植物研究雑誌
J. Jpn. Bot.
82: 296–304 (2007)

Morphological Investigation of *Perissonoë crucifera* (Kitton) Desikachary & al. Collected from Japan

Tsuyoshi WATANABE[a], Hidekazu SUZUKI[a], Tamotsu NAGUMO[b] and Jiro TANAKA[a]

[a]Department of Ocean Sciences, Tokyo University of Marine Science and Technology,
4–5–7, Konan, Minato-ku, Tokyo, 108-8477 JAPAN;
E-mail: tsu4shi_w@yahoo.co.jp
[b]Department of Biology, The Nippon Dental University,
1–9–20, Fujimi, Chiyoda-ku, Tokyo, 102-8159 JAPAN

(Received on April 28, 2007)

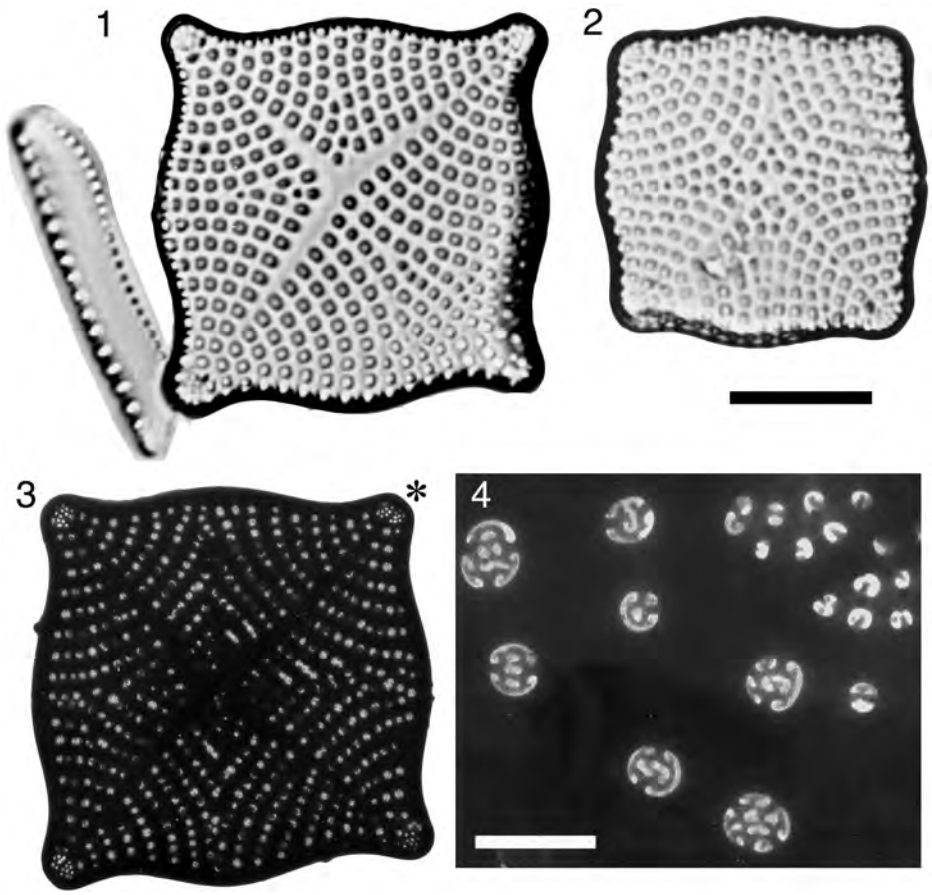
The tropical and subtropical marine diatom taxon *Perissonoë crucifera* (Kitton) Desikachary & al., was obtained from the coast of Iriomote Island, Okinawa Pref., for the first time from Japan. Frustule is quadrate in valve view, composed of two valves, a valvocopula and a pleura. Each valvocopula and pleura comprises four quarter bands (QBV and QBP) with four corner bands (CBV and CBP) covering the openings between QBs. CBP extends along the bottom sides of two pleurae. Granules are scattered over the outer surface of a valve. In a mantle, many granules and small areolae are alternately arranged in a line. On both surfaces of a QB, areolae are arranged in a single line like those on a valve. Areolae on a valve and mantle are occluded by concentric rotae and are supported by two to four spokes. In the apical pore fields, there are smaller areolae perforated by rota-like segments.

Key words: Bacillariophyta, corner band (CB), *Perissonoë crucifera*, quarter band (QB), valve morphology.

Perissonoë crucifera was originally described as *Amphitetras crucifera* Kitton (Kitton and Pritchard 1861) and later transferred to the genus *Rhaphoneis* Ehrenb. (Hagelstein 1938). The genus *Perissonoë* was established by Andrews and Stoelzel (1984) based on *Amphitetras cruciata* Janisch & Rabenhorst with the second species *P. trigona* (Grunow) Andrews & Stoelzel. Desikachary et al. (1987) noted that *P. cruciata* was a synonym of *P. crucifera*, the latter having nomenclatural priority; they added the third species *P. pentagona* Desikachary & al. *Perissonoë crucifera* is found in recent materials and *P. trigona* has been reported from both recent and fossil marine waters in the Miocene (Hajós 1974).

Perissonoë crucifera occurs in tropical and subtropical marine waters of the Indian, Atlantic and Pacific Oceans but has never been reported from Japan (Janisch and Rabenhorst 1863, Kitton 1867, Hendey 1970, Foged 1975, Li 1978, Giffen 1980, Desikachary and Prema 1987, Desikachary et al. 1987, Desikachary 1989, Beltrones and Castrejón 1999) nor have any living cells been observed. According to Andrews and Stoelzel (1984), this taxon thrives best in clear, shallow and moderately agitated marine waters of high productivity. It attaches to hard inorganic substrates.

The genus *Perissonoë* belongs in the family Rhaphoneidaceae and it is closely related to *Rhaphoneis* and *Delphineis* as they share similarities of valve structure and morphology of areolae and rimoportulae (Round



Figs. 1–4. *Perissonoë crucifera*. Fig. 1. Valve view in LM with a quarter band (left). Fig. 2. Valve view in LM. Fig. 3. Valve view in TEM. Fig. 4. Enlargement of the part with asterisk in Fig. 3. Showing the apical pore field (upper right) and areolae occlusion. Some small pores of apical pore fields having rota-like segments. Scale bars = 10 μm (Figs. 1–3), 1 μm (Fig. 4).

et al. 1990).

According to Andrews and Stoelzel (1984), the girdle of *Perissonoë crucifera*, which consists of four segments, is located at each side of the valve. Pores occur in a single row and are arranged near the margin of a segment. The segment is warped to fit the edge of the valve; each segment terminates with rounded ends. Round et al. (1990) described the apical pore fields as clusters of small areolae without vela, occurring at each angle of a valve. The rimoportulae are located in the center of each angle and have paired lips. Some taxa lack rimoportulae while others have one to four. *Perissonoë crucifera* has two in the adjacent angles.

Morphological information available for the girdle is poor, especially for araphid diatoms, although some information is available (Tanaka and Nagumo 2004). Morphological information of the girdle is sparse in *Perissonoë*. The girdle morphology of *Perissonoë* and the related genera *Rhaphoneis* and *Delphineis* have not been observed in detail (Andrews 1975, 1977, 1981, Round et al. 1990).

This study presents details of the fine structure of a valve and a girdle of *Perissonoë crucifera* for the first time.

Materials and Methods

Specimens of *Perissonoë crucifera* were obtained from three samples taken on the coast of Iriomote Island (24º21´N, 123º43´E), Okinawa Prefecture, southern Japan. One sample was collected by T. Nagumo on April 24th 1982 (TN 0500), the other two were collected by T. Watanabe on October 16th and 17th 2005 (TW 0018, TW 0025). The samples were collected from neighboring localities where the seagrass *Thalassia hemprichii* (Ehnenberg) Ascherson, and seaweeds *Turbinaria ornata* (Turner) J. Agardh, *Halimeda opuntia* (L.) Lamouroux, *Acetabularia ryukyuensis* Okamura & Yamada and *Neomeris annulata* Dikcie occur.

The samples contained hard organic matter composed mostly of sand grains and a few dead coral parts. They were treated with the bleaching method (Nagumo and Kobayasi 1990, Nagumo 1995) as described below.

The sample containing living specimens was washed 5 times with distilled water. A drop of bleaching agent or a sodium hypochlorite (NaClO) solution was added to a drop of washed sample on a slide glass and left for 1 to 3 minutes. Some specimens were selected from the sample and washed 5 times in distilled water using a glass capillary pipette on a glass slide under a light microscope (LM). The specimens were finally transferred to cover glasses that were dried on a hot plate. Some cover glasses were then mounted on to slides using mount medium (Pleurax) for LM observations while other cover glasses were mounted on stubs for scanning electron microscopy (SEM). The stubs were coated with platinum using a Hitachi E-1030 ion sputter coater. For transmission electron microscopy (TEM), the cover glass was replaced with a copper mesh grid.

LM observations were undertaken using a Nikon Optiphot. SEM observations were undertaken using Hitachi S-4000 and S-5000 at an accelerating voltage of 2 or 3 kV. TEM observations were undertaken a JOEL-2000EX.

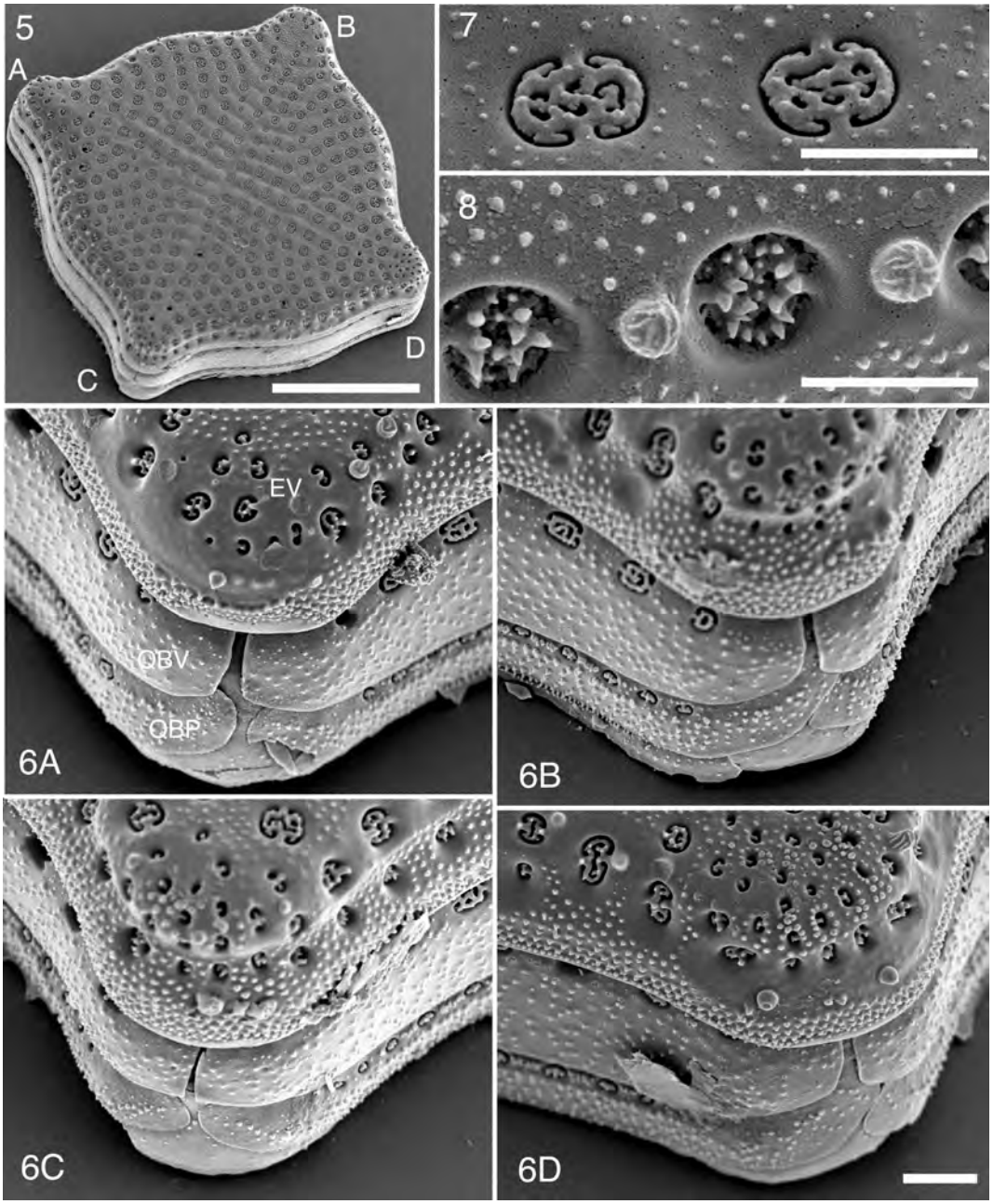
Morphological terminology follows Hendy (1959), Anonymous (1975), von Stosch (1975), Ross et al. (1979), Cox (2004) and Kobayasi et al. (2006).

Results and Discussion

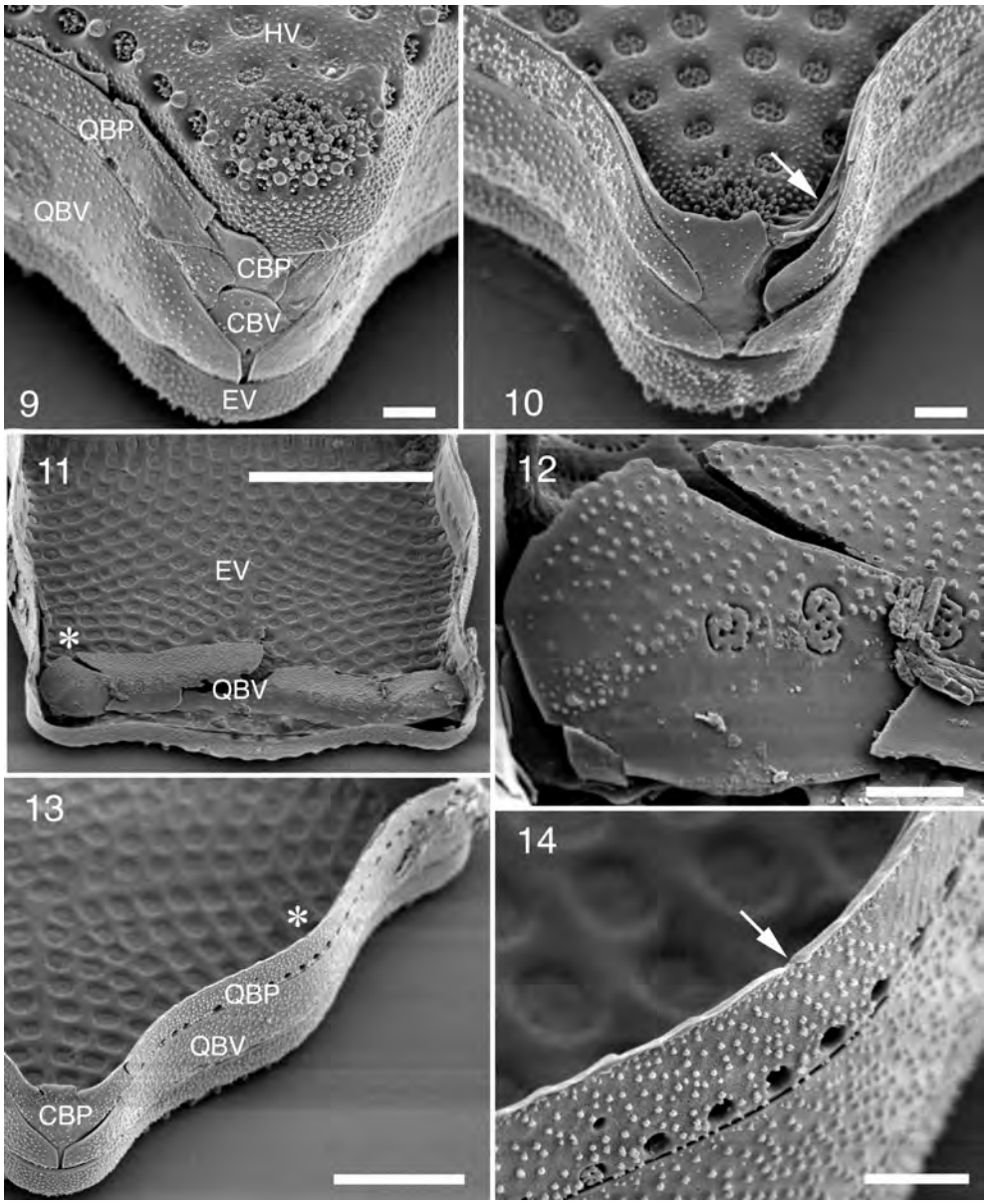
The samples from the shore of Iriomote Island were collected during the spring low tide at a water depth of 0.5–1.5 m. The water was clear and the sediments contain a few living organisms in our localities. Single frustules of *Perissonoë crucifera* attaches to the small sand grains; the species does not form colonies nor does it attach to the larger sand grains or to living organisms such as algae and seagrasses. *Delphineis* occurs prominently in eutrophic waters as long chains in the plankton (Fryxell and Miller 1978).

Andrews and Stoelzel (1984) collected *Perissonoë crucifera* from two localities in Barbados, West Indies. The first one a fringing reef near the shore at the water depth of 3–5 m, and the second one a submerged barrier reef platform about 0.6 km from the shore at a water depth of approximately 10 m. *Perissonoë* appears to thrive best in clear, shallow, moderately agitated marine waters, with a preference for a firm substrate for attachment; it does not attach to living substrates.

Perissonoë crucifera has been reported from Taiwan at about 23ºN for its northern limit (Li 1978) to Dar-es-Salaam, Tanzania at 6ºS (Foged 1975) for its southern limits. According to Andrews and Stoelzel (1984), *Perissonoë* thrives between 30º north and south in tropical and subtropical marine



Figs. 5–8. *Perissonoë crucifera*. SEM. Fig. 5. Valve view. Figs. 6A– 6D. Enlargement of four corners of A to D in Fig. 5. Note valvocopula and pleura are separated at all corners. An apical pore field consists of some small pores occluded by rota-like segments in Fig. 6D. Fig. 7. Enlargement of areolae occluded by rota. Fig. 8. Rotae and spinules on valve shoulder. EV = epivalve, QBP = quarter band of pleura, QBV = quarter band of valvocopula. Scale bars = 10 μm (Fig. 5), 1 μm (Figs. 6–8).



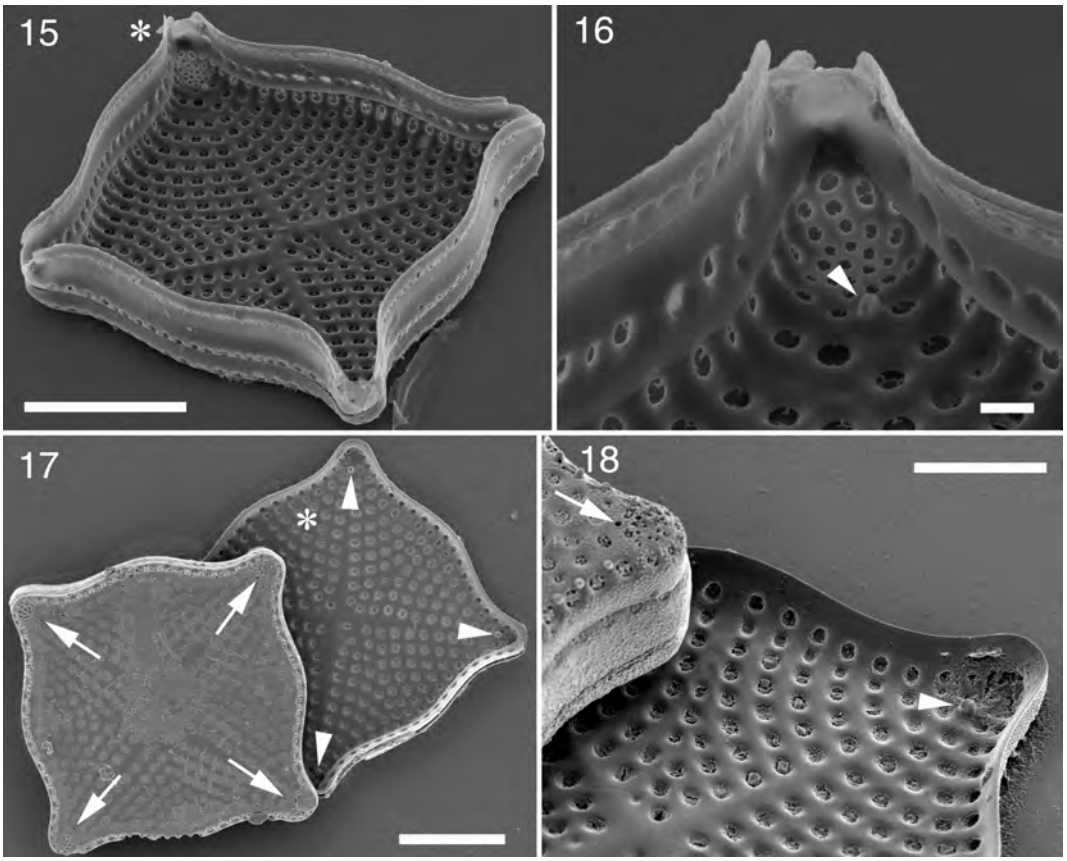
Figs. 9–14. *Perissonoë crucifera*. SEM. Figs. 9, 10. External view of the corner. Arrow shows QB side of horseshoe-shaped CB. Fig. 11. Whole a QBV. Fig. 12. Edge of QBV. Enlargement of the part with asterisk in Fig. 11. Note a single row of areolae lies on a valvocopula. Areolae shape is similar to valve ones. Fig. 13. Side view of an epitheca. Fig. 14. Enlargement of the part with asterisk in Fig. 13. Note CBP is stretched towards the center (arrow). CBP = corner band of pleura, CBV = corner band of valvocopula, EV = epivalve, HV = hypovalve, QBP = quarter band of pleura, QBV = quarter band of valvocopula. Scale bars = 1 μm (Figs. 9–10, 12, 14), 10 μm (Fig. 11), 5 μm (Fig. 13).

waters. Our localities (24º21´N) are the most northern reported so far.

The valves of *Perissonoë crucifera* are quadrate in valve view, and its margin is almost straight or undulate (Figs. 1–3, 5). The distance between opposing apices of a valve is 22 to 29 μm. The valve face is plain with a shallow mantle (Fig. 5). Granules are found on the external valve surface (Fig. 7) but not on the internal one. A single row of spinules, which are ridged, also occurs on the valve shoulder (Fig. 8, about valve shoulder see Kobayasi et al. 2006, p. 28, fig. 18). Those areolae and spinules are alternately arranged in a row (Figs. 8, 9). Granules are found on the valve surfaces of species in *Perissonoë* and *Rhaphoneis*. Spinules or round spines are found on the valve shoulder of *Perissonoë* and *Delphineis*. The spinules of *Perissonoë* have a wrinkled surface, whereas the round or blunt spines of *Delphineis* do not (see round spines of *D. surirella* (Ehrenberg) Andrews in Andrews 1981, Fig. 1; blunt spines like small teeth of *D. karstenii* (Boden) Fryxell in Fryxell and Miller 1978, Figs. 1–4). Sterna are present, radiating from the center but does not form a perfect cross (Figs. 1–3, 5). The striae are uniseriate, curving slightly towards the sterna (Figs. 1–3, 5). Areolae are round, occluded by rotae with concentric slits supported by two to four spokes, slightly indented (Figs. 5, 7, 8). Rota consisted of concentric slit with two to four spokes, is common among species in *Perissonoë* and *Rhaphoneis*. Apical pore fields occur at each of the four angles (apices) of a valve (Figs. 3–6). The specially reduced smaller areolae of the apical pore fields are perforated by rota-like segment (Figs. 4, 6). The external openings of the rimoportulae are close to the apical pore fields (Figs. 9, 10). Apical pore fields without velum is observed in species of *Rhaphoneis*. On the other hand, in *Delphineis*, there are one or two small apical pores representing the apical pore field. Interstriae and sternae are raised above the inner surface of a valve (Fig. 15). The sessile rimoportulae have paired lips and occur near the angles of apical pore fields on both surfaces of the valve (Figs. 15–18). Rimoportulae are common in three genera, but their number per a valve differs from each other. *Rhaphoneis* and *Delphineis* have two rimoportulae per a valve.

The epicingulum consists of two bands, a valvocopula and pleura (Figs. 5, 6A). The valvocopula is composed of four quarter bands (quarter band of valvocopula = QBV) and four corner bands (corner band of valvocopula = CBV), the latter covering the open ends of the QBs (Figs. 6A–D, 9, 10). The pleura has the same structure as the valvocopula (quarter band of pleura = QBP and corner band of pleura = CBP in Figs. 6A–D, 9, 10), although the valvocopula is thicker than the pleural band when widths are compared (Figs. 9, 10, 13). QBs warp to fit the edges of the valve, each band having a rounded end (Figs. 11, 12). Granules are scattered on external surface of both the valvocopula and pleura (Figs. 6A–D) but are absent from the internal surface (Fig. 12). Both QBV and QBP have a single row of round areolae (Figs. 6A, 6B). Areola of QB is occluded by rota, with a structure identical to that on the valve surface (Fig. 12). The shape of CB differs between valvocopula and pleura. CBV forms a triangle plate (Fig. 16). On the other hand, CBP extends along the lower sides of two QBPs (Figs. 9, 10, 13); the lower side is undulated to fit the pleurae (Figs. 9, 14). The ‘horseshoe’ shaped part of CB fits to QB (Fig. 10). The relationship between the various components of the epitheca is illustrated in a schematic drawing (Fig. 19).

The girdle of *P. trigona* and *P. pentagona* have never been observed, so it is not known whether their bands are separate or closed. QB and CB described in this study are not only known in any closely related genera nor



Figs. 15–18. *Perissonoë crucifera*. SEM. Fig. 15. Internal view. Fig. 16. Enlargement of the part with asterisk in Fig. 15. Note rimoportula (arrowhead) is seen between areolae and apical pore fields. Fig. 17. External and internal view of valves of the same cell. Arrows show external openings of rimoportula and arrowheads show rimoportula. Fig. 18. Enlargement of the part with asterisk in Fig. 17. Arrow shows external opening of rimoportula and arrowhead shows rimoportula. Scale bars = 10 μm (Figs. 15, 17), 5 μm (Fig. 18), 1 μm (Fig. 16).

any other diatom taxa.

It is clear that CB covers the slits of QB as a ligula. Thus, one might ask why QB is separated into four segments. It is suggested that the four segments of QBs are required to cope with the size reduction of a cell. The valves and pleurae are formed within the parent frustule, as diatoms have the peculiar property that the mean cell size usually decreases with each cell division within a population (Round et al. 1990). The pleurae are formed within a valve, and therefore the size of the daughter valves are restricted to the size of the parent pleurae. Usually a pleura is closed or open only at one side. Such pleura can never change the valve size. In *P. crucifera*, the daughter cell is not restricted to its parent valve size and can retain approximately the original size because the pleurae are free from each other by being separated as QBs.

Andrews and Stoelzel (1984) suggested that *P. crucifera* may be variant of *P. trigona*; as the latter occurs as a fossil and its morphological features appear intermediate between those two taxa. QBs and CBs of *P.*

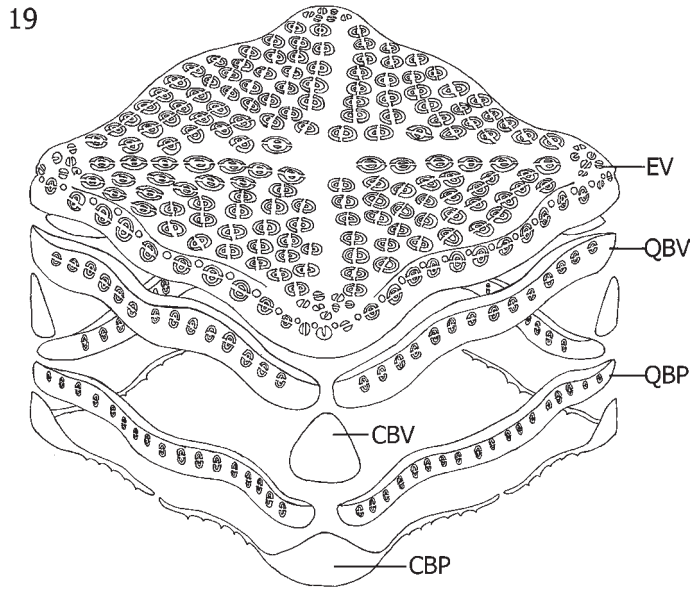


Fig. 19. Schematic drawing of several components of an epitheca in *Perissonoë crucifera*. CBP = corner band of pleura, CBV = corner band of valvocopula, EV = epivalve, QBP = quarter band of pleura, QBV = quarter band of valvocopula.

crucifera are unique among the related species. This girdle structure acquired in its evolutionary process may be one of the specialization strategies for the epipsammic niche.

Andrews and Stoelzel (1984) concluded that *Perissonoë* is closely related to *Rhaphoneis* and *Delphineis*, and *Perissonoë* is perhaps closer to the rhombic-lanceolate forms of *Rhaphoneis* than *Delphineis*. Furthermore, morphological and ecological features of *Perissonoë* differ from *Delphineis*.

We are grateful to Dr. David M. Williams (Department of Botany, The Natural History Museum) for helpful discussion and critical comments on the manuscript. We also thank Dr. Richard M. Crawford (Alfred Wegener Institut für Polar und Meeresforschung) for improving our English.

References

Andrews G. W. 1975. Taxonomy and stratigraphic occurrence of the marine diatom genus *Rhaphoneis*. Nova Hedwigia Beih. **52**: 193–227.

—— 1977. Morphology and stratigraphic significance of *Delphineis*, a new marine diatom genus. Nova Hedwigia Beih. **53**: 243–260.

—— 1981. Revision of the diatom genus *Delphineis* and morphology of *Delphineis surirella* (Ehrenberg) G. W. Andrews, n. comb. *In*: Ross R. (ed.), Proceedings of the 6th Symposium on Recent and Fossil Diatoms. pp. 81–92. O. Koeltz, Koenigstein.

—— and Stoelzel V. A. 1984. Morphology and evolutionary significance of *Perissonoë*, a new marine diatom genus. *In*: Mann D. G. (ed.), Proceedings of the 7th International Diatom Symposium. pp. 225–240. Science Publishers, Konigstein.

Anonymous 1975. Proposals for a standardization of diatom terminology and diagnoses. Nova Hedwigia Beih. **52**: 323–354.

Beltrones D. A. and Castrejón E. S. 1999. Structure of benthic diatom assemblages from mangrove environment in Mexican subtropical lagoon. Biotropica **31**: 48–70.

Cox E. J. 2004. Pore occlusion in raphid diatoms – a reassessment of their structure and terminology, with particular reference to members of the Cymbellales. Diatom **20**: 33–46.
Desikachary T. V. 1989. Marin diatoms of the Indian Ocean region. Atlas of diatoms. Fascile VI, pp. 1–27. TT. Maps & Publications Private Limited, Madras.
—— and Prema P. 1987. Diatoms from the Bay of Bengal. Atlas of diatoms. Fascile III. pp. 1–10. TT. Maps & Publications Private Limited, Madras.
——, Gowthaman S., Hema A., Prasad A. K. S. K. and Prema P. 1987. Genus *Perissonoë* (Fragilariaceae, Bacillariophyceae) from the Indian Ocean. Current Sci. **56**: 879–882.
Foged N. 1975. Some littoral diatoms from the coast of Tanzania. Bibliot. Phycol. **16**: 3–126.
Fryxell G. A. and Miller W. I. 1978. Chain forming diatoms: three araphid species. Bacillaria **1**: 113–136.
Giffen M. H. 1980. A checklist of marine littoral diatoms from Mahe, Seychelles Islands. Bacillaria **3**: 129–159.
Hagelstein R. 1938. The diatomaceae of Porto Rico and Virgin Islands. New York Acad. Sci. Survey of Porto Rico and Virgin Island **8**: 313–450.
Hajós M. 1974. Faciological and stratigraphic importance of the Miocene diatoms in Hungary. Nova Hedwigia Beih. **45**: 365–390.
Hendey N. I. 1959. The structure of the diatom cell wall as revealed by the electron microscope. J. Quekett Micr. **5**: 147–175.
—— 1970. Some littoral diatoms of Kuwait. Nova Hedwigia Beih. **31**: 107–167.
Janisch C. and Rabenhorst L. 1863. Ueber Meeres diatomaceen von Honduras, Beiträge zur näheren Kenntnis und Verbreitung der Algen I, pp. 1–16, Leipzig.
Kitton F. 1867. The genus *Amphitetras*. Hardwicke's Science Gossip III pp. 271–272, London.
—— and Pritchard A. 1861. A History of Infusoria Including the Desmidiaceae and Diatomaceae. British and Foreign. 858 pp. Whittaker and Co., London.
Kobayasi H., Idei M., Mayama S., Nagumo T. and Osada K. 2006. H. Kobayasi's Atlas of Japanese Diatoms Based on Electron Microscopy. Vol. 1. 531 pp. Uchida Rokakuho Publishing Company, Tokyo (in Japanese).
Li C. W. 1978. Notes on marine littoral diatoms of Taiwan I. Some diatoms of Pescadores. Nova Hedwigia **39**: 787–812.
Nagumo T. 1995. Simple and safe cleaning methods for diatom samples. Diatom **10**: 88.
—— and Kobayasi H. 1990. The bleaching method for gently loosening and cleaning a single diatom frustule. Diatom **5**: 45–50.
Ross R., Cox E. J., Karayeva N. I., Mann D. G., Paddock T. B. B., Simonsen R. and Sims P. A. 1979. An amended terminology for the siliceous component of the diatom cell. Nova Hedwigia Beih. **64**: 513–533.
Round F. E., Crawford R. M. and Mann D. G. 1990. The Diatoms. Biology and morphology of the genera. 747 pp. Cambridge University Press, Cambridge.
Tanaka H. and Nagumo, T. 2004. Four cyclotelloid taxa characterized by a ligula-like segment on valvocopula, *In*: Poulin M. (ed.), Proceedings of the 17th International Diatom Symposium. pp. 399–409. Biopress, Bristol.
von Stosch H. A. 1975. An amended terminology of the diatom girdle. Nova Hedwigia Beih. **52**: 1–36.

渡辺 剛[a]，鈴木秀和[a]，南雲 保[b]，田中次郎[a]：**本邦新産珪藻 *Perissonoë crucifera* (Kitton) Desikachary & al.の形態**

沖縄県西表島で採取した砂粒に着生していた *Perissonoë crucifera* (Kitton) Desikachary & al.の光学顕微鏡，走査電子顕微鏡および透過電子顕微鏡による殻微細構造を報告する．本分類群は本邦初記録である．殻形は四角形で，殻端はやや突出する．半被殻は殻，接殻帯片および連結帯片よりなる．帯片は接殻帯片，連結帯片ともに4カ所で分離する（それぞれ四分接殻帯片 QBV，四分連結帯片 QBP とする）．2種の四分帯片間の隙間にはそれを裏打ちする小片（それぞれ接殻小片 CBV，連結小片 CBP とする）がある．この四分帯片と小片は他の分類群では観察されたことがなく，*P. crucifera* に特徴的な構造である．殻面は顆粒状突起が散在し，殻肩には小針が1列に並ぶ．条線は1列の胞紋により構成され，殻套にも同様の胞紋が1列並ぶ．胞紋は輪形篩板により閉塞される．輪形篩板は同心円状の間隙をもち，2から4本の棒状体によって支持される．殻端小孔域は多数の小孔によって構成され，小孔は篩板様の構造（rota-like segment）をもつ．唇状突起の数は1から4個で，それぞれ各殻端に存在する．

（[a]東京海洋大学，
[b]日本歯科大学）