

# Phosphorus speciation in temperate basaltic grassland soils by solution $^{31}\text{P}$ NMR spectroscopy

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## Summary

Phosphorus (P) speciation in 21 basaltic and four non-basaltic Irish grassland soils was determined by NaOH–EDTA extraction and  $^{31}\text{P}$  NMR spectroscopy. Organic P in basaltic soils ranged between 30 and 697 mg P kg<sup>-1</sup> and consisted of phosphate monoesters (84–100%), DNA (0–16%) and phosphonates (0–5%). Inorganic P was mainly phosphate (83–100%) with small concentrations of pyrophosphate (0–17%). Phosphate monoesters were more important as a proportion of extracted P in basaltic soils, probably because of their greater oxalate-extractable Fe and Al contents. Phosphate monoesters appeared to be strongly associated with non-crystalline Al and increased with total soil P concentration, indicating that they do accumulate in grassland soils. In non-basaltic soils *myo*-inositol hexakisphosphate constituted between 20 and 52% of organic P, while *scyllo*-inositol hexakisphosphate constituted between 12 and 17%. These compounds were not quantified separately in basaltic soils because of poor NMR resolution in the phosphate monoester region, but appeared to represent a considerable proportion of the organic P in most samples. DNA concentrations were greater in basaltic soils compared with non-basaltic soils and were associated with acidic pH and large total C contents. The inability of the Olsen P test to assess effectively the P status of basaltic soils may result from strong phosphate sorption to Fe and Al oxides, inducing plant utilization of soil organic P. Phosphorus nutrient management should account for this to avoid over-application of P and associated financial and environmental costs.

## Introduction

Maintaining the availability of phosphorus (P) to plants whilst minimizing loss to water bodies is key to managing temperate grasslands in an economically and environmentally sustainable manner. This requires information on soil P speciation (Frossard *et al.*, 2000), which can be influenced by soil parent material (McDowell *et al.*, 2005; McDowell & Stewart, 2006). Grassland soils developed on basaltic parent material might therefore be expected to differ in P speciation from those developed on more acid igneous and sedimentary rock types, because of the large Al and Fe content of basaltic soils. Phosphorus is closely associated with Fe and Al in many soils (Karathanasis, 1991; Zhang *et al.*, 2001) and Fe and Al contents can influence P speciation (Turner *et al.*, 2003b).

There is evidence to suggest that P cycling in basaltic soils of north-east Ireland may differ from those developed on other parent materials, because conventional soil P tests, such as the

bicarbonate (Olsen) or calcium chloride extraction, are incapable of assessing their fertilizer requirements (Bailey *et al.*, 2000; Bell *et al.*, 2005b). For example, comparison of extractable soil P and herbage P indices for grass swards revealed that swards with sufficient or even surplus P could occur on soils containing low concentrations of Olsen P (Bell *et al.*, 2005a). This has both agronomic and environmental implications, because under-estimation of plant-available P may lead to excess application of mineral phosphate fertilizer, increasing both costs to farmers and the risk of P transfer to surface waters. Phosphorus loss from agricultural soils to surface waters is a major environmental concern in Ireland and elsewhere (EPA, 2004).

It is possible that organic P, which is abundant in temperate pasture soils (Turner *et al.*, 2003b), is utilized as a P source by grass growing on such soils. Apart from a small fraction of the most labile forms, organic P is not accounted for in standard soil P tests (Bowman & Cole, 1978; Olsen & Sommers, 1982; Coventry *et al.*, 2001). To investigate this, we determined P speciation in 21 temperate basaltic grassland soils from north-east Ireland using NaOH–EDTA extraction and solution  $^{31}\text{P}$

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nuclear magnetic resonance (NMR) spectroscopy. For comparison, four non-basaltic soils from the same region were analysed. By investigating differences in P speciation between basaltic and non-basaltic soils, we hoped to infer possible reasons for the failure of the conventional soil P tests on basaltic soils.

## Materials and methods

### Soil sampling and analysis

Twenty-five acidic grassland soils from north-east Ireland were sampled. Soils in the region are predominantly brown earths (Haplic Cambisols (Eutric, Dystric or Humic)) in moderate to well drained sites, or gleys (Haplic Gleysols (Eutric, Dystric or Humic)) in more poorly drained sites. All fields were under permanent perennial ryegrass used for silage production or grazing. The natural dominant vegetation is temperate deciduous forest, but deforestation and conversion to agriculture occurred on a widespread basis in Ireland from at least 3000 BC (Feehan, 2003).

Topsoil (0–20 cm) was sampled by lifting the sod with a spade over an area of approximately 1 m<sup>2</sup>. Soil was air-dried and ground to pass a 2-mm sieve. P status was determined using five methods: Olsen P, oxalate-extractable P, degree of P saturation (DPS), water-extractable P and total P. Soils were analysed for bicarbonate-extractable phosphate (Olsen P, in mg l<sup>-1</sup> soil), the standard extraction for assessing P plant-availability in agricultural soils of Northern Ireland (MAFF, 1986), using 0.5 M NaHCO<sub>3</sub> at a 1:20 soil:solution ratio, with phosphate detection by molybdate colorimetry (Murphy & Riley, 1962). Bell *et al.* (2005b) found that DPS, based on oxalate extraction of Fe, Al and P, was a better indicator of P status when comparing basaltic soils with those derived from other parent materials. For this reason, DPS was used to assess P status in addition to Olsen P. Oxalate-extractable Fe, Al and P were determined by extraction with acid ammonium oxalate (1:1.3 v/v mixture of 0.2 M oxalic acid and 0.2 M ammonium oxalate) (McKeague & Day, 1966) with detection by inductively-coupled plasma optical-emission spectroscopy (ICP-OES). Phosphorus sorption capacity (PSC) was calculated as:

$$\text{PSC} = \alpha(\text{Al}_{\text{ox}} + \text{Fe}_{\text{ox}}) \quad (1)$$

where  $\alpha$  is a scaling factor of 0.5 and Al<sub>ox</sub> and Fe<sub>ox</sub> are oxalate-extractable Al and Fe, respectively. The DPS was calculated as:

$$\text{DPS} = 100 \left( \frac{\text{P}_{\text{ox}}}{\text{PSC}} \right) \quad (2)$$

where P<sub>ox</sub> is oxalate-extractable P (Maguire *et al.*, 2001a).

Water-extractable P was determined by extracting 5 ml of soil with 50 ml of deionized water in an orbital shaker for 1 hour,

with phosphate detection by molybdate colorimetry. Total P was determined by digestion in a mixture of nitric and perchloric acids according to a modification of the method of Olsen & Sommers (1982). Soil (0.5 g) was heated with 10 ml nitric/perchloric acid (4:1 v/v mixture) in a digestion block for 3 hours at 205°C. Twenty millilitres of 0.2 M HCl was then added and samples were boiled again. Phosphate in the digest was determined by molybdate colorimetry.

Soil pH was determined using a combined glass-calomel electrode in a 1:2.5 soil to deionized water ratio (MAFF, 1986). Total soil (C) was determined by combustion using a Leco CN-2000 elemental analyser (Leco Corporation, St Joseph, MI, USA). In the case of four soils (2, 3, 4 and 23), organic matter content was determined by loss-on-ignition (Avery & Bascomb, 1974) using 10 g of sample heated to 850°C in a muffle furnace. Organic matter was converted to total carbon by multiplying by 0.58 (for two soils, total C was determined using both methods and total C derived from organic matter was found to be 0.91 of total C determined by combustion). Cation-exchange capacity (CEC) was determined by compulsive exchange of Mg from 20 ml of 25 mM MgSO<sub>4</sub> onto 1 g of a Ba-saturated soil sample (Avery & Bascomb, 1974). Magnesium remaining in the solution was determined by atomic absorption spectroscopy (AAS).

Particle size analysis by laser diffraction was carried out on 1–2 g samples of 19 of the soils using a Malvern Mastersizer 2000 particle size diffractometer (Malvern Instruments Ltd, Malvern, UK). Bulk mineralogy was determined by X-ray powder diffraction. Corundum was used as an internal standard for quantitative phase analysis and diffraction patterns were recorded from 2 to 75 °2 $\theta$  using Cobalt K $\alpha$  radiation.

### Solution <sup>31</sup>P NMR spectroscopy

Phosphorus was extracted by shaking 5 g of soil with 100 ml of a solution containing 0.25 M NaOH and 0.05 M EDTA for 16 hours at 20°C (Cade-Menun & Preston, 1996). Although it has not been tested on basaltic soils, this procedure is assumed to extract quantitatively soil organic P (Turner *et al.*, 2005a), because it extracted similar concentrations of soil organic P as other methods considered to provide the most accurate measure of soil organic P (Bowman & Moir, 1993). The extracts were centrifuged at 10 000 g for 30 minutes, and an aliquot was taken for determination of total P by ICP-OES after a 100-fold dilution. The remaining solution was then frozen immediately at –80°C, lyophilized, and ground.

For solution <sup>31</sup>P NMR spectroscopy, each freeze-dried extract (approximately 100 mg) was re-dissolved in 0.1 ml of deuterium oxide and 0.9 ml of a solution containing 1 M NaOH and 0.1 M EDTA, and then transferred to a 5-mm NMR tube. The deuterium oxide provided an NMR signal lock and the NaOH raised the pH to > 13 to ensure consistent chemical shifts and optimum spectral resolution. Inclusion of EDTA in the NMR tube reduces line broadening by chelating free Fe in solution (Turner & Richardson, 2004).

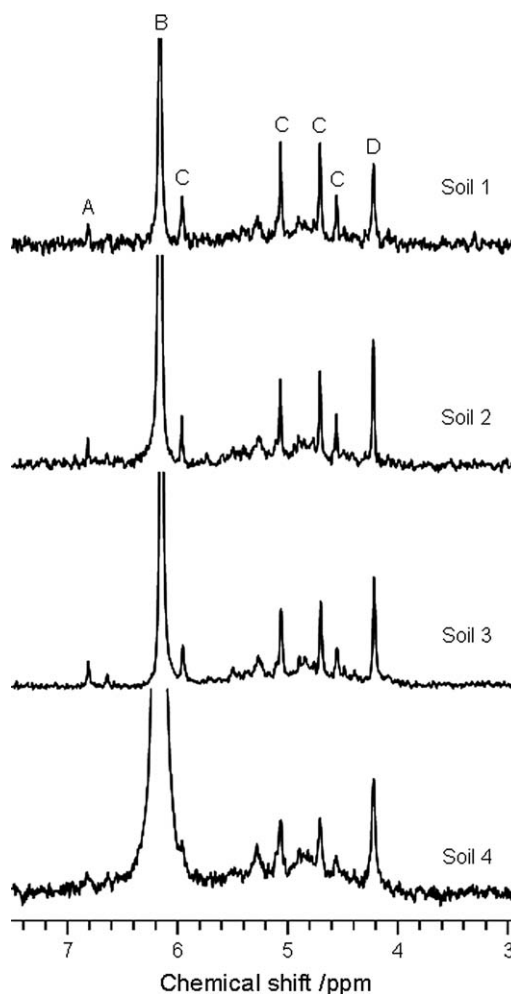
Solution  $^{31}\text{P}$  NMR spectra were obtained using a Bruker Avance DRX 500 MHz spectrometer (Bruker AXS Inc., Madison, WI, USA) operating at 202.456 MHz for  $^{31}\text{P}$  and 500.134 MHz for  $^1\text{H}$ . Samples were analysed using a 6  $\mu\text{s}$  pulse ( $45^\circ$ ), a delay time of 1.0 s, and an acquisition time of 0.2 s. A delay time of 0.8 s was reported previously to be sufficient to obtain quantitative spectra of soil NaOH–EDTA extracts (Cade-Menun *et al.*, 2002). Between 48 000 and 69 000 scans were acquired, depending on the P concentration of the lyophilized extract, and broadband proton decoupling was used for all samples. Chemical shifts of signals were determined in parts per million (ppm) relative to an external standard of 85%  $\text{H}_3\text{PO}_4$ . Signals were assigned to individual P compounds or functional groups based on literature reports (Turner *et al.*, 2003a) and signal areas calculated by integration. Spectra were plotted with a line broadening of 5 Hz, although additional spectra of non-basaltic soils were plotted with a line broadening of 1 Hz to preserve fine resolution in the phosphate monoester region. For these spectra the concentrations of *myo*-inositol hexakisphosphate (phytate) and *scyllo*-inositol hexakisphosphate were determined from the integrals of the associated signals (Figure 1; Turner *et al.*, 2003a; Turner & Richardson, 2004). We did not perform replicate extracts or collect replicate NMR spectra, but variation in triplicate analyses (including extraction and NMR spectroscopy) of mineral soils has been reported to be approximately 2% for total organic P, with greater variation associated with smaller signals such as DNA compared with larger signals such as those from phosphate monoesters (Turner, 2008).

## Results

### Soil properties

Selected soil physical and chemical properties are shown in Table 1 and soil mineralogy is summarized in Table 2. Soil pH ranged between 5.3 and 6.7, which is typical for grassland soils in north-east Ireland (Cruickshank, 1997). Total C ranged from 1.3 to 23.6%. Extractable Fe was 2.4 times greater, on average, and extractable Al was 2.5 times greater, in basaltic soils compared with non-basaltic soils, leading to larger PSCs in basaltic soils. All soils had greater extractable Fe than Al contents, as reported previously for similar soils (Maguire *et al.*, 2001b). Most of the basaltic soils (Soils 5–25) were silt loams, whereas the non-basaltic soils were sandy loams.

Mineralogical composition was mostly typical of Irish soils (Stevens & Jones, 1985). Quartz dominated, but was rare or even absent in some of the basaltic soils (Soils 5, 15 and 23), which were instead dominated by smectite, haematite, gibbsite, kaolinite and pyroxene. The other basaltic soils showed a range of proportions of quartz, with the balance consisting mostly of plagioclase, K-feldspar, muscovite and relatively high levels of smectite. The four non-basaltic soils (1–4) were dominated by quartz with lesser amounts of plagioclase, K-feldspar and clay minerals (dioctahedral clays, illite and smectite).



**Figure 1** Solution  $^{31}\text{P}$  NMR spectra of four non-basaltic soils. The spectra are truncated vertically to show fine resolution in the phosphate monoester region. Signal assignments are as follows: (a) unidentified inositol phosphates; (b) phosphate; (c) the four signals from *myo*-inositol hexakisphosphate, occurring in a 1:2:2:1 ratio; (d) *scyllo*-inositol hexakisphosphate.

### Soil phosphorus

While basaltic and non-basaltic soils had a similar range of total P concentrations (779–1838 and 616–2580  $\text{mg P kg}^{-1}$ , respectively), the range of Olsen P and water-extractable P concentrations was much smaller and more limited for basaltic (2–15  $\text{mg P litre}^{-1}$  and 0.3–3.4  $\text{mg P kg}^{-1}$ , respectively) than non-basaltic soil (13–96  $\text{mg litre}^{-1}$  and 5.4–172.0  $\text{mg kg}^{-1}$ , respectively) (Table 1). Olsen P was below the agronomic optimum of 16–25  $\text{mg litre}^{-1}$  (MAFF, 2000) for all the basaltic soils, but for only one of the non-basaltic soils.

Results of  $^{31}\text{P}$  NMR analysis are summarized in Table 3. Mean recovery of total P in the NaOH–EDTA extract was 48%. Mean recovery in non-basaltic soils was 70%, but only 44% in basaltic soils. As the NaOH–EDTA procedure is designed to extract organic P, the differences probably reflect

**Table 1** Properties of 25 Irish grassland soils

Soil	Bedrock	pH	Texture	Total C /%	Total P /mg P kg <sup>-1</sup>	CEC /cmol <sub>c</sub> kg <sup>-1</sup>	Oxalate extraction				DPS /%	Olsen P /mg l <sup>-1</sup>	Water-P /mg kg <sup>-1</sup>
							Al	Fe	P	PSC			
							/g kg <sup>-1</sup>						
1	Shale	5.3	Sandy loam	2.8	616	12.8	1.66	3.60	0.286	3.42	8.3	13	5.4
2	Shale	5.6	Sandy loam	4.4 <sup>a</sup>	1638	22.9	2.71	8.02	0.801	6.97	11.5	16	14.5
3	Shale	6.0	Sandy loam	4.4 <sup>a</sup>	1488	15.9	2.81	6.18	0.994	5.84	17.0	38	49.8
4	Sandstone	5.8	Sandy loam	6.2 <sup>a</sup>	2580	25.0	2.86	4.95	2.175	5.08	42.8	96	172.0
5	Basalt	6.7	NA	1.3	1159	17.2	1.97	7.58	0.554	4.77	11.6	15	1.4
6	Basalt	6.3	NA	2.7	779	18.0	2.00	8.16	0.284	5.08	5.6	7	0.7
7	Basalt	6.4	Silt loam	3.0	844	33.7	1.75	7.34	0.203	4.54	4.5	9	0.7
8	Basalt	6.2	NA	4.1	1330	52.0	4.18	13.50	0.439	8.84	5.0	9	1.0
9	Basalt	5.9	NA	4.3	1067	31.5	4.23	9.97	0.378	7.10	5.3	4	0.3
10	Basalt	5.9	Silt loam	4.4	1088	34.5	2.13	10.52	0.310	6.32	4.9	6	1.0
11	Basalt	5.9	Silt loam	4.4	1362	45.7	4.96	11.71	0.445	8.34	5.3	8	1.0
12	Basalt	6.0	Silt loam	4.4	1472	47.5	4.18	14.17	0.605	9.18	6.6	10	1.2
13	Basalt	6.0	Silt loam	4.8	1298	45.3	3.91	13.83	0.293	8.87	3.3	5	1.1
14	Basalt	6.3	NA	5.2	1508	44.8	6.36	18.95	0.849	12.66	6.7	15	1.0
15	Basalt	5.9	Silt loam	5.6	1458	56.0	7.78	16.82	0.664	15.99	4.2	3	2.8
16	Basalt	5.9	Silt loam	5.8	1838	56.0	8.53	18.43	0.727	13.48	5.4	2	0.3
17	Basalt	6.1	Silt loam	5.8	1685	51.0	5.21	15.36	0.788	10.28	7.7	13	1.1
18	Basalt	6.0	Silt loam	5.8	1373	48.8	7.32	14.65	0.583	10.99	5.3	5	0.4
19	Basalt	5.8	Silt loam	6.8	1647	45.5	12.36	12.71	0.863	12.54	6.9	13	0.4
20	Basalt	5.9	NA	7.0	1832	47.7	11.20	12.83	0.963	12.01	8.0	5	0.3
21	Basalt	5.9	Sandy loam	8.4	1816	48.3	10.31	9.87	0.955	10.09	9.5	10	0.4
22	Basalt	5.7	Silt loam	9.0	978	49.5	8.07	15.35	0.268	11.71	2.3	4	0.3
23	Basalt	5.5	Sandy loam	10.3 <sup>a</sup>	1698	61.7	14.35	19.89	0.879	22.26	4.0	8	3.4
24	Basalt	6.0	Silt loam	10.7	1585	61.5	6.12	26.46	0.663	16.29	4.1	6	0.5
25	Basalt	5.9	Sandy loam	23.6	1550	76.6	6.05	15.05	0.745	10.55	7.1	8	1.5

CEC, cation exchange capacity; DPS, degree of P saturation; NA, not analysed; PSC, P sorption capacity.

<sup>a</sup>Determined by loss on ignition. All others were determined by Leco total combustion.

the P composition of the soils. Greater levels of unrecovered P in the basaltic soils probably result from greater levels of inorganic P sorbed strongly to Fe and Al oxides. Recovery was negatively correlated with pH in basaltic soils ( $r = -0.65$ ) (Table 4), but positively correlated in non-basaltic soils ( $r = 0.74$ ). Recovery was positively correlated with total P, Olsen P, water-extractable P and DPS in non-basaltic soils ( $r = 0.74-0.93$ ), but much more weakly correlated in basaltic soils ( $r = -0.06-0.55$ ).

Phosphorus species identified in the soil extracts were consistent with those typically found in temperate grassland soils, including phosphate, pyrophosphate, phosphate monoesters, phosphate diesters, phosphonates and phospholipids (McDowell, 2003; Turner *et al.*, 2003b) (Figures 1, 2). Organic P concentrations ranged between 30 and 697 mg P kg<sup>-1</sup> (14–72% of the extracted P) in basaltic soils and between 188 and 592 mg P kg<sup>-1</sup> (22–53% of the extracted P) in non-basaltic soils (Table 3). The organic P pool was dominated by phosphate monoesters in all soils, representing between 84 and 100% of the total organic P in basaltic soils. Basaltic soils had smaller concentrations of phosphate monoesters (30–653 mg P kg<sup>-1</sup>, mean 204 mg P kg<sup>-1</sup>)

than non-basaltic (184–560 mg P kg<sup>-1</sup>, mean 403 mg P kg<sup>-1</sup>), but phosphate monoesters were a greater proportion of the extracted P in basaltic soils (51% compared with 41%). It is possible that some RNA and phospholipids may be hydrolyzed during extraction, contributing to phosphate monoesters measured in the extract.

The phosphate monoester region of the basaltic soil extracts was relatively poorly resolved, with the C-2 phosphate signal of *myo*-inositol hexakisphosphate at approximately 6 ppm obscured by the adjacent broad orthophosphate signal (Figure 3). Therefore, we did not attempt to further separate the phosphate monoester region of basaltic soil extracts. However, the phosphate monoester region of the non-basaltic soils was well resolved, allowing quantification of the inositol phosphates (Table 5): concentrations of *myo*-inositol hexakisphosphate ranged between 97 and 185 mg P kg<sup>-1</sup> (20–52% of total organic P) and *scyllo*-inositol hexakisphosphate ranged between 23 and 99 mg P kg<sup>-1</sup> (12–17% of total organic P). The *myo*:*scyllo* ratio varied between 0.23 and 0.85.

DNA constituted up to 16% of the organic P in basaltic soils, but was detected in only one of the four non-basaltic soils

**Table 2** Mineralogy (%) of 25 Irish grassland soils. Soils 1–4 are non-basaltic and 5–25 are basaltic

Soil	Quartz	Plagioclase	K-feldspar	Muscovite	Pyroxene	Hematite	Ilmenite	Gibbsite	Kaolinite	Illite	Chlorite	Di-smectite	Tri-smectite	Di-clay
1	68.5	9.4	8.8							1.3	1.0			7.9
2	39.5	5.3	1.9							16.1	5.9			24.1
3	40.3	10.4	4.4							6.9	3.8			17.0
4	46.0	9.9	6.6							1.6	2.8			11.9
5						31.6		68.4						
6	75.2	11.6	9.9	0.6								2.8		
7	75.0	11.4	7.3	1.6								4.7		
8	40.5	20.7	11.1	8.8								19.0		
9	69.1	15.5	10.3	3.8								1.3		
10	70.6	15.8	8.3	4.6								0.8		
11	50.8	14.9	9.8	14.3								10.3		
12	60.4	12.9	8.9	3.8								14.1		
13	47.5	26.7	11.5	2.8								11.5		
14	57.8	21.4	8.2	6.8								5.7		
15	3.5	9.1			6.9	4.6	2.3	4.8	13.5			30.1	26.9	
16	10.3	24.7	11.2	38.2								15.7		
17	51.6	20.6	8.8	11.7								7.3		
18	39.0	38.8	6.6	8.5								7.2		
19	48.2	23.6	10.1	13.1								5.0		
20	65.1	21.3	7.4	3.6								2.7		
21	62.5	16.5	12.0	7.4								1.6		
22	44.1	24.5	14.0	12.1								5.2		
23	2.5	10.3			5.0	2.4	2.3		11.3			34.9	21.9	
24	26.9	39.4	13.9	12.9								6.9		
25	25.5	36.0	14.9	14.9								8.6		

Di-clay, undifferentiated dioctahedral clay; Di-smectite, dioctahedral smectite; Tri-smectite, trioctahedral smectite.

(Table 3). Concentrations ranged from trace amounts to 53 mg kg<sup>-1</sup> in basaltic soils, but were always < 10% of total extractable P. On average, DNA constituted 5% of total organic P in basaltic and only 1% in non-basaltic soils. The mono-ester:diester ratio ranged between 5 and 46 for soils in which phosphate diesters were measured.

Phosphonates were only present in measurable quantities in four basaltic soils and three non-basaltic. Concentrations were up to 34 mg P kg<sup>-1</sup> and ≤ 3% of total extractable P (up to 5% of organic P). Phospholipids were found in trace amounts in all soils, probably resulting in part from the rapid degradation of some phospholipids in strong alkali solutions (Turner *et al.*, 2003a).

Inorganic P in the extracts ranged between 112 and 469 mg P kg<sup>-1</sup> (28–86% of total extracted P, or 46% on average) for basaltic soils and between 167 and 1845 mg P kg<sup>-1</sup> (47–78% of total extracted P, or 58% on average) for non-basaltic soils. Phosphate was the most important inorganic P species in all soils, constituting between 83 and 100% of total inorganic P in basaltic soils. Concentrations ranged between 100 and 452 mg P kg<sup>-1</sup> in basaltic soils (43% of total extracted P) and between 143 and 1837 mg P kg<sup>-1</sup> in non-basaltic soils (56% of total P in NaOH–EDTA, on average).

Pyrophosphate was the only other inorganic P species identified in the soil extracts (Table 3, Figures 1, 2). It was present in

all but one soil in concentrations ranging from 7 to 38 mg kg<sup>-1</sup>, constituting up to 7% of extractable P. On average, pyrophosphate made up 3% of extractable P, with little difference between basaltic and non-basaltic soils. Pyrophosphate constituted up to 17% of inorganic P in basaltic soils.

#### *Correlations between soil properties and soil phosphorus*

Correlation coefficients for selected soil properties and soil P in the basaltic soils are shown in Table 4. Note that the statistical significance of correlations has been determined for the 21 basaltic soils, but not for the four non-basaltic soils, on account of the low number of the latter. Therefore, correlations discussed for the non-basaltic soils should be viewed as a possible trend, rather than a confirmed relationship. PSC was positively correlated with total C ( $r = 0.42$ ) (correlation just marginally outside significance at the 0.05 level), but negatively correlated with pH ( $r = -0.66$ ). Total P in NaOH–EDTA extraction was strongly positively correlated with total soil P ( $r = 0.88$ ) and oxalate-extractable P ( $r = 0.90$ ). All three P extractions were positively correlated with PSC ( $r = 0.68$ ,  $r = 0.65$  and  $r = 0.62$ , respectively), but stronger relationships were found with oxalate-extractable Al ( $r = 0.83$ ,  $r = 0.72$  and  $r = 0.74$ ) than Fe ( $r = 0.43$ ,  $r = 0.53$  and  $r = 0.41$ ). Indeed, there was no significant correlation between oxalate-extractable P and

**Table 3** Phosphorus fractions determined by NaOH-EDTA extraction and solution  $^{31}\text{P}$  NMR spectroscopy for 25 grassland soils from north-east Ireland. Values in parentheses are the proportion (%) of the extracted phosphorus. Soils 1-4 are non-basaltic and 5-25 are basaltic

Soil	NaOH-EDTA total P	Inorganic phosphorus		Organic phosphorus			Total organic P
		Phosphate	Pyrophosphate	Phosphate monoester	DNA	Phosphonate	
		/mg P kg <sup>-1</sup> soil					
1	355	143 (40)	23 (7)	184 (52)	ND	4 (1)	188 (53)
2	877	502 (57)	9 (1)	362 (41)	ND	5 (<1)	367 (42)
3	1154	538 (47)	24 (2)	560 (49)	24 (2)	8 (<1)	592 (51)
4	2353	1837 (78)	8 (<1)	508 (22)	ND	ND	508 (22)
5	222	192 (86)	ND	30 (14)	ND	ND	30 (14)
6	311	106 (34)	7 (2)	199 (64)	TR	ND	199 (64)
7	319	100 (31)	21 (7)	188 (59)	10 (3)	ND	198 (62)
8	458	228 (50)	14 (3)	194 (42)	23 (5)	ND	217 (47)
9	528	135 (26)	11 (2)	373 (71)	10 (2)	TR	383 (72)
10	446	163 (37)	13 (3)	258 (58)	12 (3)	ND	270 (60)
11	520	250 (48)	18 (4)	247 (47)	5 (1)	ND	252 (48)
12	594	315 (53)	23 (4)	250 (42)	7 (1)	ND	257 (43)
13	468	171 (37)	32 (7)	236 (51)	29 (6)	TR	265 (57)
14	761	354 (47)	17 (2)	367 (48)	23 (3)	TR	390 (51)
15	653	228 (35)	24 (4)	401 (62)	TR	ND	401 (61)
16	712	195 (27)	22 (3)	479 (67)	15 (2)	2 (<1)	496 (70)
17	801	390 (49)	13 (2)	385 (48)	12 (2)	ND	397 (50)
18	612	288 (47)	12 (2)	289 (47)	22 (4)	TR	311 (51)
19	882	452 (51)	17 (2)	377 (43)	25 (3)	12 (1)	414 (47)
20	1016	303 (30)	16 (2)	639 (63)	24 (2)	34 (3)	697 (69)
21	1043	359 (35)	16 (2)	653 (63)	15 (2)	TR	668 (64)
22	393	144 (37)	16 (4)	196 (50)	37 (9)	ND	233 (59)
23	993	360 (36)	38 (4)	526 (53)	53 (5)	16 (2)	595 (60)
24	685	382 (56)	8 (1)	268 (39)	27 (4)	ND	295 (43)
25	793	416 (52)	25 (3)	315 (40)	37 (5)	ND	352 (44)

ND, not detected; TR, trace.

oxalate-extractable Fe. Oxalate-extractable P was also similar in magnitude to total P in NaOH-EDTA, making up 89% on average (SD = 14) when Soil 5 (with a very small total C content and a haematite and gibbsite mineralogy) was excluded.

Olsen P was positively correlated with total P ( $r = 0.87$ ) and water-extractable P ( $r = 0.99$ ) in non-basaltic soils, but poor non-significant relationships were found in the basaltic soils ( $r = 0.09$  and  $r = 0.08$ , respectively). Similarly, DPS was well correlated with Olsen P ( $r = 0.99$ ) for non-basaltic soils, but a much weaker relationship was found for basaltic soils ( $r = 0.64$ ). Olsen P was poorly and non-significantly correlated with total P in NaOH-EDTA ( $r = 0.06$ ) in basaltic soils.

Phosphate monoester concentration was positively correlated with PSC ( $r = 0.55$ ) (Table 4). A strong relationship was found in both basaltic and non-basaltic soils between phosphate monoester and oxalate-extractable Al ( $r = 0.76$  and  $0.91$ , respectively), whereas no significant correlations were found with Fe ( $r = 0.25$  and  $0.38$ , respectively). Concentrations were positively correlated with P status in both basaltic (total P,  $r = 0.75$ ; oxalate-extractable P,  $r = 0.75$ ) and

non-basaltic soils (total P,  $r = 0.74$ ; oxalate-extractable P,  $r = 0.71$ ) and showed linear relationships (Figure 4). For the regression analysis displayed in Figure 4, Soil 5 occurred as an outlier and was excluded.

Positive correlations were found between phosphate monoester concentration and Olsen or water-extractable P for non-basaltic soils ( $r = 0.64$  and  $r = 0.62$ , respectively) but no significant correlations were found for basaltic soils ( $r = -0.18$  and  $r = 0.04$ , respectively). Phosphate monoester concentration and total C were positively correlated for non-basaltic soils ( $r = 0.77$ ) and for basaltic soils ( $r = 0.56$ ) when Soil 25, which had a total C content more than twice as large as any other soil, was removed.

DNA concentration tended to increase with total C ( $r = 0.64$ ) and decrease with pH ( $r = -0.56$ ) in basaltic soils (Figure 5). Soil 25 occurred as an outlier and was excluded from the regression analysis in Figure 5. Phosphate monoester ( $r = -0.57$ ) was also negatively correlated with pH.

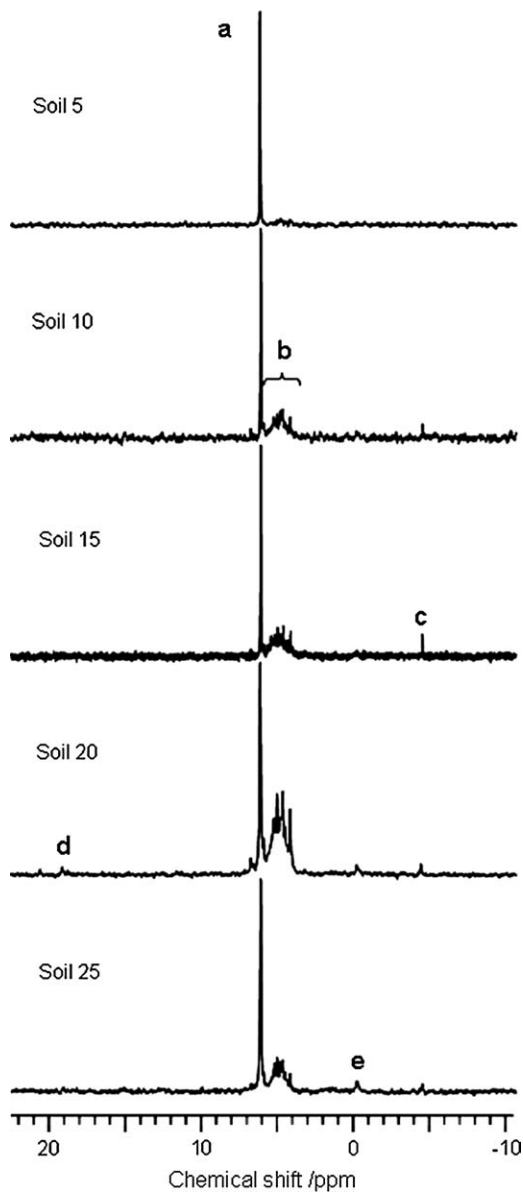
Pyrophosphate was negatively correlated with pH in basaltic soils ( $r = -0.60$ ) and with total C in non-basaltic ( $r = -0.72$ ). Pyrophosphate, as a proportion of total P in NaOH-EDTA,

**Table 4** Correlation coefficients for selected soil properties and P species determined in NaOH-EDTA extracts of basaltic soils ( $n = 21$ ). Values in parentheses are those for proportions of species in the extract

	pH	Total C	CEC	Total P	Olsen P	Water P	Oxal. P	Oxal. Al	Oxal. Fe	PSC	DPS	Total P	P Recovery	NaOH extraction		
														Ortho-phosphate	Pyrophosphate	Total Phosphate
Total C	-0.44*	1.00														
CEC	-0.61**	0.77***	1.00													
Total P	-0.41	0.38	0.66***	1.00												
Olsen P	0.50*	-0.12	-0.23	0.09	1.00											
Water P	-0.21	-0.17	0.28	0.12	0.08	1.00										
Oxal. P	-0.27	0.38	0.49*	0.91***	0.30	0.18	1.00									
Oxal. Al	-0.69***	0.38	0.55**	0.72***	-0.09	0.19	0.74***	1.00								
Oxal. Fe	-0.42	0.41	0.72***	0.53*	-0.19	0.23	0.41	0.43*	1.00							
PSC	-0.66***	0.42	0.71***	0.65***	-0.19	0.48*	0.62**	0.80***	0.82***	1.00						
DPS	0.49*	-0.05	-0.27	0.31	0.64***	-0.12	0.50*	-0.01	-0.38	-0.30	1.00					
NaOH TP	-0.57**	0.48*	0.62**	0.88***	0.06	0.13	0.90***	0.83***	0.43*	0.68***	0.21	1.00				
P Recov.	-0.65***	0.48*	0.49*	0.54**	-0.06	0.08	0.64***	0.73***	0.29	0.56**	-0.03	0.87***	1.00			
Orthophosphate	-0.31	0.55**	0.63***	0.78***	0.44*	0.16 (0.13)	0.83*** (0.11)	0.59**	0.50*	0.56**	0.34	0.79***	0.59**	1.00		
Pyrophosphate	(0.50*)	(0.02)	(-0.09)	(0.02)	(0.65***)			(-0.24)	(0.02)	(-0.14)	(0.52*)	(-0.23)	(-0.49*)			
Monos-ter P	-0.60**	0.35	0.58**	0.33	-0.23	0.56**	0.17	0.43	0.31	0.53*	-0.44*	0.39	0.39	0.19	1.00	
DNA	(-0.21)	(-0.01)	(-0.15)	(-0.30)	(-0.32)	(0.22)	(-0.50*)	(-0.16)	(-0.06)	(-0.04)	(-0.66***)	(-0.25)	(-0.11)			
Phosphate	-0.57**	0.27	0.43	0.75***	-0.18	0.04	0.75***	0.76***	0.25	0.55**	0.14	0.90***	0.83***	0.46*	0.36	1.00
Pyrophosphate	(-0.42)	(-0.10)	(-0.00)	(-0.00)	(-0.60**)	(-0.15)	(-0.05)	(0.19)	(-0.08)	(0.08)	(-0.38)	(0.24)	(0.48*)			
Monos-ter P	-0.56**	0.64***	0.64***	0.33	-0.10	0.18	0.28	0.60**	0.53*	0.63***	-0.35	0.46*	0.47*	0.42	0.53*	0.28
DNA	(-0.39)	(0.41)	(0.43)	(-0.08)	(-0.25)	(-0.06)	(-0.20)	(0.23)	(0.34)	(0.29)	(-0.59**)	(0.00)	(0.10)			
Phosphate	-0.31	0.10	0.13	0.44*	-0.08	0.04	0.51*	0.63***	0.06	0.37	0.16	0.56**	0.48*	0.27	0.20	0.58**
Pyrophosphate	(-0.32)	(0.10)	(0.13)	(0.45*)	(-0.07)	(-0.04)	(0.52*)	(0.64***)	(-0.07)	(0.38)	(0.15)	(0.57**)	(0.48*)			
DNA	-0.35	0.57**	0.66***	0.79***	0.41	0.20	0.83***	0.62**	0.52*	0.60**	0.30	0.81***	0.61**	1.00***	0.26	0.48*
Total	(0.50*)	(0.02)	(-0.08)	(-0.01)	(0.64***)	(0.16)	(0.06)	(-0.27)	(0.02)	(-0.15)	(0.47*)	(-0.27)	(-0.52*)			
Inorganic P	-0.60**	0.32	0.46*	0.75***	-0.19	0.06	0.76***	0.80***	0.28	0.59**	0.11	0.92***	0.85***	0.48*	0.40	0.99***
Total	(-0.50*)	(-0.02)	(0.08)	(0.01)	(-0.64***)	(-0.16)	(-0.06)	(0.27)	(-0.02)	(0.15)	(-0.47*)	(0.27)	(0.52*)			
Organic P																

CEC, cation exchange capacity; DPS, degree of P saturation; Oxal., oxalate extractable; PSC, P sorption capacity.

\*Significant at 0.05 level, \*\*Significant at 0.01 level, \*\*\*Significant at 0.001 level.



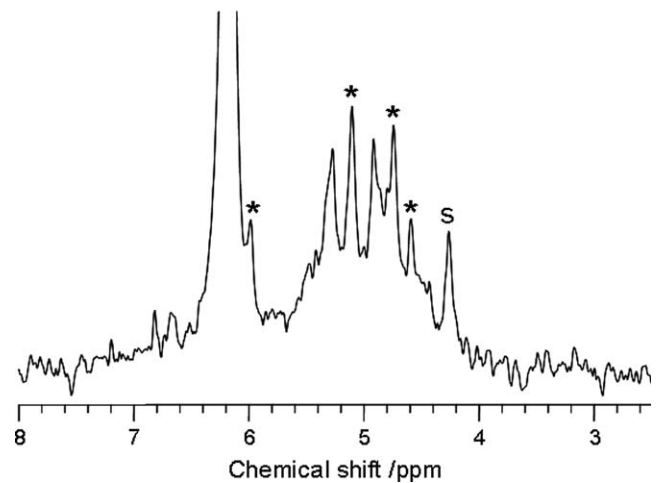
**Figure 2** Solution  $^{31}\text{P}$  NMR spectra of five basaltic soils, selected to cover the range of total C and DPS. Signal assignments are as follows: (a) phosphate; (b) phosphate monoesters; (c) pyrophosphate; (d) phosphonates; (e) DNA.

was not significantly correlated with total P, Olsen P and water-extractable P in the basaltic soils ( $-0.32 < r < 0.22$ ) but was strongly and negatively correlated in the non-basaltic ( $-0.91 < r < -0.60$ ).

## Discussion

### Phosphorus speciation and soil properties

Greater oxalate-extractable Fe and Al concentrations in the basaltic soils can be attributed to their parent material and



**Figure 3** Expanded phosphate monoester region of a NaOH-EDTA extract of a basaltic soil (Soil 25), showing the broad phosphate signal at 6.1 ppm obscuring the small signal from the C-2 phosphate of *myo*-inositol hexakisphosphate at 6.0 ppm. Signals from *myo*-inositol hexakisphosphate (\*) and *scyllo*-inositol hexakisphosphate (S) are indicated.

appear to influence P speciation. Many other studies have found that Fe and Al phases are important for P sorption (Lindsay & Moreno, 1960; Wang *et al.*, 1991; Burkitt *et al.*, 2001; Blake *et al.*, 2003). Extractable Fe and Al concentrations, however, showed little relation to the proportions of Fe- and Al-bearing minerals, suggesting that some of the Fe and Al in the basaltic soils may be in oxalate-insoluble forms. Oxalate dissolves mainly active non-crystalline Fe and Al phases (Fengmao & Yost, 1999), which appear to be more important than crystalline phases in influencing P speciation. This is highlighted by the fact that Soil 5 had a mineralogy of haematite and gibbsite (Table 2), but small oxalate-extractable Fe and Al contents (Table 1) and low phosphate monoester concentration (Table 3). This soil was an outlier in the relationship between monoester concentration and soil P status (Figure 4).

Phosphate monoesters dominated organic P, as is typical of agricultural grassland soils (Condon *et al.*, 1985; Magid *et al.*, 1996; Koopmans *et al.*, 2003). Phosphate monoesters were the dominant P species in NaOH-EDTA extracts of basaltic soils (51% monoester), unlike the non-basaltic soils in this study (41% monoester) and grassland soils of New Zealand (38% monoester) (McDowell *et al.*, 2005). Phosphate monoesters also dominated P in NaOH-EDTA extracts of pasture soils of non-defined parent material from England and Wales (50% monoester) (Turner *et al.*, 2003b). These phosphate monoesters could be derived from plant and microbial residues, animal manure or slurry application (Guggenberger *et al.*, 1996; Koopmans *et al.*, 2003). They are also likely to include the products of alkaline degradation of RNA and some phospholipids (Turner *et al.*, 2003a), which could be substantial in some soils.



**Table 5** Concentrations of inositol hexakisphosphate in NaOH-EDTA extracts of four non-basaltic soils, determined by solution  $^{31}\text{P}$  NMR spectroscopy

Soil	<i>myo</i> -Inositol hexakisphosphate		<i>scyllo</i> -Inositol hexakisphosphate		Total inositol hexakisphosphate		
	/mg P kg <sup>-1</sup>	/% organic P	/mg P kg <sup>-1</sup>	/% organic P	/mg P kg <sup>-1</sup>	/% organic P	<i>scyllo</i> to <i>myo</i> ratio
1	98	52	23	12	121	64	0.23
2	97	27	44	12	141	39	0.45
3	117	20	99	17	216	37	0.85
4	185	36	86	17	271	53	0.46

Because of their large charge density, higher-order phosphate monoesters such as inositol phosphates are more strongly sorbed by metal oxides than orthophosphate (Turrión *et al.*, 2001), which may protect them to a certain extent from phytase enzymes (Lung & Lim, 2006). Increased phosphate monoester concentrations in soils with greater PSC and greater dominance of phosphate monoesters in basaltic soils are consistent with this hypothesis. Turner *et al.* (2003b) found that *myo*-inositol hexakisphosphate was positively correlated with oxalate-extractable Fe and Al in grassland soils from England and Wales. In the Irish soils studied here, phosphate monoesters and total P were more closely associated with oxalate-extractable Al rather than Fe, suggesting that Al species are more important for P sorption. McDowell *et al.* (2005) also found that phosphate monoesters were positively correlated with oxalate-extractable Al in New Zealand grassland soils, while Turner *et al.* (2007) reported that in temperate forest soils in New Zealand inositol phosphate concentrations declined in older, strongly-weathered soils in parallel with a decline in amorphous Al and Fe.

The close relationship between phosphate monoesters and total C, as well as oxalate-extractable Al, suggests that phosphate monoesters may occur as metal oxide-organic matter complexes. Such complexes may act as a sink for P, or as a source supplying the soil solution and thus plant uptake (Pant *et al.*, 1994), possibly through organic matter degradation or accumulation by microbial activity (Evans, 1985). Stability of these complexes may decrease with increasing pH as phosphate monoester concentrations were negatively correlated with pH. This may account for some of the increase in P plant uptake that has been reported to occur after liming (Haynes, 1984). McDowell *et al.* (2005) also found negative correlations with pH and positive correlations with organic C in New Zealand grassland soils. Such correlations may also result, however, from inhibited microbial activity at acidic pH and the accumulation of phosphate monoesters along with organic matter.

Although the phosphate monoester region of spectra of basaltic soil extracts was not separated further because of relatively poor resolution, it seems certain that the extracts included considerable proportions of inositol phosphates, as indicated in Figure 3 (Koopmans *et al.*, 2003; Turner *et al.*, 2003a). Inositol phosphates are important in soil organisms and plants (Magid *et al.*, 1996) and in soil solution and leachate (Evans, 1985;

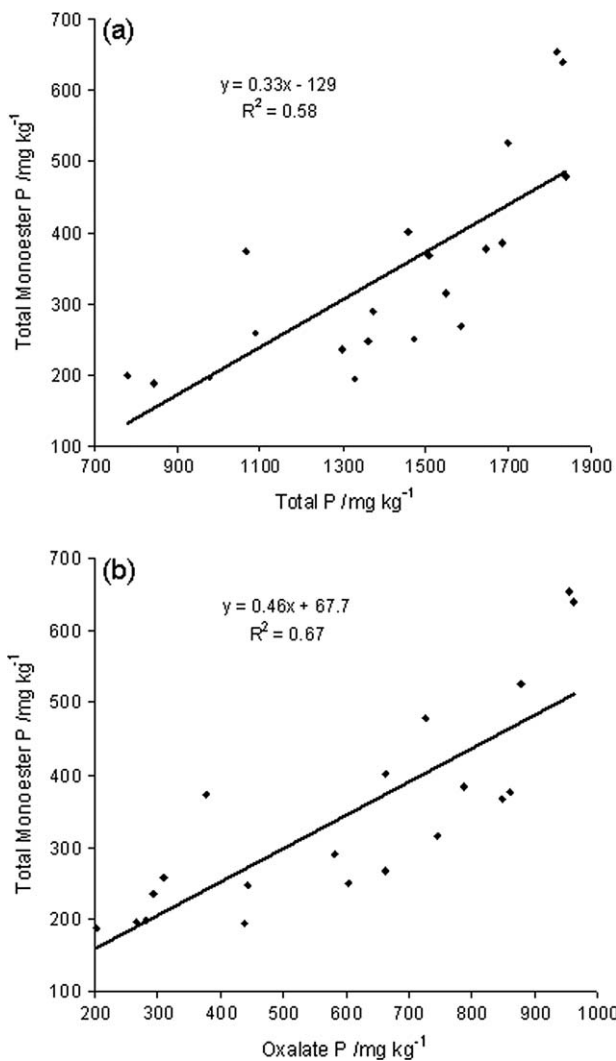
Espinosa *et al.*, 1999). A large proportion of the organic P in the non-basaltic soils was in the form of inositol hexakisphosphate. Indeed, these are some of the largest values so far reported (reviewed in Turner, 2007).

*myo*-Inositol hexakisphosphate concentrations in non-basaltic soils were greater than *scyllo*-inositol hexakisphosphate, as was found for grassland soils in England and Wales (Turner *et al.*, 2005b). However, both stereoisomers of inositol phosphate appear to be more important, quantitatively and proportionally, in the non-basaltic Irish soils than the English and Welsh soils, in which the *myo*-isomer ranged between 26 and 189 mg P kg<sup>-1</sup> (11–35% of organic P) and the *scyllo*-isomer ranged between 11 and 130 mg P kg<sup>-1</sup> (4–14% of organic P) (Turner *et al.*, 2003b; Turner *et al.*, 2005b). *myo*-Inositol hexakisphosphate was correlated with Al + Fe in the British soils (Turner *et al.*, 2003b). The non-basaltic Irish soils tended to be at the upper range of values of oxalate-extractable Al observed in the English and Welsh soils, with a mean concentration of 2509 mg Al kg<sup>-1</sup>, compared with 1644 mg Al kg<sup>-1</sup>. Aluminium oxides may sorb and stabilize greater quantities of inositol hexakisphosphate in the Irish soils. Stronger correlation with Al than Fe in both basaltic and non-basaltic soils suggests that phosphate monoesters are more closely associated with Al, rather than Fe, in Irish soils.

Greater phosphate monoester concentrations in soils with greater P status (for both basaltic and non-basaltic soils) indicate that as more P is added, more P accumulates as phosphate monoesters (Figure 4). Others have also found that monoesters accumulate in grassland pasture soils as a result of long-term P addition (Hawkes *et al.*, 1984; Condon *et al.*, 1985; McDowell & Stewart, 2006).

In terms of DNA content, the soils in our study reflect what might be expected in neutral, well-drained soils (McDowell, 2003). Although many are acidic gleysols, they are not as acid and do not have the extremely poor drainage of the peat soils (Histosols) of the region. Phosphate diesters such as DNA in soil extracts are probably derived from both microbial cells and DNA sorbed to soil (Makarov *et al.*, 2002). Indeed, DNA has been measured in humic acid extracts (Makarov *et al.*, 1996; Mahieu *et al.*, 2002).

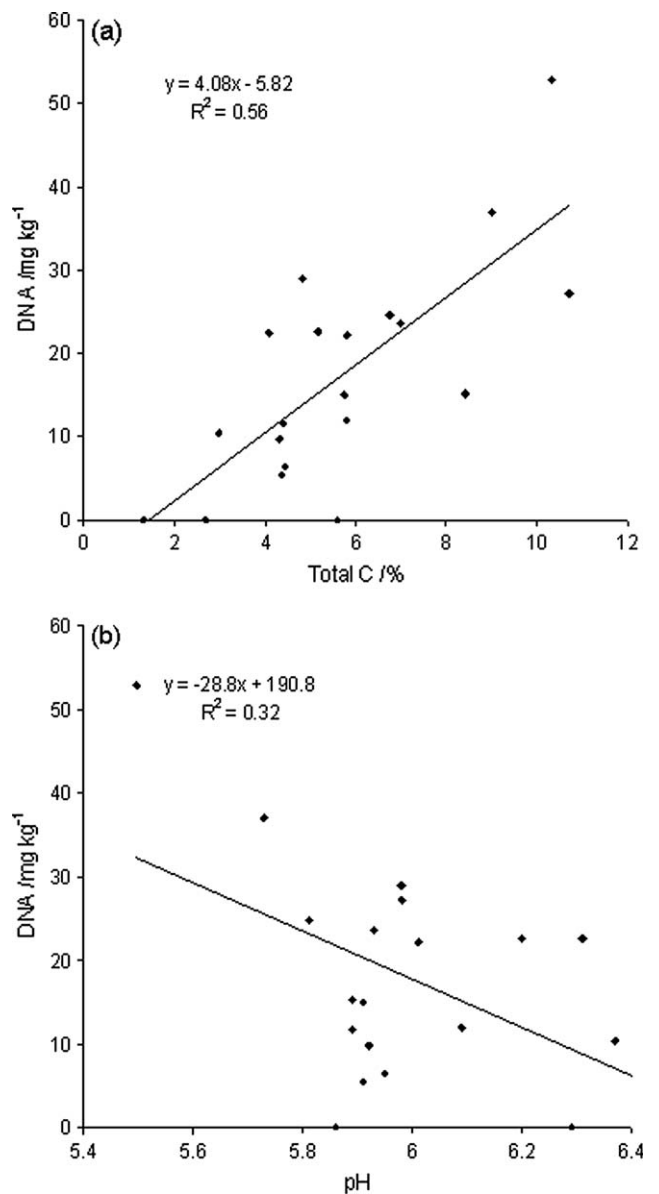
Larger-molecular-weight phosphate diesters such as DNA have a smaller charge density than phosphate monoesters and



**Figure 4** Concentrations of phosphate monoesters in basaltic soils increased with soil P status as (a) total P and (b) oxalate-extractable P. Soil 5, which had a haematite and gibbsite mineralogy, was excluded from this analysis.

are not as strongly sorbed by Fe and Al oxides (Celi & Barberis, 2005). Weak sorption makes phosphate diesters more susceptible to transport and degradation (Turner *et al.*, 2002). They are mineralized more readily than monoesters and the absence of DNA in a soil is often taken as an indicator of conditions favouring microbial activity and mineralization (Condon *et al.*, 1990; Magid *et al.*, 1996; Cade-Menun *et al.*, 2000; Makarov *et al.*, 2002; McDowell, 2003).

Increased DNA with increased total C content and decreased pH is consistent with inhibited microbial activity (Figure 5). The relationship shown in Figure 5 may not be valid for soils with large total C contents, as Soil 25 with a large C content was an outlier and was removed from the analysis. Similarly, McDowell (2003) found that phosphate diesters became more abundant with decreasing soil pH in an English grassland soil. This trend



**Figure 5** DNA concentrations increased with total C content (a) and decreased with increasing pH (b) for basaltic soils. Soil 25, which had a much higher total C content than the other soils, was excluded from this analysis. Soils in which DNA was not detected, or was found in trace amounts only, were assigned a DNA concentration of 0 mg kg<sup>-1</sup>.

may reflect the effect of soil drainage on redox and pH conditions. In contrast, a study of 29 grassland soils from England and Wales found no correlation between DNA and soil pH, although concentrations were strongly correlated with microbial biomass and total C (Turner *et al.*, 2003b). Others have also found correlations with microbial biomass (Rheinheimer *et al.*, 2002). Turner *et al.* (2007) reported recently that DNA appeared to become stabilized by association with organic matter during pedogenesis in temperate forest soils of New

Zealand. It seems, therefore, that DNA accumulates in association with organic matter, but not necessarily under conditions of inhibited microbial activity.

Recently, Makarov *et al.* (2004) showed that DNA can be stabilized in association with clay minerals that protect it from mineralization. Greater prevalence of DNA in the basaltic soils may be because of their finer texture, clay mineral (smectite, kaolinite) and Fe- and Al-oxide content. DNA was positively correlated with both oxalate-extractable Fe and Al. Similarly, McDowell *et al.* (2005) found that phosphate diester was positively correlated with oxalate-extractable Al in New Zealand grassland soils. McDowell *et al.* (2005) also found that phosphate diester concentration was negatively correlated with organic C in New Zealand soils. It would seem that DNA may accumulate in association with both organic and mineral soil fractions, as we have also hypothesized for phosphate monoesters.

The fact that pyrophosphate concentrations changed little and proportions decreased with increasing soil P status (Total P, Olsen P, water-extractable P) in non-basaltic soils, suggests that P does not tend to accumulate significantly as pyrophosphate with P loading. This pattern of distribution suggests that pyrophosphate is unstable and subject to rapid conversion to orthophosphate upon release from microbes. In basaltic soils, however, lack of change in the proportion of pyrophosphate with increasing P status suggests that some pyrophosphate may accumulate, perhaps being protected from conversion to orthophosphate by sorption to Fe and Al oxides. Pyrophosphate is a short-chain polyphosphate used as a storage compound in microbes when inorganic P is plentiful and is associated with microbial activity (Harold, 1966; Cade-Menun *et al.*, 2000). Strong correlations between pyrophosphate and microbial biomass and total C were found in British soils, indicating that pyrophosphate was associated with similar soil properties to DNA (Turner *et al.*, 2003b). This would be consistent with the negative correlation of pyrophosphate with pH found in our study. Turner *et al.* (2007) reported that pyrophosphate appeared to become stabilized by association with organic matter in New Zealand soils.

#### *Implications for P plant availability*

The Olsen extraction is inadequate for assessing P plant availability in basaltic soils, possibly because of plant-available pools of total P that are insoluble in the Olsen extraction. This would explain why all the basaltic soils were below the apparent agronomic optimum Olsen P, despite many having relatively large total P contents (Table 1). The poor relationship between total P or DPS and Olsen P is consistent with this, as is the limited range of Olsen P concentrations for basaltic soils relative to non-basaltic, despite similar ranges in total P. These observations may be attributed to the large Fe and Al contents of the basaltic soils. The Olsen test was designed for calcareous soils in which P solubility may be largely controlled by Ca–P species. In contrast,

P solubility would appear to be largely controlled by Al- and Fe–P associations in these basaltic soils.

Bell *et al.* (2005b) found that DPS was a better indicator of P plant availability on these basaltic soils and established an agronomic optimum DPS of 4.6–7.9%. Therefore, only six basaltic soils were below the agronomic optimum level. Fractions of P that are not available to the Olsen or water extraction are actually plant available. Taking account of this additional plant-available P in nutrient management may offer cost savings to farmers and reduce the risk of P loss from these soils to surface waters by reducing fertilizer application rates.

At least some of this additional plant-available P may be inorganic P associated with Fe and Al phases that is insoluble in the Olsen extract. A second possibility is that at least part of the organic P pool, which is not measured in the Olsen extract, is plant available. Organic P can be an important fraction in soil solution (Murphy, 2007) and a large part of the Fe- and Al-associated P extracted in acid oxalate to determine DPS can be organic P (Fransson, 2001). Strong positive correlations between both total P in NaOH–EDTA and total soil P (both containing large proportions of organic P) and oxalate-extractable P suggest that this may be the case. The fact that total P in NaOH–EDTA and oxalate P were so similar in magnitude suggests that they may be extracting the same pools of P. As organic P made up a large proportion of total P in NaOH–EDTA (54%, on average), this result suggests that the good relationship between DPS and P plant availability may result from, in part at least, organic P. In contrast, the poor correlation between Olsen P and total P in NaOH–EDTA indicates that Olsen P does not include this additional Fe- and Al-bound P.

Iron- and Al-rich tropical Oxisols and Ultisols strongly sorb inorganic P and the cycling of P from organic pools in these soils can be particularly significant for plant nutrition (George *et al.*, 2006). Oxalate-extractable P has been found to be a good indicator of plant-available P in such soils (Fengmao & Yost, 1999). Organic P also becomes more important in P cycling in older soils because of an increasingly limited supply of inorganic P (McDowell *et al.*, 2007). No soils in Ireland are older than approximately 13 000 years and Oxisols and Ultisols do not occur, but as has been noted, the basaltic soils have larger contents of Fe and Al. Phosphorus cycling in these temperate basaltic soils may therefore bear some similarity to that associated with tropical soils and soils at a later stage of pedogenesis. Their mineralogical composition is also unusual for temperate soils, because soils rich in haematite, gibbsite and kaolinite are generally associated with tropical climates. Tertiary palaeosols occur within the basalts (Hill *et al.*, 2001) and may contribute to their unusual mineral assemblage.

Although organic forms of P are not taken up directly by plant roots, enzyme-labile organic P can be a large soil-P pool and an important source of P for plants (Asmar *et al.*, 1995; Shand & Smith, 1997; Hayes *et al.*, 2000; George *et al.*, 2002; McDowell & Koopmans, 2006). Phosphate monoesters, in particular, are regarded typically as being relatively unavailable to plants

(Richardson *et al.*, 2000, 2001), but they may be able to contribute to plant-available P (Chen *et al.*, 2004) when more readily available forms of phosphorus are limited (Turner *et al.*, 2003b; Turner *et al.*, 2005b). As a response to small concentrations of inorganic P in soil solution, plant roots can release organic acids and enzymes such as phosphatase to increase P availability from organic pools (Antibus *et al.*, 1997; Richardson *et al.*, 2000; George *et al.*, 2006), particularly through mycorrhizal associations (Bolan, 1991; Marschner & Dell, 1994). Such biochemical responses to P deficiency appear to be coordinated by changes in cellular phosphate levels and enhanced expression of gene coding for phosphatase (Raghothama & Karthikeyan, 2005). A more P-limited soil environment may also favour organisms capable of degrading phosphate monoesters (Turner, 2007). Therefore, P that is not extracted by agronomic tests and not considered immediately plant-available may actually be available in the rhizosphere (Magid *et al.*, 1996).

The fact that phosphate monoester concentration was positively correlated with Olsen P and water-extractable P for non-basaltic soils in the present study suggests that phosphate monoesters are also taken up by plants in these soils when inorganic P is limited and that easily-soluble inorganic P is regulating P supply. Lack of a similar correlation for basaltic soils suggests that easily soluble inorganic P may not control P supply. Low solubility of inorganic P (Olsen and water extractions) in these soils with large PSCs may limit P supply to grass swards, inducing the uptake of less available forms such as phosphate monoesters. Smaller concentrations of phosphate monoesters and smaller proportions of inorganic P in the NaOH–EDTA extraction of basaltic soils are consistent with this.

DNA and other highly unstable forms of organic P could also be available to plants on basalt soils and could account for some of the shortfall between Olsen P and actual plant-available P. These compounds would be quickly cycled from soil microbial biomass to simpler, more plant-available forms (McDowell *et al.*, 2007). Their instability in soil, and possible hydrolysis during extraction, may partially account for their reduced presence in NaOH–EDTA extractions.

## Conclusions

Parent material can be an important factor in determining P speciation. Phosphate monoesters were more prevalent in basaltic soils, probably because of their larger oxalate-extractable Fe and Al contents. Phosphate monoesters may be associated with non-crystalline Fe– and, to a greater extent, Al–organic matter complexes. DNA was also more prevalent in basaltic soils, probably because of their finer texture and clay mineral (smectite, kaolinite) and metal oxide content. Speciation is also strongly influenced by relative stability, as determined by the properties of the species themselves and soil conditions such as redox and pH. DNA was associated with soils of greater total C content and more acidic pH, indicating poor drainage conditions.

Inadequacy of the Olsen P test for basaltic soils may result from, in part at least, phosphate monoesters that are not measured using this test but are plant available. Strong sorption of inorganic P in the Fe- and Al-rich basaltic soils leads to small rates of inorganic P solubility and may induce plants to access organic P. DPS would appear to be a better indicator of P status for such soils and P nutrient management on basaltic soils should take this into account to avoid over-application of fertilizer P to grasslands and the associated financial and environmental costs.

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