

COMPARISON OF ATTACHED DIATOM COMMUNITIES ON NATURAL AND ARTIFICIAL SUBSTRATES¹

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ABSTRACT

A study of attached diatom communities on artificial and natural substrates was conducted in Wheelwright Pond, New Hampshire, during 1975. There were differences in the species composition growing on artificial glass slides and natural substrates. The slides favored the accumulation of *Achnanthes minutissima* Kütz. and decreased the relative abundance of *Eunotia incisa* W. Sm. ex. Greg. and *Cocconeis placentula* v. *euglypta* (Ehr.) Cleve. Large growths of planktonic species were also noted on the slides. Compositional differences between slides positioned at 15-30 cm (upper) and 1 m (lower) from the surface of the water were minimal. A similar community composition of periphytic diatoms was found on five aquatic macrophytes.

Key index words: diatom; epiphytic algae; periphyton; species specificity; substrates, artificial; substrates, natural

Periphyton, the organisms that grow attached to macrophytes, logs, rocks and other submersed objects are important in aquatic systems. Species, primarily Bacillariophyceae (diatoms), often occur in large numbers producing slimy mats (19). Periphyton produces organic metabolites utilized by diverse

organisms in the food chain and account for 20-50% of the primary productivity of certain lakes (1,18).

Recently periphyton has been used as a tool in studying the environmental effects of heated and chemical effluents (2,10,14). Generally glass slides are favored as substrates for monitoring changes in diatom communities since the surface area and length of exposure time are easily obtained. However, it is not known if the glass slide periphyton are representative of the naturally occurring communities. Several researchers have investigated this question with conflicting results (3,4,6,16,17).

The purpose of this study is to investigate the question of species selectivity by natural and artificial substrates. Comparisons were made of attached diatom communities on glass slides and aquatic macrophytes. Since the periphytic communities were largely composed of diatoms, both in number and total cell volume, I concentrated my study on this group of algae.

MATERIALS AND METHODS

Throughout 1975 biweekly or monthly trips were made to Wheelwright Pond, Lee, New Hampshire, to collect *Potamogeton robbinsii* Oakes. Plants were harvested at depths of 0.5-1.0 m with a garden hoe and placed in buckets with lake water to minimize loss of periphyton. On 31 July four other macrophytes (*Pontederia cordata* L., *Nymphaea odorata* Ait., *Scirpus subterminalis* Torr., *Decodon verticillatus* [L.] Ell.) were

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clipped at the base, cut into sections and placed in plastic bags containing distilled water.

Diatom colonization on an artificial substrate (glass slides) was compared to that on natural substrates from March through June, 1975. To study diatom attachment on glass slides a rack was constructed consisting of a central aluminum pole ca. 2.5 m tall with wooden slabs (slide holders) at two levels attached to the central pole via small nylon rope and clips. The wooden slabs were thus suspended freely when submerged. The central pole was anchored in the sediments and suspended the slabs at 15–30 cm and 1 m from the water surface. Small styrofoam floats attached to each slab insured flotation. In each slab 20–25 slits were cut 1–2 cm deep which held slides vertically in the water. On each collection date when *P. robbinsii* plants were collected, all slides were removed to a slide box and transferred to the laboratory. The slabs were cleaned and new slides were placed into the slots. Slides were thus incubated between collection dates (2–4 wk).

Samples were processed in the following ways: a) From each of at least five randomly chosen *P. robbinsii* plants two leaves of ca. equal size were clipped at 0, 5, 10 and 15 cm intervals from the apex and placed in separate beakers, thus giving two equal sets of leaves from each interval. One set was used to obtain the total surface area of the leaves and the other for measuring diatom concentrations. The first set of leaves was photographed and the total leaf area determined gravimetrically. The second set of leaves was put through a cleaning process which oxidized the organic material (leaf substrate, diatom cell contents) yielding the diatom frustules (see following). b) Attached diatoms on the other macrophytes (sampled 31 July) were scraped or the epidermis peeled off, washed into a 100 ml beaker and oxidized. c) Periphyton on the glass slides was scraped off using a razor blade with 2–7 slides providing sufficient sample for observation.

All samples were oxidized by boiling in a mixture of distilled water, H_2SO_4 and potassium dichromate. After boiling, the sample was centrifuged 7 min at 2,500 rpm: the supernatant was removed with a vacuum decanter and microscopically examined for un sedimented frustules. The diatoms were resuspended and washed several times with distilled water and repeated centrifugation. After the last washing, the pellet was suspended in 8 ml distilled water and preserved with a few crystals of thymol to prevent fungal growth while stored (11).

A volume of 0.125–0.500 ml of the oxidized sample was pipetted onto a No. 1 coverslip and gently heated on a hot plate until dry, leaving the clean frustules on the coverslip. Each coverslip was inverted on a glass slide with a drop of Kleermount mounting medium (Carolina Biological Supply, Elton, North Carolina). A minimum of four permanent slides/sample was made and the slides with the most even distribution of frustules were chosen for quantitative analysis. These slides were scanned until 1,000 individual cells were identified and counted at $\times 1,000$ using a Wild M20 microscope.

Identification of the diatoms was according to Hustedt (9), Patrick and Reimer (11,12), Hansmann (7) and the Fritsch microfiche collection.

RESULTS

Throughout 1975 there was an increase in diatom concentration along the shoot of *Potamogeton robbinsii* from the apex to ca. 10 cm, but from 10 to 15 cm the concentration decreased slightly. The annual average concentration of diatoms on *P. robbinsii* leaves from the apex and 5, 10 and 15 cm below there were 1.9, 4.1, 6.1 and 4.7×10^5 cells·cm⁻², respectively. Spring and fall maxima were from April through June, and October through November,

while the summer minimum occurred between June and September.

A total of 133 taxa were found on *P. robbinsii* (quantitative results for each date available from the author). The three most abundant species were *Achnanthes minutissima* Kütz., *Cocconeis placentula* v. *euglypta* (Ehr.) Cleve and *Eunotia incisa* W. Sm. ex. Grég. *Anomoeoneis vitrea* (Grun.) Ross was occasionally abundant. The relative abundance of these taxa shifted as the leaves aged. Two species, *E. incisa* and *C. placentula* v. *euglypta*, had a parallel pattern of decreasing relative abundance along the shoot, whereas *Ac. minutissima* and *An. vitrea* had a parallel pattern of increasing relative abundance along the shoot.

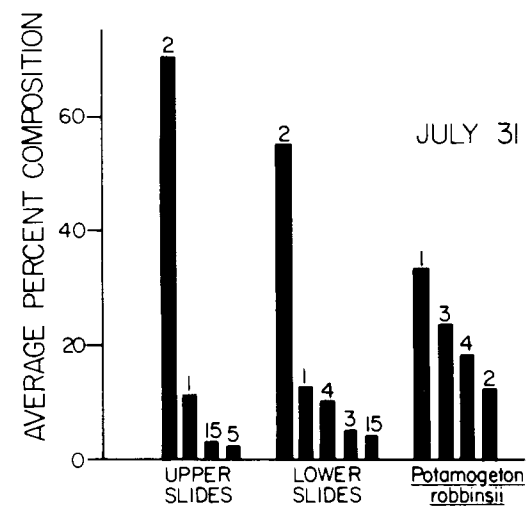
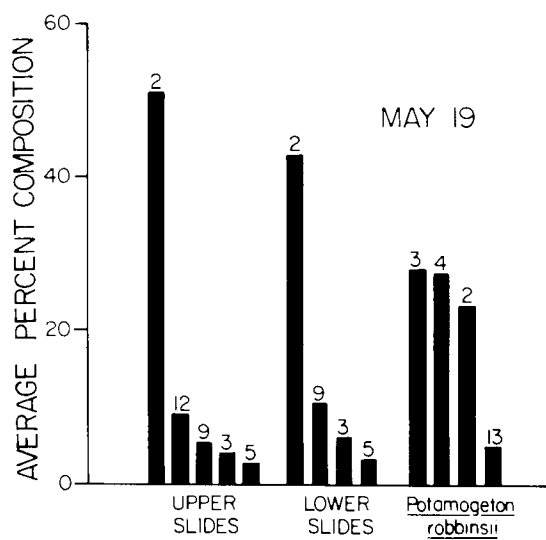
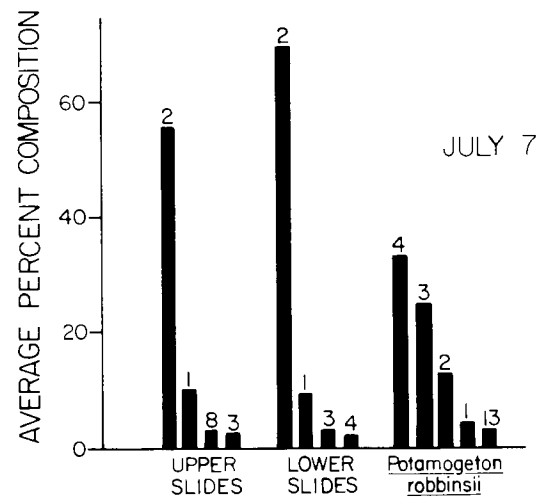
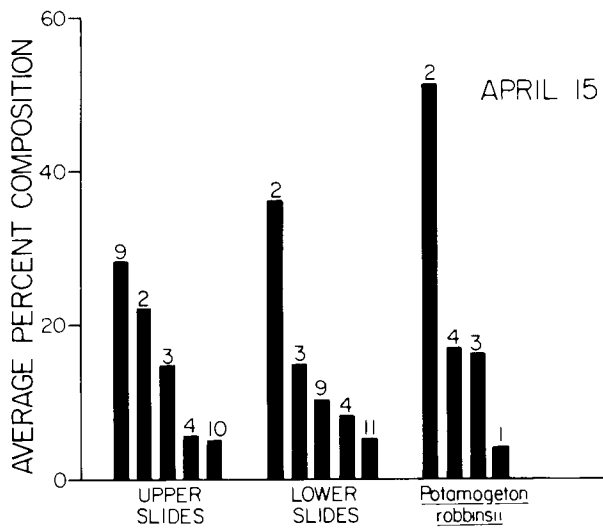
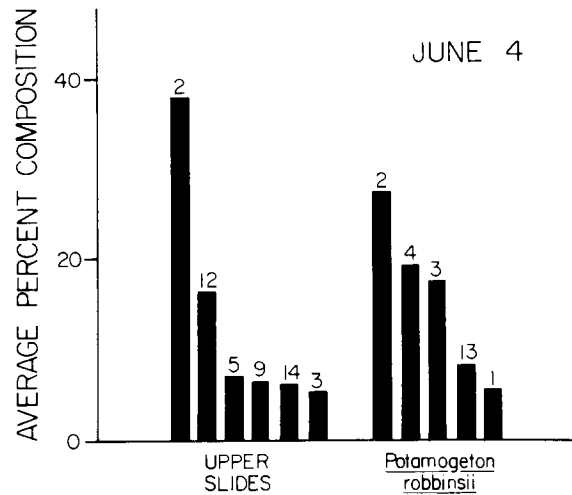
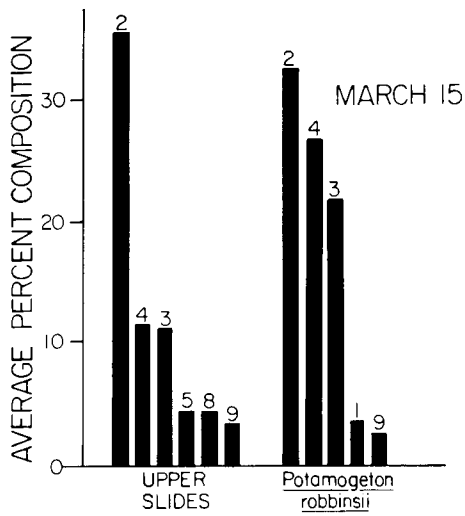
To compare the communities on *P. robbinsii* with those on the glass slides, the concentration of each taxon from each shoot segment was averaged. In general, *Ac. minutissima*, *C. placentula* v. *euglypta* and *E. incisa* composed 26, 24 and 22%, respectively of the total population on *P. robbinsii* from March through July; *An. vitrea* averaged 10% of the epiphytic populations.

Diatom colonization on glass slides and natural substrates was compared from March through July (Fig. 1). In March greater concentration occurred on the deeper slides but during April through July the reverse was true. The absolute concentrations of diatoms after 2–4 wk incubation periods were maximum at $2.5\text{--}5.0 \times 10^5$ cells·cm⁻² from April to early June. On all other dates concentrations were less than 1.0×10^5 cells·cm⁻².

In general, the relative abundance of the predominant taxon, *Ac. minutissima*, colonizing the glass slides was similar throughout the study period (Fig. 1). It comprised ca. 40% of the attached community from March through June and 50–70% during July. In contrast, it was never as dominant on *P. robbinsii*, except in April. During July, for example, *Ac. minutissima* averaged only 10% of the total epiphytic diatom population on *P. robbinsii*.

While *Ac. minutissima* grew extensively on glass slides, *E. incisa* and *C. placentula* v. *euglypta* were inhibited. These two comprised less than 10% of the glass slide communities and were never dominant. On *P. robbinsii* these diatoms generally accounted for over 40% of the total community and were codominants if not the dominant species. For example, on 19 May they comprised nearly 60% of the attached algae on *P. robbinsii* compared to less than 10% for both the upper and lower slides (Fig. 1–19 May).

At times, species which were present in the plankton became abundant on the slides but not the macrophytes. For example, on the glass slides the relative abundance of *E. flexuosa* Bréb. ex Kütz. was ca. 30% in April and *Asterionella formosa* Hass. was nearly 20% in June. Both taxa were present in large numbers in the plankton and were minor species



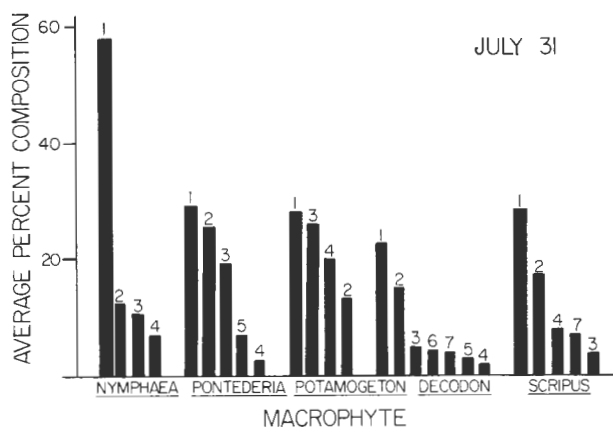


FIG. 2. Relative abundance of dominant diatoms on macrophytes, 31 July 1975: bar graph numbers as in FIG. 1.

in the *P. robbinsii* communities. Similar results were found with *Cyclotella kützingiana* Thw. and *Malomonas* sp. (nondiatom), both plankters found in the slide communities.

Only the lower slides of March and April resembled closely the communities on *P. robbinsii*. In both ice cover persisted on the pond and spring overturn had not begun. Compositional differences between the shallow and deep slides were minimal (Fig. 1–15 April, 19 May, 7 and 31 July).

On 31 July the attached diatom communities on *P. robbinsii*, *Pontederia cordata*, *Nymphaea odorata*, *Scirpus subterminalis* and *Decodon verticillatus* were compared to give an overview of the vertical and horizontal distribution patterns of naturally occurring communities. Relative abundance was averaged for all segments from each macrophyte (Fig. 2). The average composition of dominant diatoms was similar on each aquatic macrophyte. In order of abundance, the commonest species were: *Anomoeneis vitrea*, *Ac. minutissima*, *E. incisa* and *C. placentula* v. *euglypta*. In contrast, 3 wk old slide communities at this time were completely dominated by *Ac. minutissima* whereas *An. vitrea* accounted for less than 10% (Fig. 1–31 July).

DISCUSSION

Seasonal fluctuations in diatom densities occurred on *Potamogeton robbinsii* with spring, summer and

fall maxima noted. Such seasonal fluctuation is well-documented for the Bacillariophyceae (11). Most diatoms are reported to favor low light and temperature regimes and many investigators suggest that these factors are the triggering mechanisms for the increased growth (11,15). The number of attached diatoms were much lower on the slides than on the macrophytes due to the length of incubation in the pond. The slides were incubated 2–4 wk, whereas the macrophytes grew over a much greater time period, which allowed more epiphytic growth.

Little information on the substrate preferences of periphytic algae is available (5,13). However, several studies suggest that species specificity on natural substrates does occur. Prowse (13) suggests the following specificity: i) *Enhydrous* favors the growth of *Oedogonium*; ii) *Najas* favors the growth of *Eunotia*; and iii) *Utricularia* favors the growth of *Gomphonema*. Others have attempted to demonstrate species specificity on macrophytes from different ponds (5), but in such studies additional variables such as nutrient differences complicate the interpretation and yield inconclusive results. Careful analysis of species specificity should include selection of various substrates from essentially the same locality, since this would reduce many environmental differences in the final interpretation.

The community composition on all macrophytes was similar and essentially no species specificity existed. It is concluded that by sampling two or three of the aquatic macrophytes present in Wheelwright Pond the epiphytic communities can be adequately described.

Whether artificial substrates are colonized by the same species as natural substrates was studied by Castenholz (4) who found that communities on glass slides were similar to those on natural substrates. Hohn and Hellerman (8) also found that the communities growing on glass slides and styrofoam floats were the same when the water temperature was greater than 3°C. Stockner and Armstrong (16) found similar communities on glass slides and rocks except for a greater relative abundance of plankton on the slides.

Other investigators report differences in the diatom communities on glass slides and natural substrates. Tippett (17) concluded that the epiphytic diatom populations on several macrophytes differed

FIG. 1. Comparison of the relative abundance of dominant diatoms on glass slides and *Potamogeton robbinsii* from March–July 1975: upper slides refers to those positioned 15–30 cm from surface; lower slides refers to those at 1 m near the sediments and within *P. robbinsii* plants. On 15 March, lower slides lacked enough growth to sample; float from the lower slide holder became detached on 4 June samples. Number bar graph is: 1. *Anomoeneis vitrea*; 2. *Achnanthes minutissima*; 3. *Eunotia incisa*; 4. *Cocconeis placentula* v. *euglypta*; 5. *Achnanthes linearis*; 6. *Rhopalodiagibba*; 7. *Navicula falaisiensis*; 8. *Fragilaria capucina* v. *mesolepta*; 9. *Eunotia flexuosa*; 10. *Gomphonema angustatum*; 11. *Fragilaria* sp.; 12. *Asterionella formosa*; 13. *Tabellaria fenestrata*; 14. *Cyclotella kützingiana*; 15. *Gomphonema intricatum*.

from those on glass slides. Further he stated, "at its best the method can only be comparative, showing relative changes in the flora." Foerster and Schlichting (6) found that many algae growing on macrophytes were not present on slides and Brown (3) also found compositional differences in attached algal communities on glass slides and the aquatic form of *Eleocharis baldwinii* (Torr.) Chapman.

Results from the present study also indicate that the communities on the glass slides are different from naturally occurring communities on macrophytes. The slides accumulated large numbers of plankton into their periphyton. For example, *Asterionella formosa* dominated the plankton in May and June and although it formed large concentrations on the slide communities during May and June, it was not found on *P. robbinsii*.

It could be argued that the attached diatoms on the slides resemble the communities on very young *P. robbinsii* leaves since both have similar incubation periods and are smooth. This is not so since both *E. incisa* and *C. placentula* v. *euglypta* were few in number on the slide substrates yet have their highest relative abundance on *P. robbinsii* leaves at the apex. When young *P. robbinsii* leaves were examined directly diatom colonization was found only in areas of decay. This suggests nutrient release by the macrophyte may effect both epiphyte attachment and composition.

It could also be argued that only a very small species such as *Ac. minutissima* could be carried by water movement to the slides. This is not the case since the lower slides were placed in contact with the bed of *P. robbinsii* plants. Also, *E. incisa* and *C. placentula* v. *euglypta* were present in large concentrations on the March slides when the pond was still frozen and the water movements were minimal. Hence, the heavier diatoms were probable "seed" sources for the attached communities on the slides.

Thus, because glass slides in Wheelwright Pond have higher densities of some diatoms (whereas other species are absent) and accumulate plankton species, they do not reflect the composition of naturally occurring epiphytic communities. Knowing whether communities on artificial substrates represent the naturally occurring populations may be important in environmental monitoring projects which employ slide communities to predict changes in water quality. Therefore, it would be important for the investigator to check the natural diatom populations frequently.

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- Allen, H. L. 1971. Primary productivity, chemo-organography, and nutritional interactions of epiphytic algae and bacteria on macrophytes in the littoral zone of a lake. *Ecol. Monogr.* 40:97-127.
- Baker, A. L. 1974. Annual periphyton report. Northern States Power Co. St. Paul, Minnesota, 1974 Annual Report, 201-31.
- Brown, H. D. 1976. A comparison of the attached algal communities of a natural and an artificial substrate. *J. Phycol.* 12:301-6.
- Castenholz, R. W. 1961. An evaluation of a submerged glass method of estimating production of attached algae. *Int. Ver. Theor. Angew. Limnol. Verh.* 14:155-9.
- Colt, L. C., Jr. & Hellquist, C. B. 1974. The role of some Haloragaceae in algal ecology. *Rhodora* 74:446-56.
- Foerster, J. W. & Schlichting, H. E. 1965. Phycoperiphyton in an oligotrophic lake. *Trans. Am. Microsc. Soc.* 84:485-502.
- Hansmann, E. W. 1973. The diatoms of the streams of eastern Connecticut. *Conn. State Geol. Nat. Hist. Surv. Bull.* 106:1-119.
- Hohn, M. & Hellerman, J. 1963. The taxonomy and structure of diatom populations from three eastern North American rivers using three sampling methods. *Trans. Am. Microsc. Soc.* 82:250-329.
- Hustedt, F. 1930. Bacillariophyta. In Pascher, A. [Ed.] *Die Süßwasser-Flora Mitteleuropas* Vol. 10, Gustav Fischer, Jena. 466 pp.
- Lowe, R. L. 1974. Environmental requirements and pollution tolerance of freshwater diatoms. U.S. Environmental Protection Agency-670/4-74-005. 333 pp.
- Patrick, R. & Reimer, C. W. 1966. *The Diatoms of the United States*. Vol. 1. Academy of Natural Sciences, Philadelphia. Monograph No. 13. 688 pp.
- 1975. *The Diatoms of the United States*. Vol. 2, Part 1. Academy of Natural Sciences, Philadelphia Monograph No. 13(2). 213 pp.
- Prowse, G. A. 1959. Relationships between algal species and their macrophyte hosts. *Nature London* 186:1204-5.
- Raschke, R. L. & Weber, C. 1970. Use of a floating periphyton sampler for water pollution surveillance. U.S. Federal Water Pollution Control Administration, Division of Water Quality Research, Cincinnati. Report No. 20:1-20.
- Smith, G. M. 1950. *The Fresh-water Algae of the United States*. 2nd ed. McGraw-Hill Book Co., New York. 719 pp.
- Stockner, J. G. & Armstrong, F. A. J. 1971. Periphyton of the experimental lakes area, Northwestern Ontario. *J. Fish. Res. Board Can.* 28:215-29.
- Tippett, R. 1970. Artificial surfaces as a method of studying populations of benthic algae in fresh water. *Br. Phycol. J.* 5:187-99.
- Wetzel, R. G. 1964. A comparative study of the primary production of higher aquatic plants, periphyton, and phytoplankton in a large, shallow lake. *Int. Rev. Ges. Hydrobiol. Hydrogr.* 49:1-61.
- Young, O. W. 1945. A limnological investigation of periphyton in Douglas Lake, Michigan. *Trans. Am. Microsc. Soc.* 64:1-20.