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Evaluation of River Water Quality by Diatoms

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In order to evaluate simply the Japanese-type river water quality, a practicable method using differentiating diatom groups is proposed. Japanese rivers are generally shorter, shallower, and run faster. Therefore, though the present grouping is considered to be equivalent to Lange-Bertalot's (Category 1-3. *Nova Hedwigia Beih.* **64:** 285-304. 1979), considerable differences have resulted in the members of the relevant groups. For the evaluation of the water quality of rivers, the employment of the formula of Pantle-Buck giving a saprobic value of 4 to the Group A taxa, 2.5 to the Group B taxa, and 1 to the others is suggested. This method is simple and easy and the results of applying this method to the Japanese river waters were in general agreement with chemical analyses.

Key Words: diatom, differentiating diatom groups, pollution-tolerant diatom groups, water pollution, water quality evaluation

INTRODUCTION

Presently, the necessity of a simple, easy and routine method for evaluation of river water quality is emphasized as pollution is getting more and more severe and is spreading. Various devices have been proposed by various authors to solve this problem (Patrick *et al.*, 1954; Liebman, 1962; Sládečeck, 1973; Lange-Bertalot, 1979; Descy, 1979; Watanabe *et al.*, 1985, 1988, etc.).

The methods can be said to be composed of the following three main steps, namely (1) identification and counting (2) rating or grouping of the individual species in accordance with their tolerance of or affinities to the pollution (3) expression of the degree of pollution.

The first step is the most important step because the data obtained by this step constitutes the basis of all methods referred to above. Though accurate identification is especially difficult in the case of microscopic objects such as diatoms, this step cannot be disregarded. At the same time the number of valves or frustules to be counted is also important to get an accurate value of the relative frequency of each taxon (Schoeman, 1979; Kobayasi and Mayama, 1982).

In the second step, diatoms found in the samples collected from various rivers by us and the Bureau of Environmental Protection of the Tokyo Metropolitan Government (Kobayasi *et al.*, 1985) were classified into three groups. Members to be included in Group A (most pollution-tolerant group), Group B (less pollution-tolerant group) and Group C (pollution sensitive group) are listed.

In the third step, the Pantle & Buck Formula (1955) was used for the evaluation of the water quality of each sampling station giving a saprobic value of 4 to the Group A taxa, 2.5 to the Group B taxa, and 1 to the Group C taxa with some exceptions for which intermediate values should be given.

The saprobic indices obtained corresponded very well with the chemical analyses. Thus the use of the above method is suggested.

MATERIALS AND METHODS

Materials used were written or listed in our previous papers accompanied with sampling places, dates and sample numbers. These were collected from various rivers in Tokyo and its vicinity by us and the Bureau of Environmental Protection of the Tokyo Metropolitan Government (Kobayasi and Mayama, 1982; Mayama and Kobayasi, 1984; Kobayasi *et al.,* 1985). A total of 308 samples were re-examined and re-analyzed in the present studies.

BOD_5 and other physico-chemical parameters were estimated by us at the time of our sampling, but for the samples collected by the Tokyo Metropolitan Government, the data obtained by periodical estimation from the fixed sampling points of the same Government were used.

Methods of sampling, cleaning, washing, preparing objects for light and electron microscopy and counting are given in Kobayasi and Mayama (1982).

RESULTS AND DISCUSSION

In the first step, photomicrographs were taken from each slide and printed at exactly 2000 magnification. These were identified and arranged to lay out an illustrated guide for counting. In this study, about 400 taxa were recorded in the guide. All species encountered in a number of transects across the prepared slide were identified using the guide and counted until a minimum of 500 valves had been scored. The relative frequency of each taxon in each sample was calculated after the countings were completed.

A taxon quite similar to *Achnanthes minutissima,* a typical oligosaprobic species, was found frequently and sometimes abundantly in highly polluted rivers. This taxon was examined with SEM and TEM and newly described as *A. minutissima* var. *saprophila* by us (Kobayasi and Mayama, 1982; Mayama and Kobayasi, 1989).

Navicula atomus was treated by Krammer & Lange-Bertalot (1986) as composed of four varieties, var. *atomus,* var. *permitis* (Hust.) Lange-B., var. *excelsa* (Krasske) Lange-B. and var. *recondita* (Hust.) Lange-B., however, the former two varieties were treated as a single taxon in the present study (Mayama and Kobayasi, 1988).

Another difficulty is the case of identification of the small and tiny *Stephanodiscus* species (Kobayasi and Inoue, 1985; Kobayasi *et al.,* 1985a; Kobayasi *et al.,* 1985b; Kobayasi and Kobayashi, 1986; Kobayasi and Kobayashi, 1987). In Group B of the following list, all of these small species were calculated under the name *S. invisitatus.*

Difficulty in identification with LM is seen in three *Nitzschia* species which are often found together with each other in the same sample and have similar shapes and structures. Distinctions between these three species are possible only by examining the SEM structure of the striae on the canal raphe (Kobayasi, 1985). The striae of *N. frustulum* are straight and without bifurcations, *N. huntzschiana* has bifurcate striae with furcate branches composed of a single areola, and *N. romana* has bifurcate striae with furcate branches composed of two areolae. In the present study, samples containing these *Nitzschia* species were examined first by SEM. After we identified which of the species occurred alone or co-occurred with any other, LM countings were carried out.

In the second step, the relative frequency of each species was plotted against the BOD_5 value of the water from which it was collected. The distribution pattern of dots in these graphs showed three types: (1) Type A showing a rise toward a higher BOD_5 value, (2) Type B showing a normal curve with a mode within water class II (β-meso-saprobic) to III (α-meso-saprobic), and (3) Type C showing a reverse pattern to Type A (Fig.

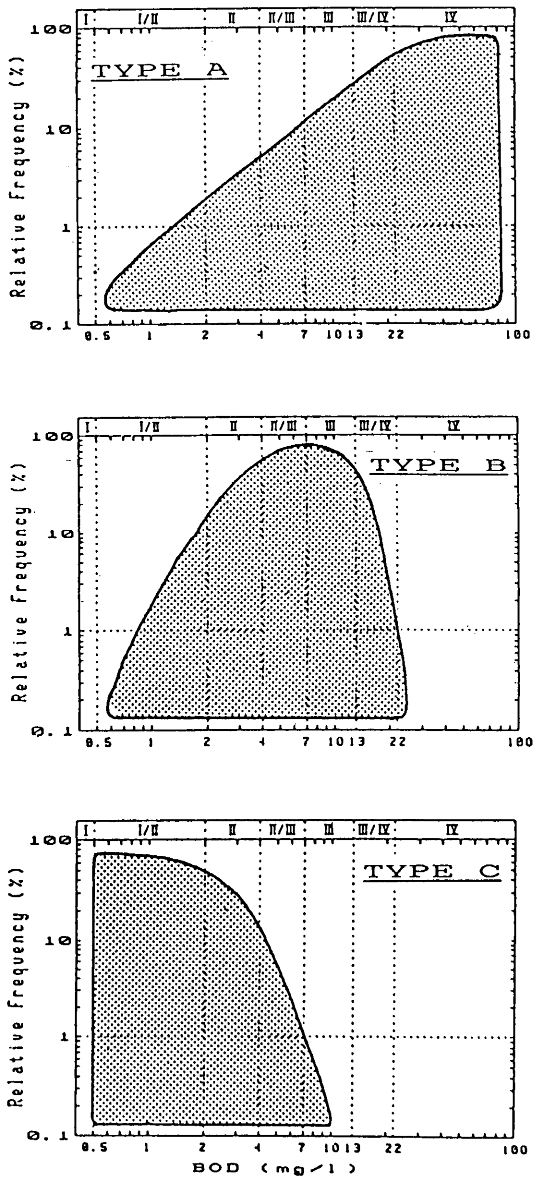


Fig. A. Distribution patterns of Type A, Type B and Type C species in relation to the relative frequency and the BOD_5.

A) (Kobayasi *et al.*, 1985).

The composition of the diatom communities is often different even if two sampling stations are only 10 cm distant from each other (Kobayasi and Mayama, 1981). Therefore, it is better to treat diatoms as groups such as group A-C mentioned above.

Taxa belonging to each group are listed below.

Group A taxa:

1. *Achnanthes minutissima* var. *saprophila* H. Kob. & Mayam. (Figs. 1-4).
2. *Gomphonema parvulum* Kuetz. (Figs. 5, 6).
3. *Navicula atomus* (Kuetz.) Grun. (Figs. 7-9).
4. *N. goeppertiana* (Bleisch) H.L. Smith (Figs. 10, 11).
5. *N. minima* Grun. (Figs. 12, 13).
6. *N. seminulum* Grun. (Figs. 14-16) [or *Sellaphora seminulum* (Grun.) D.G. Mann].
7. *N. veneta* Kuetz. (Figs. 17, 18).
8. *Nitzschia gandersheimiensis* Krasske (Figs. 19, 20).
9. *N. palea* (Kuetz.) Grun. (Figs. 21, 22).
10. *Pinnularia braunii* var. *amphicephala* (Mayer) Hust. (Figs. 23, 24).

Note: Give a saprobic rating of 4 to the taxa of this group, but give 3.25 to *N. veneta,* and 3.25 to *N. goeppertiana* only at fresh waters.

Group B taxa:

1. *Achnanthes brevipes* var. *intermedia* (Kuetz.) Cleve (Figs. 25, 26).
2. *A. delicatula* (Kuetz.) Grun. (Figs. 27, 28).
3. *A. exigua* Grun. (Figs. 29, 30).
4. *A. hungarica* (Grun.) Grun. (Figs. 31, 32).
5. *Amphora acutiuscula* Kuetz. (Figs. 33, 34).
6. *A. coffeaeformis* Ag. (Figs. 35, 36).
7. *A. luciae* Choln. (Figs. 37, 38).
8. *A. submontana* Hust. *(Figs. 39, 40).*
9. *A.* sp. (Figs. 41, 42).
10. *Bacillaria paradoxa* Gmel. (Figs. 43, 44).
11. *Cyclotella atomus* Hust. (Figs. 45, 46).
12. *C. criptica* Reiman *et al.* (Figs. 47, 48).
13. *C. meneghiniana* Kuetz. (Figs. 49, 50).
14. *C. stelligera* (Ehr.) Cleve & Grun. var. *stelligera.* (Figs. 51, 52).
15. *C. stelligera* var. *tenuis* Grun. (Figs. 53, 54).
16. *Cymbella microcephala* Grun. (Figs. 55-57).
17. *Fragilaria brevistriata* Grun. (Figs. 58, 59).
18. *F. construens* (Ehr.) Grun. var. *construens.* (Figs. 60, 61).

19. *F. construens* var. *binodis* (Ehr.) Grun. (Figs. 62, 63).
20. *F. elliptica* Schumann (Figs. 64, 65).
21. *F. pinnata* Ehr. (Figs. 66, 67).
22. *F. vaucheriae* (Kuetz.) B.-Petersen (Figs. 68, 69).
23. *Gomphonema pseudoaugur* Lange-B. (Figs. 70, 71).
24. *Melosira ambigua* (Grun.) O. Muell. (Figs. 72, 73).
25. *M. distans* var. *alpigena* Grun. (Figs. 74, 75).
26. *M. granulata* (Ehr.) Ralfs (Figs. 76, 77).
27. *Navicula accomoda* Hust. (Figs. 78, 79).
28. *N. capitata* Ehr. (Figs. 80, 81).
29. *N. confervacea* Kuetz. (Figs. 82, 83).
30. *N. excelsa* Krasske (Figs. 84, 85).
31. *N. gregaria* Donkin (Figs. 86, 87).
32. *N. margalithii* Lange-B. (Figs. 88, 89).
33. *N. molestiformis* Hust. (Figs. 90, 91). [= *N.* paucivisitata Patr., *N. twymaniana* Archib.].
34. *N. neoventricosa* Hust. (Figs. 94, 95).
35. *N. odiosa* Wallace (Figs. 92, 93).
36. *N. pupula* Kuetz. (Figs. 96, 97).
37. *N. pygmaea* Kuetz. (Figs. 98, 99).
38. *N. salinarum* Grun. (Figs. 100, 111).

Plate 1. Group A (most pollution-tolerant group). ×2000 unless otherwise stated.
Figs. 1-4. *Achnanthes minutissima* v. *saprophila*. 3, 4. TEM ×6000. Figs. 5, 6. *Gomphonema parvulum*. Figs. 7-9. *N. atomus*. 9. TEM ×8000. Figs. 10, 11. *N. goeppertiana*. Figs. 12, 13. *Navicula minima*. 13. TEM ×6000. Figs. 14-16. *N. seminulum*. 14. TEM ×8000. Figs. 17, 18. *N. veneta*. Figs. 19, 20. *Nitzschia gandersheimiensis*. Figs. 21, 22. *N. palea*. Figs. 23, 24. *Pinnularia braunii* v. *amphicephala*.

Plate 2. Group B (less pollution-tolerant group). ×2000 unless otherwise stated.
Figs. 25, 26. *Achnanthes brevipes* v. *intermedia*. Figs. 27, 28. *A. delicatula*. Figs. 29, 30. *A. exigua* Figs. 31, 32. *A. hungarica*. Figs. 33, 34. *Amphora acutiuscula*. Figs. 35, 36. *A coffeaeformis*. Figs. 37, 38. *A. luciae*. Figs. 39, 40. *A. submontana*. Figs. 41, 42. *A.* sp. Figs. 43, 44. *Bacillaria paradoxa*. Figs. 45, 46. *Cyclotella atomus*. Figs. 47, 48. *C. criptica*. 48. TEM ×7500. Figs. 49, 50. *C. meneghiniana*. Figs. 51, 52. *C. stelligera* v. *stelligera*. Figs. 53, 54. *C. stelligera* v. *tenuis*.

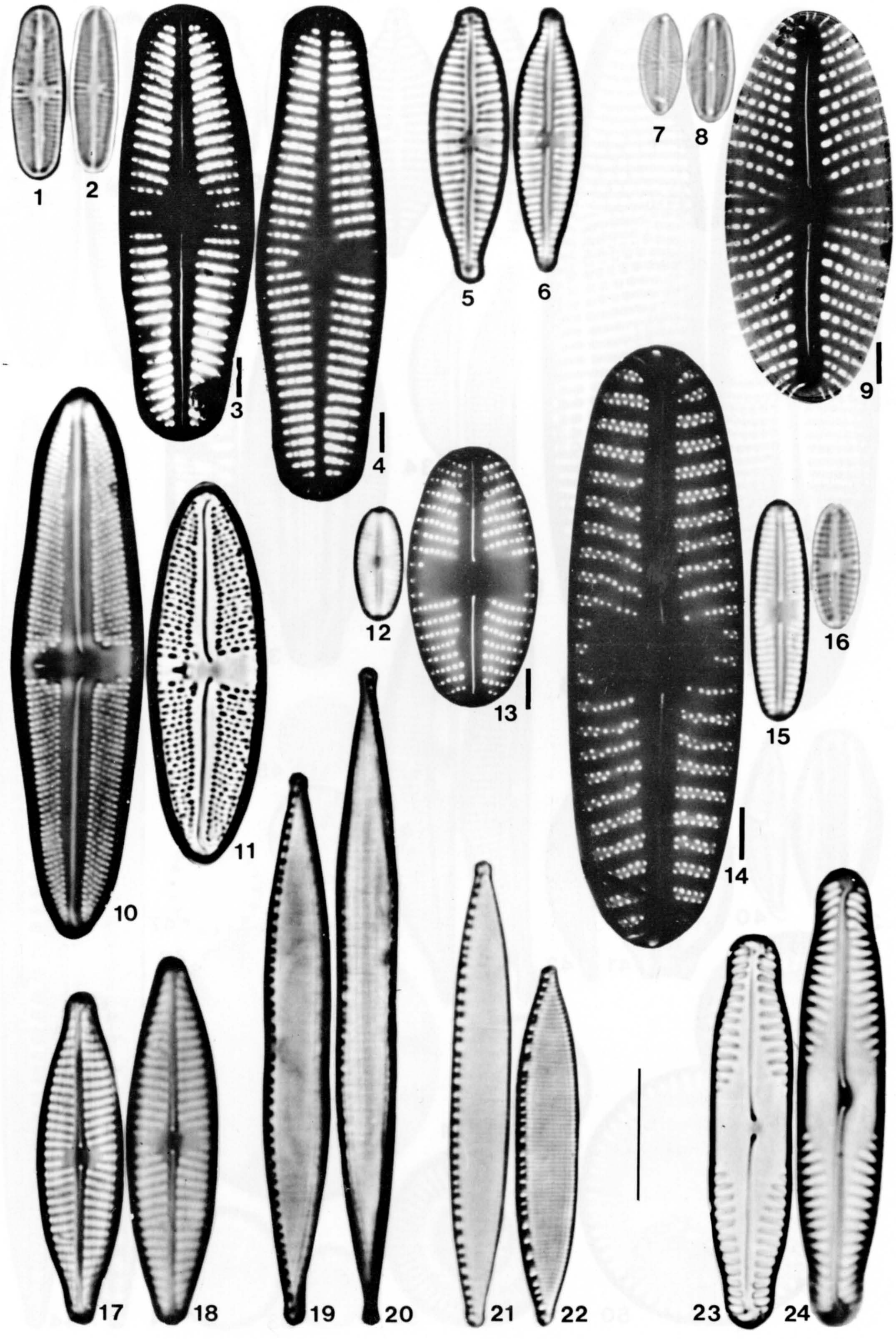
Plate 3. Group B (less pollution-tolerant group). ×2000 unless otherwise stated.
Figs. 55-57. *Cymbella microcephala*. Figs. 58, 59. *Fragilaria brevistriata*. Figs. 60, 61. *F. construens* v. *construens*. Figs. 62, 63. *F. construens* v. *binodis*. Figs. 64, 65. *F. elliptica*. Figs. 66, 67. *F. pinnata*. Figs. 68, 69. *F. vaucheriae*. Figs. 70, 71. *Gomphonema pseudoaugur*. Figs. 72, 73. *Melosira ambigua*. Figs. 74, 75. *M. distans* v. *alpigena*. Figs. 76, 77. *M. granulata*. Figs. 78, 79. *Navicula accomoda*. Figs. 80, 81. *N. capitata*. Figs. 82, 83. *N. confervacea*. Figs. 84, 85. *N. excelsa*. *85. TEM ×4000.* Figs. 86, 87. *N. gregaria*. Figs. 88, 89. *N. margalithii*. Figs. 90, 91. *N. molestiformis*. Figs. 92, 93. *N. odiosa*.

Plate 4. Group B (less pollution-tolerant group). ×2000 unless otherwise stated.
Figs. 94, 95. *Navicula neoventricosa*. Figs. 96, 97. *N. pupula*. Figs. 98, 99. *N. pygmaea*. Figs. 100, 101. *N. salinarum*. Figs. 102-104. *N. saprophila*. *102. TEM ×8700.* Figs. 105-107. *N. subminuscula*. Figs. 108, 109. *N. tenera*. Figs. 110, 111. *N. tripunctata*. Figs. 112, 113. *N. trivilis*. Figs. 114, 115. *Nitzschia filiformis*. Figs. 116-117. *N. amphibia*. Figs. 118, 119. *N. nana*. Figs. 120, 121. *N. solgensis*, Figs. 122, 123. *N. pusilla*. Figs. 124, 125. *N. tribrionella* v. *subsalina*.

Plate 5. Group B (less pollution-tolerant group). ×2000.
Figs. 126-129. *Nitzschia frustulum*. Figs. 130-133. *N. hantzschiana*. Figs. 134, 135. *N. intermedia*. Figs. 136, 137. *N. scalpelliformis*. Figs. 138, 139. *N. umbonata*. Figs. 140, 141. *Pinnularia gibba*.

Plate 6. Group B (less pollution-tolerant group). ×2000 unless otherwise stated.
Fig. 142. *Pleurosigma salinarum*. Figs. 143. *Synedra ulna*. Figs. 144, 145. *S. fasciculata*. Figs. 146, 147. *Skeletonema costatum*. 146. Living specimen, Phase contrast, ×1000. Figs. 148, 149. *Surirella angusta*. Figs. 150, 151. *Stephanodiscus invisitatus*. Fig. 152. *Thalassiosira faurii*. Fig. 153. *T. lacustris*. Figs. 154, 155. *T. weissflogii*..

Plate 1. (lengends see p.124)



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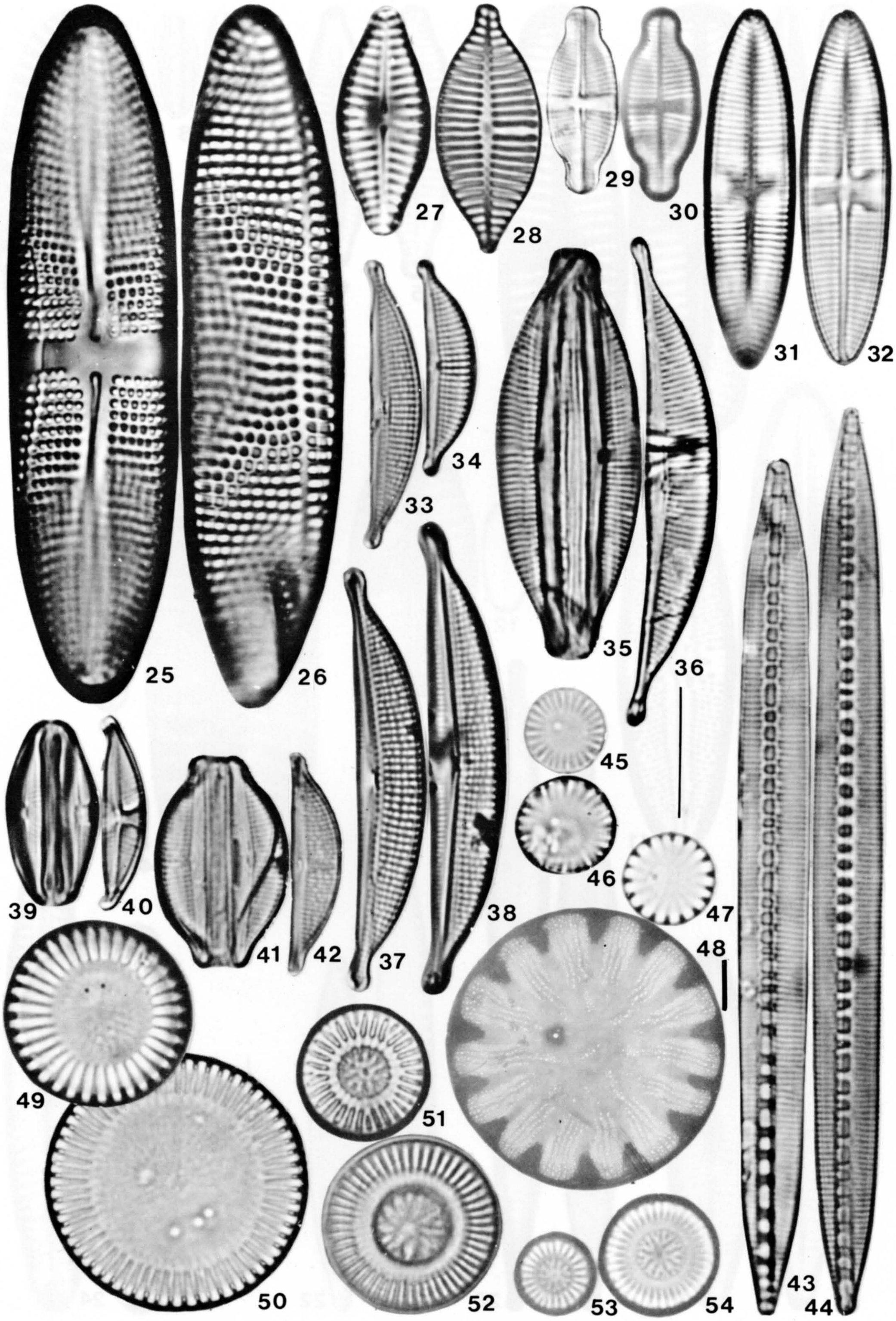

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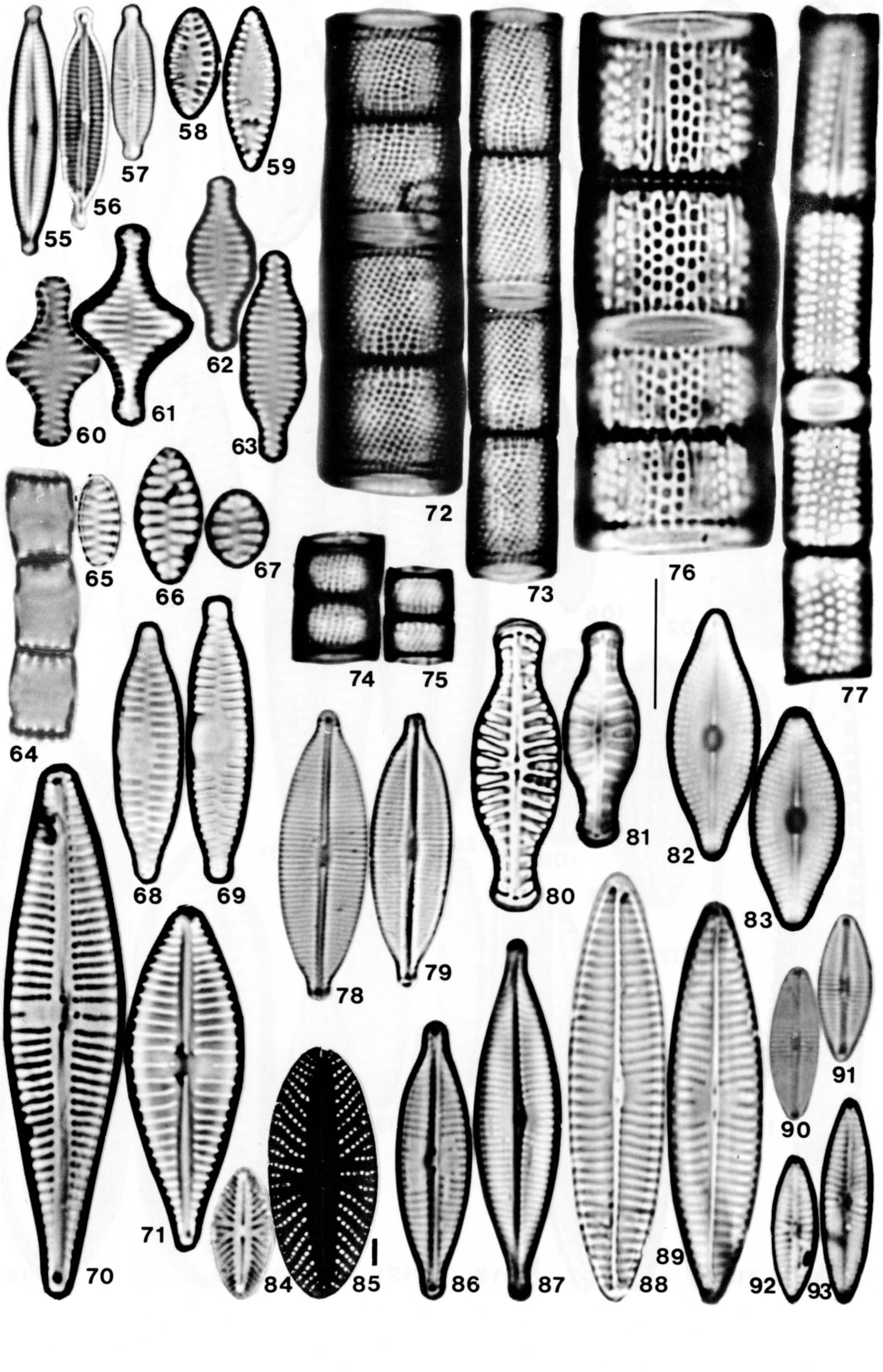

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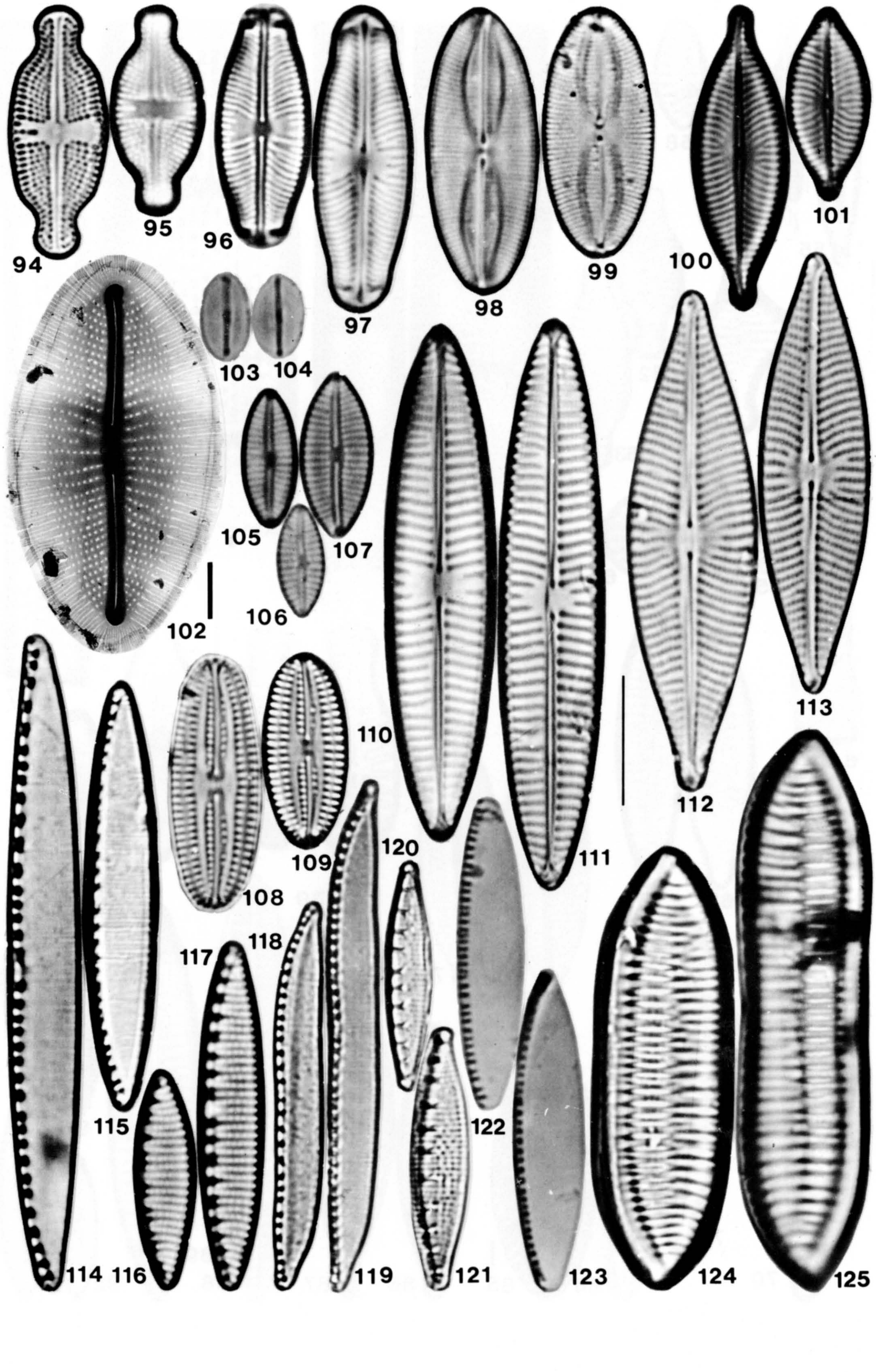


Plate 5. (lengends see p.124)

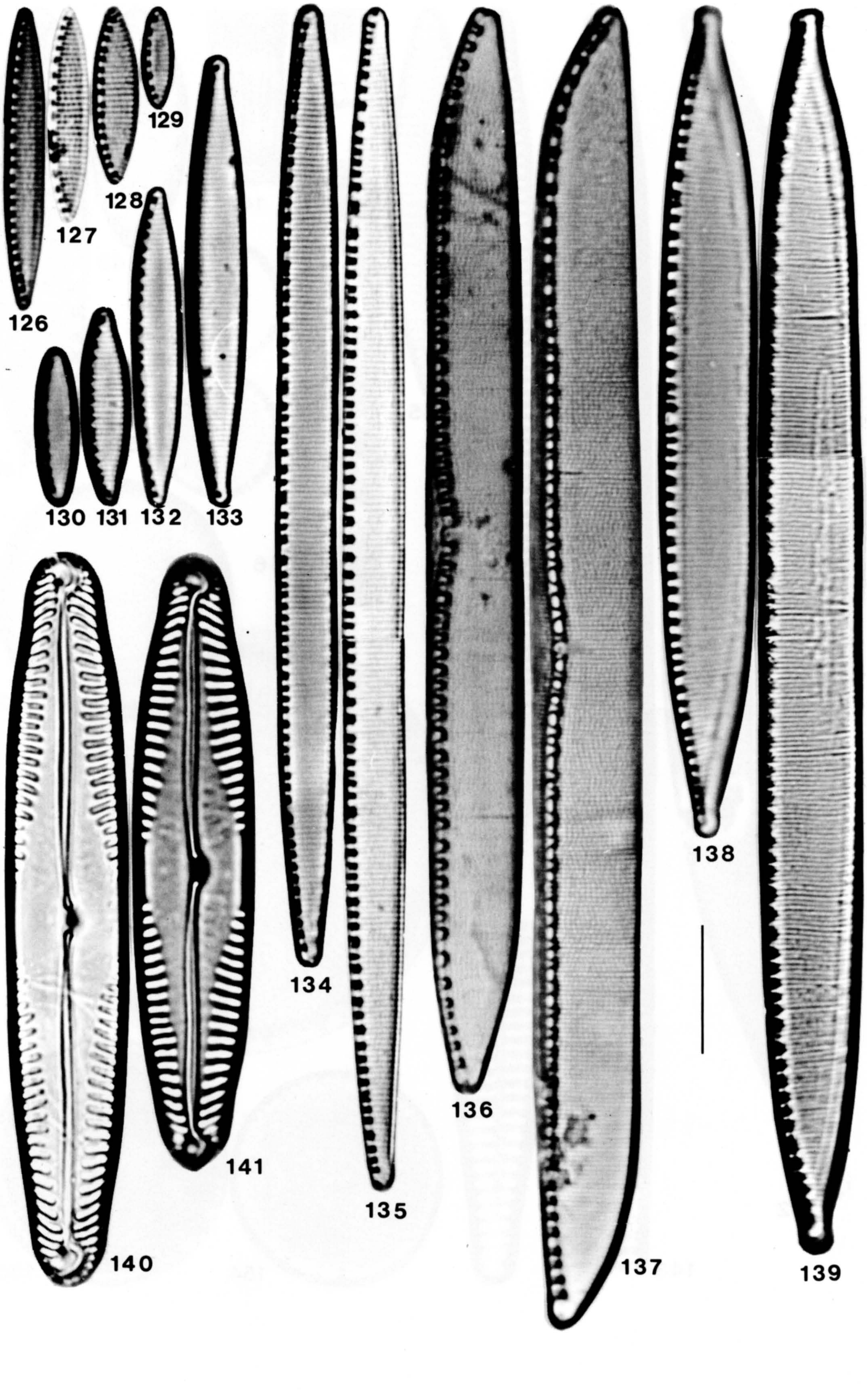
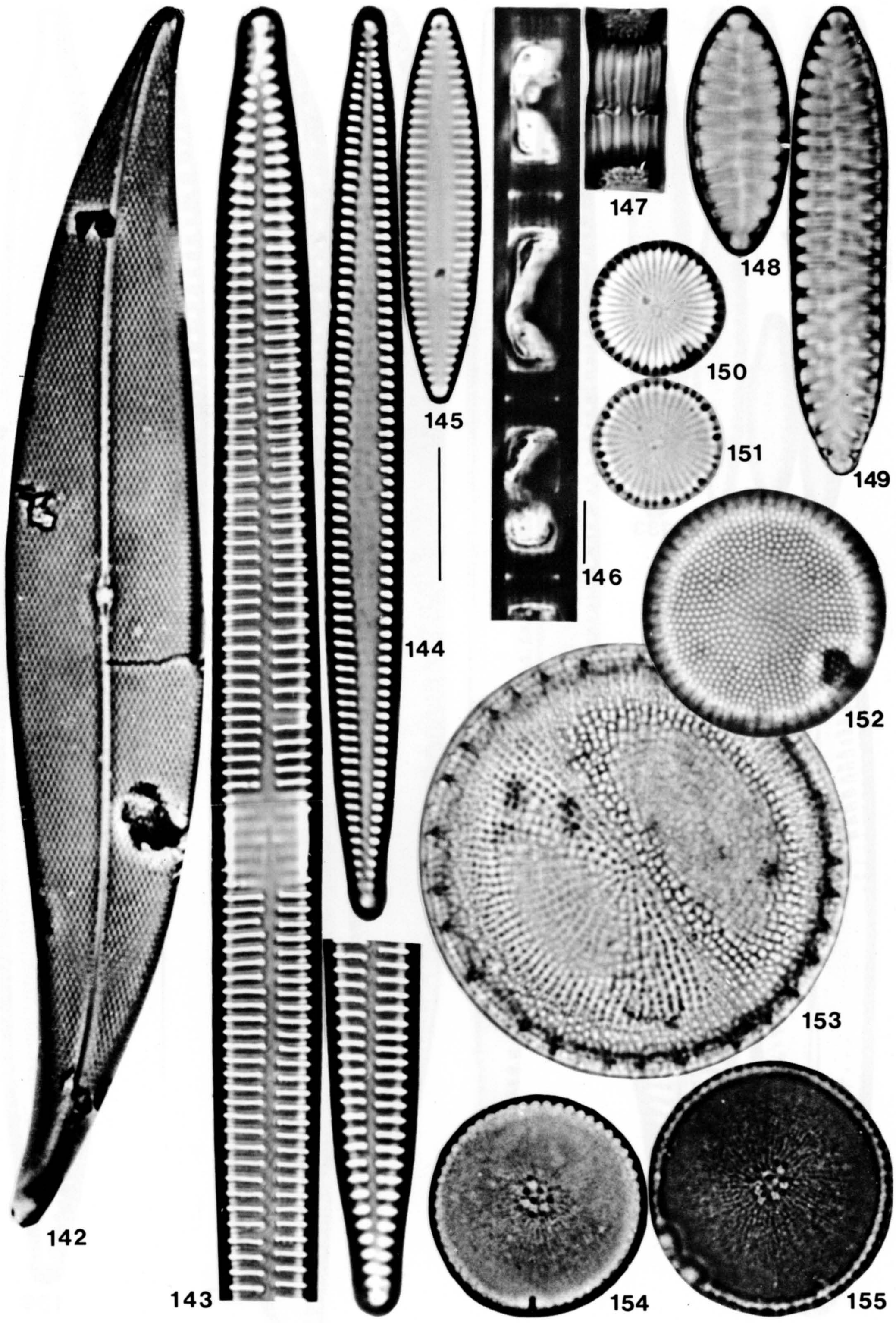


Plate 6. (lengends see p.124)



39. *N. saprophila* Lange-B. (Figs. 102-104).
40. *N. subminuscula* Manguin (= *N. frugalis* Hust.) (Figs. 105-107).
41. *N. tenera* Hust. (Figs. 108, 109).
42. *N. tripunctata* (O.P. Muell.) Bory (Figs. 110, 111).
43. *N. trivialis* Lange-B. (Figs. 112, 113).
44. *Nitzschia amphibia* Grun. (Figs. 116, 117).
45. *N. filiformis* (W. Smith) V. Heurck (Figs. 114, 115).
46. *N. frustulum* (Kuetz.) Grun. (Figs. 126-129).
47. *N. hantzschiana* Rabh. (Figs. 130-133).
48. *N. intermedia* Hantz. (Figs. 134, 135).
49. *N. nana* Grun. (Figs. 118, 119).
50. *N. pusilla* Grun. (Figs. 122, 123).
51. *N. scalpelliformis* (Grun.) Grun. (Figs. 136, 137).
52. *N. solgensis* Cleve-Euler (Figs. 120, 121). [= *N. sinuata* var. *delognei* (Grun.) Lange-B.].
53. *N. trybrionella* var. *subsalina* (O. Meara) Grun.
54. *N. umbonata* (Ehr.) Lange-B. (Figs. 138, 139).
55. *Pinnularia gibba* Ehr. (Figs. 140, 141).
56. *Pleurosigma salinarum* (Grun.) Grun. (Fig. 142).
57. *Skeletonema costatum* (Grev.) Cleve (Figs. 146, 147).
58. *Stephanodiscus invisitatus* Hohn & Hell. (Figs. 150, 151) [incl. *S. hantzschii* Grun. form. *tenuis* (Hust.) Håkansson's & Stoerm., *S. delicatus* Genkel, etc.].
59. *Surirella angusta* Kuetz. (Figs. 148, 149).
60. *Synedra fasciculata* (Ag.) Kuetz. (Figs. 144, 145) [or *Tabularia fasciculata* (Ag.) Williams & Round] [= *Synedra tabulata* (Ag.) Kuetz.].
61. *S. ulna* (Nitz.) Ehr. (Fig. 143).
62. *Thalassiosira lacustris* (Grun.) Hasle (Fig. 153).
63. *T. faurii* (Gasse) Hasle (Fig. 152).
64. *T. weissflogii* (Grun.) Fryxell & Hasle (Figs. 154, 155). [= *T. fluviatilis* Hust.].

Note: Give a saprobic value of 2.5 to the taxa of this group, but give 1.75 to the intermediate *Nitzschia hantzschiana,* and give 1 to *Nitzschia frusturum* when occurring in fresh waters and 2.5 when occurring in brackish waters.

Group C taxa: Any taxa found outside of Group A or Group B are included in this group. The list is omitted.

In the third step, the Pantle-Buck Formula for saprobic index (SI) was used (Pantle and Buck, 1955), though, no formula is used by Lange-Bertalot (1979) and another formula for the Diatom Assemblage Index (DAIpo) has been proposed by Watanabe *et al.* (1985, 1988). The use of a formula seems generally to have the advantage of getting results numerically and intuitively. Furthermore, it is easy to compare the values obtained from different places as well as different countries.

Watanabe's formula is complicated at first sight but the formula itself is basically the same as that of Pantle-Buck. One can easily convert DAIpo values to SI-values using the following equation when the relative frequency (%) is used for the occurrence rating of the Pantle-Buck Formula. The only difference between the two formulae is the maximum point which is 4 in the SI and 100 in the DAIpo.

$$SI = 4 - \frac{4}{100} DAIpo$$

Therefore, the use of the popular Pantle-Buck Formula [SI = sum total of occurrence rating(h) × saprobic zone rating(r) / sum total of occurrence rating] is recommendable. The saprobic zone rating(r) and the occurrence rating(h) are replaced with our group rating(g) and relative frequency (f%) and the SI values of our sampling stations were calculated.

$$SI = \frac{\Sigma f(\%)g \text{ (sum total of relative frequency} \times \text{group rating)}}{100\,[= \Sigma f\% \text{ (sum total of relative frequency)}]}$$

The SI values calculated are plotted against the BOD_5 values of the waters of the sampling

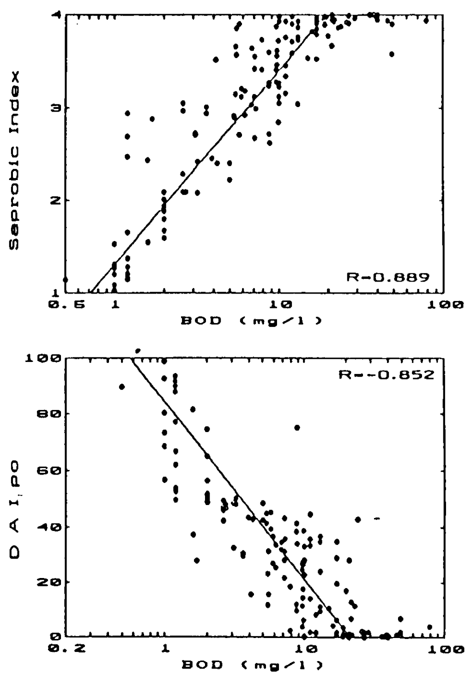


Fig. B. Correlation of SI and DAIpo with BOD_5.

stations (Fig. B). The SI values obtained correlated well with the BOD_5 values of each sampling station (R = 0.889). We found the same correlation to DAIpo values in our samples when we calculated using Watanabe's grouping method (statistical analysis). (Fig. 2, R = 0.852).

Correlations between the sum of the relative frequency of the Group A taxa in each sample and the BOD_5 values are shown in Fig. C (● = fresh water, ○ = brackish water). Taxa of the Group A occurred with high frequency on the high BOD_5 side Group B mainly occurred within the median BOD_5 range and Group C taxa occurred with high frequency on the low BOD_5 side respectively.

We suggest the use of the popular and simple Pantle-Buck Formula and our grouping A-C for the evaluation of water quality of the short, shallow, and fast flowing rivers like those of Japan.

PANEL DISCUSSION

K. Lee: How long are the rivers of Japan?

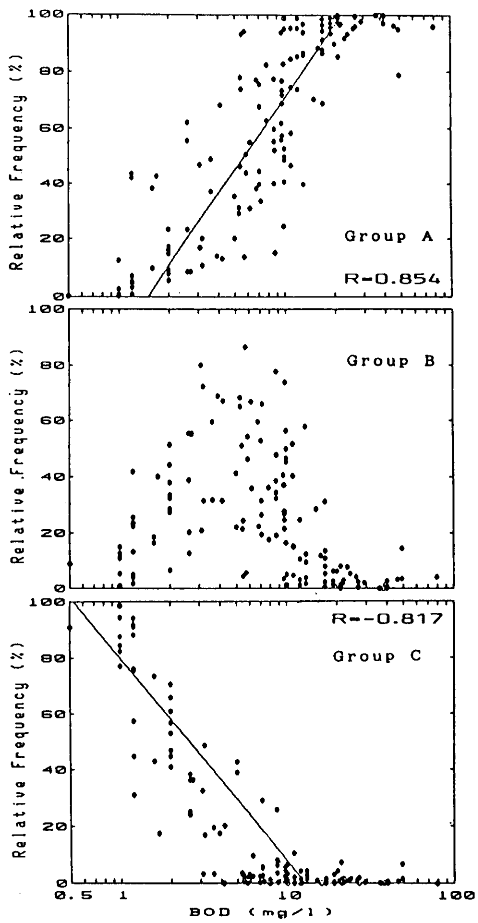


Fig. C. Correlations between sum of relative frequency of the Group A, B and C taxa in each sample and BOD_5.

Kobayasi Most are about 100 km, though there are a few rivers longer than 300 km.

K. Lee Are all species which you have handled attached or planktonic diatoms.

Kobayasi Almost all are attached forms but some species occurring in the estuaries are brackish planktons.

K. Lee Do you have any marker species as in Watanabe's paper?

Kobayasi We have no marker species because we selected the species according to their distribution patterns.

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REFERENCES

Descy, J.P. 1979. A new approach to water quality estimation using diatoms. *Nova Hedwigia, Beih.* **64:** 305-323.

Kobayasi, H. 1985. Ultrastructual differences in certain taxonomically difficult species of *Nitzschia* Section *Lanceolate* in Japan. pp.304-313. *In,* H. Hara (ed.). Origin and evolution of diversity in plants and plant communities. Academic Scientific Book, Tokyo. 396 pp.

Kobayasi, H. and H. Inoue, 1985. Fine structure and taxonomy of the small and tiny *Stephanodiscus* (Bacillariophyceae) species in Japan. 1. *Stephanodiscus invisitatus* Hohn & Hell. *Jap. J. Phycol.* **33:** 149-154. (in Japanese).

Kobayasi, H., H. Inoue and H. Kobayashi, 1985. Fine structure and taxonomy of the small and tiny *Stephanodiscus* (Bacillariopyceae) species in Japan. 2. *Stephanodiscus hantzschii* Grun. from. *tenuis* (Hust.) Håk. et Stoerm. *Jap. J. Phycol.* **33:** 233-238 (in Japanese).

Kobayasi, H. and H. Kobayashi, 1986. Fine structure and taxonomy of the small and tiny *Stephanodiscus* (Bacillariophyceae) species in Japan. 4. *Stephanodiscus costatilimbus* sp. nov. *Jap. J. Phycol.* **34:** 8-12.

Kobayasi, H. and H. Kobayashi, 1987. Fine structure and taxonomy of the small and tiny *Stephanodiscus* (Bacillariophyceae) species in Japan. 5. *S. delicatus* Genkel and the characters useful in identifying five small species. *Jap. J. Phycol.* **34:** 268-276.

Kobayasi, H., H. Kobayashi and M. Idei, 1985. Fine structure and taxonomy of the small and tiny *Stephanodiscus* (Bacillariopyceae) species in Japan. 3. Co-occurrence of *Stephanodiscus minutullus* (Kuetz.) Round and *S. parvus* Stoerm. & Håk. *Jap. J. Phycol.* **33:** 293-300.

Kobayasi, H. and S. Mayama, 1981. Comparative studies on the methods for water quality assessment using diatoms in severely polluted rivers. *Water and Waste* **23:** 1190-1198 (in Japanese).

Kobayasi, H. and S. Mayama, 1982. Most pollution-tolerant diatoms of severely polluted rivers in the vicinity of Tokyo. *Jap. J. Phycol.* **30:** 188-196.

Kobayasi, H., S. Mayama, K. Asai and S. Nakamura, 1985. Occurrence of diatoms collected from variously polluted rivers in Tokyo and its vicinity, with special reference to the correlation between relative frequency and BOD_5. *Bull. Tokyo Gakugei Univ. Sect. 4.* **37:** 21-46 (in Japanese).

Krammer, K. and H. Lange-Bertalot, 1986. Bacillariophyceae. 1. Teil: Naviculaceae. *In* H. Ettl, J. Gerloff, H. Heynig and D. Mollenhauer (eds.), Susswasserflora von Mitteleuropa. 2/1. Gustav Fischer, Stuttgart. pp.876.

Lange-Bertalot, H. 1979. Pollution tolerance of diatoms as a criterion for water quality estimation. *Nova Hedwigia, Beih.* **64:** 285-304.

Liebmann, H. 1962. Handbuch der Frischwasser- und Abwasser-biologie, Bd. 1. Gustav Fischer, Jena. pp.588. + 22pls.

Mayama, S. and H. Kobayasi, 1984. The separated distribution of the two varieties of *Achnanthes minutissima* Kuetz. according to the degree of river water pollution. *Jap. J. Limnol.* **45:** 304-312.

Mayama, S. and H. Kobayasi, 1988. Morphological variations in *Navicula atomus* (Kuetz.) Grun. pp.427-435. *In* F.E. Round (ed.), Proceedings of the Ninth International Diatom Symposium. Biopress, Bristol. pp.480.

Mayama, S. and H. Kobayasi, 1989. Sequential valve development in the monoraphid diatom *Achnanthes minutissima* var. *saprophila. Diatom Research* **4:** 111-117.

Pantle, R. and H. Buck, 1955. Die biologische Überwachung der Gewässer und die Darstellung der Ergebnisse. *Gasunt Wasserfach.* **96:** 604.

Patrick, R., H. Hohn an J.H. Wallace, 1954. A new method for diatom flora. *Not. Nat. Acad. Nat. Sci. Philadelphia* **259:** 1-12.

Schoeman, F.R. 1979. Diatoms as indicators of water quality in the upper Hennops River. *J. Limnol. Soc. sth. Afr.* **5:** 73-78.

Sládecěck, V. 1973. System of water quality from the biological point of view. *Arch Hydrobiol. Beih.* **7:** 1-218.

Watanabe, T., K. Asai and A. Houki, 1985. Epilithic diatom assemblage to organic water pollution (DAIpo) and its ecological significance. *Ann. Rep. of Graduate Division of Human Culture, Edited and Published by Doctoral Degree Program, Nara Women's Univ.* **1:** 76-94 (in Japanese).

Watanabe, T., K. Asai and A. Houki, 1988. Numerical water quality monitoring of organic pollution using diatom assemblages. 123-141. *In,* F.E. Round (ed.). Proceedings of the Ninth International Diatom Symposium. Biopress, Bristol. pp.480.

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