The Ecophysiology of Epilithic Diatom Communities of Acid Lakes in Galloway, Southwest Scotland

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The ecophysiology of epilithic diatom communities of acid lakes in Galloway, southwest Scotland

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Lake-water chemistry in Galloway, southwest Scotland is characterized by strong correlations between low calcium and high aluminium concentrations and low pH. Nitrate and silicate levels were sufficient for diatom growth but phosphate was limiting. N:P and Si:P ratios indicated severe phosphate limitation according to the Redfield ratio. Chlorophyll-*a* specific epilithic phosphatase activity expressed as a ratio of acid to alkaline phosphatase activity showed a clear relation to pH with acid phosphatase predominating at pH 5.5 and below. Acid phosphatase activity in epilithon from low pH lakes was inducible as phosphate levels decreased, and inhibited by the addition of phosphate. Inducible acid phosphatase activity clearly confers a selective advantage to epilithic diatom communities growing in oligotrophic lakes of low pH.

INTRODUCTION

Although it is widely accepted that there is a relation between lake-water pH and diatoms, and that diatom records in sediments can be used to infer past pH (Renberg & Hellberg 1982; Flower & Battarbee 1983; Charles 1985), very little is known of the physiological bases for the changes observed in the diatom communities. Possible causes for changes in the abundance of some species of diatom in lakes that have undergone surface water acidification include changes in nutrient chemistry (Dickson 1978; Nalewajko & O'Mahony 1988), and increases in the concentration of some metals, in particular, aluminium (Schindler 1988; Dillon *et al.* 1988). Increased dissolved aluminium concentrations may result in the precipitation of phosphorus reducing biologically available orthophosphate to very low levels (Hsu & Rennie 1962). Phosphorus starvation and aluminium toxicity have been suggested as factors involved in decreased algal biomass and productivity in acid lakes (Nalewajko & Paul 1985; Patterson *et al.* 1988) and phosphatase production has been used as an indicator of phosphorus limitation (Healey 1973).

Alkaline phosphatase is produced on the external surface of the plasma membrane (Doonan & Jensen 1980) and hydrolyses a wide variety of organic phosphorus compounds to orthophosphate, which is transported into the cell. Inducible alkaline phosphatase activity is widespread among the major algal groups (Owens & Esias 1976). Some species of algae may have up to five different phosphatases including acid phosphatases (Matagne *et al.* 1976; Boavida & Heath 1986). Some are synthesized in response to low external phosphate concentrations (inducible), others remain constant regardless of the external environment (constitutive). Alkaline phosphatase is not efficient below pH 5.5 and this may provide an explanation for the absence of some diatoms from low pH lakes.

Lake survey: environmental and algal characteristics

A survey of 32 lakes of varying water chemistry (Flower 1986) was undertaken during April and September 1988; four sites were monitored on a monthly basis for one year. Water chemistry and measurements of epilithon physiology were used to test the hypothesis that the success of algal communities at low pH and low inorganic nutrient levels is related to their ability to metabolize organic phosphorus via the induction of acid phosphatase activity.

Sampling and analytical methods for a full range of chemical variables (table 1) are given in Smith & Carroll (1988). Epilithic communities from each lake were assayed for acid- (pH 5.0), alkaline- (pH 9.0) and ambient- (at the pH of the lake) phosphatase activity by using *p*-nitrophenyl phosphate as substrate. Phosphatase activity and organic phosphorus concentrations were expressed as a function of the chlorophyll-*a* content of the epilithon (specific epilithic phosphatase activity, SEPA; specific epilithic organic phosphorus, SEOP).

The epilithic communities comprised bacteria and algae but were dominated by diatoms. The composition of diatom assemblages from each site is given in Flower (1986).

In the survey of the 32 lakes soluble reactive phosphate (SRP) levels were rarely above 0.3 μmol l^{-1} and the N:P ratios (as NO_3-N and PO_4-P) were often much higher than 35:1 (up to 5000:1), which would indicate severe phosphorus limitation according to the Redfield ratio (Redfield 1958). With decreasing pH the lakes typically had decreasing calcium levels (figure 1*a*) and increasing aluminium concentrations (figure 1*b*).

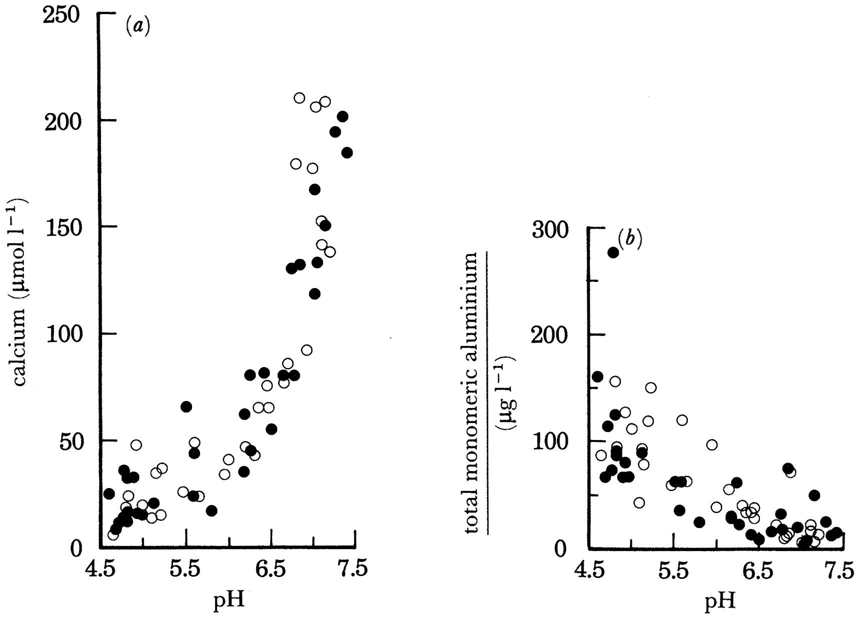

Figure 1. (*a*) Calcium and (*b*) total monomeric aluminium for the 32 lakes plotted as a function of pH; (●), April; (○), September.

Water chemistry and physiological results for the four main sites are summarized in table 1. Nitrate and silicate levels were generally higher than 4 μmol l^{-1} whereas SRP levels were below 0.1 μmol l^{-1} for up to six months in three of the lakes. Seasonal variations in SRP reflect the spring and autumn growth in planktonic and epilithic communities.

Table 1. Characteristics of water chemistry and epilithon physiology in four Galloway lakes (mean values 1988–1989)

(Abbreviations: *n*, number of samples; TAl, total aluminium; TMAl, total monomeric aluminium; N-LMAl, non-labile monomeric aluminium; SOP, sestonic organic phosphorus; SEOP, specific epilithic organic phosphorus; sepa, specific epilithic phosphatase activity.)

	Loch Whinyeon	Loch Fleet	Loch Howie	Loch Grannoch
n	12	12	12	12
colour (absorbance at 250 nm)	0.217	0.268	0.062	0.232
pH	6.96	6.29	5.82	4.68
conductivity/(μS (25 °C) cm^{-1})	71.0	47.3	59.0	51.2
alkalinity/(μeq l^{-1})	303	62.8	28.8	−24.4
NO_3-N/(μmol l^{-1})	13.5	25.7	32.5	19.4
PO_4-P/(μmol l^{-1})	0.07	0.10	0.07	0.16
Si/(μmol l^{-1})	21.0	16.8	51.3	28.1
Ca/(μmol l^{-1})	139	79.7	65.6	23.5
Mg/(μmol l^{-1})	48.6	22.2	44.8	24.3
K/(μmol l^{-1})	9.06	7.63	7.76	6.03
Na/(μmol l^{-1})	213	145	201	170
Cl/(μmol l^{-1})	309	238	329	248
SO_4/(μmol l^{-1})	54.3	42.1	75.6	43.5
TAl/(μg l^{-1})	38.3	105	130	291
TMAl/(μg l^{-1})	14.3	38.4	58.0	198
N-LMAl/(μg l^{-1})	10.7	31.4	32.3	87.1
SOP/(mmol l^{-1})	0.450	0.494	0.513	0.605
SEOP/(mmol P μg Chla^{-1})	0.021	0.011	0.044	0.049
SEPA-acid/(nmol PO_4 (μg Chl*a*)$^{-1}$ min^{-1})	1.65	2.03	7.17	3.94
SEPA-alkaline/(nmol PO_4 (μg Chl*a*)$^{-1}$ min^{-1})	6.25	2.17	4.69	1.87
SEPA-ambient/(nmol PO_4 (μg Chl*a*)$^{-1}$ min^{-1})	2.79	2.06	8.36	3.40

PHOSPHATASE ACTIVITY

At all 32 sites studied there was a clear relation with pH for both planktonic and epilithic communities when acid- to alkaline-phosphatase activity was expressed as a ratio (figure 2). In lakes with low pH and high aluminium concentrations (> 100 μg l^{-1}), phosphatase activity was low but the ratio of acid- to alkaline-phosphatase activity remained greater than 1 (Carroll

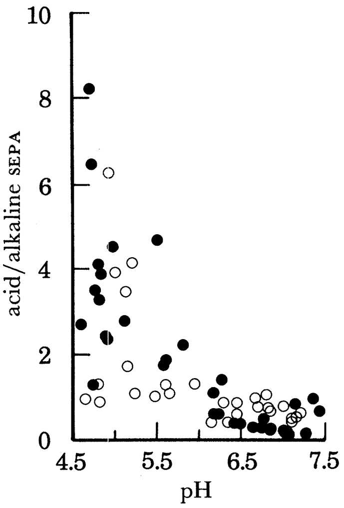

Figure 2. Acid:alkaline phosphatase activity (SEPA) for the 32 lakes plotted as a function of pH; (●), April; (○), September.

& Smith 1989). This differs from the report by Jansson (1981) that phosphatase activity was stimulated by aluminium in an acidified lake, but is consistent with the finding of Mulholland *et al.* (1986) that some of the least acidic sites with lower aluminium levels were the most phosphate limited.

Maximum phosphatase production occurred at the time of lowest SRP levels (figure 3). Phosphatase activity was high (1–5000 times) compared with other published field measurements similarly normalized for chlorophyll-*a* (Pick 1987). Healey & Hendzel (1979) showed that maximum alkaline phosphatase activities for algal cultures were dependent on external calcium concentrations of 100 μmol l^{-1} or more. Loch Whinyeon was the only lake where calcium concentrations exceeded 100 μmol l^{-1} and in Loch Grannoch the mean calcium level was 24.3 μmol l^{-1}, one quarter of that required to maximize alkaline phosphatase activity in cultures.

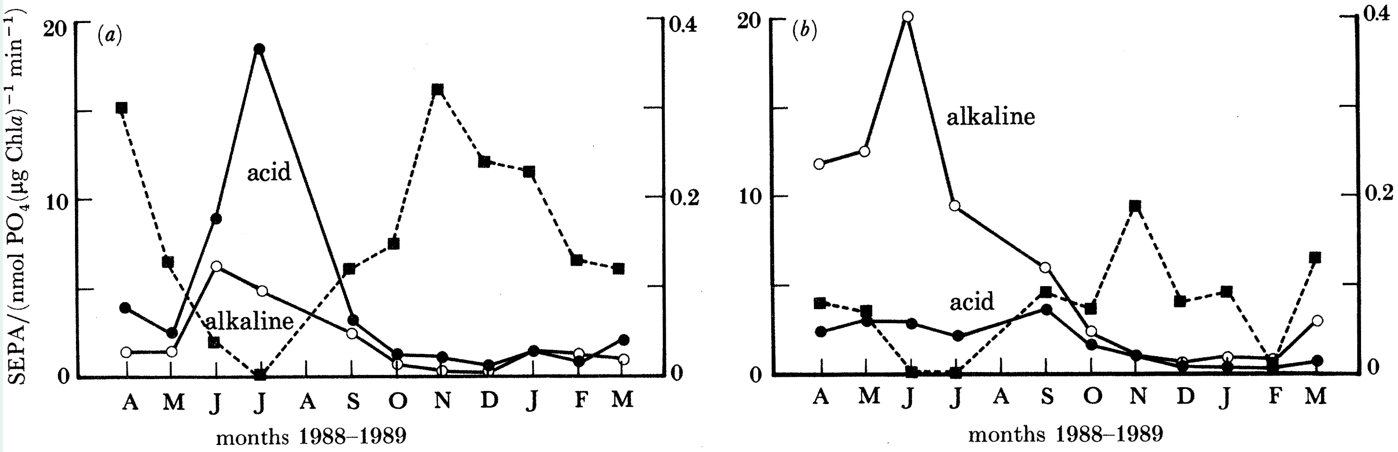

FIGURE 3. Seasonal variation in acid- and alkaline-chlorophyll-*a* specific epilithic phosphatase activity, and in soluble reactive phosphate (SRP) levels (----) for (*a*) Loch Grannoch (mean pH 4.68) and (*b*) Loch Whinyeon (mean pH 6.96); (■), SRP; (●), acid-SEPA; (○), alkaline-SEPA.

The acid phosphatase activity in Loch Grannoch epilithon increased (inducible) as SRP decreased whereas the alkaline phosphatase activity increased only slightly (constitutive) (figure 3*a*). Conversely, the alkaline phosphatase activity in Loch Whinyeon epilithon was inducible whereas the acid phosphatase activity was constitutive (figure 3*b*). Probably because there were several inducible phosphatases operating, the phosphatase data from Loch Howie and Loch Fleet did not show such clear differences. The pH for the phosphatase assays on epilithon from Loch Grannoch and Loch Whinyeon quite closely matched the optimum pH for phosphatase production by those communities and so the assays gave a good estimate of maximum phosphatase production. In the assays on epilithon from Loch Howie and Loch Fleet, for which the pH of the assay was not as closely matched to the pH of the lake, acid- and alkaline-phosphatase production may have been underestimated. The inherent- or ambient-phosphatase activity for all the lakes can be taken as the best estimate of phosphatase production. Maximum inducible phosphatase activity occurred during the summer months May to August, in all four lakes.

Inducible phosphatase activity has a greater affinity for the substrate and is inhibited to a greater extent with increasing SRP concentrations than constitutive phosphatase activity. The epilithon from Loch Grannoch showed inducible acid phosphatase activity (figure 4*a*) and the epilithon from Loch Whinyeon showed inducible alkaline phosphatase activity (figure 4*b*) with orthophosphate inhibition.

The phosphatase enzyme kinetics were examined in more detail at the peak of phosphatase production. Measurements of phosphatase activity with increasing substrate concentration and increasing SRP levels (figure 5) were analysed graphically, according to the methods of Lineweaver & Burk (1934) and Dixon & Webb (1979). At comparable SRP levels the ambient phosphatase activity for epilithon from Loch Whinyeon and Loch Grannoch had similar values. However, the major contribution to ambient phosphatase activity for Loch Grannoch epilithon was acid phosphatase, and for Loch Whinyeon epilithon it was alkaline phosphatase.

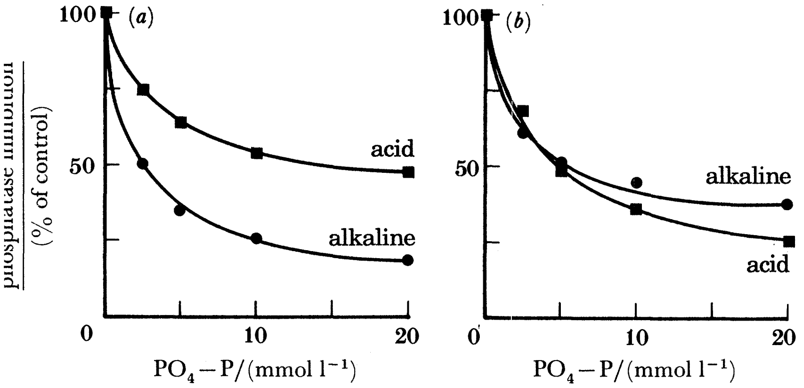

FIGURE 4.

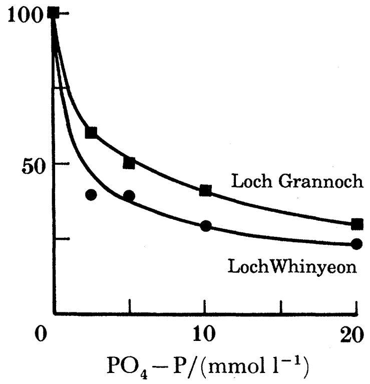

FIGURE 5.

FIGURE 4. The inhibitory effect of increasing inorganic phosphate concentration on acid- and alkaline-SEPA expressed as a fraction of the enzyme activity in the sample containing no added phosphate. The data are means of duplicates; (*a*) Loch Whinyeon, (*b*) Loch Grannoch.

FIGURE 5. Comparison of phosphatase inhibition of ambient SEPA from Loch Whinyeon and Loch Grannoch July samples at the time of maximum phosphatase activity in the lakes. The data are means of duplicates.

It would appear that as lakes become acidified, those epilithic species that depend upon alkaline phosphatase induction to utilize organic phosphorus under conditions of decreasing SRP will be at selective disadvantage. This may well lead to the loss of these particular species from the community through phosphate starvation. Epilithic algae remaining in lakes with pH 5.5 and below have inducible acid phosphatase and are able to hydrolyse organic phosphorus during periods of SRP deficiency. Inducible acid phosphatase may vary in aluminium tolerance from species to species; this is the subject of further investigation.

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