Electron-microscopical Study on Fine Structures of Diatom Frustules. XVIII

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**Rhizosolenia alata** Brightwell f. indica (Peragallo) Ostenfeld (Text-fig. 1a; Pl. I, fig. 1), Hustedt, Kieselalg. 1: 602, fig. 346 (1930); Desikachary, Mikrosk. 9: 172, figs. 10-14 (1954).

Cells much larger than the type species. In the present specimen, the frustules about 110 μ in diameter and 300 μ in length. Intercalary band scale-like, about 40 × 12 μ, arranged in longitudinal rows. In shape and number of rows of the intercalary bands, this form akin to *Rhiz. Temperei* and *Rhiz. Castracanei*, but distinctly differs from them by its tubelike, more or less curved oblique setae. Frustule pores on calyptrae and intercalary bands are locular. Loculi on the calyptra, usually round or elliptical, arranged in longer and shorter longitudinal rows about 35-42 in 10 μ. In each row, the loculi about 30-36 in 10 μ. In the present stereoscopic observation, it was ascertained that the loculi are opened inwards directly into the cell interior and closed outwards by the sieve membranes (the same is in the intercalary bands). The outer sieve membrane has 1-3 irregularly arranged round sieve pores about 40-50 μ in diameter. The inner membrane rudimental, or may be absent. The interlocular space, which is light microscopically smooth, shows super-fine spongy porous structure. Loculi on the intercalary band usually regular or non-regular hexagonal (in those surrounding the interlocular pore, often heptagonal), about 30-36 in 10 μ, arranged in different quincunxes on different parts of the band; diameter about 250-400 μ. In the present stereoscopic observation, it was elucidated that the loculus is closed outwards by a sieve membrane and half closed inwards by an inner membrane with a large central opening. (Desikachary described the structure of the loculi of this form as follows: “The areolae are partially open to the outside and on the inside is the sieve membrane.”) This description based upon his non-stereoscopic observation is quite contrary to my present stereoscopic observation, and is not correct. His misconception of the position of elements of loculi, I suppose, was caused unavoidably by his non-stereoscopic microscopy. The outer sieve membrane very thin, seems to have six marginal sieve pores (30-40 μ in diameter) at the six corners, and in addition to these, there is one or two sieve pores or poroids in the middle of the membrane which can be seen more clearly than the marginal ones through the opening of the inner membrane. Inner membrane of the loculus is apparently spongy porous, and is provided with a large round central opening about 150-250 μ in diameter. Details of the lateral membrane of the loculus could not be revealed in the present research, and it has not been possible to find if there is any pass pore on it. The depth of the loculus could not be accurately measured, but judging from the stereoscopic image, it may be concluded that the depth is nearly equal to the diameter. Interlocular pores, about 30-50 μ in diameter (the rudimentary loculi; Desikachary’s “canal pore”), are scattered about on the entire area of the intercalary band, and do not have any definite arrangement of regularity in the distance between each

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other as mentioned by Desikachary\(^1\). They are usually rectangular, and open on the inside by a rectangular opening and closed on the outside by a sieve membrane with a linear (?) sieve pore. Fine structure of the loculus of intercalary bands of the present form coincides, in its fundamental structure, with that of the type species\(^2\). On the other hand, \(f. \text{ inermis}\)\(^3\) and a doubtful form\(^1\), respectively show quite different structure in their sieve membranes. In \(f. \text{ inermis}\) the sieve membrane has two parallel lens-shaped sieve pores (correspond to Hendey's frustule structure No. 8\(^9\)), and the doubtful form has no distinct sieve pores but minute poroids (Desikachary, fig. 21). Generally speaking, such a non-coincidence of fine structure between different forms of the same species is rather exceptional, and further detailed research of these forms is expected.

**Habitat:** Marine plankton. Harimanada, Seto Inland Sea, Hyōgo Prefecture (Okuno, No. m1324. Aug. 1958).

**Rhizosolenia imbricata** Brightwell var. **Shrubsolei** (Cleve) Schröder (Text-figs. 1b, 2A; Pl. I, fig. 2), Okuno, Bot. Mag. Tokyo, 70: 104, pl. 1, figs. 4a, b (1957).

I already reported in my previous paper above mentioned, some of the electron microscopical fine structures of this variety observed by the non-stereoscopic electron microscopy, and further reviewed the Desikachary's electron micrographs which he published under the name of this variety and of *Rhiz.* sp.\(^4\) In the present stereoscopic observation, the following facts were ascertained or more correctly found: Loculi are closed on the outside by the sieve membranes and open almost fully on the inside. The ratio between the length, breadth and depth of the loculus is about 2.25:1:0.7—0.8. In loculi along the longitudinal axis of the intercalary band, the length and breadth are usually much reduced, and the directions of the diagonal sieve pores are irregular. The diagonal sieve pore is usually a single linear slit, sometimes it is divided into several minute oblong pores, or it is closed outwards by a thin membrane. The sieve membrane is somewhat thick on its outer margin and from there becomes thin gradually to the border of the sieve pore, and sometimes
has scattered minute granules impenetrable to the electron beam. Such granules were sometimes found also on the limb of the intercalary band. The sieve membrane and the limb of the intercalary band distinctly show the fine spongy porous structure.


*Mastogloia angulata* Lewis (Text-figs. 1c, 2C; Pl. I, fig. 3), Okuno, Bot. Mag. Tokyo, 70: 222, pl. 7, figs. 2a-c (1957).

In my previous paper above mentioned, I reported some of the fine structure of this species observed by the electron non-stereomicroscopy. In the present stereomicroscopy, further details of the three dimensional character of the fine structure of the frustule wall was much clearly revealed, and accordingly some of my previous description of the fine structure must be revised here. The present research reveals the following structure: Loculi in the central part of the valve, usually non-regular hexagonal, arranged in slightly radiating rows, about 9-11 in 10μ, and in each row loculi about 8-10 in 10μ. Loculus opened outwards and closed inwards by a sieve membrane. Ratio between the diameter and the depth of a loculus seems to be nearly equal to 2:1. The outer membrane narrow, marginal (about 10-15mμ broad), leaving a large round opening about 70-80mμ in diameter. The inner membrane is divided into two parts, the central dome-shaped area projected outwards (diameter about 400-600mμ) and the marginal flat area. The central area on its margin with a circular row of round sieve pores about 20-30mμ in diameter, and sometimes scattered with several pores inside the circle. The lateral membrane of the loculus seems to be non-porous. Transverse row of the hexagonal loculi ends on the margin of the valve in a double row of small incomplete loculi (the secondary loculi5)) about 2 in 1μ, and each double row represents a transversely elongated marginal compound oculus (the primary loculus5)). In the secondary loculus, the inner sieve membrane does not seem to be projected centrally, and has scattered round sieve pores over the whole area. Outer membrane of the secondary loculi indistinct, or may be absent. The marginal primary loculus seems to have a little higher lateral membrane than the central hexagonal loculi.


*Mastogloia aciculata* W. Smith (Text-figs. 1d, e. 2B; Pl. I, fig. 4), Synop. Brit. Diat. 2: 65, pl. 62, fig. 387 (1956); Hustedt, Kieselalg. 2: 515, fig. 946 (1933); Mills, Index Diat.: 898 (1933); Cleve-Euler, K. V. A. Handl. 4, no. 5: 58 (1953).

Valves elliptic-lanceolate, with more of less produced ends. Length 47-60 (40-90)μ; breadth 22 (16-24)μ. Marginal loculi about 2μ broad, quadrate, 8-9 in 10 μ, inner borders flattened, forming a band, ending near the ends of valve. Transverse rows of loculi 15-16 (15-20) in 10μ, slightly radiate; loculi 18-20 in 10μ, forming undulating longitudinal rows. The present electron stereomicroscopy reveals the following structure: Valve surface concave, submarginally highest, falls down gradually towards the raphe and abruptly towards the margin. Valve wall, doubly locular. Primary loculus which corresponds to the transverse row of areolae or alveoli of the light microscope image, is transversely elongated, and fully opened towards inside and closed towards outside by a lamina of secondary loculi. Lateral wall of the primary loculus is transverse, very thin, slightly thickened at its inner margin, and apparently hyaline, but it is finely spongy porous as the other parts of the valve wall. The transverse lateral membrane may be slightly projected outwards as a low lamella. Secondary loculus rectangular, about 500×650 mμ, 18-20 in 10 μ, arranged
in a transverse single row in each primary loculus, and closed towards outside by a sieve membrane and open freely on the inside into the primary loculus. The outer sieve membrane, with a thin round central area about 300-400 mμ in diameter and the thick margin provided with four round sieve pores (60-70 mμ in diameter) at the four angles, and in addition to these, sometimes, there is a sieve pore inside the middle of the transverse margin of the membrane. Central area of the sieve membrane very thin (often destroyed during preparation), with scattered fine pores or poroids and distinctly spongy porous. The longitudinal lateral membranes of the secondary loculi very low, slightly projected inwards, and the inner cover membrane
absent. Axial area of this species which hitherto misunderstood by its light microscopical image to be a thickened border of the raphe or a sort of rib run along the raphe, is elucidated in the present research, not to be a thickening or a rib, but to be a doubly laminar part along the raphe. The inner border of the outer membrane of the primary loculus turn inwards along the raphe and then bent back sidewards to run parallel to the outer membrane for about 1 µ. The part turned inwards which represents the side wall of the raphe is apparently smooth. The side turned, parallel running part is smooth and transparent enough for the fine structure of the outer membrane to be seen through it. Such a parallel running inner membrane of the valve on both sides of the raphe was also found by me in the raphe-valve of Cocconeis ceticola\(^7\)). The electron stereomicrographs of Achnanthes hungarica\(^8\), Ach. longipes\(^9\), and Pinnularia appendiculata\(^10\) show much thicker side wall of the raphe, and in such a case the axial area can be described being ribbed. From the present observation, it may be stated that such a doubly laminar structure of the axial area is to a certain extent common to the locular raphe-valve. In the light microscopy, the raphe of this species was considered to be a single straight fissure on its whole length and described being “straight”, but in the present electron microscopy, it was elucidated that the raphe is a vertical slit of about 50 mµ breadth, and at least in the middle one-third is folded or may be divided into the two, straight and accurate fissures by a thin oblique septum. Most of the diatoms hitherto researched with the electron microscope showed the straight raphe, and only in a few species, the folded raphe was found. In electron microscopy, the straight raphe was found in Achnanthes lanceolata\(^11,12\), Amphipleura rutilans\(^13\), Amphora ovalis var. pediculus\(^14\), Cocconeis ceticola\(^15\), Cocc. scutellum\(^15\), var. parva\(^16\), Cocc. stauroneiformis\(^17\), Diploneis chersonensis (Okuno, unpublished), Mastogloia Smithii\(^18\), Navicula pelliculosa\(^19–31\), Nav. pygmaea\(^20\), Nav. tripartita\(^28\), Nav. sp.\(^28\), Phaeodactylum tricornutum\(^24\), Rhoicosphenia curvata\(^25\). The folded raphe was found in Mastogloia Braunii\(^26\) and Navicula Trompae\(^27\), and the canal raphe was found in Epithemia sorex\(^28\), Nitzschia sp.\(^29\), Rholapodida musculus\(^30\), Surirella gemma\(^31\).


**Summary**

Fine structure of the frustules of four marine diatoms are described presenting their electron stereomicrographs.

**Rhizosolenia alata f. indica**: Both loculi on calyptra and intercalary band are closed outwards and opened inwards. Loculi on the calyptra round or elliptical, arranged in long and short longitudinal rows. The outer sieve membrane has 1–3 irregularly arranged sieve pores about 40–50 mµ in diameter. Loculi on the intercalary band hexagonal, about 250–400 mµ in diameter, in quincunxes. The outer sieve membrane has six marginal sieve pores each about 30–40 mµ in diameter. The inner membrane has a central opening about 150–250 mµ in diameter.

**Rhizosolenia imbricata** var. Shrubsolei: Loculi parallelogramatic on the intercalary band, closed outwards by a sieve membrane and almost fully opened inwards. The outer sieve membrane has a long diagonal sieve pore, which is often divided into several minute pores or closed by a delicate membrane.

**Mastogloia angulata**: Loculi hexagonal in the central part of the valve, opened upwards and closed inwards. The inner sieve membrane has a central dome-shaped
projection with a circular row of sieve pores each about 20–30 mμ in diameter. On
the margin of the valve, each row of the hexagonal loculi ends in a double row of
small incomplete loculi which represent the secondary loculi.

*Mastogloia apiculata:* Primary loculi transverse, groove-shaped, each closed
outwards by a row of secondary loculi, and opened freely inwards. Lateral membranes
of the primary loculi are spongy porous. Secondary loculi rectangular, about 500 ×
550 mμ, each closed outwards by a sieve membrane with four sieve pores at four
angles.

The electron stereomicrographs in plate I and II of my previous paper (Bot. Mag.
Tokyo, 72: 61–67) were presented by the halftone black and white printing, and when
viewed with a magnifying stereoscope, the meshed image of printing is seen distur-
bining the correct understanding of the fine structure of the diatom frustules. The fine
structure of frustules shown in such micrographs will be correctly understood by
viewing them with naked eyes. The method of such viewing was described in my
previous paper.

References


奥野 春雄：電子顕微鏡による珪藻殻細胞構造の研究 XVIII

つけの 4 種類の海産生産珪藻殻の電子顕微鏡的立体観察結果について記した。

*Rhizosolenia alata f. indicus:* 蒜殻 (Calyptra) の孔は円形または楕円形で縦列にならび、中間帯
(Intercalary band) の孔は六角形で、60° に変わる 3 縦列にならぶ。孔は壁状。中間帯のいずれのも
のも外周内開型で、その外膜は鰹膜をもつ。鰹膜はせまく、中央に大きい内孔をもつ。

*Rhizosolenia imbricata var. Shrubsolien:* 中間帯の孔は前後と開くように外周内開型で、外膜には面角線
の方向に線形の静孔が 1 つある。鰹孔はときに数個の小孔に分かれている場合もある。外膜は鰹孔近くでし
ずく、孔周辺に向ってやや厚さを増す。鰹膜に電子線の不透な不定形小粒子の散在する場合も見られた。

摘 要
内膜は不顕著である。*Mastogloia angulata*：立体写真により孔房が外周内閉型であることが判明した。著者の前報文（植物，70：222）では平面写真から推定して孔房が外周内閉型であると記したが、それは誤りであった。孔房は六角形を基本形とし、横列にならび、その外膜はきわめて広く、孔房は外へ広く開く。内膜は中央部がドーム状に外方へ隆起し、そのふちに環状にならんだ縦孔群がある。珪殻の左右線では各孔列はさらに小さい2列の孔房群に分れている。これら2列ずつの孔房は共通の横側膜をもつ集合孔房となる。*Mastogloia apiculata*：孔房は外周内開型で、しかも二層孔房となる。一次孔房 (大孔房)は横走する溝状孔房で、その外側は1列の二次孔房 (小孔房)層で閉じ、内側は全開する。二次孔房は光学顕微鏡的にAreolaeまたはAlveoliと記載されたもので、その外側は凹陥に1個ずつの縦孔をもつ縦膜でとざされ、内側は内膜なく一次孔房に全開する。

第17報（植物，72：61-67）では竜顕写真図版 (Pls. I, II)を網目写真版としたので、ルーペ式立体鏡で見た場合に印刷網目が見え、珪殻顕微組織のまぎらわしい結果となった。それらは裸眼による方法で見て、顕微構造を理解されたい。（京都工芸繊維大学繊維学部植物学研究室）
Pairs of electron stereomicrographs. Fig. 1, *Rhizosolenia alata* f. *indica* (upper part, the calyptra; lower part, the intercalary band). 2, *Rhiz. imbricata* var. *Shrubsolei* (intercalary band). 3, *Mastogloia angulata*. 4, *Mast. apiculata*. Scales: 1 μ. (When viewed through the stereoscope with parallel visual axis, Figures 1-3 show the inside view, and Figure 4 shows the outside view.)

H. Okuno: Electron-microscopical Study on Fine Structures of Diatom Frustules. XVIII