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An indoor freeze/thaw lysimeter study of phosphorus leaching from soils with four catch crops

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An indoor freeze/thaw lysimeter study of phosphorus leaching from soils with four catch crops

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Abstract

Catch crops have been found to decrease leaching of nitrates into surface and ground waters, but they also have the potential to increase phosphorus (P) loadings to natural waters as well, due to plant cell destruction caused during natural freezing and thawing events. An indoor lysimeter experiment was carried out using a clay and a sand soil with the application of four different plant species: perennial ryegrass cultivar 'Helmer' (*Lolium perenne* L.), honey herb, cultivar 'Stala' (*Phacelia tanacetifolia* L.), chicory, cultivar 'Puna' (*Cichorium intybis* L.), and oilseed radish, cultivar 'Adios' (*Raphanus sativus* L.). These plants were exposed to four simulated rainfall and three freezing events in two separate experiments, one using topsoil monoliths with applied plant material and one with plant material only. Sand and clay soils had significantly different leaching loads after 1 and 2 freezing events with total P contents in leachate equivalent to 0.71 kg ha⁻¹ for clay and 0.20 kg ha⁻¹ (P=0.0018) for sand soil, and after the second event 0.54 kg ha⁻¹ for clay and 0.30 kg ha⁻¹ for sand (P=0.0026). The combined total P leaching loads from the clay soil were significant for many of the plants and were in the order of chicory (2.6 kg ha⁻¹) > ryegrass (2.3 kg ha⁻¹) > oilseed radish (2.2 kg ha⁻¹) > honey herb (1.3 kg ha⁻¹), taken from the plant and soil experiment. Losses were of a magnitude greater from the plant only experiment with chicory (51.7 kg ha⁻¹) > oilseed radish (43.2 kg ha⁻¹) > honey herb (18.4 kg ha⁻¹) > ryegrass (10 kg ha⁻¹). A total P analysis of plant tissue from before the first lysimeter experiment and after the second plant only experiment showed that chicory lost 84%, oilseed radish 76%, ryegrass 74% and honey herb 39% of total initial biomass P. The results indicate that soil texture and plant choice can have a large impact on leaching loads that could potentially enter natural waters.

Table of Contents

Introduction.....	4
Materials and Methods	6
Soil Lysimeter Experiment.....	6
Plant Only Experiment.....	9
Calculations and Statistical Analysis.....	9
Results	10
Amounts of Leachate	10
Phosphorus Leaching Comparing a Clay and Sand a Soil	12
Phosphorus Leaching Comparing Clay and Sand Soils Including Plants	13
Phosphorus Leaching From Plant Only Experiment.....	18
Discussion	24
Conclusions.....	29
Acknowledgements.....	29
References.....	30
Appendix.....	34

Introduction

The phosphorus (P) loading to the Baltic Sea is of great concern to the surrounding Baltic nations. The non-point source agriculture is reportedly the largest contributor of P to this particular accelerated eutrophication problem, by contributing as much P as all other sources combined (Bergström et al., 2007). These losses come from a range of different sources from within agriculture, from stored manure piles to over fertilisation of crops, as well soil runoff due to poor soil management. One potential source of P losses that has been gaining more recognition is that from the use of catch crops.

Catch crops or cover crops as they are also known have been traditionally used as a 'mop up' crop, planted to remove any excess available soil nutrients, specifically nitrate-N after a summer crop has been harvested (Bergström and Jokela, 2001). This reduces the risk of nitrate leaching during the following winter period as it is 'locked' within plant residue until the following spring. Although, the large gains from some management practices aimed at reducing losses of a specific nutrient can in fact magnify the losses of another (Bechmann et al., 2005). Authors as far back as Tukey et al. (1957) realized that if plants have the ability to take up nutrients, they can also equally lose them, which is the case for P. Timmons et al. (1970) found this when conducting an experiment on nutrient losses from lucerne (*Medicago sativa*) and bluegrass (*Poa pratensis* L.), that surface runoff from these crops could contribute significant amounts of P to lakes and streams. Even though there has been increasing research into leaching losses of P (Schreiber and McDowell, 1985) the focus has been on the surface flows known as run-off (Sharpley, 1981; Sharpley and Smith, 1991; Uusi-Kamppä, 2007; Bechmann et al., 2005). These researchers have found that, although an increase of inorganic P is found in runoff, less particulate P related to soil erosion is found due to the soil stabilizing effects of the crop. Leaching of P through the soil has been found to be negligible in the majority of soils due to the sorption ability of the soil (Sinaj et al., 2002). However, the recent research suggests it still poses a threat to the subsurface water systems (Bergström et al., 2007).

The function of P in plants is vital, considering its use in the transport of energy through ADP and ATP (Whitehead, 2000). It is found in all parts of the plants, and is susceptible to loss under natural climatic conditions, such as those encountered in northern Europe. These conditions include rainfall, with which leached plant nutrients can become part of the soil-water matrix environment (Schreiber and McDowell, 1985). This theory was also acknowledged by Sharpley (1981) who stated that in order to understand soil nutrient flows we need to include the P contributions from plant material and specifically how rainfall and plant type affect leachability throughout the year.

Another major aspect of P losses from catch crops, are the increased leaching losses caused by the process of freezing and thawing, a natural winter scenario in northern Europe. Herber (1967) commented that the membranes that surround the protoplasm of plants cells are damaged by frost.

Tukey and Morgan (1963) found that when plant tissues are damaged by very low temperatures that plant leaching losses increase greatly. Plants exposed to frost released large amounts of P (Uusi-Kamppä, 2007). In other research of lucerne by Timmons et al. (1970) it was found that plant losses of soluble inorganic P from frozen and thawed plants, which were subsequently irrigated, were 46 times higher than from plants not frozen and thawed. Some results from Torstensson et al. (2006) also showed higher leaching losses of P from a soil with cover crops, than that without, which was explained by the freezing of plant material in the autumn.

Various authors have also found that the leachability and nutrient content of plant materials varies between plant varieties (Schreiber and McDowell, 1985; Jones and Bromfield, 1969) and plant age (Tukey, 1970; White, 1973). For example an experiment conducted by Miller et al. (1994) found concentrations in leachate from 2.0 – 15 mg L⁻¹ for red clover and oilseed radish, respectively. This suggests plant choice is also a critical factor in controlling the losses of P by leaching. The soil type is another factor of consideration, though most soils show little or no loss of P through the leaching process, as phosphate ions are rapidly adsorbed or precipitated by other soil fractions (Whitehead, 2000). However, some situations can allow higher concentrations of P in leachate. This can include cracking in soils can create macropores allowing surface water to travel directly to groundwater (Wiederholt and Johnson, 2005) increasing the losses of nutrients from the soil.

Farmers are being encouraged to grow catch crops through regulations such as the requirement of agricultural land owners in the southern region of Sweden to have at least 50% of their arable land covered by a winter catch crop in order to reduce nitrate leaching (Ulén, 1997). With a large part of the dynamics of catch crops and their potential P losses and their relation to soil properties not clearly understood, it seems enforcing the use of these crops may not be an ideal prevention method for P leaching losses.

The objective of this research was to determine the P losses from four different catch crops applied on two soil types as well as a bare soil under simulated freezing Swedish winter climatic conditions in an indoor lysimeter experiment. To identify if soil type had an influence on P leaching loads, a sandy and silty-clay soil were used. A second experiment was also carried out using plant material only, combined with freezing/thawing and artificial rainfall to determine P leaching loads from plant material that had not been exposed to the adsorption abilities of soil particles as in the first experiment.

Materials and Methods

Soil Lysimeter Experiment

An indoor lysimeter experiment was conducted using four plant species: perennial ryegrass cultivar 'Helmer' (*Lolium perenne* L.), honey herb, cultivar 'Stala' (*Phacelia tanacetifolia* L.), chicory, cultivar 'Puna' (*Cichorium intybis* L.), and oilseed radish cultivar 'Adios' (*Raphanus sativus* L.). The varieties were chosen due to being common Swedish catch crops with the exception of honey herb, which has been found to be efficient at removing soil nutrients from poor soils. The four plant varieties were grown in a glasshouse from the 14th January 2010 until 23rd February 2010, for a total of 40 growing days, at a temperature of 23°C between 0600 and 2300 and 18°C from 2300 to 0600. Light was used once the plants had germinated to increase the growth rate of the plants, with on an off times set to follow the daily temperature changes. All plants were sown at a rate of 8 kg ha⁻¹ to simulate a typical farming scenario. The plants were irrigated every second day and only one application of a high nitrogen based fertilizer 'Yara Mila' with a composition of 21-3-10 (NPK) was applied on the 5th February 2010. The application rate of 100 kg N ha⁻¹ gave 14.3 kg P ha⁻¹ and 47.6 kg K ha⁻¹, which was first ground up then applied, followed by irrigation of approximately 5 mm. At maturity some of the oilseed radish had started to flower, whilst the rest were still in the vegetative stage. The amount of plant material added to each lysimeter from the original growing containers was based on the sowing rate and how many plants of each variety should grow per hectare, which varied due to seed size, and then per soil area in each lysimeter (Table 1).

Table 1: The number and weights of plants added to each lysimeter. 'Tops' denotes anything above ground and 'roots' anything below ground. Note that the tops weight was targeted to be similar within each treatment and consequently the root weights varied within treatments.

Plant	Calculated plants applied per lysimeter	Mean fresh weight of plant material added per lysimeter		Mean plant top and root application kg dm ha ⁻¹
		Tops (g)	Roots (g)	
Perennial Ryegrass	8	40	15	2765
Honey Herb	12	175	2	5152
Chicory	17	175	6	4813
Oilseed Radish	2	175	7	4658

Two different soil types were used, a sandy soil (Nåntuna) from Ultuna, Sweden (59°48'42.78''N 17°40'41.88''E) classified as an Arenosol by the WRB system (IUSS Working Group WRB, 2006), and a clay soil (Brunnby) from Västerås, Sweden (59°36'51.57''N 16°39'34.08''E) classified as a possible Cambisol (no investigation was undertaken). Some of the P contents from the two sites are included in Table 2.

Twenty 200 mm diameter by 250 mm in length plastic sewer pipes were inserted in the soil at each of the two sites. At the Brunnby site (6th November 2009) a tractor with a front-end loader was used to press the pipes with a metal cutting edge at one end, into the ground. The soil was almost bare apart from a few weeds. At the Nåntuna site (13th November 2009), a sledge hammer was used to hand drive the pipes in, as the soil was mainly sand. This soil was badly contaminated with 'twitch' (*Elytrigia repens*). The tubes filled with soil were then dug out, capped at both ends and stored at 5°C, and covered from light until used.

Table 2: Some soil chemical properties of the Nåntuna and Brunnby soils.

Soil	Depth (cm)	pH (H ₂ O)	TOC (%)	AL-P	AL-Ca	AL-Al	AL-Fe
				mg 100g ⁻¹			
Nåntuna	0-10	7	1.440	15.5	211.2	11.8	18.6
	10-30	7.4	1.016	11.6	176.6	11.1	17.1
Brunnby	0-23	6.2	1.485	4.7	241	20	35

There were 3, 4 or 5 replicates used in the experiment, 3 for the 'honey herb', 4 for the ryegrass, chicory and oilseed radish, and 5 for the control treatment (bare soil only). A shortage of honey herb plant material was the reason the unused replicate was added to the control treatment instead.

Plant material, including roots, was removed from the growing containers with attached soil brushed off, weighed separately as tops and roots, and then spread on the soil surface of each of the lysimeters. An additional section of 100 mm sewer pipe was taped to the top of the original pipe to stop any water splash loss from the irrigation procedure. A 50µm nylon filter mesh was placed under the base of each lysimeter and then placed in a metal collection pot and placed in one of the 2 simulators. The collection pot was then connected to a 1 L glass collection bottle underneath to catch the leachate. The lysimeters were irrigated for 10 hours at a rate of 5 mm hr⁻¹ (total 50 mm) of tap water with collection bottles changed after 5 hours.

The irrigation water was applied in 6 second spray applications with 55 second intervals, at a pressure of 0.5 bar as this was the only way to adjust the application rate with this system. The two rainfall simulators were not exactly the same in their application of water, both within each simulator and also between simulators.

During calibration testing, the application of 5 mm of rainfall per hour was targeted, with a range of 4.7-6 mm between the two simulators achieved, which was considered acceptable. To reduce the risk of any soil or plant column being advantaged or disadvantaged, there was a random assignment of simulators.

Once irrigated, the columns were left to drain for 3 days and then placed in a freezer at -18°C for 3 days. They were then removed and allowed to thaw for 24 hours at room temperature (~20°C) and after which the process was repeated another 3 times, giving 4 irrigation events and 3 freezing events. These simulation cycles allowed for the management of the many replicates into and out of the simulators. During the 3rd irrigation, the irrigation system did not stop after the predetermined 10 hours, but instead after 22 hours, causing excessive irrigation and subsequent loss of leachate water. This occurred to 17 of the 40 lysimeters, with a mixture of treatments affected.

The leachate volume was measured and 100ml subsamples were retained for analysis of total-P and PO₄-P. Concentrations of total P and dissolved P were measured according to the method of the European Committee for Standards (1996). Plant samples were analyzed for total-P to be able to quantify the initial amount of total-P added from within the different plants. The plant material was weighed, dried at 50°C for 3 days, ground and analysed using an ICP Optima 7300 DV (Perkin Elmer, Ma. U.S.A.) (Appendix 1).

Plant Only Experiment

In an attempt to calculate how much P was potentially lost from the plant material during freezing and thawing, a second experiment was run with only plant material and no soil.

Similar amounts of plant material as in the previous experiment were placed into metal pots on top of 50 μm nylon filter mesh and were connected to glass bottles to collect. A control treatment was also used, without plant material. The material was then placed in the rainfall simulator and irrigated, frozen and thawed the same as the previous experiment. The leachate volume was measured, sub sampled and analysed as before. When the experiment was finished, the plant samples were dried at 50°C for 3 days, ground and analysed for total-P using an ICP Optima 7300 DV (Perkin Elmer, Ma. U.S.A.) (Appendix 1).

Calculations and Statistical Analysis

Leached amounts of P were calculated from leachate concentrations for each plant species using the analysis concentration and leachate volume collected for each individual lysimeter. These values were then converted into P loads in kg ha^{-1} using the surface area of the lysimeter. Values presented for each of the treatments are means of the combined replicates.

The statistical analysis included a 2-way analysis of variance to compare the soil types using SAS version 9.2 (2008). The soil comparison was done by taking the mean of all treatments within each soil type and comparing them for the separate P analysis and simulation runs. Two separate 2-way analysis of variance were completed to compare the two different soils and the plant soil interaction. The first was within each soil to see differences between simulation runs and P tests and the second was to get statistical information on all comparison possibilities between the different soils and different treatments, within each simulation run and within each P test. Significant differences identified within the report are at a 95% confidence interval, anything below this was regarded as not significant (NS).

Results

Amounts of Leachate

The initial soil columns were not adjusted to a common specific soil water content, therefore drainage results for the first rainfall simulation were low in comparison to the other three (Table 3). The first three simulations had extreme outliers in terms of very low drainage volumes collected. This occurred only in the clay soil columns, apart from the first simulation where both soils had a variable leachate loss, which could be explained by the initial possible differences in soil water content in each lysimeter (Table 3). If the collected volumes were unexplainably low during the experiment, the replicate was removed from the dataset for that particular simulation event (Table 4).

Due to the complication of the experiment caused by excessive watering of some of the replicates in the 3rd simulation, numerous replicates were removed from the analysis dataset (Table 4). This left some of the plant species with sometimes only one replicate in simulations 3 and 4. This also had an effect on simulation 4, and these effected replicates were also left out of simulation 4.

Table 3: The mean, maximum, minimum and standard error of the mean from the drainage water collected from the lysimeters from each of the two soils and four rainfall simulations. The desired application was 1.38 L (50 mm).

Rainfall Simulation	Soil	Mean (mL)	Min ((mL)	Max (mL)	Standard Error of the Mean
1	Clay	1066	58	1382	71
	Sand	769	513	961	30
2	Clay	1194	139	1577	75
	Sand	1316	1144	1591	28
3	Clay	1342	640	1624	83
	Sand	1310	1071	1497	34
4	Clay	1402	1229	1564	35
	Sand	1474	1215	1727	44

Table 4: The replicates that were removed due to being either an outlier (OUT) or damaged in simulation 3 due to excessive irrigation (DG) or were not damaged and analysed as normal (ok). Black boxes indicate no replicate in the experiment.

Soil and Plant Material	Simulation 1 Replicate					Simulation 2 Replicate					Simulation 3 Replicate					Simulation 4 Replicate					
	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5	
Clay	Ryegrass	ok	ok	ok	ok		ok	ok	ok	ok		DG	DG	ok	ok		DG	DG	ok	ok	
	Honey Herb	ok	ok	ok			ok	ok	ok			DG	DG	ok			DG	DG	ok		
	Chicory	ok	ok	ok	ok		ok	ok	ok	ok		DG	DG	ok	ok		DG	DG	ok	OUT	
	Oilseed Radish	ok	ok	ok	ok		OUT	OUT	ok			DG	DG	ok	OUT		DG	DG	ok	ok	
	Control	OUT	ok	ok	ok	ok	ok	ok	ok	ok	ok	DG	ok	ok	ok	ok	DG	DG	ok	ok	ok
Sand	Ryegrass	ok	ok	ok	ok		ok	ok	ok	ok		DG	DG	ok	ok		DG	DG	ok	ok	
	Honey Herb	ok	ok	ok			ok	ok	ok			DG	DG	ok			DG	DG	ok		
	Chicory	ok	ok	ok	ok		ok	ok	ok	ok		DG	DG	ok	ok		DG	DG	ok	ok	
	Oilseed Radish	ok	ok	ok	ok		ok	ok	ok	ok		DG	ok	ok	ok		DG	ok	ok	ok	
	Control	ok	ok	ok	ok	ok	ok	ok	ok	ok	ok	DG	ok	ok	ok	ok	DG	ok	ok	ok	ok

Phosphorus Leaching Comparing a Clay and Sand a Soil

The overall differences in leaching loads between the two soils are quite apparent in figure 1. These values were the mean of all treatments within each simulation and for each soil type. In the first simulation (figure 1A-B) showed the different P types, and that there was no statistical difference (NS) between clay and sand soils (table 5) in total and PO₄-P.

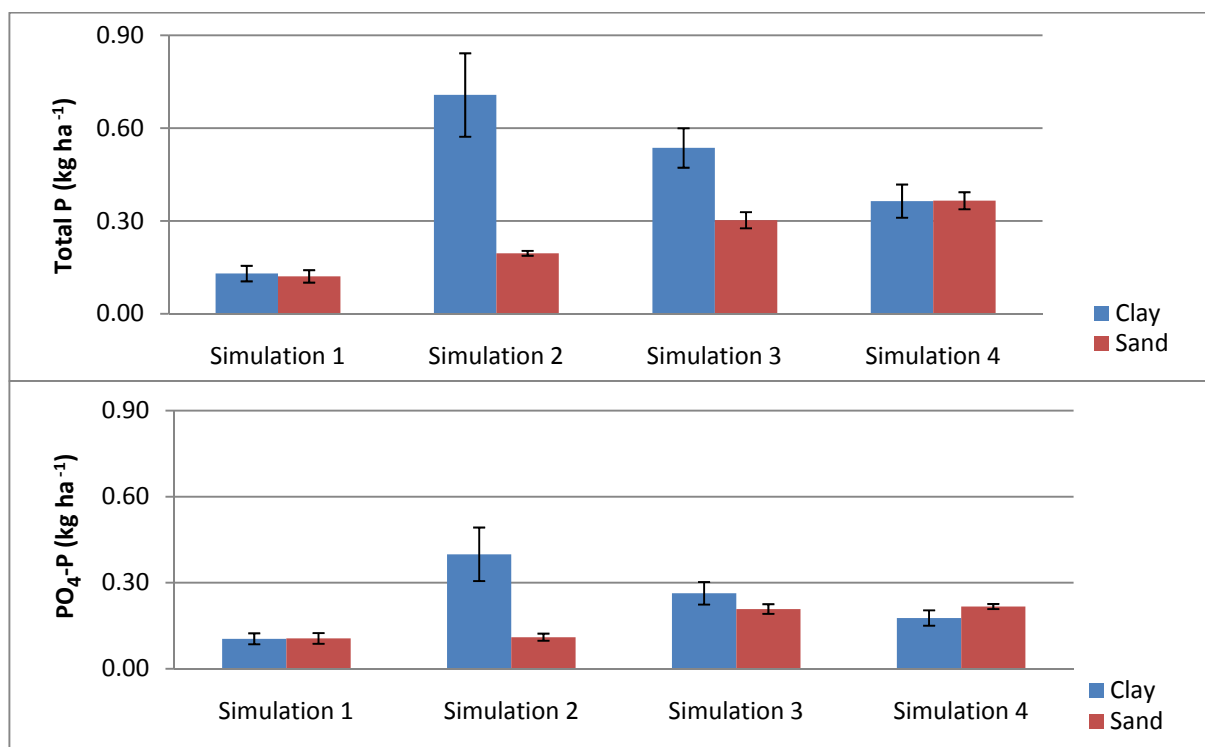


Figure 1: Mean P leaching loads from all treatments

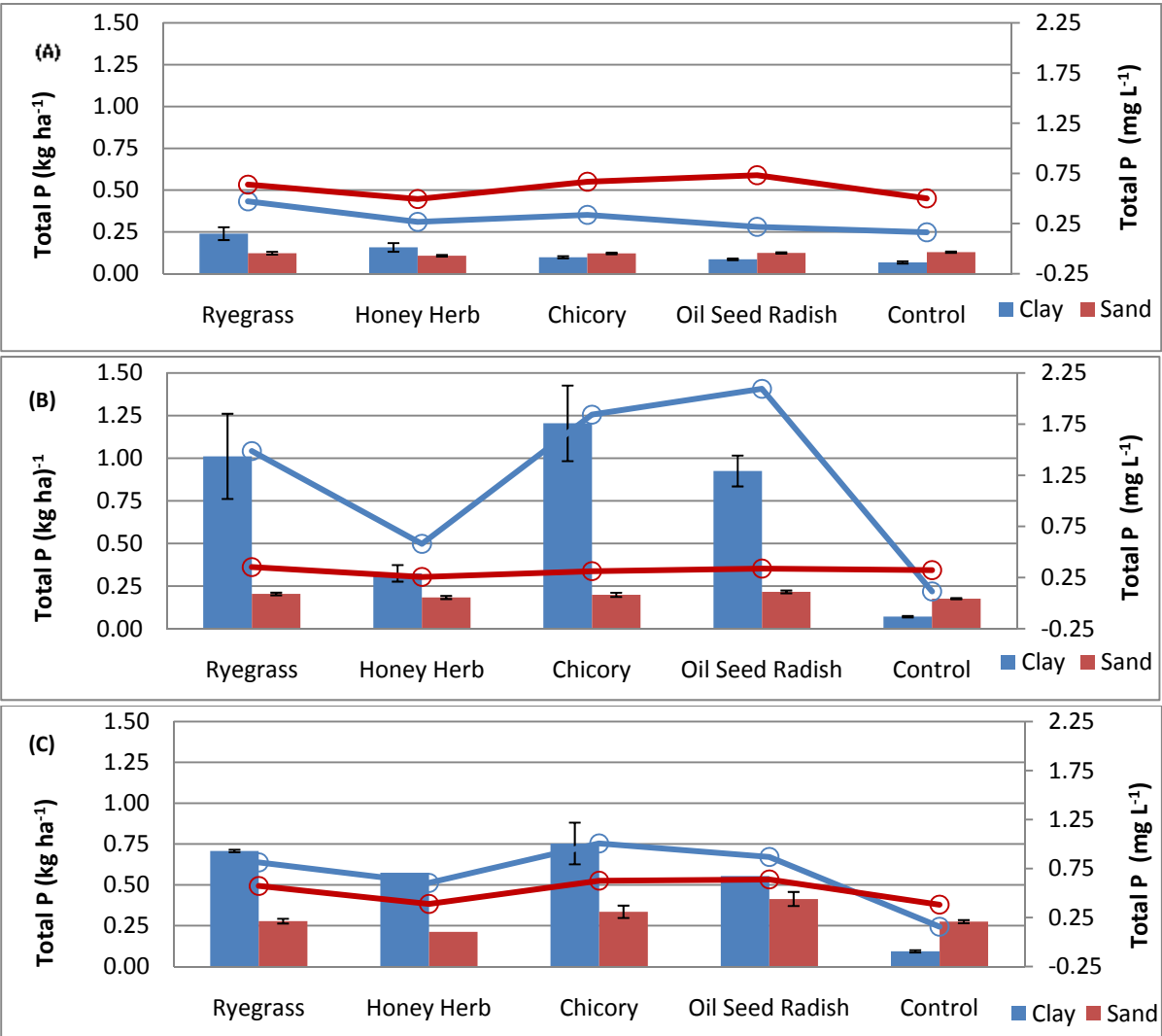
Simulation 2 showed significant differences between the two soils for both P types. Simulation 3 had a high level of significance in total P with 0.536 kg ha⁻¹ for clay and 0.302 kg ha⁻¹ for sand. Simulation 4 showed no differences in total P or PO₄-P.

Table 5: Statistical significance (P values) comparing the clay and the sand soil (from figure 1).

	Simulation 1	Simulation 2	Simulation 3	Simulation 4
Total P	NS	< 0.005	<0.005	NS
PO₄ P	NS	=0.01	NS	NS

Phosphorus Leaching Comparing Clay and Sand Soils Including Plants

The combining of the plant types to distinguish which of the soils produced more leaching showed differences between the actual soils, but not which plant had the greatest and the least effect on P loading, which would be a practical end use. Figure 2 shows in detail the comparison between the different plant and soil types used, and their total P losses. Apart from figure 2, in general the clay soil had greater variability shown by the standard error of the mean (S.E.M) bars, and also had higher leaching losses in both concentration and leaching loads. A trend occurred throughout the series of four figures (figure 2) with clay chicory, oilseed radish and ryegrass having higher leaching losses and loads than honey herb and the control treatment. The first simulation for clay ryegrass and honey herb showed to have greater loading losses than the other plant types, but after the first freezing event it changed and honey herb had significantly less leaching. The sand soil leaching loads were more consistent, with no large increases or decreases between simulation events in comparison to the clay soil.



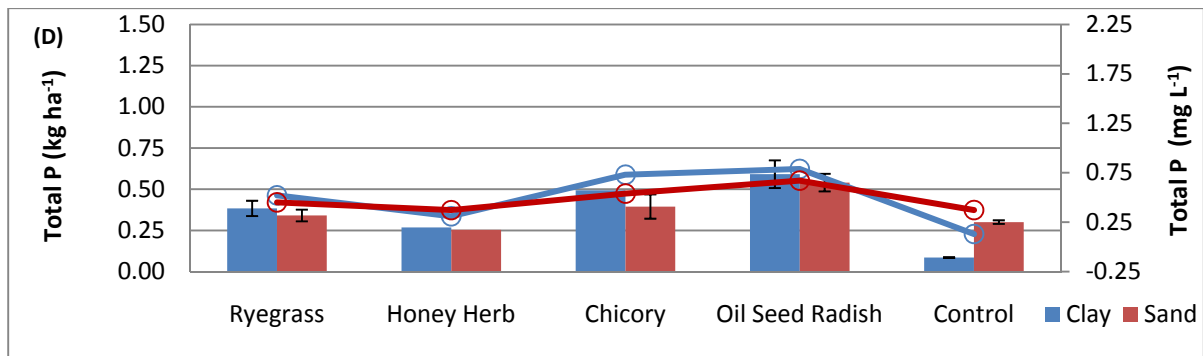


Figure 2: The total P concentrations (circles) and total-P leaching loads (columns) from the four simulations. Simulation 1= (A), simulation 2= (B), simulation 3= (C), simulation 4= (D). Standard errors of the mean bars are present on those columns that have more than one replicate.

Table 6: Statistical significance of leaching loads between clay and sand soils and their respective treatments from figure 2. A 95% confidence interval is used.

	Simulation 1	Simulation 2	Simulation 3	Simulation 4
Ryegrass	NS	=0.012	=0.006	NS
Honey Herb	NS	NS	NS	NS
Chicory	NS	=0.003	=0.007	NS
Oilseed Radish	NS	NS	NS	NS
Control	NS	NS	NS	NS

There were no statistical differences in the first simulation between any treatments. Simulation 2 and 3 showed variable significance results between the treatments (table 6) with a very high degree of significance between the chicory clay and the chicory sand treatments.

The total P leaching loads for the control treatments increased after each freezing event for the sand soil. The sand control loads increased over the four simulations from 0.129 to 0.176 to 0.275 to 0.301 kg P ha⁻¹ (figure 2), although was not significantly different from clay in any simulations (table 6). The clay soil control went from 0.068 to 0.071 to 0.093 to 0.085 kg P ha⁻¹. There was no statistical difference at a 0.05% level between the soils; however the differences do suggest an underlying reason for the variation in control treatments between the soils. Table 7 shows each of the plant treatments with their respective soils' control treatment loading removed to get an idea of what leaching is most likely from the plants and not from the soil. Simulation 1 (table 7) shows ryegrass and honey herb clay soil having the highest total P leaching loads.

Ryegrass leaching was 0.172 kg P ha⁻¹ (table 7) during simulation 1, which is nearly twice as much total P as honey herb (0.090 kg ha⁻¹). Ryegrass clay had among the greatest leaching loads throughout the 4 simulations. Honey herb clay on the other hand was one of the lowest.

The sand soil in fact shows negative numbers which of course is not realistic though the numbers are too close to suggest a difference from the control treatment. This does give an indication of the low leaching losses of the sand in comparison to the clay soil.

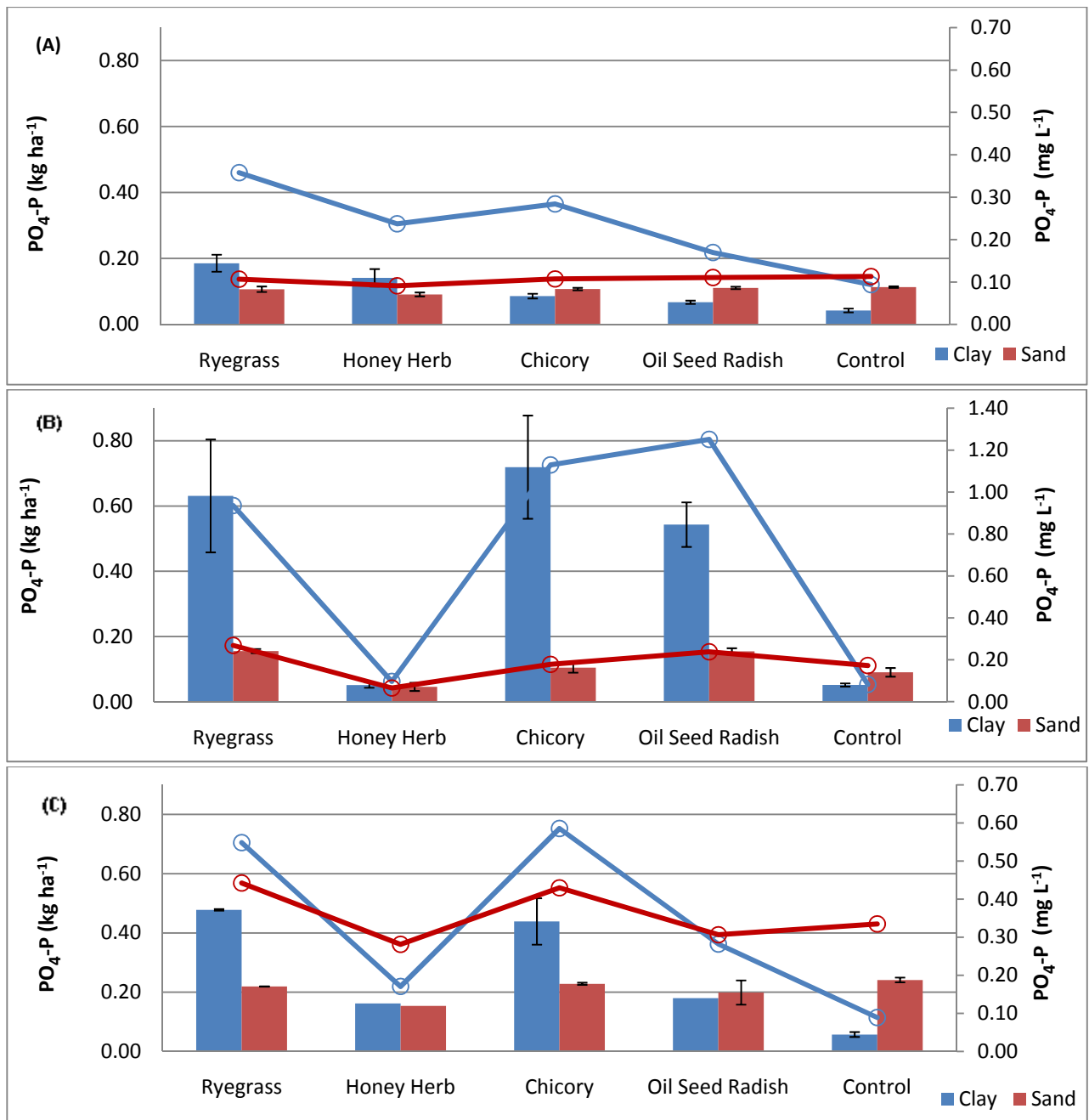
Table 7: Mean total P leaching loads from the four plant types used minus the contribution from the soil only

	Simulation 1		Simulation 2		Simulation 3		Simulation 4	
	Clay	Sand	Clay	Sand	Clay	Sand	Clay	Sand
	(kg ha ⁻¹)		(kg ha ⁻¹)		(kg ha ⁻¹)		(kg ha ⁻¹)	
Ryegrass	0.172	-0.006	0.941	0.028	0.615	0.003	0.298	0.040
Honey Herb	0.090	-0.022	0.254	0.007	0.481	-0.062	0.183	-0.047
Chicory	0.031	-0.007	1.134	0.023	0.660	0.060	0.408	0.094
Oil Seed Radish	0.019	-0.004	0.855	0.040	0.461	0.139	0.506	0.240

The total P losses from each treatment is shown in table 8, showing obvious differences between the soils and the plants. The clay soil values from the table indicate the total losses from the ryegrass, chicory and oilseed radish are all very close, but the honey herb lost the least total P of all the plants. The control treatment shows an interesting difference and is in fact the opposite from the trend of higher losses from the clay soil and lower from the sand.

Table 8: Accumulated mean total P loss from each of the treatments over the four simulations.

Ryegrass		Honey Herb		Chicory		Oilseed Radish		Control	
Clay	Sand	Clay	Sand	Clay	Sand	Clay	Sand	Clay	Sand
(kg ha ⁻¹)									
2.34	0.95	1.33	0.76	2.55	1.05	2.16	1.29	0.32	0.88



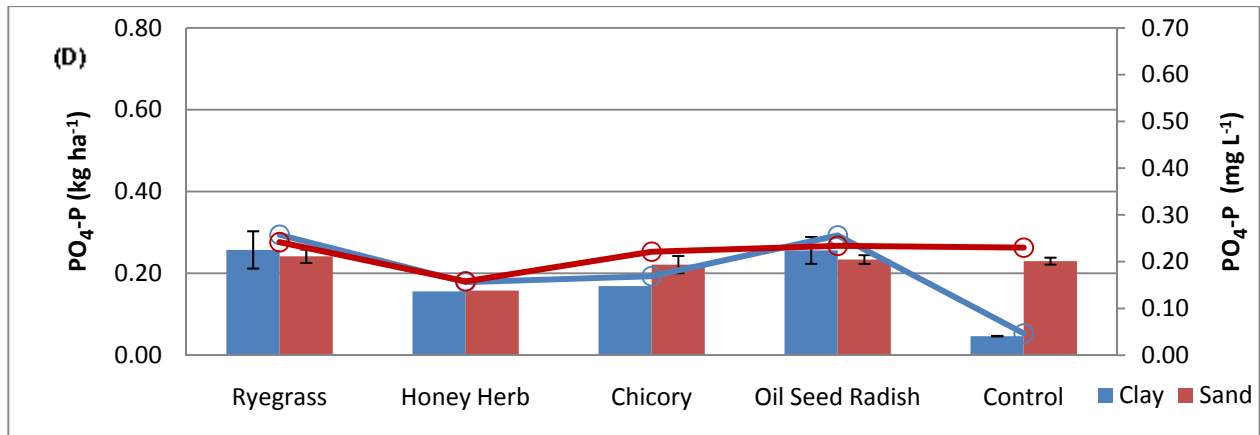


Figure 3: The PO₄-P concentrations (circles) and PO₄-P leaching loads (columns) from the four simulations. Simulation 1= (A), simulation 2= (B), simulation 3= (C), simulation 4= (D). Standard errors of the mean bars are present on those columns that have more than one replicate.

Table 9: Statistical significance between clay and sand soils and their respective treatments from figure 3 (leaching loads). A 95% confidence interval is used.

	Simulation 1	Simulation 2	Simulation 3	Simulation 4
Ryegrass	=0.021	=0.040	=0.013	NS
Honey Herb	NS	NS	NS	NS
Chicory	NS	=0.009	=0.037	NS
Oilseed Radish	NS	NS	NS	NS
Control	=0.028	NS	=0.013	=0.002

The total P trend from figure 2 carried on for the PO₄-P comparison shown in figure 3, with the same high loading plants having the greater losses again. This is due to the proportion of PO₄-P of total P in clay, measured for the first simulation being roughly 80-90%. In the second, ryegrass 62%, honey herb 16%, chicory 60%, and oilseed radish 59%. The third ryegrass 68%, honey herb 28%, chicory 58%, and oilseed radish 32%, and the fourth simulation, ryegrass 67%, honey herb 58%, chicory 34%, and oilseed radish 43%. This shows that the PO₄-P is a significant part of the total P losses.

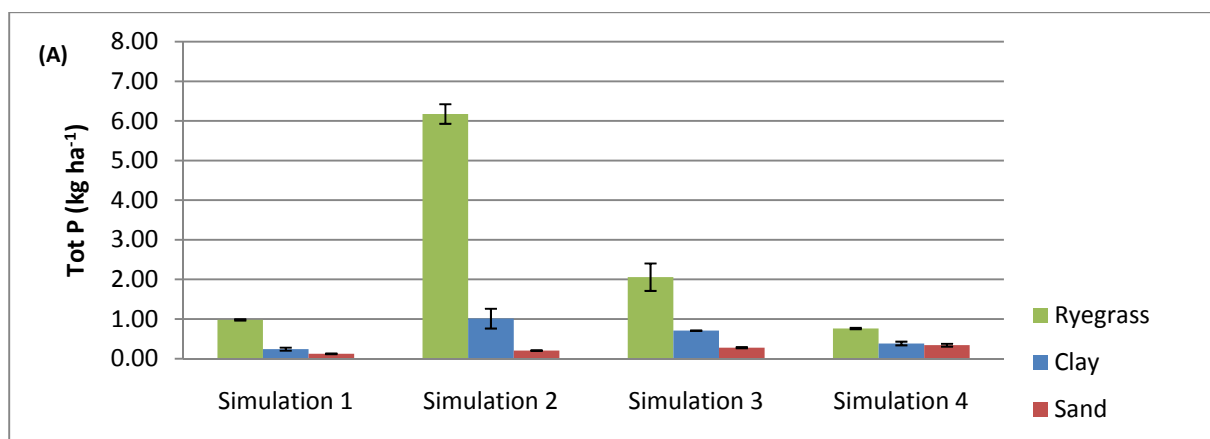
Honey herb had the least leaching losses and the clay chicory and ryegrass had the most. The clay again had the highest peaks in the second simulation, with sand leaching losses being only a fraction of the clay in the same simulation. Throughout the first three simulations ryegrass clay and sand were significantly different (table 9), with clay, again, dominating with the higher loadings. The greater loadings from the sand control over the clay control are shown clearly, with the PO₄-P form a large proportion of the total P lost (table 9).

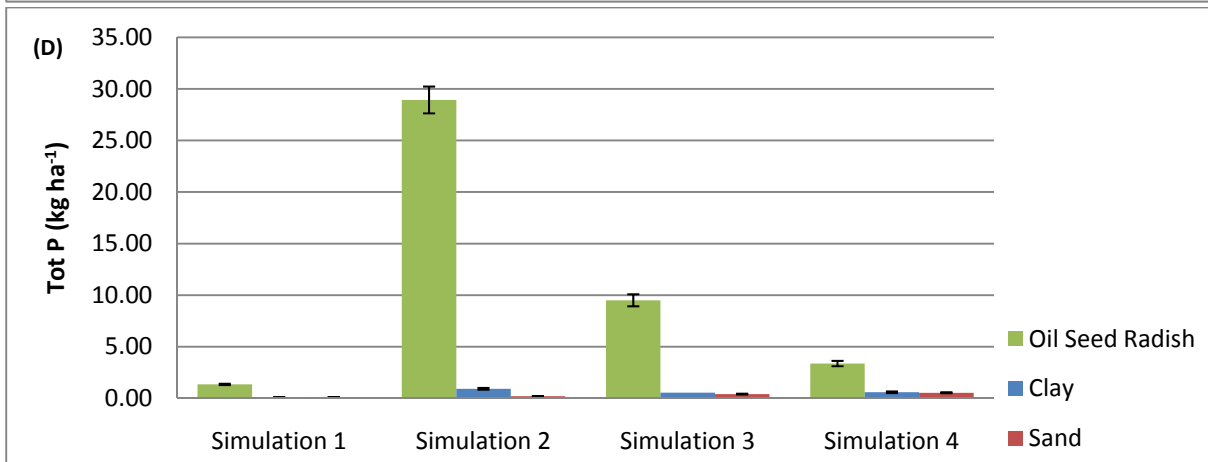
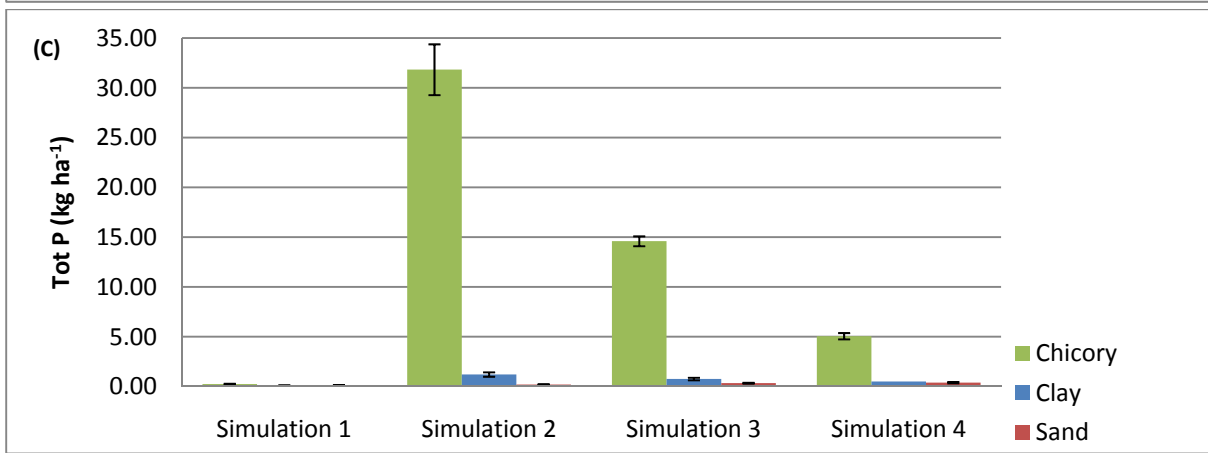
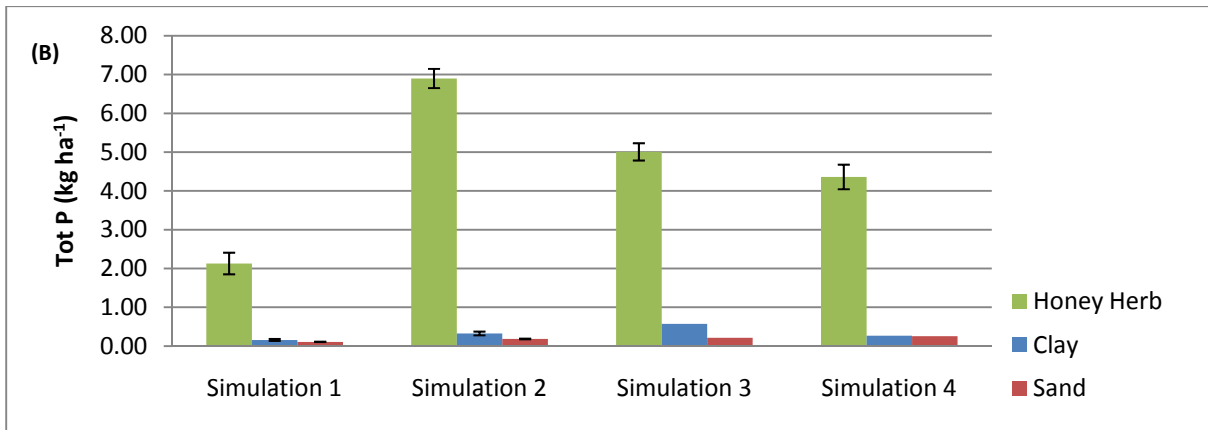
Phosphorus Leaching From Plant Only Experiment

Losses of total P from the plant species were higher than from the clay or sand soils. There was a large difference between the P amount applied to the surface (figure 4) and the amount leached.

The first simulation showed the highest mean losses of 2.1 kg P ha⁻¹ from honey herb consisting mostly of the PO₄-P fraction (figure 5B), followed by the next highest 1.3 kg ha⁻¹ from oilseed radish, even though they were not significantly different. This was followed by ryegrass at 0.98 kg ha⁻¹ (NS from oilseed radish), and chicory at a mean of 0.24 kg ha⁻¹, which was significantly different from honey herb, oilseed radish (P<0.05) and ryegrass (P<0.08).

The losses in general were again highest in the second simulation, which occurred after the first freezing event. However, as to how high the losses were, was dependent on the plant species themselves. Chicory and oilseed radish had by far the greatest losses after the second simulation (figure 4c, d), with almost five times the total P lost in the chicory and oilseed radish than the ryegrass and honey herb (P<0.001) (figure 4). Ryegrass and honey herb losses were not different from one another statistically, nor were they different (NS) from the control treatment. The pattern was still the same as with the soil simulations, in that there was a dramatic peak after the second simulation followed by a steep decrease in loading with the third then fourth simulations. This was not as defined with honey herb though as it still had lower values for the second and third simulations than the chicory (P<0.0001, P<0.0001) and oilseed radish (P=0.0001, P<0.05) but not the fourth for either (NS). During the second simulation ryegrass lost around 47% of the total plant biomass P in leaching, honey herb lost around 19%, chicory around 72% and oilseed radish around 79%.





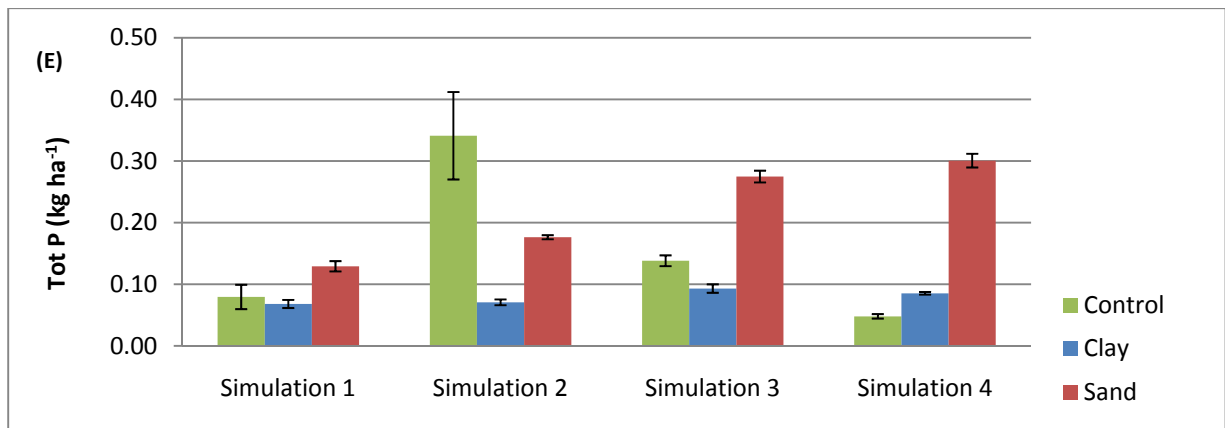


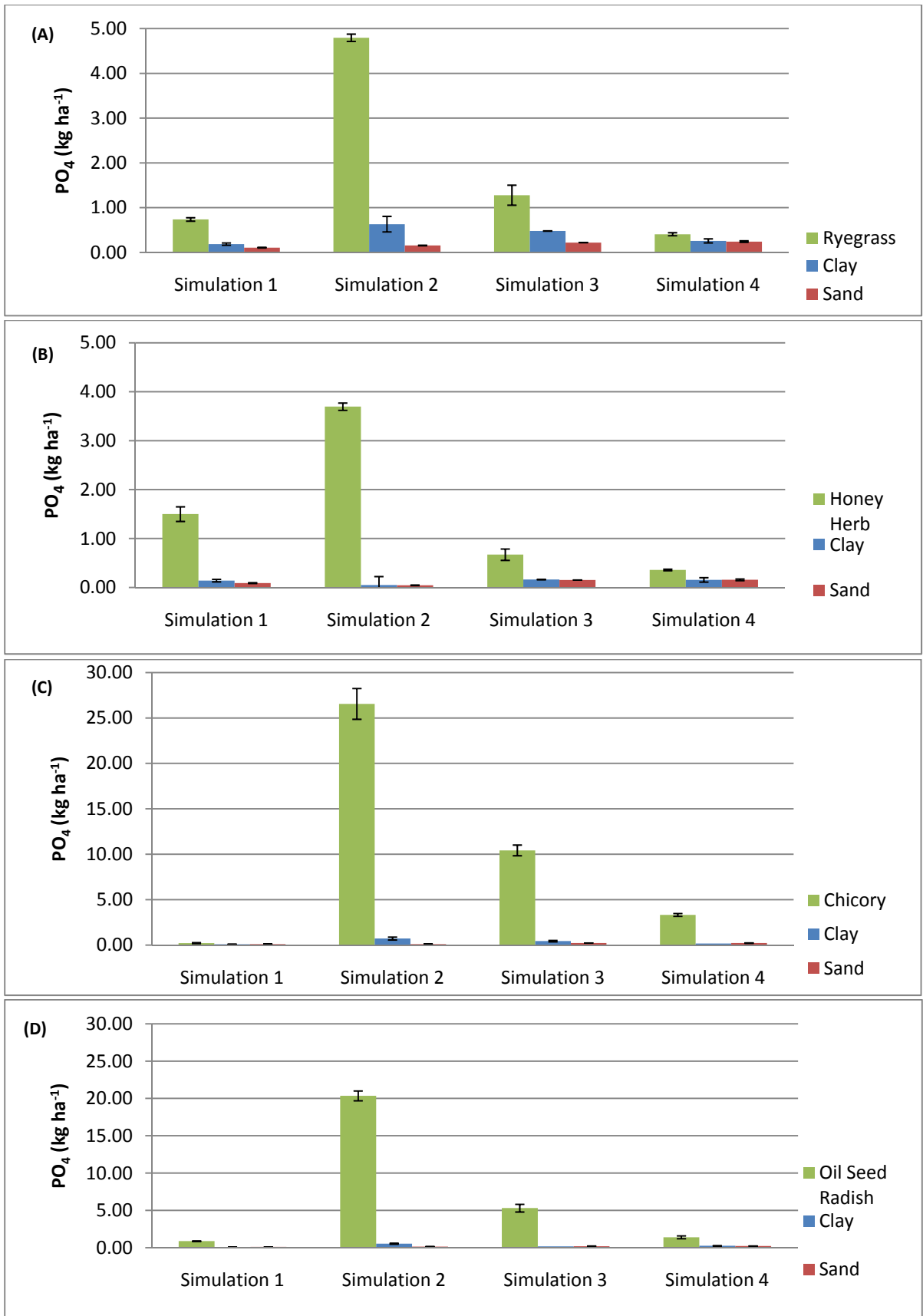
Figure 4: The comparison of mean total P leached from five treatments (without soil) with total P leached from plants through clay and sand soils. A= ryegrass, B= honey herb, C= chicory, D= oilseed radish and E= control. Bars are standard error of the mean. Note different scales for some figures.

Of the total P, most of it was in the form of $\text{PO}_4\text{-P}$ (~50-90%), with only a few exceptions in the honey herb and control treatments, where the remainder of unanalyzed forms of P dominated.

The $\text{PO}_4\text{-P}$ leaching load from the first simulation (figure 5) was significantly greater from the honey herb than any of the other treatments ($P < 0.05$). In the second simulation, honey herb, ryegrass and the control treatments were not statistically different, but the chicory and oilseed radish had significantly greater ($P < 0.0001$) leaching loads than the other three treatments, with chicory having the greatest losses overall, and significantly more than oilseed radish ($P < 0.05$). This simulation had the greatest leaching loads of the four simulations.

The third simulation showed again that chicory had the greatest leaching load, with a P value of < 0.0001 when comparing against ryegrass, honey herb and control, and a P value of < 0.0006 when comparing oilseed radish. Ryegrass and honey herb were the lowest and not different from one another, or from the control treatment.

The fourth simulation again showed chicory had the highest loading, with a P value of < 0.0001 when comparing chicory to ryegrass, honey herb and the control treatment and $P = 0.0001$ comparing chicory against oilseed radish. Ryegrass again had the lowest losses, and along with honey herb were both significantly lower than oilseed radish ($P < 0.05$).



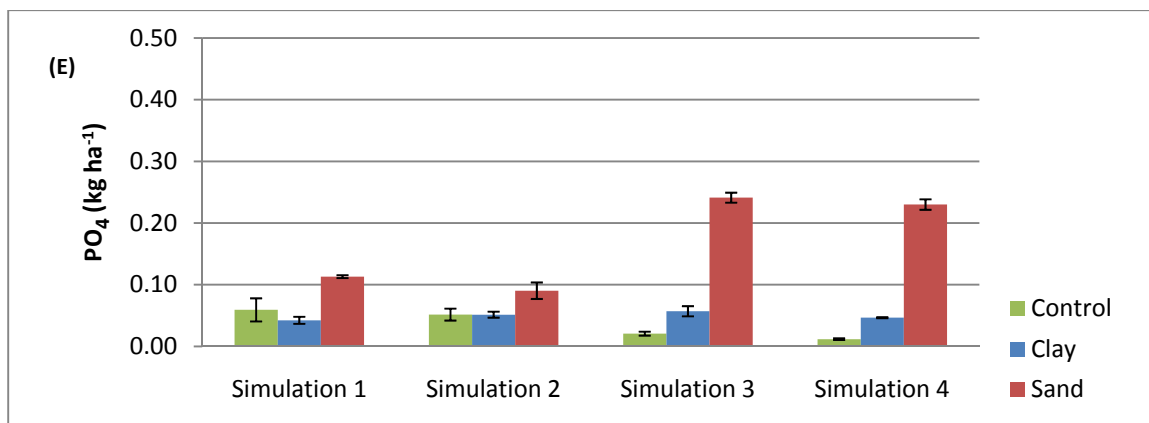


Figure 5: The comparison of mean PO₄-P leached from five treatments (without soil) with total P leached from plants through clay and sand soils. Bars are standard error of the mean. Note different scales for some figures.

It is difficult to give an overall accurate total loss from each of the treatments and experiments due to the variability of the results, though table 8 is a rough calculation from the soil and plant experiment. Table 11 shows the total P extracted from samples of each of the plant treatments before the experiment and table 12 the analysis results from the plant material left after the plant only experiment was completed. The starting value for the honey herb is minus the root value, as there was not enough root mass available to do an accurate analysis. Even though there were no root values for honey herb, table 10 shows the composition of total P from both tops and roots for the rest of the plants, and roots tend to contribute very little in the ryegrass in comparison to the tops.

Table 10: Total P content of the different plants and plant parts in mg per kg of dry weight before the experiment commenced. Also included is the total P as a percentage of total plant biomass. * indicates the data is unavailable.

Plant	Plant Part	Tot-P (mg/kg)	Total-P of Plant Material (%)
Ryegrass	Top	7287.0	0.729
	Roots	1891.8	0.189
Honey Herb	Top	7305.0	0.731
	Roots	*	*
Chicory	Top	9510.9	0.951
	Roots	6051.3	0.605
Oilseed radish	Top	7871.0	0.787
	Roots	9076.2	0.908

Table 11: The plant mean application of total P per lysimeter based on an initial total-P plant analysis. * Indicates that honey herb roots are missing.

Plant	Tops (kg tot P ha ⁻¹)	Roots (kg tot P ha ⁻¹)	Total (kg tot P ha ⁻¹)
Ryegrass	10.58	2.43	13.01
Honey Herb	36.86	*	36.86*
Chicory	42.76	1.63	44.39
Oilseed Radish	33.49	3.25	36.74

Table 12. Plant dry weight analysed Total-P from extra plant material at the experiment start compared with a plant analysis of plants after the plant only experiment. Due to a difference in applied initial plant fresh weight, the last column illustrates if all species were applied at 100g fresh weight (tops and roots) (see Table 1). Note *honey herb start of experiment does not include roots whereas all the other species do.

Plant	Mean plant Total-P at start of experiment (kg ha ⁻¹ DW)	Mean plant Total-P remaining after experiment (kg ha ⁻¹ DW)	Mean plant Total-P lost during experiment (kg ha ⁻¹)	Mean plant Total-P lost if all initial plant applied fwt is 100g (kg ha ⁻¹)
Ryegrass	13.01	3.38	9.63 (74%)	23.38
Honey Herb	36.86*	22.62	14.24 (39%)	8.02
Chicory	44.39	6.91	37.48 (84%)	22.72
Oilseed Radish	36.74	8.83	27.90 (76%)	15.86

Even though there was no statistical analysis completed to compare the losses in table 12, it is not required to identify some stand out trends. When comparing the added plant fresh weight and the included total P per lysimeter, ryegrass had one of the lowest initial total P contents and also the lowest losses of P. However when looking at the percentage of total P lost from the initial starting total P, honey herb lost only ~39% (not including root content) which is close to half the losses of the next closest, ryegrass (table 12). It is more obvious with the last column in table 12 which shows if an equal fresh weight of 100g of each plant material (both tops and roots combined) is applied to eliminate actual loadings as an issue, what the losses of total P will then be. The highest plant P uptake are from chicory, but it also has the greatest losses with 84% of the initial starting total P lost in leaching from the plant by the end of the experiment.

Discussion

This experiment is different from other previous similar studies of catch crops and P leaching in that the ability to remove the soil affect from the equation allows a true comparison carried out in this study of the different crops selected. By combining the two individual experiments carried out in this study it allows a better understanding of the different soil and plant components involved and their part in the leaching process.

It is quite obvious from the results that there are differences in leaching loads from the two different soils, though not always statistically significant (table 5). The clay soil did have greater leaching losses than sand in many occasions, but this tends to go against some authors reasoning. Syversen and Borch (2005) concluded from their experiment that P content increased only in their clay soil, stating that there is higher P adsorption potential of small clay particles, due to their relatively larger surface area. This did occur in this study, but it was much more noticeable in the sand soil which behaved more like a buffering system than the clay soil, though for possibly different reasons than the clay.

There are also many authors that found reasons for why similar actions of clay and sand soils in their research occurred. Bergström and Shirmohammadi (1999) carried out an experiment comparing clay and sand soils to see if there were any differences in flow penetration over the depth of the soil profile using a dye staining method. They found that the sand soil had a greater distribution of water across, as well as down the soil profile than the clay, with only a small fraction of the clay soil matrix being infiltrated by the dye. This is due to the occurrence of preferential flow (Kleinman et al., 2005; Thomas et al., 1997) occurring in clay soils, this being the fast movement of solutes to the lower soil layers or even to the groundwater (Sinaj et al., 2002). Flury et al. (1994) found in their experiment of observing water flow through soils using dyes, that the range of soils had quite different flow patterns, with most soils bypassing the soil matrix, thus reducing the effective sorption capacity of the soil (Thomas et al., 1997).

The preferential flow is typically caused by the presence of root channels, worm holes and cracks in the soil. This action is in comparison to the also recognized 'saturated' or 'piston' flow (Haygarth and Jarvis, 1999) common in sandy soils. During this flow type there is a higher chance that the P in solute will be slowed down or stopped by presence of more pathways and from the presence of soil particles blocking flow pathways (Tindall et al., 1986) allowing for the binding of P.

The soil test results (table 2) illustrate some similarities and some differences between the two soils. The soils are quite different in terms of the ammonium lactate P measured (P-AL), with the sandy Nântuna soil results between 11.6 and 15.5 mg 100 g⁻¹, and the clay Brunnby soil at 4.7 mg 100 g⁻¹. This is classed as a 4 (a) or 4 (b) for the sandy soil and a 3 for the clay soil out of a possible five classes used to categorize available soil P. This suggests an already larger pool of P 'available' for leaching in the sand; though further insight into this testing method is required. The test itself was designed for more acidic soils using an extraction pH of 3.75, (do Carmo Horta et al., 2010).

The two soils in this study have a pH of between 6.2 and 7 (table 2), which is clearly more neutral to alkaline. Guo and Yost (1999) found in testing the ability of this extraction method that carbonate present in some soils tended to increase the pH of the oxalate solution and therefore give an unreliable result for predicting available P. Since there is a relatively high pH in these soils and due to the high amounts of ammonium lactate extractable calcium present (table 2), which could be potentially bound to the P and immobilizing it (Neyroud and Lischer, 2003), the test could give an unreliably high estimate value of P.

The difference between the soil test results could also be a good indication as to why the sand control results in figures 2 and 3 were always higher than that of the clay soil. There could also be some P released from the freezing and thawing process affecting the buried twitch weeds that were prevalent in the sand soil, which have shown to release high concentrations of PO₄-P. Bechmann et al. (2005) found that the presence of roots in the top 0-1 cm of soil significantly increased the P content in the soil.

The differences between the leaching loads from the plant species tested were variable and large with similar trends found between the soil plant experiment and the plant only experiment. It is quite obvious that the differences in leaching loads between the plant species were a lot less in the sand soil due to the reasons mentioned in the previous section. For that reason and the fact that there are also many clay soils used in agriculture as well, the comparison of plants will concentrate only on the clay soil treatments. To try and understand why there were such high leaching loads from some plants and low from others, a review of the experiment and other authors work in similar research will be used to see if there were any unusual occurrences in this experiment.

Bechmann et al. (2005) carried out a similar experiment to this with catch crops and freeze thaw cycles and found their ryegrass had a total P content of 5800 mg kg⁻¹ dry matter, though their results concluded that the leaching losses from the ryegrass plants did not contribute to elevated P levels in the leachate. Ulén (1997) evaluated a ryegrass catch crop that had a total P of 3800 mg kg⁻¹ dry matter and also found that losses from this catch crop were too low to be of concern. In this experiment ryegrass had a total P in the tops of around 7000 mg kg⁻¹ dry matter. Belesky et al. (2001) reported P contents of 4400 mg kg⁻¹ in a young crop of chicory and 3700 mg kg⁻¹ in an older crop, in this experiment it was 7305 mg kg⁻¹. This may give some indication that the P concentrations in the plants were elevated in this trial, likely due to the ideal indoor growing conditions experienced and the good supply of P in the fertilizer applied. Table 10, gives the percent of total P in plant biomass before the leaching, ranging from 0.95 in chicory, to 0.79 in oilseed radish, 0.73 in honey herb and 0.73 in ryegrass. It was difficult to find research on biomass P levels in all the studied plants, but when comparing values found in this experiment with other previous research from different crops, these current values seem quite high.

For example Wilczewski and Skinder (2009) found 0.4% in honey herb and 0.54 in oilseed radish residues, also commenting that oilseed radish had the highest P content over the 3 years of the trial (including sunflower and honey herb). Timmons et al. (1970) reported total P contents of 0.46% in lucerne and 0.32% in bluegrass plants.

Crop residues from research carried out by Schomberg and Steiner (1999) found the P content in lucerne to be 2360 mg kg⁻¹, grain sorghum 1007 mg kg⁻¹, spring wheat 1005 mg kg⁻¹, corn to be 730 mg kg⁻¹ and winter wheat to be 450 mg kg⁻¹. Jalali and Ranjbar (2009) found sunflower residues to contain 2400 mg kg⁻¹ and potato residues 2200 mg kg⁻¹ of P. This illustrates that P contents can change depending on the plant species and the growing conditions experienced. Miller et al. (1994) noted that increased N applied to a prior wheat crop caused a substantial increase in P concentrations in leachate from a following catch crop. Sharpley and Reed (1982) concluded that increased applications of P fertilizer increased total P content of cotton (*Gossypium hirsutum* L.), sorghum (*Sorghum sudanense* (Piper) (Stapf.)), and soybean (*Glycine max* (L.) (Merr.)), but not for little bluestem (*Andropogon scoparius* Michx.). This indicates that the requirements of each plant for P, or the ability of each plant to be able to take up luxury amounts, beyond that of what is required to sustain it, is dependent on the plant itself. This may explain why the plants had the initial P contents they did (table 10).

The actual loading of the plant material used in the experiment can also be a potential cause of the high leaching loads experienced. Miller et al. (1994) found that an increase in loading of dry matter from 2000-4000 kg ha⁻¹ in both ryegrass and oilseed radish high nitrogen treatments caused a significant difference in leaching concentrations of P in their indoor plant leaching experiment, with the higher loading resulting in greater leaching losses. Table 1 shows the loading of plant material ranked from highest application with honey herb (5152 kg DM ha⁻¹) to the lowest ryegrass (2765 kg DM ha⁻¹) with chicory (4813 kg DM ha⁻¹) and oilseed radish (4658 kg DM ha⁻¹) in between. With a higher loading rate, one would logically expect higher leaching loads of P, as shown previously, which was not in fact the case (figure 2, figure 5, and table 12). Instead the highest loading rate from the honey herb gave some of the lowest leaching losses in both experiments, and even the ryegrass with the lowest applied loading (table 1) gave some of the highest losses (e.g. figure 2 a, b, c). The data from table 11 which gives the mean plant total P before the experiment began, with ryegrass being the lowest at 10.58 kg ha⁻¹, and honey herb the second to highest at 36.86 kg ha⁻¹ suggests that ryegrass should logically lose the least amount of P in leaching, but instead it was the opposite. This still does not explain the general high losses and the difference in plant leaching.

The procedure of cutting the plant into roots and tops could potentially have increased the losses of P from the plant in the first simulation but when this technique was compared to a similar experiment conducted by Timmons et al. (1970) who used lucerne plants, they found there was no difference if the cut end of the stem was submerged in water or not, suggesting it may not have an effect in this experiment.

The high losses of P from ryegrass and honey herb in the first simulation suggest a greater proportion of water soluble P present in easily accessible areas on the plant for the simulated rainfall to remove. This was also noted by White (1973) who found that leaching by rain or melting snow reduced the total amount of soluble ions in the plant.

In the second simulation all of the plants tested had much higher leaching losses than in the first simulation. This had to do with the freezing and thawing process on the plants. Heber (1967) suggested that the freezing causes damage to the membranes surrounding the protoplasm of plant cells. Timmons et al. (1970) found that one freezing and thawing cycle increased the soluble form of inorganic P by 46 times in lucerne (*Medicago sativa* L.) plants. Bechmann et al. (2005) noted that water extractable P (WEP) in ryegrass increased dramatically after freezing and thawing events, with only 0.9% of total P was actually WEP in the fresh plant material compared with 40% after freezing and thawing. Although they found that the WEP concentration increased in a log type scale with the number of freezing and thawing events, up to 6 freezing cycles, in this experiment the total P only increased after the first freezing and thawing cycle. Afterwards, total P decreased in all treatments apart from the honey herb treatment which increased loadings in leachate from the second to third simulation (figure 2). Miller et al. (1994) reported P leaching losses from oilseed radish and ryegrass were around 30% of total biomass P. Jones and Bromfield (1969) carried out an experiment on hayed off glasshouse grown phalaris (*Phalaris tuberosa* L.) and noted that up to 80% of total P was leached from the plants. Research from Uusi-Kämpä (2007) found that between 60-80% of total biomass P was lost from the buffer strips during freezing and thawing cycles in her experiment with most of it occurring after the first cycle. These results are variable and similar to this experiment in which ryegrass lost around 47% of the total plant P in leaching; honey herb lost around 19%, chicory around 72% and oilseed radish 79%, after the first freezing and thawing event.

The structural composition of the plants is another possible point to consider when determining the reasons for variations between the different plants, Schomberg and Steiner (1999) stated that plant properties such as N, cellulose and lignin contents affect the decomposition rate and therefore the ability of P to be released. Although the decomposition was not 'natural' in the sense of not being exposed to outdoor living microbial decomposition, it is still a common natural winter occurrence in the north of Scandinavia. There was no structural composition analysis undertaken in this experiment, although there were definite visual differences between the species. The obvious large thick stem of the oilseed radish plant with thick water repelling leaves compared to the light and thin leaf of the chicory plant, which must give rise to differences in degradation ability. There were also the losses of total P from the second simulations that were similar between experiments, in that chicory had the highest leaching loads followed by oilseed radish or ryegrass and lastly honey herb. In the first simulation, with no freezing and thawing events having yet occurred, it was almost the completely opposite result. Chicory had the lowest and honey herb the highest losses. This illustrates the differing plant characteristics that protect some plants from rain induced leaching, and not others.

The recommendation of suitability of these plants to be used as catch crops is met with some hesitation. The chicory, oilseed radish and ryegrass all had very high losses at different stages of the two experiments. The results were sometimes mixed in terms of which plant leached the least, depending on which experiment.

For example, the total P lost for ryegrass in the soil plant experiment was greater than the honey herb, but was the reverse for the plant only experiment. To add to the confusion, the honey herb took up more P and retained more after the freezing and thawing had finished than the ryegrass. The main clear point was that oilseed radish and chicory had the greatest leaching losses throughout the two experiments and the before and after plant analysis. This agrees with results from Miller et al. (1994) who found that oilseed radish leached more P than ryegrass or red clover.

Although the greatest losses of P were from the clay soil, it does not necessarily mean that this soil type is particularly susceptible to P losses, and that catch crops on this soil are unsuitable. Bergström et al. (2007) suggested that the presence of a catch crop is important to remove excess amounts of potential leachable P from the soil, which in Scandinavia is commonly around 15–50 kg ha⁻¹. This is similar to what was found in this experiment and if the correct plant type is chosen, it can make a noticeable difference to leaching loads reaching the groundwater. The soil lysimeters were in fact only 250 mm long, and shallower than most groundwater would tend to be, which could lead to inaccurate field comparisons. Sinaj et al. (2002) found that even though they had preferential flow in their lysimeter experiment, they had very low total P loss in leachate, due to very high P fixing abilities of the subsoil (70 cm deep) and that the main flow paths ended in the soil matrix. Turner and Haygarth (2000) found that there was no significant difference in leaching losses from a range of soils, from sand to clay tested in lysimeters. These particular lysimeters were 1.35 m deep and therefore may have had a greater chance for P to adsorb to soil particles or bind to calcium or iron present in the soil. Djodjic et al. (2004) also noted that the ability of the subsoil to retain or lose P was an important part of limiting or increasing leaching losses.

There is the overwhelming difference between the two experiments presented in this paper, illustrating the large potential leaching loads presented to the soils. However, the total P concentrations in soil leachate and plant analysis suggest the plants were subjected to saturated P growing conditions and therefore leaching loads of P maybe higher than a typical outdoor growing environment, common to that in a farming situation.

Although there was less leaching losses from the sand soil, it is not to say that growing crops on sand soil is the answer. Over time with the imbalanced inputs of P combined with the lower outputs causes a gradual accumulation within any soil, which could increase the eventual chance of P transfer to the soil water (Sharpley and Rekolainen, 1997; Nash and Halliwell, 2000).

Possible future research could involve a larger outdoor lysimeter trial looking at the same crops to see whether these particular crops act in a similar way in regard to P as shown in this experiment. Combined with that, there is the possibility to also look at the soil types and maybe use dye tracers to see where the P solute from the plants actually goes, whether preferential flow paths are active in clay and if it is a general flow into the soil matrix in sandy soils?

Conclusions

The use of catch crops can help remove excess available soil P, but careful plant selection must be taken into account. This is due to the large variation in P released from the four chosen plant species in this experiment, with honey herb and ryegrass releasing less P than chicory and oilseed radish under freezing conditions. The soil texture can also be a factor in the increased or decreased risk of losing P to natural waters. The clay soil had generally greater losses of P than the sand in this experiment. This author also must point out that although these results suggest potentially high losses of P from plants, and resulting high soil loadings, the experiments were conducted in ideal indoor conditions, with large potential soil P supplies, and similar results may not occur in a natural outdoor environment.

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Appendix

1) Inductively Coupled Plasma (ICP) Method for determining total P in plant samples

Approximately 1.5 g dry weight of plant material was transferred to a 50 ml Kjeltec tube, 10 ml concentrated nitric acid was added and the samples were left over night to start the breakdown of organic matter. The samples were boiled with stepwise increasing temperature for one hour at 60°C, one hour at 100°C and four hours at 125°C. After two hours at 125°C, an additional 5 ml concentrated nitric acid was added. The samples were diluted to 50 ml with distilled water after cooling. The samples are then passed through an ICP machine (ICP Optima 7300 DV) to give total P values.

