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UNIVERSITY OF CALIFORNIA
RIVERSIDE

Biochar's Effect on Plant Growth and Soil Nutrient Loss

A Dissertation submitted in partial satisfaction
of the requirements for the degree of

Doctor of Philosophy

in

Plant Biology

by

Elizabeth Floy Crutchfield

August 2016

Dissertation Committee:

Dr. Milton McGiffen, Chairperson

Dr. Don Merhaut

Dr. Giles Waines

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The Dissertation of Elizabeth Floy Crutchfield is approved:

Committee Chairperson

University of California, Riverside

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DEDICATION

I'd like to dedicate this dissertation and all the experiences that went with it to my late grandfather, William Yale Crutchfield I.

ABSTRACT OF THE DISSERTATION

Biochar's Effect on Plant Growth and Nutrient Loss

by

Elizabeth Floy Crutchfield

Doctor of Philosophy, Graduate Program in Plant Biology
University of California, Riverside, August 2016
Dr. Milton McGiffen, Chairperson

Recent years have shown an increased interest in biochar, a high carbon compound made from pyrolyzed biomass. Biochar is a carbon negative product that has been suggested as a soil amendment. Studies have shown disagreement on the effect of biochar on plant growth and on anion leaching from biochar amended soils. Three experiments were conducted to investigate biochar's effect on plant growth and on nitrogen and phosphorus leaching. Chapter one focuses on biochar's effect on root growth. Bread wheat (*Triticum aestivum* L.) cv. Pavon 76 and Pavon 1RS.1AL were grown in a sandy loam soil amended with sand (50% by weight) and/or biochar (~1.5% by weight). Results indicated that the 1RS.1AL plants had more roots and deeper roots and biochar addition resulted in more root growth, likely due to changes in soil texture. A second study was conducted to investigate nitrogen and phosphorus leaching from Begonias (*Begonia semperflorens* 'Viva') grown in nursery conditions. Different amounts of biochar, ranging from 0% to 30% by weight, were incorporated into potting mix and the amount of nitrate, ammonium and phosphate leached from each pot was measured. No difference in plant growth was detected,

but high rates of biochar did reduce the amount of nitrate, ammonium and phosphate leached. The last chapter investigated the biochar's ability to adsorb nitrate, ammonium and phosphate when added to tall fescue (*Festuca arundinacea*) turf. Plots were either direct seeded or transplanted as sod with high, low or no biochar applied to the plots. Although plots with transplanted plants clearly leached more nitrate, ammonium and phosphate, the effect of biochar on leaching was less substantial. However, the results do seem to indicate that the biochar does reduce some leaching. Overall, the affect biochar had minimal effect plant growth in these experiments. The begonias showed no change in shoot growth. However, wheat plants tended to have more roots when grown with biochar. Additionally, when the results of the leaching studies for both the begonia experiment and the turf experiment are taken in aggregate, it's clear that biochar can reduce the amount of nitrate, ammonium and phosphate leached.

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BIOCHAR'S EFFECT ON PLANT GROWTH AND NUTRIENT LOSS

Introduction to Biochar

Biochar is pyrolyzed biomass, much like charcoal. Pyrolysis is the thermochemical decomposition of biomass at temperatures less than 700° C in the presence of little to no oxygen (Lehmann and Joseph, 2009). When organic biomass is pyrolyzed, most of the hydrogen, oxygen, nitrogen, and some of the carbon is volatilized (Antal and Grønli, 2003). The remaining solid fraction, the biochar, is comprised of the carbon and the ash (Keiluweit et al. 2010). At pyrolysis temperatures greater than 350° C the carbon begins to turn into carbonaceous rings, and at 600° C graphene sheets form (Amonette and Joseph, 2009; Lehmann et al., 2009). These carbon ring structures are very resistant to degradation (Schmidt and Noack, 2000).

Because biochar is very resistant to degradation, one of the key benefits of using biochar is that it is carbon negative (Lehmann, 2007). Biochar half-life in soil can range from tens to hundred thousands of years (Spokas, 2010b). A life cycle analysis of biochar found that biochar made from crop residue or yard waste reduced greenhouse gas emissions by about 800 kg of CO₂ per ton of dry feedstock (Roberts et al., 2010). Biochar can be made from any kind of organic matter;

therefore it can be made from waste products, such as sewage, manure, and yard waste. This simple and ancient technology, pyrolysis, can turn waste products into a useful product, biochar, which not only reduces atmospheric CO₂ levels but has an agricultural benefit as well.

Biochar and Plant Growth

Many studies have found increased growth of plants in biochar amended soils (Chan et al. 2008a; Chan et al. 2008b; Steiner et al. 2008; Van Zwieten et al., 2010). Biochar improves plant growth by altering soil quality. Few studies have investigated biochar's direct effect on roots, but Bruun et al. (2014) found that the addition of biochar improved root growth, which was correlated to increased barley grain yield. However, Many studies have found no increases or even decreases in plant growth (Spokas et al., 2012). Some kinds of biochar, depending on how they are made, may contain toxic compounds, which may dissolve into the soil water (Kim et al., 2003). However, the majority of studies reviewed by Spokas et al. (2012) found that biochar increased plant growth.

An important way that biochar is able to affect plant growth is by improving fertilizer holding capacity of the soil. Taghisadeh-Toosi et al. (2012) used N¹⁵ to verify that the ammonia sorbed to the surface of the biochar is still bioavailable. Many studies (Chan et al. 2008a; Chan et al. 2008b; Steiner et al., 2007; Van Zwieten et al., 2010) have found that the application of biochar, with fertilizer, increases nutrient uptake in plants.

Biochar and Adsorption

Nitrogen

Understanding nitrogen (N) cycling in the soil is particularly tricky because N can easily transform from nitrate (NO_3^-) to nitrite (NO_2^-) to ammonium (NH_4^+) or be released into the air as ammonia, nitrous oxide or N gas (Marschner and Rengel, 2007). In order to understand how NO_3^- , a highly mobile and potentially detrimental, ion behaves in the soil, we have to monitor NH_4^+ as well, because the NH_4^+ to NO_3^- conversion is a common transformation found in soils.

NH_4^+ , as a cation, can be sorbed to the surface of biochars, by way of negative functional groups such as hydroxyls, amines, ethers, esters, and carboxyls (Amonette & Joseph, 2009). This is the mechanism for cation exchange capacity (CEC). Because the bonds formed between the ion and the functional groups on the biochar are relatively weak electrostatic and nonspecific interactions (Essington, 2004), ions can become attached and detached and replaced by other ions. Many studies have shown that biochar can adsorb NH_4^+ (Angst et al., 2013; Asada et al., 2002; Ding et al., 2010; Hale et al., 2013; Lehmann et al., 2003; Steiner et al., 2010). Some studies have shown that biochars pyrolyzed at lower temperatures adsorb NH_4^+ better, because those biochars have a higher CEC (Asada et al., 2002; Hollister et al., 2013). As temperatures increase less acidic functional groups, specifically carboxylic groups, are formed on the surface of the biochar (Cheng et al., 2006). However, aging can make the surface chemistry of biochar more acidic (Cheng et al., 2006).

NO_3^- leaching is a bigger problem than NH_4^+ leaching. Soils and potting media often have components that have a high CEC, but most materials don't have a high anion exchange capacity (AEC). If biochar has higher AEC than soil or potting medium, than it could be a useful tool in plant production. Some studies have found biochar had no effect on NO_3^- (Eykelbosh et al., 2015; Hale et al., 2013; Hollister et al., 2013), while others did find increased sorption (Chintala et al., 2013). Chintala et al. (2013) found that NO_3^- sorption was influenced by pH and the presence of other anions such as phosphate (PO_4^{3-}) and sulfate (SO_4^{2-}). Increases in pH were correlated with decreased NO_3^- sorption (Chintala et al., 2013), because at high pH, the functional groups on the surface of the biochar would become negative and repel nitrate. Because NO_3^- is only monovalent, multivalent anions can outcompete it. Chintala et al., (2013) suggested that sorption of NO_3^- to the surface of biochar may be due to electrostatic interactions and ionic exchange mechanism. Biochar that was better at NO_3^- sorption also had a higher point of zero net charge, high volatile organic carbon, and high base cation concentration, all of which indicates a greater positive charge of the surface functional groups. It is possible that divalent cations could provide a bridge from the negative functional group to anions such as NO_3^- (Chintala et al., 2013). Chintala et al. (2013) used biochar pyrolyzed at a higher temperature (650°C), than the studies that found no NO_3^- adsorption (575°C - Eykelbosh et al., 2015; 300-350°C - Hale et al., 2013; 350 & 550°C - Hollister et al., 2013). Kameyama et al. (2011) found that biochar pyrolyzed at 400-600°C did not adsorb NO_3^- but biochar pyrolyzed at 700+°C did.

Phosphorous

There are many forms of P in the soil: organic P, P precipitates, and ionic P (Russell, 1973). Unfortunately, only the ionic P is plant available and it is the smallest part of P in soil (Russell, 1973). Because soils typically have very little AEC (Singer and Munns, 2006), application of inorganic P fertilizer can result in considerable runoff. Biochar can also have high levels of P contained as ash; feedstocks with a relatively high P, such as animal waste biochar, will result in biochar with a high P. Unlike N, the portion of P in the biochar is much more predictable because ash isn't lost in pyrolysis. Such biochar could effectively be used as a P fertilizer. As described above, some biochar do possess comparatively high AEC. Some studies have found that some biochar is capable of reducing P leaching (Angst et al., 2013; Hale et al., 2013; Hollister et al 2013). Since biochar can change the pH of a soil (Van Zwieten et al., 2010), it can be used to make the pH of the soil more suitable for P bioavailability. Because both NO_3^- and PO_4^{3-} are anions, many of the same mechanisms apply. Biochar has greater AEC if it was pyrolyzed at a high temperature; so high temperature biochar adsorbs PO_4^{3-} better (Chen et al., 2011). Also like NO_3^- , PO_4^{3-} can become bound to the surface of the biochar through electrostatic bridges made of divalent cations (Hale et al., 2013). It is hypothesized that the main mode of sorption for PO_4^{3-} is periclase (MgO) particles on the surface of the char (Yao et al., 2011).

This research was intended to investigate the impact that biochar has on plant growth and nutrient leaching. The first chapter investigates how biochar

interacts with the soil into which it is incorporated and the subsequent impact of the growth on two wheat (*Triticum aestivum* L.) isogenic lines with differing rooting strategies. In chapter two, biochar was applied to a typical potting mix at different rates to investigate the impact it has on plant growth and nutrient leaching in a greenhouse setting. In the last chapter, biochar was applied at differing rates to a turfgrass lawn in order to assess the nutrient leaching that would occur in a field situation.

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Chapter 1

Affect of a soft wood biochar on wheat growth and root architecture

Abstract

With the increase in human populations and climatic temperatures, new ways to increase grain yield in drought affected areas are increasingly important. A study was conducted over three years to determine the effect of biochar on root growth, shoot growth, and yield of wheat in different soil mixtures using plants with different rooting strategies. Bread wheat (*Triticum aestivum* L.) cv. Pavon 76 and Pavon 1RS.1AL were grown in a sandy loam soil amended with sand (50% by weight) and/or biochar (~1.5% by weight). These plants were grown in a randomized block design at a greenhouse at UC Riverside for 5-6 months. Plants were harvested twice, either at flowering or seed maturity (65 and 92 on the Zadok's development scale). Harvested plants were measured for root mass, shoot mass and grain yield. The results show the 1RS.1AL plants tended to have more roots biomass and more deep roots, granting it greater access to water, but that this did not translate into increases in grain yield. The addition of sand changed the water-holding of the soil and therefore root and plant growth. Biochar also changed water holding to a lesser degree, but may have allowed for more root growth by reducing bulk density.

Introduction

Biochar is made through pyrolysis, the thermochemical decomposition of biomass in the presence of little to no oxygen, at temperatures less than 700 °C (Lehmann and Joseph, 2009). During the pyrolysis process much of the carbon is lost, but what is left becomes a very recalcitrant carbon ring structure (Keiluweit et al., 2010) with a half-life of upwards of 1000 years (Spokas, 2010). This makes the product carbon negative (Lehmann, 2007). Roberts et al. (2009) estimated in a life cycle analysis that plant waste biochars can reduce greenhouse gas emissions by about 800 kg of carbon dioxide each year per tonne of dry feedstock. As a result, there is a lot of interest in using biochar to fight global warming. Fortunately, it has also been found that biochar can provide a number of benefits in an agricultural setting (Lehmann and Joseph, 2009). Those benefits include high soil porosity, low bulk density (Topoliantz & Ponge, 2005), increased soil water-holding capacity (Karhu et al., 2011; Basso et al., 2013, Tryon, 1948) and increased soil nutrient retention (Glaser et al., 2002; Lehmann et al., 2003; Brockhoff et al., 2010).

Improved root system growth could allow for increased yields with minimal cost to the grower or the environment (Gewin 2010). Pavon 76 is a wheat cultivar developed during the Green Revolution. A translocation of the short arm of rye (*Secale cereal* L.) chromosome 1 (1RS) into the genome of Pavon 76 has been shown to increase root biomass (Ehdaie et al., 2012). Some studies have shown that this translocation also leads to an improvement in grain yield (Edhaie et al., 2003; Edhaie et al., 2012) or at least no loss of yield (Kaggwa et al. 2015). Waines (2012)

linked increased root biomass with yield for some wheat cultivars. Improving root growth could be a key to maintain high yields in increasingly water-limited environments.

Root growth is dependent on water-holding capacity (WHC) and bulk density of soil. Grasses, like wheat, tend to grow shallow, adventitious roots in wet environments, and when soil is dry, seminal roots penetrate more deeply to draw water up from deeper zones (Loomis and Connor, 1992). High WHC can lead to a waterlogged environment, where plants tend to have less roots. Biochar additions can reduce WHC of soil from 9% to 15% depending on the biochar and the soil (Karhu et al., 2011; Dugan et al., 2010; Basso et al., 2013). Malik et al., (2001) found reductions in length and number of seminal and adventitious roots in wheat plants grown in waterlogged conditions. High soil bulk density can also reduce root growth (Jones, 1983).

By improving root growth, biochar could sequester carbon indirectly. Biochar was shown to prevent the decomposition of associated labile carbon (Cross et al., 2011). Carbon inputs from roots were retained in soils better than carbon from leaf fall (Schmidt et al., 2011). Deeper root growth increased carbon sequestration because deeper soil profiles are more likely to have slower decomposition turnover rates (Lorenz and Lal, 2005). Soils currently provide a sink for 21 to 52 gigatons of carbon, which is 50% to 66% of historic rates (Lal, 2004).

This study investigates how biochar impacts plant growth and yield in very sandy soils and silty soils with a deeper rooting iso-translocation line versus a

classic parental wheat cultivar. To this end, wheat was grown in cooled greenhouse conditions in tall PVC tubes to allow for more natural root growth.

Materials and Methods

Experimental Setup

This experiment was conducted in a greenhouse at UC Riverside. Wheat was grown in media filled tubes as described in Ehdaie et al. (2010). Seeds were germinated in Petri dishes and seedlings were transplanted after approximately five days. A plastic sleeve was filled with 8.5 kg of the appropriate soil media type and placed within a 0.75 m tall, 0.10 m diameter polyvinyl chloride (PVC) pipe. The bottom of each plastic sleeve had two holes for drainage and a filter paper to prevent soil loss. Prior to planting, 2 L of deionized water were added to each tube.

The experiment was repeated three times during the winter and spring of 2012, 2013 and 2014, using a randomized complete block design with 4 replicates. The experiment utilized two accessions of wheat (*Triticum aestivum* L.), a semidwarf cv. Pavon 76, (a spring bread wheat from the breeding program of Centro Internacional de Mejoramiento de Maiz y Trigo, Mexico) and a its translocation line Pavon 1RS.1AL. In Pavon 1RS.1AL, the short arm of chromosome 1 A of Pavon 76 was replaced with the short arm of chromosome 1 (1RS) of rye (*Secale cereale* L.). The original rye translocation was from a Petkus rye selected in Germany and included in cv. Kavkas bread wheat. The resulting plants tend to have longer roots and less plastic root system (Ehdaie et al., 2003).

The biochar used in this experiment was produced by pyrolyzing white pine at 400 °C, resulting in a final product with a dry weight ash content of 14.4% and a carbon content of 85.5%. The soil used in this experiment was an Arlington Sandy Loam, a mildly alkaline, fine textured sandy loam, harvested from the Citrus Research Station at UC Riverside. The sand used was washed Grade 30 silica sand.

In the first year, there were eight different treatments, a full factorial design with two wheat lines: Pavon 76 (P76), and the translocation line Pavon 1RS.1AL (P1RS); two base soil types: the soil only (SO), and 50% soil : 50% sand by weight (SS); and two biochar amounts: no biochar (B0), and the equivalent of 20 tons/hectare biochar incorporated to a depth of 15 cm (B15). For example, treatment P76-SS-BC15 has a mixture of sand and soil with biochar incorporated to a depth of 15 cm and a Pavon 76 wheat plant growing in it. Additionally, in 2013 and 2014, another biochar treatment was added: biochar incorporated fully throughout the soil column at ~1.5% by weight, equivalent to the BC15 treatment (called B75). Therefore in 2013 and 2014, there were twelve treatments a full factorial of two wheat lines x two soil types x three biochar amounts.

Crop Management

After seedlings were planted, they were fertigated with half-strength Hoagland's solution. Irrigation was discontinued at maturity, when the main tiller was devoid of green color. In 2012, plants were watered based on a visual approximation of when the soil looked dry and the plants were slightly water

stressed. The SS treatments dried out much more quickly therefore the two soil types were watered differently, but all SS treatments were watered the same and all SO treatments were watered the same. In 2013, all plants were watered the same. As a result, the substantial difference between the WHC of the SS and SO treatments meant that many of the plants grown with sand were water stressed while the plants without sand were waterlogged. The irrigation regime for 2014 will be described below under “Soil Moisture Probes.”

In 2012, after 9 weeks, when the plants were flowering, the shoots were harvested and oven dried to obtain shoot biomass. The soil from the plastic sleeve was gently washed off the roots and the roots were collected and dried to obtain the root biomass. Roots were subdivided into shallow roots (shorter than 30 cm) and long roots (longer than 30 cm). In 2013, half the replicates with harvested at 13 weeks and the other half at 20 weeks. In 2014, the harvests were at 13 and 17 weeks. The second harvest allowed for collection of yield measurements: grain mass, and number of grains, in addition to the root and shoot data.

Soil Moisture Probes

Soil moisture probes (Watermark from Irrrometer) were placed in the soil column at three different levels: near the bottom, at the middle, and at the top. These soil moisture probes were used to determine how much water each treatment would receive, in order to maintain some drought pressure; treatments

were only watered when the soil moisture dropped below 40 centibars. Table 1.1 shows the total amount of water added by the first harvest and the second harvest.

	Soil	Soil + 15 cm biochar	Soil + all biochar	Soil + sand	Soil + sand + 15 cm biochar	Soil + sand + all biochar
First Harvest	2950 mL	3050 mL	5200 mL	7000 mL	6600 mL	4300 mL
Second Harvest	6750 mL	5750 mL	9100 mL	12500 mL	12600 mL	8950 mL

Table 1.1: This table shows the total amount of water applied over the experiment to each potting mix type from 2014.

Statically analysis

The total root biomass was calculated as the addition of the shallow and deep roots, and likewise the total plant biomass was calculated as the addition of root and shoot biomass. The deep to shallow root ratio was the quotient of the deep roots biomass divided by the shallow root biomass, and used as an indicator of allocation of biomass to either water acquisition or nutrient acquisition. Root to shoot ratio was calculated as the quotient of the root biomass divided by the shoot biomass and was used as an indicator of the allocation of resources to light acquisition or water/nutrient acquisition.

Analysis of variance was performed using SAS on the data after transformation to improve normality. In the data from 2012, shoot weight was analyzed on the inverse scale; deep root weight was analyzed on the natural log scale; and all other dependent variables was analyzed on the original scale. While in

the early harvest of 2013, deep root weight and the deep : shallow root ratio were analyzed on the natural log scale, all other dependent variables was analyzed on the original scale. In the late harvest of 2013, shallow root weight, total root weight, and the root : shoot ratio were analyzed on the natural log scale; the shallow to deep root ratio was analyzed on the inverse scale; all other dependent variables was analyzed on the original scale. In the first harvest of 2014, the shallow root weight was transformed on a log scale; while everything else was left at it's original scale. In the second harvest of 2014, the shoot weight, shallow root weight, total root weight, the deep : shallow root ratio, the root : shoot ratio, and the seed yield (g) were transformed on the log scale; while everything else was left at it's original scale. A P-value of 0.05 or less was considered significant.

Results and Discussion

2012

Soil Type

The soil type had a significant impact on plant growth in 2012. There was more shallow root biomass in the wheat grown in SO than the SS mixture, increasing from 0.642 g to 0.814 g (Figure 1.1a). However, deep root mass was not significantly different and, as a result, total root biomass for SO and SS was equivalent (Figures 1.1b and d); and in turn, the effect of media on the ratio of deep to shallow roots was not significant (Figure 1.1c). There was, however, a significant impact on shoot biomass, the SO treatments contained almost double the above

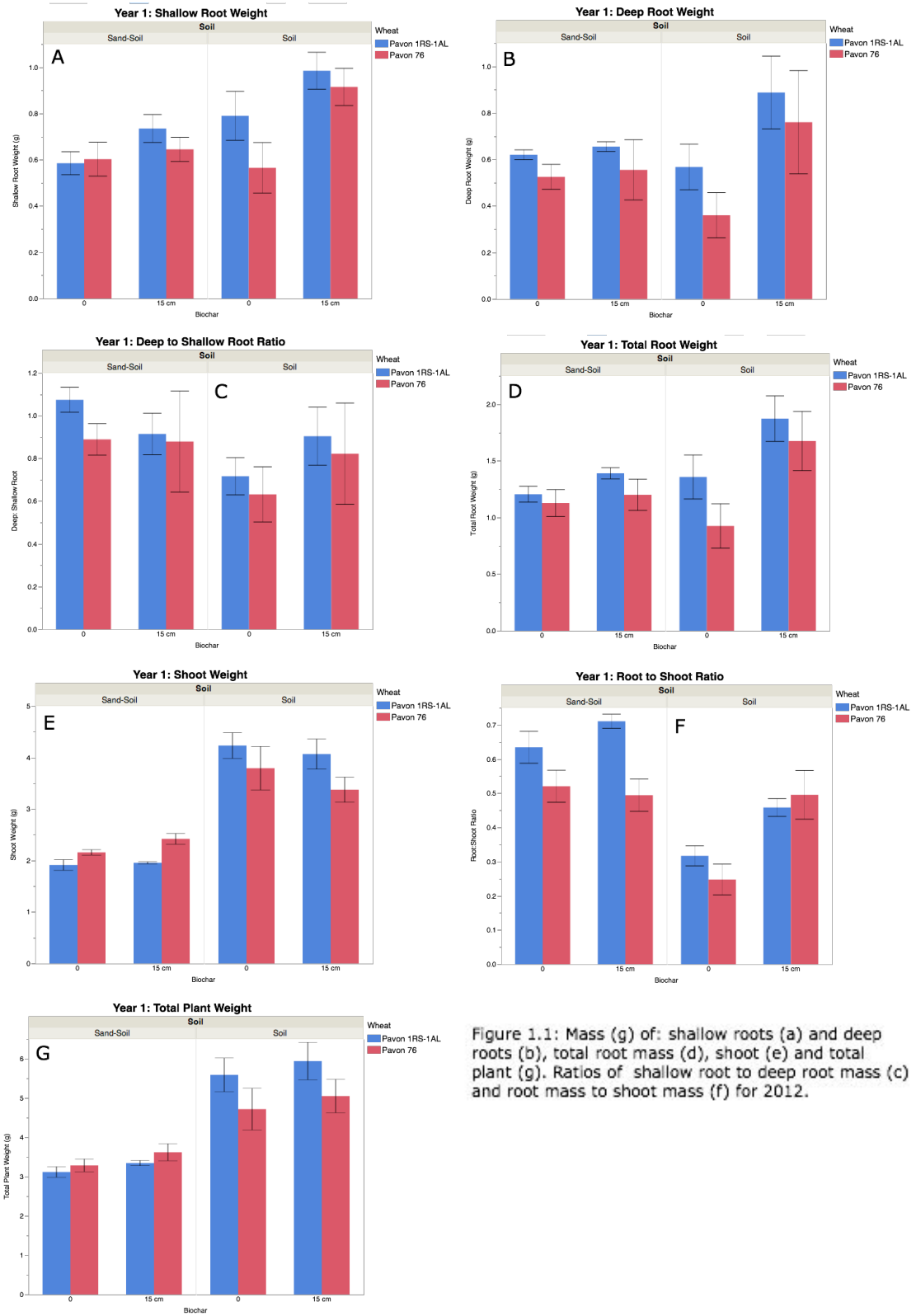


Figure 1.1: Mass (g) of: shallow roots (a) and deep roots (b), total root mass (d), shoot (e) and total plant (g). Ratios of shallow root to deep root mass (c) and root mass to shoot mass (f) for 2012.

ground biomass of the SS treatments (Figure 1.1e). This resulted in an overall increase in biomass across the whole plant, the plants grown in the SS mixture were only 60% the size of the SO treatments (Figure 1.1g), resulting in a greater root-to-shoot ratio for the SS treatments (Figure 1.1f). The greater WHC of SO compared to SS, gave plants grown in SO more water and therefore greater growth. The plants grown with SS had less plant available water and therefore allocated more resources to their deep roots.

Genotype

As expected based on past research, P1RS had more and deeper roots. P1RS plants had about 25% more deep root biomass than the P76, although the shallow root mass was not significantly different (Figure 1.1a and b). However, there was not a significant change in total root biomass (Figure 1.1d) nor in the allocation of resources to shallow or deep roots (Figure 1.1c). In SO, P1RS had more shoot growth, 4.15 g compared to P76's 3.58 g, but less if they had been grown in SS, 1.93 g compared to 2.29 g (Figure 1.1e). Therefore, there was greater total plant biomass for P1RS-SO plants (5.77 g) compared to the P76-SO plants (4.88 g; Figure 1.1g). The P1RS variety had a higher root to shoot ratio compared to P76 if grown in SS (Figure 1.1f). P1RS plants tended to have more shoot growth if they had been grown in SO, perhaps because P1RS plants grew more deep roots, but greater root mass is more advantageous when there was more available water.

Biochar

In 2012, B15 had more shallow roots, 0.82 g compared to B0's 0.636 g, regardless of the variety or soil type (Figure 1.1a). And B15 had roughly double the amount of deep roots if the plants were grown in SO (Figure 1.1b), but no significant effect if grown in SS. This led to a 55% increase in overall root growth for SO-B15 (Figure 1.1d). There was no effect of biochar on the relationship of deep roots to shallow roots (Figure 1.1c), nor on the overall shoot growth (Figure 1.1e). Therefore there was no effect on the total biomass of the plant, because the majority of the biomass was in the shoots. But due to the increased root growth in the SO-B15 treatments, the data shows an increased root to shoot ratio for those treatments. For 2012 overall, the wheat plants were able to grow more roots if the soil was amended with biochar, either due to increased water holding or reduced bulk density that allowed better root penetration.

Summary

The SO treatments had greater bulk density but also more plant available water, which led to greater growth. Reduced access to water in the SS treatments meant the plants allocated more resources to root growth. P1RS plants tended to produce more deep roots regardless of what medium they were grown in, which was of greater benefit when grown in SO, a relatively water rich environment. Biochar improved root growth in SO treatments without sand, presumably because it reduced bulk density, allowing for increased root penetration, but this did not have an effect on shoot growth.

2013: First Harvest

In 2013, all treatments were watered equally. Because of the great disparity in WHC between SO and SS treatments, the SS treatments were under drought conditions while the SO treatments were overwatered. This had a profound effect on the results, as a result they are very different from the 2012 results.

Soil Type

While there was no effect on the shallow root growth (Figure 1.2a), plants grown in SS had 30% more deep root growth than SO (Figure 1.2b). P1RS-B75 and P76-B0 showed more root growth in the SS treatments than the SO treatments (Figure 1.2d). This was likely due to waterlogged conditions at the bottom of some of the SO columns, which may explain the greater deep root to shallow root ratio in SS compared to the SO treatments (Figure 1.2 c). In P1RS-SS-B75, the waterlogging appears to have negatively impacted shoot growth. It reduced shoot biomass from the average of 33.6 g to 27.3 g (Figure 1.2e) and total plant biomass was reduced from the average of 36.6 g to just 28.9 g (Figure 1.2g). The treatments grown in SS had a higher root to shoot ratio, either because of the waterlogging in the SO treatments or because drought stress in the SS mixtures lead to more allocation to root biomass (Figure 1.2f). Better drainage in the SS mixture treatments led to more overall growth, especially of the deep roots.

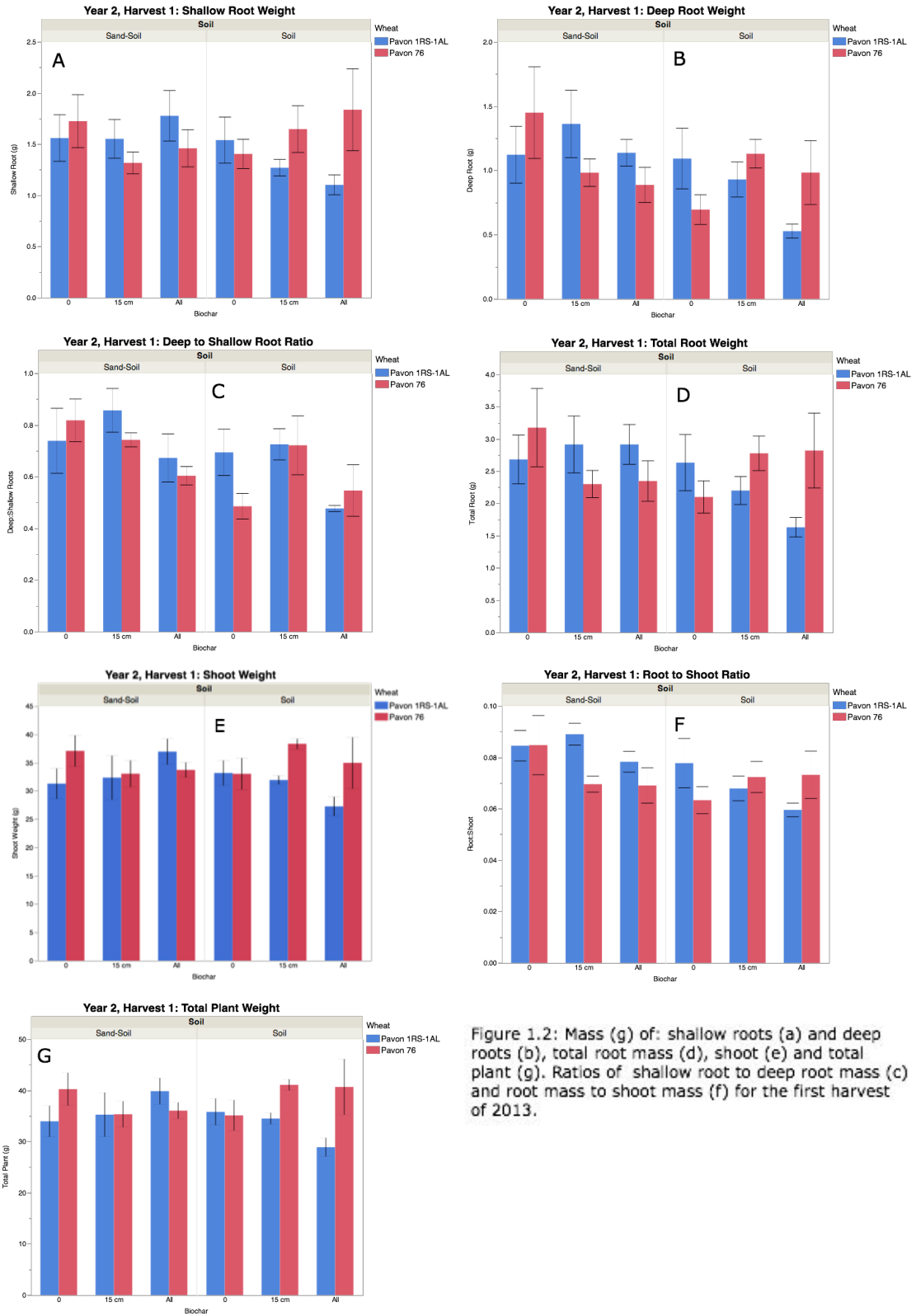


Figure 1.2: Mass (g) of: shallow roots (a) and deep roots (b), total root mass (d), shoot (e) and total plant (g). Ratios of shallow root to deep root mass (c) and root mass to shoot mass (f) for the first harvest of 2013.

Genotype

The genotype of wheat did not affect the shallow or deep root growth in the 2013 experiment (Figures 1.2a and b). But the P1RS-SS plants had particularly low overall root growth (Figure 1.2d). P76-SO-B15 and P76-SO-B75 had greater shoot biomass than the coresponding P1RS plants, by 20% and 28%, respectively (Figure 1.2e). This resulted in greater overall biomass for the P76-SO-B15 and P76-SO-B75 plants compared to the P1RS versions, 40.7 g compared to 28.9 g (Figure 1.2g).

Biochar

While there was no effect on the shallow roots (Figure 1.2a), the P1RS-SO-B0 plants had about double the deep root biomass compared to P1RS-SO-B75 (Figure 1.2b). But total root growth was unaffected (Figure 1.2d). There was proportionally more biomass allocated to deep roots in B15 treatments compared to B75 (Figure 1.2c). This seems to be due to waterlogging killing the deep roots and skewing the results. There was no effect of the biochar on above ground biomass, total biomass of the plant, or the root to shoot ratio (Figures 4.1e – g).

Summary

In 2013, the SO treatments held more water than the SS treatments, which may have led to waterlogged conditions that could not support deep roots. Perhaps because of the watering scheme and root death, the effect of the genotype and biochar was not very apparent.

2013: Second Harvest

Soil Type

In the second harvest of 2013, SS treatments tended to have about 40% more deep roots compared to their SO counterparts (Figure 1.3b). But there was no effect on the shallow roots or the total roots (Figures 1.3a and d). As a result, the plants grown in SS had allocated about 40% more biomass to deep roots compared to shallow roots (Figure 1.3c). There was no effect of the soil type on the amount of shoot tissue, overall plant growth, the ratio of roots to shoots or grain yield (Figures 1.3 e - j).

Genotype

There was no effect of wheat genotype on the deep root growth or the overall root biomass produced (Figures 1.3 b and d), but there was significantly more shallow root biomass produced by the P76-B0 wheat plants grown compared to the P1RS-B0 plants (Figure 1.3a). Regardless, there was no significant effect on the ratio of shallow and deep roots (Figure 1.3c). There was no significant effect of wheat genotype on the aboveground biomass, the overall biomass or the root to shoot ratio (Figure 1.3e – g). However, P76 plants produced more grain yield compared to P1RS, 24.6 g and 22.1 g per plant on average, respectively (Figure 1.4a). But this did not affect the overall number of seeds or the average weight of individual seeds (Figures 1.4b and c).

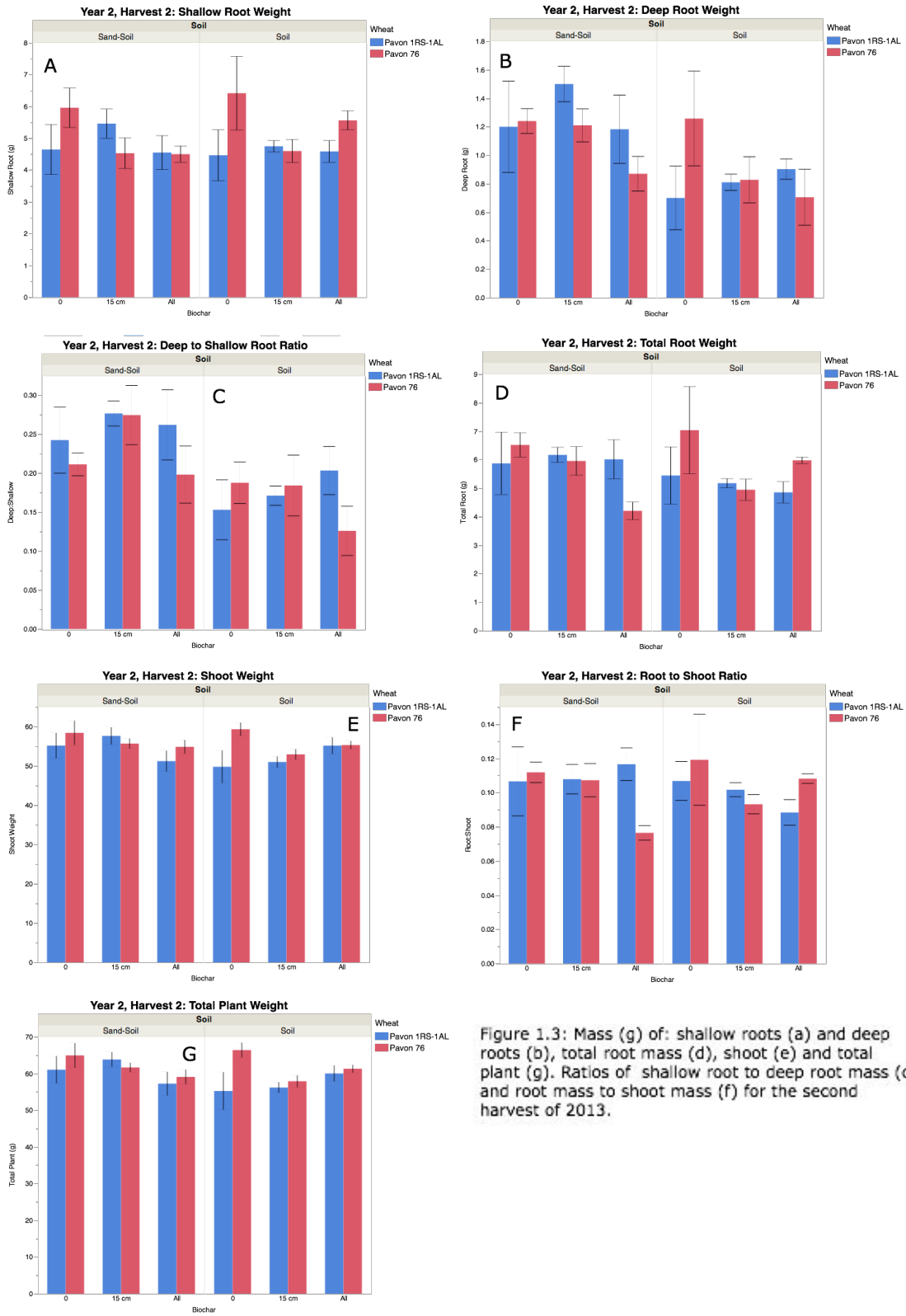


Figure 1.3: Mass (g) of: shallow roots (a) and deep roots (b), total root mass (d), shoot (e) and total plant (g). Ratios of shallow root to deep root mass (c) and root mass to shoot mass (f) for the second harvest of 2013.

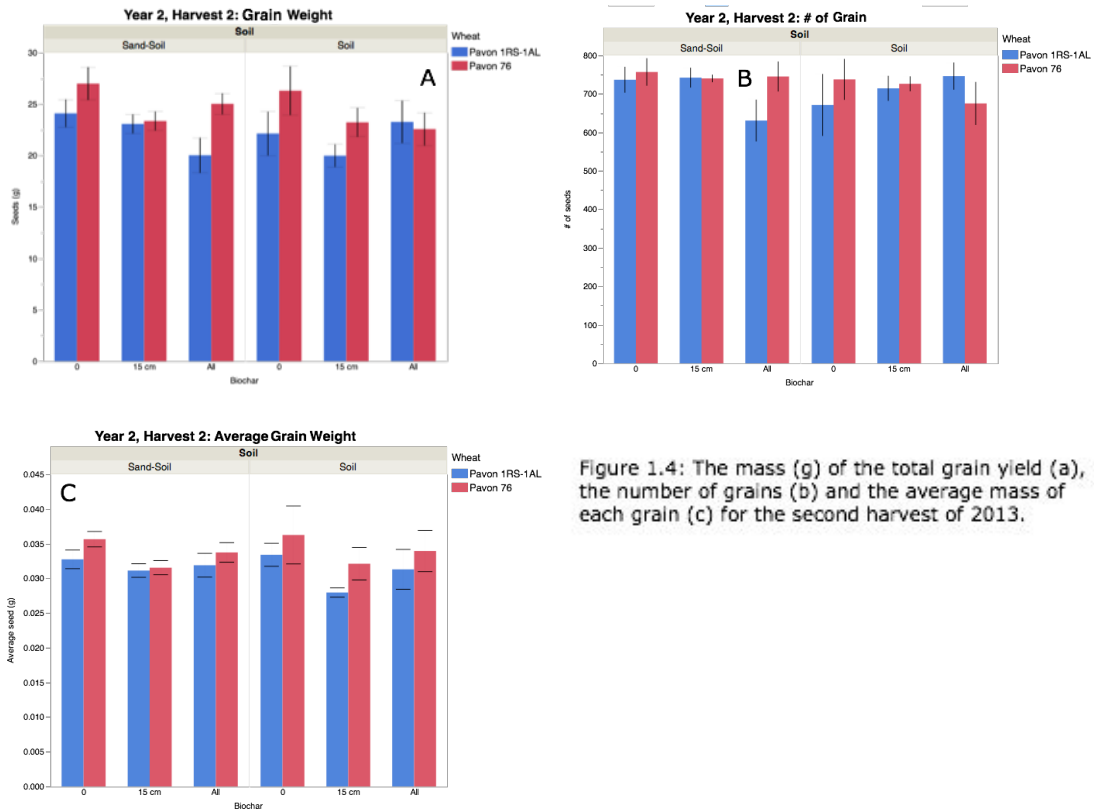


Figure 1.4: The mass (g) of the total grain yield (a), the number of grains (b) and the average mass of each grain (c) for the second harvest of 2013.

Biochar

There was no effect of biochar on the amount of deep roots or the total root biomass (Figures 1.3b and d), but P76-B0 plants had more shallow roots than P76-B15 (Figure 1.3a). However, this did not affect the deep to shallow root ratio (Figure 1.3c). Biochar had no effect on the above ground growth of the plants, vegetative or reproductive (Figures 1.3e – g and Figures 1.4 a -c). Although biochar did not have a positive impact, it did not decrease yield either.

Summary

Just as with the early harvest, the uneven watering scheme lead to waterlogging in the S0 treatments, causing deep roots to die. As a result, SS plants had better root growth. Again, there was not much effect on the growth from the

genotype or biochar application. Although, the data seems to indicate that the P76 genotype is a better grain producer than the P1RS genotype in this situation.

2014: First Harvest

The use of water sensors in 2014 meant that water was only applied when needed, preventing the possibility of waterlogged soil. Therefore, results were more comparable with 2012 than 2013.

Soil Type

In 2014, B75 grew more shallow and deep roots if they were S0 treatments, but the opposite was true for the B15 and B0 treatments (Figures 1.5a and b). This trend was clearly shown in the total root biomass data (Figure 1.5d), but there was no difference in allocation to deep or shallow roots based on the soil type (Figure 1.5c). This same trend was seen in the aboveground biomass and, therefore, the total plant biomass (Figures 1.5e and g). However, there was significantly more root biomass per unit shoot biomass in the sandy treatments, an increase of 16% (Figure 1.5f). Sand's lower WHC meant less plant available water, so even if the SS treatments were getting watered more often, those treatments were drier between watering, which seem to have led to greater root production to acquire more water.

Genotype

Although the wheat genotype did not affect shallow roots (Figure 1.5a), there were significantly more deep roots in the P1RS plants. The P1RS plants produced

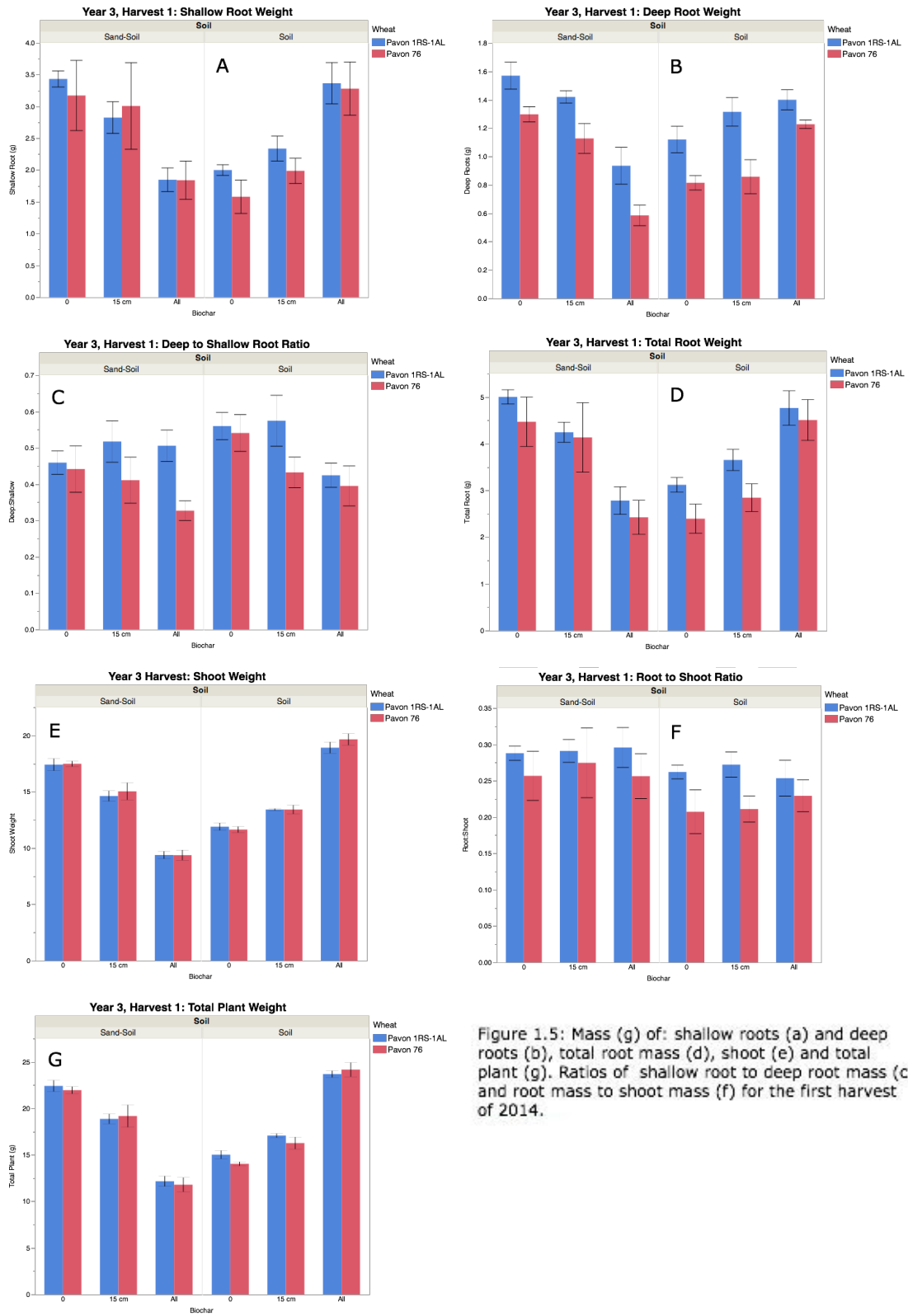


Figure 1.5: Mass (g) of: shallow roots (a) and deep roots (b), total root mass (d), shoot (e) and total plant (g). Ratios of shallow root to deep root mass (c) and root mass to shoot mass (f) for the first harvest of 2014.

1.29 g of deep roots compared to P76's 0.985 g (Figure 1.5b). And there was a corresponding increase in total root biomass, 3.93 g in P1RS to 3.46 g in P76 (Figure 1.5d). And the P1RS plants had 20% higher deep to shallow root ratio compared to the P76 plants (Figure 1.5c). The 1RS.1AL translocation is known to affect root characteristics, but in this case those affects do not translate into changes in shoot growth. Shoot growth was not affected by genotype nor the overall growth of the plants (Figure 1.5e and g). But the greater root growth in the P1RS plants translated into a root to shoot ratio 16% greater than the P76 plants (Figure 1.5f). The translocated plants allocated more biomass to deep root than wheat Pavon 76, as was expected based upon previous studies (Edhaie et al., 2003 and 2012).

Biochar

The roots of wheat plants grown in the SS mixture, both shallow and deep, tended to grow better if they were either B0 or B15 treatments (Figures 1.5a, b, and d). B0 treatments averaged 3.3 g of shallow roots and 1.43 g of deep roots, and the B15 treatments had 2.92 g and 1.27 g, while B75 treatments had only 1.84 g and 0.76 g. When wheat, of either genotype, was grown in S0, it grew best if it had B75 (Figures 1.5a, b, and d). The B75 treatment averaged 3.32 g of shallow roots and 1.31 g of deep roots, while B15 had 2.16 g and 1.09 g and B0 treatments had 1.79 g and 0.986. In general, B0 plants had a 20% higher deep root to shallow root ratio than B75 (Figure 1.5c).

Aboveground biomass followed a similar trend. Plants grown in the SS had the most growth if they were grown in B0 and growth decreased with increasing

amounts of biochar, 17.4 g for B0, 14.8 g for B15, 9.38 g to B75 (Figure 1.5e). Plants grown in SO had more shoot growth as the amount of biochar increased, 11.8 g, 13.4 g and 19.3 g, respectively (Figure 1.5e). Total plant growth followed the exact same pattern as the shoot growth (Figure 1.5g). But biochar did not affect the overall root to shoot ratio (Figure 1.5f). It seems that growth was largely dependent on the amount of water added to the columns, as the growth trends seen in figures 4.4 follow the total irrigation amounts shown in table 4.1.

Summary

In general, the SO treatments tended to have greater growth with more biochar but in SS treatment biochar decreased growth. Perhaps the SO treatments soil was too fine for optimal root production, making it difficult for roots to grow to deeper depths. But the addition of biochar, and thus decreased bulk densities, lead to greater root growth. Since sand has greater porosity, biochar had little effect in the SS treatments. With water content being held constant, the data clearly shows that the translocated plants grew more deep roots than the control.

2014: Second Harvest

Soil Type

The mature plants followed almost the exact same trends in 2014 that the flowering plants did. B0 and B15 plants tended to grow more roots, both shallow and deep, than if grown with SS (Figures 1.6a and b). But B75 plants preferred SO (Figures 1.6a and b). As a result, B75 plants had more root biomass if grown in SO,

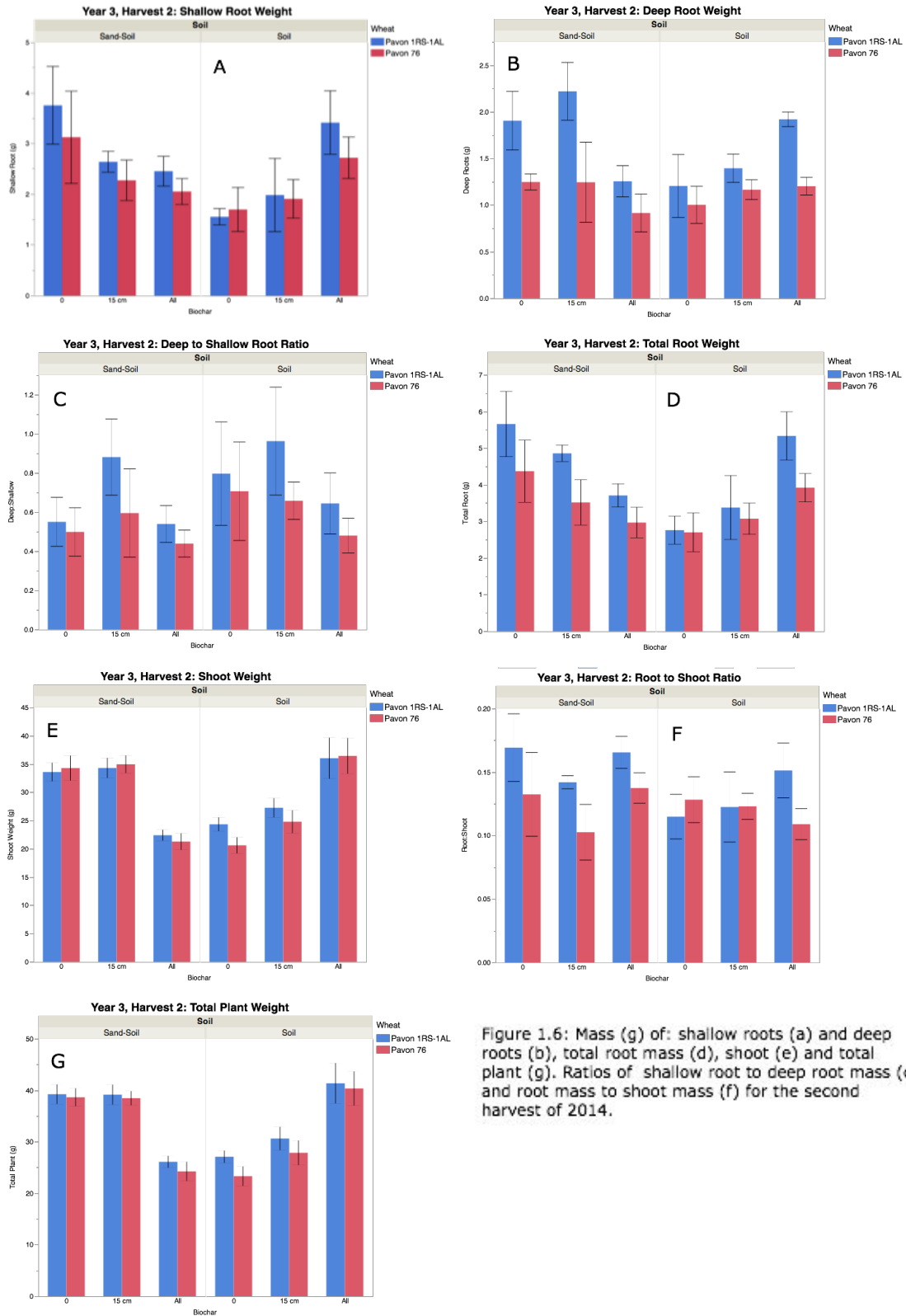


Figure 1.6: Mass (g) of: shallow roots (a) and deep roots (b), total root mass (d), shoot (e) and total plant (g). Ratios of shallow root to deep root mass (c) and root mass to shoot mass (f) for the second harvest of 2014.

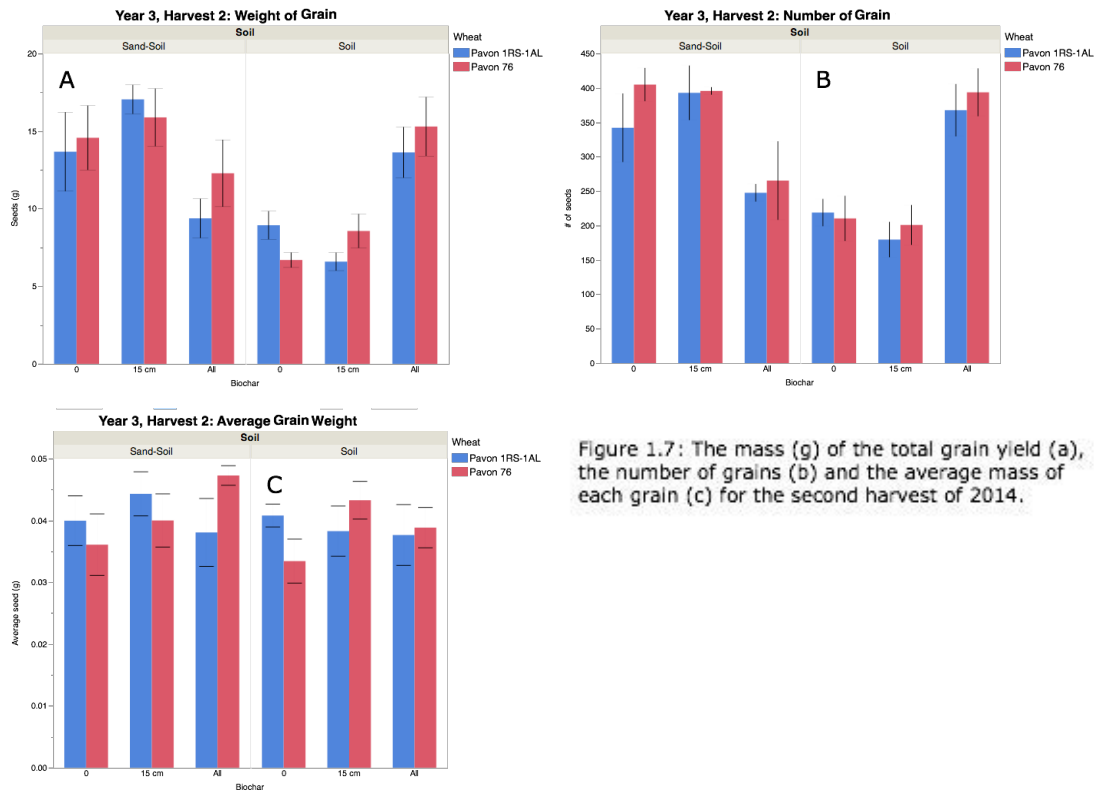


Figure 1.7: The mass (g) of the total grain yield (a), the number of grains (b) and the average mass of each grain (c) for the second harvest of 2014.

but B0 plants had more root biomass if grown with SS (Figure 1.6d). There was no effect on the ratio of shallow and deep roots (Figure 1.6c). The same trend found in the roots was shown in the above ground biomass and, of course, in the total plant biomass (Figures 1.6e and g). But unlike in the younger plants, no difference was found in the root to shoot ratio (Figure 1.6f). The yield data also followed the same trend, B75 produced less grain in sandless media, while B0 or B15 produced more grain in sandy media, both for the total mass of grain and the number of grains (Figure 1.7a and b). There was no effect of the soil type on the average mass or the grain (Figure 1.7c). Just like in the younger plants, growth was likely determined by access to water.

Genotype

The genotype of wheat only impacted the total root biomass, with P1RS averaging 4.28 g and Pavon 76 averaging only 3.42 g (Figure 1.6d). Otherwise genotype had no statistical impact on any measured aspect of plant growth.

Biochar

Again the impact of the media was very similar between the younger and older plants. If grown in SS, the shallow roots were better in B0 or B15, while SO treatments had more shallow roots if grown with B75 (Figure 1.6a). The deep root biomass followed the same pattern for the SS, but there was no impact in the SO treatments (Figure 1.6b). Total root biomass for SO plants followed the same trend as the shallow roots (Figure 1.6d). While for SS plants the total root biomass was greater for B0 plants than the plants with B75 (Figure 1.6d). B15 plants had a greater deep to shallow root ratio than B75 plants (Figure 1.6c). If grown in SS, increasing the amount of biochar in the soil mixture lead to increasing amounts of above ground biomass. But SO treatments had the opposite trend (Figure 1.6e). And the total plant biomass behaved the same as the vast majority of the total biomass was from the above ground portion (Figure 1.6g). There was no effect of the biochar on the root to shoot ratio (Figure 1.6f). For the yield, if grown in SS, the B75 level of biochar lead to more grain, and greater grain yield, than the other treatments, while having no effect on the average grain weight (Figures 1.7a - c).

Summary

The trends observed in the younger plants held true for the mature plants. P1RS had more roots than P76. And more biochar meant more growth for plants grown in SO but less for plants grown in with SS. These results are strongly tied to the water application rates (Table 1.1)

Conclusion

The main way soil medium affected root growth in this experiment was through its WHC. In 2013, the SO treatments had too much water resulting in less root growth. It seems that anoxia from waterlogging may have killed roots. If the plants were very water stressed as in the SS treatments from 2012 they tended to grow a lot of roots to find the available water. But when plants that had access to an adequate amount of water, such as in the 2012 soil treatments, they tended to have the most shoot growth. However if the B0 treatments in 2014, the SO treatments did more poorly. Because water availability was being held constant, this suggests that other factors are responsible for this effect. I propose that it was due to lower root penetration from high bulk density.

In both 2012 and 2014, it was clear that the P1RS line makes produces more root and more deep roots than P76. But greater root growth did not necessarily translate into greater grain yield (Figure 1.5h). In 2013, the P1RS plants actually produced less grain than P76 (Figure 1.3h). This was counter to what Edhaie et al. (2003) found, but it was comparable to what Maheepala et al. (2015) found.

In 2013, there was almost no effect of biochar on any treatment. But in 2012 and 2014, the data showed that biochar improved root growth for plants grown in the SO treatments with higher bulk density. In 2012, this did not translate into greater shoot growth but it did in 2014. But even in 2014, biochar additions did not result in greater grain yield. However, in 2014, SS-B15 and SS-B75 plants did more poorly than SS-B0 plants. This may indicate that without the impact of the WHC or improvements to soil texture, the biochar used in this experiment had a negative impact on plant growth. This could be due to toxic compounds left on the biochar from the pyrolysis process (Hajaligol et al., 2001). But, whatever was the negative effect the biochar may be causing, it was greatly outweighed by the positive effect it had in the denser soil.

Based on the results of this experiment, biochar could be very beneficial in heavy soils that compact greatly. In very loose soils, biochar may not improve plant growth. And in waterlogged soils, the biochar application rates used in this experiments were not sufficient to rescue soil productivity. However, in very sandy soils biochar was able to reduce water needs fairly substantially (Table 4.1), reducing water supplied by 30% in this experiment. But for the SO treatments the water application increased by 35%. Although, there was no significant impact on grain yield, wheat straw still has some commercial uses. Some growers incorporated straw back in to the soil (Ocio et al., 1991), which in addition to the additional root growth, could lead to increases in soil organic matter, which could help to build tilth. Some wheat straw is used for livestock forage (Gebrehiwot and

Mohammed, 1989; Lujia et al., 2003). Additionally, that straw could be used to make biochar.

A long-term field study would help to elucidate what would happen in practice. Biochar applications could provide additional benefits, once it has been “aged” (Major et al., 2010), that were not tracked in this four month experiment. In addition, although the PVC pipes used in this experiment were much deeper than standard pots, wheat roots can grow much deeper than 75 cm. As a result, field trials, where biochar incorporation to a depth of 75 cm would be difficult, may see a reduced effect from biochar amendment since less of the roots would be in contact with the biochar.

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Chapter 2

Effects of biochar on nutrient leaching and begonia plant growth in a nursery setting.

Abstract

Fertilizer runoff into surface water can affect human and ecosystem health. As a result, laws like the Clean Water Act of 1972 have been passed to try to protect waterways. A study was conducted over three years to determine the ability of biochar to reduce fertilizer runoff from nurseries. Biochar is a high carbon substrate that is highly adsorptive. Standard potting mix was augmented with biochar at different rates, ranging from 0% to 30% by volume biochar, and some treatments were planted with *Begonia semperflorens* 'Viva'. The pots were fertilized with a modified Hoagland solution and watered four times a week. The leachate was collected from each pot after watering, and aggregated into weekly samples. Leachate from each week was analyzed photometrically for nitrate, ammonium and ortho-phosphate concentrations. The results suggest that biochar can reduce leaching from the pots of all three ions. The amount of biochar had no bearing on plant growth or nitrogen or phosphorus content of the plant material.

Introduction

Fertilizer runoff from agricultural land and into streams and rivers can cause significant impacts on ecosystems and human health. However, important plant nutrients, such as nitrogen (N) and phosphorus (P), are necessary for plant growth. Therefore, growers need to find with new ways to prevent fertilizer runoff.

A possible solution is a soil amendment called biochar. Biochar is pyrolyzed biomass, much like charcoal. Pyrolysis is the thermochemical decomposition of biomass at temperature upwards of 250° C in the presence of little to no oxygen (Lehmann and Joseph, 2015). One of the key benefits of using biochar is that it is carbon negative (Lehmann, 2007). Biochar half-life can range from tens to hundred thousands of years depending on the O:C ratio (Spokas, 2010). In addition to biochar's carbon sequestering ability, it is often touted as having properties that promote plant growth. Of these properties, most important to this study is its nutrient holding capacity.

Nutrient holding capacity is attributed to biochar in a number of different ways. The highly porous nature of biochar means that it can physically hold dissolved compounds in its water filled pore space (Major et al., 2009). These ions would not be tightly bound but flow out easily with the addition of water. This high surface area means that there is a lot of space where chemical interactions can take place relative to the volume of the biochar.

N found in the soil solution comes in two main forms, nitrate (NO_3^-) and ammonium (NH_4^+). Many studies have shown that biochar can adsorb NH_4^+ (Angst et al., 2013; Asada et al., 2002; Ding et al., 2010; Hale et al., 2013; Lehmann et al., 2003; Steiner et al., 2010). NH_4^+ , as a cation, can be sorbed to the surface of biochars, by way of negative functional groups such as hydroxyls, amines, ethers, esters, and carboxyls (Amonette & Joseph, 2009). This is the mechanism for cation exchange capacity (CEC). Because the bonds formed between the ion and the

functional groups on the biochar are relatively weak electrostatic and nonspecific interactions (Essington, 2004), ions can become attached and detached and replaced by other ions.

Soils and potting media often have components that have a high CEC, but most materials do not have a high anion exchange capacity (AEC), making it useful for NO_3^- adsorption. Several studies have investigated biochar's ability to adsorb NO_3^- . Some of these studies have found no effect (Eykelbosh et al., 2015; Hale et al., 2013; Hollister et al., 2013), while others did find an effect (Chintala et al., 2013; Kameyama et al. 2011). Chintala et al. (2013) suggested that sorption of NO_3^- to the surface of biochar may be due to electrostatic interactions, an ionic exchange mechanism. It is also possible that divalent cations could provide a bridge from the negative functional group to anions such as NO_3^- (Chintala et al., 2013). Also high temperature biochars seem to have higher NO_3^- adsorption (Kameyama et al., 2011). Biochar could also reduce NO_3^- leaching by adsorbing NH_4^+ , therefore decreasing the pool of reactive N in the soil.

If biochars possess relatively high AEC compared to soil, they could be used to prevent P leaching. Some studies have found that biochar is capable of reducing P leaching (Angst et al., 2013; Hale et al., 2013; Hollister et al 2013). Because both NO_3^- and PO_4^{3-} are anions, many of the same mechanisms apply.

This study seeks to understand how well biochar is able to adsorb N and P liquid fertilizer in a nursery setting. This is determined by comparing the amount of NO_3^- , NH_4^+ , and PO_4^{3-} leached out of peat moss based potting mix either augmented

with biochar or not. This study will hopefully give insight into biochar's utility in reducing fertilizer runoff for nursery growers.

Materials and Methods

Cultural Practices

A greenhouse study was conducted over nine weeks in spring and summer of 2010, 2013 and 2014. There were nine treatments in total. Two of them had no plants in them: just the potting mix (N0) and potting mix plus 10% by volume biochar in a layer at the bottom of the pot (N10). The other seven had begonias planted in them: normal potting mix (Y0), potting mix with 5% by volume biochar mixed in (Y5), potting mix with 10% biochar (Y10), 15% biochar (Y15), 20% biochar (Y20), 25% biochar (Y25), 30% biochar (Y30). *Begonia semperflorens* 'Viva' was used because they are very hardy and have very few pests. In spite of this, the begonias in this experiment developed powdery mildew in both 2013 and 2014. In 2014, the disease was kept in check, but in 2013, it had a noticeable impact of plant growth. Each pot sat inside of a bucket on top of a PCV ring, which allowed for the pots to drain into the bucket without the pots sitting in water. The experiments were set up on April 28, 2010; June 7, 2013; and April 29, 2014. Each week, pots were fertilized twice a week with a modified Hoaglands solution (Table 2.1) and watered twice more per week as much as necessary to ensure leaching. Immediately following each watering, the leachate was collected from the bucket and stored in a freezer.

Substrate, Biochar and Fertilizer

The potting media used in 2010 was Sunshine Mix 2, a mix of Canadian sphagnum peat moss, coarse grade perlite, and dolomitic lime. Due to some confusion, in 2013 and 2014, Sunshine Mix 4 was used, which included a fertilizer charge. As a result, the data collected from years 2013 and 2014 have much higher concentrations than 2010. The biochar used was a mix of wheat straw and hardwood biochar obtained from Alterna Energy, pyrolyzed at 650°C for 2 hours. The plants were fertilized with a modified Hoagland solution (Table 2.1) at ~400 mL twice a week and watered with deionized water until the media was saturated.

NH ₄ -N	NO ₃ -N	P	K	S	Ca	Mg	Cl
49.86	50	21.84	83.08	45.85	50.10	25.03	101.40

Table 2.1: The number of ppms of the nutrients in the modified Hoagland solution used in this experiment

Data Collection and Analysis

Leachate from each week was analyzed for pH and EC. Then the leachate was analyzed photometrically for NO₃⁻, NH₄⁺, and PO₄³⁻. At the end of the experiment above ground plant material was collected, dried and weighted. Plant and potting mix samples were analyzed for N and P content. Plants were analyzed with total Kjeldahl Nitrogen (Jones, 1991) for N content, and for P content a “wet ash” method (Kirkpatrick and Bishop, 1971) was used. In 2010 and 2013, NO₃⁻ was extracted using calcium sulfate and NH₄⁺, using potassium chloride. In 2014, both the NO₃⁻ and NH₄⁺ were extracted from the biochar and/or potting mix with a 1M KCl

extraction; and %P was analyzed after a nitric acid and heat digestion, similar to EPA 3050B (EPA, 1996).

Statistical Analysis

NO_3^- , NH_4^+ , total inorganic nitrogen (TIN), and PO_4^{3-} data were all transformed by $y=x^{0.15}$ to improve normality. %N and %P from plant samples and extracted NO_3^- and NH_4^+ and %P from media were not transformed. Means were compared using a Dunnett test on JMP. A P-value of 0.05 or less was considered significant.

Results and Discussion

2010

Ammonium

WITHOUT BEGONIA PLANTS

In 2010, the amount of NH_4^+ leached from the N0 treatment was significantly higher than N10 in weeks 1, 2, 3, 4, 5, and 8 (Figure 2.1a). The total NH_4^+ leached from N0 was 34.0 mg significantly more than N10's just 9.2 mg (Figure 2.1a). The media in N0 and N10 had the same amount of NH_4^+ extracted (Figure 2.1b). However, significantly less NH_4^+ was extracted from the biochar in the N10 treatment than from the potting media (Figure 2.1b). This was perplexing based on the leaching data. If the media was binding the same amount of NH_4^+ and more NH_4^+ was held by N10 we would expect that the biochar is adsorbing more than the potting mix.

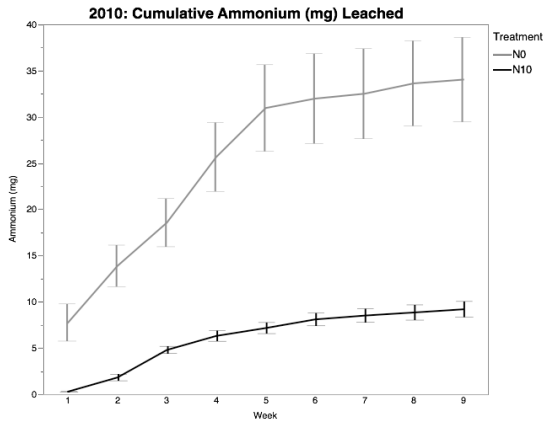


Figure 2.1a

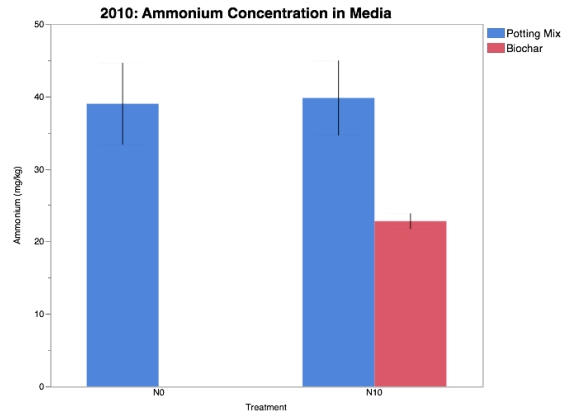


Figure 2.1b

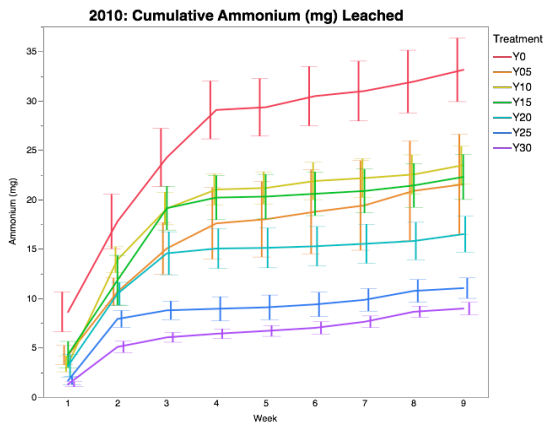


Figure 2.1c

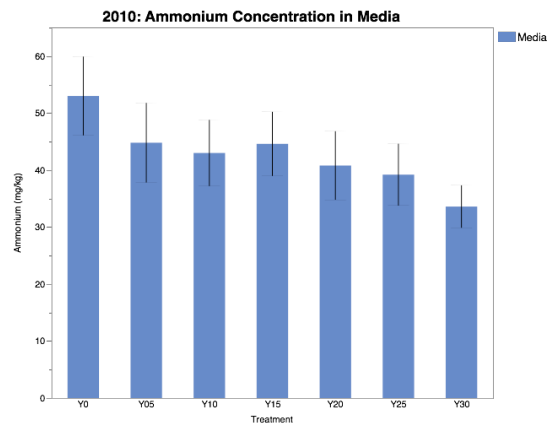


Figure 2.1d

Figure 2.1: The cumulative mg of ammonium leached from the pots for each week of the experiment for the treatments without plants (a) and with a begonia (c). The mg/kg of ammonium extracted from the potting media of those without plants (b; blue bars) and those with a begonia (d; blue bars), in the biochar layer (b; red bar).

WITH BEGONIA PLANTS

In week 1, the Y20, Y25 and Y30 leached significantly more NH_4^+ than Y0, with Y25 and Y30 leaching only about 1.5 mg to the control's 8.6 mg (Figure 2.1c). In week 2, Y30 again leached significantly less NH_4^+ (3.8 mg) than the control (7.2 mg). In week 3, Y25 and Y30 leached less than 1 mg compared to the control

6.4 mg. In week 4, Y10, Y15, Y20, Y25 and Y30 all leached significantly less NH_4^+ than the Y0, with Y30 leaching 0.4 mg and Y0 4.8 mg. The amount of NH_4^+ leached began to plateau for all the treatments and the amount leached was equivalent for all treatments in weeks 5 and 7. In weeks 6 and 8, Y20 had significantly less NH_4^+ leachate than the control but the difference was less than a gram. And in week 9, Y25 and Y30 again had significantly less NH_4^+ leaching than the control, 0.3 mg to 1.2 mg. The cumulative amount of NH_4^+ leached over all nine weeks was significantly lower for Y5, Y15, Y20, Y25, and Y30 compared to Y0 (Figure 2.1c). Y30 leached only about 9.0 mg compared to Y0's 33.1 mg, about one third the amount. However, the amount of NH_4^+ found in the soil mixture at the end of the experiment was statistically equivalent (Figure 2.1d).

Nitrate

WITHOUT BEGONIA PLANTS

The amount of NO_3^- leached was significantly greater in N0 compared to N10 in weeks 1, 5 and 6 (Figure 2.2a). N0 accumulated 139.1 mg of NO_3^- over the 10 weeks significantly more than N10's total of 92.3 mg (Figure 2.2a). The 10% biochar layer cause a reduction of 34% NO_3^- leaching. Unlike the NH_4^+ , NO_3^- leaching was low initially and increased in weeks 4 through 6 before tapering off at the end. NO_3^- concentrations probably increased in the middle due to nitrification of NH_4^+ in the media. Like NH_4^+ , there was no statistical difference between the amount NO_3^- held in the potting mix for the N0 and N10 treatments (Figure 2.2b). Although there was more NO_3^- extracted from the biochar in the N10 treatment, it

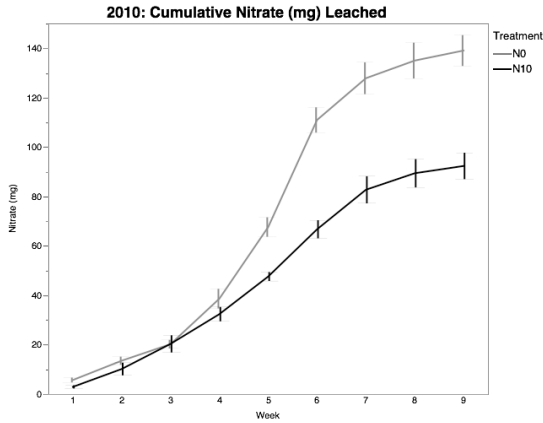


Figure 2.2a

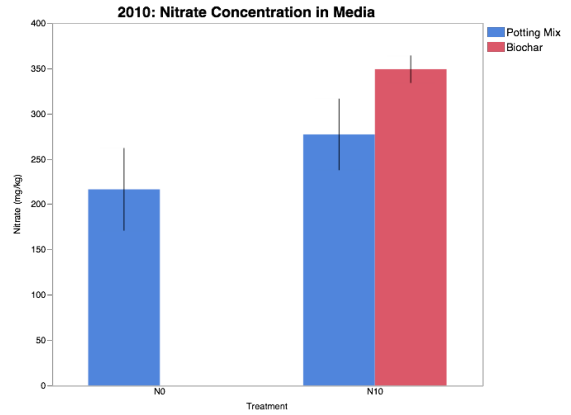


Figure 2.2b

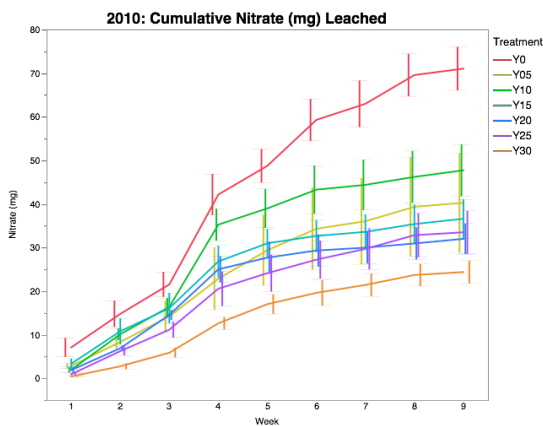


Figure 2.2c

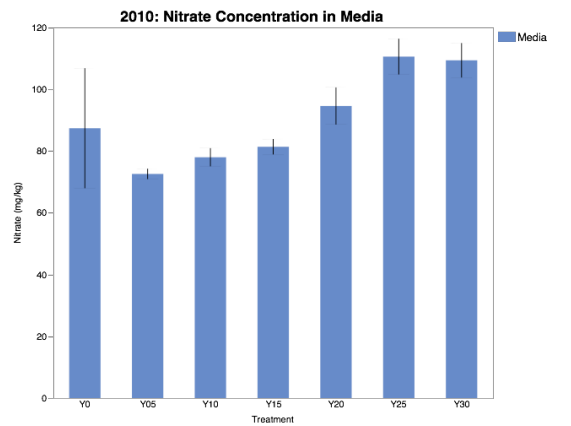


Figure 2.2d

Figure 2.2: The cumulative mg of nitrate leached from the pots for each week of the experiment for the treatments without plants (a) and with a begonia (c). The mg/kg of nitrate extracted from the potting media of those without plants (b; blue bars) and those with a begonia (d; blue bars), in the biochar layer (b; red bar).

was not significantly greater than that found in the potting mix (Figure 2.2b). The reduction in NO_3^- leaching indicates that, like the NH_4^+ .

WITH BEGONIA PLANTS

In the first week, Y10, Y20, Y25 and Y30 all leached significantly less NO_3^- than the control, with Y30 leaching 0.4 mg to Y0's 7.1 mg. The following week, only Y30 leached significantly less than Y0, 2.3 mg to 7.7 mg. But in weeks 3, 5, and 7, there was no statistical difference between the treatments. In week 4, Y5 and Y30 leached significantly less NO_3^- , 8.7 mg and 6.7 mg to 20.6 mg. In week 6, Y15, Y20, and Y30 leached 1.7 mg, 1.6 mg, and 2.6 mg, respectively, significantly less than Y0's 10.5 mg. In week 8, Y10 and Y15 leached about 1.8 mg, and Y20 leached about 1 mg, significantly less than the control, 6.6 mg. In the last week, Y25 and Y30 leached slightly, but significantly less NO_3^- than the control; about 0.7 mg to 1.5 mg. As a result, the cumulative amount of NO_3^- leached from all the biochar treatments was significantly less than the control, with Y30 leaching a total of 24.4 mg, only 34% of Y0's 71.0 mg. However none of the biochar treatments had significantly more or less NO_3^- extracted from the media compared to the control (Figure 2.2d).

Total Inorganic Nitrogen

WITHOUT BEGONIA PLANTS

The TIN is the sum of NH_4^+ and NO_3^- (this assumes that the nitrite levels were low enough to be insignificant). The N levels in N10 were significantly less than N0 in weeks 1, 4, 5, and 6 (Figure 2.3a). Cumulatively, N10 leached 101.5 mg, 59% of N0's 173.1 mg (Figure 2.3a). Just like with the NO_3^- , the amount of TIN found in the

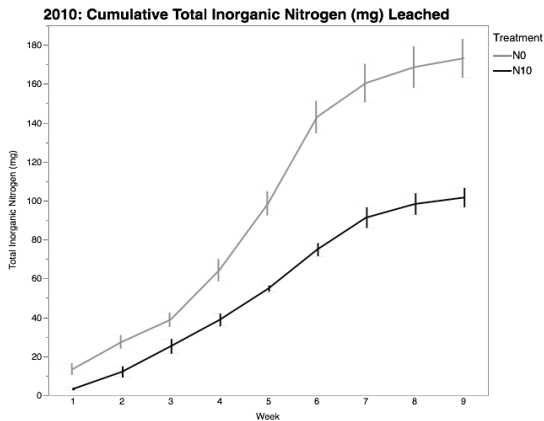


Figure 2.3a

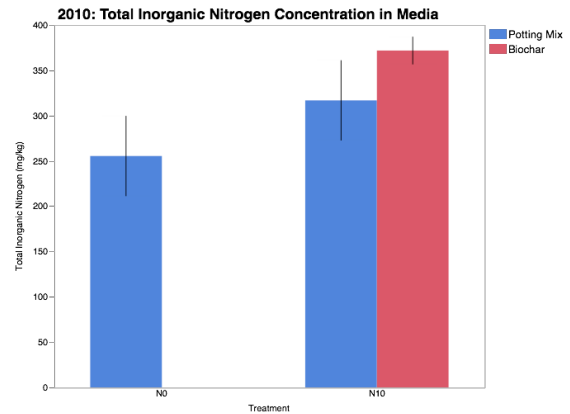


Figure 2.3b

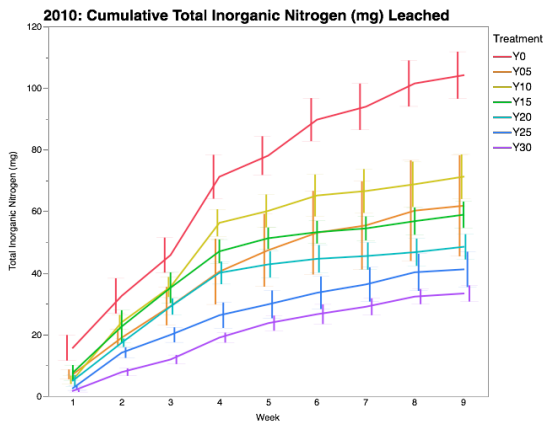


Figure 2.3c

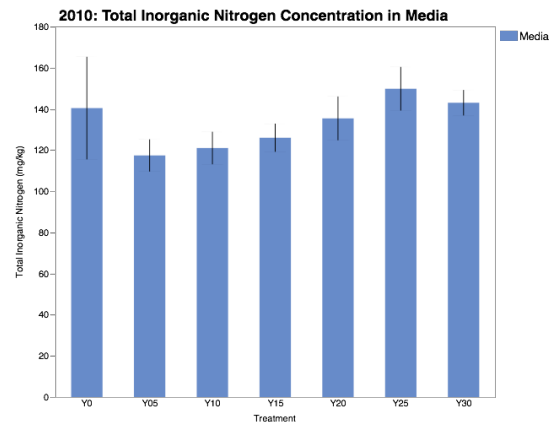


Figure 2.3d

Figure 2.3: The cumulative mg of total inorganic nitrogen leached from the pots for each week of the experiment for the treatments without plants (a) and with a begonia (c). The mg/kg of total inorganic nitrogen extracted from the potting media of those without plants (b; blue bars) and those with a begonia (d; blue bars), and in the biochar layer (b; red bar).

N0 and N10 were equivalent. TIN was also equivalent in the biochar and potting mix in N10 (Figure 2.3b).

WITH BEGONIA PLANTS

In week 1, Y10 (5.3 mg), Y20 (5.0 mg), Y25 (2.6 mg), and Y30 (1.7 mg) all had significantly less N leached out than Y0 (15.7 mg; Figure 2.3c). In week 2 and 3, only

Y30 had significantly less TIN leaching. Y30 leached 6.1 mg and 4.1 mg, in weeks 2 and 3 respectively, compared to the control's 16.8 mg and 13.2 mg. In week 4, Y5, Y20, Y25 and Y30 all leached significantly less TIN than Y0, with Y30 leaching only 7.1 mg compared to Y0's 25.4 mg. But in weeks 5 and 7, none of the treatments were significantly different from each other. In week 6, Y15, Y20 and Y30 all leached significantly less TIN than Y0, with Y30 leaching just 2.9 mg compared to Y0 with 11.7 mg. In week 8, Y10, Y15 and Y20 leached significantly less TIN than Y0 did; Y20 leached 1.2 mg compared to Y0 at 7.6 mg. And in the last week, Y25 and Y30 leached the least, with about 1.0 mg TIN, significantly less than Y0 (2.7 g). Overall, Y30 leached a total of 33.3 mg, which was only 31.9% of Y0's 104.2 mg (Figure 2.3c). Again there was no significant difference between the different treatments on TIN extracted from the media (Figure 2.3d). Nor were there any differences in the percent N in the begonias (Figure 2.3d).

Phosphate

WITHOUT BEGONIA PLANTS

N10 leached less PO_4^{3-} than N0 in almost every week, 1, 2, 4, 5, 6, 7, and 8 (Figure 3.4a). After week 6 the amount of PO_4^{3-} leached tapers off, similar to the NO_3^- and NH_4^+ curves (Figure 2.4a). The total amount of PO_4^{3-} leached from N0 was 26.2 mg, significantly more than N10, which leached only 10.5 mg (Figure 2.4a). Additionally, the biochar in N10 was found to contain more P than the potting mix (Figure 2.4b). And the N0 potting mix had significantly less P than N10 (Figure

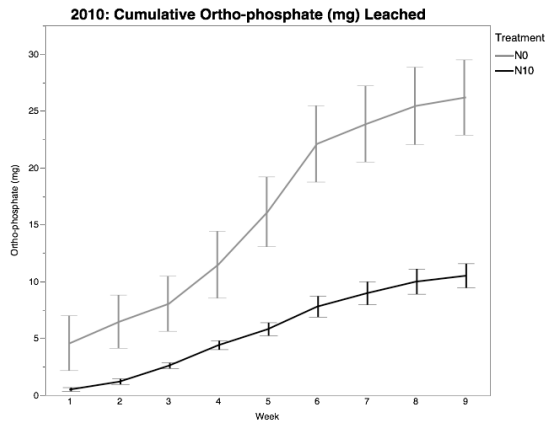


Figure 2.4a

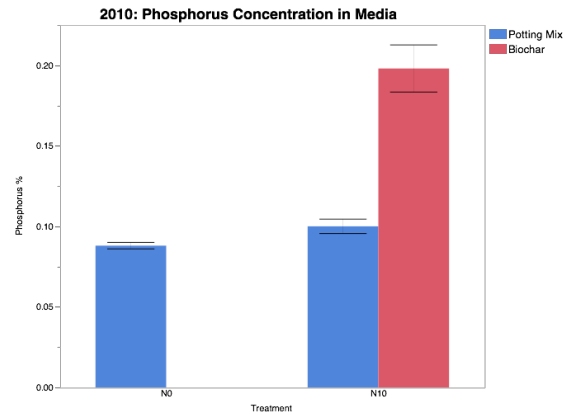


Figure 2.4b

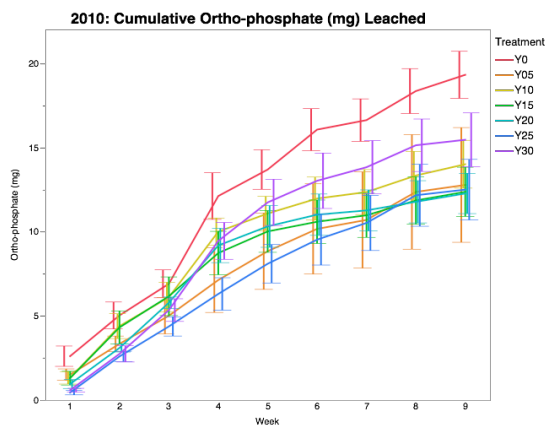


Figure 2.4c

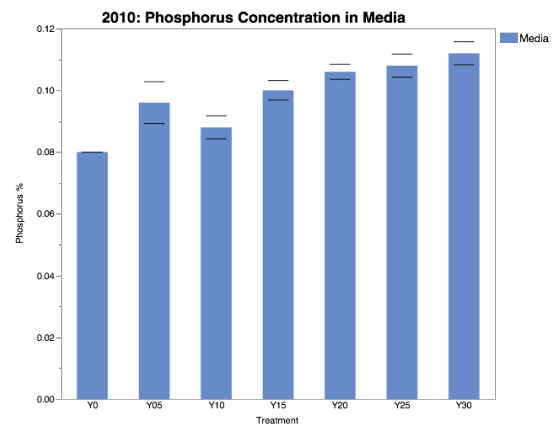


Figure 2.4d

Figure 2.4: The cumulative mg of ortho-phosphate leached from the pots for each week of the experiment for the treatments without plants (a) and with a begonia (c). The percent phosphorous found in the potting media of those without plants (b; blue bars) and those with a begonia (d; blue bars), and in the biochar layer (b; red bar).

2.4b), presumably because the biochar had a greater holding capacity for P, which moved from biochar active sites to potting mix active sites.

WITH BEGONIA PLANTS

Although the relationship between P uptake and percent biochar seems very strong in the treatments without plants, it was much weaker in the treatments with begonias growing in them. Most weeks, 2, 3, 5, 6, and 7, there was

no significant difference between the amounts of PO_4^{3-} leached from any of the treatments (Figure 2.4c). In the first and last weeks, Y25 and Y30 had significantly less PO_4^{3-} leaching compared to Y0. Y0 leached 2.6 mg while Y25 leached 0.4 mg and Y30 leached 0.6 mg in week 1; in week 9, Y25 and Y30 each leached about 0.3 mg compared to Y0's 1 mg. In week 4, Y5 leached significantly less than the control and, in week 8, Y20. Although Y25 only leached an average of 12.3 mg compared to Y0's 19.3 mg there was no significant difference between any of the treatments. Although we can see from the treatments without plants that biochar did have an impact on PO_4^{3-} leaching, with the addition of plants to complicate the picture, that impact becomes insignificant. There was significantly more P in the media of Y5, Y15, Y20, Y25 and Y30 than Y0 (Figure 2.4d).

2013

Ammonium

WITHOUT BEGONIA PLANTS

Unlike in 2010, the amount of NH_4^+ leached from the plant-less treatments was rarely significant (Figure 2.5a). Only in week 8 did N10 (1.4 mg) leach less than N0 (3.5 mg). And there was no difference between the two treatments in NH_4^+ leached over the nine weeks (Figure 2.5a). Similar to 2010, the amount of NH_4^+ found in the potting mix from N0 and N10 were similar and the biochar had significantly less NH_4^+ than the potting mix for N10 (Figure 2.5b).

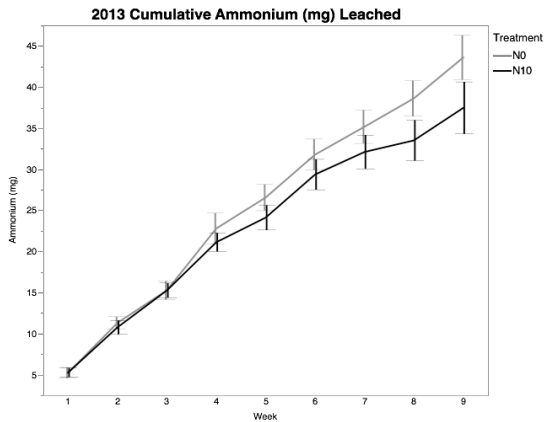


Figure 2.5a

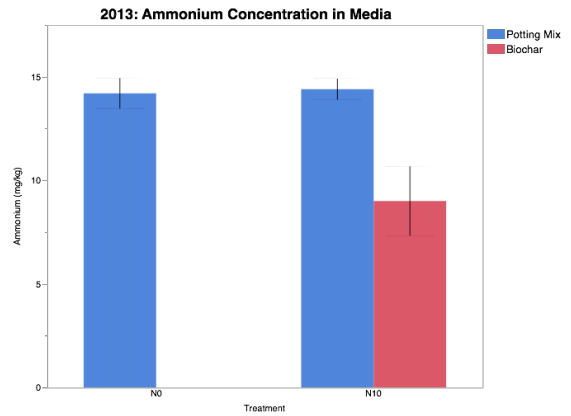


Figure 2.5b

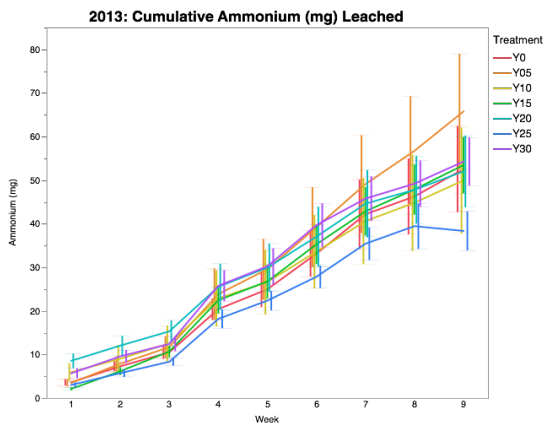


Figure 2.5a

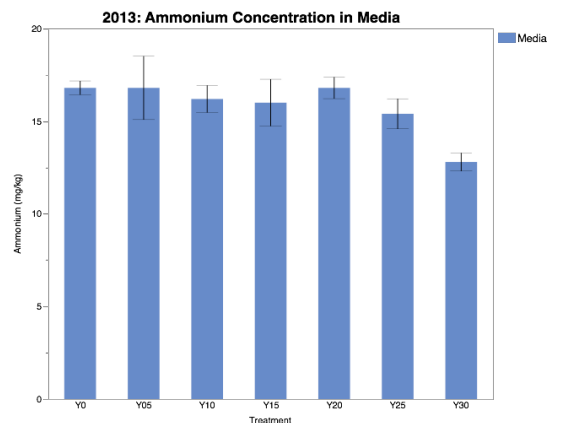


Figure 2.5b

Figure 2.5: The cumulative mg of ammonium leached from the pots for each week of the experiment for the treatments without plants (a) and with a begonia (c). The mg/kg of ammonium extracted from the potting media of those without plants (b; blue bars) and those with a begonia (d; blue bars), in the biochar layer (b; red bar).

WITH BEGONIA PLANTS

Again unlike in 2010 the amount of NH_4^+ leached was rarely significant in the treatments with plants; only in week 1, Y20 leached significantly less than (Figure 2.5c). And the sum of all the NH_4^+ leached over the nine week experiment was not significantly different between the treatments. Also the total amount of NH_4^+

leached in 2013 was about twice than of 2010. However there was significantly less NH_4^+ extracted from Y30 compared to Y0 (Figure 2.5d). Given that the amount of NH_4^+ extracted from the biochar in the N10 treatment this seems a reasonable finding.

Nitrate

WITHOUT BEGONIA PLANTS

Similar to 2010, the amount of NO_3^- leached from N10 was significantly less than N0 in weeks 1, 2, 3, 4, 6 and 8. Making the total amount of NO_3^- leached from N10 significantly less than N0 (Figure 2.6a). However, the amount leached was substantially more than what was leached during 2010. N0 leached 1223.5 mg in 2013, but only 139 mg in 2010, and N10 leached 712.1 mg in 2013, but 92 in 2010 (Figures 2.2a and 2.6a). This was likely due to the addition of fertilizer to the potting mix. However the results for the NO_3^- extraction from the potting mix and biochar were comparable to 2010. There was no significant difference between the biochar and potting mix or between the potting mix of N0 and N10 (Figure 2.6b).

WITH BEGONIA PLANTS

However in 2013, there was no significance in the amount of NO_3^- leached from the pots with begonias in any week, nor was there any significance found when the weeks were summed (Figure 2.6c). This was in contrast to the data found in the N0 and N10 treatments and the data from 2010. In addition the amount of NO_3^- leached was about a factor of ten greater in 2013 than it was in 2010. Again this

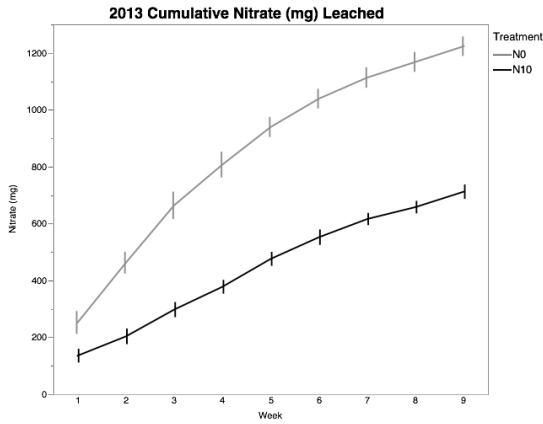


Figure 2.6a

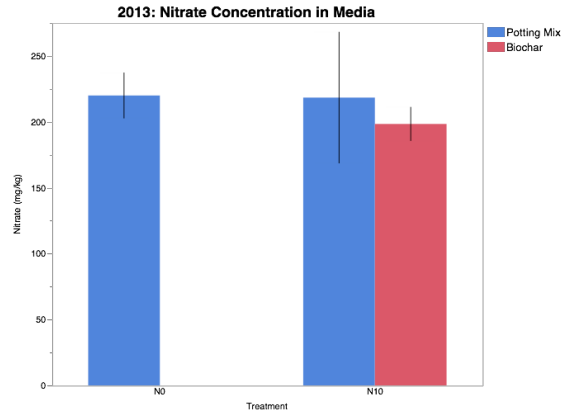


Figure 2.6b

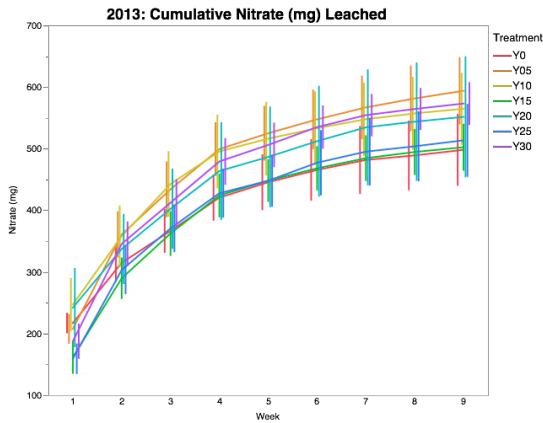


Figure 2.6c

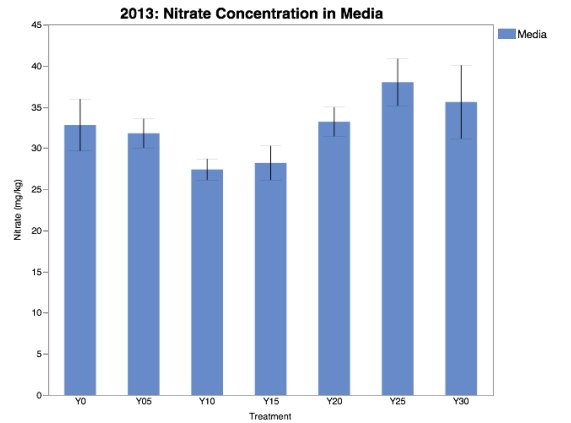


Figure 2.6d

Figure 2.6: The cumulative mg of nitrate leached from the pots for each week of the experiment for the treatments without plants (a) and with a begonia (c). The mg/kg of nitrate extracted from the potting media of those without plants (b; blue bars) and those with a begonia (d; blue bars), in the biochar layer (b; red bar).

may be due to the fertilizer charge in the potting mix or potentially because of the powdery mildew weakened the begonias, reducing nitrate uptake by the plant. And in the soil, there was no significant difference between the different treatments (Figure 2.6d), this trend matches up well with the data from N0 and N10, as well as the data from 2010.

Total Inorganic Nitrogen

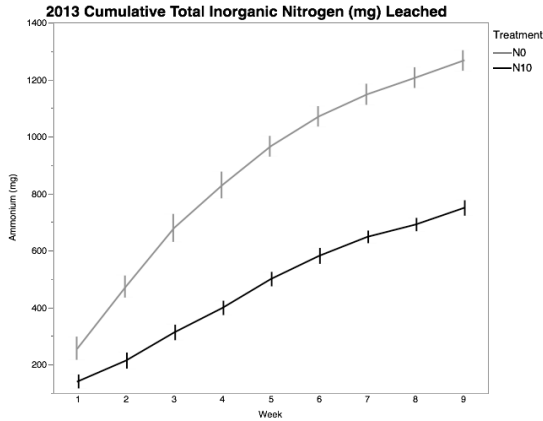


Figure 2.7a

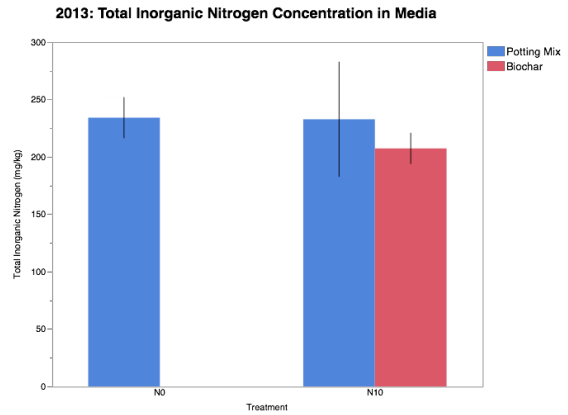


Figure 2.7b

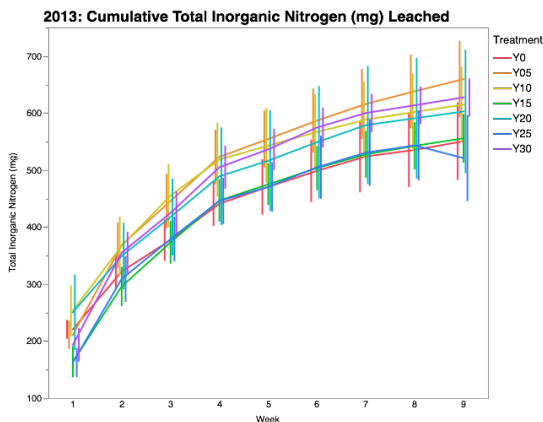


Figure 2.7c

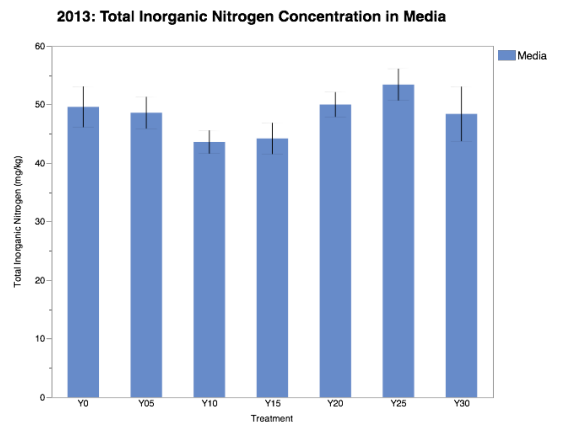


Figure 2.7d

Figure 2.7: The cumulative mg of total inorganic nitrogen leached from the pots for each week of the experiment for the treatments without plants (a) and with a begonia (c). The mg/kg of total inorganic nitrogen extracted from the potting media of those without plants (b; blue bars) and those with a begonia (d; blue bars), in the biochar layer (b; red bar).

WITHOUT BEGONIA PLANTS

The amount of TIN leached from N10 was significantly less than that from N0 in weeks 1, 2, 3, 4, 6, and 8 (Figure 2.7a), which makes sense considering the vast majority of the TIN came from NO_3^- (Figure 2.6a). The total amount of TIN leached from N10 was 749.5 mg, about 59% of what leached from N0 (Figure 2.7a). And just like with the nitrate, there was no difference between the amount of N extracted

from the biochar and the potting mix for N10, or between the potting mix in N0 versus N10 (Figure 2.7b).

WITH BEGONIA PLANTS

Since the amount of NO_3^- and NH_4^+ leached from the different treatments across the nine weeks was not significantly different, it was not surprising to see that there was no significance to be found in the amount of TIN leached in individual weeks or the sum of those weeks (Figure 2.7c). There was also no significant difference in the amount of TIN extracted from the media or found in the plant tissue for the different treatments (Figure 2.7d).

Phosphate

WITHOUT BEGONIA PLANTS

Although N0 leached slightly more than N10, there was no significant difference in leaching in any of the weeks or in the sum of the leachate (Figure 2.8a). And the total amount of PO_4^{3-} leached was about four times or more as much as was leached in 2010, potentially due to a fertilizer charge in the potting mix. There was no statistical difference in the amount of P found in the potting mix and biochar of N10 or in the potting mix of N0 and N10 (Figure 2.8b).

WITH BEGONIA PLANTS

Just like with NO_3^- and NH_4^+ there was no significant difference between any treatments over the nine weeks (Figure 2.8c). Again suggesting that something went wrong with the fertilizer status of the initial ingredients, most likely in the potting mix, but potentially the DI water could have been contaminated. There was

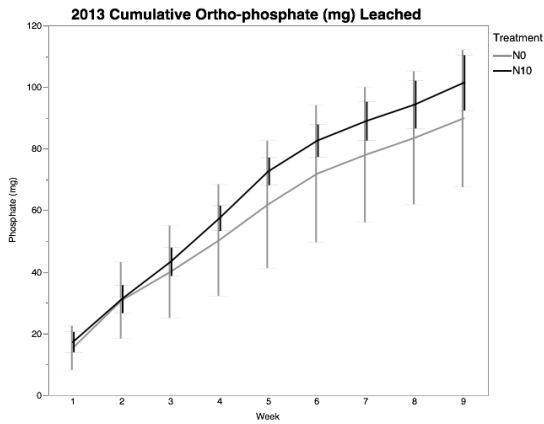


Figure 2.8a

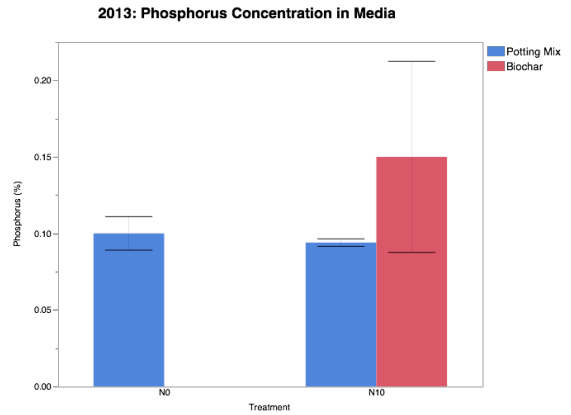


Figure 2.8b

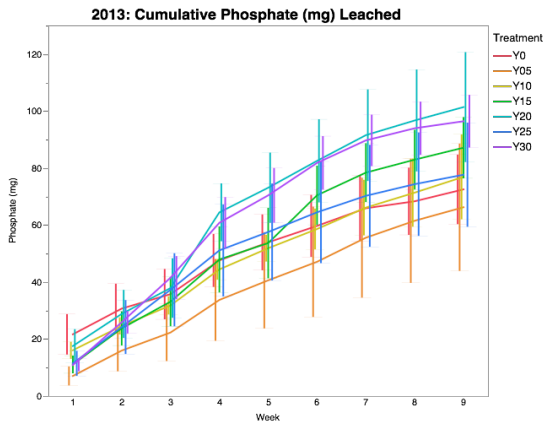


Figure 2.8c

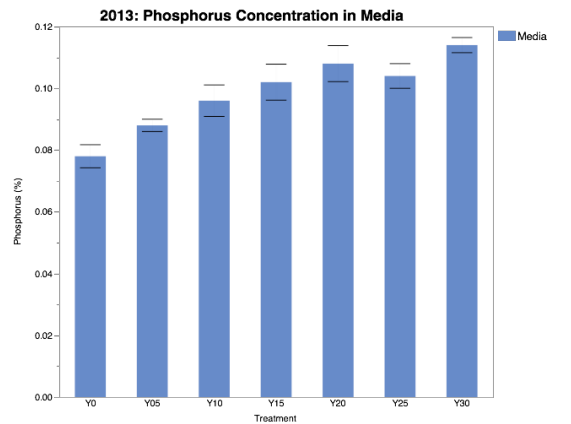


Figure 2.8d

Figure 2.8: The cumulative mg of ortho-phosphate leached from the pots for each week of the experiment for the treatments without plants (a) and with a begonia (c). The percent phosphorous found in the potting media of those without plants (b; blue bars) and those with a begonia (d; blue bars), and in the biochar layer (b; red bar).

an increase in the amount of P found in the media depending on how much biochar was added (Figure 2.8d). Y10-Y30 all had significantly more P than Y0.

2014

Ammonium

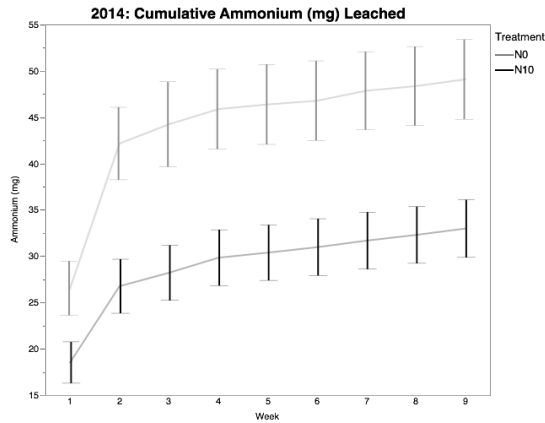


Figure 2.9a

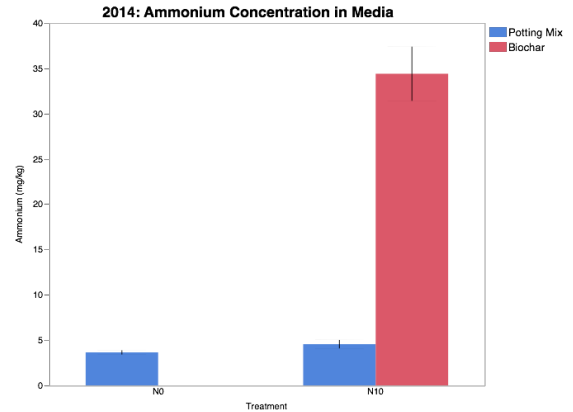


Figure 2.9b

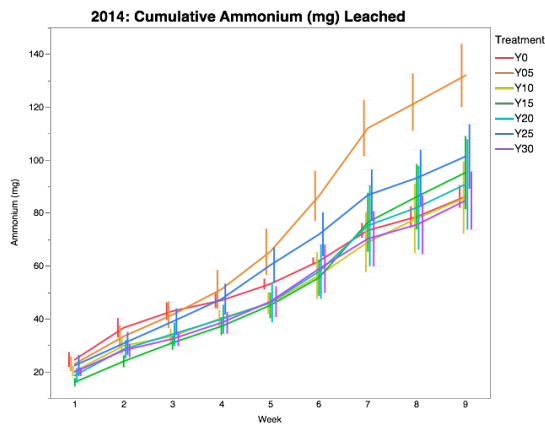


Figure 2.9c

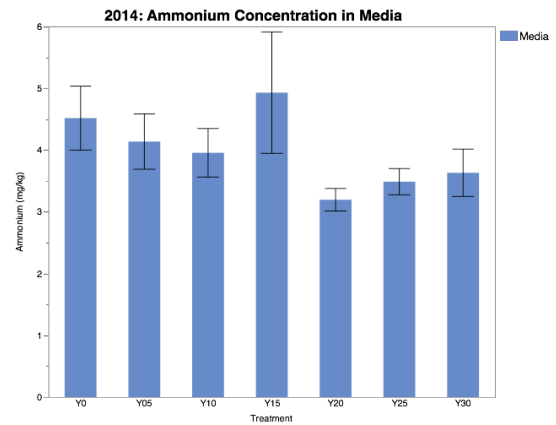


Figure 2.9d

Figure 2.9: The cumulative mg of ammonium leached from the pots for each week of the experiment for the treatments without plants (a) and with a begonia (c). The mg/kg of ammonium extracted from the potting media of those without plants (b; blue bars) and those with a begonia (d; blue bars), in the biochar layer (b; red bar).

WITHOUT BEGONIA PLANTS

In the last year, the amount of NH_4^+ leached out of N10 was significantly less than N0 in the first two weeks (Figure 2.9a). However that was enough to make the total amount of NH_4^+ leached significant. N10 leached 33.0 mg, about two-thirds the

amount leached from N0. There was also significantly more NH_4^+ extracted from the N10 biochar than from the potting mix (Figure 2.9b). This was very different from the 2010 and 2013 results, but a different extraction method was used in this year. In this year, the extraction data helps support the idea that the concept that the biochar was adsorbing the NH_4^+ , preventing leaching from the pots.

WITH BEGONIA PLANTS

Just as with 2013, the amount of NH_4^+ leached in 2014 was not significantly affected by the presence of biochar. There was no statistical difference between the amounts of NH_4^+ leached from the different weeks except in week 5 where Y5 leached more NH_4^+ than Y0 (Figure 2.9c). As a result, there was no significant difference in the total NH_4^+ leached over the nine weeks. And there was no statistical difference in the amount of NH_4^+ extracted from the potting mix of these different treatments (Figure 2.9d).

Nitrate

WITHOUT BEGONIA PLANTS

Unlike in 2010 or 2013, there was no significant difference in the NO_3^- leaching from N0 and N10, not in any individual week, nor in the sum of the weeks (Figure 2.10a). However there was a significant difference in the amount of NO_3^- extracted from the biochar and the potting mix in N10 (Figure 2.10b). This stands in contrast to the results found in 2010 and 2013, but, as mentioned previously, the extraction method was different.

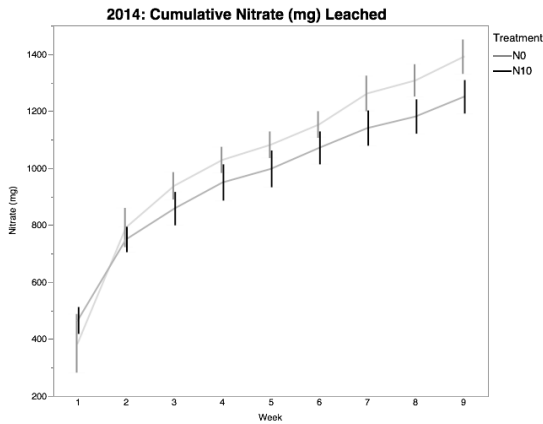


Figure 2.10a

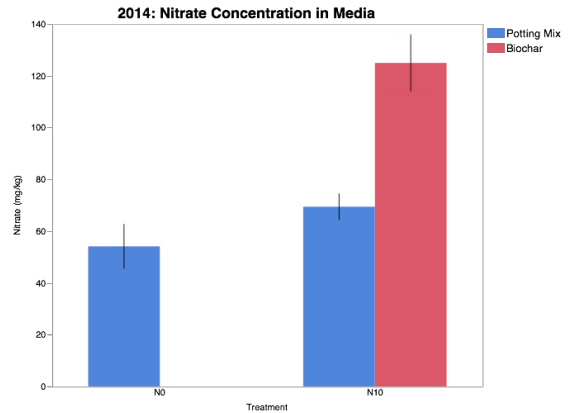


Figure 2.10b

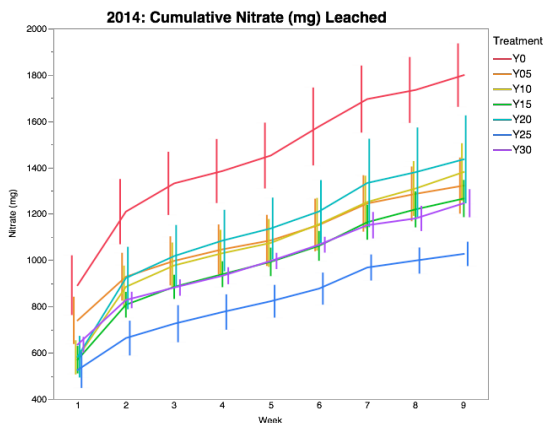


Figure 2.10c

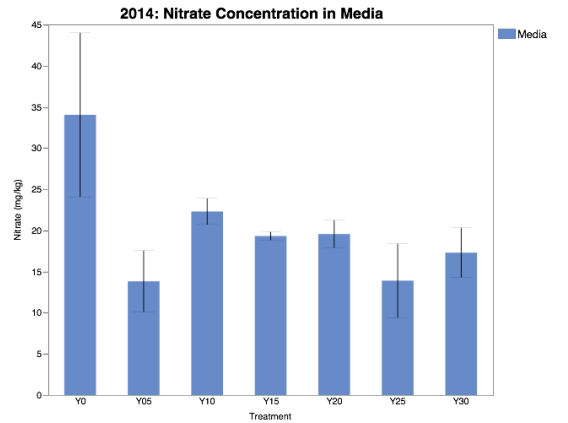


Figure 2.10d

Figure 2.10: The cumulative mg of nitrate leached from the pots for each week of the experiment for the treatments without plants (a) and with a begonia (c). The mg/kg of nitrate extracted from the potting media of those without plants (b; blue bars) and those with a begonia (d; blue bars), in the biochar layer (b; red bar).

WITH BEGONIA PLANTS

Even though in 2014 the amount of NO_3^- leached was not affected by the presence of biochar for N0 and N10, when begonias were added, there was a correlation between the presence of biochar and nitrate leaching (Figure 2.9c). In week 1, Y25 had significantly less NO_3^- leaching than Y0. Y25 leached only 525.8 mg to Y0's 891.9 mg. While there were differences in the amounts leached, the amounts

were much more than 2013, which was in turn more than 2010. The exact cause of this increase is unknown, but the most likely due to the fertilizer charge added to the potting mix. In week 2, Y25 again had the least leaching, with only 137.7 mg, significantly less than Y0 at 317.7 mg. In week 3, Y5, Y15, Y25 and Y30 all leached significantly less than Y0. Y30 leached just 53.8 mg, much less than 122.0 mg from Y0. However, after week 3 there was no difference between treatments until week 9. In week 9, only Y25 (28.8 mg) leached significantly less NO_3^- than Y0 (71.7 mg). Over the nine weeks of the experiment, Y5 (1322.0 mg), Y10 (1380.6 mg), Y15 (1266.0 mg), Y25 (1027.0 mg), and Y30 (1245.8 mg) all leached less NO_3^- than the control (1799.0 mg). Contrary to our expectations, the amount of NO_3^- found in the potting mix was significantly less in Y5 and Y20 (Figure 2.10d). In previous years, there was no significant difference found in the amount of NO_3^- extracted from the Y treatments. This also runs contrary to the findings of the N treatments (Figure 2.10b). Perhaps healthier plants were able to extract the NO_3^- from the media reducing what was left to extract after the experiment was over.

Total Inorganic Nitrogen

WITHOUT BEGONIA PLANTS

Just as with the NO_3^- there was no difference in the amount of TIN leached from N0 and N10 (Figure 2.11a). This makes sense given how much NO_3^- contributes to TIN. And as with the NO_3^- , the amount of N in the biochar of N10 was significantly more than that of the potting mix (Figure 2.11b).

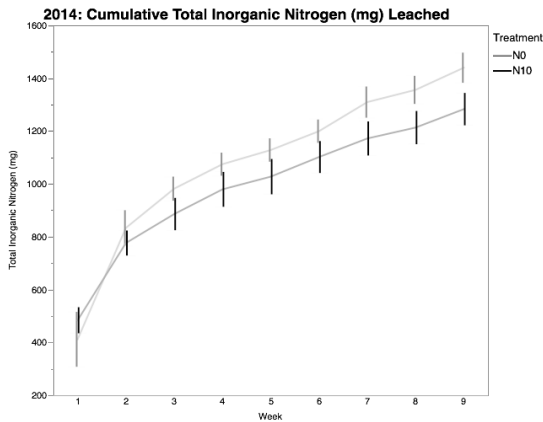


Figure 2.11a

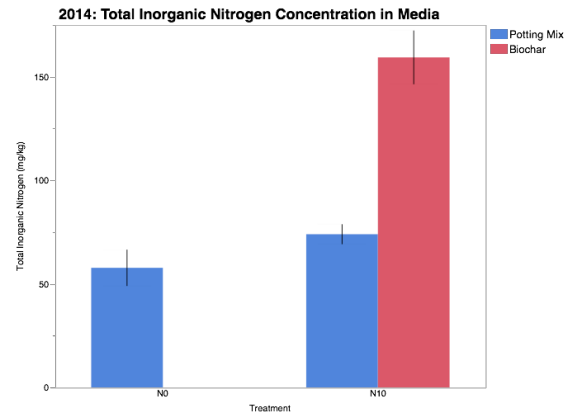


Figure 2.11b

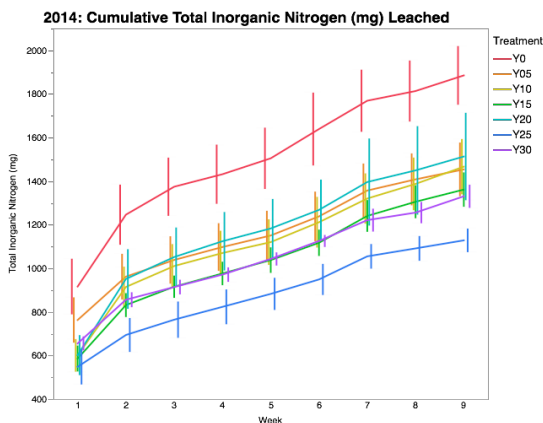


Figure 2.11c

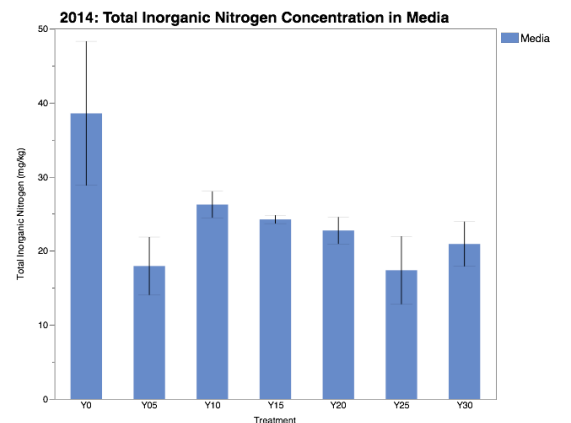


Figure 2.11d

Figure 2.11: The cumulative mg of total inorganic nitrogen leached from the pots for each week of the experiment for the treatments without plants (a) and with a begonia (c). The mg/kg of total inorganic nitrogen extracted from the potting media of those without plants (b; blue bars) and those with a begonia (d; blue bars), and in the biochar layer (b; red bar).

WITH BEGONIA PLANTS

The amount of TIN leached from Y25 was significantly less in weeks 1 and 2 compared to Y0 (Figure 2.11c). In week 1, Y25 leached 548.1 mg and Y0, 916.4 mg; and in week 2, 146.1 and 329.7mg respectively. In week 3, Y5, Y15, Y25 and Y30 all leached less than Y0, just as with NO_3^- . Y30 leached 58.1 mg, much less than Y0, at 128.2 mg. And in week 9, Y25 leached only 36.9 mg, which was significantly less

than Y0's 79.0 mg. Over the nine weeks, Y15, Y25 and Y30 all leached significantly less than Y0. Y25 leached 1128.3 mg, which was about 60% of the control, which leached 1885.2 mg (Figure 2.11c). Y5 and Y25 had significantly less N than did the control (Figure 2.11d).

Phosphate

WITHOUT BEGONIA PLANTS

The amount of PO_4^{3-} leached from the N10 was less than N0 in the first few weeks of the experiment. In weeks 2, 3, and 5, N10 leached less PO_4^{3-} than N0 (Figure 2.12a). Over all nine weeks the total amount of PO_4^{3-} leached from N0 was 154.0 mg while N10 leached 112.3 mg, a 27% decrease (Figure 2.12a). In this case as well, the data shows that the biochar from N10 had significantly more P than the potting mix did, although the amount of P in the potting mix from N0 was not different from N10 (Figure 2.12b).

WITH BEGONIA PLANTS

Unlike in 2013, there was a significant impact of the biochar on the PO_4^{3-} leaching from the planted treatments (Figure 2.12c). In week 1, all the treatments with biochar leached significantly less PO_4^{3-} than Y0. Y30 leached 17.8 mg, much less than Y0, which leached 101.7 mg. In week 2, Y5, Y15, Y25 and Y30 leached significantly less than the control. Y30 leached 16.0 mg compared to Y0, which leached 52.1 mg. In week 3, Y5 (6.5 mg), Y10 (6.8 mg), and Y30 (6.4 mg) all leached significantly less PO_4^{3-} than Y0 (13.0 mg). But after week 3, there was no statistical

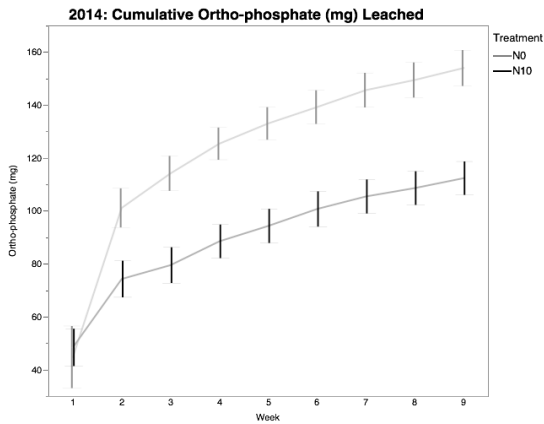


Figure 2.12a

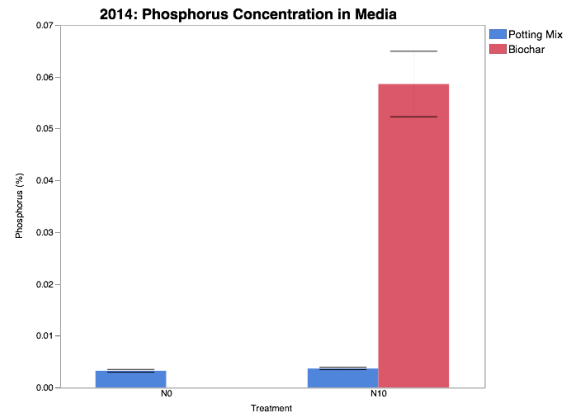


Figure 2.12b

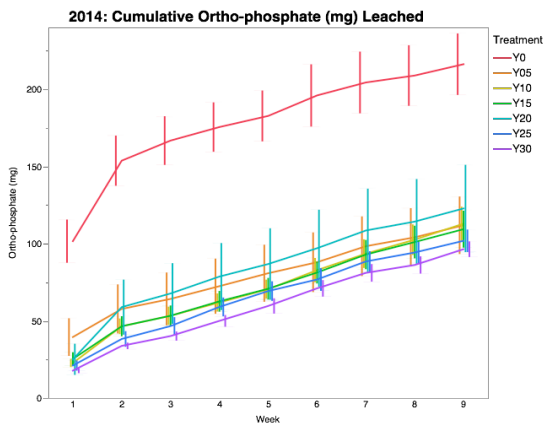


Figure 2.12c

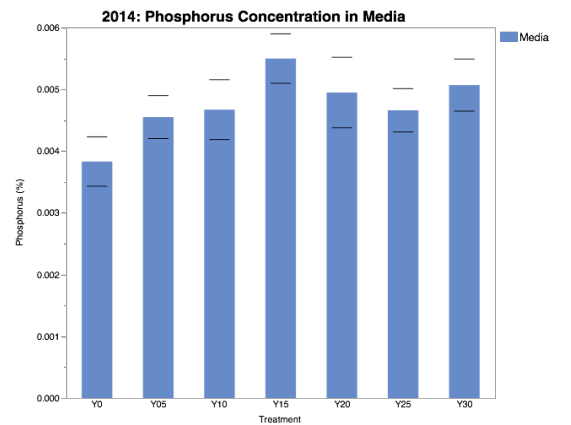


Figure 2.12d

Figure 2.12: The cumulative mg of ortho-phosphate leached from the pots for each week of the experiment for the treatments without plants (a) and with a begonia (c). The percent phosphorous found in the potting media of those without plants (b; blue bars) and those with a begonia (d; blue bars), in the biochar layer (b; red bar) and the in the begonias themselves (d; red bars).

difference between the treatments. However, because of the strong affect of the biochar towards the beginning of the experiment there was a statistical difference between all the biochar treatments and the control (Figure 2.12c). Y30 leached the least, 96.4 mg, which was only about 44.6% of Y0 (216.3 g). However, unlike in previous years where P content of the media increased with biochar content, there was very little difference in the amount of P in the soil (Figure 2.12d).

Conclusion

Although biochar is touted as a plant growth promoting amendment, in this experiment we found no evidence to support the claim. All of the data collected from the plant tissue showed all treatments to be the same, dry shoot biomass, %N and %P (Data not shown). Although in this experiment biochar did not improve plant growth, nor did it cause any detriment to the begonias. This is not an unusual finding; Spokas et al. (2012) found that approximately 30% of studies reviewed showed no effect of biochar on plant growth.

The presence of biochar was significantly correlated to reductions of NH_4^+ leaching in 2010 and 2014. In 2010, the 10% biochar disk reduced NH_4^+ leaching in a system without plants by 73%, and, in 2014, by 33%. In 2010 for treatments with begonias growing in them, 5% biochar reduced leaching by 35%, 15% biochar resulted in a 33% reduction, 20% biochar lead to a 50% reduction, 25% biochar lead to a 67% reduction and 30%, 73%. But in the following years there was no effect on NH_4^+ leaching. And in the cases where biochar did have an effect on the NH_4^+ leaching it was mostly towards the beginning of the experiment. In 2010, when the overall concentrations of NH_4^+ leached were low, there were more weeks where the biochar had a significant impact. But in 2013 and 2014, when concentrations were unexpectedly high, there was either no effect of biochar or it was only effective in the every beginning of the experiment. The amount of NH_4^+ extracted from the media was almost never significantly different from the control (except Y30 in 2013), which is expected, given that NH_4^+ leaching was reduced and

the plant growth was not changing (or was not an issue). This may be explained by the extraction of NH_4^+ from the biochar layer of N10. In 2010 and 2013, the biochar in N10 had significantly less NH_4^+ extracted than the potting mix from the same pots. Since in 2010, the amount of NH_4^+ leached was less in the N10 treatment, but the NH_4^+ extracted was less in the N10 treatment, this suggests there may be something wrong with extraction method, since the layer of biochar was the only difference between the N0 and N10 treatments. Many groups have found that NH_4^+ is adsorbed by biochars (Angst et al., 2013; Hale et al., 2013).

The total amount of NO_3^- leached from the pots was significantly different for the biochar treatments compared to the control treatment in 2010 for both the treatments with and without begonias, in 2013 for the treatments without begonias and in 2014 for the treatments with begonias. These results are comparable to Chintala et al. (2013). The N10 treatments had 34% less NO_3^- leaching than N0 in 2010 and 42% less in 2013. In 2010 in the Y treatments, 5%, 10%, 15%, 20%, 25% and 30% biochar lead to 43%, 33%, 48%, 70%, 53%, and 66% reduction, respectively. In 2014, there were significant reductions in 5%, 10%, 15%, 25% and 30% biochar incorporations, 27%, 23%, 30%, 43%, and 31%, respectively. Since the biochar tended to have a greater impact of NO_3^- leaching than NH_4^+ leaching, perhaps the biochar was better at adsorbing anions than cations, but is still better at adsorbing cations than the regular peat moss based potting mix. It is also possible that the impact of the biochar on NO_3^- leaching was more pronounced than NH_4^+ because the concentrations of NO_3^- were higher. Because in 2010 and 2013, there

was a significant difference between N0 and N10, but not between the biochar and potting mix, and the only difference between N0 and N10 was the biochar, again suggests that the method of extraction used in 2010 and 2013 was not successfully extracting NO_3^- . There was no difference in the amount of NO_3^- extracted from any of the potting mix of any of the treatments except in 2014, Y5 and Y25 had significantly less NO_3^- extracted than Y0. Which runs contrary to the finding that the biochar had greater extractable NO_3^- than the potting mix in N10 for 2014. However the differences there were not very significant.

Because NH_4^+ nitrifies into NO_3^- it is important to analyze them together. In this experiment, the vast majority of the nitrogen found was in the form of NO_3^- . Therefore, the TIN follows the pattern of NO_3^- fairly closely. In both 2010 and 2013, the 10% biochar disk reduced TIN leaching by 41%. And in 2010, Y25 and Y30 reduced TIN leaching by 60% and 66%, respectively, compared to the control. In 2014, 15%, 25% and 30% biochar and the begonia plant leached 28%, 40% and 29%, respectively, less TIN compared to just the begonia. Because these two species can interchange, when we see reductions in NO_3^- it maybe due to NH_4^+ absorption, and vice versa. But whether it was through anion or cation adsorption, TIN was reduced in the presence of biochar in 2010, both with and without begonias, in 2013, without begonias, and in 2014, with begonias.

As with NO_3^- and NH_4^+ , PO_4^{3-} leaching was reduced in the presence of biochar, some of the time. These results are comparable to those of Hale et al. (2013) and Angst et al. (2013). In 2010 and 2014, the 10% disk treatment reduced PO_4^{3-}

leaching by 60% and 27%, respectively, compared to the control, but in 2013 there was no effect. And in 2014, 5%, 10%, 15%, 20%, 25%, and 30% biochar by volume in the treatments with begonias reduced leaching by 48%, 48%, 49%, 43%, 53% and 55%, respectively, compared to the control, but there was no effect in 2010 or 2013. The % P found in the treatments with biochar was higher than the control, which makes sense because the biochar itself had higher P content than the potting mix.

In total, the treatments with biochar saw reductions in leaching of all three tested ions in 2010, but very little effect in 2013, and in 2014, all three ions were impacted again. This seems to imply that biochar has the capacity to adsorb anions, like PO_4^{3-} and NO_3^- , and cations like NH_4^+ .

Therefore, biochar could be an important tool in reducing fertilizer run off in a nursery setting. However, the amount of biochar used has to be proportional to the amount of fertilizer used. At a certain point biochar is not able to adsorb any more fertilizer. Perhaps biochar amended potting mix could be used in conjunction with a biochar water filter, or other filtering methods.

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CHAPTER 3

The effect of biochar on the ammonia, nitrate and phosphate concentrations in soil water in a turfgrass system.

Abstract

Fertilizer run-off into surface and ground water can affect humans and other organisms that rely on those water systems for consumption and habitat. A study was conducted over two years to determine the effect of biochar and establishment methods on nitrogen and phosphorus levels in turfgrass soil water solutions. Treatments were arranged in a randomized complete block design on a sandy loam soil seeded with tall fescue (*Festuca arundinacea*). The grass was either direct seeded at the UC Riverside field site or started at a sod production farm and transplanted as sod to the field site. For each establishment method, there were three levels of biochar application: control (0 tonnes/hectare), low (6.25 tonnes/ha), and high (31.25 tonnes/hectare). Plots were fertilized with Simplot BEST Turf 15-5-8 fertilizer and irrigated four times a week. Soil water solution was extracted from the plots using suction lysimeters. Those samples were then analyzed photometrically for nitrate, ammonium and orthophosphate concentrations. The results showed that plots with plants scalped and transplanted as sod had higher concentrations of fertilizer in the soil solution compared to the plots that had been direct seeded. The high rate of biochar application decreased the concentration of all three tested ions in the first sampling season and that effect continued for an additional year for the nitrate concentration. The establishment

method did not affect biochar's ability to decrease fertilizer concentrations in the soil water solution.

Introduction

One of the main contributors to global warming is the increase in atmospheric carbon dioxide (Rodhe, 1990). Biochar is labile organic carbon, (i.e. plant or animal waste) transformed through pyrolysis into recalcitrant organic carbon (Lehmann and Joseph, 2009), which can then be used as a soil amendment. The half-life of biochar can be over 1,000 years (Spokas, 2010). By applying biochar to soil, carbon is sequestered for 100s to 10,000 of years.

Biochar could be a benefit to homeowners because it has the capacity to provide benefits like reducing water needs (Tryon, 1948; Brockhoff et al., 2010), and increasing soil nutrient retention (Glaser et al., 2002; Lehmann et al., 2003; Brockhoff et al., 2010). To reduce the burden on homeowners, turfgrass production facilities could apply biochar during sod production. Biochar would then be added to lawns when the biochar-enriched sod is installed.

Because of intensive and widespread management of turfgrass, fertilizer application to lawns has lead to ground and surface water contamination (Petrovic, 1990). Nitrogen (N) and phosphorus (P) can cause problems, like eutrophication (Sharpley et al., 1994; Caperon et al. 1972; Howarth et al., 2011) and methemoglobinemia (Majumdar, 2003), if they leach in to waterways. In order to

understand N cycling in soils, it was necessary to monitor ammonium (NH_4^+) and nitrate (NO_3^-), for P cycling, phosphates.

NH_4^+ can leave the soil solution through absorption by living organisms, adsorption by soil particles, volatilization into the atmosphere, nitrification, or leaching. Plants, fungus and bacteria all absorb NH_4^+ . Although N can be made unavailable to plants (immobilized), if microbial community consumes it before the plants can, usually when the carbon to nitrogen ratio in the soil is greater than 25 (Loomis and Conner, 1992). NH_4^+ can become strongly adhered to soil particles if the cation exchange capacity (CEC) of the soil is high or the NH_4^+ content of the soil is low. NH_4^+ could also be converted into ammonia and if it is near the surface it could volatilize out of the system (Marschner and Rengel, 2007). NH_4^+ could be leached out of the system if the concentration of NH_4^+ is high, the soil's CEC is low, or the water flow is high (Marschner and Rengel, 2007). Or the NH_4^+ could be nitrified, converting into nitrate (Marschner and Rengel, 2007).

NO_3^- can leave the system by absorption by plants, denitrification or leaching. Plants and other soil organisms can take up NO_3^- . It could be denitrified and converted into nitrogen gas or nitrous oxide, which could diffuse out of the system (Marschner and Rengel, 2007). But NO_3^- is also very mobile in soils and can be easily leached away (Marschner and Rengel, 2007).

There are three pools of P with varying degrees of availability. There is usually a large pool of P in the soil that is fixed, either as insoluble inorganics from the parent material of the soil or organic matter that is resistant to mineralization

(Stevenson and Cole, 1999). There is also a pool of active P in the soil that is dissolvable but not available to plants because it is adsorbed to a soil particle or has formed precipitates that are easily dissolved (Stevenson and Cole, 1999). The last pool is the solution P pool, ionic P dissolved in the soil solution (Loomis and Conner, 1992). When dissolved in soil water, P is usually in the form of di hydrogen phosphate (H_2PO_4^-) or hydrogen phosphate (HPO_4^{2-}), depending on soil pH. These ionic forms of P are available uptake by the plant (Marschner and Rengel, 2007); however, this plant available pool is a very small fraction of the total P in the soil (Stevenson and Cole, 1999). It is quickly depleted by the organisms in the soil and replenished by the easily dissolved active P pool (Stevenson and Cole, 1999).

P in the solution P pool is controlled by a couple of mechanisms. It could be taken up by plants, microbes or fungi, and sequestered as organic P (Frossard et al., 2000). The P could be adsorbed to soil particles, like clay lattices (Loomis and Conner, 1992). Or it could be precipitated out of solution by calcium, aluminum or iron (Loomis and Conner, 1992). If the P is in an aqueous state then leaching can play a key role. Although the amount of P is small, this is usually inconsequential (Kertesz and Frossard, 2014). But if there are large amounts of P being added to the soil leaching could still play a significant role in P loss (Stevenson and Cole, 1999).

This study investigates the effect of biochar on N and P fertilizer run off from turf grass grown from seed or transplanted as sod. This was determined by quantifying the concentration of the aforementioned ecologically important ions as collected from the soil solution by suction lysimeters over a two-year study. This

experiment will hopefully give insight into the utility of biochar application in sod for consumers.

Materials and Methods

Study Location and Experimental Setup

In the fall of 2012, an experiment to determine the effect of biochar on nutrient retention of turfgrass was initiated at the UCR Citrus Research Station. All three biochar treatments were direct seeded (SEEDED): 0 tonnes/ha biochar (CONTROL), 6.25 tonne/ha biochar (LOW), 31.25 tonne/ha biochar (HIGH). Those same three biochar treatments were established in West Coast Turf's Escondido farm (SOD). The mix of Tall fescue grass (*Festuca arundinaceae*) called 'West Coaster' was used in this . Turf grass was seeded in October 2012 (FALL) and March 2013 (SPRING) at both locations. The fall treatments were transplanted to the Research Station in February 2013 and the spring treatments were transplanted in October 2013. Therefore there were six treatments in the first year of the study: FALL-SEEDED-CONTROL, FALL-SEEDED-LOW, FALL-SEEDED-HIGH, FALL-SOD-CONTROL, FALL-SOD-LOW, and FALL-SOD-HIGH. And in the second year of the experiment there were twelve: FALL-SEEDED-CONTROL, FALL-SEEDED-LOW, FALL-SEEDED-HIGH, FALL-SOD-CONTROL, FALL-SOD-LOW, FALL-SOD-HIGH, SPRING-SEEDED-CONTROL, SPRING-SEEDED-LOW, SPRING-SEEDED-HIGH, SPRING-SOD-CONTROL, SPRING-SOD-LOW, and SPRING-SOD-HIGH.

The experiment was a randomized block design with four replications. Each plot (replicate) was 3m x 3m, and all samples were taken from the center of the plot. The biochar was lain down on the top of the plots and not incorporated so that the biochar would be scalped along with the grass when the sod producers harvested the grass. Biochar applicartion was done within a couple days of planting.

The soil at UC Riverside's Citrus Experiment Station is a Hanford coarse sandy loam and the soil at West Coast Turf's Escondido farm is Tujunga Sand. The biochar came from Blue Sky Biochar, which is pelletized biochar and wood dust. For this experiment the turf was fertilized with a 15-5-8 ammonium based fertilizer microgreen from Simplot BEST Turf. Plots were fertilized at a rate of 651.88 kg/ha. All the plots were watered to 80% ET consistently 4 times a week based on the evapotranspiration rate and mowed twice a week for the entire course of the experiment.

Lysimeter sampling and soil water analysis

Ten cm long suction lysimeters (IRROMETER SSAT) were placed in the center of each plot. Soil water solution samples were taken immediately prior to fertilization and after each irrigation event for the following two weeks, during each of four sampling periods: summer 2013, fall 2013, summer 2014 and fall 2014. However, in the summer of 2013 no samples were taken prior to fertilization and the samples were taken over the course of two months instead of two weeks. Samples were frozen and later analyzed photometrically for NO_3^- , NH_4^+ and PO_4^{3-} .

using an Astoria Pacific Autoanalyzer. The analysis methods converts all forms of phosphate into PO_4^{3-} .

Weather

Season	Week	Average Air Temperature (°C)	ET ₀ (mm)	Precipitation (mm)	Irrigation (mm)
Summer 2013	Before	20.9	6.09	0	43.18
	Week 1	27.9	7.10	0.4	51.56
	Week 2	25.2	6.48	0	51.56
Fall 2013	Before	15.6	2.68	0	24.64
	Week 1	17.1	3.27	0	18.80
	Week 2	17.7	2.37	0	18.80
Summer 2014	Before	25.4	7.24	0	49.28
	Week 1	24.2	6.13	0	49.28
	Week 2	22.1	5.78	0	49.28
Fall 2014	Before	18.0	2.42	0	18.54
	Week 1	15.9	2.68	0	23.11
	Week 2	16	2.77	3.1	23.11

Table 3.1: “Before” refers to the week immediately prior to fertilization of the plots, while “Week 1” and “Week 2” refer to the first and second weeks of the experiment, respectively. ET₀ stands for potential evapotranspiration. Temperature and ET₀ are averaged over the week while precipitation and irrigation are cumulative. These values are given for the two weeks of the experimental time and the week immediately before the plots were fertilized.

Wind speeds averaged 1-2 m/s although each fall had one incidence of wind speeds as high as 4 m/s. Soil temperature during the summer ranged from 22 °C to 24 °C, and 12 °C to 17.5 °C in the fall. Average air temperature during the summer was 19 °C to 30 °C, and during the fall, 13 °C to 23 °C. Evapotranspiration was 4-8 mm during the summer and 1-5 mm during the fall. There were only two rain events while measurements were being taken, 0.4 cm of rain on July 1, 2013, and 3.1 cm of rain on November 21, 2014.

Statistical analysis

In order to understand the movement of ionic N in the system, NO_3^- and NH_4^+ were summed to make total inorganic nitrogen (TIN). This calculation assumes that amounts of nitrite in the soil would be very small. Statistical analysis was done with a mixed model with a temporarily correlated covariance structure to analyze the treatment effect and temporal trend on SAS ($\alpha = 0.05$). The NH_4^+ and TIN results were transformed by the - 0.2 power. The NO_3^- results were transformed by the -0.3 power. The PO_4^{3-} results were transformed by a logarithm. All transformations were to improve normality of the data.

Results and Discussion

Ammonium

Effect	Summer 2013	Fall 2013	Summer 2014	Fall 2014
Planting Date	n.a.	n.a.	0.0198	n.s.
Establishment Method	n.s.	n.s.	n.s.	n.s.
Biochar Amount	n.s.	0.0708	n.s.	n.s.
Sampling Date	<0.0001	<0.0001	<0.0001	<0.0001
Planting Date * Biochar Amount	n.a.	n.a.	n.s.	0.0688
Establishment Method * Biochar Amount	0.0045	n.s.	n.s.	0.0129
Establishment Method * Sampling Date	n.s.	n.s.	n.s.	0.0409
Biochar Amount * Sampling Date	n.s.	0.0488	n.s.	n.s.

Table 3.2: Probability statistics of interaction variables for mixed model analysis. Values given are the probabilities that the ammonium concentrations would be similar for the variables described. The following variables were removed from the table because they were never significant: Planting Date * Establishment Method, Planting Date * Sampling Date. "n.s." stands for "not significant," at $p = 0.05$; "n.a." stands for "not applicable" as 2013 samples were only taken from the fall planting date onward.

General Observations

The fertilizer used is NH_4^+ based; therefore an immediate spike in soil water NH_4^+ concentration was expected and observed in each of the first three seasons. However, in the fall on 2014, there does not appear to be any spike in NH_4^+ (Figure 3.1d), it was possible that the NH_4^+ was immobilized by the microbial community rapidly enough that it was not picked up by the lysimeters. Since grass trimmings had been building up for more than two years, it was possible that a higher carbon

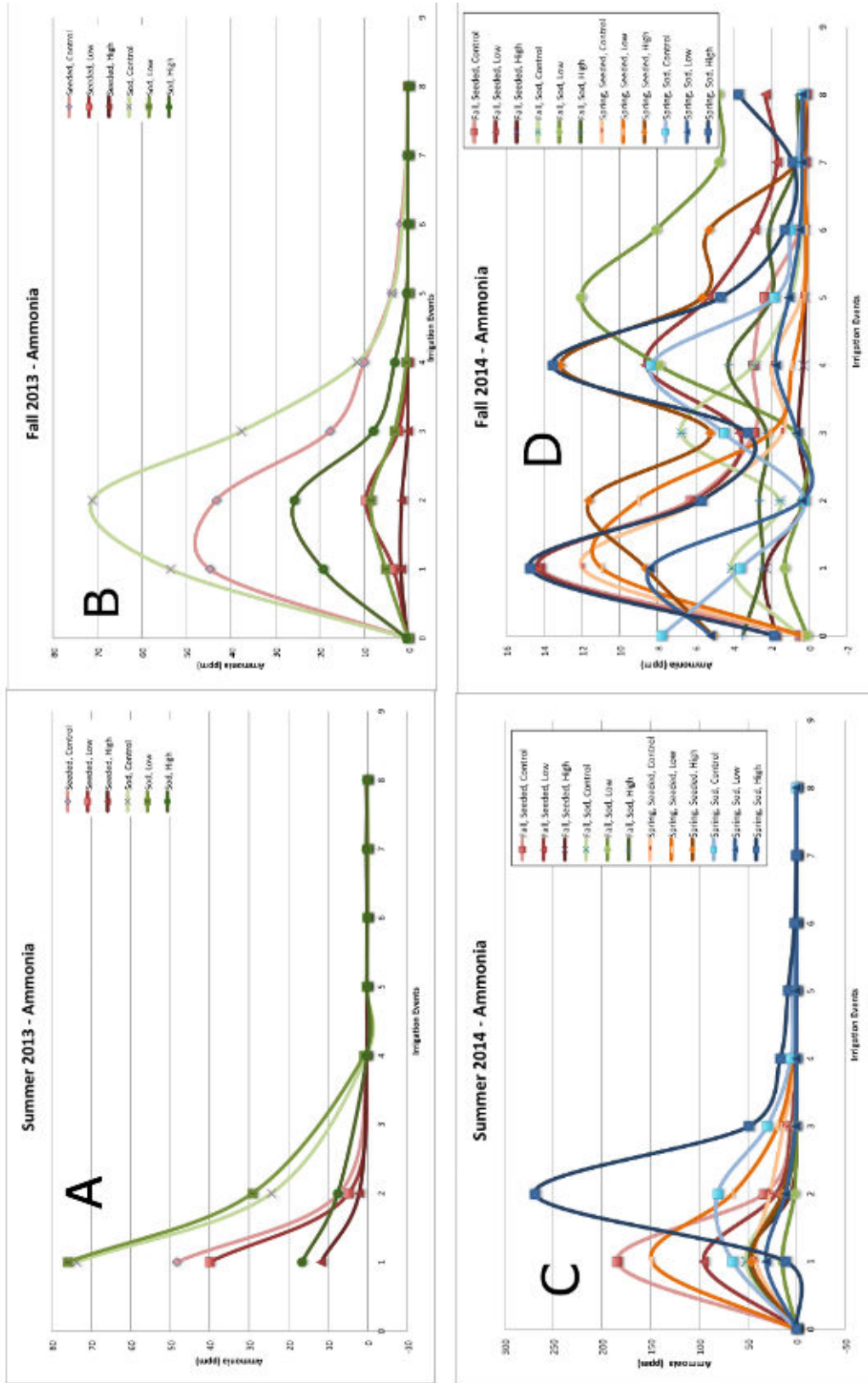


Figure 3.1. The amount of ammonia in the soil solution in ppm for each season of data. "Fall" and "Spring" refer to the time of planting, in figures 3.1a and 3.1b all the plants are from the fall planting. "Seeded" and "Sod" refer to the establishment method, either direct seeding or transplanting as sod. "Control," "Low," and "High" refer to the amount of biochar applied to the treatment. The control had 0 tonnes/ha, the low had 6.25 tonnes/ha and the high rate had 31.25 tonnes/ha.

to nitrogen ratio in the soil resulted in the majority of the applied N to be incorporated into the microbial biomass.

In the summer of 2013, no baseline measurement was taken before the plots were fertilized; the first collection was taken after the first irrigation event. The concentration of NH_4^+ at the first irrigation was the peak NH_4^+ level in the soil water. The concentrations of the NH_4^+ in the HIGH plots (10-20 ppm) were roughly a fourth of the CONTROL (45-75 ppm) and LOW (40-75; Figure 3.1a). And the SEEDED plots had lower concentrations of NH_4^+ compared to the SOD plots (Figure 3.1a). After the first irrigation concentrations of NH_4^+ dropped significantly, by the fourth irrigation event soil water NH_4^+ concentrations were consistently near zero (Figure 3.1a).

Pre-fertilization baseline samples were taken in fall 2013, summer 2014, and fall 2014. The baseline level of NH_4^+ across treatments in the fall of 2013 was less than 1 ppm, and reached a peak after the second irrigation. Biochar reduced soil NH_4^+ concentrations; CONTROL plots maxed at 45 to 70 ppm while the HIGH and LOW treatments ranged from 1-25 ppm. While the SOD plots generally had higher NH_4^+ concentrations than the SEEDED plots (Figure 3.1b). The levels of NH_4^+ drop to near zero levels by the sixth irrigation.

Summer 2014 baseline NH_4^+ levels were also low prior to fertilization (Figure 3.1c). For SPRING-SOD planting most treatments reached peak NH_4^+ levels in the first irrigation event, with some treatments maxing at 100s of ppm of NH_4^+ ,

significantly higher than the previous year (Figure 3.1c). The NH_4^+ levels in the soil decreased to near zero by the sixth irrigation again in this summer.

In the fall of 2014, soil water NH_4^+ concentrations remained fairly low through out the entire measured period. The FALL-SOD-LOW, FALL-SOD-HIGH, and FALL-SEEDED-HIGH were consistently very low, with no peaks (Figure 3.1d). The SEEDED-CONTROL for both plantings as well as the SPRING-LOW all peaked once at the first irrigation (Figure 3.1d). All the other rates had two peaks, once at the first irrigation and the other at the fourth or fifth irrigation (Figure 3.1d).

Biochar

In the first season, the LOW rate had significantly lower NH_4^+ levels compared to the CONTROL rate for SEEDED plots (Figure 3.1a). In the fall 2013, the CONTROL had higher concentrations compared to both levels of biochar treatment in the third and fourth irrigation events, when the concentration of the NH_4^+ in the soil was still fairly high (Figure 3.1b). In the second year there was no effect of the biochar treatments in the summer (Figure 3.1c). But in the next fall, the HIGH rate had significantly higher concentrations of NH_4^+ compared to the LOW and CONTROL rates for SPRING plots (Figure 3.1d). At the beginning of the experiment, in the first two seasons, the biochar treatments had some effect on reducing the concentration of NH_4^+ in the soil solution. In the second year, there was no effect in the summer, but in the fall it had the opposite effect, but the values are all very low compared to what was expected.

Establishment Method

In the first season (Summer 2013; Figure 3.1a), the concentration of NH_4^+ in the soil solution of the SEEDED plots was significantly less than that of the SOD plots for the LOW treatments. But in fall 2013 and summer 2014 there was no effect due to the planting style. In the fall of 2014, the SEEDED plots had a greater concentration of NH_4^+ compared to the SOD for the first irrigation, but this was due an outlier. There does not seem to be too much effect of the planting style on the NH_4^+ adsorption by the plant-soil system after the first sampling season. Potentially NH_4^+ was not as affected by the establishment method as well as NO_3^- was (see below) because NO_3^- is the more mobile ion.

Nitrate

General Observations

Without the addition of NO_3^- -N to the system, the main source of the NO_3^- in the soil solution of this system should be from the nitrification of the NH_4^+ fertilizer.

Therefore, there should be a spike of NO_3^- at about the time the NH_4^+ concentration starts to decrease. This was seen in all four tested seasons. Interestingly, while the concentration of NH_4^+ was much lower than previous seasons, the concentration of NO_3^- in the fall of 2014 was much greater (Figure 3.2d). If the NH_4^+ applied as fertilizer was immobilized as theorized in the previous section, it was possible that N was mineralized into nitrate as the bacteria died off.

Effect	Summer 2013	Fall 2013	Summer 2014	Fall 2014
Planting Date	n.a.	n.a.	0.0405	0.0405
Establishment Method	<0.0001	n.s.	0.0427	0.0427
Biochar Amount	0.0013	n.s.	n.s.	n.s.
Sampling Date	<0.0001	0.0207	<0.0001	<0.0001
Planting Date * Biochar Amount	n.a.	n.a.	0.0237	0.0237
Establishment Method * Biochar Amount	0.0001	n.s.	n.s.	n.s.
Establishment Method * Sampling Date	n.s.	n.s.	n.s.	n.s.
Biochar Amount * Sampling Date	n.s.	n.s.	n.s.	n.s.

Table 3.3: Probability statistics of interaction variables for mixed model analysis. Values given are the probabilities that the nitrate concentrations would be similar for the variables described. The following variables were removed from the table because they were never significant: Planting Date * Establishment Method, Planting Date * Sampling Date. "n.s." stands for "not significant," at $p = 0.05$; "n.a." stands for "not applicable" as 2013 samples were only taken from the fall planting date onward.

In summer 2013, NO_3^- levels remain fairly low through out the experiment, peaking in the first irrigation (21 ppm) for the SOD-LOW rate or between the second and fourth irrigations for the other treatments. In general, the HIGH rate resulted in a lower concentration of NO_3^- . And the SEEDED plots had less NO_3^- than the SOD ones (Figure 3.2a).

The fall 2013 pre-fertilization baseline for NO_3^- was roughly one ppm. The NO_3^- in the soil water peaked during the fourth to sixth irrigations. As with NH_4^+ , peak NO_3^- levels were less in the plots with biochar compared to those without. And the SOD plots had consistently higher levels of NO_3^- compared to the SEEDED plots (Figure 3.2b).

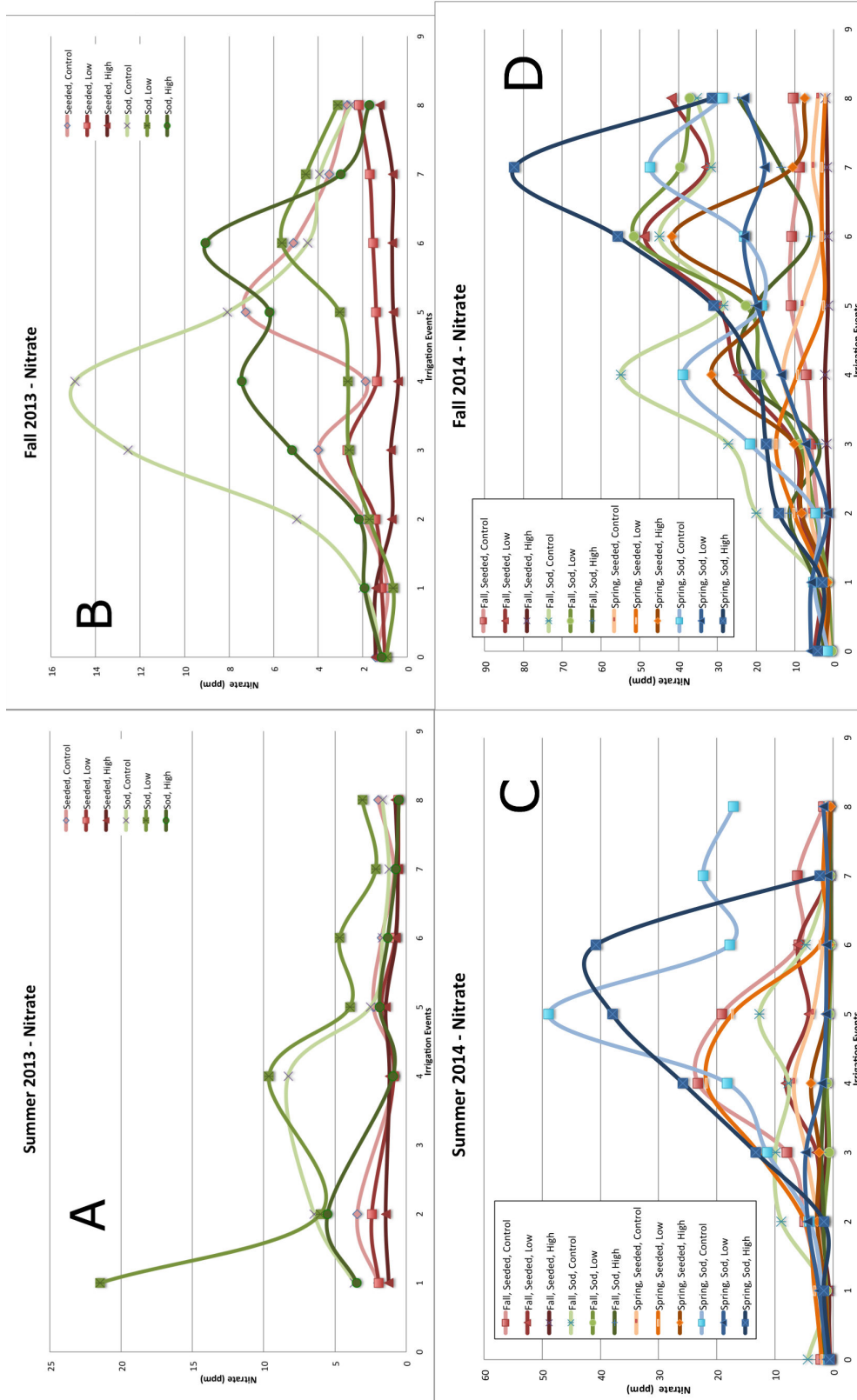


Figure 3.2. The amount of nitrate in the soil solution in ppm for each season of data. "Fall" and "Spring" refer to the time of planting, in figures 3.1a and 3.1b all the plants are from the fall planting. "Seeded" and "Sod" refer to the establishment method, either direct seeding or transplanting as sod. "Control," "Low," and "High" refer to the amount of biochar applied to the treatment. The control had 0 tonnes/ha, the low had 6.25 tonnes/ha and the high rate had 31.25 tonnes/ha.

The following summer, NO_3^- levels remained low from the baseline measurement to the second irrigation. Again the NO_3^- levels peaked in the fourth to sixth irrigation. The SPRING-SOD-CONTROL and SPRING-SOD-HIGH had peaks of 40 to 50 ppm, which was more than three times higher than the previous year. FALL-SEEDED-CONTROL and SPRING-SOD-LOW peaked at about 22 ppm, roughly twice peak levels of the previous year. Trends for the other treatments were more similar to the previous fall - in general, the CONTROL biochar rates had greater concentrations of NO_3^- but SPRING-SEEDED-LOW and SPRING-SOD-HIGH were also very high in NO_3^- (Figure 3.2c).

In the fall of 2014, NO_3^- concentrations began very low but started to increase by the second irrigation. The concentration of NO_3^- peaked between the third and seventh irrigation events. The peak NO_3^- concentration was again higher in this instance than it had been in any previous sampling time period. SPRING-SOD-HIGH topped out at over 80 ppm, roughly twice the measured peak of the previous summer. FALL-SEEDED-LOW, FALL-SOD-LOW, FALL-SOD-CONTROL, SPRING-SEEDED-HIGH, and SPRING-SOD-CONTROL all peaked at 40-55 ppms, similar to the highest peaks of the previous summer. The rest of the treatments resembled the previous fall's data, peaking at less than 25 ppm. This overall increase in NO_3^- (Figure 3.2d) concentrations correlates to an overall decrease in NH_4^+ levels (Figure 3.1d)

Biochar

In the summer of 2013, plots that had been SEEDED and incorporated with biochar had lower NO_3^- concentrations than the SEEDED-CONTROL; but SOD-LOW had significantly higher soil water NO_3^- concentration than the HIGH or CONTROL (Figure 3.2a). In the next season there was no effect (Figure 3.2b). But in the 2014, plants that had been planted in the FALL showed significantly more sorption of the NO_3^- for the HIGH rate of biochar compared to the CONTROL rate (Figure 3.2c and Figure 3.2d).

Fresh biochar often has a relatively high AEC (Lawrinenko, 2014), which may explain why the plots with biochar applied to them sorbed more NO_3^- in the first season. The aging process for biochar lowers the AEC and increases the CEC (Hale et al., 2011). Perhaps the biochar adsorbed more NH_4^+ , due to its relatively high CEC which lead to less NO_3^- from nitrification and therefore the lower values of NO_3^- observed in the fall planted plots in 2014 which were 22 months old in the summer and 26 in the fall.

Establishment Method

In the first summer, the concentration of NO_3^- in the soil solution of the biochar treatments was higher for SOD treatments compared to SEEDED (Figure 3.2a). There was no effect in the fall of the first year. However, across both the fall and summer of the second year, the concentration of NO_3^- was consistently higher in the SOD treatments compared to the SEEDED ones (Figures 3.2c and 3.2d). The damage done to the grass roots during the scalping process would have impaired

the ability of the plants to absorb nutrients, accounting for the differences seen here. This agrees with Geron et al. (1993)'s study on NO₃⁻ leaching from seeded and sodded turf grass.

Total Inorganic Nitrogen

Effect	Summer 2013	Fall 2013	Summer 2014	Fall 2014
Planting Date	n.a.	n.a.	0.0607	0.0607
Establishment Method	<0.0001	n.s.	n.s.	n.s.
Biochar Amount	0.0121	n.s.	n.s.	n.s.
Sampling Date	<0.0001	<0.0001	<0.0001	<0.0001
Planting Date * Biochar Amount	n.a.	n.a.	0.0366	0.0366
Establishment Method * Biochar Amount	0.0046	n.s.	n.s.	n.s.
Establishment Method * Sampling Date	n.s.	n.s.	0.0112	0.0112
Biochar Amount * Sampling Date	n.s.	n.s.	n.s.	n.s.

Table 3.4: Probability statistics of interaction variables for mixed model analysis. Values given are the probabilities that the total inorganic nitrogen concentrations would be similar for the variables described. The following variables were removed from the table because they were never significant: Planting Date * Establishment Method, Planting Date * Sampling Date. "n.s." at p = 0.05; "n.a." stands for "not applicable" as 2013 samples were only taken from the fall planting date onward.

General Observations

Because TIN was calculated as the combination of the NO₃⁻ and NH₄⁺ concentrations, and NH₄⁺ concentrations in 2013 and the summer of 2014 were significantly higher than NO₃⁻ concentrations, the TIN concentration was fairly similar to the NH₄⁺ concentration. There was a significant peak just subsequent to

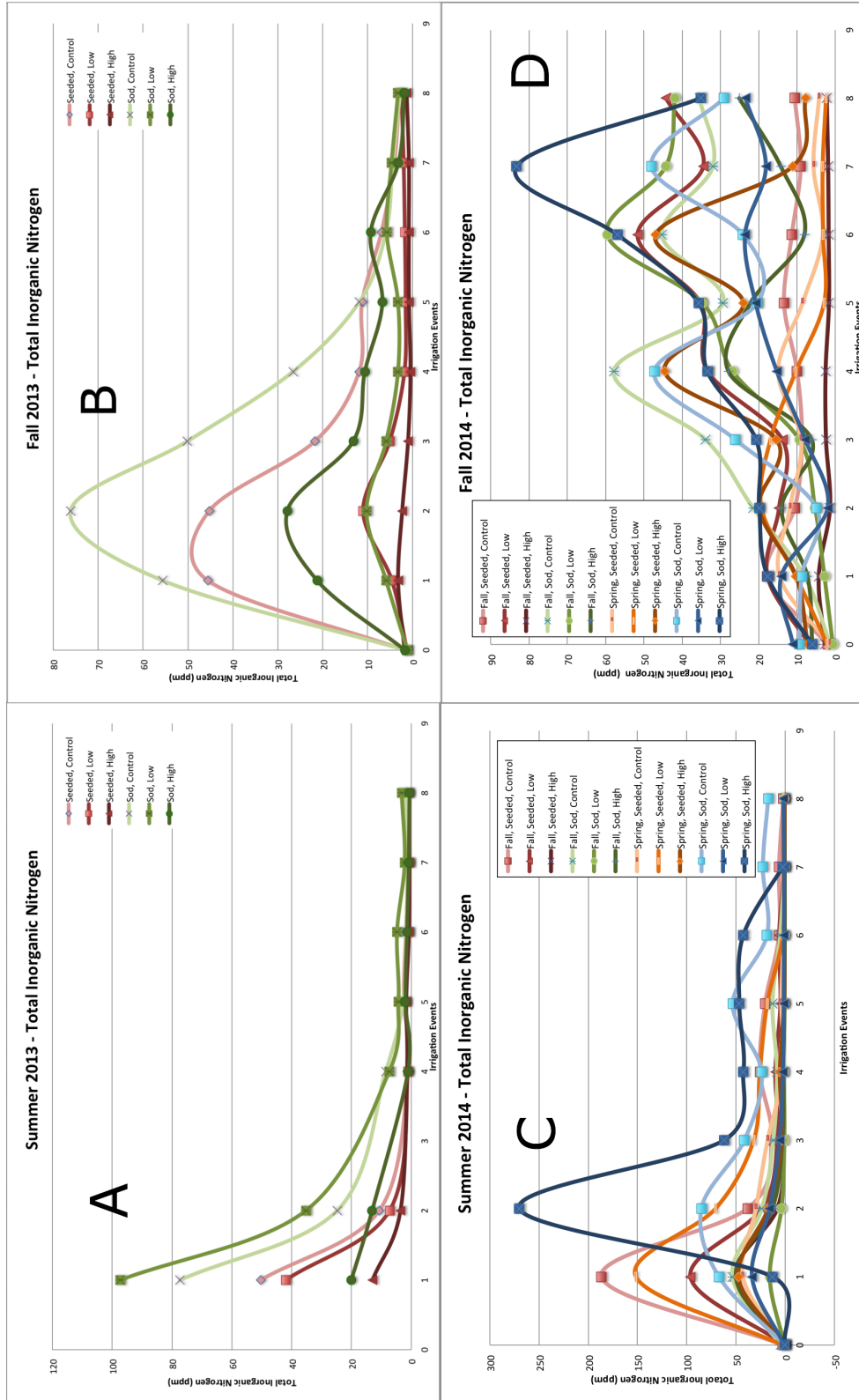


Figure 3.3. The amount of total inorganic nitrogen in the soil solution in ppm for each season of data. "Fall" and "Spring" refer to the time of planting, in figures 3.1a and 3.1b all the plants are from the fall planting. "Seeded" and "Sod" refer to the establishment method, either direct seeding or transplanting as sod. "Control," "Low," and "High" refer to the amount of biochar applied to the treatment. The control had 0 tonnes/ha, the low had 6.25 tonnes/ha and the high rate had 31.25 tonnes/ha.

fertilization. But because the NO_3^- peaks later, there was slightly slower taper to the curve as the nitrogen was absorbed by the plants, volatilized, leached, or denitrified. However, in the fall of 2014, the NO_3^- level was higher than NH_4^+ ; therefore the TIN follows the NO_3^- concentration more closely, generally peaking in the fourth to sixth irrigation (Figure 3.3d).

Biochar

In the summer of 2013, SOD plots had greater concentrations of N in the LOW rate compared to the CONTROL and HIGH (Figure 3.3a). There was no effect in the following season. But in the summer and fall of 2014, the CONTROL rate had a significantly greater concentration of N compared to the HIGH rate.

Establishment Method

In the summer of 2013, the transplanting effect was apparent in the TIN data, the treatments with biochar had greater concentrations of N in the soil solution if the plants had been SOD compared to the SEEDED plots (Figure 3.3a). But this effect was not apparent in any other season.

Phosphate

General Observations

P never had the high peaks observed with NO_3^- or NH_4^+ . P can easily precipitate out of solution or strongly adsorb to soil particles, and was thus less likely to have a large increase in soil water concentration following application. The

Effect	Summer 2013	Fall 2013	Summer 2014	Fall 2014
Planting Date	n.a.	n.a.	n.s.	0.0028
Establishment Method	<0.0001	0.0427	n.s.	0.0153
Biochar Amount	n.s.	0.0571	n.s.	n.s.
Sampling Date	<0.0001	<0.0001	<0.0001	<0.0001
Planting Date * Biochar Amount	n.a.	n.a.	0.0014	n.s.
Establishment Method * Biochar Amount	n.s.	n.s.	n.s.	n.s.
Establishment Method * Sampling Date	0.0014	n.s.	0.0182	n.s.
Biochar Amount * Sampling Date	n.s.	n.s.	n.s.	n.s.

Table 3.5: Probability statistics of interaction variables for mixed model analysis. Values given are the probabilities that the orthophosphate concentrations would be similar for the variables described. The following variables were removed from the table because they were never significant: Planting Date * Establishment Method, Planting Date * Sampling Date. "n.s." stands for "not significant," at $p = 0.05$; "n.a." stands for "not applicable" as 2013 samples were only taken from the fall planting date onward.

P peak for summer 2013 was 1 to 2.5 ppm and occurred during the first and second irrigations. The high rate of biochar treatments peak was about half the PO_4^{3-} peak of the CONTROL or LOW treatments. SOD plots had higher levels of soil water PO_4^{3-} compared to the SEEDED treatments.

In fall 2013, the SOD-CONTROL plots had much higher concentrations of PO_4^{3-} than the other treatments, peaking around 3 ppm. The other treatments never exceeded 1.5 ppm. The control plots had greater concentrations of PO_4^{3-} compared to the biochar treatments, and in general, the SOD plots had greater concentrations of PO_4^{3-} than the SEEDED plots.

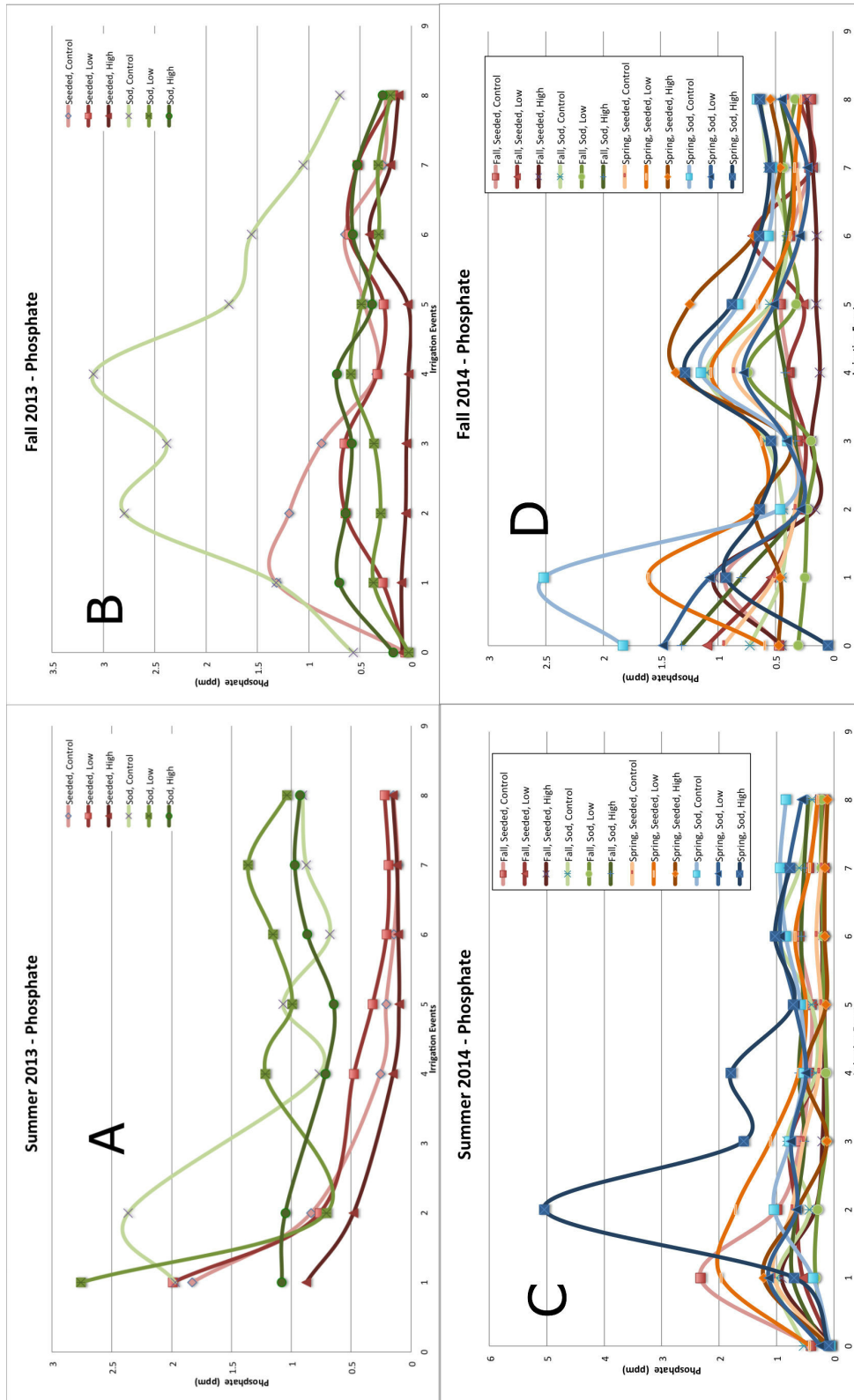


Figure 3.4. The amount of ortho-phosphate in the soil solution in ppm for each season of data. "Fall" and "Spring" refer to the time of planting, in figures 3.1a and 3.1b all the plants are from the fall planting. "Seeded" and "Sod" refer to the establishment method, either direct seeding or transplanting as sod. "Control," "Low," and "High" refer to the amount of biochar applied to the treatment. The control had 0 tonnes/ha, the low had 6.25 tonnes/ha and the high rate had 31.25 tonnes/ha.

In the summer of 2014, the PO_4^{3-} concentrations behaved very similarly to previous seasons. The SPRING-SOD-HIGH peaked at 5 ppm, but all other treatments peaked at less than 3 ppm at either the first or second irrigation. In this season, there were not noticeable differences between the different treatments.

In the last season, fall 2014, PO_4^{3-} peaked in the first irrigation, as it had for the previous seasons. The SPRING-SOD plots had the highest values for PO_4^{3-} concentration with the CONTROL plot peaking at 2.5 ppm. As in the previous three seasons, PO_4^{3-} levels returned to less than 1 ppm by the end of the measurement period.

Biochar

No effect of the biochar on soil water PO_4^{3-} concentration was observed in the summer of 2013 or the fall of 2014, the first and last seasons. But in the fall of 2013, the HIGH treatments had a significantly lower concentration of PO_4^{3-} compared to the CONTROL (Figure 3.4b; $P=0.0571$). In the summer of 2014, the LOW rate had significantly lower concentrations of PO_4^{3-} compared to the CONTROL and HIGH rates for plots planted in the FALL but higher for SPRING plots. This may indicate that there was no reliable effect of biochar on PO_4^{3-} adsorption in this system.

Establishment Method

The damage done to the roots when transplanting the sod, was apparent in summer and fall of 2013 and the fall of 2014. In the first season of sampling, the SOD treatments had greater concentrations of PO_4^{3-} in the soil compared to the

SEEDDED plots after the fourth irrigation event (Figure 3.4a). But in the fall of both years, the SOD treatments had greater concentrations of PO_4^{3-} compared to the SEEDDED plots (Figures 3.4b and 3.4d). Only in the summer of 2014 did the trend shift. In this season, the comparison was not significant except for on the first irrigation event where the concentration in the seeded plots was actually higher (Figure 3.4c).

Conclusion

Immediately after the fertilizer was applied, NH_4^+ and PO_4^{3-} spiked. Because the NH_4^+ in the soil solution was nitrified, the NO_3^- concentrations peak a few days later. As the nitrogen is taken up by the grass, denitrified, volatilized or leached out of the system and the PO_4^{3-} was absorbed by the roots, leached out or precipitates out, the concentrations of these ions gradually taper out (Stevenson and Cole, 1999). This pattern was seen in almost every season and treatment. The exception was the last season, fall 2014, where the concentration of NH_4^+ never got as high as previous seasons and the NO_3^- concentration gradually increased to much higher concentrations than previous years. By the fall of 2014, the plots had been set up for about two years, long enough to build up a significant thatch layer. The increased carbon to nitrogen ratio could explain why the NH_4^+ was not collected by the lysimeters initially. Once the NH_4^+ converted into organic matter by the microbial community it could later be mineralized into NO_3^- , explaining the later peak in NO_3^- (Stevenson and Cole, 1999).

Turf grass is often sold as sod. Sod producers grow the grass and then scalp the grass to sell it. Only a relatively small amount of the grasses roots are included with the above ground matter. This means that the transplanted sod will likely have less ability to absorb nutrients due to their limited amount of roots. The experiment was carried out at least six months after the sod was transplanted into the field. Despite that the effects were still apparent. In many cases the plots that had been transplanted had greater concentrations of nutrients in the soil solution indicating that those plants adsorbed less than the plants that had been direct seeded. This effect was apparent even into the last season (Figure 3.2d). Interestingly, the concentration of NH_4^+ was only affected by the production method in the first season, while the NO_3^- concentration was influenced in most seasons. The PO_4^{3-} was also significantly effected in some seasons, although not as consistently as the NO_3^- . This agrees with a study from Ohio that found that once the seeded plots were established, they leached less NO_3^- than sodded plots (Geron et al., 1993).

Biochar has been shown in other studies to adsorb NO_3^- , NH_4^+ and PO_4^{3-} (Angst et al., 2013; Asada et al., 2002; Chintala et al., 2013; Ding et al., 2010; Hale et al., 2013; Lehmann et al., 2003; Steiner et al., 2010). Therefore, theoretically, biochar would adsorb them in field conditions, like those tested in this experiment, removing them from the soil solution. The observations show that in some cases biochar did seem to lower the concentration of the nutrients in the soil solution especially at the times when the concentrations were high. But there were also several cases where the treatments did not behave as expected. Especially with the

low treatment having either significantly greater concentration compared to the other treatments or, in one case, significantly lower, like with the PO_4^{3-} concentration in summer 2014. Because of problems with acquiring the sod for the experiment and the difficulty removing the water samples, it was possible that there was not enough replication to prevent type I errors.

Based on the results from this study, it seems that if fertilizer run-off or leaching is a concern, establishing turf from sod is unwise. The damage done to the roots is still noticeable after about two years. The addition of biochar can help to mitigate some leaching from the root zone but not substantially.

Biochar can make a statistically significant improvement to both N and P fertilizer loss. But the quantity of P that is available to leach or run off from this system is so low that it's probably not going to be a concern. If larger quantities of P were being moved through the system, perhaps the biochar would make a greater difference. But biochar could reduce the NO_3^- or NH_4^+ -N peaks by more than 50%, if applied in high enough quantities.

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GENERAL DISCUSSION AND CONCLUSIONS

The goal of this research was to investigate biochar's influence on nutrient leaching and plant response to biochar in multiple systems. Chapter 1 investigated how biochar influences root growth in wheat plants. Chapter 2 looked at nitrogen and phosphorus leaching in a nursery plant system. Chapter 3 considered nitrogen and phosphorus leaching from a lawn or turf-grass setting. Together these chapters give an idea of how biochar may benefit growers in a variety of different settings.

Results from chapter 1 showed that biochar increased root biomass in water stressed conditions. And that the root mass was increased even in areas that were not amended with biochar, either because of changes in hydraulics of the soil, overall improved growth of the plant, or as systemic response. But that did not necessarily translate into shoot growth and never lead to increases in grain yield. Similarly, Bruun et al. (2014) showed that rooting in barley increased when similar rates of biochar (1% by weight) were applied in their sandy soils, although they found a corresponding increase in yield. Bruun et al. (2014) attributed this increase in growth and yield to increased plant available water and reduced soil bulk density. Unexpected decreases in plant growth in both the roots and shoots were found when biochar was combined with the Arlington sandy loam without any sand. Spokas et al. (2011) found more than 140 volatile organic carbon compounds associated with biochars. Some studies have found that biochar can contain toxic poly aromatic hydrocarbons (Hajaligol et al., 2001). Although biochar can greatly improve growth, end users of this product need to be aware of potential problems.

Biochar effects were not the only variable tested in chapter 1. Wheat genotype also had an effect on growth. As demonstrated in several other studies (Ehdaie et al., 2012; Maheepala et al., 2014), the Pavon 1RS.1AL translocation line had significantly greater root growth and mostly in the roots that were longer than 30 cm. However this increase in root growth did not dependably translate into increases in shoot growth or grain yield. There was no consistent interaction between isogenic line, biochar amount and soil type.

Amending the soil with sand reduced water holding, increased drainage, and reduced bulk density. When water availability was held constant, plant growth was best in the sand amended soils. Presumably, the reduced bulk density allowed for greater root penetration and therefore access to resources. When biochar was added, there was more overall growth in the unamended soil. Implying that biochar can provide a similar benefit as the sand does.

In chapter 2, fertilizer solution was added to potting mix or a layer of potting mix and a layer of biochar. The biochar layer reduced nitrate, ammonium and ortho-phosphate leaching by as much as 73%. Reduction in cation ammonium concentration in the leachate was likely due to biochar's high cation exchange capacity and the resulting electrostatic interactions (Xu et al., 2012; Mukherjee et al., 2011). Nitrate, an anion, responded in a similar way. Chintala et al. (2013) and Kameyama et al. (2011) showed that biochar has both a cation exchange capacity and an anion exchange capacity. Phosphate adsorption is regulated by ligand

exchange, Yao et al. (2011) suggested that biochar's colloidal and nano-sized periclast particles were capable of adsorbing the phosphate.

Even with the introduction of a *Begonia sempervirens* plant into the system, the highest rate of biochar was able to reduce leaching of nitrogen and phosphorus. In this system, biochar had no effect on plant growth or the nitrogen or phosphorus content of the plants. This is not unexpected; potting mix is very different from field soil. Potting mixes are designed to be ideal conditions in which to grow plants. As expected, the addition of a plant reduced fertilizer leaching. The addition of biochar and a plant did not compound this; the leaching was not reduced substantially more when a plant was added to a treatment with biochar. This is not surprising; biochar had no effect on plant growth.

Chapter 3 investigated an even more complicated system; turf grass grown under field conditions. Ammonium and phosphate concentrations in the soil water solution spiked immediately after fertilization and the nitrate concentration peaked a few days later when the ammonium had nitrified. Except in the last season, where the nitrogen concentration did not peak immediately, but increased greatly over the two-week period. I suspect that the thatch layer that had built up over the two-year period since planting shifted the carbon to nitrogen ratio enough to cause an immobilization of nitrogen in the soil that was subsequently released over time.

In this system as well, results show some decreases in nitrate, ammonium and phosphate leaching. The peak concentrations were decreased as much as fifty percent, although this effect was most noticeable in the first season, and not

consistent over time. However, leaching was more consistently reduced in the plots that had been started as seed compared to those transplanted as sod. Geron et al. (1993) found that once direct seeded plots are established they reduce nitrate leaching compared to sod plots. The damage done to the roots at the time of scalping impaired the ability of the grass sod to adsorb nutrients compared to plants that had not been cut for sod, and this effect lasted for over two years.

Biochar is simple to produce; it is possible that growers could produce it on their own property from plant wastes (Lehmann et al., 2006). It has the capacity to make a substantial impact in carbon sequestration (Lehmann, 2007). And biochar could be a very effective tool in the hands of growers. In this research, I showed that biochar can improve soil quality, increasing plant growth. And that biochar is an effective way of reducing nitrate, ammonium and phosphate leaching, effective both in the greenhouse and in the field. However, the rates of biochar used were not sufficient to negate leaching of these ions. Biochar should be used in conjunction with other practices to prevent outflow of fertilizer especially after fertigation events. Growers need to be careful about the circumstances in which they employ biochar. Potential toxic substances could be generated in the manufacture of biochar (Hajaligol et al., 2001). None the less, biochar has been shown in this research and other research to be a useful tool for plant production.

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