

Report for 2001NY1081B: Microbial indicators: new tools for assessing phosphorus eutrophication

- Conference Proceedings:
 - Wolfe, BE, 2002, Responses of extracellular enzyme activity to phosphorus addition in two calcareous fens, in The 17th annual Cornell Undergraduate Research Forum Proceedings, Spring 2002.
- Other Publications:
 - Wolfe, BE and CT Chapin, April 2002, Responses of extracellular enzyme activity to phosphorus addition in two calcareous fens, Invited seminar, Department of Crop and Soil Science, Cornell University.
 - Chapin, CT; and BE Wolfe, June 2-7, 2002, Microbial enzymatic response to phosphorus addition in two calcareous fens, Poster presentation at the Society of Wetland Scientists, Lake Placid, New York.
- Articles in Refereed Scientific Journals:
 - Chapin, CT and BE Wolfe, Microbial enzymatic response to phosphorus addition in two calcareous fens, Journal of Environmental Quality, in press.

Report Follows:

Problems and Research Objectives:

Wetlands often lie at the interface between terrestrial and aquatic systems and can act effectively to buffer downstream environments from upland nutrient loading. Consequently, changes in wetland biogeochemical processes can serve as early warning signals to the potential degradation of downstream water-quality. In minerotrophic wetlands, those receiving mineral-rich waters, phosphorus (P) is often tied up in geochemical forms by mineral cations. Although minerotrophic systems are often highly buffered, continuous loading of P into a wetland will reduce the soil buffering capacity which then would increase levels of labile P. Measurement of wetland microbial processes and pool sizes may serve to alert management agencies of changes in natural nutrient cycles, and enzyme activity in particular may be a key indicator of P dynamics within a wetland. The measurement of microbial enzymes can be linked to detrital characteristics and rates of decomposition, as these extracellular enzymes serve to hydrolyze complex plant compounds into simple organic molecules that are more readily utilized by the plant community. Hence, as available P becomes more limiting, more enzyme activity is required to acquire organically-bound forms. Indeed, alkaline phosphatase activity (APA) has been reported higher in wetland soils with lower phosphate content, and has been shown to decrease after P loading in a Florida Everglades system.

We tested the efficacy of APA as a possible bio-indicator in two calcareous peatlands that have been subject to experimental phosphorus addition.

Methodology:

In situ fertilization

We have been adding an available form of nitrogen (NH_4NO_3) separately and in combination with different forms phosphorus to two calcareous wetlands in the Finger Lakes Region for the last two years (Fish Fen) and three years (Belle School Fen). The treatments include; labile P (equal molar ratios of $\text{NaH}_2\text{PO}_4 + \text{Na}_2\text{HPO}_4$), Fe-P (FePO_4), Ca-P (CaHPO_4), and an organic form of P (β -glycerophosphate, $\text{C}_3\text{H}_5(\text{OH})\text{PO}_4\text{Na}_2$). Each treatment and the control consists of 5 replicates. Nitrogen is added at $6 \text{ g/m}^2/\text{yr}$ and phosphorus at $2 \text{ g/m}^2/\text{yr}$.

Labile soil nutrients were monitored with ion-exchange resin bags. We used a Hedley fractionation technique modified for wetland soils to estimate pools of mineral and organic P. We measured microbial biomass C, N, and P within each plot one week after the addition of fertilizer in mid-June. Microbial P was determined by the change in quantity of inorganic P removed by extraction following a direct chloroform addition. We are also measured microbial C and N using a separate K_2SO_4 extract following chloroform addition. Microbial C (by difference) was analyzed on a Shimadzu TOC analyzer and microbial N (by difference) was analyzed by digesting the extracts with $\text{K}_2\text{S}_2\text{O}_8$ followed by analysis using a Lachat. We used methods outlined in Tabatabai (1962. Agronomy Monographs No. 9) to determine levels of alkaline phosphatase activity within soil samples from each plot at the time of microbial biomass measurements. This method is a low specificity assay which measures *p*-nitrophenol released by phosphatase activity of soil incubated with sodium *p*-nitrophenol phosphate.

Laboratory incubation

Sub-samples of the top 15 cm of the rhizosphere of Belle School Fen were homogenized into one composite sample following the removal of plant roots and rocks. We placed 30 (100g wet weight peat) samples into acid-washed mason jars and added one of five P additions or distilled deionized water as a control. We had a total of 31 jars (n=5) and one blank (no soil + water). The treatments were as follows: $0.5 \text{ g/m}^2 = 39 \text{ } \mu\text{g P/jar}$, $1 \text{ g/m}^2 = 78 \text{ } \mu\text{g P/jar}$, $2 \text{ g/m}^2 = 156 \text{ } \mu\text{g P/jar}$ (comparable field application rate), $4 \text{ g/m}^2 = 312 \text{ } \mu\text{g P/jar}$, $8 \text{ g/m}^2 = 624 \text{ } \mu\text{g P/jar}$. All P was applied as Na_2HPO_4 in 1 ml applications and mixed thoroughly with the soil. Jars were anaerobically incubated in the dark at room temperature for 28 days. Immediately after the incubation, we sub-sampled the peat for microbial biomass P, C, and APA activity as described above.

Principle Findings and Significance :

With respect to the Belle School Fen field samples, the only fertilization effect was a decrease in APA in N+P (all forms) treatments from that off the control at Belle School Fen ($p = 0.022$, Dunnett's test significant). The lack of an effect with the most labile form of phosphorus suggests that either the phosphorus was made unavailable due to rapid geochemical sorption, or as the response by all N+P (all forms) treatments suggests, the microbial community was not under nutrient constraints, but limited by labile carbon. Our primary concern to wetland integrity in this region is phosphorus eutrophication. However, since we did not add any carbon amendments to the soil, and soils receiving many types of effluents would also receive bio-available carbon, considering carbon as a supplement to phosphorus loading may be important in future studies of possible wetland bio-indicators. In the lab incubation of Belle School Fen soil, we did not alter APA or microbial pool sizes despite a wide range of simulated P loading in these soils. It is interesting to note that we did not see changes in extractable phosphorus until we had added P equivalent to over 1g/m^2 . This 1g/m^2 is the theoretical threshold for P retention in many wetland soils.

Our results indicate that the response of the microbial community depends on limitations within the microbial pool and not on perceived limitation of the ecosystem as it pertains to the plant community.

Notable Achievements :

Ben Wolfe who was a junior this past year, worked extensively on this project to be used as the basis for his undergraduate research project. In conjunction with Carmen Chapin he designed a laboratory experiment to help clarify results from field sampling. His research was presented at the Undergraduate Research Symposium April 15, 2002 as well as s an invited talk at the Department of Crop and Soil Science seminar series April 30, 2002.