

Benthic grazing and functional compensation in stressed and recovered lakes

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Abstract: During ecosystem recovery, grazing pressure is expected to increase as larger herbivores become reestablished. Alternatively, grazing pressure may remain unchanged during recovery as large consumers replace and functionally compensate for more abundant populations of smaller, tolerant herbivores. We tested these hypotheses by conducting a 90-day experiment in which three size categories of benthic consumers were excluded from producers in three chemically stressed and three recovered lakes. Our findings showed that consumers did not significantly affect producer biomass in either type of lake. However, exposure to larger and more abundant grazers did induce a physiognomic shift towards less edible producers in the recovered lakes. In comparison, recovered lakes contained significantly greater producer biomass and diversity. Comparison of the observed subtle effects of consumers and pronounced negative impact of ecosystem stress on benthic producers suggest that they can compensate for natural disturbances (e.g., grazing), but not for the other multiple stressors associated with anthropogenic acidification of the Killarney lakes.

Résumé : Durant la restauration des écosystèmes, on s'attend à ce que la pression de broutement augmente, à cause du retour des grands herbivores. D'un autre côté, la pression de broutement peut rester inchangée durant la restauration, parce que les consommateurs de grande taille remplacent les populations plus abondantes d'herbivores tolérants plus petits et compensent fonctionnellement pour eux. Nous avons testé ces hypothèses lors d'une expérience de 90 jours durant laquelle trois tailles de consommateurs benthiques ont été séparées des producteurs dans trois lacs en stress chimique et trois lacs restaurés. Nos résultats montrent que les consommateurs n'affectent pas significativement la biomasse des producteurs dans les deux types de lacs. Cependant, une exposition à des brouteurs plus abondants et de plus grande taille induit un changement physiognomique qui favorise les producteurs moins comestibles dans les lacs restaurés. En comparaison, les lacs restaurés possèdent une biomasse et une diversité de producteurs significativement plus importantes. La comparaison des effets subtils des consommateurs que nous observons et l'important impact négatif du stress de l'écosystème sur les producteurs benthiques indiquent que ces derniers peuvent compenser pour les perturbations naturelles (par ex., le broutement), mais pas pour les autres multiples facteurs de stress reliés à l'acidification anthropique des lacs de Killarney.

[Traduit par la Rédaction]

Introduction

Ecological processes are expected to become more efficient as species diversity and size structure increase in ecosystems during recovery from anthropogenic stressors (Odum 1985). More diverse consumer assemblages can better exploit food resources (Naeem and Li 1998; Norberg 2000; Duffy 2002), while larger consumers often exert stronger predation pressure than do small-bodied, stress-tolerant species (Brooks and Dodson 1965; Cattaneo and Kalff 1986; Diehl 1992). For example, reestablishment of a more diverse assemblage of larger herbivores has been hypothesized to increase grazing pressure in recovering acidified lakes (Fairchild

and Sherman 1992; Vinebrooke et al. 2003a; Hogsden and Vinebrooke 2005).

Alternatively, ecological processes may remain unchanged during ecosystem recovery owing to compensatory species dynamics (i.e., functional compensation; Frost et al. 1995; Klug et al. 2000; Fischer et al. 2001). For instance, sparse populations of large, sensitive consumers replace higher densities of smaller, acid-tolerant species in recovering acidified lakes (Hogsden and Vinebrooke 2005), thereby possibly maintaining net secondary production and grazing pressure. Further, producers may also functionally compensate or even overcompensate for shifts in grazing pressure and other environmental changes in acidified lakes (Vinebrooke 1996; Klug et al. 2000; Vinebrooke et al. 2003b).

Functional compensation involving shifts in the body sizes of benthic consumers has not been addressed empirically in anthropogenically stressed lakes. Previous experiments that used only coarse-meshed (250 µm) exclosures showed that consumers affected producers in circumneutral lakes, but not in acidified lakes where large-bodied grazers were in low abundance (Vinebrooke 1996; Graham and Vinebrooke 1998). Therefore, we conducted an experiment in which three size categories of benthic consumers were excluded from producers to determine their relative impacts in stressed and recovered

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Table 1. Physical characteristics and acidification histories for six lakes included in the experiment (mean \pm standard error (SE)).

Lake	Lake surface area (ha)	Depth (m)		Acidification history (pH)*		
		Mean	Max.	1971–1973	1992	2003
Stressed						
Acid	19.6	10.9	29	—	5.1	5.1
Norway	63.3	15.1	33.6	4.7	4.5	5.4
Ruth Roy	54.5	4.3	18	4.5	4.2	5.8
Mean	45.8	10.1	26.9	4.6	4.6	5.4
SE	13.4	3.1	4.6	0.1	0.3	0.2
Recovered						
A.Y. Jackson	6.5	7	9.8	5	5.6	6.2
Carlyle	156.7	5.7	14.6	5	5.1	6.3
Little Sheguiandah	4.5	1.7	2.7	5	6.3	6.9
Mean	55.9	4.8	9	5	5.7	6.5
SE	50.5	1.6	3.5	0	0.3	0.2

*Acidification history data (1971–1973 and 1992) were taken from Vinebrooke and Graham (1997); 2003 data is from Hogsden and Vinebrooke (2005). Note that data was not available for Acid Lake for 1971–1973.

lakes. We hypothesized that a compensatory size shift from numerous, small, tolerant consumers towards fewer, larger, sensitive consumers maintains grazing pressure as lakes recover from acidification. We also expected that a shift in the size of grazer would alter feeding selectivity, thereby affecting producer diversity. Further, we predicted that relatively diverse producer assemblages would show greater functional compensation than species-poor, higher consumer groups, such as herbivores (e.g., Tilman 1999; Klug et al. 2000; Vinebrooke et al. 2003b).

Materials and methods

Study area

The experiment was conducted from May to August 2003 in six lakes located in Killarney Provincial Park (46°3'N, 81°21'W), southwest of Sudbury, Ontario, Canada. Reduction of anthropogenic emissions of sulphur dioxide has resulted in chemical improvements in many of these acid-stressed lakes (Snucins et al. 2001; Findlay 2003; Keller et al. 2003). Lakes were selected based on their historical and current acidities, which are useful abiotic determinants of ecosystem recovery from anthropogenic acidification (Vinebrooke and Graham 1997; Keller et al. 2003). The “stressed” lake group consisted of three acidic waterbodies (Acid, Norway, and Ruth-Roy) situated on orthoquartzite bedrock, while the “recovered” group included three chemically recovered (i.e., returned to pre-acidification pH), circumneutral lakes (A.Y. Jackson, Carlyle, and Little Sheguiandah) located in lowland areas with limestone, granite, or sandstone bedrock (Snucins et al. 2001). All lakes in the stressed and recovered lake groups became acidified below pH 5 in the early 1970s but currently differ in chemical recovery status (Table 1; Vinebrooke and Graham 1997; Vinebrooke et al. 2003a; Hogsden and Vinebrooke 2005).

Lake water chemistry

Chemical analyses were performed on water samples collected monthly during summer 2003 from the site of the experiment in each lake. Each study area was located on a

south-facing bedrock outcropping that provided a shallow-sloping littoral habitat and a 1 m submerged rock ledge that ran parallel to the shore. Duplicate samples were analyzed at the Limnology Laboratory at the University of Alberta (Edmonton, Alberta, Canada). In the lab, pH was measured using a Fischer Accumet model 925 meter and compared with values recorded during summer sampling. Conductivity was determined using a radiometer (Copenhagen model CDM83). Samples for total nitrogen (TN) samples were pre-digested with alkaline potassium persulfate according to Ameel et al. (1993) and determined by flow injection analysis on a Technicon Autoanalyzer™. Total dissolved phosphorus (TDP) was measured using standard colorimetric procedures following digestion with potassium persulfate (Menzel and Corwin 1965). Dissolved organic carbon (DOC) and dissolved inorganic carbon (DIC) were both analyzed using a Shimadzu TOC-5000A analyzer.

Consumer communities

Zoobenthos were collected monthly from the shallow (~1 m depth) littoral area using a D-frame dip net (500 μ m mesh size). Three time-standardized sweeps were performed with the net along a 10 m transect adjacent to the experimental site and parallel to the shoreline. Samples were pooled and stored in coolers until sorting was performed later in the day. Predators were immediately separated from the samples during sorting. A dissecting microscope was used at 40 \times and 100 \times magnifications to sort and identify all invertebrates to the family level. Many invertebrates were collected as early instars and were difficult to identify to genus or species; therefore, family level was used for all statistical analyses. Body size was measured at the widest part of the body (usually head or abdomen) and was divided into four size classes (<0.5, 0.5–2.5, 2.5–10, and >10 mm) based on accessibility into the different experimental enclosures (see below). Consumers were also classified according to published dietary descriptions and functional feeding groups (Merritt and Cummins 1984; Thorp and Covich 2001). We use the term consumer to refer collectively to all functional feeding groups (i.e., consumer = predators + grazers + omnivores).

For statistical analyses, the grazer fraction is considered separately when compared with other consumers (i.e., predators plus omnivores). After drying at 60 °C for 96 h, samples were weighed. Dry mass for snails and crayfish were shell-free.

Experimental design and setup

A two-factor, split-plot design with repeated measures was used to determine the effects of chemical recovery and size structure of benthic consumer communities on autotrophic biomass and species composition. The lake-type treatment (plot) consisted of two levels (stressed pH ~5, recovered pH ~7) that were replicated three times for a total of six lakes. The lakes served as independent experimental units because of the absence of connecting streams. Four consumer treatment levels (subplot) were created using open, coarse (10 mm), intermediate (2.5 mm), and fine (0.5 mm) mesh enclosures (40.6 cm long × 27.9 cm wide × 15.2 cm deep) that were closed on the bottom. Consumer treatment levels were not replicated within any of the lakes. A single partial cage for each level of the consumer treatment was also deployed in each lake to account for the physical presence of the enclosure material (i.e., cage effect). Partial cages were constructed using the same design and materials of the complete enclosures; however, they lacked vertical walls. Each enclosure contained a matrix of 12 unglazed ceramic tiles (56.25 cm² each), which simulated the predominant type of algal habitat (quartzite–granitic bedrock) that exists along the shallow shorelines of the study lakes. All ceramic tiles were acid washed (10% HCl) for 48 h prior to deployment. In each lake, cages were randomly deployed along a submerged rock ledge and secured using bricks in early May 2003.

Sampling and enumeration of producers

Four tiles from each enclosure were retrieved randomly and processed monthly for a total of three sampling events. Producers were removed from the four tiles with a hard-bristled toothbrush, rinsed with filtered lake water, and pooled together. The harvested material was transferred from the tray to a 500 mL graduated cylinder and was filled to 200 mL with filtered lake water. The top of the cylinder was covered and inverted several times to mix thoroughly and was subsampled in the following sequence: 50 mL was filtered and concentrated on Whatman GF/C glass microfiber filter papers and frozen for chlorophyll *a* (Chl *a*) analysis, and 100 mL was preserved with Lugol's solution for taxonomic analysis. Producer biomass was quantified using fluorimetry and biovolume estimates (Wetzel and Likens 2000). For Chl *a* determinations, samples were extracted in 95% ethanol for 24 h. Tubes were centrifuged at 5 °C and 4400 r·min⁻¹ for 10 min, and dilutions, if required, were made with 95% ethanol. Extracts were read on a Shimadzu model RF-1501 spectrofluorometer at 436 nm and 680 nm, using a 1 cm path-length quartz cell. For biovolume estimates, samples were well mixed and diluted depending on the density of algal and detrital material. After 1 h of settling into the base of an Utermöhl chamber, a minimum of 300 viable cells were counted under 400× or 1000× magnification on an inverted microscope. Cell viability was based on chloroplast integrity for eukaryotic cells and pigmentation for cyano-

Table 2. List of periphytic producers used in canonical correspondence analysis (CCA).

No.	Taxon	Algal group	Physiognomic group
1	<i>Anabaena</i>	C	LU
2	<i>Aphanocapsa</i>	C	LU
3	<i>Chroococcus</i>	C	LU
4	<i>Lyngbya</i>	C	LU
5	<i>Merismopedia</i>	C	LU
6	<i>Oscillatoria</i>	C	LU
7	<i>Phormidium</i>	C	LC
8	<i>Rhaboderma</i>	C	LU
9	<i>Dinobryon</i>	CHR	LU
10	<i>Actinotaenium</i>	DE	LU
11	<i>Bambusina</i>	DE	LC
12	<i>Closterium</i>	DE	LU
13	<i>Cosmarium</i>	DE	LU
14	<i>Cylindrocystis</i>	DE	LU
15	<i>Desmidium</i>	DE	LC
16	<i>Euastrum</i>	DE	LU
17	<i>Gonatozygon</i>	DE	LU
18	<i>Netrium</i>	DE	LU
19	<i>Penium</i>	DE	LU
20	<i>Pleurotaenium</i>	DE	LU
21	<i>Staurastrum</i>	DE	LU
22	<i>Xanthidium</i>	DE	LU
23	<i>Achnanthes</i>	DI	FU
24	<i>Actinella</i>	DI	FU
25	<i>Cymbella</i>	DI	FU
26	<i>Eunotia</i>	DI	LU
27	<i>Fragilaria</i>	DI	LC
28	<i>Frustulia</i>	DI	LU
29	<i>Gomphonema</i>	DI	FC
30	<i>Melosira</i>	DI	LC
31	<i>Navicula</i>	DI	LU
32	<i>Nedium</i>	DI	LU
33	<i>Pinnularia</i>	DI	LU
34	<i>Semiorbis</i>	DI	LU
35	<i>Stenopterobia</i>	DI	LU
36	<i>Surirella</i>	DI	LU
37	<i>Synedra</i>	DI	FU
38	<i>Tabellaria</i>	DI	LU
39	<i>Bulbochaete</i>	FG	FC
40	<i>Klebsormidium</i>	FG	LC
41	<i>Mougeotia</i>	FG	LC
42	<i>Oedogonium</i>	FG	FC
43	<i>Spirogyra</i>	FG	LC
44	<i>Stigeocolonium</i>	FG	FC
45	<i>Zygonium</i>	FG	LC
46	<i>Chlamydomonas</i>	G	LU
47	<i>Pediastrum</i>	G	LU
48	<i>Scenedesmus</i>	G	LU
49	<i>Stichococcus</i>	G	LU

Note: Taxon identification numbers correspond to those used in CCA plots. Taxa are identified as cyanobacteria (C), chrysophytes (CHR), desmids (DE), diatoms (DI), filamentous green algae (FG), and green algae (G) and are grouped in to loose understory (LU), loose canopy (LC), firm understory (FU), or firm canopy (FC) categories based on their growth form and inferred attachment potential.

Table 3. Water chemistry for stressed and recovered lakes (mean \pm standard error (SE)) over the course of the 90-day experiment based on sampling in littoral area.

Variable	Stressed	Recovered	Lake type		Time		Time \times lake type	
			<i>F</i>	<i>P</i>	<i>F</i>	<i>P</i>	<i>F</i>	<i>P</i>
pH	5.43 \pm 0.18	6.42 \pm 0.16	12.24	0.02	1.29	0.32	0.13	0.94
DOC (mg·L ⁻¹)	2.02 \pm 0.15	3.92 \pm 0.25	14.21	0.02	0.66	0.59	3.09	0.07
DIC (mg·L ⁻¹)	0.88 \pm 0.27	1.20 \pm 0.21	0.51	0.52	1.64	0.23	0.21	0.89
TDP (μ g·L ⁻¹)	1.93 \pm 0.35	2.89 \pm 0.66	167.68	0.04	4.87	0.11	1.12	0.47
TN (μ g·L ⁻¹)	248.42 \pm 10.23	307.16 \pm 12.15	11.96	0.03	1.50	2.64	0.35	0.80
Conductivity (μ S·cm ⁻¹)	25.19 \pm 2.46	26.73 \pm 2.24	0.03	0.88	0.74	0.55	0.20	0.89

Note: DOC, dissolved organic carbon; DIC, dissolved inorganic carbon; TDP, total dissolved phosphorus; and TN, total nitrogen.

Table 4. Level of accessibility to experimental enclosures for consumer families in stressed and recovered lakes.

Family	Stressed				Recovered			
	Fine	Intermediate	Coarse	Open	Fine	Intermediate	Coarse	Open
Hyallidae		×				×		
Gammaridae							×	
Ranidae								×
Dytiscidae (adult)	×						×	
Dytiscidae (larvae)			×					
Gyrinidae			×					
Cyprinidae								×
Cambaridae			×				×	
Chironomidae	×				×			
EphemereIIDae						×		
Heptageniidae						×		
Viviparidae							×	
Corixidae			×					
Notonectidae			×					
Hydrachnida		×				×		
Asellidae		×				×		
Corydalidae			×				×	
Sialidae						×		
Libellulidae			×				×	
Gomphidae			×				×	
Coenagrionidae		×					×	
Limnephilidae			×				×	

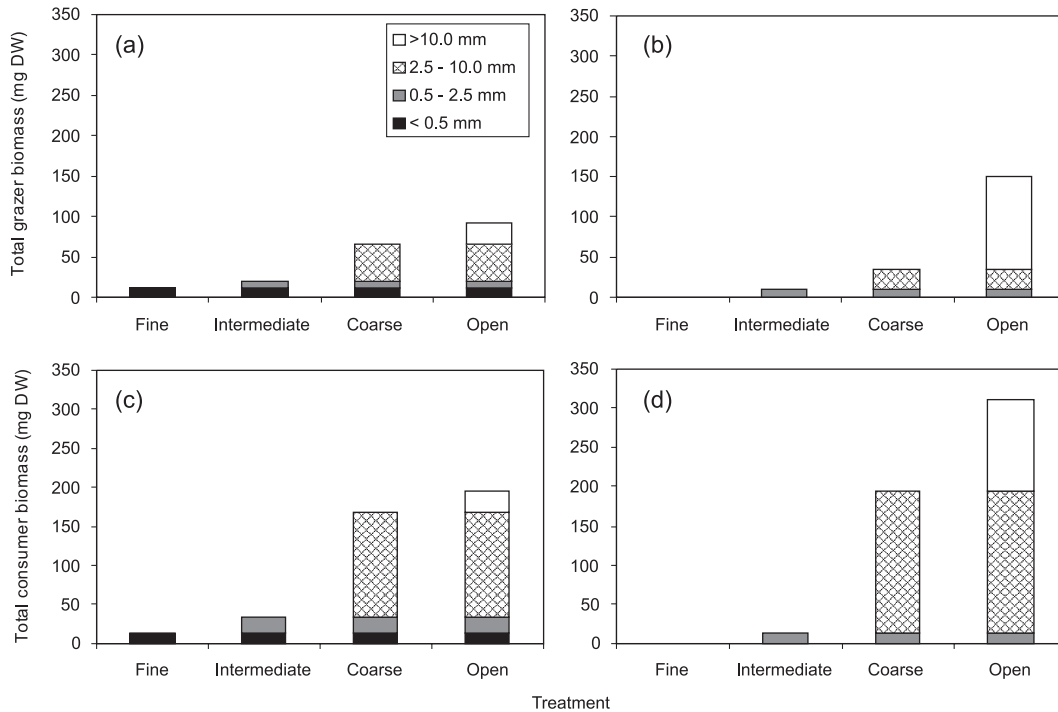
Note: Taxa missing from table indicate families that were absent from each lake type.

bacteria. Cell counts were converted to biovolumes by approximating cell volume using geometric formulae (Hillebrand et al. 1999; Sun and Liu 2003). Biovolume was estimated on a per-cell basis for solitary and filamentous cells. A minimum of 20 cells per taxon was used to determine the mean dimensions used in geometric formulae, where sufficient cell numbers were available. Biovolume estimates were converted to biomass assuming the specific gravity equaled one and mean values derived from duplicate or triplicate enumerations. Comparison of benthic producers that had been sampled from acid-washed tiles and freshly collected rocks during a 2003 survey of these lakes indicated that producer communities on tiles contained greater average species richness (49 taxa vs. 30 taxa) (Hogsden and Vinebrooke 2005). *Homoeothrix*, a relatively rare and slow-growing cyanobacterium, was the only genus found on the rocks that was absent from the tiles. Therefore, representative sampling of the benthic producer community was achieved using tiles in each lake. All algal

samples were identified to genus according to Prescott (1982) and Wehr and Sheath (2003). Identification to this level of taxonomic resolution generalized species within a genus and was a limitation of our study design. Duplicate counts of each sample were done, and if species richness (i.e., number of species) varied by more than 20%, a triplicate sample was counted.

To assess ecological responses by producers to grazing and lake type, taxa were grouped into four physiognomic categories based on their individual morphologies and abilities to attach to surfaces (Table 2; Peterson 1996; Steinman 1996; Graham and Vinebrooke 1998). The physiognomy of benthic producers serves as a highly reliable predictor of their responses to several environmental factors, including grazing (Steinman 1996), resource availability (Burkholder 1996), and physical disturbance (Peterson 1996). Taxa that possessed a stalked attachment structure or specialized basal holdfast were termed firmly attached. In contrast, other taxa

Fig. 1. Biomass of each size fraction of (a) grazers in stressed lakes, (b) grazers in recovered lakes, (c) consumers in stressed lakes, and (d) consumers in recovered lakes that can access each enclosure treatment level (data presented are mean biomass (DW, dry weight) over entire experiment).



that lacked tightly adhering structures were defined as weakly attached. Firmly and loosely attached groups were subdivided into canopy and understory categories based on the stature of each taxon. Canopy taxa included filamentous and long-stalked growth forms, while understory taxa were prostrate or short-stalked.

Although the physiognomic approach can help simplify interpretations of ecological responses by producers, several caveats pertain to its use. For example, mucilaginous excretions by taxa that lacked specific attachment structures may have enabled them to better adhere to the tiles. Secondly, heterotrichous taxa (e.g., *Stigeoclonium*) can produce both erect canopy and prostrate understory filaments. However, this alga was classified as a canopy producer because its erect filaments were more abundant than basal cells. Thirdly, certain genera were represented by species that differ in their growth forms (e.g., stalked and nonstalked *Achnanthes* spp). In these cases, the relative abundance of identifiable species and their associated growth form were used to classify each genus into one of the four physiognomic groups.

Statistical analyses

Repeated measures analysis of variance (RM-ANOVA) was used to evaluate differences in water chemistry, the effectiveness of experimental exclusion of consumers, and the effects of lake type and consumer size on producer biomass (Chl *a* and biomass) (Scheiner and Gurevitch 2001). Lake type was included as a between-plot factor, and consumer size was included as a within-plot factor. Statistical analysis was performed with SYSTAT Version 10 (SPSS Inc., Chicago, Illinois) using the general linear model procedure. If the assumption of sphericity was violated, we used conservative Greenhouse–Geisser-corrected significance levels ($P < 0.05$).

Repeated-measures multivariate analysis of variance (RM-MANOVA) was used to determine differences in the physiognomic and taxonomic composition of the producers between stressed and recovered lakes and grazer treatment levels. All data were \log_{10} -transformed to improve normality and homogeneity of variance, which were verified using Shapiro–Wilks and Levene's tests, respectively.

A specific set of statistical results were required to support our hypothesized effect of a compensatory size-shift by consumers maintaining grazing pressure across acidified and recovered lakes. First, we would have to detect a significant time-dependent interaction between lake type and consumer size treatments. If so, then multiple-comparison testing would be required to determine the nature of the interaction. Specifically, pronounced grazing by small, acid-tolerant consumers would be demonstrated if producers remained significantly more abundant in the fine-meshed enclosures compared with both open controls and coarser-meshed cages placed in the acidified lakes. In comparison, a significantly negative difference of a similar magnitude between coarser-meshed enclosures and open controls in the recovered lakes would indicate a compensatory size shift towards larger grazers.

Canonical correspondence analysis (CCA) was conducted using CANOCO version 4.51 (2003, Biometris-Plant Research International, Wageningen, the Netherlands) to examine the effect of water chemistry variables and grazer biomass on producer assemblages. All algal, grazer, and water chemistry data, with the exception of pH, used in the ordination were \log_{10} -transformed to stabilize variances (Morin and Cattaneo 1992) and to reduce the influence of large-celled taxa (e.g., filamentous green algae (FGA)), which would have dominated the ordination (ter Braak 1986). To remove ecologically irrelevant and insignificant or redundant environmental

variables, data were screened prior to the final CCA using forward selection. Monte Carlo permutation testing (499 iterations) was used to test for the statistical significance of the final ordination. Algal taxa that did not occur in at least three samples were excluded from ordination analysis.

Results

Water chemistry

The water chemistry differed substantially between the two lake types (Table 3). Stressed and recovered lakes differed by approximately a full pH unit. Recovered lakes also contained significantly higher concentrations of TDP, TN, and photo-protective DOC (RM-ANOVA, lake type, $P < 0.05$). Conductivity and DIC levels were not significantly different across all six lakes.

Consumer communities

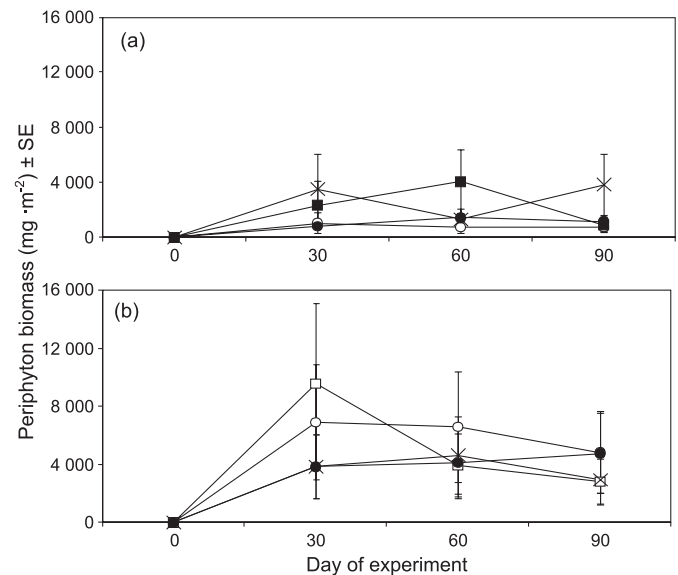
The fine-meshed enclosures potentially excluded most consumers, except chironomids and hydrachnids, from producers in both stressed and recovered lakes (Table 4). Small dytiscids could also enter the fine-meshed enclosures, but only in the stressed lakes. Intermediate-meshed enclosures could exclude ephemeroptera and heptagenids that were only present in the recovered lakes. Similarly, coarse-meshed enclosures could prevent larger consumers (amphipods, viviparids, gammarids) accessing the tiles in the recovered lakes. The size of some consumers differed between stressed and recovered lakes and affected the treatment enclosure that could be accessed. For example, small coenagrionids entered the intermediate-meshed enclosures in the stressed lakes, while larger coenagrionids were excluded in the recovered lakes (Table 4).

Use of enclosures significantly reduced potential exposure of producers to all consumers (RM-ANOVA, consumer effect, $F_{[3,16]} = 7.28$, $P = 0.003$), but not all grazers (RM-ANOVA, consumer effect, $F_{[3,16]} = 5.74$, $P = 0.39$) during the entire experiment (Fig. 1). Multiple pairwise comparisons (Tukey's test) revealed significant differences in consumer biomass between each treatment level ($P = 0.005$), with biomass increasing as enclosure size increased. However, potential exposure did not differ significantly between acidified or recovered lakes for all total consumers (RM-ANOVA, lake type – consumer–time interaction, $F_{[6,32]} = 1.6$, $P = 0.17$) or grazers ($F_{[6,32]} = 0.7$, $P = 0.65$). There was an apparent shift from smaller grazers (e.g., chironomids) in stressed lakes to fewer but larger taxa (e.g., mayflies, tadpoles, minnows) in recovered lakes (Figs. 1c, 1d); however, it was not statistically significant (ANOVA, $P > 0.05$).

Lake-type and consumer-size effects on producer biomass

Exclusion of different size categories of consumers did not significantly affect total producer biomass in either stressed or recovered lakes (RM-ANOVA, lake type – consumer–time interaction, $F_{[6,24]} = 0.3$, $P = 0.95$; Fig. 2). In particular, total biomass did not differ significantly ($t = 0.76$, $df = 7$, $P = 0.47$) between fine-meshed enclosures and open controls by day 90 of the experiment. Chlorophyll-inferred producer biomass did not differ significantly between the partial-cage

Fig. 2. Benthic producer biomass in each enclosure in (a) stressed lakes ($n = 3$) and (b) recovered lakes ($n = 3$) over the 90-day experiment. Open circles, open enclosures; open squares, coarse enclosures; solid circles, intermediate enclosures; ×, fine enclosures.



treatments and open controls (RM-ANOVA, lake type – consumer–time interaction, $F_{[6,20]} = 0.5$, $P = 0.81$).

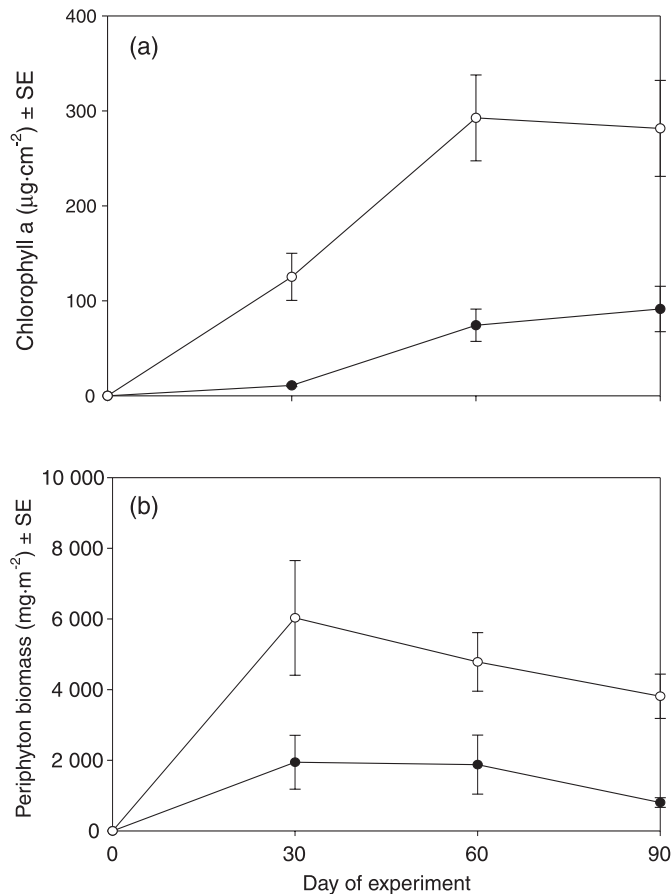
Acidified lake conditions significantly suppressed the total abundance of benthic producers based on differences in Chl *a* (RM-ANOVA, lake-type effect, $F_{[1,22]} = 31.4$, $P < 0.001$) and algal biomass ($F_{[1,24]} = 38.4$, $P < 0.001$) between the stressed and recovered lakes (Fig. 3). At the end of the experiment, both metrics revealed producer biomass had increased in the recovered lakes and remained relatively unchanged in the stressed lakes (Fig. 3). Total Chl *a* and producer biomass were positively correlated across all treatments ($R^2 = 0.25$, $P < 0.001$). Total producer biomass on experimental tiles was considerably less than recorded biomass on surveyed rocks during the same time period in both stressed and recovered lakes (Hogsden and Vinebrooke 2005).

Lake-type and consumer-size effects on producer diversity

The physiognomic structure of benthic producers did not differ significantly between stressed and recovered lakes or among the three enclosure types (Fig. 4; RM-MANOVA, lake type – consumer–time interaction, $F_{[33,132]} = 0.6$, $P = 0.94$). Loosely attached canopy taxa represented the most abundant growth form across all treatments.

Acidified lake conditions significantly altered producer diversity based on changes in the abundance of major algal groups (Fig. 5; RM-MANOVA, lake type – time interaction, $F_{[14,224]} = 7.0$, $P < 0.001$). Specifically, significantly more desmids (RM-ANOVA, lake-type effect, $F_{[1,22]} = 21.21$, $P < 0.001$), diatoms ($F_{[1,22]} = 53.95$, $P < 0.001$), FGA ($F_{[1,22]} = 13.61$, $P = 0.001$), and other green algae ($F_{[1,22]} = 9.50$, $P = 0.05$), but not cyanobacteria ($F_{[1,22]} = 1.02$, $P = 0.32$) occurred in recovered lakes. FGA was the most abundant algal group in the recovered lakes, representing over 90% of the total producer biomass by day 90. Diatoms were the second most abundant group in the recovered lakes, accounting for 7% of the final total producer biomass. In comparison,

Fig. 3. (a) Chlorophyll *a* and (b) producer biomass in stressed lakes (solid circles) and recovered lakes (open circles) over the 90-day experiment.



FGA were less abundant by a full order of magnitude in the stressed lakes in which they still represented over 80% of the total producer biomass by day 90. Consumers did not significantly affect the abundance of major algal groups (RM-MANOVA, consumer–time interaction, $F_{[42,224]} = 0.4$, $P = 0.98$). Other green algae included all chlorophytes that did not belong to the Zygnematales or Oedogoniales. A summary of RM-ANOVA and RM-MANOVA analyses is provided in Table 5.

Chemical differences between the stressed and recovered lakes significantly altered producer diversity at the genus level based on CCA (Fig. 6). A total of 62 genera from six major taxonomic groups (cyanobacteria, chrysophytes, diatoms, desmids, FGA, and other green algae) were detected in the experiment. Chrysophytes made up less than 1% of total biomass and were excluded from further analysis. CCA was performed on 49 genera that accounted for 99% of the total producer biomass across all enclosures (Table 2). Fourteen genera were excluded from analysis, as they occurred in fewer than three samples. The first CCA axis explained 18.7% of the total taxonomic variance (Fig. 6a). CCA axis 1 ($\lambda_1 = 0.30$) represented a significant gradient ($F = 13.79$, $P < 0.005$) that was best described by DOC ($r = -0.95$) and pH ($r = -0.79$). The first axis contrasted genera (*Euastrum* (No. 16), *Pleurotaenium* (No. 20), and *Pinnularia* (No. 33)) that were common in recovered lakes, from filamentous green algae

(*Stigeoclonium* (No. 44)) and desmids (*Actinotaenium* (No. 10) and *Cylindrocystis* (No. 14)) that were more abundant in the stressed lakes.

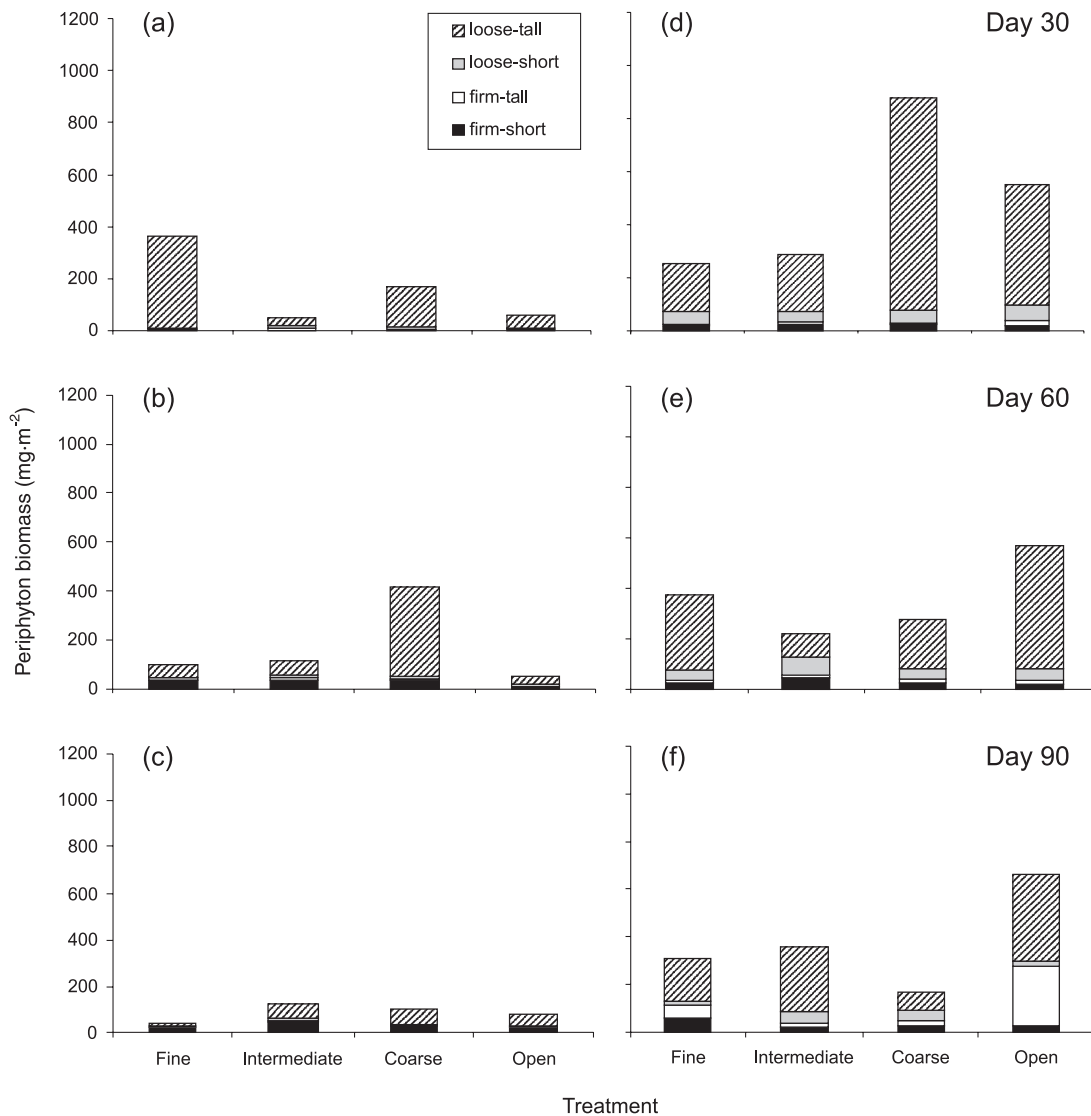
Consumers also significantly influenced producers at the genus level (Fig. 6a). Grazer biomass was retained as an independent, significant explanatory variable that was correlated ($r = 0.41$) with CCA axis 2 ($\lambda_2 = 0.10$). The second CCA axis explained 9% of the total taxonomic variance and was also related to DIC ($r = 0.78$) and conductivity ($r = 0.56$). CCA axis 2 separated grazer-resistant, firmly attached genera (*Gomphonema* (No. 29), *Achnanthes* (No. 23), and *Bulbochaete* (No. 39)) and *Xanthidium* (No. 22) in recovered lakes from loosely attached taxa, such as *Lyngbya* (No. 4) and *Penium* (No. 19), in stressed lakes. Clear separation of recovered from stressed lakes was evident based on producer diversity (Fig. 6b).

Discussion

Our results provided partial support for the hypothesis that a shift from numerous small benthic consumers to fewer large grazers maintains secondary production and grazing pressure in recovering acidified lakes. Lack of a significant difference in total grazer biomass between the lake types was associated with large taxa (larval mayflies, tadpoles, minnows) being more abundant in the recovered lakes, while the stressed lakes contained greater densities of smaller grazers (chironomids). However, we did not detect a significant interactive effect of consumer size and lake type on producer biomass. Therefore, grazing pressure did not depend on different size categories of consumers in acidified and recovered lakes. Further, consumers did not exert a significant main effect on producer biomass across all lakes. Instead, the influence of grazing was only detected at the taxonomic (i.e., genus) level in recovered lakes based on ordination analysis. In contrast, other environmental stressors significantly suppressed producer biomass and altered taxonomic composition in the acidified lakes. These findings suggest that benthic producers functionally compensated for weak grazing pressure, but not for anthropogenic acidification. Below, we consider potential explanations for weak benthic grazing effects and differences in functional compensation by producers to environmental stressors and grazing in relatively unproductive lakes.

Benthic grazing may have been difficult to detect in these lakes because consumer–producer interactions can be weak in oligotrophic systems (Elser and Goldman 1991; Lamberti 1996; but see Rosemond et al. 1993). Low productivity combined with predation by benthivores (e.g., crayfish, odonates, minnows) can suppress smaller herbivorous invertebrates, thereby minimizing their effect on producers in stressed lakes without piscivorous fish (Appelberg et al. 1993). Further, the prevalence of omnivory (Gu et al. 1997) and flexibility in feeding strategy (Dangles 2002) likely weaken the reliance of many grazers on producers in stressed ecosystems. For example, stable isotopic analyses reveal that most benthic invertebrates are not obligate herbivores in unproductive lakes (Vander Zanden and Rasmussen 1999; Vinebrooke et al. 2001). Therefore, low population densities of omnivorous, benthic invertebrates likely preclude detection of grazing effects in oligotrophic systems.

Fig. 4. Benthic producer biomass based on physiognomic groups in (a–c) stressed lakes and (d–f) recovered lakes for each exclosure treatment level.



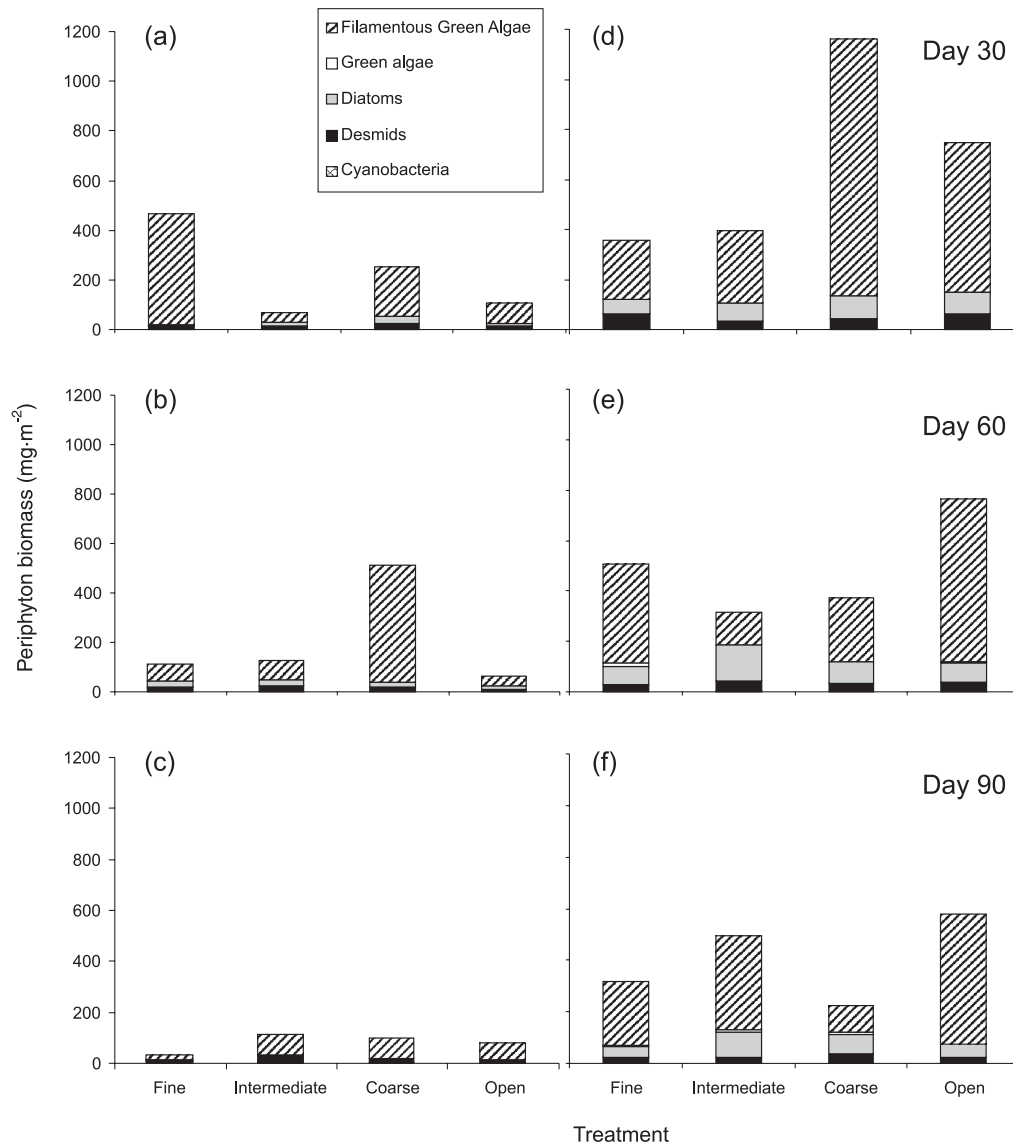
High abundances of potentially edible producers in all lakes also suggested that ambient grazing pressure was weak during the study. Specifically, filamentous canopy producers were abundant in both stressed and recovered lakes despite their reported susceptibility to large-bodied grazers (Gelwick and Matthews 1992; Vinebrooke 1996; Graham and Vinebrooke 1998). Compensatory shifts involving edible (zygnematacean taxa) and relatively grazer-resistant filamentous algae (oedogoniacean taxa) may also have offset the effects of benthic grazers on producer biomass (e.g., Vinebrooke 1996; Graham and Vinebrooke 1998). In addition, production of mucilaginous substances and other potentially antiherbivory chemicals may also have decreased the edibility of certain filamentous producers, thereby weakening grazing pressure (Steinman 1996).

Alternatively, lack of statistically significant grazing effects on producer biomass may have been a consequence of low study power. Thus, we performed a post hoc power analysis using a conventional effect size of 0.8 (Quinn and Keough 2002). Based on our level of replication ($n = 3$),

power analysis revealed an approximate 55% probability of detecting an 80% difference in autotrophic biomass between our open controls and fine-meshed exclosures. Based on the variance of total biomass responses by producers to the consumer-size treatment, replication would need to be doubled (i.e., $n = 6$ or 12 cages) to detect a significant grazing effect. Morin and Cattaneo (1992) recommended a minimum level of replication of 20 for benthic algal studies using biomass as their response variable, indicating that intense sampling would be required to detect weak grazing effects. In addition, we may have underestimated cyprinid and mollusc biomass because of the D-net sweep method used to sample zoobenthos, which may be reflected in our nonsignificant grazing effects.

Another possibility is that the difference in nutrient concentrations between lake types may have been sufficient to positively affect producer biomass in recovered lakes. Evidence from recovered Killarney lakes has shown that increased producer biomass is positively related to higher TDP (Hogsden and Vinebrooke 2005); however, this is not

Fig. 5. Benthic producer biomass divided into taxonomic groups found in different enclosure treatments in (a–c) stressed and (d–f) recovered lakes over the 90-day experiment.



likely linked to differences in acidification histories, which do not affect TDP levels in boreal lakes (Vinebrooke et al. 2003a).

Nevertheless, our ordination analyses suggested that grazers were more abundant in recovered lakes, affecting producers at the genus level. This finding agreed with correlations between body size and feeding selectivity of benthic grazers (France et al. 1991; Vinebrooke 1996; Graham and Vinebrooke 1998). Also, Cattaneo and Kalff (1986) showed that large invertebrate grazers altered benthic producers by selectively feeding on large diatoms and filamentous producers, while not affecting total biomass. In contrast, studies in recovering acidified lakes have shown that high abundance of large benthic grazers can suppress producer biomass (Graham and Vinebrooke 1998). Similarly, Heneberry (1997) reported that the lack of large grazers (e.g., crayfish) resulted in massive accumulation of producer biomass in acid-stressed lakes.

Although planktonic producers exhibit functional compensation in stressed lakes, (Klug et al. 2000; Vinebrooke et al.

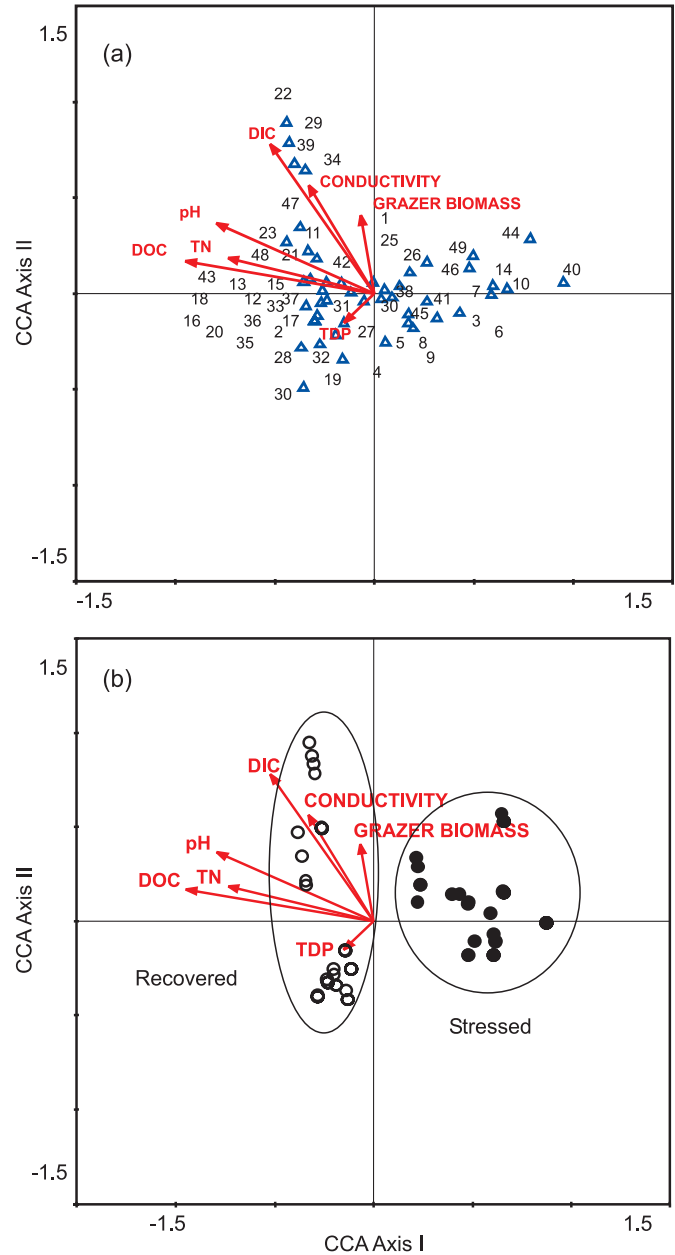
2003b), acidified lake conditions significantly suppressed benthic producer biomass in our study. The spatial and temporal extent of widespread acidification over several decades may have limited the compensation potential of benthic producers by depleting the regional species pool (Minns et al. 1990; Arnott et al. 2001; Vinebrooke et al. 2003b). In contrast, Findlay et al. (1999) and Vinebrooke et al. (2003b) have documented recovery by benthic producers from an 8-year experiment in which acid was added to a single lake located among other pristine lakes. Further, small acid-tolerant producers may not fully compensate for larger sensitive taxa, in terms of total biomass (Cattaneo et al. 1998; Klug et al. 2000).

The difference between the impacts of benthic consumers and abiotic stress on producer biomass versus community composition highlight the strong capacity of algae to compensate for weak, natural disturbances (e.g., grazing) but not potentially chronic, novel disturbances (e.g., anthropogenic acidification). Therefore, we must consider interactions among

Table 5. Summary of repeated measures analysis of variance (RM-ANOVA) and repeated measures multivariate analysis of variance (RM-MANOVA) results including all model effects and interactions.

Effect	Exclosure effectiveness (RM-ANOVA)		Autotrophic biomass (RM-ANOVA)		Chl <i>a</i> biomass (RM-ANOVA)		Partial cage		Autotrophic community (RM-MANOVA)		Taxonomic composition	
	Consumer	Grazer	Overall	Overall	Overall	Overall	F	P	F	P	F	P
Lake type	0.01	0.94	0.01	0.94	31.40	<0.001	12.35	0.01	44.59	<0.001	26.08	<0.001
Consumer	7.28	0.003	1.08	0.39	1.73	0.22	0.01	1.00	0.33	0.81	0.28	0.84
Time	6.21	0.01	5.83	0.01	50.37	<0.001	10.56	0.001	22.31	<0.001	49.42	<0.001
Lake type × consumer	0.29	0.84	0.30	0.83	1.29	0.33	0.43	0.74	1.40	0.29	0.18	0.91
Lake type × time	0.92	0.41	0.24	0.79	5.37	0.01	4.01	0.03	0.78	0.66	7.01	<0.001
Consumer × time	1.34	0.27	1.46	0.23	1.50	0.22	0.21	0.97	0.34	1.00	0.39	0.98
Lake type × consumer × time	1.62	0.17	0.70	0.65	5.19	0.00	0.48	0.81	0.63	0.94	0.50	0.97

Fig. 6. Association of (a) producers and (b) lake sampling scores with environmental variables in exclosures in six lakes based on canonical correspondence analysis (CCA) using log₁₀-transformed producer biomass, water chemistry (except pH), and grazer biomass data. For panel b, open circles are from recovered lakes, and solid circles are from stressed lakes. DOC, dissolved organic carbon; DIC, dissolved inorganic carbon; TDP, total dissolved phosphorus; and TN, total nitrogen.



abiotic stressors and altered trophic interactions to predict community responses to long-term stress (Frost et al. 1995; Hulot et al. 2000).

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