Jap. J. Phycol. (Sôrui) 29: 85-93. June 20, 1981

Taxonomic studies on *Scenedesmus* in Japan 1. On *Scenedesmus acuminatus* (LAG.) CHOD. and its varieties and *S. javanensis* CHOD.

Kaiichi OOSHIMA

Biological Laboratory, College of Agriculture and Veterinary Medicine, Nihon University, Fujisawa, Kanagawa, 252 Japan

OOSHIMA, K. 1981. Taxonomic studies on *Scenedesmus* in Japan 1. On *Scenedesmus acuminatus* (LAG.) CHOD. and its varieties and *S. javanensis* CHOD. Jap. J. Phycol. 29: 85-93.

In order to determine the circumscription of *Scenedesmus acuminatus* and its closely related taxa, a number of morphological features and their variations were examined based upon sixteen isolates cultured in seven kinds of media. Consequently, it was confirmed that the following characters were apparently stable: 1) the shape of cells: lunate or sigmoid, 2) the arrangement of cells in the 4- and 8-celled coenobia: linear, alternate or zigzag, 3) the form of colonies: flat, curved or twisted. On the basis of these features, *S. acuminatus* var. *acuminatus*, *S. acuminatus* var. *falcatus*, *S. acuminatus* var. *tortuosus*, and *S. javanensis* were recognized as distinct taxa.

Key index words: *Green algae; Chlorococcales; Chlorophyceae;* Scenedesmus; S. acuminatus; S. javanensis; *taxonomy*.

Since MEYEN (1829) established the genus *Scenedesmus*, four monographs have been published by CHODAT (1913, 1926), G. M. SMITH (1916) and UHERKOVICH (1966). Numerous species and infraspecific taxa belonging to *Scenedesmus* have been described based only on a few samples from natural collections. Consequently, the taxonomy of the genus, which contains more than 100 species, is currently in confusion.

On the other hand, TRAINOR and coworkers have investigated many strains of polymorphic *Scenedesmus* and obtained excellent results, especially on morphological variability and the control of unicell formation (TRAINOR 1966, 1967, 1969, 1979; TRAINOR and ROSKOSKY 1967; TRAINOR and ROWLAND 1968; TRAINOR and SHUBERT 1974; SHUBERT and TRAINOR 1974; TRAINOR *et al.* 1976). SHUBERT (1975) has made a new attempt to describe a polymorphic species on the basis of morphological and physiological characteristics observed in the cultures. The use of culture methods is today of primary importance to taxonomic studies of *Scenedesmus*. In order to determine the circumscription of the individual species and infraspecific taxa, it is necessary to obtain as many cultures as possible and to examine the stability and flexibility of related characteristics.

The present paper constitutes the first of a series of taxonomic studies on *Scenedesmus* in Japan, and deals with the morphology and taxonomy of *S. acuminatus* (LAG.) CHOD. and several closely related taxa, maintained in unialgal cultures.

Materials and Methods

The unialgal cultures used for this study were originally isolated from the algal habitats summarized in Table 1. All the cultures were immediately established from a 4- or 8-celled coenobium by using PRINGSHEIM's micropipette-washing method (STEIN

Table 1. Source of unialgal cultures investigated.

Strain No.	Localities	Dates	Cell No. / colony
421	Senzokuike pond, Tokyo	Dec. 10, '75	4
452	Sanaruko-lake, Shizuoka	May 12, '76	4
459	Sanaruko-lake, Shizuoka	May 12, '76	4
462	Sanaruko-lake, Shizuoka	May 12, '76	4
473	Sanaruko-lake, Shizuoka	May 17, '76	8
492	Ashigaike pond, Aichi	May 18, '76	4
495	Ashigaike pond, Aichi	May 18, '76	4
501	a small pond, Okayama	Oct. 16, '75	8
505	a small pond, Okayama	Oct. 16, '75	8
510	a small pond, Okayama	Oct. 16, '75	4
539	Ashigaike pond, Aichi	Aug. 24, '76	8
555	a small pond, Aichi	Aug. 25, '76	8
562	Ashigaike pond, Aichi	Aug. 25, '76	4
630	a small pond, Ibaraki	May 30, '77	8
699	a small pond, Saitama	May 12, '78	4
703	a moat around the castle, Shimane	Aug. 21, '78	8

Table 2. Composition of media (mg/*l*).

Constituent \ Medium	A	B	C	D10	D200	F	AWB
KNO_3	300	300	300	30	1.5	300	300
NH_4NO_3	200	200	200	20	1	200	200
KH_2PO_4	30	—	20	2	0.1	20	20
K_2HPO_4	—	30	10	1	0.05	10	10
$MgSO_47H_2O$	50	50	50	5	2.5	50	50
$NaHCO_3$	20	20	20	2	0.1	20	20
Fe-citrate	—	—	—	—	—	0.5	0.1
Soil Extract	—	—	—	—	—	—	10 ml
pH	4.6 ~ 4.8	8.2 ~ 8.4			6.8 ~ 7.0		

1973), and were maintained at 20°C under fluorescent lights of ca. 3000 lux intensity with a 12/12 hr light-dark cycle.

Four weeks after the beginning of incubation, a drop of the original cultures was suspended by shaking and was transferred into a series of test tubes containing the seven kinds of media shown in Table 2. Medium A, B and C were different in acidity. Medium D10 and D200 were made by diluting medium C and were the same in acidity. Medium F was also made by adding ferric citrate to medium C. Medium AWB with 0.6% agar was employed as an agar medium for the agar-water biphasic culture medium, which was composed of two phases, a solid phase of agar and a liquid phase of water (OOSHIMA 1975). This medium was also employed for isolation. The others were used as liquid media. All the media were sterilized by autoclaving. These series of test tubes inoculated were kept under the same culture conditions mentioned above and were gently shaken every other day.

After two to four weeks, a total of 200–300 individual organisms in each of the test tubes was examined; the percentages of coenobial types were recorded, and the shape, size and arrangement of cells and the form of coenobia were observed under high magnification. The length of the cells was measured and recorded as that of the longitudinal axes of the cells.

Results and Discussion

Scenedesmus acuminatus (LAG.) CHOD. and the related taxa have been classified based on the following characters: the shape and size of cells, the arrangement of cells in each colony, and the form of the 4- and 8-celled coenobia. Concerning the stability and flexibility of these characters, the results obtained in the present investigation examining each strain in various media provides useful information.

Unicells: The strains used in this study frequently produced a high percentage of unicells in medium B, and seldom in the others. In the older cultures of medium B, there was a tendency toward a higher percentage of unicells. These formed colonies when transferred into fresh media. The colonies were also similar in shape and size to the original cell. These results suggest that the unicells observed are quite different from those in the polymorphic strains of spiny *Scenedesmus*, reviewed by TRAINOR *et al.* (1976).

Cell shape and size: The culture conditions employed in the present investigation produced little abnormal change in the shape of cells in all the vigorous cultures. Cells from newly released colonies were more or less lunate or sigmoid, tapering to a point. They increased gradually in dimension with age. When the cells matured, the proportions between length and width had changed to some degree. They were about one and half times as long as the young cells, and about twice in width. However, they retained the lunate or sigmoid shape. Consequently, the shape of cells proved to be one of the stable characteristics for all strains examined.

On the other hand, the size of cells was variable at each stage of the cultures, especially in the mature condition. The bigger mature cells had a strong tendency to produce 8-celled coenobia.

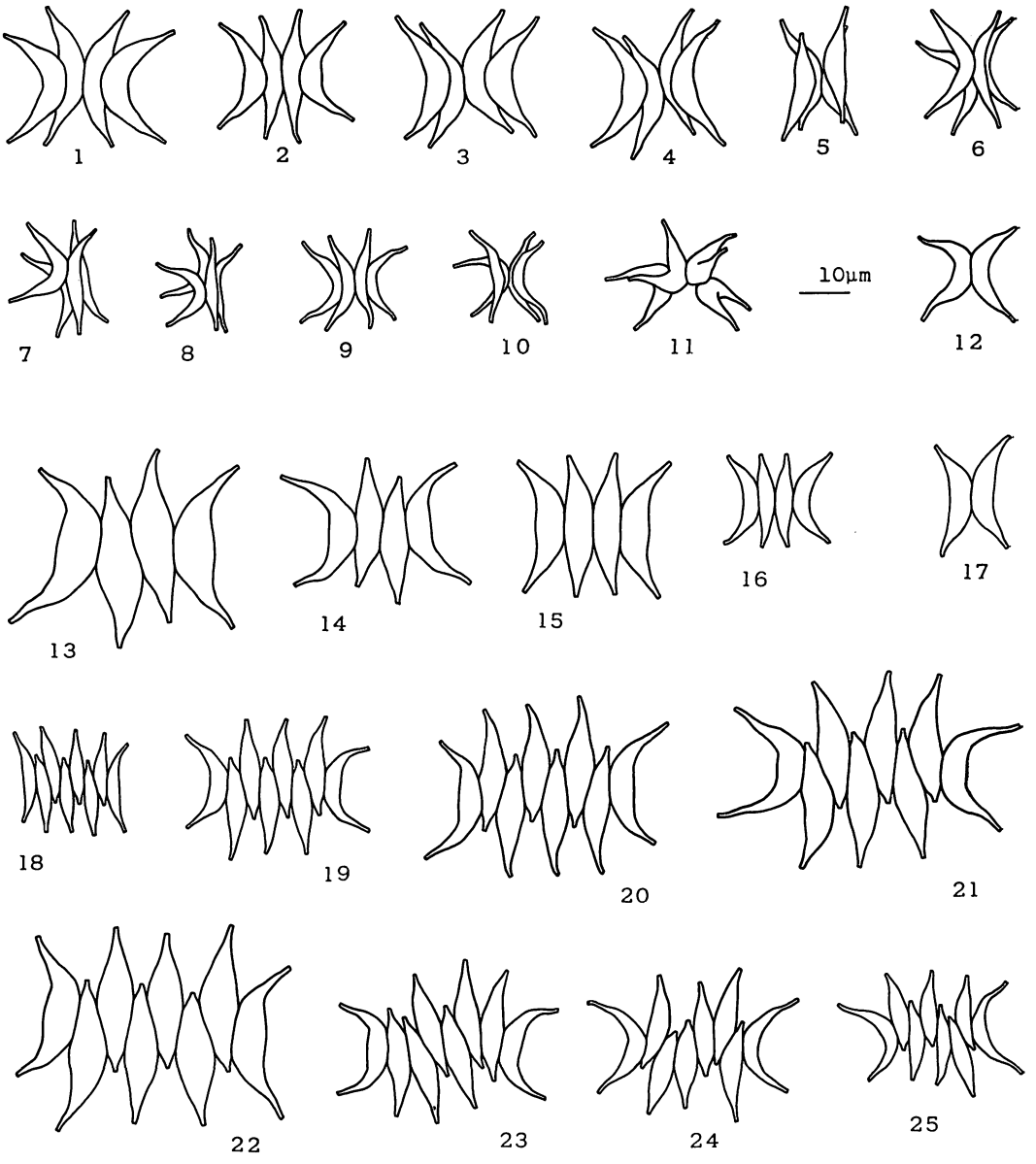
Cell arrangement: The cells were regularly arranged in the 4- and 8-celled coenobia in the front view. All the cultures of strains 501, 505 and 510 constantly showed a unique zigzag arrangement of cells in the 4- and 8-celled coenobia (Figs. 40, 43). In the other strains, the cells were arranged either in a linear series or in an alternate series, and never in such a double series as *S. costulatus* CHOD. or *S. ecornis* var. *disciformis* CHOD. The alternate arrangement was more conspicuous in the 8-celled coenobia (Figs. 18–22). This character was stable in each strain examined.

Colony form: In vertical view, we can clearly observe the form of colonies in which the cells join one another just like a raft. Each strain kept the form of 4- and 8-celled coenobia constant. The fifteen strains produced either the flat or curved coenobia, while strain 492 distinctively produced the twisted coenobia (Figs. 26–31). The form of the 2-celled coenobia, however, showed strikingly similarity for all strains observed (Figs. 12, 17, 37). According, the form of the 4- and 8-celled coenobia proved to

Table 3. Comparisons of four taxa investigated in the present study.

Taxa	Cell Shape T : M	Cell Size T : M	Cell arrangement 4-C	8-C	Form of 4-C & 8-C
S. acuminatus var. *acuminatus*	L L	≒	1-a	—	curved
var. *falcatus*	L L	>	1-a	a	flat
var. *tortuosus*	L L	≒	1-a	a	twisted
S. javanensis	L S	>	z	z	flat

* T, terminal cells; M, median cells; L, lunate; S, sigmoid; 4-C, 4-celled coenobia; 8-C, 8-celled coenobia; a, alternate series; 1, linear series; z, zigzag series.



Figs. 1-12. *S. acuminatus* var. *acuminatus*. 1-5. 4-celled mature coenobia; 6-10. 4-celled young coenobia; 6-8. side view of 4-celled coenobia; 11. vertical view; 12. 2-celled coenobium.

Figs. 13-25. *S. acuminatus* var. *falcatus*. 13-15. 4-celled mature coenobia; 16. 4-celled young coenobium; 17. 2-celled coenobium; 18. 8-celled young coenobium; 21 and 22. 8-celled mature coenobia; 23-25. irregular arrangement of cells in 8-celled coenobia.

be one of the stable characteristics.

On the basis of the stable characters described above, the sixteen strains examined were grouped into four taxa, as summarized in Table 3.

1. *Scenedesmus acuminatus* (LAG.) CHOD. var. ***acuminatus*** MAT. Fl. Crypt. Suiss. **1**(3): 211. *fig.* 88. 1902.

——**Figs. 1-12**

In Table 4, the percentages of coenobial

types observed in the 2-week-old cultures are given for strain 452. The number of cells forming colonies was always two or four in all the media, and the 8-celled coenobia were never found during this investigation. Unicells appeared only in medium B. The 4-week-old cultures of medium B, which produced 29% unicells, were no longer vigorous, and the cells in these cultures were brownish in color and abnormal in shape. Similar results were obtained with strains 421, 459, 539 and 562.

Each cell forming a coenobium was little different in shape and size. The cells in the young coenobia were 19-21 μm long and 2-2.5 μm broad, while the largest cells in the mature coenobia were up to 34.5 μm long and 5.5 μm broad. The dimensions of the mature cells were approximately equal to those in some descriptions (LAGERHEIM 1882; CHODAT 1902, 1926; SMITH 1920). The shape of the cells was more or less lunate. The terminal cells in the 4-celled coenobia were somewhat more curved than the median cells (Figs. 6-8).

The cells of the 4-celled coenobia were never arranged in a plane. The coenobia were always curved and sometimes semicircular in vertical view (Fig. 11). Although it changed to some degree with age, the curvature of the 4-celled coenobia was constant in all the vigorous cultures. In the front view of the coenobia, on the other hand, the cells were arranged in a linear series (Figs. 1, 2), and sometimes in an alternate series (Figs. 3-5).

From the results of the present investigation, the five strains examined were definitely identified with *S. acuminatus* (LAG.) CHOD. (1902). These results, moreover, supported the opinion of LAGERHEIM (1882), CHODAT (1902, 1926) and SMITH (1916, 1920) that the curvature of the 4-celled coenobia was one of the important characteristics for this species. Accordingly, this taxon can be clearly distinguished from the others.

2. *S. acuminatus* var. *falcatus* (CHOD.) OOSHIMA nov. comb.

S. falcatus CHOD. Rev. Hydrol. **3**: 146-147. *figs.* 36-37. 1926. ——**Figs. 13-25**

The results obtained from strain 462 are shown in Table 5. The 4-week-old cultures of the strain were not vigorous in medium B and D200. High percentages of the 8-celled coenobia were recorded especially in medium AWB and D10. Unicells were always produced in medium B, and they were rarely found in the others, *e.g.* in medium AWB (strain 462) and in medium C (strain 473). Similar results were obtained with strains 473, 495, 555, 630, 699 and 703.

In each of the 4- and 8-celled coenobia, the terminal cells were different from the median cells in shape and size. The shape of the median cells was slightly lunate, while the terminal cells showed the characteristic curved shape. In most cases, curving was more conspicuous in mature coenobia (Figs. 21, 22) than in young ones (Fig. 18). The terminal cells were longer than the median cells, and were about the same in width. In the largest of the 8-celled coenobia, the terminal cells were up

Table 4. Percentages of unicells and coenobia produced in seven kinds of media (strain 452: *S. acuminatus* var. *acuminatus*).

2 w.	AWB	A	B	C	D10	D200	F
unicells	0%	—	1%	0%	0%	0%	0%
2-celled	6	—	15	31	52	67	42
4-celled	94	—	84	69	48	33	58
8-called	0	—	0	0	0	0	0

*A: dead

Table 5. Percentages of unicells and coenobia produced in seven kinds of media (strain 462: *S. acuminatus* var. *falcatus*).

2 w.	AWB	A	B	C	D10	D200	F
unicells	0%	—	1%	0%	0%	0%	0%
2-celled	0	—	3	0	0	0	0
4-celled	12	—	84	51	43	79	50
8-celled	88	—	12	49	57	21	50

*A: dead

to 35.5 μm long and 7 μm broad, and the median cells were up to 32.5 μm long and 7 μm broad. In the 4-celled coenobia, the terminal cells were up to 42 μm long and 8.5 μm broad, and the median cells were up to 34 μm long and 9 μm broad.

The form of the 4- and 8-celled coenobia was flat and rarely curved in some degree. The cells in the 8-celled coenobia were always arranged in a markedly alternate series and never in a linear series (Figs. 18-22). In some of the coenobia, however, they were arranged in irregular series (Figs. 23-25). The percentage of the irregularly arranged coenobia was about 6% in strain 462. In the 4-celled coenobia, on the other hand, the arrangement of the cells was variable, from linear (Figs. 15, 16) to strikingly alternate (Figs. 13, 14).

The seven strains examined were clearly identified with *S. falcatus* CHOD. (1926). Although many authors have regarded it as synonymous with *S. acuminatus*, the above-mentioned results give good grounds for the recognition of this taxon. *S. falcatus* differs from *S. acuminatus* in the form of 4-celled coenobia, but closely resembles it in other respects: the shape of cells and the arrangement of cells in 4- and 8-celled coenobia. Taking the similarities into consideration, it would be more natural to consider this taxon as a variety and to name *S. acuminatus* var. *falcatus* (CHOD.) OOSHIMA.

Some of the 8-celled coenobia observed in the vigorous cultures (Fig. 20) were similar to those of *S. acuminatus* f. *gyoparosiensis* (KISS) UHERK. (1966), which was originally described based only upon the 8-celled coenobia. It seems open to doubt whether the characteristic structure of the 8-celled coenobia, which has been used as the only criterion for characterizing the taxon, is stable.

3. *S. acuminatus* var. *tortuosus* (SKUJA) OOSHIMA nov. comb.

S. falcatus* f. *tortuosa SKUJA Act a Hort. Bot. Univ. Latv. **2**: 83. *fig.* 4. 1927. ***S. acuminatus* f. *tortuosus*** (SKUJA) UHERK. *Scenedesmus*-Art. Ung. 43. *figs.* 71-78. 1966. **——Figs. 26-37**

The percentages of coenobial types recorded for strain 492 are presented in Table 6. The 4-celled coenobia were always dominant in all the media, while the 8-celled coenobia were found only in medium C and F.

The form of the 4- and 8-celled coenobia was distinctively twisted. Viewing them from different angles, the 4-celled coenobia varied in appearance (Figs. 26-30, 32-36). The twisted form was more conspicuous in the side or vertical view (Figs. 26, 29). The cells in the 4-celled coenobia were arranged in a linear or slightly alternate series, and the 8-celled coenobia showed a strikingly alternate arrangement of the cells which was quite similar to that of the var. *falcatus* (Fig. 31).

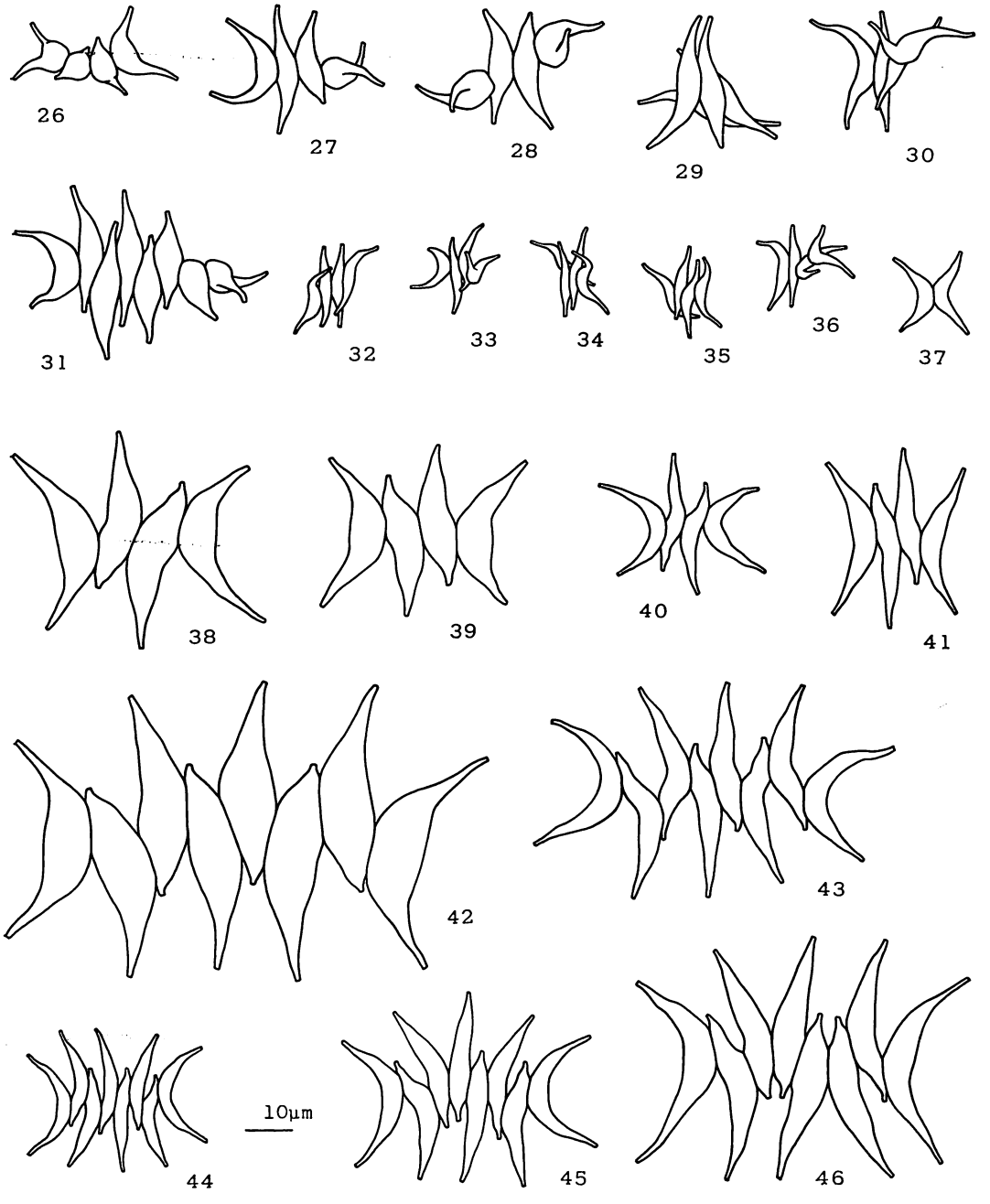
The cells of each colony were usually alike both in shape and size. Most of them were approximately the same in length and width though the terminal cells were sometimes a little longer than the median cells. The large cells in the mature coenobia were up to 35.5 μm long and 6.5 μm broad. The dimensions measured in strain 492 were smaller than those in SKUJA's description (1927). The shape of the cells was lunate and sometimes looked sigmoid or α-shaped in appearance because of the twist of each colony.

Strain 492 was identified with *S. acuminatus* f. *tortuosus* (SKUJA) UHERK. The results of the present investigation confirmed

Table 6. Percentages of unicells and coenobia produced in seven kinds of media (strain 492: *S. acuminatus* var. *tortuosus*).

2 w.	AWB	A	B	C	D10	D200	F
unicells	0%	—	30%	0%	0%	0%	0%
2-celled	2	—	17	0	2	5	0
4-celled	96	—	53	90	98	05	93
8-called	0	—	0	10	0	0	7

*A: dead



Figs. 26-37. *S. acuminatus* var. *tortuosus*. 26-30. 4-celled mature coenobia; 31. 8-celled mature coenobium; 32-36. 4-celled young coenobia; 37. 2-celled coenobium.

Figs. 38-46. *S. javanensis*. 38 and 39. 4-celled mature coenobia; 40 and 41. 4-celled young coenobia; 42. 8-celled mature coenobium; 44. 8-celled young coenobium; 45 and 46. irregular arrangement of cells in 8-celled coenobia.

the original description of SKUJA (1927) and clarified the circumscription of this taxon, which has been misunderstood because of the too fragmentary description. On the basis of the characteristic features mentioned above, especially the unique form of colonies,

the taxon should be raised to the higher rank and named *S. acuminatus* var. *tortuosus* (SKUJA) OOSHIMA.

4. *S. javanensis* CHOD. Rev. Hydrol. **3**: 157. *fig.* 47. 1926. ——**Figs. 38-46**

In strain 510, the 8-celled coenobia were always dominant in all the vigorous cultures, while the 2- and 4-celled coenobia were sometimes observed in the older cultures. As shown in Table 7, the 4-week-old cultures of medium B produced not only three types of coenobia but also 5% unicells which were brownish in color and abnormal in shape. Similar results were obtained with strains 501 and 505.

In the 4- and 8-celled coenobia, the terminal cells were lunate in shape and the median cells were sigmoid. The sigmoid shape was more unusual in the median cells of the younger coenobia than in those of the mature coenobia. As they matured, the median cells showed a tendency to become lunate in appearance (Fig. 42). Strains 501, 505 and 510 all possessed this feature.

Cells in the 4- and 8-celled coenobia were about the same in width, but the terminal cell was always longer than the median cells. The terminal cells in the 4-celled coenobia were up to 55 μm long and 11 μm broad, and the median cells were up to 46.5 μm long and 11.5 μm broad. In the 8-celled coenobia, on the other hand, the terminal cells were up to 43 μm long and 8.5 μm broad, and the median cells were up to 36 μm long and 8.5 μm broad. These dimensions are much larger than those in CHODAT's description (1926).

Table 7. Percentages of unicells and coenobia produced in seven kinds of media (strain 510: *S. javanensis*)

4 w.	AWB	A	B	C	D10	D200	F
unicells	0%	—	5%	0%	0%	—	0%
2-celled	0	—	1	0	0	—	0
4-celled	26	—	6	0	0	—	2
8-celled	74	—	88	100	100	—	98

*A and D200: dead

The form of the 4- and 8-celled coenobia was flat and rarely curved a little. In some cases, however, the terminal cells were at an angle to the whole coenobium. The arrangement of the cells was very characteristic and it proved to be a stable, species-specific feature. The two median cells in the 4-celled coenobia and in the center of the 8-celled coenobia were in contact with each other only at the middle, and were joined with the outer cells by their apices. As a result, the cells were distinctively arranged in a unique zigzag series (Figs. 40, 43).

The irregular arrangement of the cells in the 8-celled coenobia was observed in the vigorous cultures (Figs. 45, 46). In the 2-week-old cultures of strain 501, the percentage of the irregularly arranged coenobia was on the average about 7% and at most 15% in medium F.

The three strains examined were definitely identified with *S. javanensis* CHOD. (1926). The original description is quite similar to the young coenobia observed in these cultures. Considering the above-mentioned features, especially the distinctive zigzag arrangement of cells and the sigmoid shape of median cells, this taxon should be recognized as a distinct species.

This species bears some resemblance to *S. bernardii* SMITH (1916) in the zigzag arrangement of cells, but is quite different from it in the shape and size of cells. Based on these characters, *S. bernardii* is similar to *S. acutus* MEYEN (1829) rather than to *S. javanensis*.

Acknowledgements

The author would like to express his appreciation to Dr. H. KOBAYASI of the Tokyo Gakugei University for his suggestions and encouragement. His thanks are also due to Dr. S. KOMIYA and Mr. T. NAGUMO of the Nippon Dental University and Dr. S. KATO of the Tokyo Metropolitan

University for their generous aid in collecting some of the original samples.

References

CHODAT, R. 1902. Algues vertes de la Suisse. Pleurococcoides-Chroolépoides. Mat. Fl. Crypt. Suiss. 1(3) : 1-373.

CHODAT, R. 1913. Monographie d'algues en culture pure. Mat. Fl. Crypt. Suiss. 4(2) : 1-266.

CHODAT, R. 1926. *Scenedesmus*. Étude de génétique, de systématique expérimentale et d'hydrobiologie. Rev. Hydrol. 3 : 71-258.

LAGERHEIM, G. 1882. Beidrag till kännedomen on Stockholmstraktens Pediastréer, Protococcacéer och Palmellacéer. Öfv. Vet.-Ak. Verh. 39 : 47-81.

MEYEN, F.J.F. 1829. Beobachtungen über einige niedre Algenformen. Nova Acta Phys.-Med. Acad. Caes. Leop.-Car. 14 : 771-778.

OOSHIMA, K. 1975. The agar-water biphasic culture medium for obtaining clonal cultures of microscopic algae. Bull. Nipp. Dent. Coll. Gen. Ed. 1975 : 275-286.

SHUBERT, L.E. 1975. *Scenedesmus trainorii* sp. nov. (Chlorophyta, Chlorococcales) : a polymorphic species. Phycologia 14 : 177-182.

SHUBERT, L.E. and TRAINOR, F.R. 1974. *Scenedesmus* morphogenesis. Control of the unicell stage with phosphorus. Br. Phycol. J. 9 : 1-7.

SKUJA, H. 1927. Vorarbeiten zu einer Algenflora von Lettland. III. Acta Hort. Bot. Univ. Latv. 2 : 51-116.

SMITH, G.M. 1916. A monograph of the algal genus *Scenedesmus* based upon pure culture studies. Trans. Wisc. Acad. Sci. 18 : 422-530.

SMITH, G.M. 1920. Phytoplankton of the inland lakes of Wisconsin. Part 1. Bull. Wisc. Geol. Nat. Hist. Surv. 57 : 1-243.

STEIN, J.R. 1973. Handbook of phycological methods. Cambr. Univ. Press, London.

TRAINOR, F.R. 1966. A study of wall ornamentation in cultures of *Scenedesmus*. Amer. J. Bot. 53 : 995-1000.

TRAINOR, F.R. 1967. Spine pattern in several clones of a *Scenedesmus*. Trans. Amer. Microsc. Soc. 86 : 16-21.

TRAINOR, F.R. 1969. *Scenedesmus* morphogenesis. Trace elements and spine formation. J. Phycol. 5 : 185-190.

TRAINOR, F.R. 1979. *Scenedesmus* AP1 (Chlorophyceae) : polymorphic in the laboratory but not in the field. Phycologia 18 : 273-277.

TRAINOR, F.R. and ROSKOSKY, F. 1967. Control of unicell formation in a soil *Scenedesmus*. Can. J. Bot. 45 : 1657-1664.

TRAINOR, F.R. and ROWLAND, H.L. 1968. Control of colony and unicell formation in a synchronized *Scenedesmus*. J. Phycol. 4 : 310-317.

TRAINOR, F.R. and Shubert, L.E. 1974. *Scenedesmus* morphogenesis. Colony control in dilute media. J. Phycol. 10 : 28-30.

TRAINOR, F.R., CAIN, J.R. and SHUBERT, L.E. 1976. Morphology and nutrition of the colonial green alga *Scenedesmus* : 80 years later. Bot. Rev. 42 : 5-25.

UHERKOVICH, G. 1966. Die *Scenedesmus*-Arten Ungarns. Akad. Kiad. Budapest.

大島海一：日本産セネデスムス属の分類学的研究 1. *Scenedesmus acuminatus* とその変種および *S. javanensis* について

7種類の培地を用いて *S. acuminatus* に類似する16株について単藻培養実験を行ない，その形態的特徴の安定性および変異を調べた。その結果，次の三形質は安定していることが明らかになった。1) 細胞の形：三日月形あるいはS字形。2)定数群体内における細胞の配列パターン：直線的配列，交互配列あるいはジグザグ配列。3) 定数群体全体の形：平面的か，湾曲しているかあるいはねじれ面を形成しているか。これらの諸形質に基づいて，*S. acuminatus* var. *acuminatus*, *S. acuminatus* var. *falcatus*, *S. acuminatus* var. *tortuosus*, *S. javanensis* の4分類群が明確に識別された。(252 藤沢市亀井野1866 日本大学農獣医学部生物学研究室)