Studies on Genera *Mallomonas*, *Synura* and Other Plankton in Fresh-water with the Electron Microscope VI
Morphological and ecological observations on genus *Synura* in ponds and lakes in Yamagata Prefecture

Eiji TAKAHASHI

(Laboratory of Electron Microscope,
Faculty of Agriculture, Yamagata University,
Tsuruoka, Japan)

Received Aug. 31st, 1966

Electron microscopical studies on *Synura* covered with minute silica scales have not yet precisely been made in Japan, excepting a few publications by TAKAHASHI (1959, 1960, 1961).

The present writer is going to explain the results of his investigation on morphology and distribution of *Synura Petersenii*, *Synura Petersenii* var. *glabra*, *Synura spinosa*, *Synura sphagnicola*, *Synura echinulata* and its forma and *Synura uvella* in ponds at Tsuruoka Park*, in ponds of swamp at Mt. Gassan and other lakes in the mountainous region, and paddy-field consisting of rice nursery, rice field and narrow ditch, and also on seasonal fluctuation of *Synura Petersenii* and its var. *glabra* and *Synura spinosa* in ponds at Tsuruoka Park during a period from 1961 to 1962.

Sincere thanks are due to Professer Dr. Noboru ABE for his guidances, and also to Mrs. Katharine HARRIS of University of Reading, and Professer Dr. Shōichiro SUZUKI of Yamagata University for their invaluable advices.

Materials and Methods

Two bottles of water were collected from several ponds and lakes (Table I).

Table 1. Data of collectioning of water.

Locality	Date	Wt (°C)	pH	Altitude(m)
Ponds at Tsuruoka Park *	May, 1954 - Apr., 1955 June, 1961 - Sept., 1962	0. 8–29. 8	6. 0– 9. 5	16
Paddy-field	May - Aug., 1954 Aug., 1954 - April, 1955 Nov., 1962 - July, 1963	10. 5–37 1. 3–24 4. 5–29. 7	6. 4– 8. 4 6. 3– 7. 3 6. 3– 6. 9	16
Ponds in swamp at Mt. Gassan Midagahara pond group Other ponds	May-Nov., 1964 August, 1964	0. 8–28 30	4. 4– 7. 3 4. 7– 5. 9	1450 - 1500 1550 - 1900
Lake Sakozuki at Mt. Zaō	August, 1957, 1961		6. 8	840
Lake Ōtori-ike at Mt. Itō	July, 1959	19 –20. 4	6. 2– 6. 4	963
Pond of Motodate River Higashine	August 1962 July, Oct., 1963 and April, 1964	— 4. 1–20	6. 6– 7. 0	100

*Former name "Ditch at Tsuruoka Park"

Organisms of one of these bottles (0. 5 lit. of water) concentrated by centrifuging at about 3,000 rpm. A drop of concentrate was examined under the light microscope, and the living organisms were identified as nearly as possible and drawn and counted the number of cell of *Synura*. For examination of whole scales of one cell of *Synura*, one or few cells were mounted on the carbon film, and then before they are dried up, organic matter was disolved away by dropping down 1/2 to 1/4 of a drop of dilute solution of chloral hydrate.

The other fixed sample of water (0. 5 lit.) was concentrated to 10 cc; the number of organism in 0. 05 cc or 0. 1 cc of concentrated water was counted under the light microscope; then the water sample was mounted on 3 to 6 meshes for thoroughgoing examination with the electron microscope.

Results

1. Morphology of *Synura*

(1) Colony and cell

Colony of *Synura Petersenii* is spherical, 35 to 50 μ in diametre, sometimes it consists of sparcely placed cells with long stalk (Fig.A, 3. 4). Cell with two unequal flagella, ovoidal to spherical, 9–12×7. 5–10. 5 μ (Fig.A, 9–12).

Colony of *S. spinosa* is spherical, about 30 μ in diametre, or long ellipsoidal (in April), 30μ×60μ and 35μ×96μ (Fig.A, 1. 2), and in February spherical colony which contained small daughter cells is found. Cell with two unequal flagella, long ovoidal, angular ellipsoidal or spherical, 13–19μ×9–13μ (Fig.A, 5–7).

By centrifuging, colony disintegrates into separate cells, losing their stalk, and changing their form from ellipsoidal and ovoidal to spherical of 8. 5μ to 10μ (*S. petersenii*) or 14μ to 20μ (*S. spinosa*) in diametre.

Encysted cell of *S. spinosa* is found in February; cyst is spherical, 14μ in diametre (Fig.A, 8).

(2) Flagella (Plate I)

Two flagella of *S. Petersenii* are unequal. The longer one is pantonematic (pleuronematic), 20μ to 35μ in length and its long terminal filament 5 μ to 6μ (Pl. I, Fig. 2). The fine structure of the two closely resemble those of *S. spinosa* (FOTT, 1957) and *S. Petersenii* (MANTON, 1954. BOURRELLY, 1957).

(3) Lorica and scale

The armour (Lorica) of *Synura* is constructed of minute silica scales which disposed regularly on the surface of the cell. The intact armour of *S. spinosa* and *S. sphagnicola* is shown in Plate VII, II and XI. Scales of the two are longitudinally disposed around the cell, on the other hand, scales of *S. Petersenii* obliquely relative to the long axis of the cell, at an angle of about 40°–45° to it

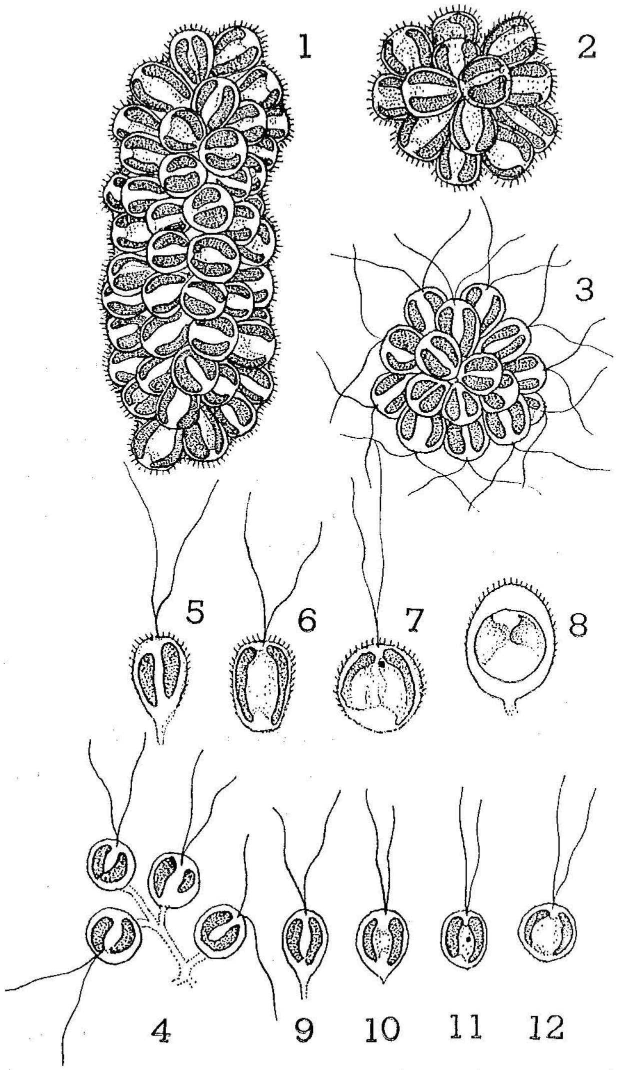


Figure A. Colony and cell of *Synura* in Ponds at Tsuruoka Park (——···10μ)

Synura spinosa

1. Long-ellipsoidal colony (April 12, 1962)
2. Spherical colony (Dec. 11, 1961)
5. Oval cell with peduncle (stalk) (April 24, 1962)
6. Ellipsoidal cell in colony (Jan. 25, 1962)
7. Disintegrated spherical cell (Jan. 25, 1962)
8. Encysted cell, electron micrograph of this cell is shown in Plate II, Fig. 5. (Feb.6, 1962)

Synura Petersenii var. *glabra*

3. Spharical colony (June 20, 1961)
4. Small colony which consists of sparcely placed cells with a long stalk (peduncle) (Dec. 11, 1961)

9.10. Oval cell with stalk (peduncle). (Jan. 25, 1962)

11.12. Disintegrated cell (March 8, 1962)

(Flagella are omitted from Figs. 1 and 2.)

(KORSHIKOV, 1926. TAKAHASHI, 1960 Figs. 10, 11)

Spine of ***Uvellae*** and thorn of ***Peterseniana*e** project outwards toward anterior portion of cell, consequently cells of the two groups show respectively spinose and serrate border (Plates I, II). The armour is constructed of apical, body (median) and stalk scales which vary in shape though the characteristic structure of species is recognized clearly.

Synura in this region were identified to following species on the base of scale: *Synura Petersenii* and var. *glabra*, *Synura uvella*, *Synura spinosa* f. *spinosa* and f. *curtispina*, *Synura echinulata* and new forma and *Synura sphagnicola*.

***Peterseniana*e**

Synura Petersenii and var. *glabra* are included to this group. Scale is constructed out of a hollow cavity with a apical thorn, radial ribs, an upturned edge on the outer side of perforated plane, and a pore of 0.3 to 0.4 μ in diametre at the base of thorn at the inner side (Pl. III, Figs. 20, 21).

Replica of scale is shown in Figures 19–21.

Synura Petersenii KORSHIKOV

Several scales of one cell in Ponds at Tsuruoka Park are shown in Plate III, Figures 9~17. Apical scale is nearly oval (Figs. 9, 10), body scale elliptic and stalk scale very small (Figs. 16, 17). Typical scales of *Petersenii* are shown in Figure 18. Scales with interconnected ribs (Figs. 22, 30, 31) and smaller scales (Figs. 31–33) were found in some localities.

Synura Petersenii KORSH. var. *glabra* (KORSH.) H-PESTALOZZI

The constructional elements of scale are identical with those of *S. Petersenii* (Figs. 23~28), but var. *glabra* is distinguished from *S. Petersenii* by the following points: (1) a less silicified scale, (2) shape is more oval, (3) a less developed middle cavity.

The intermediate scales of *Synura Petersenii* and its var. *glabra* were abundant in materials of ponds. And abnormal scales which three scales combined into one (Pl. V, Fig. 34), and also those of large size with abnormally developed cavity (Pl. V, Fig. 35) were found.

Uvellae

Species with spined scale are included to this group. Scales are oval with upturned edge, a more or less strongly developed hollow spine, which forms an angle about 45° with the scale, on the outer side, and a pore at the base of spine at the inner side; and on the front plane, there are such ornament as hexagonal mesh (*S. uvella*, *S. spinosa*), linear thickenings (*S. echinulata*), many papillae (*S. echinulata* forma) and membrane with linear openings (*S. sphagnicola*, Pl. X Fig. 67a).

Replica of scales of *S. spinosa* and *S. echinulata* are shown in Figures 50, 51 and 39–40.

Synura uvella EHRENBERG emend. KORSHIKOV

Few scales only were found in pond of Motodate (Pl. VI, Figs. 36, 37).

Synura echinulata KORSHIKOV

Scale presents a spine ending in one point and linear thickenings which seem to be covered by thin membrane (Fig. 39. HARRIS and BRADLEY 1958, Fig. 26). Stalk scales are narrow, and at the rear end of cell it becomes slipperlike scale without spine and linear thickenings (Pl. VI, Fig. 41).

Synura echinulata forma (Pl. VII, Figs. 42–47)

The present writer found the scales, which have never been illustrated, with many papillae in place of linear thickenings of type, in materials of ponds in swamp at Mt. Gassan and paddy field (Figs. 42–47). The dimension of scale is nearly identical with those of *S. echinulata*. Also he found comparatively shorter rodlike (tubular) scales (3. 8–7. 2μ in length, 0. 2–0. 3μ in thickness).

Synura spinosa KORSHIKOV

S. spinosa KORSHIKOV f. *spinosa* PETERSEN

A part of armour (lorica) is shown in Figure 48. Scales of one cell vary from anterior towards posterior portion of cell as is shown in Figures 49 to 58. The present species has the hollow rod-like (tubular) scale (5. 7 μ -11. 5 μ in length, 0. 2 μ-0. 3 μ in thickness) which the upturned edges of both sides of narrow thin silica membrane overlap each other (Plate IX, Figs. 59 to 60). The curved and thinner end of rod-like (tubular) scale seems to be connected with a triangular stalk scale (Fig. 61). Prolonged scale (Fig. 62), spheric thin scale and smaller scales were found (Figs. 63, 64).

Synura spinosa f. *curtispina* PETERSEN

Scales have shorter spine (1. 5μ to 1. 6μ in length) with 3 to 4 teeth at apical end (Fig. 65). The hexagonal mesh pattern and upturned edge of scales in this region are more well developed than those in Denmark which illustrated by PETERSEN (1956). Scales shown in Figure 65 rather resemble to scales of *Synura favus* BRADLEY 1966.

Synura sphagnicola (KORSHIKOV) KORSHIKOV

The present species occurs in such acid water only as ponds in swamp at Mt. Gassan, Lake Ōtori-ike and Lake Sakazuki-ko in the mountainous region.

Scale with evenly perforations, upturned edge and few linear openings, has yet not been pointed out; it shows the most simple structure in spined scales.

Scales of *S. sphagnicola* are abundant in variety in 7 stations at Mt. Gassan and Lake Sakazuki-ko and Lake Ōtori-ike; namely oval scale with nearly hexagonal perforations (Fig. 69), narrow scale with well developed upturned edge (Figs. 67, 68), and scales with unevenly perforations (Fig. 72).

The writer divids these scales into 4 types on the base of form, and then measured their size. The results are shown in Table II.

Table II. Comparison of scales of 4 types of *S. sphagnicola*.

Type	Scale length	Scale breadth	Upturned edge breadth of posterior end	* Extent of upturned edge : length of scale
a	3 - 3. 5	2. 2 - 2. 6	0. 18	3/5
b	2. 8	2. 1	0. 3	4/5
c	2. 8 - 3. 4	2. 2 - 2. 5	0. 47	3. 5/5
d	2. 7 - 3. 8	1. 7 - 2. 3	0. 49	4/5

Type a···oval, unevenly perforated scale (Fig. 72)

Type b···oval scale in lakes (Figs. 8, 76)

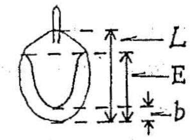
Type c···scale with well developed upturned edge (Fig. 66)

Type d···narrow scale with well developed upturned edge (Fig. 67)

* L···Length of scale

E···Extent of upturned edge

b···breadth of posterior end of upturned edge



Synura sp. (Pl. XII, Fig. 77)

Few only scales found in paddy-field and River Higashine.

Size of scales of all *Synura* species in this region is shown in Table III.

Table III. Size of scales of *Synura* in Yamagata Prefecture.

Species	Locality	Apical & Body scales		Stalk scale		Rod-like scale (tubular scale)
		length	breadth	length	breadth	
Synura Petersenii	Ponds at Park	4. 8 (μ)	2. 6 (μ)	1. 9 (μ)	0. 85(μ)	(μ)
	Paddy-field	2. 3–4	0. 75–2	4	1. 26	
	Pond-Motodate	4. 2–4. 8	1. 9–2. 4			
	R-Higashine	2. 7–3. 5	1. 5–2. 3	2. 2	0. 8	
var. *glabra*	Ponds at Park	3. 1	4. 5	1. 7	0. 6	
	Paddy-field	2. 2–3. 4	0. 9–2. 1	2	0. 9	
S. uvella	P-Motodate	3. 8	3. 3	4. 3×3. 3 *		
Synura echinulata forma	Paddy-field	2. 6–3. 8	0. 9–2. 8			
	Ponds in swamp	2. 5–3. 5	1. 8–2. 5	2. 0–3. 6	0. 4–1. 7	3. 8–7. 2×0. 2–0. 3
S. spinosa f. *spinosa*	Ponds at Park	2. 6–5. 2	1. 2–4. 5	2. 6–5. 6	0. 9–2. 8	5. 7–11. 5×0. 3
f. *curtispina*	R-Higashine	3. 3–5. 5	2. 1–3. 3	3. 7	1. 8	
	Ponds at Park	3 – 3. 4	2 – 2. 5			
	Paddy-field	3 – 4. 1	2. 3–2. 9			
Synura sphagnicola	Lake-Sakazuki	2. 3–2. 8	1. 7–2. 1			8×0. 25
	Ponds in swamp	2. 7–3. 8	1. 7–2. 8	3 –3. 6	1. 6–1. 9	
Synura sp.	Paddy-field	2. 6×1. 7(spine : 0. 66μ in length, 0. 29μ in thickness)				

*···Body scale of *S. uvella*

2. Ecology of *Synura*

(1) Distribution of *Synura* in this region

The results are shown in Table IV. *Synura Petersenii* and its var. *glabra,* and *S. spinosa* are commom in ponds in the plain land, *S. sphagnicola* common in lakes and ponds in the mountainous region. *S. echinulata* is comparatively rare, *S. uvella* very rare. In River Higashine and paddy-field, scalcs of many species are found but living cell is rare.

Excepting that *S. Petersenii* occurs in ponds at Tsuruoka Park in the period of high water temperature and alkaline water, the others are common in acidwater of low water temperature (less than 20°C).

Table IV. Distribution of *Synura* in Yamagata Prefecture

Species	syn.	Ponds at Tsuruoka Park	Paddy-field	Swamp at Mt. Gassa Midahara others Ponds	Lake Otori-ike	Lake Sakaju-ki	Pond of Motoda-te	River Higashi-ne
S. Petersenii	*S. uvella*; *S. caroliniana*	C	RR C				*	*
var. *glabra*	*S. glabra*	C	RR R					
S. uvella	*S. uvella* *S. reticulata*; *S. verrucosa*						*	
S. echinulata	*S. conradii*		R					
f. or var.			R *	R				
S. spinosa (*f. spinosa*)	*S. Bioretii*; *S. uvella*	C	R *					*
f. curtispina		RR *	RR *					*
S. Sphagnicola	*Skadowskiella sphagnicola* *Syncrypta volvox*; *S. uvella*		*	C RR *	C	CC		*
S. sp.			*					*

* Scale only

(2) Seasonal fluctuation of *Synura* in Ponds at Tsuruoka Park

Synura in concentrated living materials were counted as the number related with the number of cell of such common species as *Scenedesmus, Trachelomonas* etc., which are observed easily at low magnification under the light microscope in fixed materials. And then in order to identify accurately *Synura* species, the greater parts of the counted cell of *Synura* and *Synura*-like species were transferred on the carbon film with the micro glass tube, and confirmed under the electron microscope.

The results of his investigation during the period from August in 1961 to September in 1962 are shown in Figures B, C, D, and Table V.

In three stations of Ponds at Tsuruoka Park, *S. Petersenii* occurs from May to November, var. *glabra* from autumn to spring, and *S. spinosa* from December to March.

Total number of cell of *Synura* is the largest and the period of occurrence the longest in Station 3, in which pond water shows acidity during the longest period among three stations, on the contrary, the number is the fewest and the period the shortest in Station 1 as is shown in Fig. C.

In Stations 2 and 3, peak of occurrence of var. *glabra* is found during the period from November to December and in April, that of *S. spinosa* in February

Fig. B

Fig. C

Fig. D

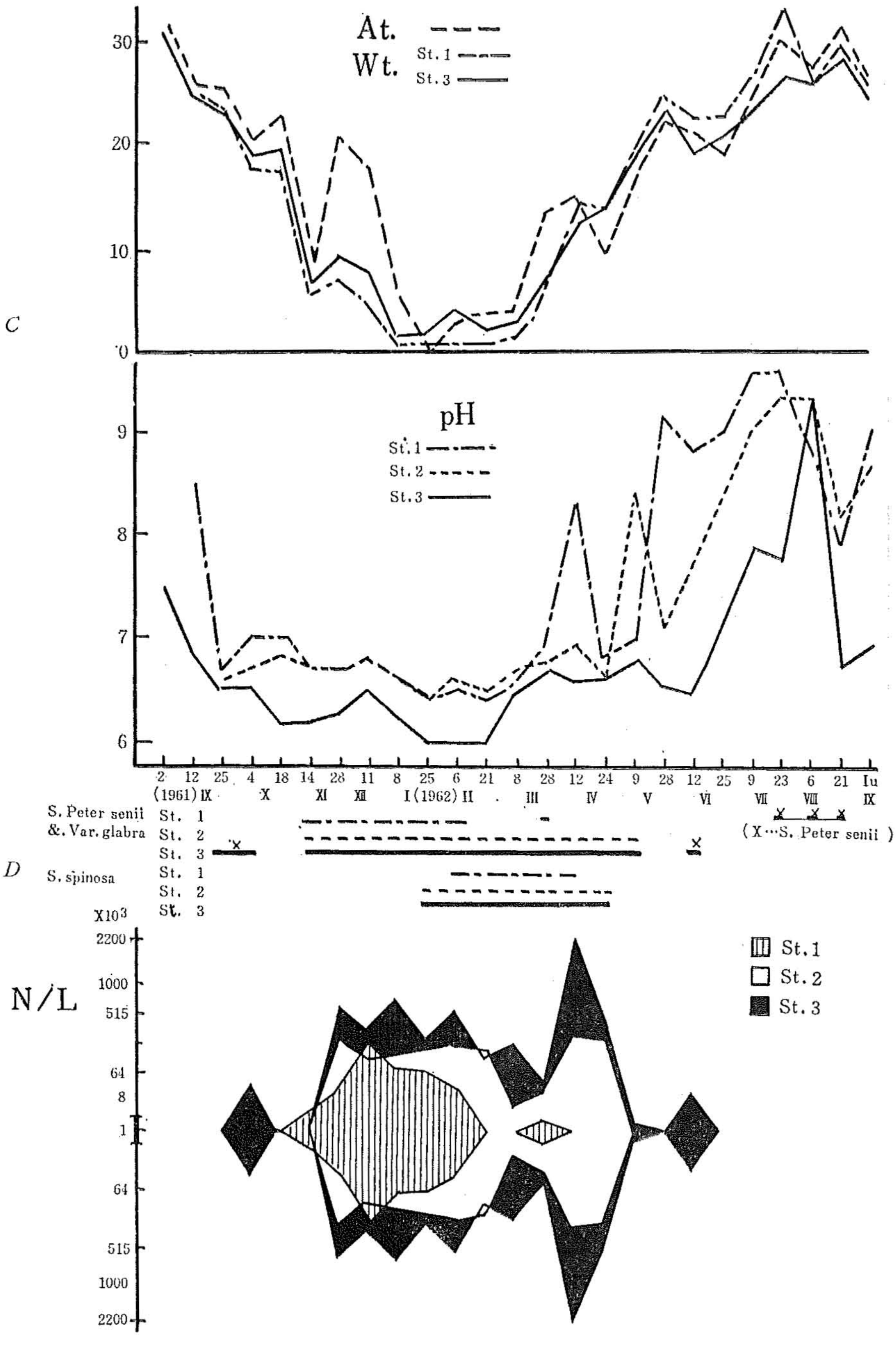


Fig. B. Seasonal change of Temperature, Fig. C. pH of water, and Fig. D. Seasonal fluctuation of *Synura* in Ponds at Tsuruoka Park.

Table V. Seasonal fluctuation of *Synura* in Ponds at Tsuruoka Park

Station		IX	X		XI		XII	I	
		-25	4	18	14	28	11	8	25
1	Total Phyto-pl.	1,494,000	148,980	2,711,640	630,260	1,267,640	1,062,580	1,094,920	3,138,180
	Synura	0	0	0	1,420	20,100	185,400	82,800	74,800
	S/T %				0.22	1.6	16	7.5	2.4
2	Total	3,225,540	—	5,942,740	2,690,020	940,000	2,028,620	—	1,270,880
	Synura	0		0	0	302,000	338,520		179,000
	S/T %					32.2	6.4		14.1
3	Total	2,198,860	2,262,500	910,220	992,400	2,148,040	2,983,060	2,155,400	1,484,900
	Synura	0	30,400	0	0	667,520	338,520	710,600	239,400
	S/T %		1.35			31.2	11.3	33.0	16.2

Station		II		III		IV		V	
		6	21	8	28	12	24	9	28
1	Total	1,689,700	12,565,220	1,774,460	297,080	4,298,720	324,700	833,220	3,077,660
	Synura	29,700	0	0	800	0	0	0	0
	S/T %	1.76			0.3				
2	Total	1,449,640	405,880	506,020	449,600	799,720	704,360	2,208,400	10,637,520
	Synura	201,000	182,800	4,000	20,000	287,400	246,600	0	0
	S/T %	13.8	45.0	1.26	4.5	36.0	35.0		
3	Total	5,698,120	887,120	1,427,080	404,360	3,281,840	9,509,560	19,427,780	4,581,640
	Synura	594,820	126,400	214,200	40,000	2,456,000	554,600	800	0
	S/T %	10.5	14.2	15.0	9.9	75.0	5.8	0.004	

Station		VI		VII		VIII		IX
		12	25	9	23	6	21	10
1	Total	11,513,360	4,296,960	9,193,120	8,116,060	42,940,560	2,466,860	13,017,360
	Synura	0	0	0	0	0	0	0
	S/T %							
2	Total	4,339,840	12,142,640	5,643,220	8,466,200	89,054,000	13,831,600	7,441,640
	Synura	0	0	0	0	0	0	0
	S/T %							
3	Total	3,597,400	1,702,500	4,486,620	5,194,580	8,214,940	11,616,800	8,545,800
	Synura	21,600	0	0	0	0	0	0
	S/T %	0.6						

(Figures indicate the number of cell per litre.)

and April, but in Station 1, peak in April is only one day and the number of cell is very small.

Comparing with the number of cell of *Synura* and total phytoplankton, *Synura* takes first to forth order during a period from December to April and 0.6 to 75% in Station 3, takes low order and 0.3 to 16.0% in Station 1, and takes the intermediate value of them both in Station 2.

These facts seem to show that there is close relationships between fluctuation of *Synura* and change of such environmental factors as pH and temperature of water or increase and decrease of micro-phytoplankton as *Microcystes*, *Aphanocapsa*, *Anabaena*, diatoms, *Scenedesmus* and *Chlamydomonas*.

It snowed on December 13th, in 1961, and Ponds were covered with ice of a few centimetres and snow of 5 to 30 centimetres in depth untill February in 1962, during that period, *Synura* with deep coloured chlomatophora occurred in abundance, together with *Chlamydomonas*, *Uroglena* and *Mallomonas* etc.

In ponds of swamp at Mt. Gassan, *Synura sphagnicola* occurs abundantly in May (Wt. 12°C, pH 6.5) and in September (Wt. 13°C, pH 4.6) but in August (Wt. 25°C pH 7.0), it is scarcely found, while this species occurs comparatively abundant in summer in Lake Sakazuki-ko and Lake Ōtori-ike (Wt. about 20°C) (TAKAHASHI 1959, 1960). This seems to show that the present species prefers lower water temperature than 20°C.

General consideration

In 1926, *Synura uvella* has been divided into 4 species by KORSHIKOV based on his investigation on its scale, after 20 years, taxonomy of *Synura* with the light microscope was worked out by HUBER-PESTALOZZI (1946), and BOURRELLY (1957) published results of his precise investigations on morphology, phylogeny.

PETERSEN who has called attention to the fact that the covering of *Synura* cell is built up of scales in 1918, reexamined scales of *Synura* with the electron microscope and confirmed 7 species of *Synura*, and he said that all species of the genus which have been established without taking into account the structure of the scale must be considered doubtful (1957, 1958). From the results of investigations with the electron microscope by HARRIS and BRADLEY (1957, 1958), MANTON (1954), and FOTT and LUDVIK (1957), FOTT attained to a conclusion (1961) that excepting the following species : *Synura Petersenii, S. uvella, S. spinosa, S. sphaqnicala, S. echinulata* and *S. lapponica*, other *Synura* species are doubtful. But in 1963, *Synura splendida* KORSHIKOV which has been described by KORSHIKOV, was examined under the electron microscpe by KRISTIANSEN in Denmark, and also by PETERFI (1965) in Rumania. Recently BRADLEY (1966) published a new species under the name of *Synura favus* which closely related to *S. spinosa*. On the other hand, the present writer has shown an electron micrograph of *Synura*-like scale in previous paper (TAKAHASHI 1959, Pl. IV Fig. 33), and though he has carried on since then a reseach for the living organism in numerous materials with the light and electron microscope, he could not yet find or confirm it, so he shows here only the scale with complex structure and shorter spine. These facts show that there are yet some minute living organisms as *Synura* involved in obscurity, and also it is needed to make a further investigation with the light and electron microscope.

On the rod-like (tubular) scale

The present writer found the rod-like (tubular) scale ($3.8\text{-}11.5\mu \times 0.2\text{-}0.3\mu$) in *Synura echinulata* forma., *S. spinosa* and *S. sphagnicola* of ***Uvellae*** group, but this scale is absent in species of ***Peterseniauae***. In addition to difference of constructed elements of scale between two groups, it presence or absence seems to be worth noticing as characteristic of each groups.

On the structure and support of spine

Spine seems to be constructed of two membranes as is shown in Figures 38, 42, 72 and 78, and teeth at apex of spine are also consisted of outer thin membrane, as is shown in Figure 78. Few perforations at median part of spine of *Synura echinulata* are clear already, however, high resolved power of electron microscope reveals minute regularly arranged perforations placing at distance of about 60 A in transversal rows as is shown in Plate XII, Figure 76 ; HARRIS & BRADLEY (1958) Figure 21. The membrane with perforations reminds the writer of the outer sieve membrane of *Melosira*. On the other hand, it seems that the spine of *Synura spinosa* is supported by ribs, which placed centripetally towards the base of spine as those of *S. splendida* (KRISTIANSEN, 1963), and also the spine of *S. echinulata* by membrane, covering the linear thickenings, and that of *S. spagnicola* by membrane with linear openings.

But it is necessary to carry on the further examinations of the ultra thin sectioned materials on above mentioned structure and function.

Disposition of scales

Scales of the armour of *S. sphagnicola*, *S. spinosa*, and *S. echinulata* (HARRIS and BRADLEY 1958) are disposed to longitudinal direction of the cell, which different from that of *S. Petersenii*. In this point the two groups of *Synura* are distinguished clearly each other in addition to the structure of scales. Also it is intresting that such longitudinal disposition of scales of uvellae group is similar to that of *Mallomonas akrokomos* which divided as isolated species from many *Mallomonas* species of 4 groups by HARRIS and BRADLEY (1960).

On *Synura echinulata* forma

The scale differs from *S. echinulata* in sculptural element, and from BOURRELLY'S forma in size (BOURRELLY, 1957), and none of scales with linear thickenings of type species have been found in ponds of swamp at Mt. Gassan throughout the year, consequently the present writer wishes to divide it into new forma of *Synura echinulata*.

On *Synura Petersenii* and its var. *glabra*, and *S. sphagnicola*

Synura Petersenii and var. *glabra* occur at different season in the ponds, but the intermediate scales of both are found in abundance, the identification of them was very difficult by reason of the gradual changing of scales. By comparing with many electron micrographs of the writer's and foreign materials, he recognizes that scales of *S. Petersenii* and var. *glabra* show large variance in form (Plates 3, 4, 5, and Fig. E-1, 2, 7) respetively. KORSHIKOV (1926) described that the cells of *Synura Petersenii* kept in cold condition became more slender, due to the decrease in the amount of leucosin. It is impossible that characters of *S. Petersenii*

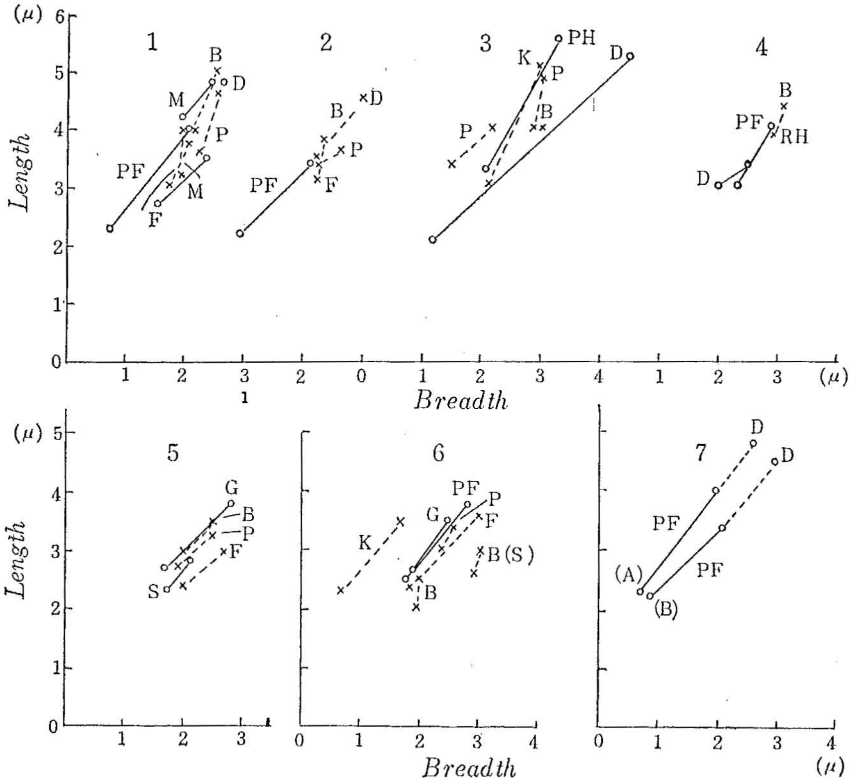


Figure E.
Size of scales of *Synura* in various localities in Japan and foreign country

1···*Synura Petersenii*
2···*S. Petersenii* var. *glabra*
3···*S. spinosa* f. *spinosa*
4···*S. spinosa* f. *curtispina*
○—○ Takahashi's results
5···*Synura sphagnicola*
6···*S. echinulata*
(G···*S. echnulata* forma)
7···*S. Petersenii* (A) and var. *glabra* (B)
D···Ponds at Tsuruoka Park
G···Ponds in swamp at Mt. Gassan
M···Pond of Motodate
PF···Paddy-field
RH···River Higashine
S···Lake Sakazuki-ko
×···× Foreign localities
B···France (BOURRELLY)
B(S). Sweden (BOURRELLY)
F···CSSR (FOTT & LUDVIK)
K···Soviet Union (KORHIKOV)
P···Denmark (PETERSEN & HANSEN)

and its var. *glabra* are independant of the environment. Though it is needed more precise investigations on that point, the gradual change of scale of them may be considered as the phenomena of cyclomorphosis.

Scales of *Synura sphagnicola* in various localities, namely many ponds of swamp at Mt. Gassan, Lake Sakazuki-ko and Lake Ōtori-ike, show a large variety as is shown in Plates X, XI. Scales in lakes of Ōtori-ike and Sakazuki-ko, closely resemble the scale of type which is illustrated by FOTT or PETERSEN, but other scales show various forms (Pls. X, XI, Figs. 66, 67, 68, 72. Table II) in small and shallow ponds in swamp which undergo immediatly influences of change of such various environmental factors as air temperature, rain fall, wind, radiant heat. It seems that such effect is produced from physiological reaction of cell against the changeable pond conditions.

The present writer figures here on size of scale of *Synura* in this region and foreign localities (Figure E), and he recognizes that scale of *Synura* closely resemble one another in size, excepting that scale of *S. spinosa* in ponds at Tsuruoka Park is wider in breadth than others, and scales of *S. echinulata* of Soviet Union (KORSHIKOV) and Sweden (BOURRELLY) are different from others as is shown in Figure E-3, 6. This fact shows clearly that the wide-spread species possess nearly identical specific characteristics in each species throughout the country.

Ecology of *Synura*

From the present writer's and foreign worker's results that *S. Petersenii* is very common in acid and alkaline water and in eutrophic ponds, and *S. sphagnicola* in ponds or sphagnum bogs with scid water ; it is clear that *S. Petersenii* possesses the great tolerance to acid and alkaline as is pointed out by BOURRELLY (1957) and also to high and low temperature of water, and *S. sphagnicola* is special acidophile, and both species are the commonest species throughout the country. It seems that the two species to be found the most commonly in Japan as well as in this region. On the other hand *Synura spinosa* is described as rare or very rare species excepting that it is common in acid water in France (BOURRELLY 1977). This species also seems to prefer acid water and low temperature in eu- and mesotrophic ponds. *Synura echinurata* forma which cohabited with *S. sphagnicola* in some ponds of swamp at Mt. Gassan, is comparatively rare, and also form of scale of which type, illustrated by PETERSEN (1956) and FOTT (1957) and this forma, is different from that of scale of KORSHIKOV's figure as is pointed out by BOURRELLY (1957). *Synura uvella*, which is common in fresh- and brackish-water (CONRAD, 1926) in Europe, in Malaya (PROWSE 1962) and South Viet-Nam (BOURRELLY 1957) in the tropics, and in Hokkaido (HADA 1959) and at Marugame (MIZUNO 1961) in Japan, is very rare in this region. On these species, it is required to investigate many materials in various localities.

Up to date, distribution of *Synura* is well documented in the Northern Hemisphere from Soviet Union to the tropics (Table VI), but ecological investigation of *Synura*, which tend to favour cold climate in general, is scarcely ever carried out. It is very interesting to know results on this points on *Synura* in various localities, especialy in the tropics and the polar region.

Table VI. Distribution of *Synura*

Species \ Locality	Japan	Den-mark	Czecho-slovakia	Eng-land	Scot-land	Ice-land	Rum-ania	France (1)	Malaya (2)	S. Viet-Nam (3)
S. Petersenii	●	●	●	●	●	●	●	●		
var. *glabra*	●	●	●				●	●		
S. uvella	●	●	●	●	●		●	●	●	●
S. echinulata	●	●	●	●	●	●	●	●		
forma or var.	●									
S. spinosa f. *spinosa*	●	●	●	●	●	●	●	●		●
f. *curtispina*	●	●	●							
S. sphagnicala	●	●	●	●	●		●	●		
S. sp.	●									
S. splendida		●					●	●		
S. favus					●					
S. lapponica		●								
Species number	9	9	7	5	6	3	7	7	(1)	(2)

(1)~(3)···Light microscopical investigation only.
(1), (3)···BOURRELLY's results

It is attempted to make the typological classification of ponds and rivers, many plankton species are chosen as indicator, *Synura uvella* is selected as indicater species of β-mesosaprobic to oligosaprobic or eutrophic to disharmonious waters (TSUDA 1964, MIZUNO 1961). The present writer can not mention on this species, because this is scarecely found in this region, but it seems to appropriate to choose *Synura Petersenii* as indicator of eu- to mesotrophic water, and *S. sphagnicola* as that of oligotrophic water.

The present writer has noticed on the importance of microplankton as a producer in the community in ponds and lakes (TAKAHASHI 1959), but by reason that he confound the number of cell with that of scale, the value in previous paper (1959) is shown higher than the true value, on this point he is to give nearly true value in near future. As a producer, the present writer must notice *Synura* species, namely such as *S. Petersenii* var. *glabra* and *S. spinosa* in ponds of Park, *S. sphagnicola* in Lake Ōtori-ike, Lake Sakazuki and in ponds of swamp at Mt. Gassan, and *S. uvella* in lakes and ponds in Hokkaido (HADA 1959), are very important in eutrophic ponds in cold season or lakes of oligotrophic type or ponds of swamp where plankton community is simple and the number of cell of phytoplankton as photosynthetic plant is rather small.

Summary

Synura species found in various localties in Yamagata Prefecture are *S. Petersenii* and its var. *glabra*, *S. uvella*, *S. spinosa* f. *spinosa* and f. *cutispina*,

S. *echinulata* and its famra, *S. sphagnicola*. The results of investigations on morphology and ecology (distribution and seasonal fluctuation in Ponds at Park) were described in this paper.

1. On colony, cell, flagella, lorica and scale of *Synura*, and rod-like (tubular) scales of *S. echinulata*, *S. spinosa* and *S, sphagnicola*, and scales of *S. echinulata* forma or varietas, electron microscopical structures are shown.

2. In this region, *S. Petersenii* and its var. *glabra* and *S. sphagnicola* are the commonest, *S. echinulata* comparatively rare, *S. uvella* very rare.

3. Excepting that *S. Petersenii* occurs in summer in the Ponds at Park other species prefer water of low temperature and acid.

4. Scales with papillae in place of linear thickenings of *S. echinulata* seems to be appropriate to divide into new forma or varietas of *S. echinulata*.

5. Various scales of *S. sphagnicola* seem to be produced from physiological reaction of cell against changeable pond conditions.

6. Size of scale of *Synura* in this region and foreign localities (Fig. E) closely resemble one another.

7. In Ponds at Tsuruoka Park, *Synura* occupied 0.3 to 75% of the number of total phytoplankton in cold seasen. Peak of occurrence of *Synura* are found in a period from November to December and in a piriod from March to April.

Literature cited

BOURRELLY, P. (1957). Recherches sur les Chrysophycées. Rev. algol. Mémoire hors. Ser. 1. p. 412

BRADLEY, C.E. (1966). Observations on Some Chrysomonads from Scotland. J. Protozoology, Vol. 13, 143—145

CONRAD, W. (1926). Recherches sur les Flagellates de nos eaux saumâtres. 2e partic; Chrysomonadines. Arch. Protistenkunde 56, 197—234

FOTT, B. & J. LUDVIK (1957). Die submikroskopische Struktur der Kieselschupen bei *Synura* und ihre Bedeutung fur Taxonomie der Gattung. Preslia 29, 5—16

——— & ——— (1961). Submicroscopical structure of silica-scales in Chrysomonads and its use in Taxonomy. Progress in Protozoology

HADA, Y. (1959). The Flagellata of the Freshwater Plankton in Hokkaido. Bull. Suzugamine Women's College No. 6, 21—69

HARRIS, K. & D.E. BRADLEY (1956). Electron Microscopy of *Synura* scales. Discovery, Vol.XVII, No. 8

——— & ——— (1958). Some unusual Chrysophyceae Studied in the Electron Microscope. J. gen. Microbiol. 18, 71—83

KORSHIKOV, A.A. (1929). Studies on the Chrysomonads I, Arch. Protistenkunde, 67, 273—290

Kristiansen, J. (1963). Observations on the structure and Ecology of *Synura splendida*. Bot. Tidsskrift 58, 281—289

Mizuno, T. (1961). Hydrobiological Studies on the Artificially Constructed Ponds ('Tame-ike' Ponds) of Japan. Jap. J. Limnol., Vol. 22, 67—192

Peterfi, L.S. (1965). Observations on *Synura* Ehrenb. in Rumania with special attention to *Synura splendida* Korshik. Rev. Algologique no. 1, 25—55

Petersen, J.B. & J.B. Hansen (1956). On the scales of some *Synura* species I. Biol. Medd. Dan. Vid. Selsk. 23, no. 2,

——— & ——— (1958). Ibid. II. ibid. no.7

Prowse, G.A. (1962). Further Malayan Freshwater Flagellata. Gardens' Bull., Singapore, vol. XIX, Part I

Smith, G.M. (1950). Fresh-water Algae of the United States. McGraw Hill.

Takahashi, E. (1959). Studies on Genera *Mallomonas, Synura,* and Other Plankton in Fresh-water with the Electron Microscope I. Bull. Yamagata Univ. (Agric. sci.) Vol. 3, no. 1, 117—151.

——— (1960). Ibid. II. Ibid. no. 2, 25—38

——— (1961). Ibid. III. Ibid. no. 3, 33—51

Tsuda, M. (1964). Biology of Polluted waters. Hokuryu-Kan (in Japanese)

摘　　　要

Mallomonas, Synura 属および他の淡水産プランクトンの電子顕微鏡による研究 (VI)
山形県の各湖沼の *Synura* 属の形態学的，生態学的研究

高　橋　永　治

(山形大学農学部農学科)

鶴岡市公園堀，水田とその側溝，月山湿原御田原の沼，頂上附近の沼，大鳥池，蔵王盃湖，等のプランクトンとして採集される *Synura* 属の鱗片の電子顕微鏡的微細構造を研究して来た．また，*Synura* 属の生態は，不明な点が多いので，鶴岡市公園堀の3堀を選び，1961年6月から翌年9月までの1か年間毎月2回採集し，季節的消長を観察した結果を報告する．

方法：各池沼から1l または0.5l 瓶2本に採水し，1本は直ちに固定し，20cc あるいは10ccに濃縮し，普通の方法で，プランクトン全種の計数を行なつた．他の1本は生のまゝ持ち帰り，3,000rpm で遠沈し，1～2cc に濃縮し，直ちに光学顕微鏡で検鏡し，*Scenedesmus* など，その時期に比較的数が多く，固定試料から容易に同定し，計数できる種との比較数で，*Synura* を計数した．計数された *Synura* または *Synura* らしい種の細胞の大部分は，そのたびにコロジオンまたはカーボン膜を張つたスライドグラスあるいはメッシユに移され，電子顕微鏡下で，種の同定が行なわれた．光学顕微鏡で見のがしたかも知れないことを考慮して，生および固定した試料の少量を数メッシユに移し，乾燥し全視野を詳細に電子顕微鏡で観察した．

(1)　コロニーと細胞の形は変異に富み，平滑に近い輪郭を示す ***Peterseninanae*** と刺の

ある *Uvellae* の二つの群の識別は容易であるが，種の同定は不可能であつた．鞭毛は不等2本でこの構造は *S. Petersenii* で確かめられた（Plate. I).

(2) 鱗片を基にして，*S. Petersenii,* と var. *glabra, S. uvella, S. echinulata,* その forma, *S. spinosa* f. *spinosa,* f. *curtispina, S. sphagnicola* の生息を確認した．各種の種的特徴を持つ鱗片の構造は Plates に示される．種的特徴は，細胞の先端部（鞭毛のある部分）の鱗片に最も明瞭で，後端部になるにつれて，薄れてゆき，形も変化してゆく．

(3) *S, echinulata* の鱗片は，前平面部に条線構造を持つ．月山湿原の池沼のものは，その代わりに多数のイボを持ち（Plate. VII. fig. 42）明らかに区別することができる．この鱗片は全く報告されていない．*S. echinulata* の forma あるいは変種として分けたいと思う．

(4) *S. spinosa, S. echinulata* forma, *S. sphagnicola* の3種は，細胞の末端部，梗の部分に細長い棒状鱗片を持つことがはじめてわかつた（Figs. 45, 47. 58—61, 70)．長さが異なる点以外，構造・形は3種に共通なものである．この構造はレプリカにより Fig. 57に示される．この鱗片は，末端部の3角形に近い形の鱗片と結合し，(Fig. 61) 梗の軸となり，コロニーの各細胞の連結を固くする役を果たしているようにも見える．

(5) *S. sphagnicola* の鱗片は，月山湿原の各池沼，盃湖，大鳥湖の細胞で，Pls. X, IX に示されるような，形態上の変異に富んでいることがわかつた．これは，盃湖（蔵王）大鳥池の二つの鱗片は全く同じであり，大きい湖の型と言うことができる．しかし，浅い，それほど大きくない月山湿原の池沼のものに変異が多いことは，四季または一日の環境変化（Wt. 0.8—28°C pH. 4.4—7.3）が直接に影響を与える結果のように思われる．

(6) 分布は第II表に示される．平地の富栄養的な公園堀に *S. Petersenii,* var. *glabra* と *S. spniosa* が普通にみられ，*S. sphagnicola* は，山地の酸性の貧栄養湖，湿原の沼に普通であつた．*S. Petersenii* と *S. sphagnicola* は，世界的に広く分布している種である．*S. echinulata* はまれであるがその forma は，比較的に稀な程度である．普遍的に分布していると言われる *S. uvella* は非常にまれで，元楯沼の試料から少数の鱗片を見つけただけであつた．外国では，*S. echinulata, S. uvella* は普通にみられるものであり，今後広い範囲の採集が必要である．日本では，今まで *S. uvella* が記録されているだけであるが，羽田（1964）も指摘するように，鱗片の観察によつて，当地方に見られるすべての種は，広く分布しているものと考えられる．

(7) 公園堀での *Synura* の季節的消長は，環境要因と共に図 B～D に示される．*S. spinosa* は冬期間に限つて出現し，*S. Petersenii* は殆ど周年出現するが *S. Petersenii* は夏期，var. *glabra* は，冬期に限られ，その中間の時期には，似ていて同定が困難な中間の型の鱗片を持つものが多く，鱗片の漸進的変化が認められる．KORSHIKOV (1929) の実験からも推測されるように *Petersenii* は夏型，var. *glabra* は冬型として，季節的変異と考えてもよいようである．(5)の *S. sphagnicola* の変異とともに，環境要因の変化との関係については，今後の興味ある問題である．

(8) *S. spinosa* は比較的まれな種として報告されているが，今回の結果では比較的栄養化の進んだ池の，冬期から春先の短い時期に出現するものであり，採集時期によつて出現の有無，量の多少など，異なった結果になつたものと考えられる．

(9) 指標種として，*S. uvella* が採り上げられている．当地方の結果からは，*S. uvella* について言及できないが，北欧での報告も参考にして，*S. Petersenii* は富栄養湖沼の，そして *S. sphagnicola* は貧栄養湖の指標種として選ばれてよい種であろう．

(10) 公園堀の3堀での *S. Petersenii* の出現の山は11月末から12月と，3月から4月にあり *S. spinosa* は2月と3・4月にある．その数は全植物プランクトンの16.0％ (St. 1)，32.2％ (St. 2)，33.0％ (St. 3) を占め，2－3月には，出現期間の短い St. 1 では少数であつたが，St. 2，3では45％ (St. 2)，75.0％ (St. 3) を占めた．4月12日の山が多いのは，*S. spinosa* と *S. Petersenii* の二つの山が重なり合つたことによる．出現期間，出現数も三つの堀で異なり St. 3 は最も長く最も多いのに，St. 1 は反対であつた. St. 3 は，水位が降雨によつて上がれば，表層の各種藻類と共に表層水は土管を通つて St. 2→St. 1 と流れる．また，St. 1 には夏期ヒシやウキクサが開水面の98％に達するのに，St. 3 には全くなく，St. 3 は水が一番澄んでおり，酸性である期間が最も長い．このことと，*Synura* の分布がほとんど酸性域であつたことから，*Synura* は一般的に好酸性種であると言うことができる．そして冬期に出現数が多い *S. spinosa* と var. *glab*ra はまた，冷水を好む種であると言うことができる．

(11) 先に生産者として *Synura* を含めて微小プランクトンの重要性を指摘した（高橋，1959)．前に著者は細胞数(個体数)と鱗片数とを混同した点もあり，実際の値より高い値を示してしまつた．次報において，正確な値を報告したいと思う．*Synura* については，Ⅲ表に示すように11月から春先までの間，群集構造が単純な時期の公園堀で，全植物プランクトンの14から75％もの高い値を占める．そしてまた，高山湖の大鳥湖，盃湖などの貧栄養湖，月山湿原の池沼等の単純構造で個体数の少ないプランクトン群集の中で，高い出現数を示すものであり，生産者として *Synura* は重要なものの一つであると言うことができる．

Explanation of plates

Plate. I Flagella of *Synura*

Fig. 1. Flagella of *S. Petersenii*, arrow indicates terminal filament of shorter flagellum (acronematic one). ×2, 000 (Ponds at Park, June 9, '61)

Fig. 2. Shorter acronenatic flagella. ×10, 000

Fig. 3. Colony of *S. Petersenii* with serrated border. ×1, 000 (Ponds at Park, Nov. 14, '61)

Fig. 4. Mastigonemes and acronematic flagella.. ×20, 000

Plate. II Cell and Lorica

Fig. 5. Encysted cell of *S. spinosa*, *a* indicates rod-like scale. ×2, 000 (Ponds at Park Feb. 6, '62)

Fig. 6. *S. spinosa* ×1, 5000

Fig. 7. Spine of *S. spinosa*, ×10, 000

Fig. 8. A part of lorica of *S. sphagnicola* (arrow indicates direction of the longitudinal axis of cell). ×8, 000 (Lake Sakazuki-ko, Aug. '59)

Plate. III *S. Petersenii*

Figs. 9 to 17. Scales at anterior to posterior portions of one cell ×10, 000 (Ponds at Park, Sept. 25, '61)

Fig. 18. The most typical scales of apical and stalk of *Petersenii* ×10, 000 (Paddy field, Sept. 26, '62)

Fig. 19. Outer side of scale ×15, 000

Fig. 20. Inner side of scale of *Petersenii* ×10, 000

Fig. 21. Inner side of scale of var. *glabra* ×10, 000

Plate. IV (All figers ×10, 000)

Fig. 22. Apical (A) and body scales of *S. Petersenii*

Fig. 23. Scales of var. *glabra* with ribs at wider distance than Fig. 22 a. (Ponds at Park St. 2. Apr. 12, '61)

Fig. 24. Scales of *S. Petersenii* (Long shadow indicates that the thorn projects at right above the scale).

Fig. 26. Scales at median part of var. *glabra* (Ponds at Park st. 1. March 22, '55)

Fig. 27. 28. Stalk scales of var. *glabra* (Ponds at Park st. 2, Janu. 5, '62)

Plate. V. Various form of scales of *S. Petersenii* and its var. *glabra* (All figers × 10, 000)Fig. 29. *S. Petersenii*

Fig. 30. Scale with interconnected rib. (Motodate August.)

Fig. 31—33. Smaller scales (R-Higashine, Apr. 8, '64)

Fig. 34. Abnormally shaped scales (Ponds at Park June 9, '61)

Fig. 35. Abnormally develloped scales and ordinary scales of var. *glabra* (Pond at Park April 12, '62)

Plate. VI *Synura uvella* and *Synura echinulata*

Fig. 36. Apical scale of *S. uvella* ×10, 000 (Motodate)

Fig. 37. Basal scale of *S. uvella* ×10, 000 (Motodate)

Fig. 38. Apical scale with well developed linear thickenings ×20, 000 (Paddy field)

Fig. 39. Replica of *S. echinulata* of outerside of scale ×20, 000

Fig. 40. Replica of inner side of scale of *S. echinulata* ×10, 000

Fig. 41. Stalk scale of *S. echinulata* ×10, 000 (Paddy field)

Plate. VII. *Synura echinulata* forma and lorica of *S. spinosa*

Fig. 42. Apical scale with papillae at front plane of *S. echinulata* forma ×20, 000 (Pond St. 6 in Swamp, July 16, '64)

Fig. 43.—45. Slipper and spathulate scales of *S. echinulata* forma ×10, 000 (43. 44··· Pond St. 3 in swamp, 45··· st. 6)

Fig. 46. Rod-like (tubular) scale ×10, 000 (Pond St. B in swamp, Oct. 11, '64)

Fig. 47. Apical and rod-like (tubular) scales ×10, 000 (Pond St. B in swamp, Sep. 13, '64)

Fig. 48. A part of lorica of *S. spinosa* ×3, 000 (Ponds at Park St. 3, March 18, '62)

Plate. VIII. *S. spinosa* f. *spinosa* in Ponds at Park (All figures ×10, 000)

Fig. 49. Apical scale with well developed hexagonal mesh pattern

Fig. 50. Replica of outer side of scale

Fig. 51. Replica of inner side of scale

Fig. 52. Median scale with shorter spine

Fig. 53. Triangular stalk scales (distance of transversal lines at back ground is 1/960 mm)

Fig. 54. Replica of outer side of stalk scale (arrow indicates a pore of upturned edge)

Fig. 55. Replica of inner side of stalk scale

Fig. 56—57. Stalk scales

Fig. 58. Rod-like (tubular) scale of *S. spinosa*

Plate. IX. *S. spinosa* f. *spinosa* in Ponds at Park and f. *curtispina* in Paddy field (All figures ×10, 000)

Fig, 59. Replica of rod-like (tubular) scale

Fig. 60. Scattered rod-like (tubular) scale, it shows that this scale consists of one silica membrane

Fig. 61. Stalk and rod-like (tubular) scales which connected with each other.

Fig. 62. Apical scale which a front plane prolongs

Fig. 63. 64. Spheric and minute scales

Fig. 65. Apical scales of f. *curtispina* (Paddy field, Aug. 2, '54)

Plate. X *S. sphagnicola*

Fig. 66. Oval scales with well developed upturned edge ×10, 000(Pond St. 5 in swamp)

Fig. 67. Long ovale scales, **a** ··· a part of linear opening on front plane, **b** ··· five teeth at apex of spine ×20, 000 (Pond St. 9.5 in swamp)

Fig. 68. Narrow median scale with abruptly cutted apex of spine ×10, 000 (Pond St. 3 in swamp)

Fig. 69. Oval and triungular stalk scales ×10, 000 (Lake Sakazuki)

Fig. 70. Elliptical and rod-like (tubular) scales (a) ×10, 000 (Lake Sakazuki)

Fig. 71. Rear scale (Pond St. 1 in swamp)

Plate. XI *Synura sphagnicola*

Fig. 72. Oval, unevenly perforated scales with slender upturned edge ×20, 000 (Pond St. 2 in swamp)

Fig. 73. Stalk scale ×10, 000 (Pond St. 2 in swamp)

Fig. 74. Narrow scale ×10, 000 (Pond St. 4 in swamp)

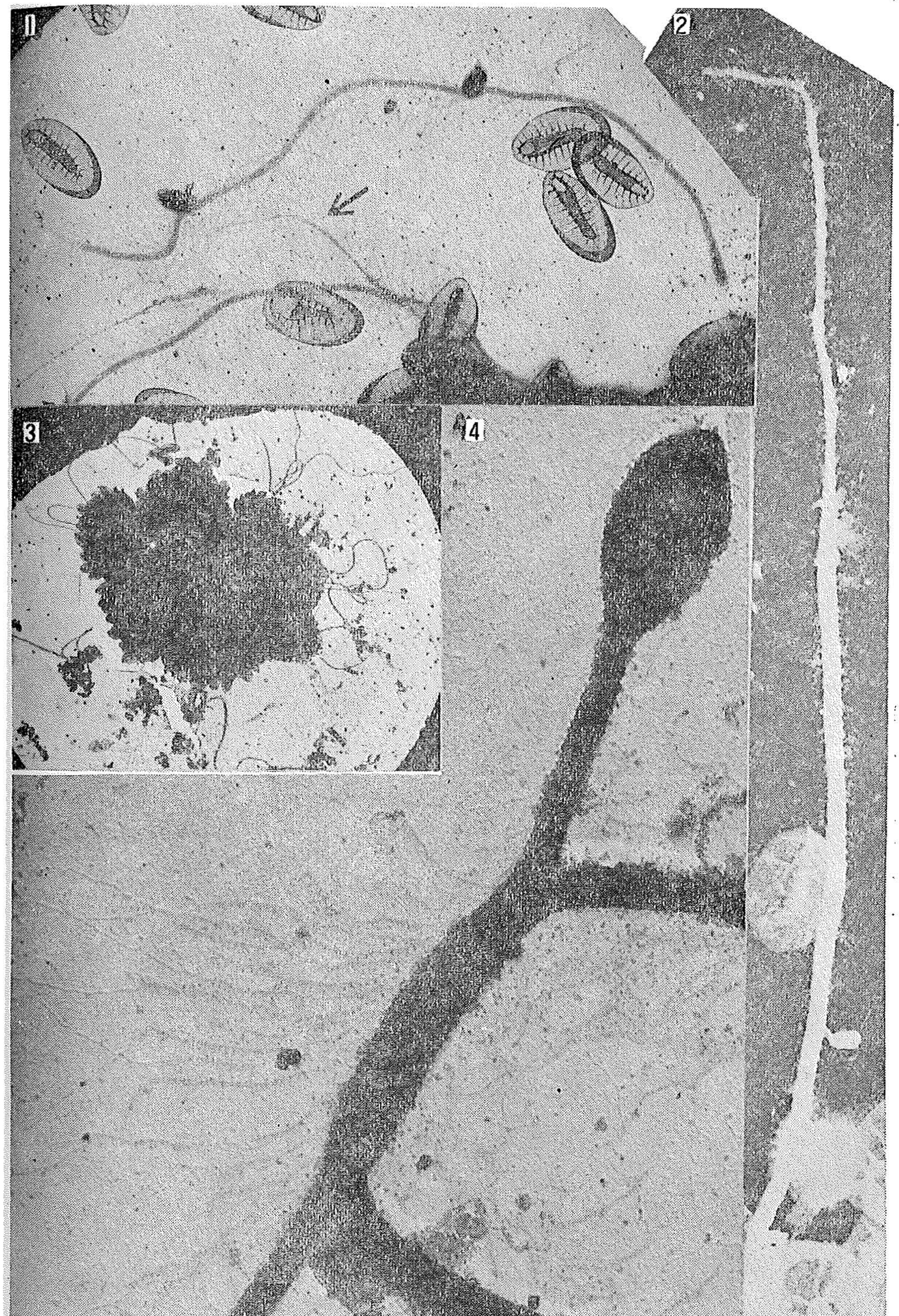
Fig. 75. The widest scale into which two scales united. ×10, 000 (Pond St. 4 in swamp)

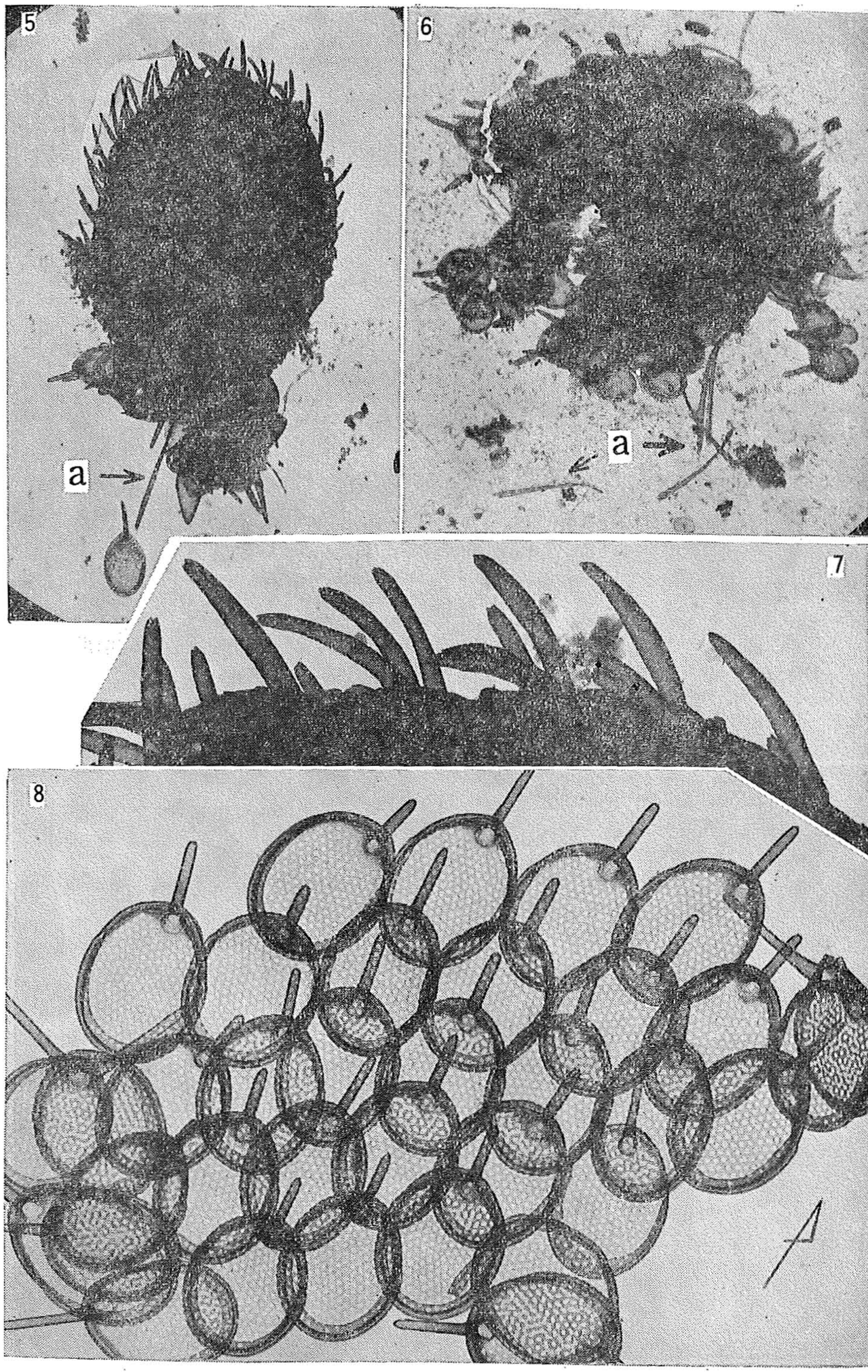
Fig. 76. Replica of outer side of part of lorica ×10, 000 (Lake Sakazuki)

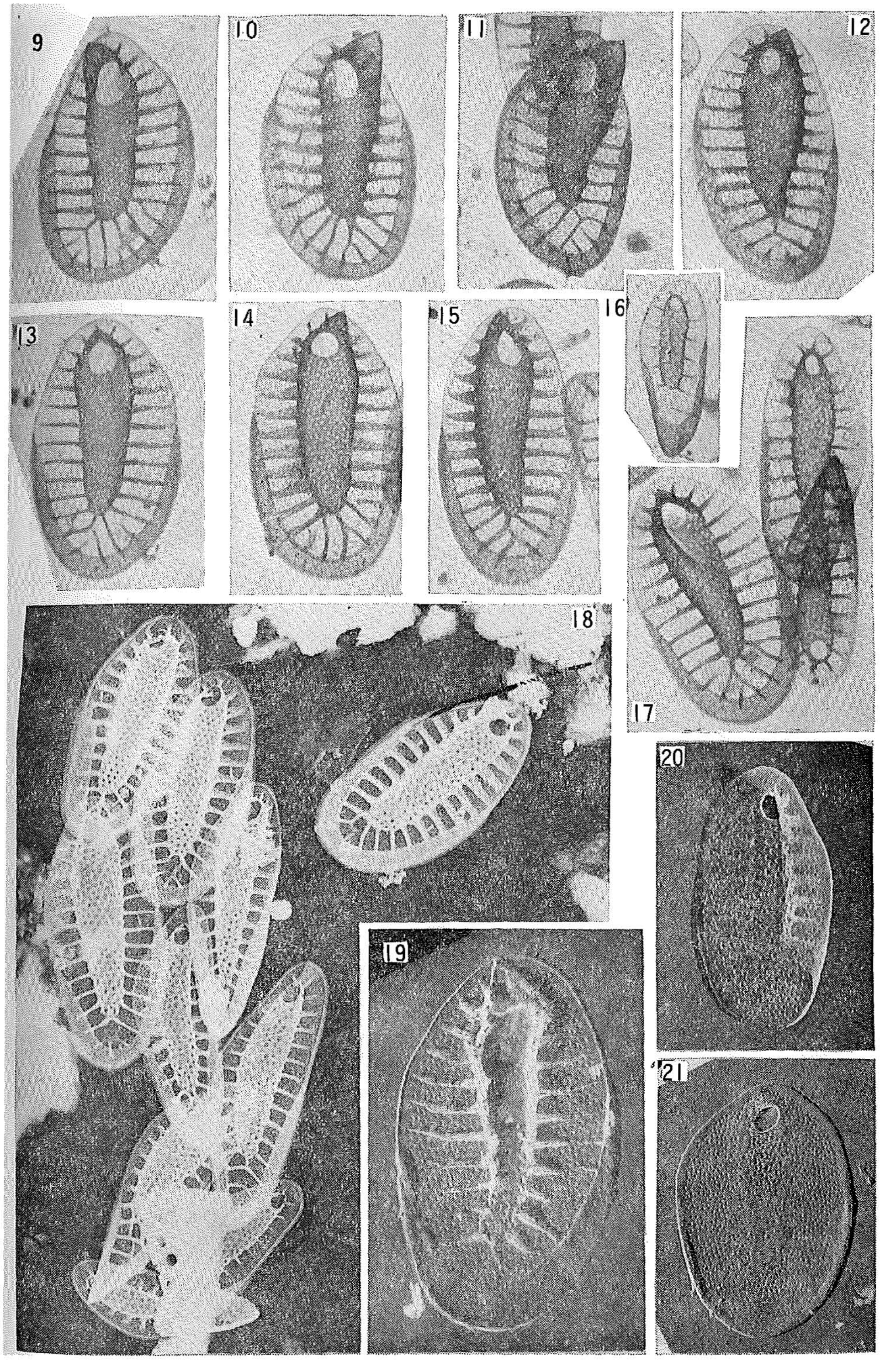
Plate. XII

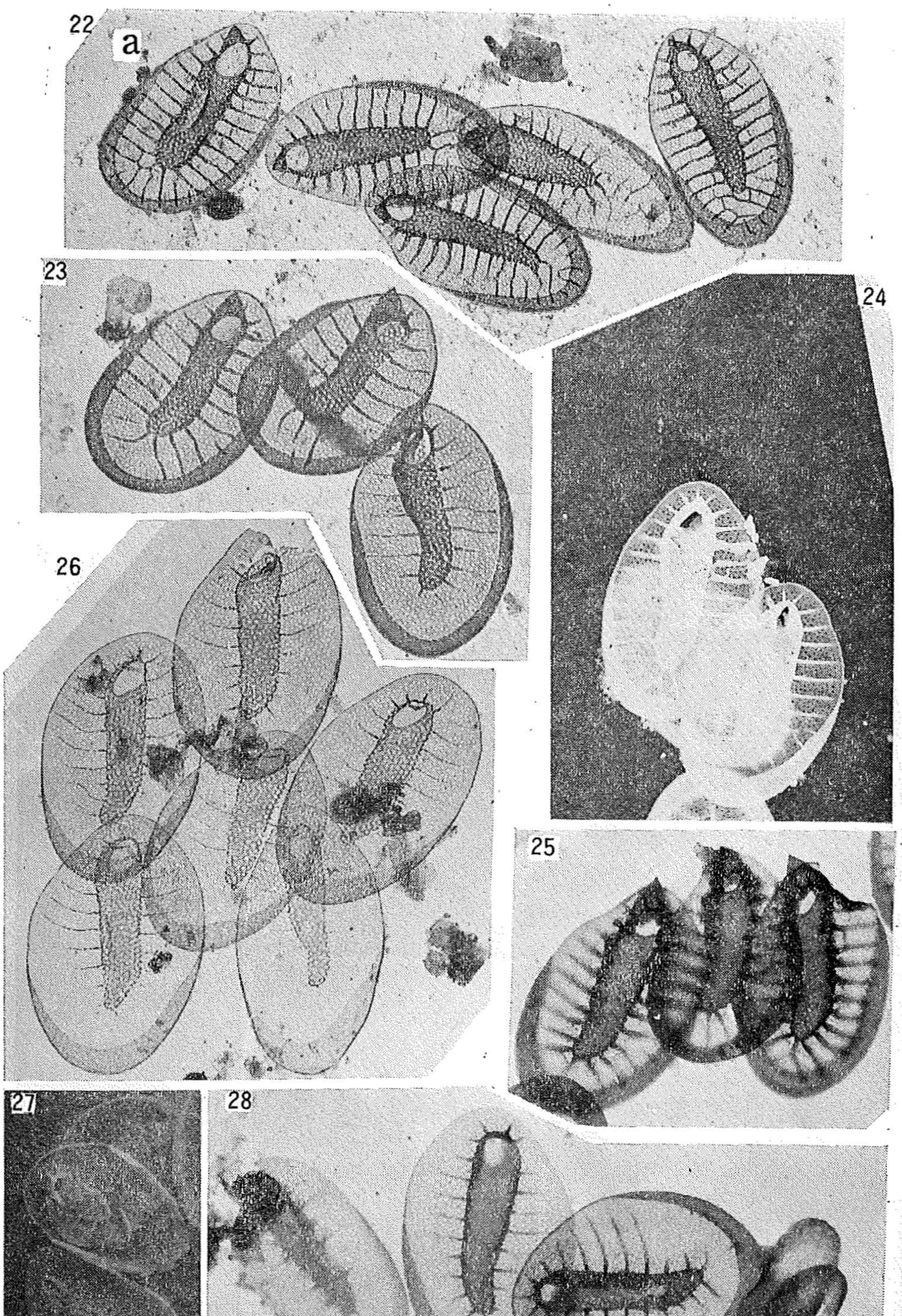
Fig. 77. Scale of *Synura* sp. (Paddy field) ×50, 000

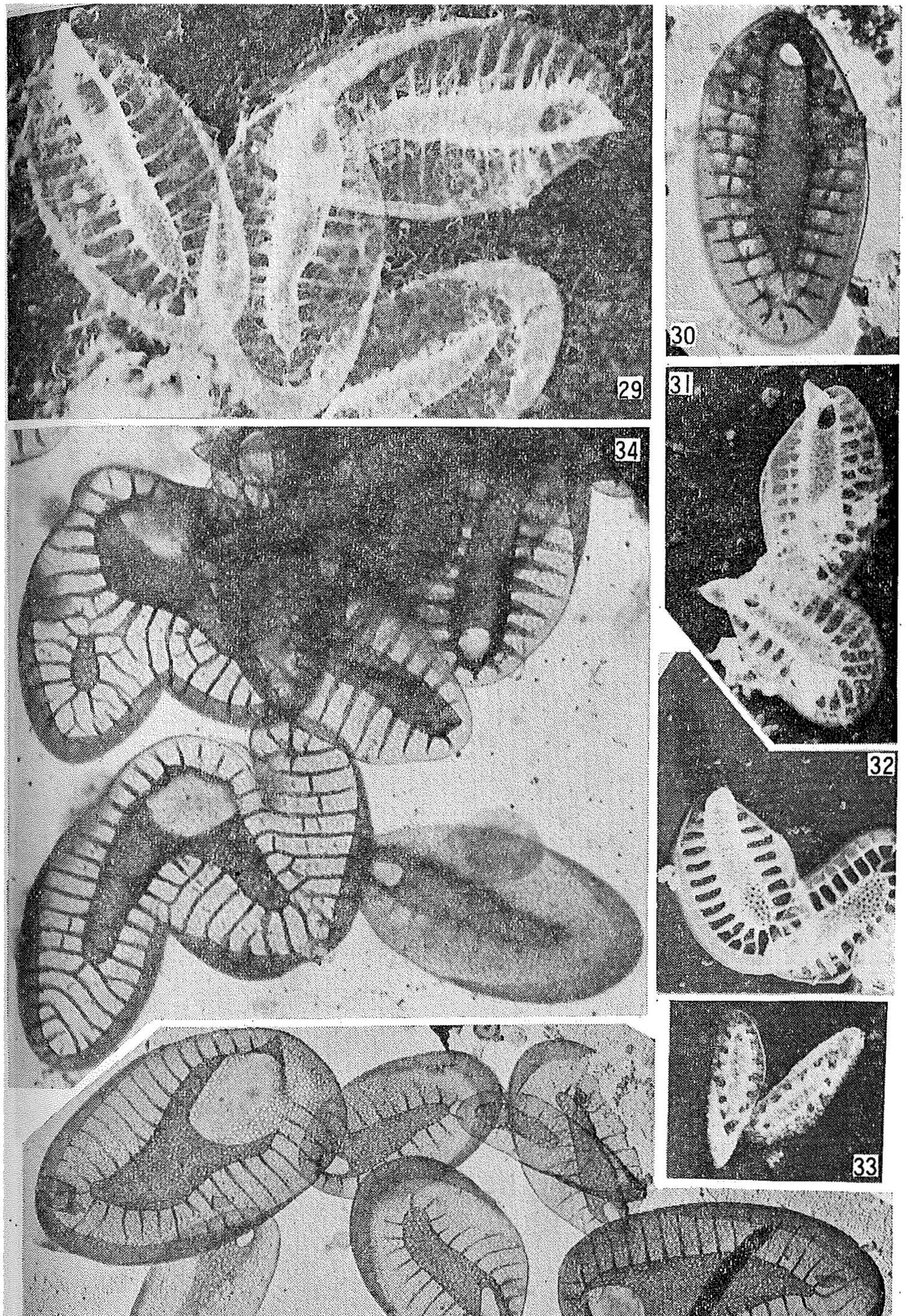
Fig. 78. Spine of *Synura echinulata* forma

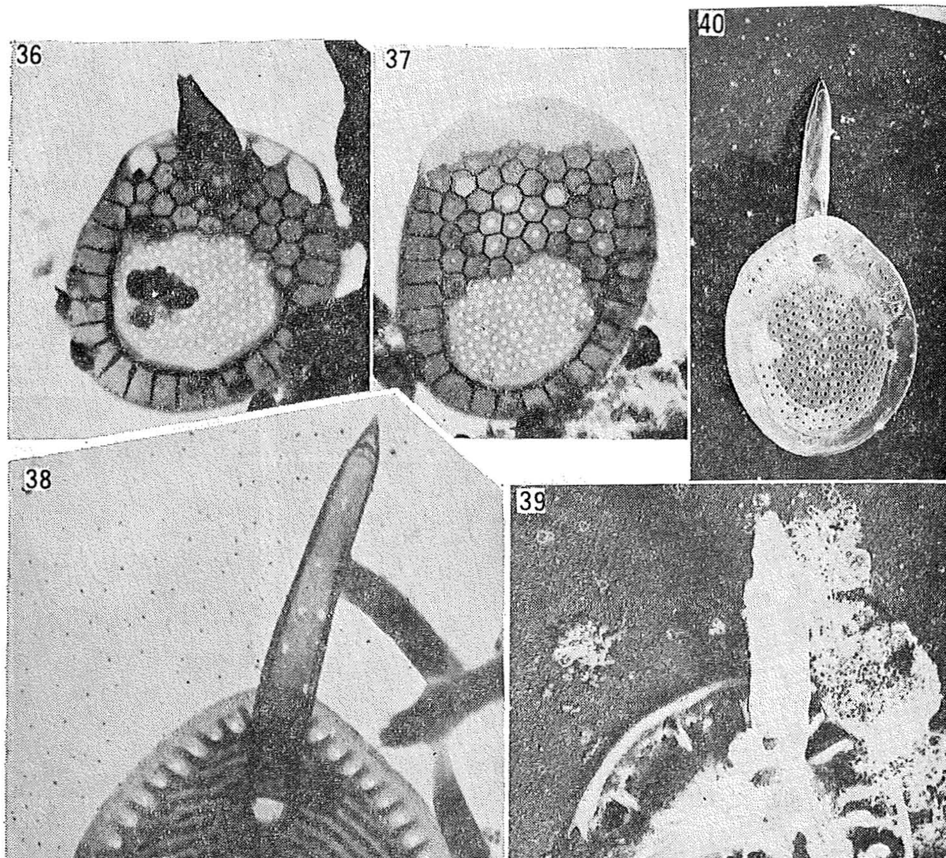

1
2
3
4

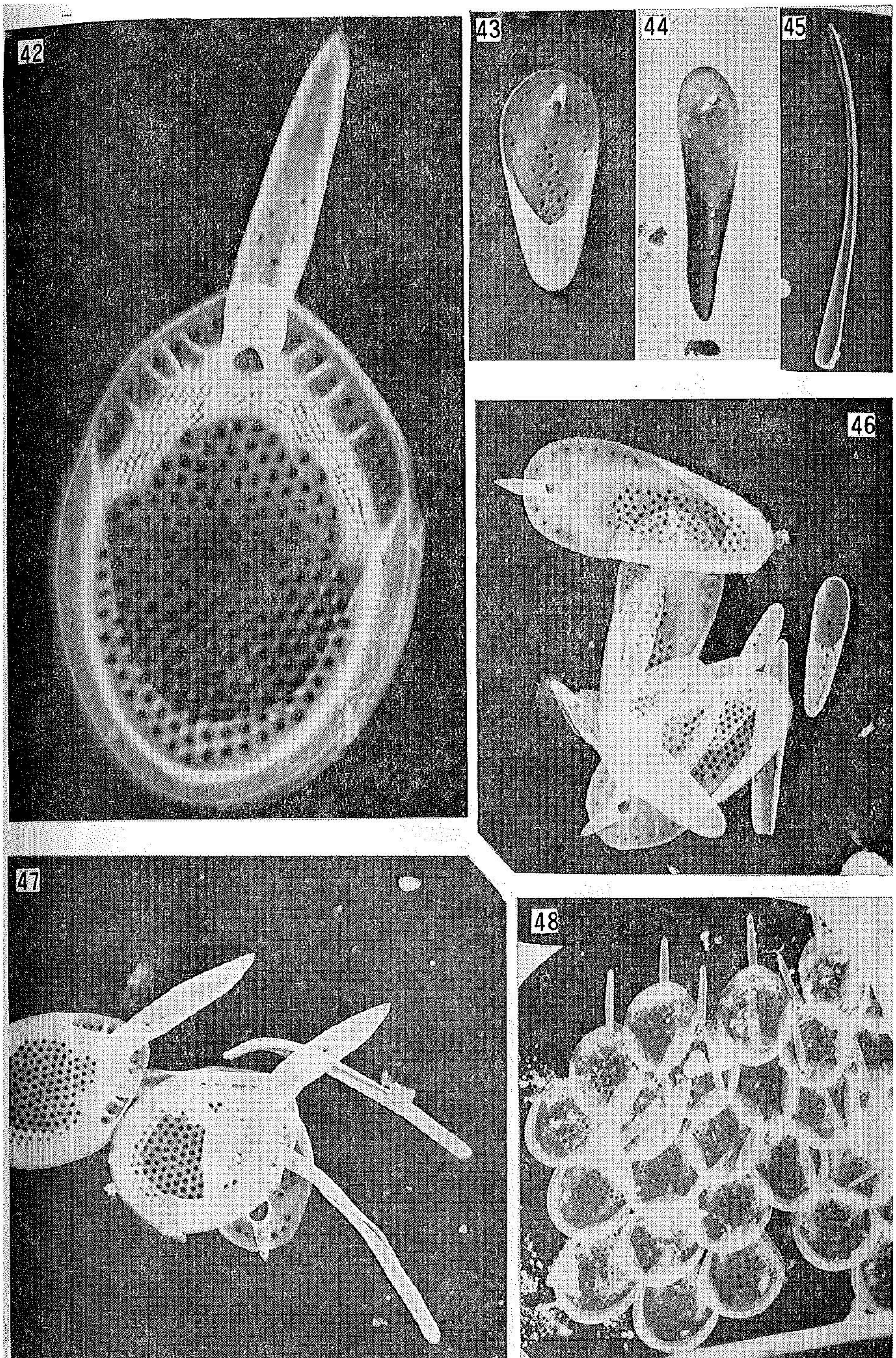

5
a
6
a
7
8

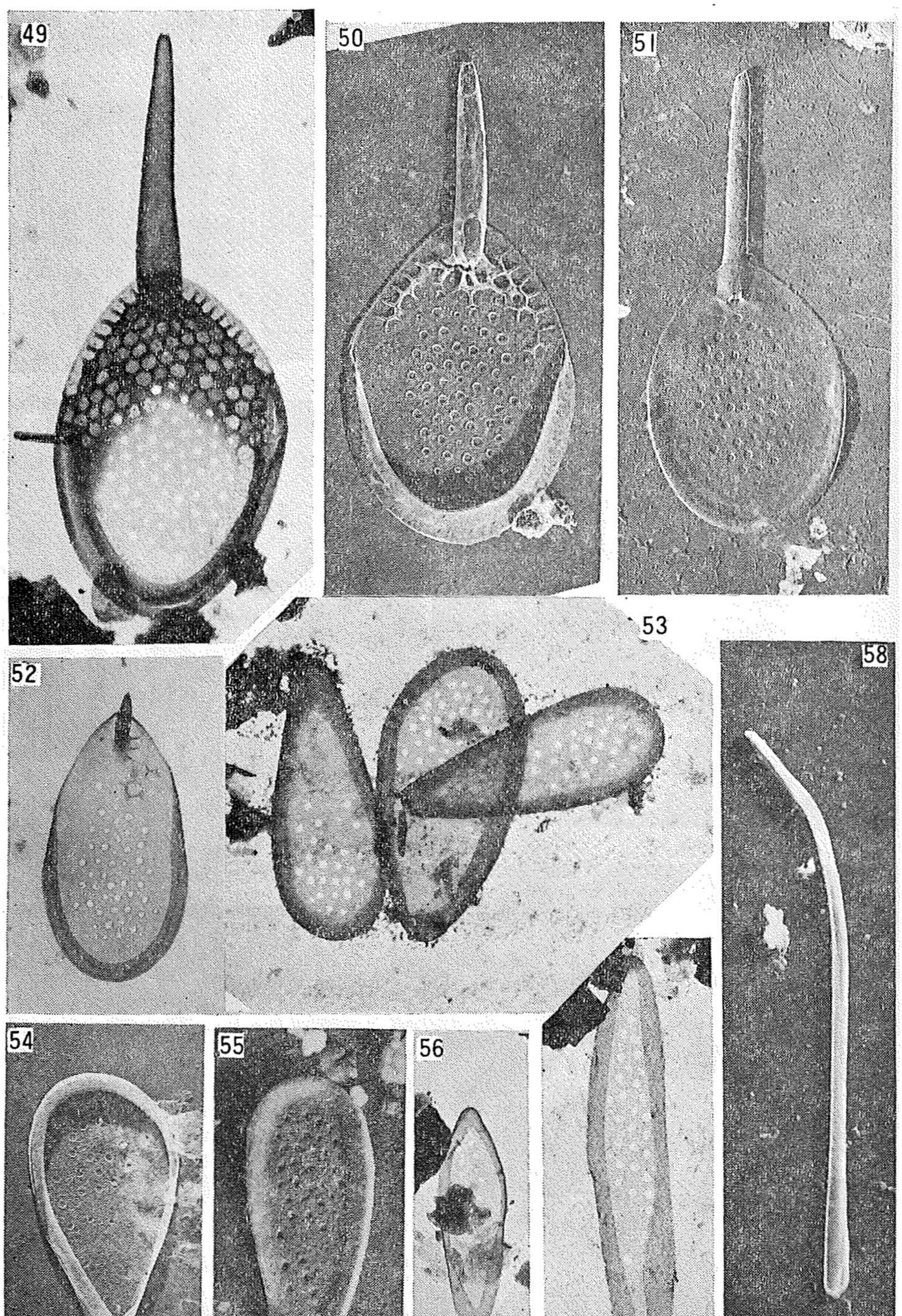

9
10
11
12
13
14
15
16
17
18
19
20
21

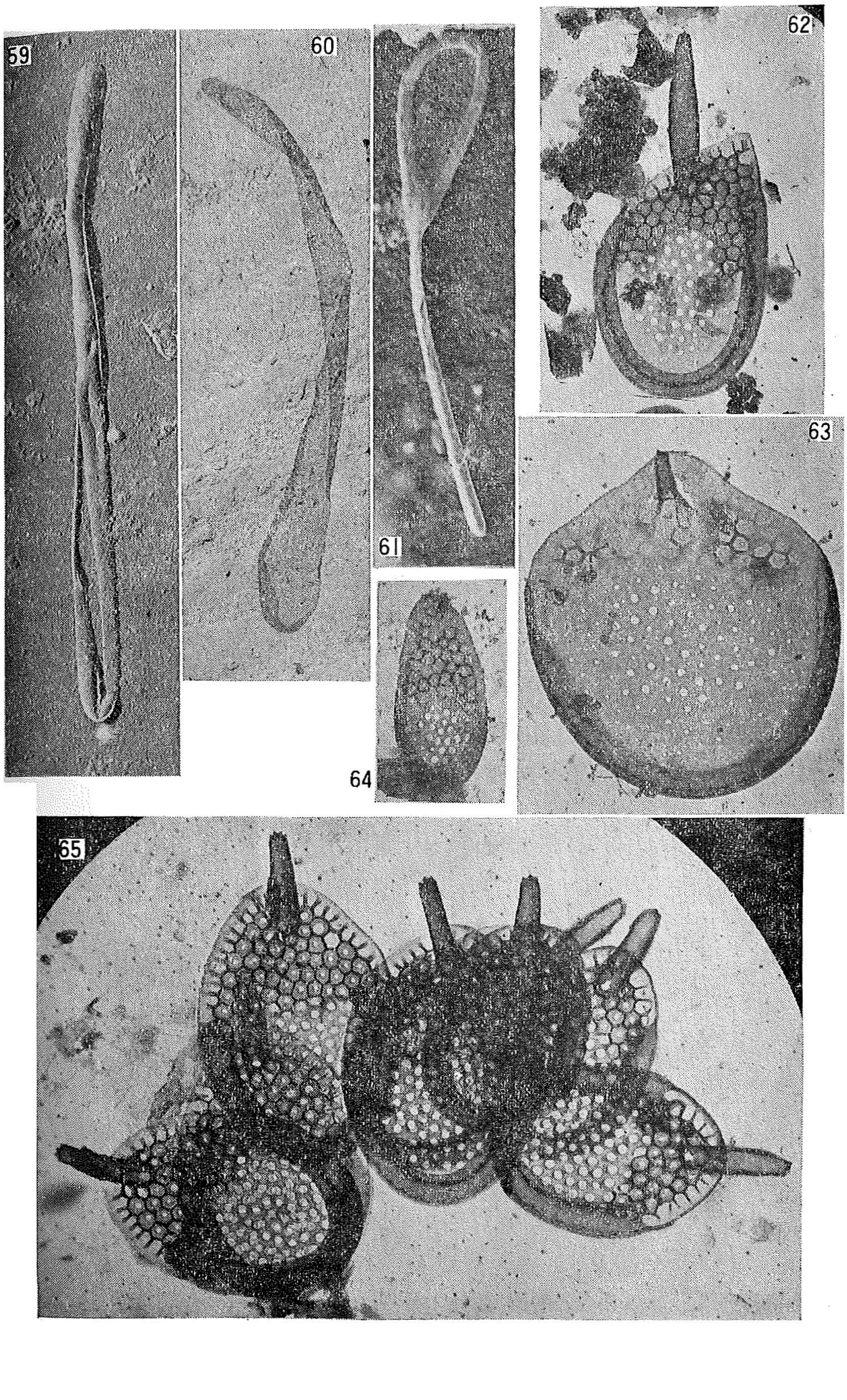

22
a
23
24
26
25
27
28

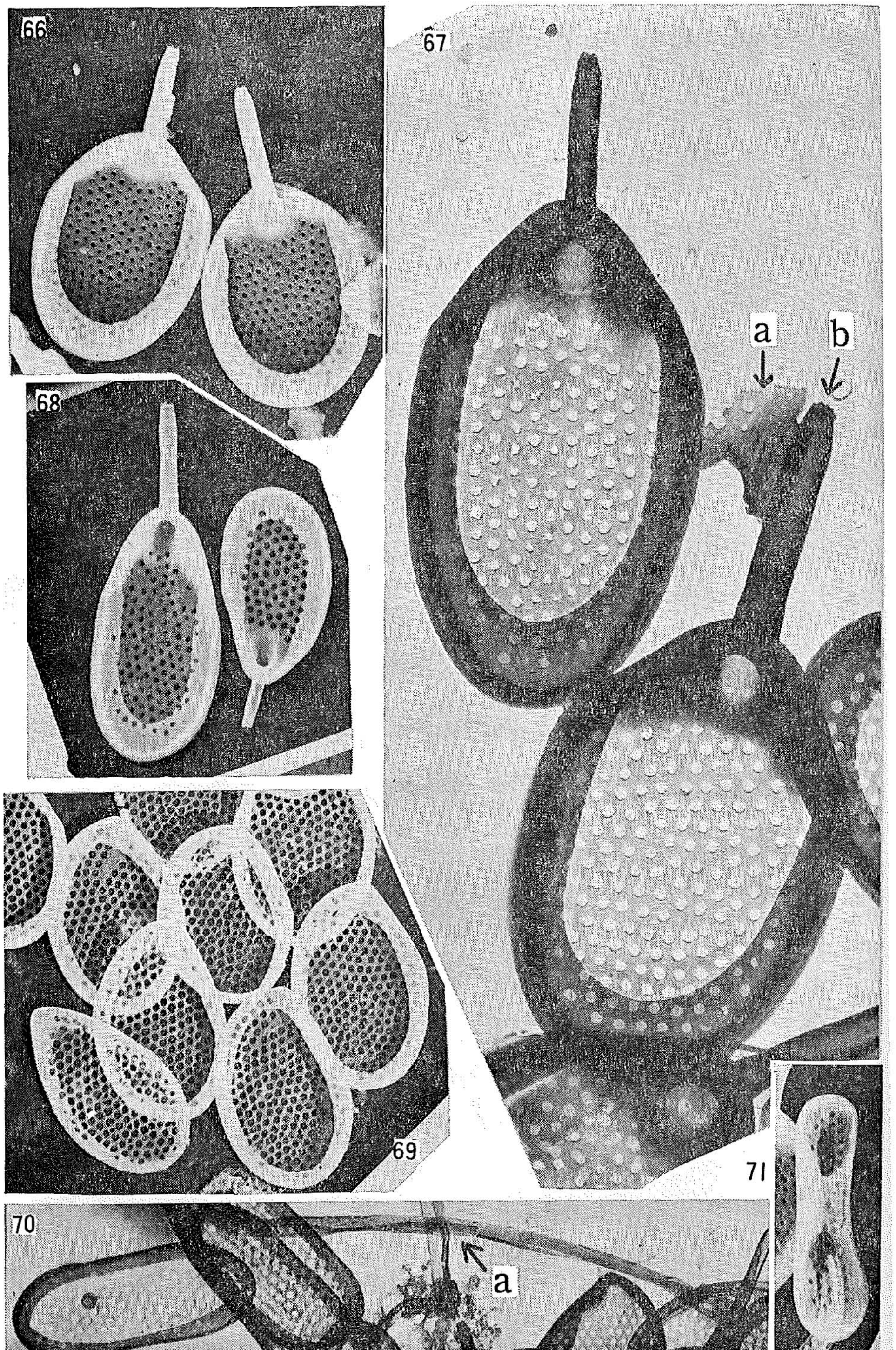

29
30
31
34
32
33
35

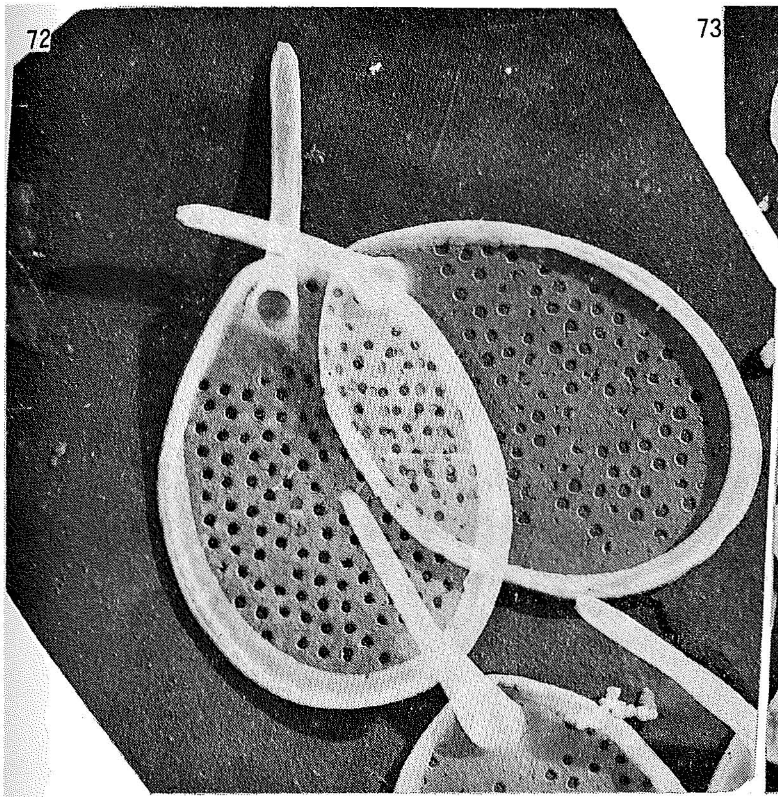

36
37
40
38
39
41

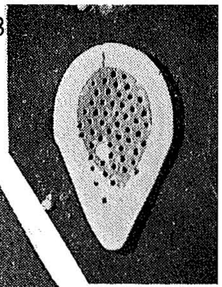

42
43
44
45
46
47
48


49
50
51
52
53
54
55
56
57
58

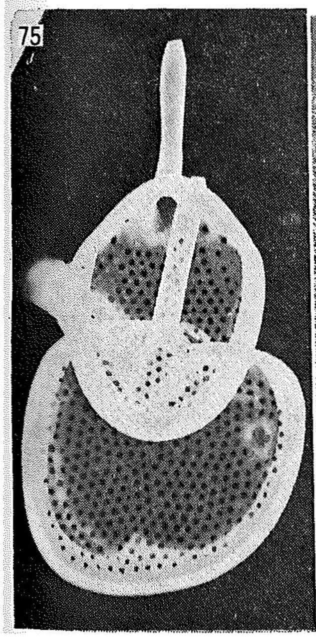

59
60
61
62
63
64
65

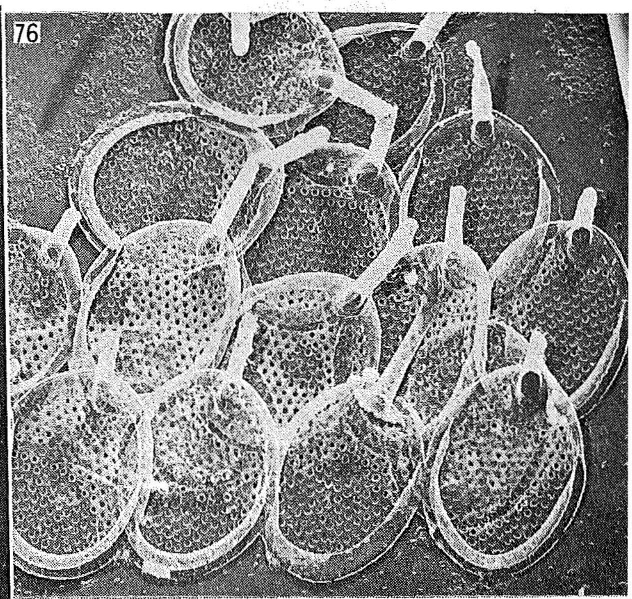

66
67
a
b
68
69
70
a
71

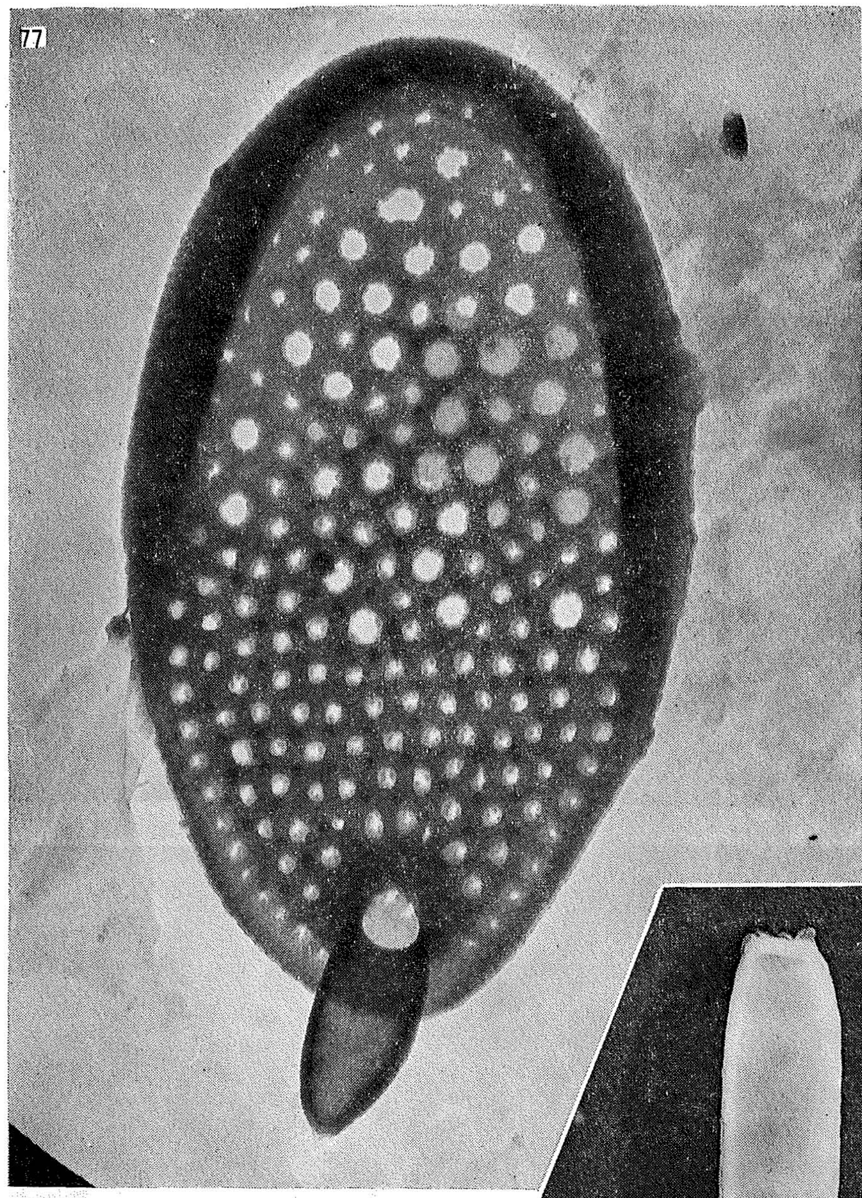

72

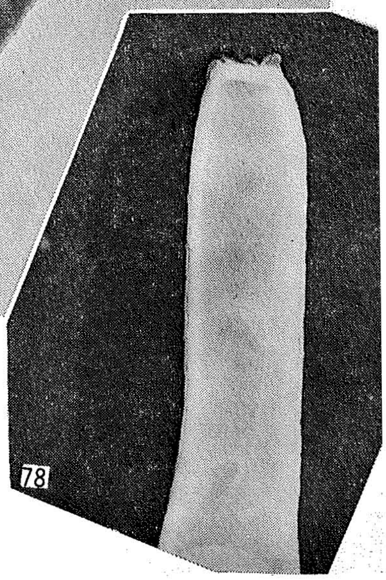

73


74


75


76


77


78