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Project Title: ACID PHOSPHATASE ACTIVITY AS AN INDICATOR OF PHOSPHORUS STATUS IN RIPARIAN FOREST SOILS

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Problem and Research Objectives

Increased phosphorus concentrations have accelerated the eutrophication of inland lakes and reservoirs in the northeastern U.S. (Frink 1991). Forested riparian areas are among the best management practices (BMPs) recommended for amelioration of nutrients and other pollutants in runoff (National Research Council 1993). Riparian wetland areas, because of their flatter slopes and high surface roughness, tend to accumulate sediment-bound P that originates from upland areas (Lowrance et al. 1984; Peterjohn and Correll 1984; Vought et al. 1994). In addition to sediment trapping, P removal in riparian areas occurs via plant and microbial uptake and adsoprtion to soil particles (e.g. Lyons et al., 1998). Long-term exposure of riparian areas to elevated P levels can affect the ability of plants, microorganisms, and soil particles to act as sinks for P. There is a need for sensitive, fast, and inexpensive method to evaluate the performance of riparian forest soils with respect to P status. The National Research Council's committee on Long-Range Soil and Water Conservation has identified the long-term effectiveness of riparian zones in nutrient and sediment removal as a major concern (National Research Council 1993).

Soil acid phosphatases catalyze the hydrolysis of organic phosphate esters to *ortho*-phosphate, and thus constitute an important link between biologically unavailable and bioavailable P pools in the soil (Speir and Ross 1978). Acid phosphatase is ubiquitous in soil and is produced by microorganisms in response to low levels of inorganic P, and its production and activity are inhibited by elevated levels of inorganic P. To test the performance of acid phosphatase as an indicator of P status, we evaluated: (1) the sensitivity of acid phosphatase activity to inputs of inorganic P, and inorganic P and N in simulated runoff over the course of a year within a riparian forest area, as a function of landscape position, and (2) the temporal

variation (within and between seasons) of the response of acid phosphatase activity to inorganic P, and inorganic P and inorganic N. We investigated the effects of inorganic P, and P and N, on short and long-term turnover pools of acid phosphatase activity.

This research addresses Research Priority Area A, Watershed/Ecosystem Management, and specifically subsection d, Effective management strategies for riparian zones and wetlands protection and assessment of their role in the retention and recycling of nutrients and toxicants. We evaluated soil acid phosphatase activity as an indicator of the P status of soil, and thus of potential P saturation and reduced biological removal in riparian forest soils. To the best of our knowledge, the only study on phosphatase activity in riparian forest soils is the one published recently by Amador et al. (1997). Thus, the present study is the first one on the potential use of phosphatase activity as an indicator of P status of riparian forest soils. The results of this study will be of use to land and water managers and land-use planners in monitoring the performance of riparian areas and in assessing their role in water quality enhancement.

Methodology

The study was conducted in moderately well drained (MWD) and somewhat poorly drained (SPD) soil within a drainage catena in a forested riparian area of the Peckham Farm research area of the University of Rhode Island in Kingston, RI (approx. 41°30' N, 71°45' W). The soil in the upland portion of the catena is mapped as a Hinckley sandy loam (sandy-skeletal, mixed, mesic Typic Udorthent), whereas the soils in the lower portions of the catena are mapped as Walpole sandy loam (sandy, mixed, mesic Aeric Haplaquept) and Scarboro mucky sandy loam (sandy, mixed, mesic Histic Humaquept) (Soil Survey Staff 1981).

The experiment consisted of three treatments: (I) control amended with distilled deionized water; (II) simulated runoff containing P only; and (III) simulated runoff containing P and N. Both drainage classes received all three treatments. Within each drainage class, each treatment was replicated four times in a randomized block design. Treatment replicates consisted of 1 m X 1 m square plots separated from each other by a 0.5-m wide buffer. Plots received 1 cm of simulated runoff. Application of simulated runoff was made monthly from September through November 1999 and April through October 2000 for a total of eight applications. Simulated runoff was applied evenly to the surface of the plot using a watering can. In all treatments the litter layer was placed on a removable screen that was replaced after the treatments were applied. Nutrients were applied at a rate of 0.75 kg PO_4-P/ha (Treatment II) or 0.75 kg PO_4-P/ha and 3 kg NO_3-N/ha (Treatment III) on every application.

Soil cores (2-cm dia.) were collected below the litter layer (0-5 cm) from each plot in September, October, November, and December 1999 and March, April, May, June, July, August, September, October, and November 2000 approximately four weeks after simulated runoff treatments were applied. In April 2000, soil samples were collected one, two, and four weeks after treatments were applied. Four samples were collected and bulked randomly from within each plot. Soil samples were screened through 2-mm-mesh sieve, placed in sealable plastic bags, and stored in the dark at 4°C for no more than one week. Storage of soil samples for six to eight weeks under these conditions has been shown to have no significant effect on phosphatase

activity (Speir and Ross 1975; Gerritse and van Dijk 1978).

Phosphatase activity was assayed twice for each sampling time: once using field-moist soil samples immediately after collection ("Total" activity, TPASE) and a second time using soil samples that were air-dried for two months ("Recalcitrant" activity, RPASE). The difference between Total and Recalcitrant activity is referred to as "Labile" activity (LPASE). Phosphatase activity was assayed using the method of Tabatabai and Bremner (1969), modified as described by Duxbury and Tate (1981) and Amador et al. (1997) for soils with a high organic matter content. Soil organic matter content was determined by loss-on-ignition for 4 h at 550°C (Karam 1993). Soil moisture content was determined gravimetrically at 105°C (Parent and Caron 1993). Soil pH was measured using a 1:10 soil/water (wt:vol) ratio and a pH meter (Hendershot et al. 1993). The amount of bicarbonate-extractable inorganic P was determined colorimetrically (Alpkem 1986) after extraction of soil (Olsen and Sommers 1982).

Principal Findings and Significance

Total phosphatase activity (TPASE) was higher in MWD than in SPD soil throughout the course of the study (Fig. 1). Both MWD and SPD soils exhibited the highest levels of TPASE in April and May of 2000, with minima apparent in October of 1999 and in July of 2000. No statistically significant differences were observed in TPASE activity between the control treatment and treatments that received either P or N + P applications on any of the sampling dates in either soil. Temporal trends in TPASE were identical for all three treatments in both MWD and SPD soil.

Levels of bicarbonate-extractable phosphate (Fig. 2) were significantly higher in MWD than in SPD soil on all sampling dates. The highest levels of phosphate were observed in June of 2000 and the lowest levels in April of 2000 in both MWD and SPD soils. No statistically significant differences were observed in levels of phosphate between control treatments and treatments receiving either P or N + P applications on any of the sampling dates in either soil. Temporal trends in phosphate levels were identical for all three treatments in both MWD and SPD soil.

Recalcitrant phosphatase activity (RPASE) was higher in SPD than in MWD soil throughout the course of the study (Fig. 3). RPASE followed the same temporal trend in MWD and SPD soil, with the highest levels of activity observed in April of 2000 and the lowest levels in October 1999 and July 2000. No statistically significant differences in RPASE were observed between the control treatment and treatments receiving either P or N + P on any sampling date for either soil. Temporal trends in RPASE were identical for all three treatments in both MWD and SPD soil.

Labile phosphatase activity (LPASE) was higher in MWD than in SPD soil on most sampling dates, particularly after December 1999 (Fig. 4). LPASE appeared to have a different temporal trend in MWD and SPD soil. LPASE maxima in SPD soil were apparent in October 1999 and April and July 2000, with the lowest levels observed in December 1999. By contrast, in MWD soil LPASE activity was highest in May 2000 and lowest in October 1999. There were

no statistically significant differences in LPASE between the control treatment and treatments containing P or N + P in either MWD or SPD soil. Temporal trends in LPASE activity were similar for all three treatments within a particular soil.

None of the phosphatase activities tested appeared to respond to additions of either P or N+P at any time during the course of a year. The levels of P and N+P applied are on the order of those found in runoff waters in the Northeastern U.S. Thus, it appears that phosphatase activity may not be a good indicator of P pollution at the levels normally found in runoff in this region. Examination of the levels of bicarbonate-extractable phosphate shows that additions of either P or N+P had no significant effect on P levels in either soil. The absence of an effect of either P or N + P inputs on soil P levels can be attributable to rapid uptake by both plants and microorganisms and/or complexation by iron and aluminum oxides in the soil. The latter mechanism would be expected to be more important in MWD soil because of the presence of higher levels of metal oxides in these soils (e.g. Lyons et al., 1998).

The absence of enhanced levels of P in these soils may explain the lack of response in phosphatase levels to nutrient additions. Phosphatase production is halted by the presence of elevated levels of phosphate in the soil. Since nutrient additions failed to increase P levels, phosphatase levels -- especially those for the total and labile enzyme activity – would not be expected to decrease. The absence of a response to nutrient inputs by recalcitrant activity was not unexpected, since this activity is attributable to enzyme that is complexed by soil particles and thus not subject to metabolic control by either microbial or plant cells.

In conclusion, total, labile, and recalcitrant phosphatase activity in soil from a riparian forest failed to respond to P and N and P inputs at levels commonly found in runoff in southern New England. As such, it appears that phosphatase activity may not be a good indicator of P pollution in these landscape features.

Resulting Publications

Savin, M. C., H. Taylor, J. H. Görres, and J. A. Amador. 2000. Seasonal variation in acid phosphatase activity as a function of landscape position and nutrient inputs. *Agronomy Abstracts* **92:** 391.

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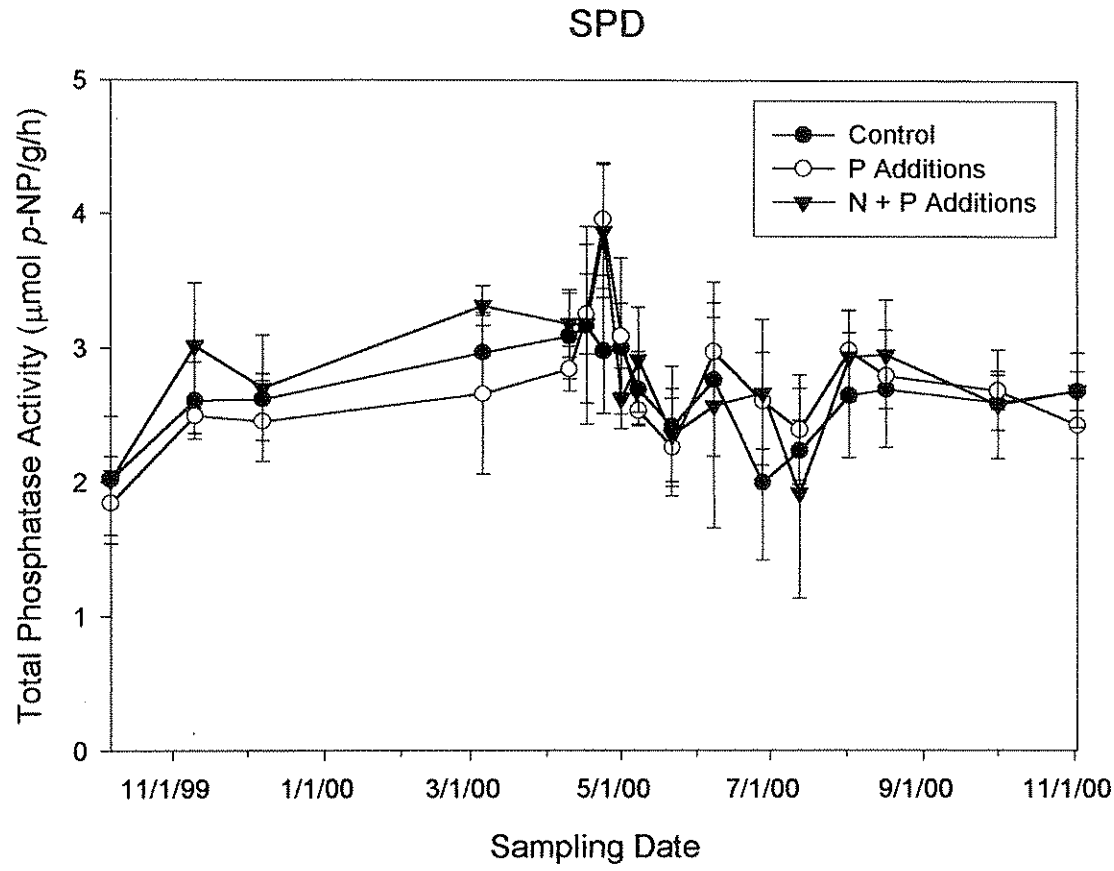
Application of Results

The data collected in this study willbe of use to state and federal agencies charged with monitoring and enhancing water quality (e.g. RIDEM and USEPA). It may also be of use to land managers involved in making decisions about landscape features and their importance of riparian areas to water quality. Our resultsindicate that the threshold for a statistically significant response of soil phosphatase activity to either P or N + P additions has not been reached. Higher levels of P may be required to saturate the capacity of these ecosystems to take up P. These data will be used to establish a threshold for response of phosphatase activity to P inputs in these soils.

Fig. 1


SPD
Total Phosphatase Activity (μmol p-NP/g/h)
0
1
2
3
4
5
11/1/99
1/1/00
3/1/00
5/1/00
7/1/00
9/1/00
11/1/00
Sampling Date
Control
P Additions
N + P Additions

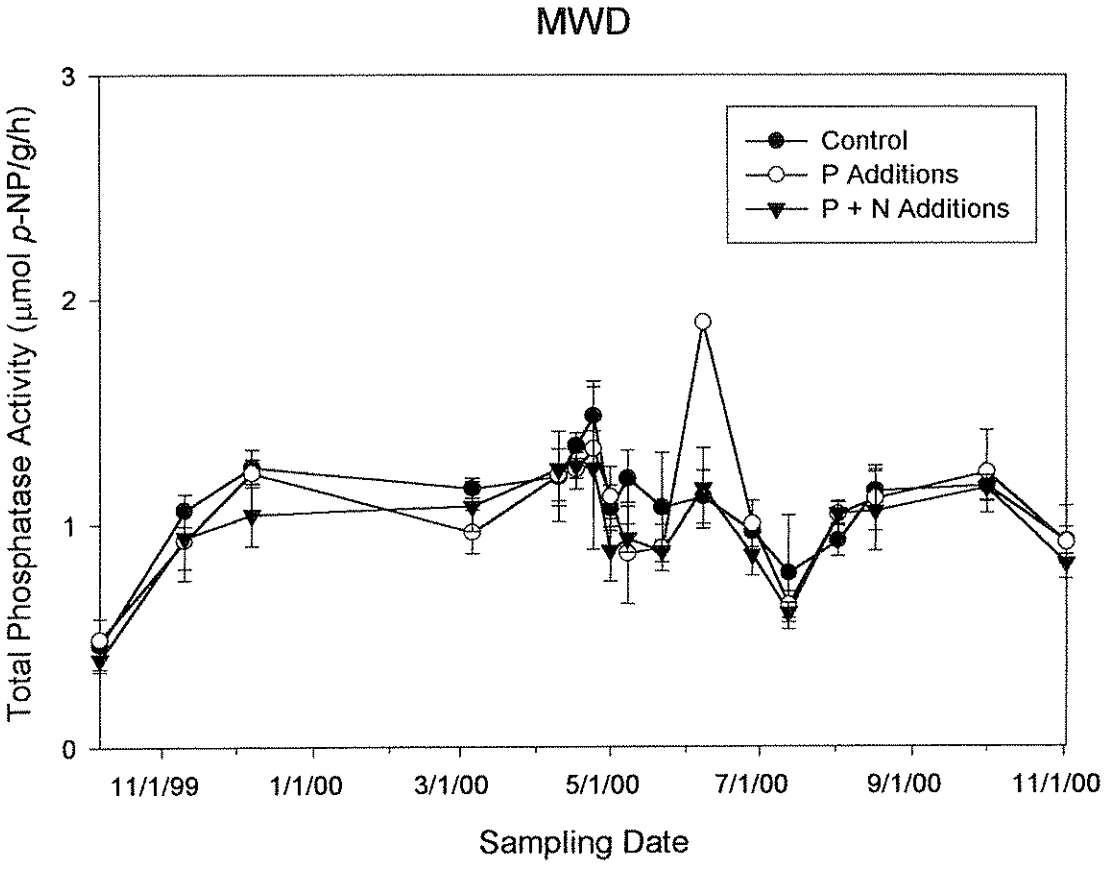
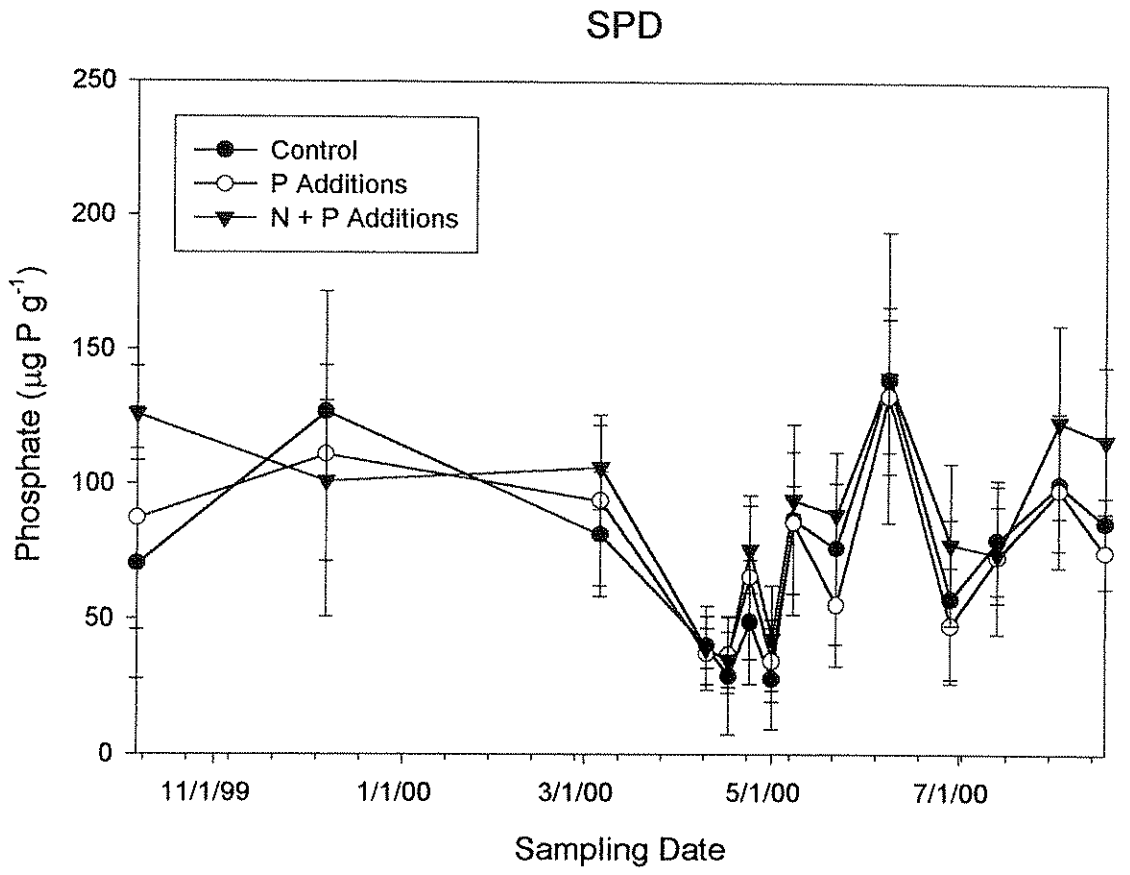

MWD
Total Phosphatase Activity (μmol p-NP/g/h)
0
1
2
3
11/1/99
1/1/00
3/1/00
5/1/00
7/1/00
9/1/00
11/1/00
Sampling Date
Control
P Additions
P + N Additions

Fig. 2


SPD
Control
P Additions
N + P Additions
Phosphate (µg P g-1)
250
200
150
100
50
0
11/1/99
1/1/00
3/1/00
5/1/00
7/1/00
Sampling Date

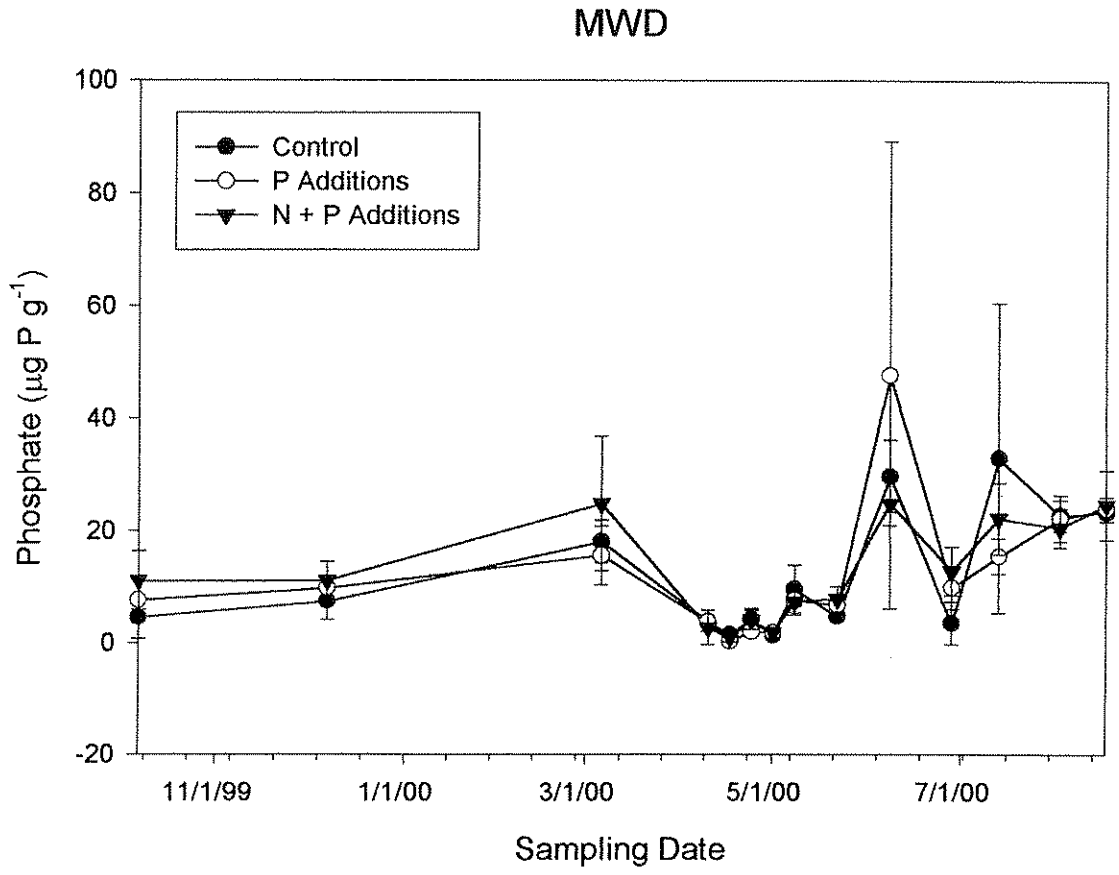
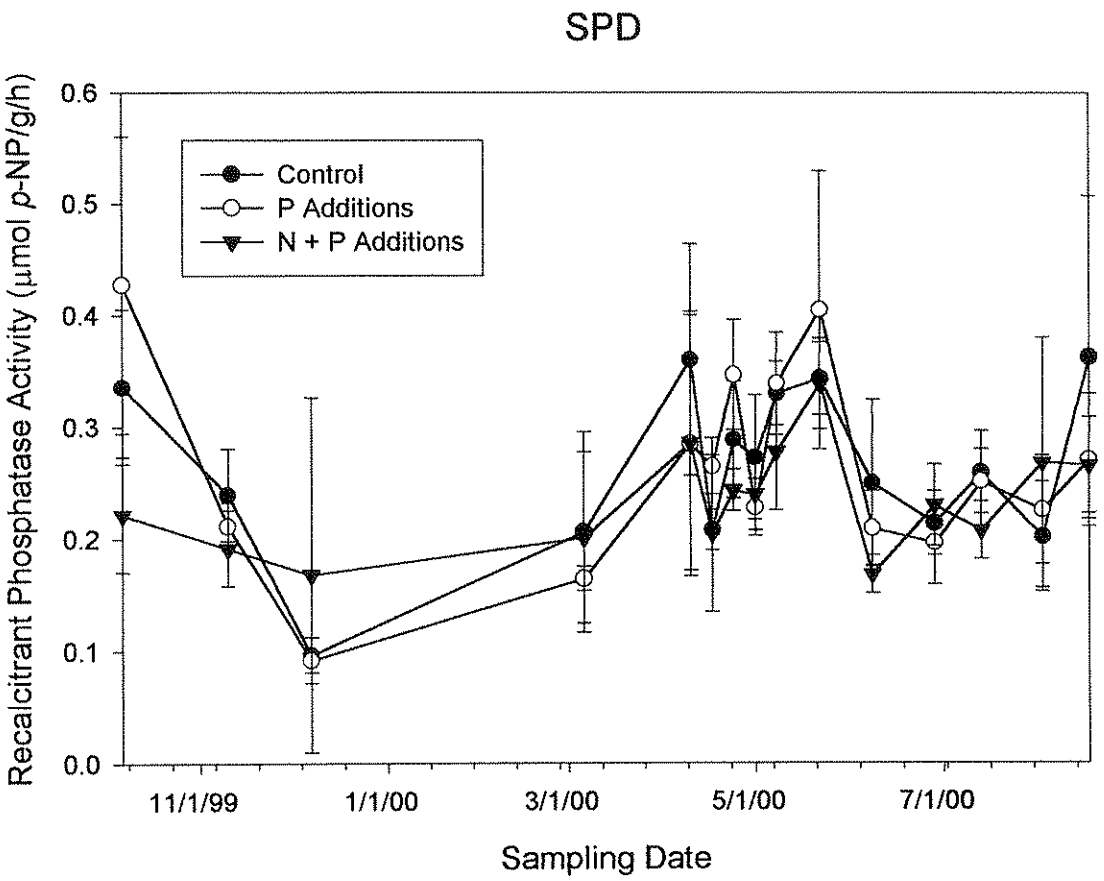
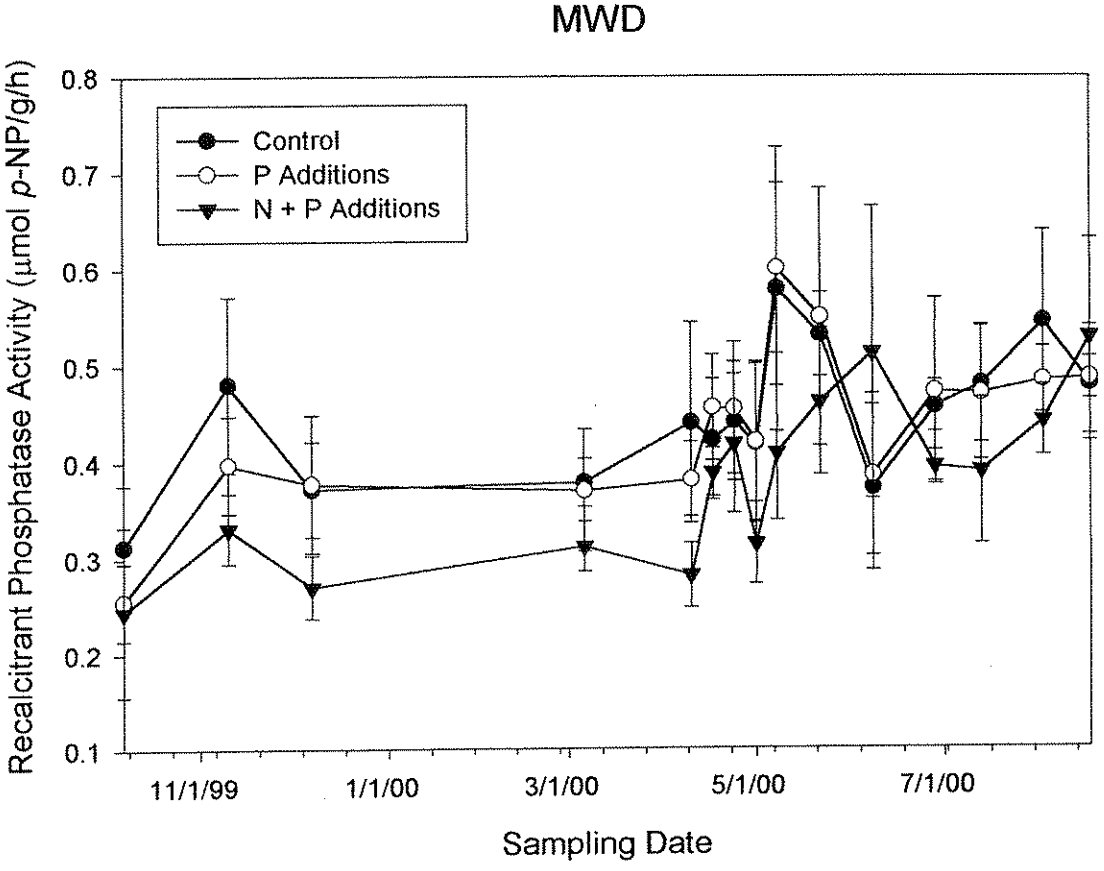

MWD
Control
P Additions
N + P Additions
Phosphate (µg P g-1)
100
80
60
40
20
0
-20
11/1/99
1/1/00
3/1/00
5/1/00
7/1/00
Sampling Date

Fig. 3


SPD
Recalcitrant Phosphatase Activity (µmol p-NP/g/h)
0.0
0.1
0.2
0.3
0.4
0.5
0.6
Control
P Additions
N + P Additions
11/1/99
1/1/00
3/1/00
5/1/00
7/1/00
Sampling Date


MWD
Recalcitrant Phosphatase Activity (µmol p-NP/g/h)
0.1
0.2
0.3
0.4
0.5
0.6
0.7
0.8
Control
P Additions
N + P Additions
11/1/99
1/1/00
3/1/00
5/1/00
7/1/00
Sampling Date