

## MICROATTACHMENT PATTERNS OF DIATOMS ON LEAVES OF *POTAMOGETON ROBBINSII* OAKES<sup>1</sup>

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SIVER, P. A. 1980. Microattachment patterns of diatoms on leaves of *Potamogeton robbinsii* Oakes. *Trans. Amer. Micros. Soc.*, 99: 217-220. A unique microzonation pattern on young leaves of aquarium-grown *Potamogeton robbinsii* Oakes was documented. Over 80% of the diatoms colonized the margins of the leaves. Several mechanical and physiological mechanisms are suggested. Diatoms colonized the midrib portions of the leaves only after cytoplasmic decay was initiated. *Cocconeis placentula* v. *euglypta* and *Eunotia incisa*, both with superior attachment mechanisms, were the only epiphytes.

Only recently have microdistributional patterns of epiphytes on macrophyte hosts been observed. Cattaneo (1978) found that diatoms epiphytic on *Potamogeton richardsonii* preferred the edges. Since the same pattern was observed on artificial plants, he concluded that the preference for the edge was a physical and not a biological effect.

Other questions concerning the microattachment patterns of epiphytes remain unanswered. Is attachment of epiphytes random, or does it follow a pattern determined by physical, chemical, and/or biological factors? Do epiphytes cause the epidermal cells of their macrophyte hosts to decay, or do they colonize the host only after cell decay has begun? This paper discusses the microattachment patterns on leaves of *Potamogeton robbinsii* Oakes and lends support for Cattaneo's findings (1978). In addition, the effect of the epiphytes on macrophyte cellular decay is discussed.

### MATERIALS AND METHODS

*Potamogeton robbinsii* plants were taken from Wheelwright Pond, Lee, New Hampshire in April 1975 and placed in an aquarium in the laboratory with lake water and sediments. The aquarium was aerated and the water was circulated gently. The plants were tagged and only leaves formed while the plants were in the aquarium were used. The plants were incubated at 20°C for 8-10 weeks, harvested, and examined directly to determine the specific attachment pattern on the leaves. Leaves from the apex, and 5, 10, 15, and 20 cm from the apex were clipped from 10 plants and observed at 400× magnification. The attached diatoms were identified, counted, and mapped along transverse scans at the tip, middle, and basal portions of the leaves. Cellular decay was defined as any epidermal leaf cell that had turned brown in color.

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*P. robbinsii* is a loosely rooted submerged pondweed that grows horizontally on the sediments. Thus, leaves on individual plants have approximately the same light and temperature regimes. Differences in light quantity for the adaxial and abaxial leaf surfaces also were minimal due to the leaves' vertical orientation.

#### RESULTS AND DISCUSSION

A unique microzonation pattern of attached diatoms on the aquarium-grown *P. robbinsii* leaves was observed. The microattachment pattern consisted of a marked "edge effect" where over 80% of the diatoms colonized the outer margins of the leaves. Distributional patterns were similar for transects at the tip, middle, and basal portions of the leaves (Figs. 1-3). Diatoms were rarely observed in the midrib portions of the leaves. The "edge effect" was present on both the abaxial and adaxial sides, and was very prominent on the leaves from 0 to 15 cm. At 20 cm from the apex, the leaves had started to decay and the "edge effect" became less distinct.

Cytoplasmic decay of epidermal cells first occurred at the margins and secondarily in the midrib areas. Epidermal cell decay in the midrib portions of the leaves preceded colonization of large numbers of diatoms. Hence, decay could not have been caused by the epiphytes. The average concentration of diatoms increased as the leaves aged; the highest concentrations of diatoms were present on leaves located 20 cm from the stem apex.

The initially incubated plants had over 50 attached diatoms with *Eunotia incisa* W. Sm. ex Greg. and *Cocconeis placentula* v. *euglypta* (Ehv.) Cleve. as co-dominants (Siver, 1978). By contrast, after the 8-10 week incubation period, only the co-dominants *E. incisa* and *C. placentula* v. *euglypta* were present on the aquarium-grown plants.

There are several possible explanations for the "edge effect." First, the marginal leaf cells are chemically and anatomically distinguishable from the midrib cells. It was observed that the marginal cells contained fewer chloroplasts, had thicker cell walls, and oxidized in a potassium dichromate and  $H_2SO_4$  solution at a slower rate than the midrib cells. When entire leaves were oxidized in potassium dichromate and  $H_2SO_4$ , the marginal cells were the most resistant and required a longer period to complete oxidation. Perhaps the marginal cells produced a different substance (a lipid), or one in larger amounts than the midrib epidermal cells, and perhaps this substance provided a better attachment surface for diatoms. By contrast, midrib cells with their thinner cell walls do not provide attachable surfaces for diatoms. Marginal cells of *P. robbinsii* leaves have single-celled trichomes (extensions of the epidermal cells) (Fig. 4). The trichomes assist in capturing both mucilage and silt, which also create a better surface for attachment by diatoms.

A second possible explanation for the "edge effect" is that there is more nutrient exchange between the leaves and the epiphytes in the marginal regions, making the interface more conducive to colonization. However, since thicker cell walls on marginal cells probably slow down nutrient exchange, this is not as probable as an explanation for the "edge effect" as the attachment hypothesis noted above. Further, Cattaneo (1978) suggested that diatom preference for the edge was a physical (and not a biological) phenomenon because it occurred on both natural and artificial leaves. Cattaneo further hypothesized that the leaf margins may favor "nutrient renewal."

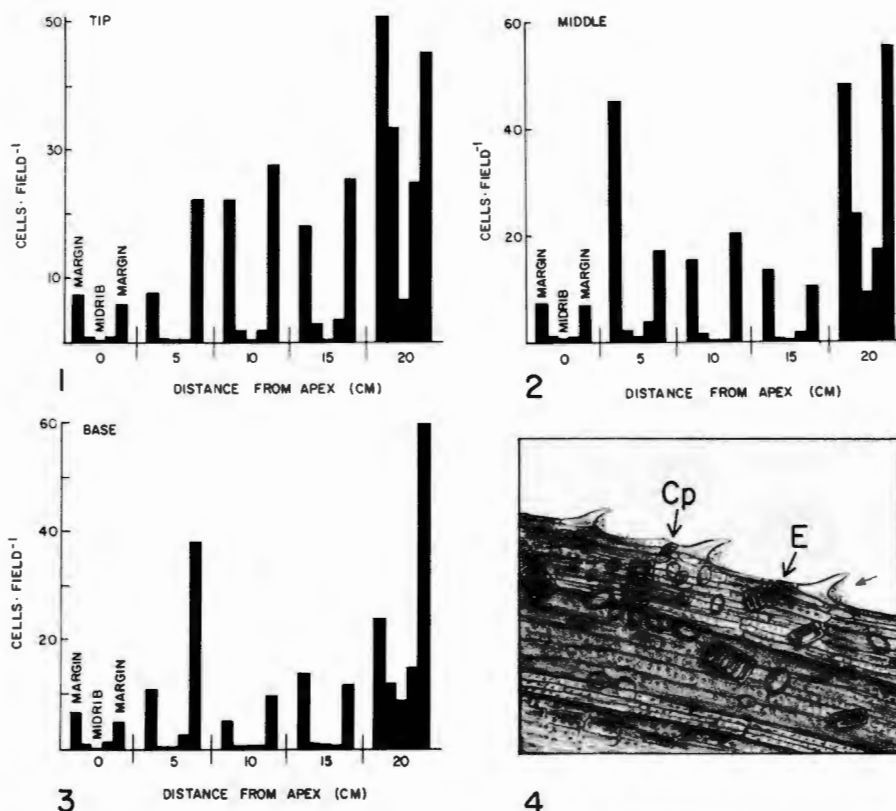


FIG. 1. Concentrations of diatoms along transverse sections at the tip of *Potamogeton robbinsii* leaves from 0 to 20 cm along the shoot (plants incubated 8 weeks in an aquarium). Leaf age increases with distance from apex. FIG. 2. Concentrations of diatoms along transverse sections at the middle of *Potamogeton robbinsii* leaves from 0 to 20 cm along the shoot (plants were incubated 8 weeks in an aquarium). FIG. 3. Concentrations of diatoms along transverse sections at the base of *Potamogeton robbinsii* leaves from 0 to 20 cm along the shoot (plants were incubated 8 weeks in an aquarium). FIG. 4. The "edge effect" on a *Potamogeton robbinsii* leaf. Note solitary *Cocconeis placentula* v. *euglypta* (Cp) cells, filaments of *Eunotia incisa* (E), and trichomes (arrow).

A third possibility is that the midrib cells may secrete a substance that inhibits colonization of epiphytes. Sieburth & Conover (1965) found that *Zostera* released a tannic substance which retarded fouling of the plant by epiphytes. A combination of factors most likely established the "edge effect."

The "edge effect" was not as apparent at 20 cm from the apex because of an increase of epiphytes in areas of decay inward from the margins. Since breakdown of the leaves occurred in these areas prior to colonization by diatoms, it cannot be argued that the physical presence of diatoms was the cause of leaf decay. Rather, leaf decay appeared to promote diatom colonization. The mechanism of promotion may be the release of nutrients from the decaying leaf cells, a reduced production of toxins (e.g., tannins), or a more conducive surface for diatom attachment. The aquarium-grown plants had fewer epiphytes than those in the pond, thus enabling the initial attachment

patterns to be revealed. Similar patterns probably occur on the macrophytes in nature.

There are several possible explanations why *C. placentula* v. *euglypta* and *E. incisa* were the only taxa found on the plants incubated in the aquarium. I originally thought that *C. placentula* v. *euglypta* and *E. incisa* were the only species that survived the sudden switch from the pond (7°C) to the aquarium (20°C). This was not the case because many of the diatom species occurring naturally on the plants (Siver, 1978) were found inhabiting the mud and plankton of the aquarium. Thus, the additional species undoubtedly were present as "seed" populations for colonization on the leaves. Apparently, only *C. placentula* v. *euglypta* and *E. incisa* can attach and remain on the new leaves under these conditions.

Superior attachment mechanisms for *C. placentula* v. *euglypta* and *E. incisa* are evident. *C. placentula* v. *euglypta* has a broad, flat, curved morphology and secretes a layer of mucilage (Smith, 1950; Stockner & Evans, 1972). *E. incisa* secretes large quantities of mucilage, has a curved morphology, and can form filaments, all of which enable it to adhere to fresh substrates. Further evidence regarding superior attachment mechanisms of these two diatoms has been documented (Siver, 1978).

Although laboratory phenomena may not fully represent nature, it is of interest to note that the two diatom species found in this study are identical to those reported by Cattaneo (1978). He found *C. placentula* and *E. incisa* as the only diatoms colonizing leaves of naturally occurring plants. Perhaps these two diatoms are important as initial colonizers in other aquatic systems.

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