

The effects of long-term detrital manipulation on micronutrients in soil solution

INTRODUCTION

Soil organic matter (SOM), which is derived from microbial and dead plant material, is a major pool of carbon in the biosphere and plays a crucial part in the global carbon cycle. It is an important determinant of soil aeration and moisture capacity. Immobilization and release of certain plant nutrients are also dictated by SOM, making SOM a key regulator of plant growth. SOM assumes many essential roles, but very little is understood about the biological, physical, and chemical processes that affect SOM dynamics.

SOM levels are determined by a balance between carbon inputs to soils as litter (i.e. leaves, wood, roots, and root exudates) and outputs from soils as particulate organic matter (POM) from erosion, dissolved organic matter (DOM) from leaching, or carbon dioxide from respiration. SOM will either be stabilized or lost depending on how well the litter inputs can withstand degradation pressures. Many short-term studies (2 to 5 years) have been done to understand this balance, but no one knows how the quantity and quality of litter might change over decades or centuries. With climate change, increasing levels of carbon dioxide and atmospheric nitrogen, air pollution, drought, and plant pathogens threatening to significantly alter the quantity and quality of future detrital inputs, it is important now more than ever to study long-term changes in litter inputs and the subsequent effect on SOM levels.

In order to observe these long-term changes in detrital inputs and how these changes will affect SOM accumulation and stabilization, the DIRT (Detrital Input and

Removal Treatments) experiment was established by Francis Hole in 1956 in the Wisconsin Arboretum. The experiment involved plots with different litter treatments, each of which were to be analyzed over an extensive timeline. These plots are still well-maintained today. Inspired by this experiment, several other plots were established in other parts of the country, including the H.J. Andrews Experimental Forest in western Oregon in 1997. Currently, the H.J. Andrews plots are the largest of all the DIRT establishments. Six treatments are in place in the H.J. Andrews: Control, No Litter, Double Litter, Double Wood, No Roots, and No Inputs. Lysimeters are installed in the H.J. Andrews sites, which allows for the study of leaching losses from the soil. Other DIRT establishments, such as those in the Harvard Forest in Massachusetts, do not have lysimeters. Instead, there are long-term incubations of the Harvard soils that are used to measure carbon respiration and leaching losses. Despite the different methods used in different DIRT locations, all the DIRT establishments ultimately allow researchers to assess how different detrital inputs dictate the SOM dynamics over decadal time scales.

The focus of most of the analyses thus far has been on carbon and nitrogen dynamics. As a result, we have analyses of SOM response to carbon inputs and removals as well as the effects of litter manipulation on nitrogen and dissolved organic carbon (DOC) presence in leachate. For example, a DIRT plot with no roots (i.e. lower carbon inputs) results in greater nitrogen release through leaching. Not only is plant uptake of nitrogen eliminated, but microbes cannot immobilize nitrogen without available carbon (Lajtha et al., 2005). Carbon and nitrogen has been studied intensively in DIRT, but detrital manipulation affects more than just carbon and nitrogen. Nevertheless, no other biogeochemical component has been studied to date in DIRT.

Calcium is one of these unstudied components. It is just as significant to forest ecosystems as carbon and nitrogen. Calcium, as well as other base cations like Mg^{2+} , K^+ , and Na^+ , helps to neutralize acidic conditions in forest floors. This is extremely important considering that atmospheric acid deposition of anions is increasing, especially in the eastern United States and the east coast of Europe. Unfortunately, calcium is being depleted in many forest soils. Hamburg et al. (2003) attributes this decline in calcium to the aging of forest stands in many acid rain affected parts of the world. Older stands cannot sufficiently mobilize calcium from the mineral soil. Less exchangeable calcium in the O horizon results in the mobilization of aluminum to the O horizon instead, causing destruction to forest vegetation (Cho et al., 2010).

In addition, no studies in the DIRT plots were done on the relationship between litter inputs and phosphorus availability or turnover. Phosphorus is released by the weathering of bedrock, after which it is biocycled by plants or precipitated out as compounds such as aluminum and iron oxyhydroxides (Ippolito et al., 2010). Wetter climates experience more weathering, but these conditions also show increased aluminum mobilization, sometimes producing more aluminum-bound phosphorous and smaller pools of available phosphorus. Phosphorus availability is also limited in older soils and when phosphorus advection through the soil column can no longer keep up with phosphorus loss and uptake (Porder et al., 2007). Thus, phosphorus, after nitrogen, is one of the most limiting nutrients in ecosystems.

We know that calcium can be affected by acid deposition and we know that phosphorus availability is related to weathering, biocycling by plants, and aluminum mobilization. What we do not know is how detrital inputs affect calcium and phosphorus

levels. DIRT showed us that the amount and types of detrital inputs altered SOM levels, but will this transformed carbon pool alter calcium and phosphorus loss like it did nitrogen loss? I answered this question by focusing my thesis research on how different quantities and qualities of detrital inputs affect calcium and phosphorus dynamics.

METHODS

My thesis is separated into two parts: measuring calcium and phosphorus levels first from the H.J. Andrews Forest in lysimeter leachates and second from laboratory incubations in soils from the Harvard Forest DIRT plots.

DIRT plots of the H.J. Andrews Forest in western Oregon were set up in an undisturbed old-growth Douglas-fir-western hemlock stand. The soils of the H.J. Andrews are basaltic, phosphorus rich, and weathers rapidly due to the 2370 mm of precipitation per year (1973-2002 period). At the H.J. Andrews, each of the six detrital treatments is replicated 3 times. Those treatments are Control, No Litter, Double Litter, Double Wood, No Roots, and No Inputs (Table 1). All plots contain five Prenart Superquartz tension lysimeters, three at 30 cm and two at 100 cm. One other treatment, OA-Less, is present at the H.J. Andrews, but there are no lysimeters installed in those plots and therefore will not be involved in my research. The main objective for the H.J. Andrews DIRT plots in 2011 is to collect and analyze lysimeter samples. I, along with a graduate student involved in DIRT, went to the H.J. Andrews to collect soil solutions from lysimeters. To collect soil solutions, bottles were attached to tubing that runs into each lysimeter. A handheld pump was used to introduce 15 psi of negative pressure, allowing leachate to flow into the bottle. The bottles were left on site for 48 hours, after which they were collected, brought back to the laboratory, and stored in sample vials.

DIRT plots of Harvard Forest in Petersham, Massachusetts were set up in a mid-successional oak-maple-birch forest in 1990. Like the DIRT experiment in the H.J. Andrews Forest, Harvard Forest has Control, No Litter, Double Litter, No Roots, No

Inputs, and OA-Less treatments (Table 1). However, there is no Double Wood treatment. Soils were collected in 2010 as part of a larger study to examine changes in plots after 20 years of DIRT manipulations. I assisted a graduate student who is incubating the Harvard soils to measure SOM lability. Incubations from the O horizon contained approximately 40 g of dry soil and incubations at depth contained approximately 70 g of dry soil. Using the same incubation setup, I leached the soils every two weeks for the first two sampling events and then every month for the last two samplings. 80 ml of leachate solution was poured into each of the 95 incubations. The leachate was comprised of 4 mM $\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$, 2 mM KH_2PO_4 , 1 mM K_2SO_4 , 1 mM $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$, 25 μM H_3BO_3 , 2 μM $\text{MnSO}_4 \cdot \text{H}_2\text{O}$, 2 μM $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$, 0.5 μM $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$, 0.5 μM $\text{Na}_2\text{MoO}_4 \cdot 2\text{H}_2\text{O}$, and 0.2 mM $\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$. After the leachate was allowed to sit for 1 hour, the incubations were placed on a vacuum. The vacuum drained the leachate solution out of the incubations and into side arm flasks. The leachate was poured into sample vials, and like the H.J. Andrews lysimeter samples, were analyzed for phosphorus and calcium.

Table 1. Detrital treatments for the H.J. Andrews and Harvard Forest DIRT experiment

Treatment	Method
Control	Normal litter inputs are allowed.
No Litter	Aboveground inputs are excluded from plots by raking.
Double Litter	Aboveground inputs are doubled by adding litter removed from No Litter plots.
Double Wood (only present at HJA)	Aboveground wood inputs are increased by adding large shredded wood pieces based on rates of C addition to the Double Litter plots.
No Roots	Roots are excluded by impenetrable barriers extending from the soil surface to the top of the C horizon.
No Inputs	Aboveground inputs are prevented as in No Litter plots; Belowground inputs are prevented as in No Roots plots.
OA-Less (will be used from Harvard only)	Organic and A horizons are replaced with B horizon soil at the start. Normal inputs are allowed thereafter.

In addition to measuring calcium and phosphorus, the H.J. Andrews lysimeter and Harvard incubation samples were analyzed for nitrate and ammonia. Phosphorus, nitrate, and ammonia were analyzed using an Alpkem Flow Solution IV. Data were organized using WinFLOW V3 software (WinFLOW V3 Manuals and Methods, 2008). Calcium was analyzed using an AAnalyst 100 Atomic Absorption Spectrometer (AAnalyst 100 User's Guide, 2000).

Phosphorus was measured as orthophosphate as an antimonyphosphomolybdate complex. Ascorbic acid reduces this complex, resulting in a blue color that is measured at 880 nm (Low-Level Orthophosphate by Segmented Flow Analysis).

Nitrate was measured as nitrite after cadmium reduction. The nitrite is then determined as an azo dye at 540 nm after its diazotization with sulfanilamide and coupling with N-1-naphthylethylenediamine dihydrochloride (Nitrate+Nitrite Nitrogen and Nitrite Nitrogen). As a modification to this method, a column containing cadmium granules was used instead of a cadmium hollow tube.

Ammonium was measured as ammonium nitrogen. Ammonium in the sample reacts with a hypochlorite-containing alkaline solution (originated from dichloroisocyanurate). Chloramine forms and reacts salicylate and nitroprusside at 37°C, which results in a blue-green dye that is measured at 640nm (Ammonium Nitrogen by Segemented Flow Analysis).

The amount of calcium present in the samples was determined by atomic absorption. An air-acetylene flame is used to convert the sample aerosol into atomic vapor. The atoms are then placed in a light path of 422.7 nm. The amount of light

absorbed is used to determine the amount of calcium present (Standard Atomic Absorption Conditions for Ca, 2000).

The Alpkem Flow Solution IV and the AAAnalyst 100 were provided by the Institute for Water and Watersheds (IWW) Collaboratory at Oregon State University. All general lab protocols are in accordance with the EPA (Willamette Research Station Analytical Laboratory Quality Assurance Plan, 2004).

ANOVA statistics were performed for all collected data using R (R 2.13.0, 2011).

RESULTS

Harvard Forest incubations *Calcium*

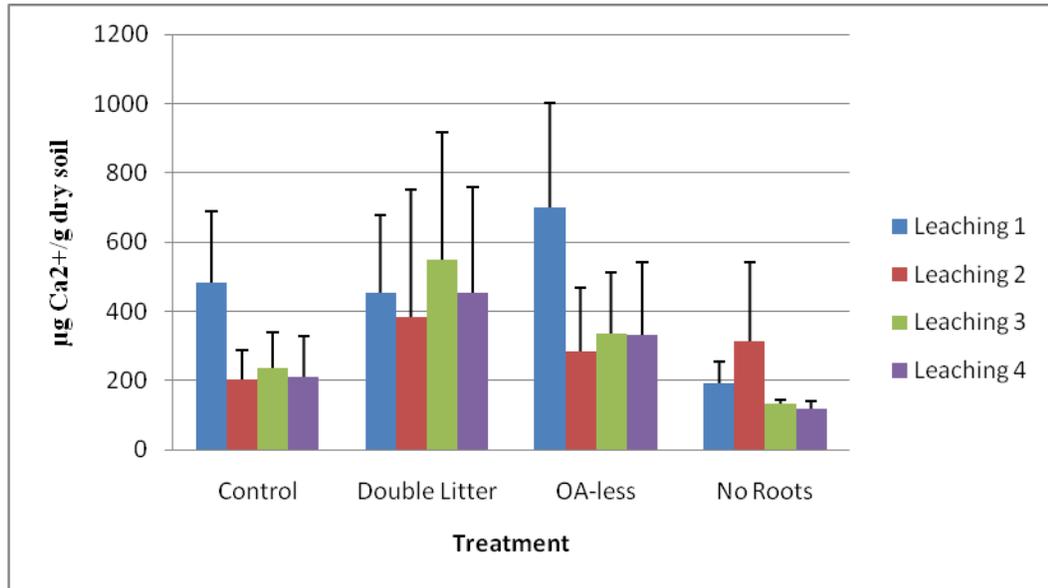


Figure 1.1 Average calcium levels of Harvard leachate from the O horizon. No Roots plots produced the least amount of calcium in leachate, an indication of the high calcium levels in root cell walls. The No Roots plot is statistically significant in the first and third leaching events.

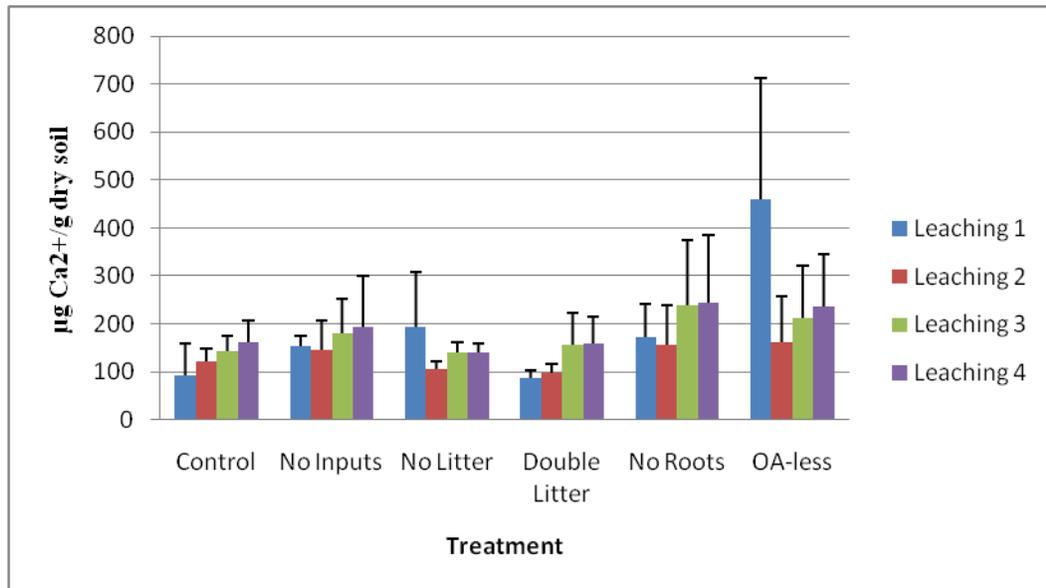


Figure 1.2 Average calcium levels of Harvard leachate at 0-10 cm depth. The overall consistency in calcium levels between the treatments suggests the absence of calcium-containing root material at lower depths. The OA-less plots are statistically significant in the first leaching event, which indicates possible calcium in mineral layers.

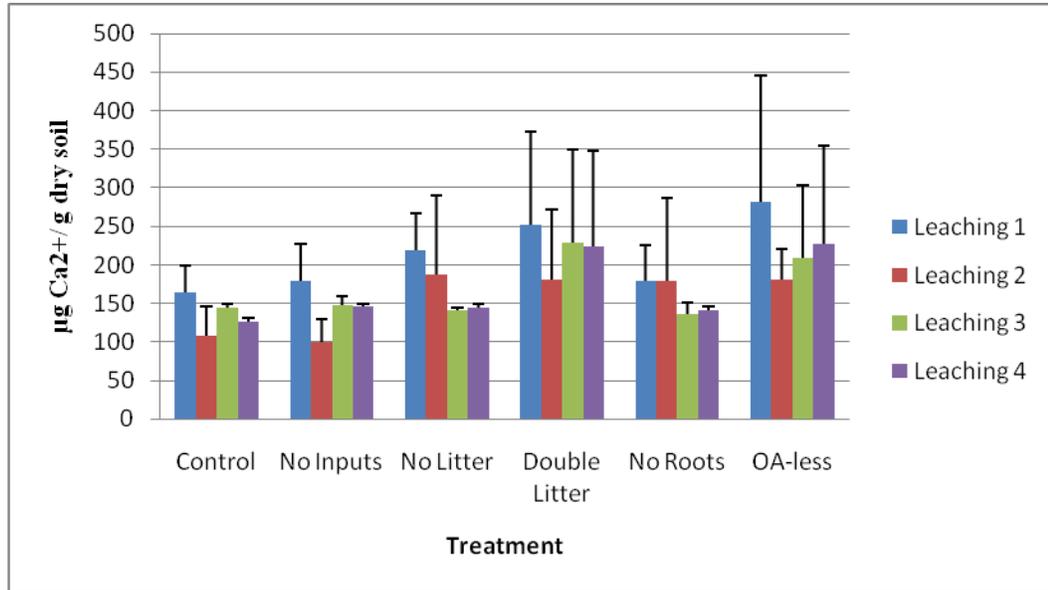


Figure 1.3 Average calcium levels of Harvard leachate at 10-20 cm depth. There are no statistically significant differences in calcium levels between the treatments. Again, this suggests the absence of calcium-containing root material at lower depths.

Phosphorus

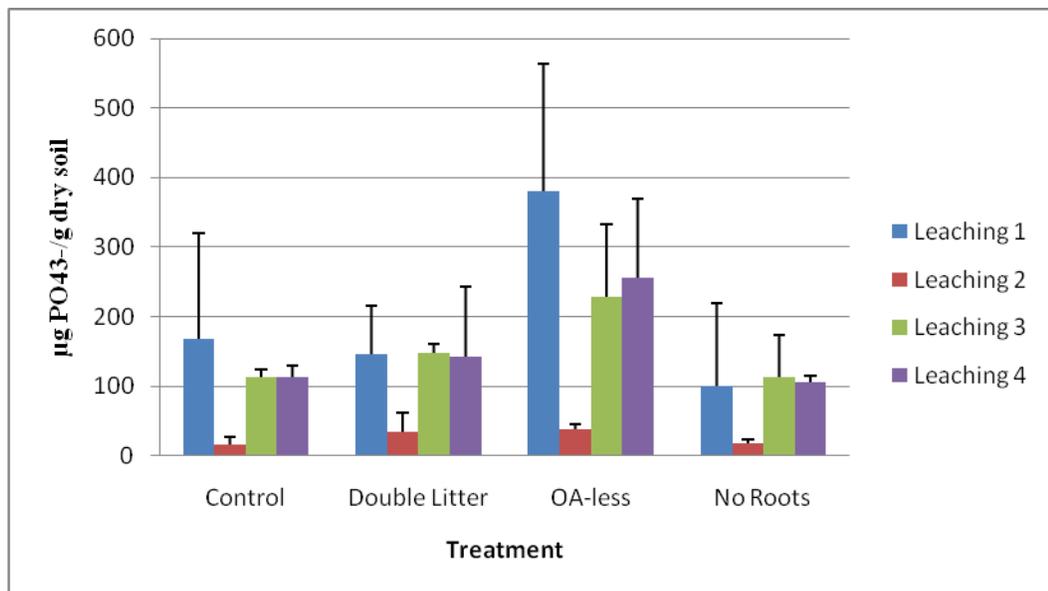


Figure 2.1 Average phosphorus levels of Harvard leachate at the O horizon. The OA-less plots appears to have produced more calcium in leachate than the other treatments, but these plots were not statistically significant in comparison to the other treatments.

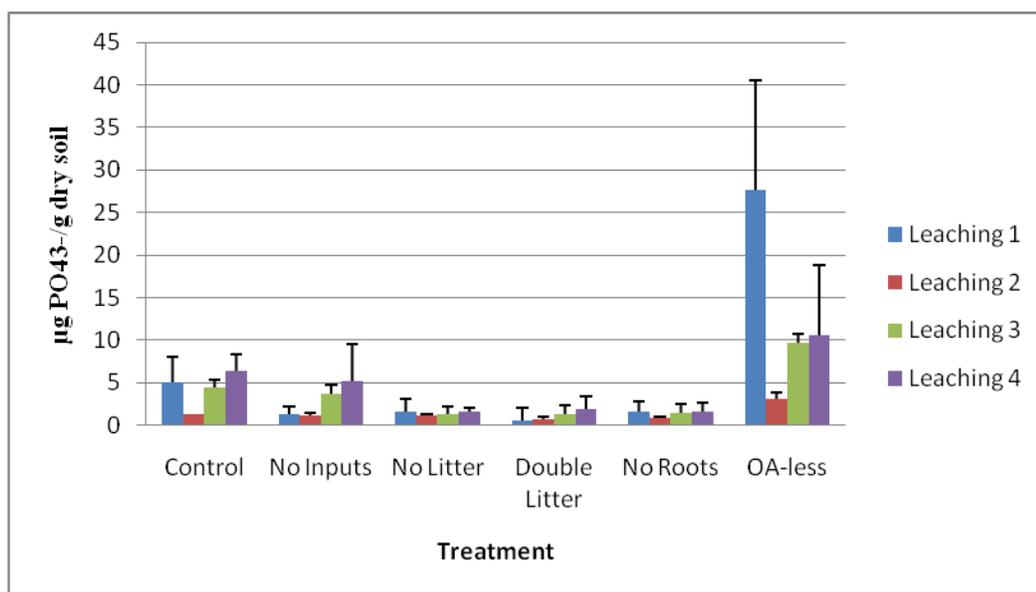


Figure 2.2 Average phosphorus levels of Harvard leachate at 0-10 cm depth. All four leaching events for the OA-less plots are statistically significant; these plots produced more calcium in leachate than the other treatments. This is an indication of bedrock in B horizon soil.

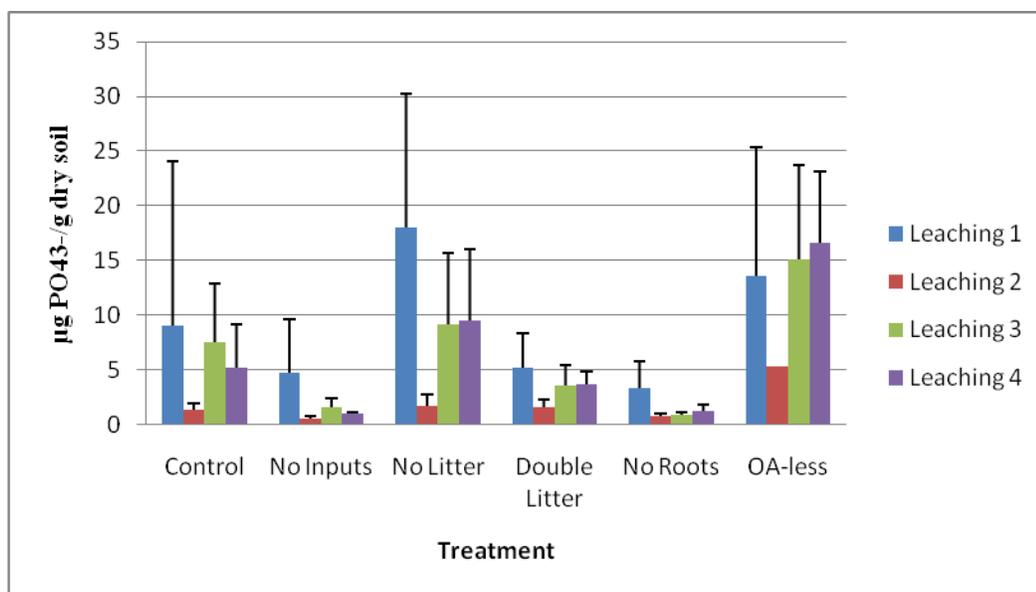


Figure 2.3 Average phosphorus levels of Harvard leachate at 10-20 cm depth. The second and fourth leaching events for the OA-less plots are statistically significant; these plots produced more calcium in leachate than the other treatments. Although not all the leaching events were significant, this is still a likely indication of the bedrock in B horizon soil.

Nitrate

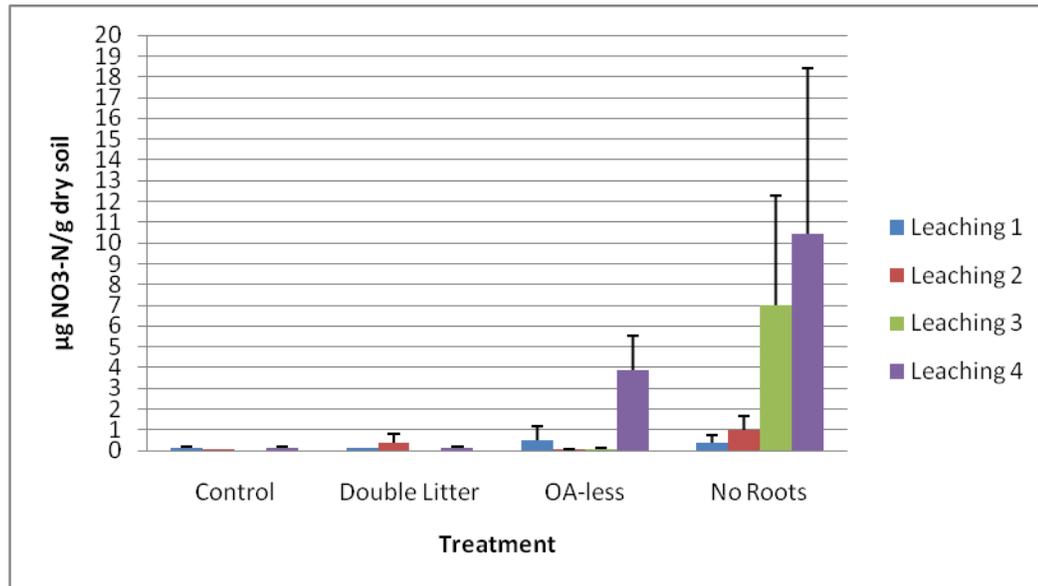


Figure 3.1 Average nitrate levels of Harvard leachate at the O horizon. Overall, higher nitrate levels were detected in the No Roots plots in comparison to the other treatments. This reflects the idea that less organic matter results in lower nitrogen immobilization. The third leaching event for the No Roots plots is statistically significant.

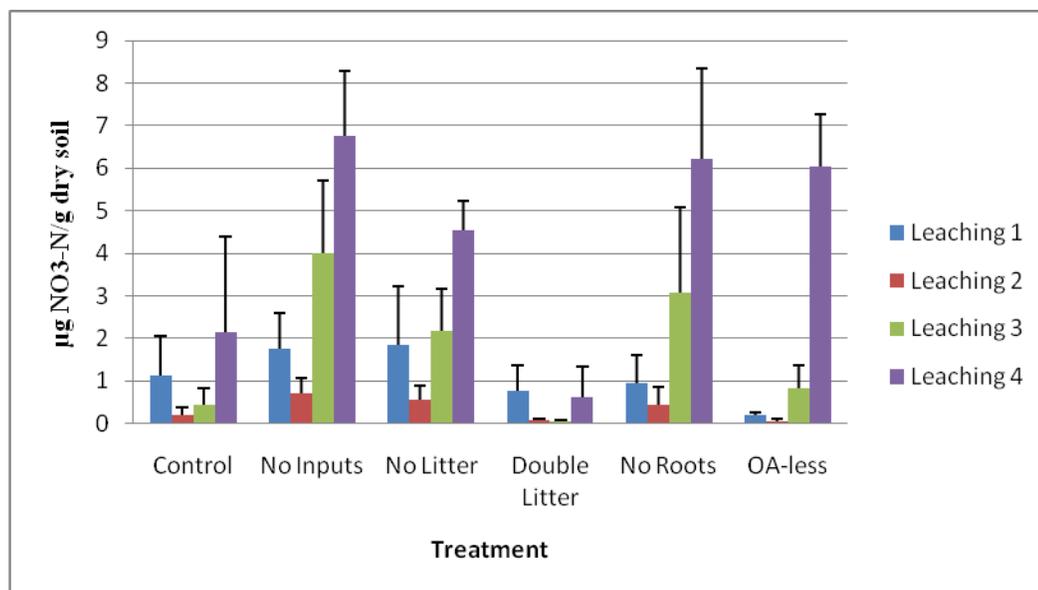


Figure 3.2 Average nitrate levels of Harvard leachate at 0-10 cm depth. The Control and Double Litter plots had the lowest levels of nitrate, with the Double Litter plots being significant for the third and fourth leaching events. This demonstrates the increased nitrogen immobilization seen with increased organic matter input. Likewise, the No Inputs, No Litter, and No Roots plots had leaching events in which they had higher nitrate levels in leachate, which illustrates the lower nitrogen immobilization seen with the absence of organic matter.

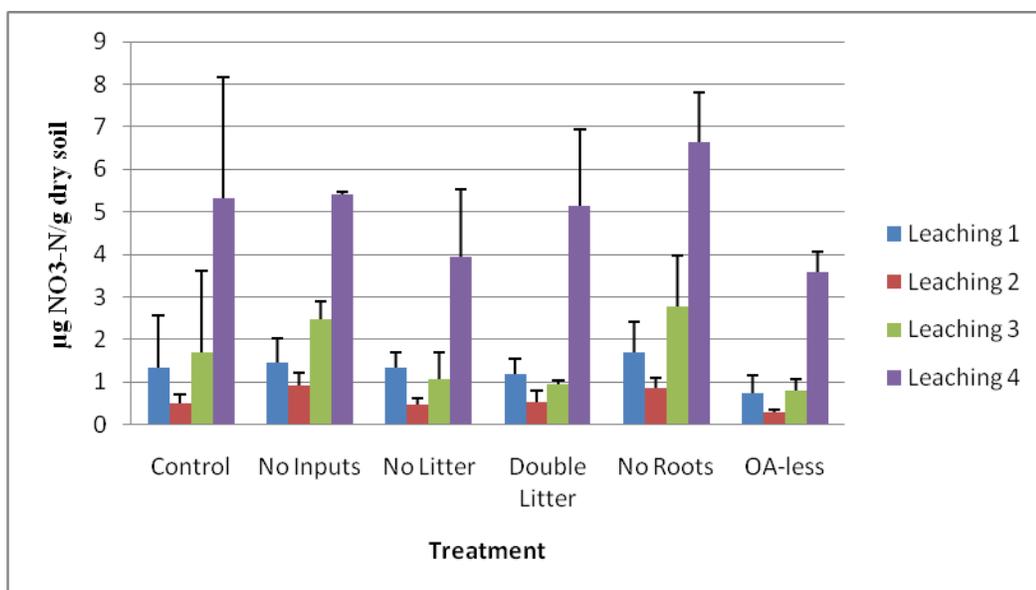


Figure 3.3 Average nitrate levels of Harvard leachate at 10-20 cm depth. There are no statistically significant differences in nitrate levels between the treatments. However, by the last leaching event, all the treatments had a high level of nitrate being leached out, which shows that nitrate is being mobilized regardless of treatment. This is likely due to increased activity from nitrifying bacteria.

Ammonium

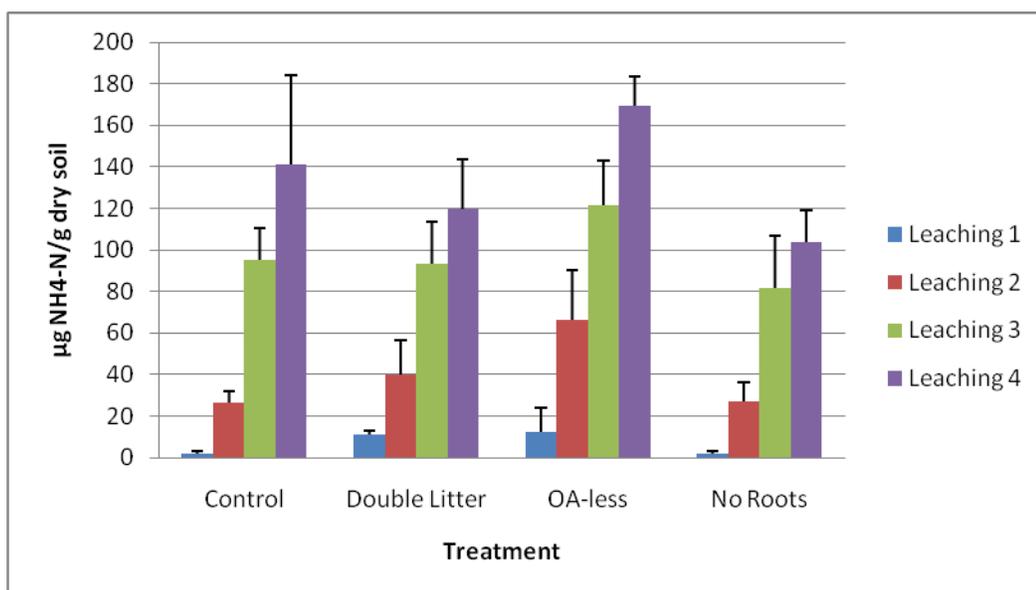


Figure 4.1 Average ammonium levels of Harvard leachate at the O horizon. There are no statistically significant differences in ammonium levels between the treatments. This is likely due to the cation exchange capacity of the soil, limiting ammonium's mobility.

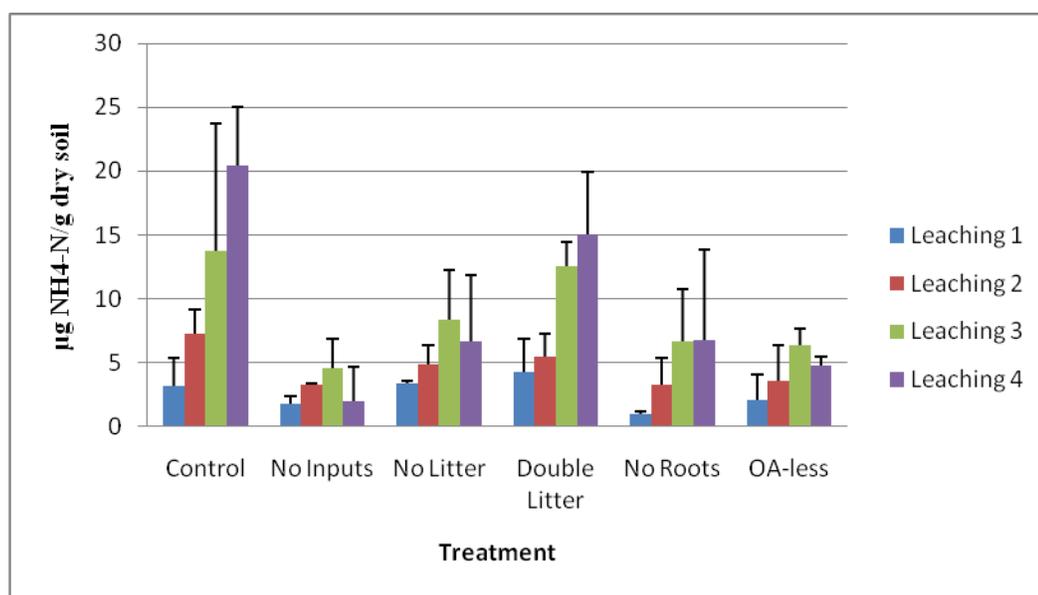


Figure 4.2 Average ammonium levels of Harvard leachate at 0-10 cm depth. Unlike Figure 4.1, the third and fourth leaching events for No Inputs plots are statistically significant. This is likely due to the cation exchange capacity of the soil, limiting ammonium's mobility, as well as the fact that No inputs plots do not have the organic matter necessary to produce ammonium.

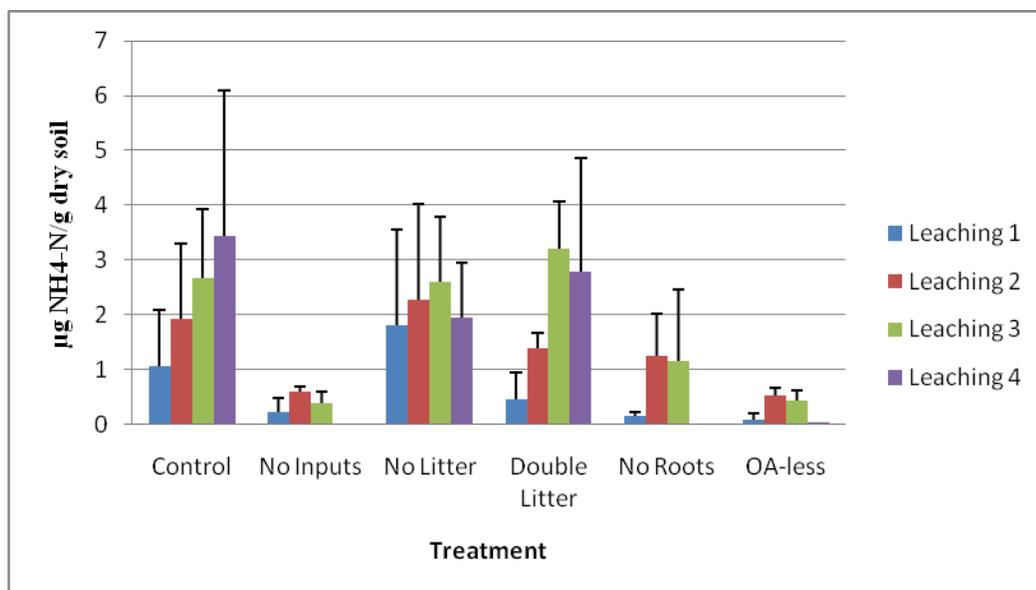


Figure 4.3 Average ammonium levels of Harvard leachate at 10-20 cm depth. The No Inputs and OA-less plots had lower levels of nitrate in their leachate than the other treatments, with the No Inputs plots being statistically significant in the third leaching event. The No Inputs plots have less organic matter to produce ammonium, resulting in less ammonium to be leached out. The B horizon soil of the OA-less plots likely contains a lot of clay particles that are binding ammonium. In general, with more depth, less ammonium is observable since one is moving further away from the litter layer.

H.J. Andrews lysimeter samples

Calcium

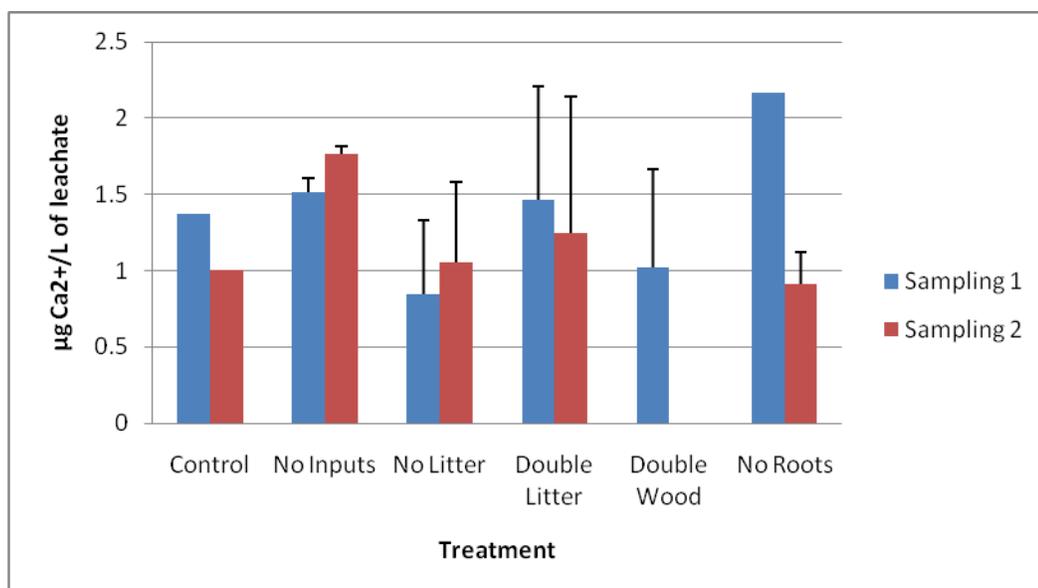


Figure 5.1 Average calcium levels of HJA leachate at 30 cm depth. For sampling 2, no Double Wood plot produced enough leachate for analysis. There are no statistically significant differences in calcium levels between the treatments. At 30 cm, there may be less root material; thus, calcium levels are likely too small to be affected by detrital manipulation.

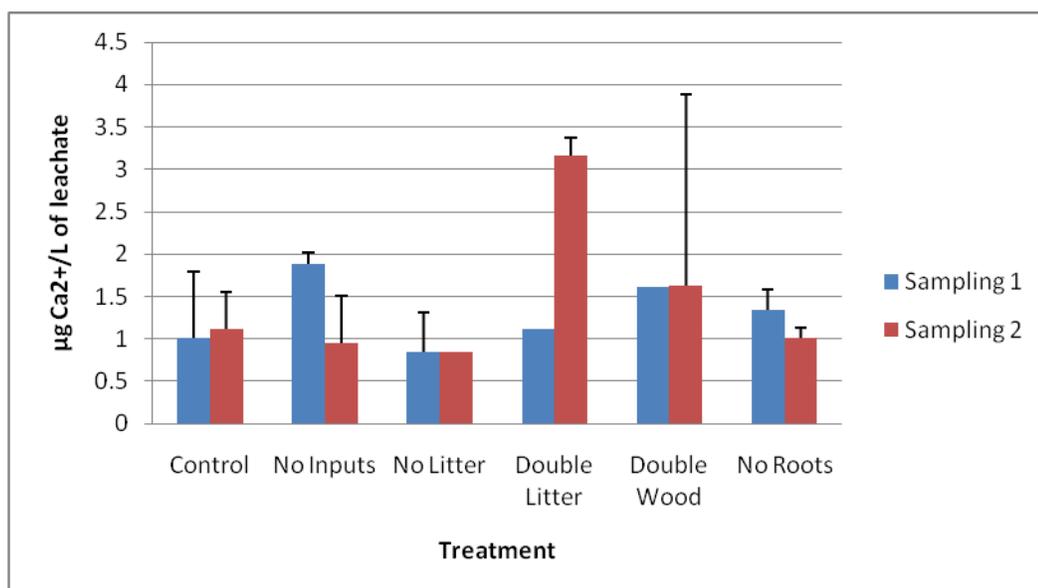


Figure 5.2 Average calcium levels of HJA leachate at 100 cm depth. Like Figure 5.1, there are no statistically significant differences in calcium levels between the treatments. At 100 cm, an even deeper sampling, root material would be scarce. Again, calcium levels are likely too small to be affected by detrital manipulation.

Phosphorus

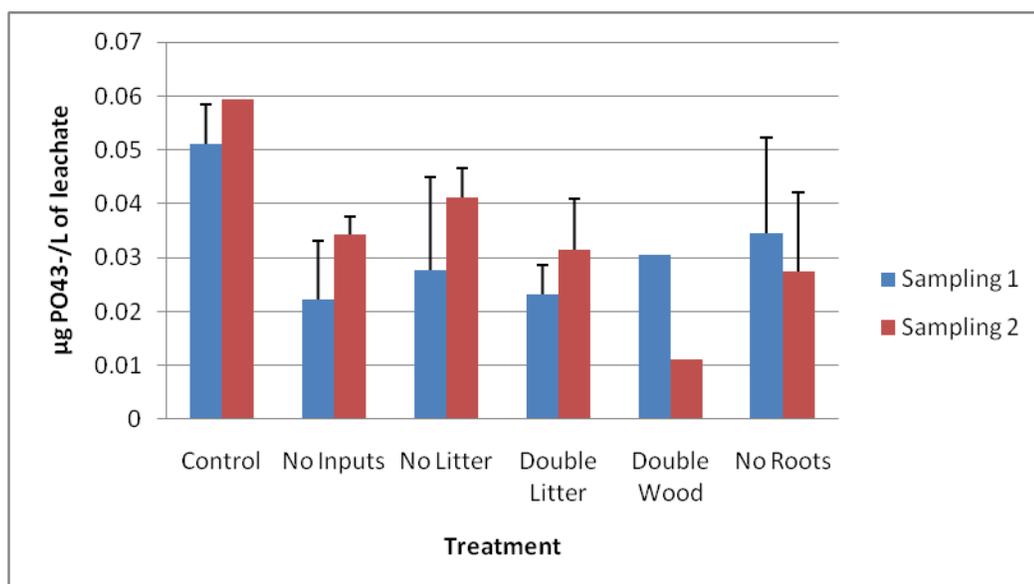


Figure 6.1 Average phosphorus levels of HJA leachate at 30 cm depth. There are no statistically significant differences in phosphorus levels between the treatments. They all produced a similar low level of phosphorus. We learned from the Harvard incubations that the OA-less plots were the determining factor for phosphorus presence in soils. However, no OA-less plots is available for comparison here.

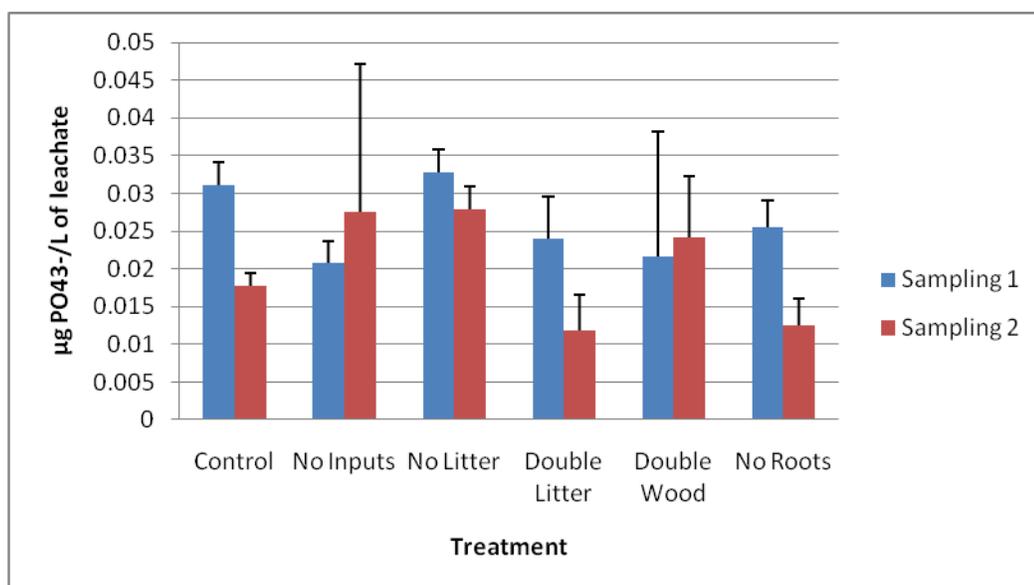


Figure 6.2 Average phosphorus levels of HJA leachate at 100 cm depth. Like Figure 6.1, there are no statistically significant differences in phosphorus levels between the treatments. Each treatment has a low level of phosphorus. OA-less plots are absent, but if there were OA-less plots available, one would expect to see a significant contrast in phosphorus levels to the treatments shown here.

Nitrate

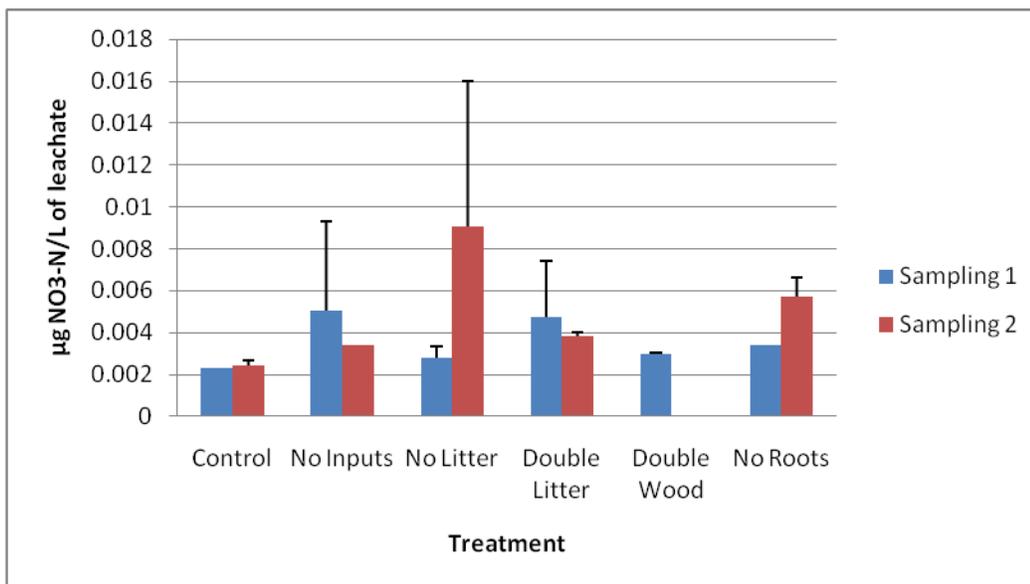


Figure 7.1 Average nitrate levels of HJA leachate at 30 cm depth. For sampling 2, no Double Wood plot produced enough leachate for analysis. There are no statistically significant differences in nitrate levels between the treatments. The extremely low levels of nitrate being analyzed by the Alpkem may have made it difficult for the detector to establish clear peaks.

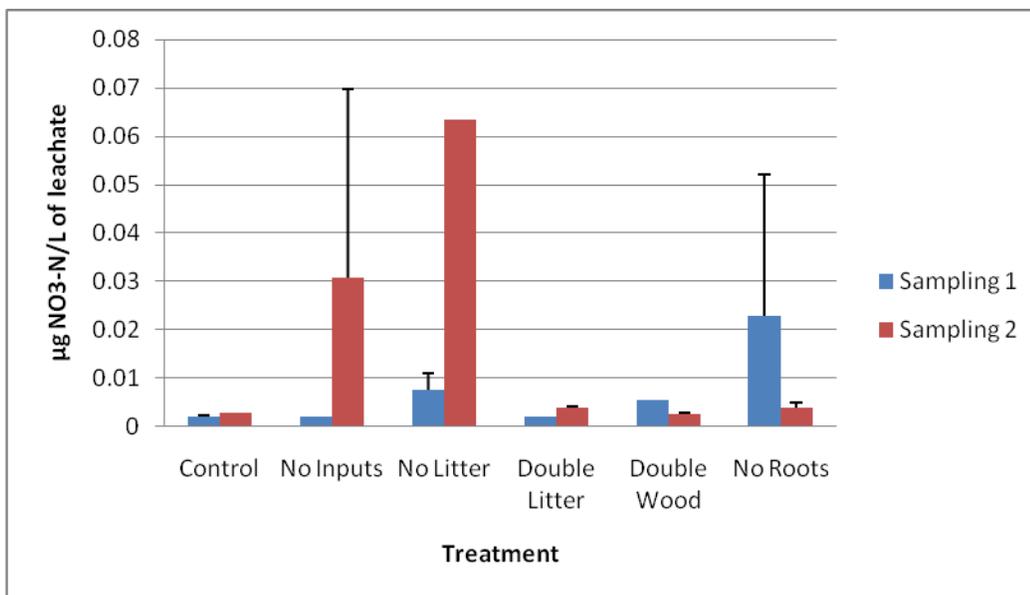


Figure 7.2 Average nitrate levels of HJA leachate at 100 cm depth. There are no statistically significant differences in nitrate levels between the treatments, but the No Inputs, No Litter, and No Roots plots appear higher in nitrate than the other treatments, which is more like the pattern we expect to see in a nitrate analysis. These treatments have more nitrate being leached out due to the lack of organic matter and therefore nitrogen immobilization.

Ammonium

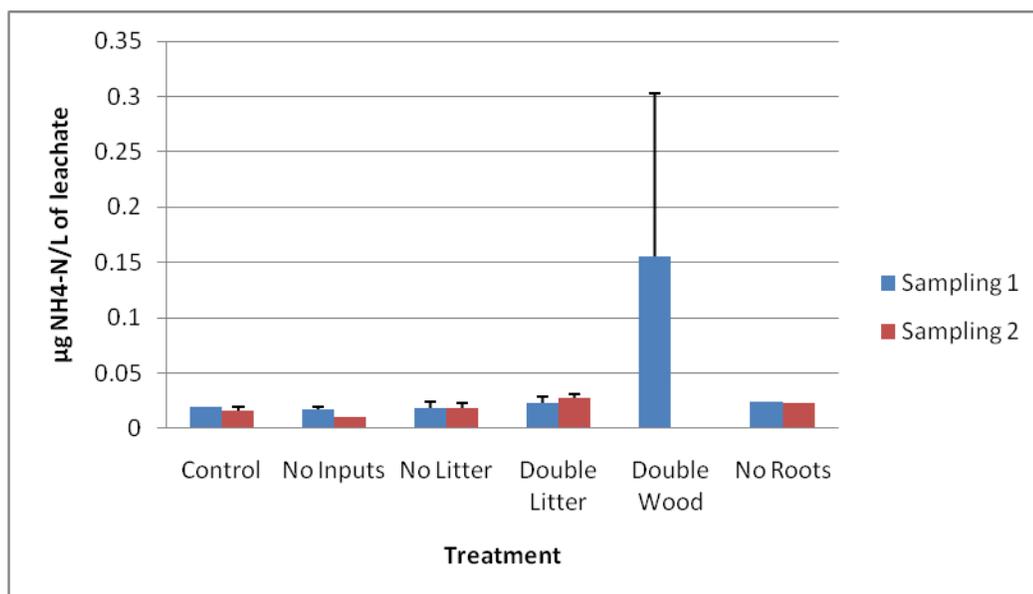


Figure 8.1 Average ammonium levels of HJA leachate at 30 cm depth. For sampling 2, no Double Wood plot produced enough leachate for analysis. There are no statistically significant differences in nitrate levels between the treatments. This is likely due to the cation exchange capacity of the soil, which limits the mobility of ammonium. The Double Wood plots have an extremely high level of nitrate in the first sampling, which may be random, but the absence of a second sampling makes it difficult to be certain.

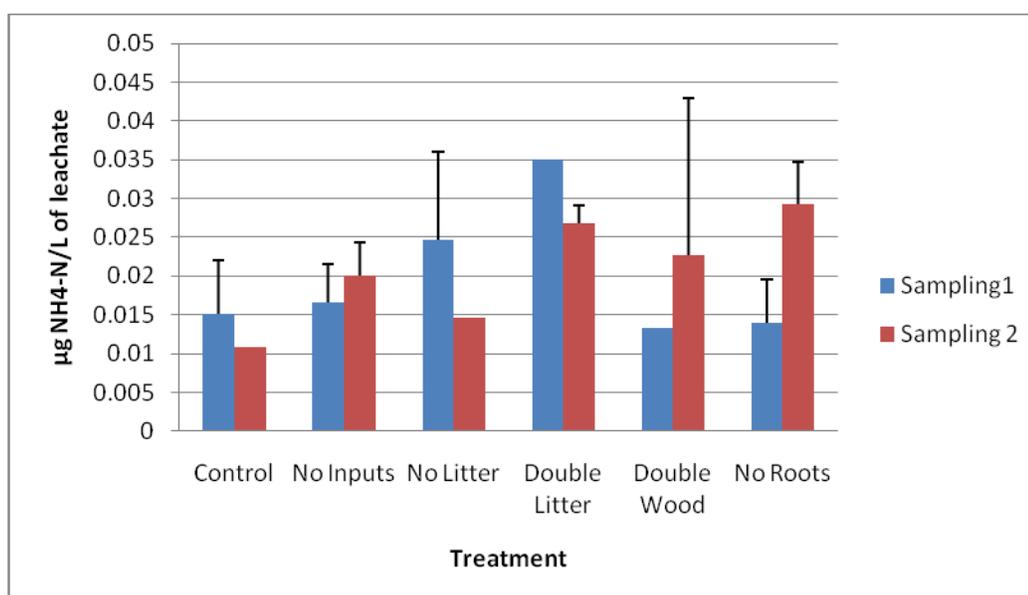


Figure 8.2 Average ammonium levels of HJA leachate at 100 cm depth. There are no statistically significant differences in nitrate levels between the treatments. Again, the cation exchange capacity of the soil is likely limiting the mobility of ammonium. The ammonium levels here are lower than at 30 cm depth, which shows how ammonium levels decrease as we get further away from the litter layer.

DISCUSSION

Harvard Forest incubations

Calcium

Mean calcium levels at the O horizon were particularly lower in the No Roots plots. The first and third leaching events of the No Roots plots are statistically significant. Calcium is a major component of plant cell walls. In root cell walls, calcium is present in the form of calcium pectate, which gives roots structural strength. Figure 1.1 demonstrates the great prevalence of calcium in roots, since the consequence of removing them is a marked decrease in calcium levels. Thus, at the O horizon, the presence or absence of calcium is a good indicator of root material.

Interestingly, this relationship did not translate at lower depths. The first leaching event of the OA-less plots in Figure 1.2 is statistically significant, likely from calcium in mineral soil. However, on the whole, all the treatments show relatively similar calcium levels at 0-10 cm and 10-20 cm depth. Thus, detrital input manipulation had no effect on calcium levels as depth increased. It appears that depth takes preference over roots or other calcium-containing plant material in determining calcium levels. At depths below the O horizon, it is probable that calcium levels are consistent between treatments since roots are not located at lower depths. Roots of deciduous tree stands are concentrated in the O horizon, which means calcium levels are highest in the O horizon where it can be used for neutralization of acidic forest floors. This explains why the No Roots plots were only lower in the O horizon; taking away roots in the 0-10 cm and 10-20 cm depths did not show a statistical difference from the other treatments since there are fewer roots at

those depths anyway. Therefore, detrital manipulation does not have any major effect on calcium levels until it is present at the surface where it is most needed for acid neutralization.

Phosphorus

Phosphorus was not significantly affected at any depth by aboveground or belowground litter manipulations. However, in OA-less plots where O and A horizon soil were removed and replaced with B horizon soil, the levels of phosphorus increased greatly. All OA-less plots produced significantly higher phosphorus levels than the other treatments. In Figure 2.2, all leaching events for the OA-less plots were statistically significant. In Figure 2.3, the second and fourth leaching events for the OA-less plots were statistically significant. This is indicative of the importance of B horizon soil as a source of phosphorus. B horizon soil contains bedrock. Bedrock, when weathered, releases phosphorus for plant use. With B horizon soil at the surface of OA-less plots, the bedrock is directly subjected to rain and other modes of weathering, which releases more available phosphorus into the surroundings. This higher level of phosphorus is clearly evident in all the figures. All the other treatments at every depth contained fairly low levels of phosphorus; the low levels did not range much between treatments either. This is again consistent with the idea that the presence or absence of bedrock is the sole factor affecting phosphorus levels. Without bedrock, no phosphorus is available to plants.

Another reason why phosphorus may be so low in other treatments is if aluminum is being mobilized and is binding phosphorus. In cases where a lot of weathering occurs, aluminum mobilization is high. Thus, the levels of phosphorus detected in leachate will

be extremely low. Only B horizon soil will have lots of phosphorus because the amount released into the soil outweighs the amount lost to either plants or aluminum precipitates. Could the presence of B horizon soil be masking the real problem of aluminum mobilization by making phosphorus levels appear significantly high in those plots?

Nitrate

Nitrate analyses proved that detrital inputs have a significant effect on nitrogen levels. In Figure 3.1, the Control, Double Litter, and OA-less plots all had minuscule amounts of nitrate leaching out. The No Roots plot was notably higher. The third leaching event for the No Roots plots was statistically significant. This is reflective of the idea that greater levels of organic matter allow for greater nitrogen immobilization. Organic matter serves as a carbon source for soil microbes. Microbes need carbon for nitrogen immobilization. Therefore, when organic matter levels are high, nitrogen immobilization is also high. The Control, Double Litter, and OA-less plots all produced low levels of nitrate because a significant amount of carbon was made available to the microbes from the litter. Microbes could then go on to immobilize nitrogen, which ultimately leads to only trace amounts of nitrate being leached out. In addition to microbes, plants also take up nitrogen. By removing roots, we are removing plant uptake of nitrogen. Furthermore, plant uptake of nitrogen is assisted by mycorrhizae, fungi that colonize roots and increase the surface area for absorption. The added assistance of the mycorrhizae makes plants a strong competitor against microbes for nitrogen, but removing roots means that whatever nitrogen microbes do not immobilize becomes leached out instead.

In Figure 3.2, a similar story exists. The Control and Double Litter plots had extremely low levels of nitrate, with the Double Litter plots being statistically significant in the third and fourth leaching events. This is consistent with the fact that higher carbon inputs allow for greater nitrogen immobilization. On the other hand, No Inputs and No Roots plots presented high levels of nitrate in leachate since these treatments lacked the carbon level needed for good nitrogen immobilization.

It appears that detrital manipulations do not affect nitrogen levels at greater depths. At the 10-20 cm depth, no statistically significant differences exist between the treatments (Figure 3.3). However, a similar pattern is evident in all the treatments from the first leaching event to the last; by the last leaching, all the treatments had a high level of nitrate being leached out, which indicates that nitrogen is being mobilized no matter what the detrital manipulation is. In fact, at all depths, there are multiple treatments that showed this pattern in which nitrate showed a small increase at the first leaching, decreased on the second leaching, and then increased substantially for the remaining events. The first little increase in the data is likely residual nitrate from the disturbance event at the time of collection and incubation of the soils. The continual increase in nitrate in the remaining leaching events suggests an increase in activity from nitrifying bacteria. Over time, the number of nitrifying bacteria multiplies, resulting in more oxidation of ammonia into nitrite, and ultimately nitrate.

Ammonium

At the O horizon, the ammonium levels did not vary significantly between treatments. This is likely due to ammonium's limited mobility in soils. Ammonium is being attracted to negatively charged clay particles in the B horizon below. This

interaction is quantified by the cation exchange capacity. The higher the cation exchange capacity of the soil, the more it will hold onto ammonium and other cations. Thus, in Figure 4.1, cation exchange capacity takes precedence over detrital manipulation in affecting ammonium levels, which is why the data appears consistent between treatments.

In Figure 4.2, the No Inputs plots are statistically significant for the third and fourth leaching events. This can not only be attributed to the cation exchange capacity of the soil, but also to the lack of organic matter producing ammonium in these plots.

At the 10-20 cm depth, ammonium was lower in the No Inputs plots and OA-less plots in comparison to the other treatments. Again, the low ammonium levels of the No Inputs plots may be due to the lack of organic matter producing ammonium. In No Inputs plots at higher depths, ammonium-producing organic matter in the A horizons makes up for the removal of detrital inputs. Hence, we do not see a low ammonium level in leachate at the O horizon. In the OA-less plots, B horizon soil comprises much of the soil profile. B horizon is rich in clay. Because OA-less plots have more clay in the surface horizons than the other plots, the chances of ammonium being bound to any clay particle are increased, which is likely why there is less ammonium is being leached out.

As depth increases, the amount of ammonium generally decreases in all plots. This again supports the idea that B horizon soil is located deeper in soils, which allows cation exchange capacity to affect ammonium levels. Also, with more depth, one gets further away from the litter layer where there is ammonium-producing matter.

H.J. Andrews lysimeters

Calcium

Unlike the Harvard incubations, no statistically significant differences in mean calcium levels exist between the treatments at either depth. This may be due to the fact that soils were collected at depths of 30 cm and 100 cm, deeper than where you would find the O and A horizon. If it is true that majority of calcium would be found at surface soils due to root activity, then this may be why calcium levels are consistent from one treatment to the next; calcium could be minute enough at 30 and 100 cm that detrital manipulation has no effect.

Phosphorus

Like calcium, mean phosphorus levels did not vary significantly among treatments at either depth. No treatments are statistically significant. In the Harvard incubations, the OA-less plots produced significantly high levels of phosphorus because of the amount of bedrock in B horizon soil. All the other treatments produced lower levels of phosphorus that were similar from treatment to treatment. The H.J. Andrews results reflect this pattern of low-level phosphorus between treatments. Unfortunately, there is no OA-less plots with lysimeters in them, so no comparison can be made as with the Harvard incubations. If there were OA-less plots that could be analyzed from the H.J. Andrews, we would expect to see the same sort of drastic increase in phosphorus levels as we did with the incubations.

Nitrate

Figure 7.1 does not show the same pattern as in nitrate analyses from the Harvard incubations. Rather, no statistically significant differences are evident between the

treatments. This may be due to the fact that the levels were thousandths of a microgram; the Alpkem Flow Solution IV had more difficulty establishing clear peaks for each sample since only trace amounts of nitrate were present, which may have affected the actual concentration calculation against the linear curve.

However, in Figure 7.2, we see the pattern that we expect in a nitrate analysis. No treatments are statistically significant, but like the Harvard incubations, the No Inputs, No Litter, and No Roots plots appear to be higher in nitrate than the treatments that had greater levels of organic matter. Again, this is due to the fact that removal of organic matter (and therefore a source of carbon) results in the elimination of plant uptake of nitrogen as well as microbial immobilization of nitrogen.

Ammonium

There are no statistically significant differences in average ammonium levels between treatments at either depth. Again, this likely due to the cation exchange capacity of the soil, clay binding ammonium so that it can be taken up by plants rather than leached out.

The Double Wood plots have an extremely high level of ammonium in the first sampling in Figure 8.1. This may be a random result; however, this cannot be for certain since there is no second sampling to compare it to.

CONCLUSION

For the Harvard incubations, only nitrate showed a significant difference between treatments. It coincided with the relationship known between organic matter, immobilization, and plant uptake: more organic matter leads to more nitrogen immobilization and plant uptake and lower nitrate levels in leachate. Calcium, phosphorus, and ammonium were not significantly affected by the detrital treatments. Rather, they were primarily affected by depth and soil profile, such as the importance of B horizon soil in detecting phosphorus.

Some of the same patterns in the Harvard incubations showed up in the H.J. Andrews lysimeter samples, which was that a lot of the elements were not affected by detrital manipulation. No treatments at any depth for any element were statistically significant. Surprisingly, even nitrate did not reflect its expected relationship with organic matter as strongly. At 30 cm, nitrate seemed not to be affected by the treatments at all, but at 100 cm, the results looked more similar to the incubation data. Here, it is important to note that working with extremely minute amounts of an element can be difficult for instruments to measure accurately.

Calcium and phosphorus, the two elements at the center of my research, was, on the whole, not affected by litter manipulations. However, they were affected by changes in the soil profile and depth, suggesting that they are good indicators of cell walls in plant material, bedrock, and other minerals of the B horizon.

In the future, it would be beneficial to install lysimeters in the H.J. Andrews OA-Less plots. It would have been interesting to see if the OA-less plots in the H.J. Andrews

would have reproduced the high phosphorus results that we saw in the Harvard incubations.

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