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Abiotic and biotic regulation of periphyton in recovering acidified lakes

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Abstract. Anthropogenic acidification of lakes alters periphyton growth and taxonomic composition. I tested whether these changes were regulated by abiotic (i.e., resources, acidity) and biotic (i.e., herbivory, competition) conditions, or both. Periphyton was reciprocally transplanted across lakes of low acidity (pH~6.3), moderate acidity (pH~5.6), and high acidity (pH < 5) undergoing recovery from anthropogenic acidification. Transplants were also placed in exclosures that excluded either macrograzers or all grazers and potential algal competitors. Data from 4-wk incubations suggested that periphyton growth and taxonomic composition were primarily regulated by abiotic conditions. Total biomass was significantly lower for periphyton that originated from, or was transferred into, the most acidic lake. Canonical correspondence analysis showed abiotic regulation of periphyton in acidified lakes was best explained by dissolved inorganic carbon availability. Regulation by biotic factors was detectable only in the least acidic lake where macrograzers suppressed loosely attached taxa, but not total biomass, owing to compensatory species replacements. When periphyton was transferred into less acidic lakes, only taxa that were already native to the host lake persisted through the 4-wk incubation, suggesting that algal recolonization during recovery is not impeded by dispersal barriers. My findings suggest that abiotic regulation of epilithic periphyton is superseded by biotic control as acid-sensitive macrograzers recolonize recovering acidified lakes.

Key words: periphyton, acidified lakes, recovery, abiotic regulation, dissolved inorganic carbon, biotic regulation, grazers, species exclusion.

Recent improvements in the water quality of many acid-stressed lakes in North America have prompted the search for evidence of concomitant biological recovery (Gunn and Keller 1990, Keller et al. 1992a, 1992b, Nicholls et al. 1992, Locke et al. 1994). Biological recovery is expected to commence at the microbial level, owing to fast growth rates and a hypothesized lack of dispersal barriers (Schindler 1987, Yan and Welbourn 1990). Unfortunately, the roles of abiotic and biotic factors in the recovery process are not well understood.

Anthropogenic acidification of lakes alters the growth of periphyton. Although filamentous green algae consistently replace diatoms and cyanobacteria, changes in total periphyton production are less certain (see review in Stokes 1986). Abiotic control of primary production under acidic conditions involves reduced availability of dissolved inorganic carbon (DIC) (Turner et al. 1987, 1991, 1994, Fairchild and Sherman 1992, 1993), and increased acidity (Müller 1980) and aluminum toxicity (Nalewajko and Paul 1985). Alternatively, comparative studies suggest that the proliferation of filamentous green algae as periphyton and unattached benthic clouds of metaphyton in the littoral zone is due to reduced densities of benthic herbivores in acidified lakes (Howell et al. 1990, Jackson et al. 1990, France et al. 1991, Appelberg et al. 1993). To date, the relative importance of abiotic (resources, acidity) and biotic (herbivory, competitive exclusion) factors in regulating periphyton in acidified lakes has not been experimentally tested.

My objective was to determine the importance of abiotic and biotic regulation of periphyton in lakes of different acidities that are currently undergoing recovery from anthropogenic acidification. I conducted a survey of these lakes to relate patterns in periphyton assemblages to environmental variables using canonical correspondence analysis (CCA). I then performed a reciprocal transplant experiment using exclosures to assess the relative importance of abiotic and biotic controls of periphyton biomass and taxonomic composition in 3 lakes of differing acidity. Relative importance was defined as the proportion of total assemblage variance explained by a particular process (sensu Menge and Sutherland 1987).

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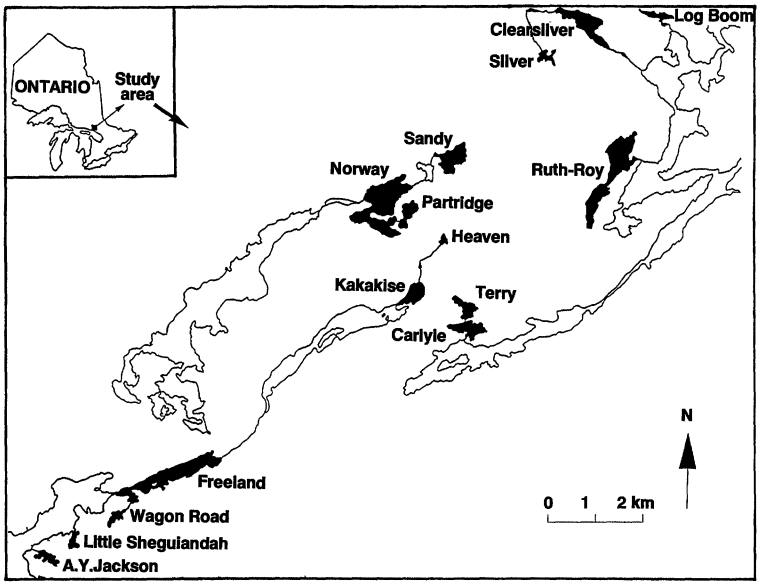

FIG. 1. Study area showing locations of the 15 survey lakes (black shading) of Killarney Provincial Park. Sampling of periphyton was confined to shallow littoral areas within each lake.

Methods

Study area

All 15 survey lakes were located in Killarney Provincial Park (46°3'N, 81°21'W), a wilderness park ~50 km southwest of Sudbury, Ontario (Fig. 1). The lakes lie among predominantly quartzite and granitic ridges of the LaCloche Mountains (Debicki 1982) in catchments consisting of northern mixed forest on podzolic soils. Lake selection was based on data from earlier surveys (Sprules 1975, Bleiwas et al. 1984), and included only lakes of low alkalinity (Table 1). The acidification history of these lakes has been extensively documented, and shows that they are experiencing both chemical and biological recovery (A. S. Dixit et al. 1992, S. S. Dixit et al. 1992, Locke et al. 1994).

Lake water chemistry

Chemical analyses were performed on water samples obtained monthly from the main outflow of each lake between May and September 1992. I chose to sample at the outflow to obtain an integrated sample of epilimnetic water chemistry for the shallow littoral zone of each lake. Alkalinity, pH, and conductivity of unaerated samples were determined in the field immediately upon collection. Alkalinity was expressed as acid neutralizing capacity (ANC), using the Gran titration procedure (Wetzel and Likens 1991). Water samples for metal and nutrient analyses were filtered using a 60-mL polypropylene syringe equipped with an acetyl resin filter holder (Gelman Sci.) and a glass-microfiber filter (Whatman GF/F). Filtrate was collected in acid-washed 125-mL polyethylene bottles that were first rinsed with filtrate. Samples were stored in ice until chemical analysis. Aluminum and phosphorus concentrations were determined using inductively coupled plasma mass spectrometry, and other cations were detected with mass atomic absorption spectrophotometry (Ontario Ministry of the Environment 1981).

Dissolved inorganic carbon (DIC) and organic carbon (DOC) were measured by total carbon infrared gas analysis (Mulholland et al. 1986, Kaplan 1992). Duplicate water samples for DIC and DOC analyses were drawn from each lake through Teflon tubing into a 60-mL polypropylene syringe. Filtered (Whatman GF/F) lake water was stored in 40-mL amber borosilicate vials for DOC analysis, whereas unfiltered water was used for DIC analysis. Samples were stored in darkness on ice and analyzed within 1 mo of collection. Total inorganic carbon was determined by infrared absorption of CO_2 gas

TABLE 1. Water chemistry for the 15 Killarney survey lakes based on sampling of lake outflows. Lakes are arranged in order of increasing DIC. DIC = dissolved inorganic carbon, DOC = dissolved organic carbon, ANC = acid-neutralizing capacity, Al = aluminum, Ca = calcium, and P = phosphorus. Lake codes are those used in the ordination plots.

Lake	DIC (mg/L)	DOC (mg/L)	pH	ANC (μeq/L)	Al (μg/L)	Ca (mg/L)	P (μg/L)
Ruth-Roy (RR)*	0.07	0.9	4.2	−52	332	0.71	7.8
Sandy (SA)	0.08	1.1	4.7	−19	178	1.42	8.2
Norway (NO)	0.09	0.9	4.5	−39	207	1.39	7.3
Clearsilver (CS)	0.13	1.7	4.3	−43	314	0.82	7.3
Silver (SI)	0.14	1.1	4.1	−48	292	0.56	6.9
Heaven (HE)	0.18	8.6	3.9	−99	299	0.34	8.2
Freeland (FE)	0.25	2.9	4.8	−24	123	2.01	7.2
A.Y. Jackson (AY)*	0.27	3.8	5.6	2	108	1.97	9.5
Terry (TE)	0.29	6.6	4.7	−12	207	1.44	7.6
Log Boom (LB)	0.32	3.4	5.1	−9	114	2.35	7.8
Partridge (PA)	0.32	2.7	4.9	−17	116	2.68	8.7
Carlyle (CA)	0.52	5.2	5.1	1	125	2.16	7.7
Kakakise (KA)	0.64	2.7	6.1	26	74	2.05	8.2
Wagon Road (WR)	0.99	10.2	5.4	31	106	1.49	8.3
Little Sheguiandah (LS)*	1.04	5.4	6.3	75	77	1.86	8.2

* Lakes used in the reciprocal transplant experiment

released by acidification of the sample. Total organic carbon was measured as the amount of CO_2 released by oxidation using potassium persulfate and heating of the sample to 100°C.

Periphyton survey

In each lake, I located epilithic periphyton habitats consisting of submerged bedrock ledges at ~0.5 m depth in sheltered bays to serve as sites of deployment for introduced substrata. Ten unglazed quartzite tiles (100 cm^2) were separately deployed across such sites in each lake during early May 1992. These tiles were chosen to closely resemble the surfaces of the bedrock in the study area, thereby allowing for periphyton development that best compared with epilithon. Tiles were vertically inclined (~80°) and against the bedrock so as to minimize accumulations of phytoplankton and detritus. Thus, tile habitats were standardized for nonerosional, moderate levels of water movement and low rates of deposition and nutrient recycling (see Turner et al. 1994). Tiles were retrieved during mid September by placing a plastic container (10 × 10 cm) directly over each tile to minimize disturbance of loosely attached taxa. Periphyton was easily removed using a glass scraper and hard-bristle toothbrush, rinsed with distilled water, and preserved with Lugol's solution.

Reciprocal transplant experiment

Periphyton was reciprocally transplanted across Little Sheguiandah Lake, A.Y. Jackson Lake, and Ruth-Roy Lake. These lakes were selected because each represented a different chemical and biological state of acid-stress recovery. For example, the abundance of benthic macrograzers differed across the 3 lakes (Table 2). Macrograzer abundances were visually estimated along 10-m transects that each ran parallel to the shoreline and over a site where the tiles had been deployed. Both mid-day and evening surveys were each conducted once at each site for 30 min from a canoe in August 1992. The highest abundances of observed benthic macrograzers, including green frog tadpoles (*Rana clamitans*), cyprinids, pumpkinseeds (*Lepomis gibbosus*), and brown bullhead (*Ictalurus nebulosus*) were found in Little Sheguiandah Lake, the least acidic lake. The moderately acidic A.Y. Jackson Lake supported fewer cyprinids, pumpkinseeds, and tadpoles. Zoobenthos in the highly acidic Ruth-Roy Lake contained few macrograzers, which consisted primarily of chironimids, odonates, and crayfish. Crayfish (*Cam-*

TABLE 2. Abundance of observed benthic macrograzers in the 3 experimental Killarney lakes during August 1992. Results are means (±1 SE) from pooled mid-day and evening surveys that were conducted along 3 transects within each lake.

Lake	Crayfish	Cyprinids	Pumpkinseeds	Tadpoles
Ruth-Roy	5.1 (1.3)	0	0	0
A.Y. Jackson	3.2 (0.8)	2.1 (0.7)	3.2 (0.9)	5.1 (0.6)
Little Sheguiandah	4.8 (0.9)	3.8 (1.2)	2.8 (0.7)	25.0 (6.3)

barus robustus, *C. bartoni*) densities were comparable in all lakes.

A 3-factor experiment was conducted to determine the effects of origin, lake acidity, and biotic conditions on periphyton biomass accrual and species composition over a 1-mo incubation period. The 27 treatment combinations (3 origins × 3 lake acidities × 3 exclosures) were randomly assigned and replicated 3 times for a total of 81 tiles. Blocks of replicates were achieved by reciprocally transplanting across 3 correspondingly numbered habitats within each lake. All tiles were deployed at near-vertical inclinations along rock ledges that were submerged at a depth of ~0.5 m.

Periphyton origins were established by deploying sets of 9 individual tiles in 3 different sites within each of the lakes during mid May 1992. Periphyton origin allowed me to look for persistent differences between assemblages that had developed under different acidities (low, moderate, high). A significant origin effect would suggest that mid-summer periphyton growth is influenced by conditions that affect its development in early summer.

The lake-acidity treatment allowed me to examine the differing overall effects of fluctuating acidities on periphyton and was implemented by reciprocal transplant of individual tiles among the 3 lakes in mid July. I retrieved tiles, removed macroinvertebrates with forceps, and transferred tiles in sealed individual plastic containers with 400 mL of their lake water of origin. Five tiles from each lake were harvested before transplantation to quantify initial standing crop, and to evaluate the efficiency of the removal of macroinvertebrates. Tiles were held for 24 h in coolers at ambient lake temperatures (~20°C) prior to transfer. The rationale for this holding period was to equalize transplant shock for tiles from the 3 lakes, and to allow sufficient time for redistribution of tiles across the lakes.

The exclosure treatment allowed me to test for the effects of grazing and algal interactions on periphyton development. Each exclosure housed 1 tile, and consisted of a 12 × 12 × 10 cm polyethylene framework with open top and side panels around which different exclosure materials were affixed. Different biotic conditions were achieved by assigning tiles to complete exclosures, or to 2.5-mm-mesh macrograzer exclosures, or to open containers. Macrograzer exclosures were made using fiberglass mesh. The complete exclosures consisted of cellulose dialysis membrane (Spectra/Por4; molecular weight cutoff = 12,000 to 14,000). Dialysis membrane was rinsed with distilled water, stored in 5% nitric acid, and washed with lake water before use (LaZerte 1984). Light attenuation by these materials was measured using a Li-Cor Quantum photometer (LI-185B) and Li-192SB sensor. After a 4-wk incubation period, photosynthetically active radiation was reduced by ~10% of the complete and macrograzer exclosures.

The importances of physiological tolerance and competitive exclusion to community composition were assessed by comparing communities transplanted to complete exclosures with those transplanted to open and macrograzer exclosures. Complete exclosures prevented colonization by non-native algal species and all grazers, allowing me to determine the extent to which the community was tolerant of abiotic changes associated with transplantation into a different lake. Furthermore, if the community was tolerant of the new abiotic environment but did not persist in the macrograzer exclosure, this outcome would suggest that species composition was affected by other biotic factors, such as competitive exclusion or micrograzers. Also, if an introduced taxon did persist on transplanted tiles, but was not a resident within the lake, this result would be taken as evidence of physical dispersal barriers.

Quantification of periphyton

Total and species-specific algal biomass within periphyton accrued over 4-wk incubations were quantified using biovolume, the most precise measure of periphyton abundance (Morin and Cattaneo 1992). Algal counts were performed using Utermöhl chambers. Taxa lists were first constructed by scans of settled samples at 100× and 250×. Large colonial and filamentous taxa were enumerated separately at 250× to improve the accuracy of density estimates (Stevenson and Lowe 1986). Other algae were enumerated at 400×. A minimum of 300 viable algal units (i.e., those with intact chloroplasts) were counted per sample.

Cell densities were converted to areal biovolumes by estimating cell volume using geometric volume formulae (Wetzel and Likens 1991). Diatom biovolumes were estimated by taking planimetric measurements of valve surface areas and girdle widths for taxa illustrated in Patrick and Reimer (1966, 1975). Average cell dimensions were based on measurements of 30 cells of each taxon. Counting precision was evaluated using coefficients of variance (CV) of triplicate enumerations of randomly chosen samples. CV for total biovolume was <10% of the mean.

Major taxonomic references consulted were Patrick and Reimer (1966, 1975) for diatoms; Prescott et al. (1972, 1975, 1977, 1981a, 1981b) and Croasdale et al. (1983) for desmids; Wei et al. (1989) for the Zygnemataceae; Bourrelly (1970) for cyanobacteria; and Bourrelly (1968) and Prescott (1982) for all others. Identification of diatoms was aided by reference slides of cleared frustules.

Scanning electron microscopy

Scanning electron microscopy (SEM) was used to qualitatively assess differences in the structure properties of periphyton among treatments. Granitic cubes were fixed to tiles using epoxy putty. Cubes were retrieved and fixed with 5% gluteraldehyde in a cacodylate buffer solution, and serially dehydrated with ethanol. Cubes were placed on aluminum SEM mounts, sputter-coated with gold and palladium, and examined with a Hitachi S-2500 SEM microscope. Each cube was surveyed with transects at low magnification (2500×). Representative areas of the epilithon from various treatments were photographed.

Statistical analyses

CCA was conducted using CANOCO (Version 3.12, Agricultural Mathematics Group, Wageningen) to determine which combinations of environmental variables best explained patterns in periphyton assemblages along the gradient of acidified lakes (ter Braak 1986). Algal biovolume and all environmental variables, except pH, were $\log_{10}$-transformed prior to ordination, to stabilize variance (Morin and Cattaneo 1992) and to downweight large-celled species that otherwise would have dominated the ordination (ter Braak 1991). The data-screening protocol of Hall and Smol (1992) was used to remove superfluous environmental variables, thereby preventing the occurrence of the distorting "arch effect". Monte Carlo permutation testing was used to establish the significance of eigenvalues (ter Braak 1991).

Differences in total and species-specific algal biovolume among 4-wk assemblages from the reciprocal transplant experiment were examined by analyses of $\log_{10}$-transformed data using randomized-block analysis of variance (RB-ANOVA) and CCA. Duncan's multiple range tests ($p = 0.05$) with Bonferroni adjustment were used for a posteriori multiple comparisons. CCA was used to determine the significance and strength of response of periphyton assemblages to reciprocal transplantation. Only taxa that occurred in at least 3 samples were ordinated. CCA was chosen over linear discriminant analysis, or canonical variate analysis, because CCA does not require that the number of samples exceed the number of species of treatments (ter Braak 1991). Monte Carlo permutation testing was used to determine the significance of the reciprocal transplant on taxonomic composition. Classification of communities from the transplant experiment was performed using the TWINSPAN option contained with COINSPAN (Version 0.5), a computer program developed by Carleton et al. (1996).

Results

Pheriphyton survey

CCA was performed using the 27 most abundant taxa, accounting for over 90% of total pe-

TABLE 3. The 27 most abundant algal taxa across the 3 experimental lakes. Taxon identification numbers correspond to those used in the ordination plots. BG = blue-green alga (cyanobacterium), D = diatom, FG = filamentous green alga, and G = nonfilamentous green alga.

No.	Taxon	No.	Taxon
1.	*Aphanocapsa* spp. (BG)	15	*Euastrum* spp. (G)
2.	*Calothrix* spp. (BG)	16.	*Spondylosium planum* Wolle (FG)
3.	*Homoeothrix juliana* (Menegh.) Kirchn. (BG)	17.	*Achnanthidium minutissimum* (Kütz.) Czar. (D)
4.	*Bulbochaete* spp. (FG)	18.	*Brachysira serians* Kütz. (D)
5.	*Coleochaete scutata* de Breb. (G)	19.	*Cymbella microcephala* Grun. (D)
6.	*Oedogonium* spp. (FG)	20.	*Eunotia* spp. (D)
7.	*Pediastrum* spp. (G)	21.	*Fragilaria acidobiontica* Charles (D)
8.	*Mougeotia* spp. (FG)	22.	*Frustulia rhomboides* (Ehr.) Det. (D)
9.	*Spirogyra* spp. (FG)	23.	*Gomphonema acuminatum* Ehr. (D)
10.	*Zygogonium ericetorum* Kütz. (FG)	24.	*Pinnularia* spp. (D)
11.	*Z. tunetanum* Gauthier-Liévre (FG)	25.	*Surirella* spp. (D)
12.	*Actinotaenium cucurbita* (Nägeli) Teiling (G)	26.	*Synedra* spp. (D)
13.	*Cosmarium* spp. (G)	27.	*Tabellaria quadrisepta* Knuds. (D)
14.	*Cylindrocystis brebissonii* Menegh. (G)		

riphyton biovolume in the 3 experimental lakes (Table 3). The first 4 CCA axes accounted for 63% of the total variance in the species data and 95% of the joint variance in the periphyton and water chemistry data (Table 4). The 1st axis was significant ($p < 0.01$) and contrasted the few acidobiontic species, such as *Homoeothrix juliana* (No. 3) and *Zygogonium ericetorum* (No. 10) with taxa having broader acidity tolerances (e.g., *Pinnularia* spp.) or lower acidity optima (e.g., *Cymbella microcephala*) (Fig. 2). The 1st axis represents an acidity gradient best explained by DIC (52%), pH (12%), and Al concentration (10%). The 2nd axis is a less-well-defined gradient of DOC (13%). Humic lakes (i.e., Freeland Lake, Wagon Road Lake) and their associated periphyton assemblages, containing algae such as *Zygogonium tunetanum* (No. 11) and *Euastrum* spp. (No. 15), are separated from clearwater (< 2 mg DOC/L) lakes and their assemblages along the 2nd axis.

Ordination of lakes showed that the 3 experimental lakes selected for periphyton transplants span the primary acidity gradient (Fig. 2). Little Sheguiandah Lake, the low-acidity system, was negatively associated with the 1st axis, while A.Y. Jackson Lake was of moderate acidity, occurring nearer the origin of the CCA plot. Ruth-Roy Lake was the most acidic site, occurring to the extreme right along axis 1.

Reciprocal transplant of periphyton: total biovolume accrual

The degree of change in algal biovolume following reciprocal transplant differed significantly ($p < 0.001$) across periphyton from the 3 lakes (Fig. 3, origin effect). Specifically, periph-

TABLE 4. Summary of CCA of periphyton algal assemblages and water chemistry across 15 lakes.

	Axis 1	Axis 2	Axis 3	Axis 4	Total
Eigenvalues	0.36	0.11	0.07	0.04	0.91
Species–environment correlations	0.97	0.96	0.82	0.81	
Cumulative % variance					
species data	40.7	51.4	58.8	63.3	
species-environment	61.1	77.3	88.4	95.2	
Sum of unconstrained eigenvalues					0.91
Sum of constrained eigenvalues					0.61

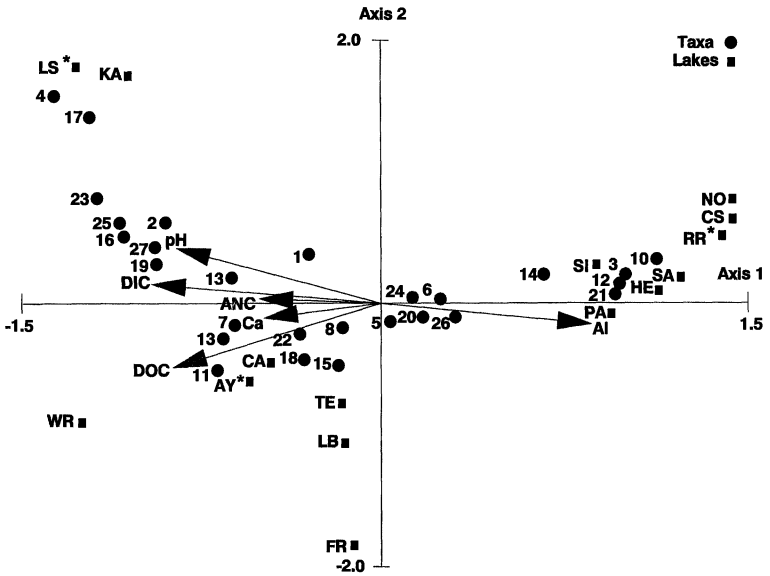

FIG. 2. Association of algal species and lakes from a CCA of periphyton and water chemistry across lakes. For lake code see Table 1. Taxa identification numbers appear in Table 3.

yton from lakes of low and moderate acidity accrued significantly ($p < 0.05$) more biovolume than periphyton from the most acidic lake. Periphyton from the most acidic lake showed negligible or negative growth under most conditions. No significant ($p = 0.37$) blocking effect was detected.

Algal growth was significantly ($p < 0.001$) suppressed when periphyton was grown in the lake of highest acidity (Fig. 3, lake acidity effect). Periphyton from the less acidic lakes decreased in biovolume over the 1-mo incubation in Ruth-Roy Lake. Only periphyton that was native to Ruth-Roy Lake did not show net declines in growth. A significant ($p < 0.01$) interaction between origin and lake acidity factors occurred because periphyton from the low and moderately acidified lakes accrued significantly less biovolume when grown in the most acidic lake.

RB-ANOVA showed that the effect of exclosures on periphyton biomass was not significant ($p = 0.11$). Although the lack of a significant interaction between lake acidity and exclosure ($p = 0.27$) suggested that periphyton biomass was not affected by biotic factors, growth tended to be greater in macrograzer exclosures in the least acidic lake for periphyton from the low and moderately acidic lakes (Fig. 3). Further, periphyton from the most acidic lake consistently showed reduced growth after 1 mo in the complete exclosures.

Reciprocal transplant of periphyton: taxonomic composition

CCA of reciprocally transplanted periphyton identified significant ($p < 0.01$) differences in taxonomic composition associated with certain treatments. The 1st CCA axis (37%; $\lambda_1 = 0.44$) separated assemblages based on both lake acidity and origin of periphyton (Fig. 4). Taxa that either originated in, or survived after being transplanted into, high acidity sites were separated from those that grew best in either the moderate or low acidity sites. The 2nd axis (14%; $\lambda_2 = 0.17$) separated native assemblages within the low acidity lake on the basis of exposure to macrograzers (Fig. 4). Assemblages grown in complete exclosures were not distinct from those grown in either open or macrograzer exclosures.

TWINSPAN classified periphyton into 4 communities, each represented by different indicator taxa and contrasting structural organization (Fig. 5). The influence of macrograzing in the least acidic lake was shown by the firmly attached indicator taxa *Bulbochaete* and *Coleochaete scutata*, along with a monolayer of short-stalked

Achnanthidium minutissimum. In contrast, periphyton in macrograzer exclosures had greater structural complexity. Ungrazed communities contained the loosely attached indicator taxa *Pediastrum* and *Spondylosium planum.* Periphyton grown in the more acidic lakes had relatively simple community structures. *Actinotaenium cucurbita* was identified as an indicator of increased acidity. Communities exposed to high acidity conditions consisted of dense filamentous canopies of zygnematacean algae, including the acidobiontic indicator species *Zygogonium ericetorum* that extended over a sparse understorey of diatoms, such as *Fragilaria acidobiontica,* and a filamentous cyanobacterium (*Homoeothrix juliana*).

Discussion

The abiotic environment appears to be the primary regulator which inhibits epilithic periphyton growth in acidified Killarney lakes. These findings are consistent with others showing low periphyton photosynthesis (Turner et al. 1987, 1994) and biomass (Turner et al. 1991, Cattaneo 1992) in acidified lakes. In situ studies of epilithon in unperturbed and acidified lakes of low alkalinity have also shown carbon limitation of productivity (Turner et al. 1987, 1991, 1994). However, severe carbon limitation is replaced by nitrogen and phosphorus limitation with increasing lake alkalinity (Fairchild and Sherman 1992). Thus, epilithic periphyton growth should be enhanced by recovery in severely acidified lakes as a result of increased DIC availability.

Algal growth was suppressed when periphyton was transplanted into the most acidic lake. Stokes (1981) reported similar decreases in periphyton biomass when algae were transplanted into lakes of high acidity. In contrast, growth of lotic periphyton is enhanced when communities are grown in low pH habitats (Mulholland et al. 1986, Planas et al. 1989). The discrepancy between the effects of acid stress on lentic and lotic periphyton may reflect differences in boundary layer dynamics. Species with high affinities for CO_2 uptake may proliferate in acidified streams in which DIC is being continually replenished within the thin boundary layer by turbulence. In contrast, lentic periphyton must contend with a thicker boundary layer in which gas exchange is reduced. Although filamentous green algae do proliferate in these acidified Killarney lakes, they occur primarily as metaphyton near the surface of the water column, as epilithon along the shoreline splash zone, or as epipelon directly above organic sediments where DIC availability is likely high due to aerobic decomposition.

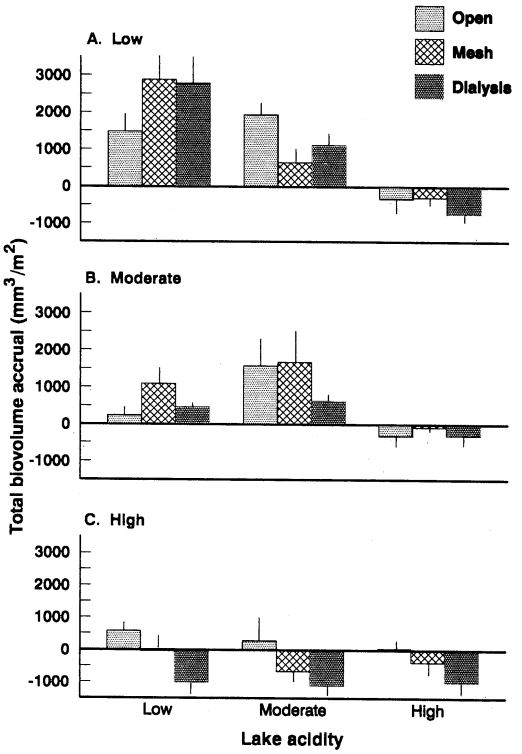

FIG. 3. Mean (±1 SE) biovolume accrual by periphyton from lakes of (A) low acidity, (B) moderate acidity, and (C) high acidity when grown in each of the three lakes, either open to grazers and algae (stippled bars), in 2.5-mm-mesh macrograzer exclosures (hatched bars), or in complete exclosures that were lined with dialysis membrane (solid bars).

Periphyton moved from the most acidic lake accrued significantly less biomass than periphyton that was native to the less acidic lakes. Thus periphyton growth did not recover from the initial inhibitory effects of high acidity despite being transferred and grown under less acidic conditions for 1 mo. Perhaps this result involves a trade off between responsive growth potential and increased acid tolerance in the periphyton from Ruth-Roy Lake, causing the algae to be relatively unresponsive to decreases in lake acidity. Furthermore, periphyton adapted

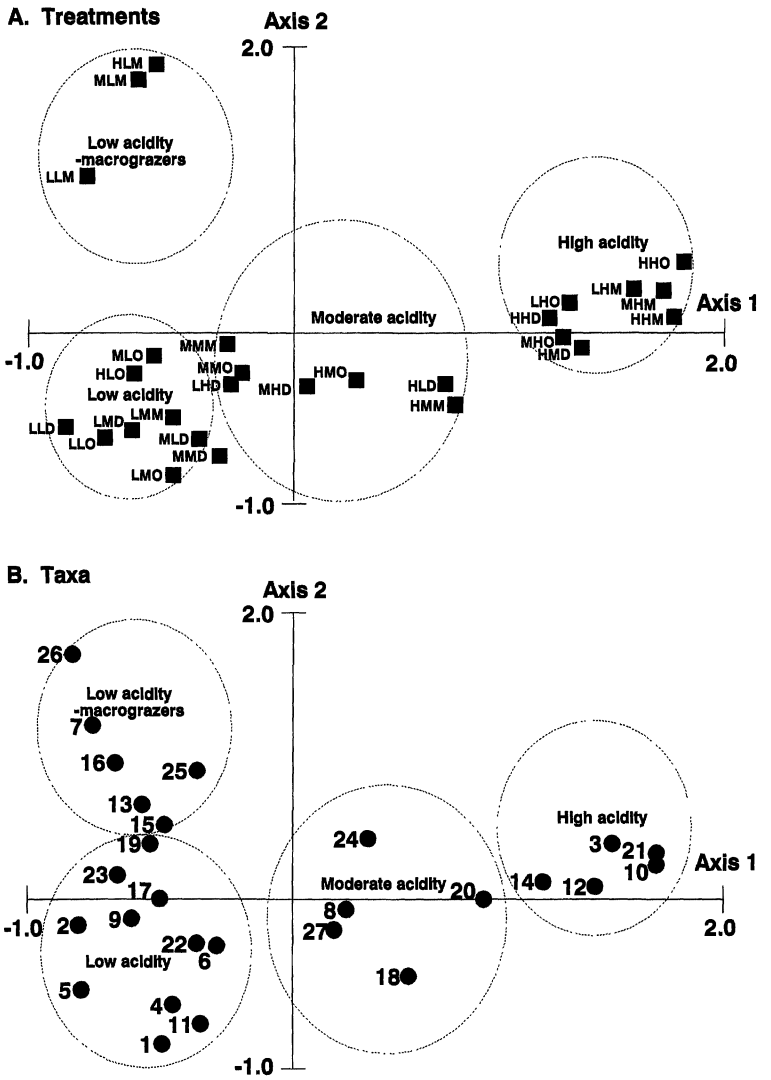

FIG. 4. Association of (A) treatment combinations and (B) taxa resulting from reciprocal transplant of periphyton in different exclosures across 3 lakes. Three-letter codes in (A) indicate treatment combinations of periphyton origin and lake acidity (L = low; M = moderate; H = high), followed by exclosure type (D = dialysis membrane that excluded grazers and algae; M = mesh that excluded macrograzers; O = open). Numbers in (B) refer to taxa identification numbers that appear in Table 3. TWINSPAN classifications of 4 experimental periphyton communities are enclosed by the hatched circles.

to the most acidic lake conditions consistently showed negative growth when placed in the complete exclosures. Although dialysis membranes will allow for the diffusion of nutrients to algal cultures, the movement of nutrients is decreased in complete exclosures (Turner et al. 1991). Thus, water movement likely plays an important role in minimizing carbon limitation of periphyton growth, especially in severely acidified lakes.

Macrograzers did not have a significant effect on periphyton biomass across the 3 experimental lakes. However, the exclusion of macrograzers tended to enhance periphyton growth in the least acidic lake. Macrograzers removed loosely attached and large-celled algal forms in the least

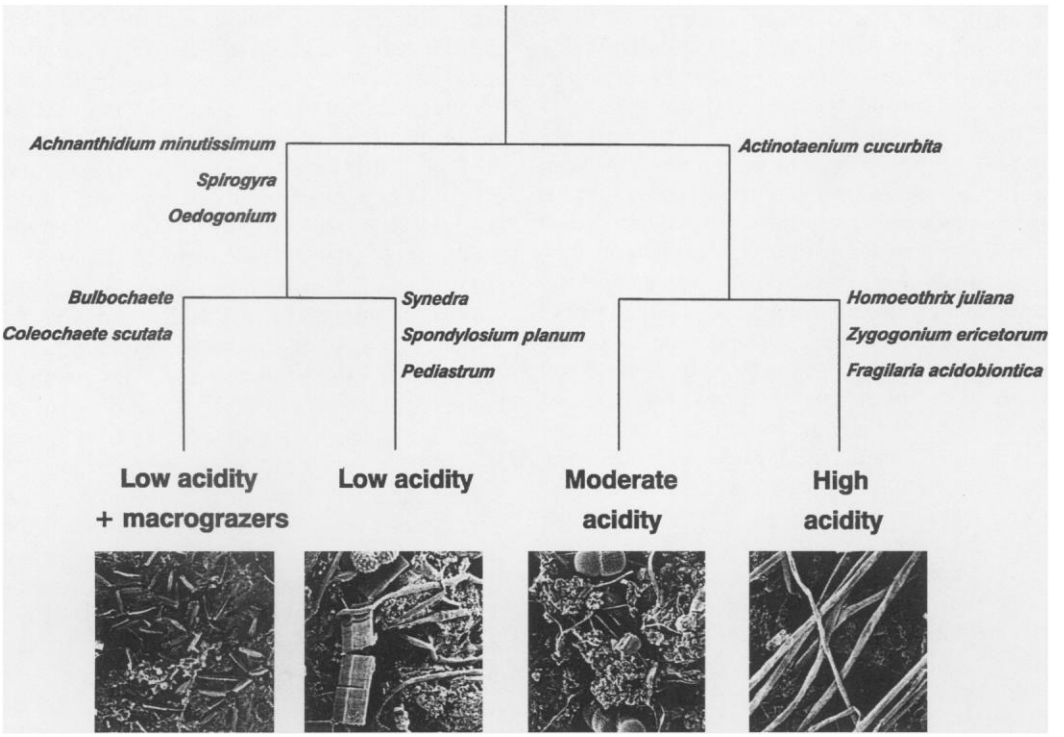

FIG. 5. TWINSPAN of the different experimental periphyton communities. Structural organization of the 4 different periphyton communities are shown that resulted from the reciprocal transplanting of periphyton in open, macrograzer, and complete exclosures across lakes of different acidity. Indicator taxa appear above the experimental periphyton communities with which they have a strong association.

acidic lake, apparently facilitating the growth of small adnate diatoms and large prostrate colonial forms. Cattaneo and Kalff (1986) also found large invertebrates altered lentic periphyton species composition by selectively feeding on large and filamentous algae without reducing periphyton biomass. Species replacements likely compensated for, and thereby, prevented significant losses in periphyton biomass due to herbivory. It is also possible that disturbance and enhanced nutrient recycling through macrograzing stimulated algal growth so as to offset the suppressive effects of grazing on periphyton biomass (see Feminella and Hawkins 1995).

Taxonomic composition of periphyton in the 3 experimental lakes appeared to be regulated primarily by abiotic conditions. CCA of species and treatment data showed taxonomic composition responded significantly to changes in acidity among the 3 lakes. The 1st CCA axis, representing an acidity gradient, accounted for the highest percentage (37%) of variance within the periphyton data. Furthermore, CCA of periphyton taxa and limnological data from the 15-lake survey showed that DIC availability best explained species responses to changes in acidity. That periphyton assemblages are strongly regulated by abiotic factors, as my data suggest, supports the paleolimnological use of benthic diatom assemblages for reconstructing the acidification histories of these and other lakes (A. S. Dixit et al. 1992, S. S. Dixit et al. 1992).

Unlike biomass accrual, taxonomic composition of the periphyton from the most acidic lake was significantly altered when grown in either of the less acidic lakes. Therefore, changes in periphyton taxonomic composition likely precede changes in standing crop during recovery in severely acidified lakes. As a caveat, my experiment exposed periphyton to rapid changes in lake acidity which may not have allowed sufficient time for physiological adaptation to new abiotic conditions. In contrast, natural recovery in these acidified lakes will take decades (Keller

et al. 1992a). Slow chemical recovery will likely facilitate adaptation, and thereby reduce some of the significant changes I observed in periphyton biomass and taxonomic composition.

Biotic regulation of the taxonomic composition of periphyton was evident only in the least acidic lake. The results support the hypothesis that macrograzers suppress the proliferation of zynematacean filamentous green algae in low-acid-stress lakes (Stokes 1986). Specifically, macrograzers reduced periphyton to a monolayer of grazing-resistant taxa, such as small-celled *r*-strategists (*Achnanthidium minutissimum*), and those with specialized holdfast morphologies (*Bulbochaete, Coleochaete scutata*). In contrast, periphyton in macrograzer exclosures was composed of loosely attached filamentous desmids (*Spondylosium planum*), pseudofilamentous colonies of diatoms (*Synedra*), and large-celled taxa (*Pediastrum*) within a thick bacteria-laden detrital matrix. Thus, macrograzers may impeded the formation of metaphyton in late summer by grazing filamentous periphyton during midsummer.

Comparison of macrograzer communities across the 3 experimental lakes suggests that the most important herbivores were green frog (*Rana clamitans*) tadpoles. Dickman (1968) also found that tadpoles ingested loosely attached desmids and zygnematacean filaments. Although crayfish may be expected to regulate the growth of filamentous periphyton in some lakes (France et al. 1991), the fact that crayfish abundances in the 3 experimental lakes were comparable suggests crayfish grazing was not responsible for the observed effects of macrograzing. Moreover, periphyton constitutes only a minor proportion of the crayfish diet, which consists mainly of vascular macrophytes and macroinvertebrates (e.g., Hanson et al. 1990). Suppression of filamentous periphyton by *R. clamitans* in some Canadian Shield lakes of low acidity can persist until metamorphosis of tadpoles in late summer (M. D. Graham, University of Toronto, unpublished data).

Ordination and classification of periphyton taxa failed to detect significant differences between assemblages transplanted into either open or complete exclosures, suggesting effects of species exclusion were minimal. Stokes (1986) hypothesized that proliferation of filamentous zygnematacean species in acidified lakes alters patterns of interspecific competition and allows these filamentous greens to exclude other species. Filamentous green algae may outcompete adnate and prostrate epilithic forms by forming an upper canopy that reduces light and nutrient availability (Hudon and Legendre 1987, Turner et al. 1991). Although competitive exclusion may exist within acid-stressed periphyton, my findings suggest that its influence on taxonomic composition after a 4-wk incubation period is not significant. However, transplant shock could have sufficiently disturbed the experimental periphyton so as to negate the potential for more subtle algal interactions, such as competition. Longer incubation periods, or more stable assemblages, may be required for competitive exclusion to be detected within periphyton.

Taxa did not persist in periphyton that was transplanted into lakes of differing acidities unless they already were native to those lakes. Thus, species exclusion due to physical dispersal barriers was not evident. Instead, species' tolerance of abiotic conditions was likely the primary determinant of distribution across the acidity gradient. This finding supports the common assumption that microbial communities are not constrained by physical dispersal barriers (McCormick and Cairns 1991). If dispersal barriers existed they would have significant ramifications for biological recovery, causing periphyton recolonization to lag behind chemical recovery, producing a "bottleneck". In the absence of species exclusion, spatial patterns in periphyton assemblages along a gradient of lake acidity should reflect temporal patterns of recovery within an individual lake (Keller et al. 1992a).

My findings suggest that abiotic control of periphyton is superseded by biotic control during recovery in acidified lakes (Fig. 6). The recolonization of recovering acidified lakes by algal species should result in increased periphyton growth. In the early stages of recovery from severe acidification, epilithic periphyton growth should respond positively to renewed DIC availability, which should increase food resources and facilitate recolonization by benthic herbivores. In the advanced stages of recovery, macrograzers may help maintain periphyton growth by suppressing the development of filamentous green algae that otherwise can detach to form metaphyton that may outcompete periphyton for light and nutrients.

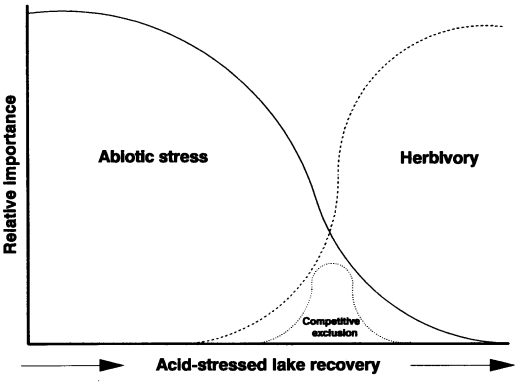

FIG. 6. Conceptual model showing changes in the relative importance of abiotic and biotic regulation of periphyton in recovering acidified lakes. Physiological acid-stress is replaced by herbivory as a regulator of periphyton in recovering acidified lakes. Competitive exclusion of periphyton species by filamentous green algae may occur near the acidity threshold that marks the proliferation of these algae.

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