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22. Concordance of Phosphorus Limitation in Lakes: Bacterioplankton, Phytoplankton, Epiphyte–Snail Consumers, and Rooted Macrophytes

Robert E. Moeller, Robert G. Wetzel, and Craig W. Osenberg

The characterization of many unpolluted lakes as aquatic "deserts" (Whittaker, 1975) reflects the well-recognized role of phosphorus and nitrogen in controlling the abundance and growth rates of *planktonic* algae (Schindler, 1974, 1978; Kalff and Knoechel, 1978). Deficiencies or reduced availability of these elements may be less severe in the littoral zone because of marked morphological and physiological adaptations that result in metabolic and community mechanisms for nutrient recycling and retention (Wetzel, 1990a, 1993). As a result, few limiting nutrient studies have been performed in the littoral zone, and many are directed toward specific components (e.g., Fairchild and Everett, 1988; Fairchild and Sherman, 1993).

In Lawrence Lake, located in southwest Michigan, submerged macrophytes are responsible for less than one-quarter of the annual net primary productivity of 180 g C/m^2. The rest originates from phytoplanktonic (16%) and epiphytic (70%) algae (Burkholder and Wetzel, 1989). To evaluate the extent of phosphorus limitation in these three communities as well as the bacterioplankton, we provided local additions of phosphate separately to subsamples of each community. For comparability within the natural light and temperature regimes, all results are for vegetation and microbial communities at a depth of 2 m. Rooted vascular plants grow as deep as 6 m in this 12-m deep lake (Rich et al., 1971; Wetzel et al., 1972; Burkholder and Wetzel, 1989).

Phytoplankton assays consistently demonstrated a deficiency of phosphorus during most of the year (Wetzel, 1981). Addition of 100 µg P/L to samples of

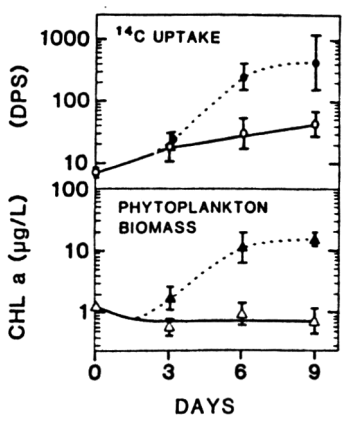

Figure 22.1. Progressive response of summer phytoplankton to P additions. Relative rates of photosynthesis (upper, as relative ^{14}C incorporation, in photosynthesis, as disintegrations per second, DPS) and biomass (lower, as µg chlorphyll *a*/L) for enriched (●, ▲) and control cultures (○, △). Values are means ± 95% CI for n = 7 dates from June 7 to September 13. Phosphate addition was 50 µg P/500 ml Pyrex flask, as K_2HPO_4. Cultures were maintained on a rotating tray at ambient lake temperatures and light (16 h light/8 h dark) (Wetzel, 1981). Chlorophyll *a* was corrected for phaeopigments (Wetzel and Likens, 1991). The ^{14}C activity of filtered algae from short-term labeling assays is a relative measure of photosynthesis: incubation times, volumes filtered, and specific activities were consistent throughout. Similar results were obtained with enrichments by organic phosphorus as β-glycerophosphate (Wetzel, 1981).

lakewater incubated in the laboratory caused consistent increases in phytoplankton biomass and photosynthesis over the next 9 days (Fig. 22.1). Alkaline phosphatase activity of enriched cultures increased at 9 days, after about 50% suppression of activity at 3–6 days, which suggested a return to phosphorus-limited conditions (Wetzel, 1981). The 10-fold stimulation of biomass of phytoplankton demonstrated a strongly limiting role for phosphorus under ambient planktonic conditions, which included abundant dissolved nitrate (nitrate averaged 30 µM and total dissolved P only 0.1–0.3 µM in summer at 2 m). Phytoplankton development in 1984, when the littoral vegetation was studied, was very similar to that in 1976, as had been the condition when the phytoplankton enrichments were conducted. Variations in the annual phytoplanktonic mean chlorophyll *a* concentrations and annual in situ rates of photosynthesis varied less than 15% over an 18-year period of continuous measurement (Wetzel, 1983).

In situ rates of productivity of bacterioplankton from the same sites were determined by incorporation rates of tritiated thymidine into DNA (Wetzel and Likens, 1991). Growth rates were also evaluated by changes in frequency of cell division. Nutrient enrichment experiments with bacterioplankton also indicated an enhanced rate of growth in response to phosphorus (Coveney and Wetzel, 1988, 1992, 1995).

A continuous and localized addition of phosphorus to the natural epiphyte community was accomplished during July–September 1984 in a 2-m-diameter site (plot A) dominated by a submerged sedge, *Scirpus subterminalis* Torr., the dominant macrophyte of the lake (Rich et al., 1971; Wetzel et al., 1972). Vertical rods (41 rods, 1.7 g P/rod positioned on July 13; 5 rods added near center on August 20) coated with resin-encapsulated pellets of calcium phosphate (Sierra Chemical Co., Milpitas, CA) were positioned 60 cm above the sediments throughout the plot; phosphate released from the pellets over 3 months was at a rate of 0.5–1% of the annual loading of phosphorus to the lake. At a nearby site (plot B), rods coated with pellets of mixed fertilizer (supplying N, P, K, S, and Ca) were embedded in the sediment (30 cm below the surface (104 rods, 0.5 g P/rod on June 12) with 60% of the rods 0.5 m from center of plots). Plot B was an attempt to stimulate phosphorus release from growing macrophyte tissue, which is known to be a direct although variable source of phosphorus for overlying epiphytes (Carignan and Kalff, 1982; Moeller et al., 1988; Burkholder and Wetzel, 1990).

A visible increase in epiphytes occurred within 5 days when phosphate was released above the sediment. After 10 weeks, we measured a 40-fold increase of epiphyte biomass at the center of plot A compared with biomass immediately outside the plot (Fig. 22.2). Epiphyte biomass was unchanged by the below-sedimentenrichment at plot B, although additional phosphorus was incorporated by *Scirpus*. Leaf phosphorus increased from 0.14 to 0.51% of dry wt within the plot ($P < .001$). Evidently, the conservative retention of phosphorus by growing macrophytes (Carignan and Kalff, 1982; Moeller et al., 1988; Burkholder and Wetzel, 1990; Wetzel, 1990a) was not significantly relaxed as excess phosphorus accumulated. In plot A, the local addition of phosphorus to the strongly phosphorus-deficient natural epiphyte vegetation led to a massive algal proliferation supported by nitrate, dissolved silica, and other nutrients continuously transported into the unenclosed site.

Epiphyte biomass increased in plot A despite an increase in the intensity of grazing by gastropods (Fig. 22.2). Snail biomass per unit area of lake bottom increased 73% (115 vs. 66 mg dry wt/0.05 m^2; $P < .05$). Snail biomass per unit of macrophyte biomass increased 46% (34 vs. 23 mg dry wt/g organic wt; $P < .1$). The response was attributable to two prosobranchs, *Amnicola limnosa* and *Valvata tricarinata*, which feed on epiphytic microflora and detritus and made up more than 80% of the total snail biomass. Both species live only 1 year and had reproduced before the enrichment started. Snail densities per unit area of lake bottom or unit of macrophyte biomass did not change ($P > .2$), indicating that neither immigration nor reproduction significantly contributed to the response. Instead, individuals of both common species were significantly larger ($P < .005$) within the zone of increased epiphyte biomass (Fig. 22.2), which demonstrated that snail growth rates responded positively to an increased food supply. Subsequent experiments replicated these results for epiphytes and snails in 2 additional years (Osenberg, 1988, 1989). Under natural conditions, therefore, food-limited grazers fed on phosphorus-limited algae. The intensity of grazing may have been sufficient to reduce algal biomass secondarily, as demonstrated elsewhere for

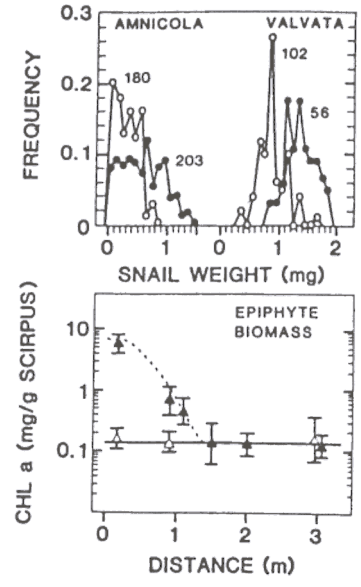

Figure 22.2. Response of epiphytic algae and snails to 10 weeks of P addition. Mean epiphyte biomass (± 95% CI, $n = 7$) increased within water-column enriched sites (▲) but not at sediment-enriched sites (Δ). Weight-frequency distributions of the two most common snails were shifted to larger sizes in the enriched zone (● = enriched zone <m from center of plot, o = control zone 2 m from center; $n = 56$–203 snails). Epiphytes were delicately brushed and rubbed from entire tillers of *Scirpus* collected using SCUBA, then centrifuged, lyophilized, and extracted by grinding in 90% basic acetone. Chlorophyll *a* was corrected for phaeopigment degradation products (Wetzel and Likens, 1991). For epiphytes, seven replicates were collected between September 18 and October 3 from each of nine distances from centers of plots. Snails were collected in mid-October from 0.05-m^2 quadrats of surficial sediment with overlying macrophytes; $n = 4$ enriched quadrats <m from center of plot A and 4 control quadrats >2 m from center. New growth of *Scirpus* was reduced by heavy epiphyte load inside the enrichment zone, so five–six entire stems of *Potamogeton illinoiensis* were also collected both inside and outside the enrichment zone to better compare snail biomass per unit macrophyte. Shell lengths were measured and converted to tissue dry mass based on regressions of length-mass.

littoral (Cattaneo, 1983; Cuker, 1983; Lowe and Hunter, 1988; Osenberg, 1988, 1989; Brönmark, 1989; Lodge et al., 1994) or pelagic algal communities (Carpenter and Kitchell, 1984; Wright and Shapiro, 1984; Lehman and Sandgren, 1985). Our nutrient enrichments were intentionally of sufficiently short duration that phytoplankton and epiphytes could outgrow their grazer populations, which revealed the primary underlying control by phosphorus.

Aquatic macrophytes respond more slowly than microalgae to changing nutrient availability, so we allowed a full year between initial enrichment and final determination of biomass at five sites around the lake. Each site included 10

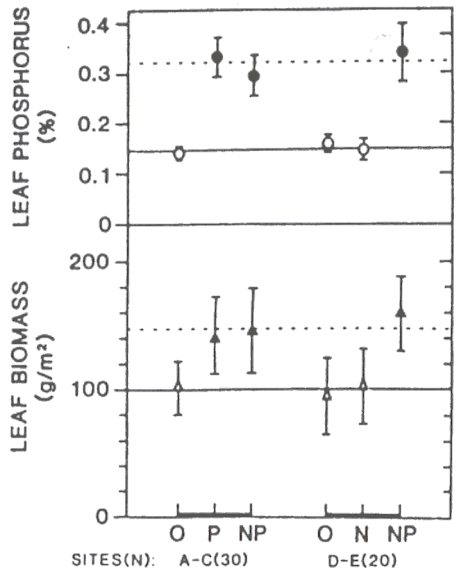

Figure 22.3. Response of the dominant submerged macrophyte *Scirpus subterminalis* to 1 year of P addition. Upper: P concentration in leaf tissue (% carbonate-free dry wt). Lower: Leaf biomass (g dry wt/m^2). Means with 95% CI are compared for three sites with a parallel P-alone enrichment and for two sites with a N-alone enrichment (0 = control, NP = mixed fertilizer, N = +nitrogen, P = +phosphorus). Fertilizer rods were as in Figure 22.2, one rod/quadrat on each date; nitrogen rods carried pellets of ammonium nitrate with some ammonium sulfate (2.6 g N/rod). F-tests for site effects were insignificant (P >.2) for all treatments; thus, quadrats were treated as replicates.

control quadrats 0.008 m^2 in area, 10 quadrats receiving mixed fertilizer in November 1983 and June 1984, and 10 separate quadrats receiving either calcium phosphate or ammonium nitrate on the same dates (Fig. 22.3). Because rooted macrophytes obtain most of their phosphorus from the sediment in nutrient-poor waterbodies (Barko and Smart, 1980; Carignan & Kalff, 1980; Carignan, 1982; Wetzel, 1990b), we avoided a proliferation of epiphytes by inserting fertilizer beneath the sediment surface.

The biomass of the wintergreen leaves of *Scirpus subterminalis* as measured in late October 1984 represents much of their annual net production (Rich et al., 1971; Wetzel et al., 1972; Moeller and Wetzel, unpublished data). This biomass was homogeneous among sites within treatments. The aggregated data (Fig. 22.3) displayed a statistically significant increase caused by mixed fertilizer (P <.01) and by phosphorus alone (P = .05). Nitrogen added without phosphorus had no effect (P >.2). *Potamogeton illinoiensis* Morong made up 30% of total macrophyte biomass at control sites and apparently responded similarly to the mixed fertilizer. The increase from 44 to 60 g dry wt/m^2 for this more heterogeneously distributed plant was not statistically significant (P >.1).

The phosphorus additions effectively saturated plant demand for that element, as demonstrated by the accumulation of excess phosphorus in leaves (Fig. 22.3). The small size of the subsequent increase in biomass (about 50%) probably means that only the lowest leaf phosphorus concentrations encountered in Lawrence Lake (e.g., the 27% of analyses <0.12% of dry wt) truly represent phosphorus deficiency. Concentrations of nitrogen and potassium did not increase after addition of mixed or nitrogen-only fertilizers, which suggested that the enrichments may not have increased their availability. Therefore, it is possible that these elements may also have a limiting role unresolved by the less effective enrichments in these elements.

In the emergent macrophyte wetland surrounding this lake, a dominant species is the common cattail *Typha latifolia* L. Detailed analyses of tissue nutrient analyses and population and growth dynamics in response to fertilization experiments revealed that *T. latifolia* was principally phosphorus-limited in the open calcareous marshes along the eastern side of the lake (Grace and Wetzel, 1981; Dickerman and Wetzel, 1985). Populations of *T. latifolia* under shaded conditions within wooded areas of the western side of the lake were light rather than nutrient-limited (Grace and Wetzel, 1981).

Qualitatively, then, phosphorus operates concordantly as a growth-regulating factor across all biotic components examined. Quantitatively, however, phosphorus is moderately available in the anoxic calcareous sediments of Lawrence Lake that allows modest development of macrovegetation. Experiments with potted *Scirpus* grown in the lake showed that biomass can be more than doubled to 500 g dry wt/m^2 within one growing season; physical disturbance plus mixed fertilizer, not phosphorus alone, was required. The heavily developed epiphytic algal component of the littoral zone shares a phosphorus-deficient medium with the phytoplankton. Rooted macrophytes occupying the same spatial habitat as their epiphytes are functionally detached, by the nutrient geochemistry of sediments, from the aquatic desert of the water column.

Conclusions

We address the question of how pelagial and littoral habitats are integrated into functional lake ecosystems by asking if a single plant nutrient, phosphorus, can play the same biomass-limiting role for littoral submerged macrophytes and their epiphytes, as well as growth of representative consumers, that it does for the plankton. Enrichments of phosphorus to the environment of natural microbial and vegetation communities from a hardwater lake demonstrate that phosphorus can play a concordant growth-regulating role across many aquatic growth-forms. Epiphytic algae as well as phytoplankton and bacterioplankton were strongly limited by phosphorus in summer. Growth and biomass of submerged macrophytes increased significantly to phosphate released within the calcareous littoral sediments, but not to inorganic nitrogen. Phosphorus availability did not suppress growth in the rooted vegetation as severely as it did in the microbial communities. Growth rates of two dominant snail species increased rapidly in response to increased epiphytic algal food supplies under enriched conditions. These results indicate that phosphorus operates uniformly as a growth-regulating factor among many growth-forms within the aqueous portions of the ecosystem.

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