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SHORT COMMUNICATION

Effects of long-term grassland management on the chemical nature and bioavailability of soil phosphorus

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Abstract Relationships between the relative solubility of soil phosphorus (P) and short-term plant P uptake were investigated using soils obtained from a field trial that had been maintained under contrasting mowing regimen (no mowing, mowing with clippings left, mowing with clippings removed) for 15 years. In a glasshouse pot experiment, P uptake by red clover and Italian ryegrass was found to be 40% lower for the clippings removed treatment compared with the no mowing treatment, which was consistent with the fact that concentrations of readily extracted inorganic P were 42% lower in the clippings removed treatment soil. However, P uptake was 51–54% higher for the clippings left treatment soil compared with no mowing, despite the fact that levels of readily extracted soil inorganic P were similar in both treatments. This indicated that biological and biochemical processes associated with enhanced mineralisation of organic P and turnover of P through the microbial biomass made a greater contribution to increased plant P uptake in the clippings left soil compared with the other treatments. These findings highlight the importance of soil biological processes in determining the P nutrition and productivity of managed grasslands.

Keywords Soil phosphorus fractionation · Bioavailability · *Trifolium pratense* · *Lolium multiflorum*

Introduction

Phosphorus (P) is an essential plant and animal nutrient and continued inputs of P are required to maintain and increase the productivity of managed ecosystems (Condron 2004). Phosphorus is a non-renewable resource, and recent and predicted future increases in the cost of P fertilisers have been linked to increased demand and declining reserves of readily available phosphate rock (Cordell et al. 2009).

Phosphorus is present in inorganic and organic forms in soil, and in many soils, the organic fraction accounts for up to half the total P (Condron et al. 2005). Plants and microbes can only obtain inorganic P from soil solution, which contains very low concentrations of P at any one time and must be replenished with time from soil mineral and biological P reserves. Accordingly, the fate and bioavailability of native and applied P in the soil–plant system is mainly governed by a combination of chemical (adsorption–desorption, precipitation–dissolution) and biological–biochemical (immobilisation–mineralisation) processes (Frossard et al. 2000).

The assessment and quantification of bioavailable P in soil has been the subject of extensive research for many years. This includes the development and application of simple chemical extraction methods for determining the bioavailable or 'labile' pool of inorganic P in soil. Examples of these methods are the widely used Olsen P (0.5 M sodium bicarbonate) and Mehlich P (Mehlich-3: 0.015 M ammonium fluoride/0.2 M acetic acid/0.25 M ammonium nitrate/0.013 M nitric acid/0.001 M ethylenediaminetetraacetic acid) soil tests, which are designed to desorb and dissolve the fraction of soil inorganic P which may be released to soil solution to meet plant demands over a growing season (e.g. 3–4 months) (Beegle 2005). However, while such soil tests have been found to be useful general indices of relative P fertility and are used to determine P fertiliser requirement, their accuracy is open to question in many soils due to the fact that they take little account of the contribution to short-term plant P requirements from organic and microbial P pools. A more detailed understanding of P dynamics and bioavailability in natural and managed ecosystems has been

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gained from the development and application of fractionation schemes for soil inorganic, organic and microbial P based on sequential extraction with a series of neutral, alkaline and acid reagents (e.g. Sanyal and DeDatta 1992; Cross and Schlesinger 1995; Pierzynski et al. 2005; Negassa and Leinweber 2009). However, for organic P in particular relationships between chemical form as determined by sequential extraction and plant availability over different time scales are poorly understood (Magid et al. 1996; Condron et al. 2005; McDowell et al. 2008; Condron and Newman 2011).

The objective of the research described in this manuscript was to assess how differences in soil P availability determined by chemical extraction relate to plant P uptake. This involved growing contrasting plant species (red clover, Italian ryegrass) in soils taken from different treatments included in a long-term field trial.

Materials and methods

Field trial

In September 1994, a field trial was established at Lincoln University, New Zealand (S 43°38′51, E 172°28′05) on a Wakanui silt loam soil (Mottled Immature Pallic [NZ]; Udic Ustochrept [USDA]) to investigate the impacts of grassland management on soil properties, plant diversity and insect ecology. The trial area had been under mixed cropping for many years and the P fertility of the site at establishment was considered ‘medium’ (Olsen P – 28 mg Pkg^{-1}) for lowland agricultural soils in New Zealand. The site was cultivated and sown with a mixture of red clover (*Trifolium pratense* L. cv. Pawera), white clover (*Trifolium repens* L. cv. Tahora), perennial ryegrass (*Lolium Perenne* L.) and cocksfoot (*Dactylis glomerata* L. cv. Kahu). Treatments were established on 5×5 m plots arranged in randomised blocks with four replicates per treatment. The trial included the following three treatments: no mowing, mowing with clippings left, and mowing with clippings removed. Mowing was carried out when the sward reached a height of approximately 20 cm (five to six times per annum), and the trial was not grazed and did not receive any fertiliser or irrigation.

Soil sampling and analysis

After almost 15 years (April 2009), 20 soil cores (0–7.5 cm) were taken from each replicate plot of the no mowing, clippings left and clippings removed treatments and bulked together. They were then sieved <4 mm to remove plant debris for use in the glasshouse pot experiment described below. Subsamples were dried in a forced air oven for 72 h and ground <2 mm. Fresh soil was used to determine concentrations of microbial biomass carbon C and N, while dried soil was used for pH, total C and total N analyses (Blakemore et al. 1987; Carter 1993). Fractionation of inorganic P (Pi) and organic P (Po) was carried on dried soil using the method described by Chen et al. (2000). This involved sequential extraction of soil with 1 M sodium chloride (NC-Pi), 0.5 M sodium bicarbonate (pH 8.5) (B-Pi, B-Po), 0.1 M sodium hydroxide (N1-Pi, N1-Po), 1 M hydrochloric acid (H-Pi), 0.1 M sodium hydroxide (N2-Pi, N2-Po), followed by ignition-1 M hydrochloric acid extraction (RES-P). Total soil P was determined as the sum of total P in the NC, B, N1, H, N2 and RES P fractions.

Glasshouse pot experiment

Phosphorus uptake by red clover *(T. pratense* L. cv. Pawera) and Italian ryegrass *(Lolium multiflorum* L. cv. Grasslands Moata) was determined for soil taken from each of the three field trial treatments (no mowing, clippings left, clippings removed) in a pot experiment over 5 months (June–October). Each replicate ‘pot’ was comprised of six plastic cells (30 mm diameter, 30 mm deep) which each contained one clover or ryegrass plant (i.e. six plants per replicate) grown from seed. Four replicates of each plant species–soil combination were arranged in randomised blocks (2 plant species×3 soils×4 replicates=24 pots) in a glasshouse (minimum temperate 10°C, maximum temperate 25°C). Water was added to maintain soil moisture at >80% of water-holding capacity. Above-ground biomass was harvested on four occasions over 5 months, and the plant material from each harvest was dried at 60°C for 48 h and weighed. Ammonium sulphate solution was applied to each cell after each harvest at a rate equivalent to 40 kg ha^{-1} of N and sulphur (total 160 kg N and S ha^{-1}) to confine the agronomic

Table 1 Mean data for selected properties determined for soils taken from the different grassland management treatments after 15 years (*n*=4; LSD 0.05= least significant difference at 5% probability)

	pH	Total C (%)	Total N (%)	Total C/N	Microbial C (mg kg^{-1})	Microbial N (mg kg^{-1})	Microbial C/N
No mowing	5.7	3.43	0.29	11.8	668	82	8.1
Clippings left	5.8	3.88	0.36	10.8	1,084	144	7.5
Clippings removed	5.6	3.35	0.29	11.6	854	103	8.3
P	NS	0.019	0.001	–	NS	NS	–
LSD 0.05	–	0.364	0.029	–	–	–	–

NS not significant

Table 2 Mean concentrations (in milligrammes per kilogramme) of inorganic P (Pi) and organic P (Po) determined in different fractions (NaCl (NC), bicarbonate (B), NaOH I (N1), HCl (H), NaOH II (N2), residual (RES), total P (sum of fractions)) for soils taken from the different grassland management treatments after 15 years (n=4; LSD 0.05=least significant difference at 5% probability)

	NC-Pi	B-Pi	B-Po	N1-Pi	N1-Po	H-Pi	N2-Pi	N2-Po	RES-P	Total P
No mowing	2.8	74.3	124.5	169.7	355	114.7	31.7	29.7	46.1	949
Clippings left	1.1	68.6	141.9	152.9	389	119.3	31.7	36.0	60.6	1,001
Clippings removed	0.8	30.0	102.2	112.9	355	101.1	30.2	26.7	49.6	809
P	0.011	<0.001	0.05	0.002	NS	NS	NS	NS	0.002	NS
LSD 0.05	1.22	17.56	30.84	25.72	–	–	–	–	6.37	–

NS not significant

response to differences in soil P availability. The dried plant material from each harvest for individual replicates was combined to give the cumulative dry matter yield (grammes per pot). The P concentration of the herbage was determined by acid digestion and inductively coupled plasma optical emission spectrometry, which enabled calculation of P uptake (milligrammes P per pot).

Statistical analysis

Differences in soil properties and agronomic performance between treatments were determined using one-way analysis of variance with least significant differences (LSD) carried out using GenStat v11.

Results

Soil pH, C and N data determined for the different treatments after 15 years are shown in Table 1. While mowing regimen had no effect on soil pH, the retention of clippings significantly increased concentrations of total organic C and N compared with the no mowing and clippings removed treatments. As expected, a similar trend was evident in the microbial C, N and C to N ratio data, although differences between treatments were not significant.

Corresponding data for soil P fractions are presented in Table 2. Concentrations of inorganic P in the various fractions were consistently lower in the clippings removed treatment compared with the no mowing and clippings left treatments. These differences were greater and most significant for the B-Pi and N1-Pi fractions, whereby B-Pi and N1-Pi were 56–60% and 26–33% lower under the clippings removed treatment compared with the other treatments, respectively. Organic P was consistently higher in soil from the clippings left treatment compared with the no mowing and clippings removed treatments, although the differences were only significant for the B-Po fraction compared with clippings removed. Residual soil P was also significantly higher under the clippings left treatment compared with no mowing and clippings removed, and while total P was higher under clippings left (1,001 mg P kg^{-1}) compared with no mowing (949 mg Pkg^{-1}) and clippings removed (809 mg Pkg^{-1}), differences between treatments were not significant.

Agronomic results for dry matter yield and P uptake for red clover and Italian ryegrass grown in soils taken from the different mowing treatments are presented in Table 3. Yield and P uptake were consistently highest for the clippings left treatment and lowest for the clippings removed treatment (except for ryegrass yield). All differences between the clippings left and the other two treatments were significant, while differences in ryegrass yield and P uptake between the no mowing and clippings removed treatments were also significant. Phosphorus uptake for the clippings left treatments were 51–54% higher than the no mowing treatment, while P uptake for the clippings removed treatments were 38–40% lower than the no mowing treatment.

Discussion

The P fertility gradient established between the three contrasting mowing regimen within long-term grassland management field trial provided an ideal and unique template to investigate how soil P extractability–solubility relates to short-term plant P uptake. Thus, the no mowing treatment

Table 3 Mean data for total red clover (RC) and Italian ryegrass (IR) dry matter yield (YLD—grammes per pot) and P uptake (PU—milligrammes P per pot) determined for soils taken from the different grassland management treatments after 15 years (n=4; LSD 0.05=least significant difference at 5% probability)

	RC-YLD	IR-YLD	RC-PU	IR-PU
No mowing	1.333	0.652	1.94	3.51
Clippings left	1.772	1.045	2.94	5.40
Clippings Removed	1.095	0.895	1.17	2.18
P	NS	<0.001	0.002	<0.001
LSD 0.05	–	0.1078	0.772	1.031

NS not significant

was effectively a control, while the clippings left and clippings removed treatments represented 'restorative' and 'depletive' management systems, respectively. In the context of managed grassland ecosystems, the clippings left treatment was akin to grazing, while the clippings removed treatment was similar to continuous hay or silage cropping with minimal nutrient inputs or returns.

By comparing the relative differences in soil P pools between mowing treatments with corresponding P uptake by red clover and Italian ryegrass, it was possible to assess how soil P solubility related to actual plant availability. Based on previous research, it is reasonable to assume that the sum of NC-Pi, B-Pi and N1-Pi fractions represents the potential pool of soil inorganic P that is available to plants over a 3-month period (Chen et al. 2003). This bioavailable P pool was similar for the no mowing (247 mg P kg^{-1}) and clippings left (223 mg P kg^{-1}) treatments, which in turn were significantly higher than the clippings removed treatment (144 mg P kg^{-1}) ($P<0.001$; LSD 0.05=36.50). Compared with no mowing (100%), values for this bioavailable inorganic P pool for the clippings left and removed treatments were 90% and 58%, respectively. Results for relative P uptake by red clover and Italian ryegrass were remarkably consistent for the different mowing treatments, and revealed that plant P uptake for the clippings left and clippings removed treatments were 151–154% and 60–62% compared with no mowing, respectively (Table 3).

These findings indicate that for the clippings removed treatment, there was close agreement between the quantities of bioavailable inorganic P determined by sequential extraction (58%) and plant P uptake (60–62%) relative to the no mowing treatment. Thus, the supply of P to plants in this organic matter and nutrient-depleted soil was primarily determined by desorption of inorganic P adsorbed on iron and aluminium oxide surfaces, with limited contribution from organic and microbial P sources.

Results for the clippings left treatment were markedly different from those observed for the clippings removed treatment. Plant P uptake was 51–54% higher for this treatment compared with no mowing despite the fact that bioavailable inorganic P was actually 10% lower (Tables 2 and 3). The fact that total N was significantly higher (24%) in the clippings left soil compared with the other treatments may be attributed to a combination of enhanced biological N fixation and N retention. Despite this, it is unlikely that enhanced N availability contributed significantly to increased plant P uptake from the clippings left treatment due to the addition of large amounts of soluble fertiliser N (160 kg N ha^{-1}) to all soils in the glasshouse pot experiment.

The enhanced P uptake observed for the clippings left treatment indicated that actual P availability was underestimated by sequential chemical extraction of inorganic P from soil with a combination of neutral and alkali reagents. This, in turn, suggested that a significantly ($P<0.002$ (clover), $P<0.001$ (ryegrass)) greater proportion of the P taken up by plants from the clippings left treatment was derived from organic and biological sources in the soil compared with the no mowing and clippings removed treatments. These sources include mineralisation of abiotic organic P and microbial biomass P turnover and release (Condron et al. 2005; Oberson and Joner 2005). This is consistent with the fact that concentrations of total C, microbial biomass C, B-Po and N1-Po were all higher in clippings left soil compared with other treatments, although the differences were only significant for total C ($P<0.019$) and B-Po ($P<0.05$) between the clippings left and clippings removed treatments (Tables 1 and 2). Thus, B-Po was 38% higher in soil from the clippings left treatment compared with clippings removed.

The increased plant P uptake observed for the clippings left treatment can be mainly attributed to enhanced return of organic matter and associated nutrients compared with no mowing and clippings removed. This concurs with observations that rates of organic P mineralisation and P cycling through the microbial biomass were higher in soils under cropping that received inputs of organic manure compared with mineral fertiliser (Oehl et al. 2001, 2004). Furthermore, enhanced biological P cycling in soil in response to increased organic matter inputs is consistent with data from studies on the impacts of green manure crops on P dynamics. For example, Randhawa et al. (2005) measured gross organic P mineralisation rates for control and green manure-amended soils and found that daily gross organic P mineralisation was five times higher in the green manure soil compared with the control. Further work is underway to investigate the precise mechanisms responsible for enhanced P availability in soil under the clippings left regime compared with no mowing and clippings removed, including using isotope tracers to quantify differences in biological P cycling (Frossard et al. 2011).

In conclusion, the findings of this study demonstrate the importance of maintaining and enhancing organic matter inputs in relation to soil fertility, together with the significant contribution that soil organic and biological P pools play in determining P availability and productivity in managed grassland systems.

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