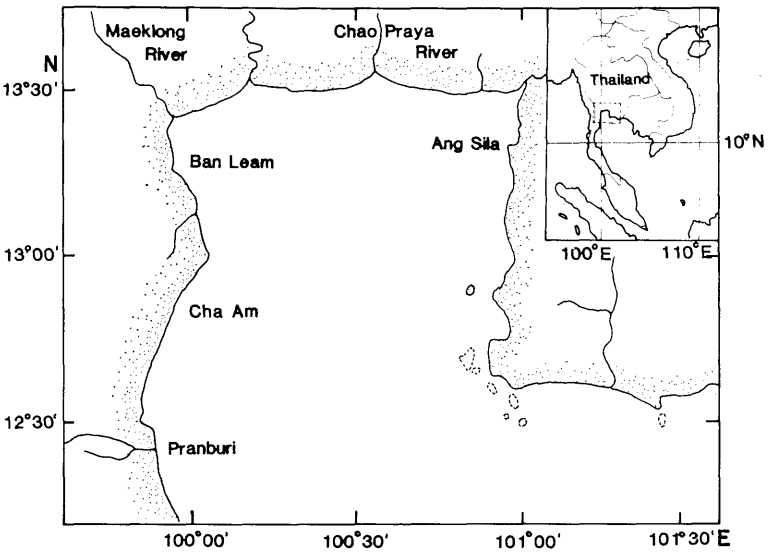

Fig. 1. Location of surveyed areas in the inner part of the Gulf of Thailand.

Morphological characteristics of *Protogonyaulax* were observed under a light microscope NIKON XF-NT21 with differential interference contrast after dissecting thecal plates using 5% sodium hypochlorite solution and staining with a solution of chloral hydrate, potassium iodine and iodine (FUKUYO & IMAMURA 1987). Cells in unialgal and clonal culture established from a planktonic cell in the Gulf of Thailand were also treated with the same procedure. Identification was ascertained by the original description of each species and publications concerning systematics of *Protogonyaulax* such as BALECH (1964, 1967, 1985), TAYLOR (1979, 1985), FUKUYO (1985a, b), and FUKUYO et al. (1985).

Observations

Division PYRRHOPHYTA Pascher
Class DINOPHYCEAE Fritsch
Order PERIDINIALES Haeckel
Family GONYAULACACEAE Lindemann
Genus *Protogonyaulax* Taylor, TAYLOR 1979, p. 51
Syn: *Gessnerium* Halim 1969, p. 619
Alexandrium Halim 1960, p. 102, emend. Balech 1985, p. 53

Plate formula is APC (apical pore complex), 4′, 6″, 6c, 8-11s, 5‴, 2⁗. APC is composed of two platelets, apical pore plate (Po) and apical closing plate (Pc). Pc plate partly closes ellipsoidal apical pore (a.p.). As Pc plate connects Po plate along right ventral margin, shape of a.p. changes to fishhook-like. Anterior attachment pore (a.a.p.) is found in Po plate of cells forming a chain, and is sealed when cells swim individually. An upper corner of first apical

plate (1′) touches APC. Some species have a ventral pore (v.p.) in 1′ plate or on suture between apical 1′ and 4′. Cingulum deeply concave and equatorial, descending its own width. The cingulum is composed of six plates nearly equal in length. Sulcus is composed of eight plates. Some species have three minute additional accessory platelets in sulcus. Large sulcal posterior plate (s.p.) is located at the center of hypotheca. Posterior attachment pore (p.a.p.) is found in s.p. Gap runs from margin to p.a.p. A.a.p. and p.a.p. are sealed up when cell division ceases. Second antapical plate does not contact sixth postcingular plate.

Large C-shaped nucleus is situated beneath cingulum. Cells making a chain have protoplasm connection through a.a.p. and p.a.p.

Protogonyaulax cohorticula (Balech) Taylor (Pl. I, 1-6, 9)

TAYLOR 1979, p. 51

Syn: *Gonyaulax cohorticula* Balech; BALECH 1967, p. 111, pl. 6, figs. 117-119, pl. 7, figs. 120-122.

Gonyaulax fratercula Balech; TAYLOR 1976, p. 101, pl. 35, fig. 392; *non* BALECH 1964, p. 31, t. 4, figs. 47-58.

Gessnerium cohorticulum (Balech) Loeblich & Loeblich; LOEBLICH & LOEBLICH 1979, p. 44.

Alexandrium cohorticula (Balech) Balech; BALECH 1985, p. 34, fig. 5.

Cell is round to subpentagonal in ventral view, and slightly wider than long (Pl. I, 1, 9). Left and right shoulders of epitheca are slightly convex. Hypotheca is rectangular. Sulcus is deeply impressed and fairly wide posteriorly. High wing-like lists develop at both right and left margins of sulcus. Thecal plates are sparsely porulated.

APC is triangular, narrower ventrally, and is rarely truncated at ventral end (Pl. I, 3). A.p. is narrow drop-shape and is located in left half of APC of cells having a.a.p., or at center of APC of cells without a.a.p. A.a.p. is large, round to short ellipsoidal, and is located between right side margin of a.p. and that of APC. P.a.p. is round and located at center of s.p. plate (Pl. I, 6). V.p. is clearly found near right corner of apical 1′ on suture between apical 1′ and 4′ (Pl. I, 3). Sulcal anterior plate (s.a.) has triangular or rectangular anterior expansion, which invades into epitheca (Pl. I, 2, 4, 5).

Length of cell is 30.3 to 50.0 μm, and width is 30.0 to 57.5 μm. Chains of more than eight cells are often found. Especially in culture, chains of more than 64 cells are formed.

This species was collected from off Ang Sila in July 1984 and 1985. This species occurs mainly at the center, offshore area in the Gulf of Thailand, and some cells in the innermost area of the Gulf (Fig. 2). KODAMA et al. (in press) found toxicity from two unialgal cultures originated from planktonic cells collected at Ang Sila.

Protogonyaulax fratercula (Balech) Taylor (Pl. I, 7-12)

TAYLOR 1979, p. 51; FUKUYO 1980, p. 50, figs. 1.8-2.c, 1.8-3.c.; FUKUYO 1985b, p. 24, fig. 2. 3. 4; FUKUYO et al. 1985, p. 29, figs. 16-19.

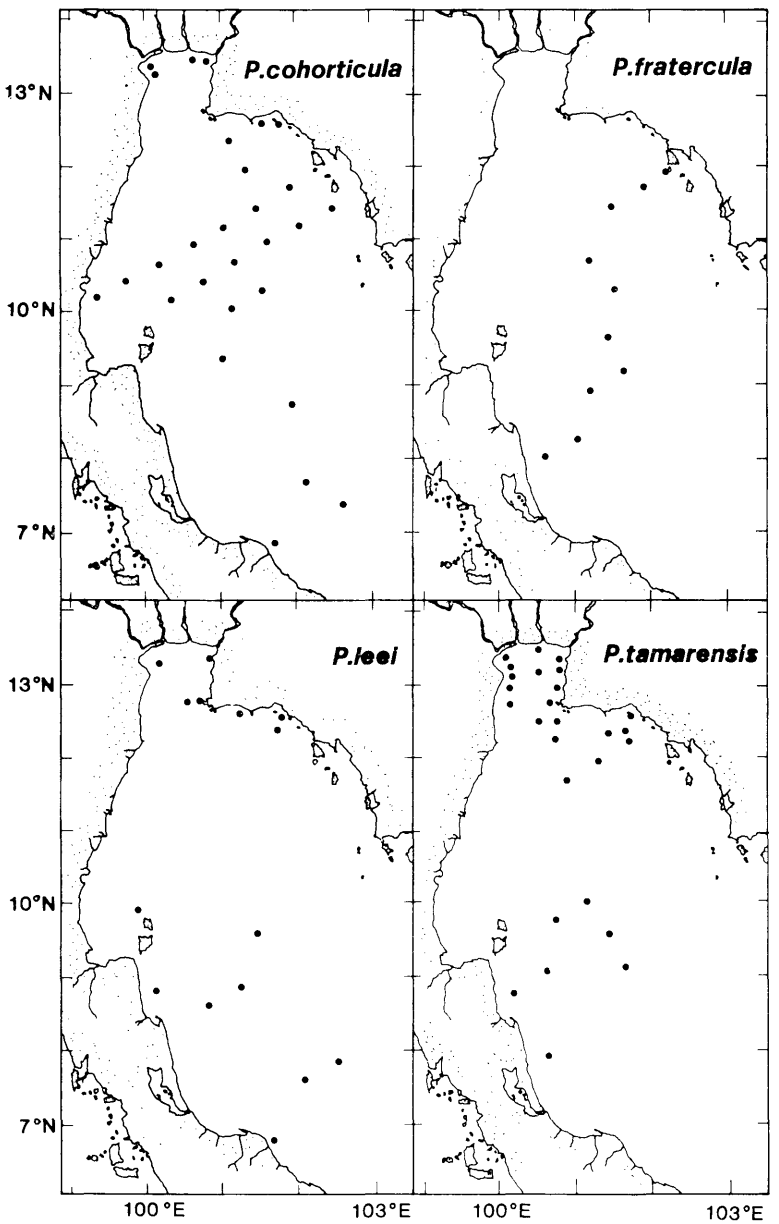

Fig. 2. Distribution of *Protogonyaulax* in the Gulf of Thailand. Solid circles show plankton sampling stations where each species was collected.

Syn: *Gonyaulax fratercula* Balech, BALECH 1964, p. 31, pl. 4, figs. 47-58.

Gesssnerium fraterculum (Balech) Loeblich & Loeblich; LOEBLICH & LOEBLICH 1979, p. 44.

Alexandrium fraterculus (Balech) Balech; BALECH 1985, p. 34, fig. 4.

Cell is round to subpentagonal in ventral view, and slightly longer than wide (Pl. I, 8, 9).

Both left and right shoulders of epitheca are slightly convex. Sulcus is deeply excavated and is widening posteriorly. High wing-like lists develop at both right and left margins of sulcus. Thecal plates are sparsely porulated.

APC is fusiform and pointed both ventrally and dorsally (Pl. I, 10). A.p. is small, narrow, and located in the left half of APC. A.a.p. is round and situated in right half of APC. P.a.p. is round and located at center of sulcal posterior plate (Pl. I, 12). V.p. is clearly found at middle of suture between apical 1′ and 4′ (Pl. I, 11).

Length of cell is 27.5 to 42.5 μm, and width is 27.5 to 42.5 μm. Chains of more than eight cells are often found (Pl. I, 7).

This species occurs mainly at the center, offshore area in the Gulf of Thailand (Fig. 2).

Protogonyaulax leei (Balech) comb. nov. (Pl. II, 1-5)

Basionym: *Alexandrium leei* Balech; BALECH, 1985, p. 37, fig. 8.

Cell is round, slightly longer than wide (Pl. II, 1). Epitheca is hemispherical. Shape of hypotheca is asymmetrical, height of left half is longer than that of right. Sulcus is shallowly impressed, and curves in the middle toward the right (Pl. II, 4). Cingulum is narrow in width, and excavated deeply. Thecal plates are porulated.

APC is narrow triangular to rectangular, prolonged dorso-ventrally and curves to left at middle (Pl. II, 2). Large, drop-shaped a.p. occupies the center of APC. Some pores are found in marginal area of APC. Two attachment pores, a.a.p. and p.a.p., are rarely found. A.a.p. cuts APC at middle of right margin. P.a.p. is small and located in right half of s.p. plate (Pl. II, 5). Weak gap is also found between p.a.p. and anterior margin of s.p. V.p. is located inside apical 1′. Pore does not touch suture between apicals 1′ and 4′ (Pl. II, 3). Narrow gap is sometimes observed between suture and pore.

Length of cell is 35.0 to 40.0 μm, and width is 35.0 to 42.5 μm. Chains of two cells are rarely found.

This species was collected from Ang Sila in February, 1986. This species is found in the innermost area and at the middle of the Gulf of Thailand.

Protogonyaulax tamarensis (Lebour) Taylor (Pl. II, 6-11)

TAYLOR 1979, p. 51; FUKUYO 1980, p. 50, figs. 1.8-2.a, 1.8-3.a.; FUKUYO 1985a, p. 531, figs. 2A-G; 1985b, p. 22, fig. 2.3.1; FUKUYO at al. 1985, p. 28, figs. 4-10.

Syn: *Alexandrium tamarense* (Lebour) Balech; BALECH 1985, p. 38, fig. 20.

See FUKUYO (1985a) for other synonyms.

Cell is round and slightly longer than wide (Pl. II, 1). Sulcus is weakly impressed, and widened posteriorly. Antapex is slightly concave. Cingulum is deeply concave. Thecal plates are thin and sparsely porulated (Pl. II, 7, 9).

APC is triangular or rectangular, narrower ventrally, with large drop-shaped a.p. (Pl. II, 7, 8). A.a.p. is round to short ellipsoidal and is located dorsally between right margin of a.p. and APC (Pl. II, 7). P.a.p. is found in the right half of s.p. plate near margin between fifth

posterior plate (Pl. II, 10). A.a.p. and p.a.p. are sometimes sealed (Pl. II, 8, 11). V.p. is clearly found at near middle of suture between apical plates 1′ and 4′ (Pl. II, 8, 9).

Length of the cell is 25.0 to 32.5 μm; width is 25.0 to 32.5 μm. Chains of two cells rarely found.

This species was collected from the Pranburi River estuary on June 21, 1983 and February 7, 1984. This species was also found from offshore the Gulf of Thailand, mainly innermost area (Fig. 2). KODAMA et al. (1987a) could not find any toxicity from four unialgal cultures originated from planktonic cells collected at Pranburi and Ban Laem.

Discussion

The shapes of species belonging to the genus *Protogonyaulax* are so similar that it is almost impossible to distinguish and identify by looking only the outline of motile cells. The criteria used for species differentiation are the shape of the apical pore complex, position of the ventral pore, position of the anterior and posterior attachment pores, and the shape of the sulcal anterior plate.

LOEBLICH & LOEBLICH (1979) and BALECH (1985) considered the contact between the first apical plate and the APC as a criterion for species but not genus and they independently adopted the genus name *Gessnerium* and *Alexandrium*, respectively. However, the presence or absence of the contact affects plate configuration which is one of the most basic criteria of the rank of genus but not of species. Therefore, we adopt *Protogonyaulax* for species having the contact, and *Alexandrium* which is senior synonym of *Gessnerium* for ones lacking the contact. Here we place the following species in *Protogonyaulax: Protogonyaulax tropicale* (Balech) comb. nov., basionym: *Alexandrium tropicale* Balech, BALECH 1985, p. 37, fig. 7; *Protogonyaulax leei* (Balech) comb. nov., basionym: *Alexandrium leei* Balech, BALECH 1985, p. 37, fig. 8; *Protogonyaulax lusitanicum* (Balech) comb. nov., basionym: *Alexandrium lusitanicum* Balech, BALECH 1985, p, 37, fig. 16.; *Protogonyaulax fundyense* (Balech) comb. nov., basionym: *Alexandrium fundyense* Balech, BALECH 1985, p. 37, fig. 18.

Among the four species described in this paper, *P. fratercula* and *P. cohorticula* make chains of more than eight cells, although *P. tamarensis* and *P. leei* (Balech) comb. nov. less than four cells. Between the former two species *P. cohorticula* is distinguished from *P. fratercula* by the anteriorly expanded sulcal anterior plate. In the latter two species *P. leei* (Balech) comb. nov. is distinguishable from *P. tamarensis* by its dorso-ventrally elongate APC.

P. leei (Balech) comb. nov. has a ventral pore inside the first apical plate instead of those on the suture found in the other three species. This characteristic is also found in *Protogonyaulax kutnerae* (Balech) Sournia, but the latter is distinguishable from the former species by its anteriorly expanded sulcal anterior plate (BALECH 1979). APC of *P. fratercula* is dorso-ventrally prolonged as same as that of *P. leei* (Balech) comb. nov., but it differs in that both dosal and ventral ends are pointed and a small apical pore in the left half is present.

The APC of *P. tamarensis* and *P. cohorticula* are both triangular to rectangular and it is impossible to distinguish them by this characteristic. However, the position of the posterior

attachment pore in the sulcal posterior plate is different in these two species. *P. tamarensis* has this pore in the right half, and in *P. cohorticula* it is at the center. The prolonged sulcal anterior plate is a distinctive character of *P. cohorticula* and can be used to separate it from *P. tamarensis*.

Reports of the occurrence of the four species are rare in tropical waters. As for *P. cohorticula*, there are only two records. One is the original description by BALECH (1967). He found for the first time in the plankton samples from the Gulf of Mexico and described intensively the morphological characteristics. The second finding is TAYLOR's (1976) monograph on *P. fratercula* in the Andamann Sea. His clear drawning of anteriorly expanded sulcal anterior plate is the most prominent characteristic of *P. cohorticula* according to BALECH's (1967) criteria. In addition, his description on the shape of the sulcal anterior plate shows that his *P. fratercula* is reconsidered as *P. cohorticula*. Therefore, his report shows that *P. cohorticula* is distributed in the Andamann Sea as well as in the Gulf of Mexico. Our results show the occurrence of the species in the Gulf of Thailand, suggesting that it is widely distributed in tropical waters. As KODAMA et al. (in press) found toxin production in *P. cohorticula*, we have to monitor cell densities of not only *Pyrodinium bahamense* var. *compressum*, which is a well-known toxic species endemic in tropical waters (HARADA et al. 1982), but also *P. cohorticula* for research on paralytic shellfish poisoning occurring in tropical areas.

P. leei (Balech) comb. nov. was originally reported from Korea by BALECH (1985). Our finding is the second one. Although his specimen is larger than that of this report, the authors consider the difference is caused by the life stage of the species, i.e. vegetative cell and planozygote. The authors (FUKUYO & YOSHIDA unpublished) found the vegetative cell and planozygote of the same species from Kagoshima Bay in Japan, and noticed that the wide variation of cell size was caused by the large planozygote.

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Explanation of Plate I

Planktonic cells and their thecal characteristics. (Scale bar=10 μm)

1-6, 9, *Protogonyaulax cohorticula*

1: Ventral view of a two-cell chain.
2: Ventral area, showing displacement of a cingulum and an anterior expansion of a sulcal anterior plate.
3: Apico-ventral view, showing APC and a ventral plate.
4, 5: Ventral view, showing a ventral pore and an anterior expansion of a sulcal anterior plate.
6: Sulcal posterior plate with a posterior attachment pore.

7-12, *Protogonyaulax fratercula*

7, 8: Ventral view of a chain.
9: Ventral view of a chain (left), comparing to that of *P. cohorticula* (right).
10: APC with an anterior attachment pore.
11: Epithecal ventral plates (1′, 4′, 6″), showing a ventral pore.
12: Hypothecal plates (s.p., 2‴′), showing a posterior attachment pore.

Explanation of Plate II

Planktonic cells and their thecal characteristics. (Scale bar=10 μm)

1-5, *Protogonyaulax leei* (Balech) comb. nov.

1: Ventral view of a cell.
2: APC without an anterior attachment pore.
3: Apical 1′, showing a ventral pore.
4: Antapico-ventral view, showing a sulcus bending toward right.
5: Sulcal platelets, showing a sulcal posterior plate after being sealed a posterior attachment pore.

6-11, *Protogonyaulax tamarensis*

6: Ventral view of a two-cell chain.
7: APC with an anterior attachment pore and two apical plates, (2′, 3′).
8: Epithecal plates consisting of APC, 1′, 3′, 4′, and 1″-6″.
9: Apicals 1′ and 4′, showing a ventral pore.
10: Sulcal posterior plate having a posterior attachment pore.
11: Sulcal posterior plate after being sealed the posterior attachment pore.

PLATE I

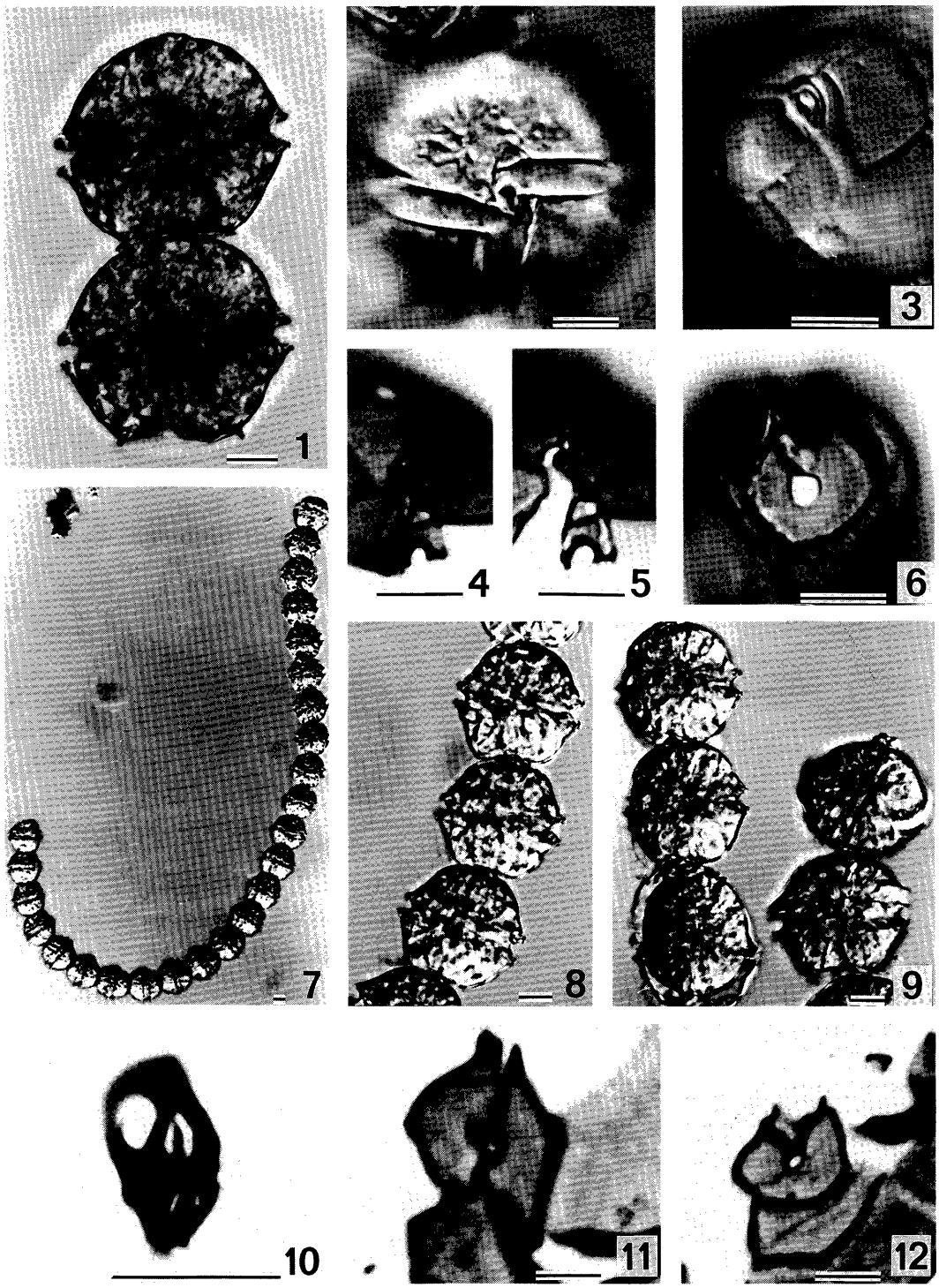

PLATE II