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Biochar and high-carbon wood ash effects on soil and vegetation in a boreal clearcut

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Abstract Additions of fire residues in the form of charcoal and wood ash may better emulate natural disturbance processes in managed boreal forests. We examined the effects of a poplar wood biochar and a high-carbon wood ash on soil and vegetation in a 3-year experiment in NW Ontario, Canada. Both soil amendments increased soil pH and soil Ca levels; high-carbon wood ash also increased soil Cu, Zn, B, S, and Pb. Amendments had large effects on plant community composition, favoring a subset of ruderal species including raspberry (*Rubus ideaus*) and goldenrod (*Solidago canadensis*). Addition of high-carbon wood ash resulted in declines in growth of planted white spruce (*Picea glauca*); a path analysis suggests this was due to effects of toxic elements rather than indirect effects of competition. We conclude that high-carbon wood ash, while qualifying as a type of biochar and having some beneficial effects on soil properties, can enhance toxic metals in boreal forest soils, with negative consequences to early tree growth. Differences in plant species responses to biochars, and potential for toxicity effects and indirect effects mediated by plant competition, will require screening and field trials of potential biochars prior to their use in operational forestry and forest restoration.

Key words: charcoal, heavy metals, natural disturbance emulation, vegetation, competition

Introduction

Wildfire is the primary disturbance factor in the boreal forest and a main determinant of ecosystem structure and function. The absence of wildfire due to human-induced fire suppression results in boreal ecosystems with altered successional patterns (Zackrisson, Nilsson, and Wardle 1996), decreased productivity (Wardle, Zackrisson, and Nilsson 1998), and shifts of species dominance (Mallik 2003). Much attention has been paid to the relative importance of fire in maintaining boreal carbon balance and ecosystem function (Hart and Luckai 2013); however, the contribution of the charcoal and ash residues that remain following a fire have only recently been identified as potentially meaningful contributors to these processes (Thomas 2013; Pluchon et al. 2014).

Natural disturbance emulation (NDE) is a framework for silvicultural management that seeks to emulate the spatial and temporal effects of natural disturbances on the landscape. In this framework, harvest impacts on the ecosystem are limited and forest regeneration is enhanced through the incorporation of management strategies to which the ecosystem is naturally adapted (Sibley et al. 2012). NDE has been embraced as a guiding philosophy of Sustainable Forest Management (SFM) in Canada, whereby large-scale disturbances such as wildfire provide the rationalization for clear-cut harvesting in the boreal forest region (Kuuluvainen and Grenfell 2012). Despite this commitment to SFM, in 2013 only 64% of Ontario’s 131,000 ha of harvested forests, 80% of which is boreal, were regenerated to the planned forest type and considered a 'silvicultural success' (Ontario Ministry of Natural Resources and Forestry 2018a). Large-scale regeneration failure is not limited to Ontario, and has been identified in the literature as a pervasive issue across managed boreal ecosystems throughout Canada (Cyr et al. 2009) and worldwide (Nordlander et al. 2011).

While boreal forest regeneration dynamics following natural disturbances are relatively well studied (Greene et al. 1999), the effects of harvesting on forest regeneration are site-specific and complex, limiting our ability to effectively predict and mitigate potential impacts on landscapes managed through coarsely defined NDE protocols (Gartner, Lifers, and Macdonald 2011). Causes of regeneration failure in the literature have been attributed to a number of factors including substrate quality and soil nutrient availability (Kokkonen et al. 2018), pests (Nordlander et al. 2011), and competing vegetation (Mallik 2003). The unpredictable and often synergistic effects of these factors may be exacerbated by the increasing threat of climate warming on boreal ecosystems (Fisichelli, French, and Reich 2013), particularly as invasive species experience range expansions as a result of their ability to withstand warmer northern winters (Kent, Dresser, and Bello 2018).

Boreal forest regeneration in Ontario is predominantly achieved through a process of site preparation followed by planting, with one-third of planted stands receiving herbicide application to manage competing vegetation (Thompson and Pitt 2011). Due to the absence of seed-bank-destroying fire in clear-cut sites, fire-adapted boreal species can lose their competitive advantage over understory vegetation, resulting in the development of a dense understory and permanently stunted succession of shade-intolerant boreal tree species (Zoladeski and Haycock 1990). Ericaceous shrub species in particular have been documented as significant inhibitors of conifer regeneration in a number of studies from across North America and Europe due to early competitive advantage resulting from effects on nutrient balance, allelopathy, and enhanced soil acidity (Mallik 2003). The use of herbicides in silvicultural management is controversial due to conflicting reports on the long-term potential threats to human (Romano et al. 2010) and ecosystem health (Lautenschlager and Sullivan 2002) associated with their persistence in the environment, and there is a need for alternative approaches to vegetation management in regenerating forest stands (Helander, Saloniemi, and Saikkonen 2012).

Charcoal produced from wood is highly recalcitrant and can enhance retention of nutrients and water in forest soils (Jeffrey et al. 2011). Charcoal can also sorb undesirable compounds within the soil such as heavy metals (Zhang et al. 2013), allelochemicals (Sujeeun and Thomas 2017), salts (Thomas et al. 2013), and other contaminants (Uchimaya et al. 2010), and provide habitat to soil microbial communities within its porous structure (Zackrisson et al. 1996). Wood ash has been documented as driving increases in soil pH (Perkiomaki et al. 2003) and contributing large quantities of cations to soils following wildfire disturbance (Bodí et al. 2014), although our understanding of the specific role of wood ash and chars in boreal forest soils is limited (Hart and Luckai 2013). In recognition of these beneficial properties, charcoal produced through controlled pyrolysis, hereafter referred to as ‘biochar’, has been utilized extensively in agriculture as a climate-friendly option to remediate nutrient-poor (Rajkovich et al. 2012) and contaminated soils (Beesley et al. 2011). Similarly, biochar amendment in boreal forest soils has been suggested as an opportunity for both carbon sequestration (Lehmann 2007) and productivity enhancement (Stavi 2013). Existing studies, with some exceptions (e.g., Sarauer and Coleman 2018) have reported positive effects of biochar amendment on northern forest soils including improvements to soil carbon stocks and soil fertility (Palviainen et al. 2018) and enhanced tree seedling growth and survivorship (Pluchon et al. 2014) with neutral or positive associated effects on soil nutrients and microbial communities (Gundale et al. 2016). A meta-analysis of tree growth responses to biochar found an average 41% increase in biomass, with the largest responses occurring in tropical and boreal systems (Thomas and Gale 2015). Although the results of these studies are promising, most data are from short-term greenhouse pot trials that do not adequately represent soil processes and biota. Further, pot trial investigations are unable to account for potential effects of biochar soil amendment on competing vegetation, a critical component of successful boreal forest regeneration as discussed above. To date, field trials investigating biochar amendment effects on boreal forest regeneration are limited to a single study in which vegetation was found to be unresponsive to biochar soil amendment (Gundale et al. 2016). Given the known importance of vegetation community structure and composition on boreal forest regeneration, and potentially significant contributions of biochar to soil nutrient cycling and forest regeneration, field studies examining the effects of biochar amendment on soil nutrients and vegetation communities are a research imperative.

The utilization of wood ash, a waste product of bioenergy production, as a “high ash biochar”, is a promising alternative to commercially manufactured biochar. Wood ashes, in particular bottom ash from certain wood gasification systems, can be very high in carbon content (e.g., 20-60% C) and thus can qualify as a type of biochar, meeting current standards for soil amendments of the Canadian Food and Inspection Agency and the International Biochar Initiative (CFIA 2018, IBI 2012). Wood ash soil amendments are common in some northern forest systems in Europe and have exhibited long-term (>10 years) positive effects on growth and survivorship of tree seedlings, soil nutrient cycling (Demeyer, Nkana, and Verloo 2001), and microbial communities (Peltoniemi et al. 2016). In Canadian forest systems, wood ash soil amendment is an emerging practice and there is a recognized need for further research in this area (Hannam et al. 2018)

Of the studies currently available examining wood ash effects on Canadian forest systems, the majority are short term and with highly variable results. In general, wood ash amendment results in positive effects on soil pH, but the associated short-term effects on tree growth are often negative (Hannam et al. 2018). A recognized challenge associated with wood ash soil amendment that likely contributes to these results is the heterogeneity of the material and the presence of trace elements resulting from the highly variable feedstocks and combustion systems associated with bioenergy production (Omil, Pineiro, and

Merino 2013). There are contrasting reports of the effects of heavy metal accumulation from wood ash on vegetation and soil, with results ranging from negligible to severe; impacts depend on a range of site-specific factors that are highly dependent on alkaline species content and the threshold(s) at which adverse effects may be manifest (Maresca, Hyks, and Astrp 2017). Given the previously observed positive effects of biochar soil amendment on boreal forest soils, successful use of wood ash amendment in European systems, and logistical incentives associated with wood ash utilization in the Canadian boreal, comparative field studies of both wood ash biochar and commercially produced biochar amendments are called for. Particular attention to nutrient and toxic element characterization is necessary to test and, if possible, optimize the potential use of these soil amendments in managed boreal forests in Canada.

The present study investigates early forest regeneration outcomes of additions of two different biochar soil amendments: 1) a wood ash composed of 22% carbon, hereafter referred to as a high-carbon wood ash biochar and, 2) a slow pyrolysis produced biochar from trembling aspen (*Populus tremuloides* Michx.) chip feedstock (71% carbon), hereafter referred to as poplar biochar. Biochars were applied to a clear-cut harvest site in Northern Ontario Canada planted with white spruce (*Picea glauca* [Moench] Voss). We test three hypotheses: (i) Biochar amendments will enhance soil fertility by increasing pH of acidic soils and available mineral nutrients, in particular P, K, and Ca; (ii) Poplar biochar and high-carbon wood ash biochar amendments will differ in their impacts on seedling growth due to different effects on availability of soil nutrients and toxic metals; and (iii) Poplar biochar and high-carbon wood ash biochar amendments will alter community composition of non-tree vegetation, favoring nutriphilous species and species associated with more alkaline soils.

Materials and Methods

Study site

The study plots were established in September 2014 near Kakabeka Falls in Northwestern Ontario, Canada (48° 23’N, 89° 46’W) in a recently harvested 45-year-old white spruce (*Picea glauca*) plantation on the Precambrian Shield located within the Pigeon River Ecoregion within the Boreal Shield ecozone. The substrate is a silty clay loam soil consisting of dystric brunisols and grey luvisols derived from granitic parent material (pH 4.4) (Ontario Ministry of Natural Resources 2018b). The climate in this area is cool and relatively dry with annual precipitation between 674 – 838 mm and mean daily temperatures in January of -15°C and in July of 17°C (Ontario Ministry of Natural Resources 2018b). The site was clearcut in December 2013 and re-planted with white spruce seedlings during the week of June 22 – 28, 2014. Containerized 1+1 white spruce stock from Hill’s Greenhouse in Thunder Bay, ON was planted using provincial standards at an average 2.2 m spacing (2066 trees/ha). The site had a gentle slope of ~5%, facing west.

Biochars

Poplar biochar was generated specifically for the purposes of the experiment. Feedstock consisted of trembling aspen (*Populus tremuloides*) wood chips harvested and processed at Haliburton Forest and Wild Life Reserve, Ltd. A rotating kiln closed slow pyrolysis system was used, with a residence time of 2 hours and a peak temperature of ~350°C. Poplar biochar was rinsed following production and manually fragmented to pass through a 1.5-cm sieve prior to application. High-carbon wood ash biochar was obtained from Wood Ash Industries as a waste product from Kirkland Lake Power, a wood-fired co-generation power plant located near the study site in Kirkland Lake, Ontario. Kirkland Lake Power utilizes mixed feedstocks consisting of wood wastes (sawdust and bark) from local softwood (dominant species *Picea glauca* and *Picea mariana*) forestry operations; the wood gasification system operates at a temperature of ~1100°C.

Biochars were characterized according to the following protocols: total carbon (TC) and total nitrogen (TN) were measured by combustion analysis (CN 628, LECO Instruments, Canada) of ground biochar samples. Biochar pH and electrical conductivity (EC) were determined from 1:20 biochar to water solution (Denver Instruments UB-10 Ultra Basic analyzer fitted with a Bluelab pH electrode). Total biochar moisture, volatile matter, and ash content were determined following standardized methodology (ASTM D1762-84) by oven drying at 105°C, muffle furnace combustion at 750°C, and heating at 950°C, respectively. Elemental content (Al, Ag, As, Au, Ba, Be, Bi, Ca, Cd, Ce, Co, Cr, Cu, Cs, Fe, Hf, K, La, Li, Na, Nb, Ni, P, Rb, Pb, S, Mg, Mn, Mo, Sb, Sc, Sn, Sr, Ta, Th, Ti, U, V, W, Y, Zn) was determined from oven-dried samples. Samples were ground to a fine powder and then analyzed by a 4-acid digestion (hydrofluoric acid, followed by a mixture of nitric and perchloric acids, and solubilized using a mixture of nitric and hydrochloric acid) followed by Inductively Coupled Plasma Mass Spectrometry (ICP-MS) completed by Activation Laboratories Ltd. (Ancaster, Ontario Canada). All analyses were conducted in triplicate; properties of the two biochars are listed in Table 1.

Experimental design

The experiment was carried out as a fully randomized unbalanced complete block design with seven replicates of each treatment. The plots measured 5 m x 5 m and were arranged in two blocks, the upslope block consisting of three replicates of each of the three treatments (n = 9) and downslope block consisting of four replicates of each of the three treatments (n = 12) with a minimum buffer zone of 5 m between plots. Plots were positioned so as include at least 2 operationally planted seedlings in each plot. Stumps were avoided so they did not occur within the study plots and did not affect amendment application rates. Treatments were randomly assigned within each block. The three treatments included: 1) control (C), 2) poplar biochar (P) and, 3) high-carbon wood ash (W). Both biochars were applied at a dosage of 5 t/ha, which is representative of natural inputs of char following wildfire (Hart and Luckai 2014) though it corresponds to a low addition rate compared to prior agricultural studies (Jeffrey et al. 2011). Treatments were applied by hand as a top dressing to the soil in Sept. 2014 at which point vegetation cover on the site was <10%..

Measurements

Tree seedling height and root collar diameter were recorded to the nearest 1 cm and 1 mm, respectively, in September 2014 and October 2017. In August 2017 understory plants were identified and percent cover visually estimated in 1-m^2 quadrats in three locations within each plot. Cover estimates were made to the nearest 0.5% for values below 10%, and to the nearest 5% for values above 10%.

Soil samples were obtained in September 2017 from the top 15 cm of soil in three randomly selected points within each plot using a 5-cm diameter stainless steel soil corer. Samples from each plot were pooled and homogenized. Soil moisture was determined gravimetrically by weighing fresh and dried soil samples to the nearest 0.001 g; soil samples were dried for 24 hours at 60°C in a forced air oven and moisture content (%) calculated as [(Wet mass – Dry mass)/ Wet mass] x 100. Soil pH was determined on 1:10 soil to deionized water mixtures (Denver Instruments UB-10 Ultra Basic analyzer fitted with a Bluelab soil pH electrode). Soil bulk density of the top 15 cm of mineral soil was determined by drying soil samples at 60° C for 48 hours, weighing and dividing mass of dry soil by the sample volume. Soil total carbon (TC) and total nitrogen

(TN) of the top 15 cm of mineral soil were measured on ground and homogenized samples using an elemental analyzer/combustion analysis (C:N 628, LECO Instruments, Canada). To measure supply rates of both cations and anions (including NO_3^-, NH_4^+, PO_4^{3-}, SO_4^{2-}, and cationic forms of Ca, Mg, K, Fe, Mn, Cu, Zn, B, Pb, Al, and Cd), Plant Root Simulator Probes (PRS) (Western Ag Innovations Inc, Saskatoon, SK, Canada) were installed in at depths of 5 and 10 cm in the soil in September 2014 and collected 8 weeks following treatment in October 2014. Additionally, soil temperatures were measured using temperature loggers programmed to record temperature at 90-minuite intervals. Log-tag (LogTag TRIX-8 Data Logger) soil temperature loggers were installed at ~5 cm depth in each plot in September 2014 and collected in November 2015. Sensors were sealed in 0.09-mm polypropylene sleeves. Lab calibration trials of identical sensors show an accuracy of ±0.11°C RMSE in the range -10-35°C (Halim and Thomas 2018).

Statistical analysis

Data were analyzed using multivariate and univariate statistics in the R Version 1.1.465 statistical programming environment (R Core Team 2016).

To compare the effects of the biochar treatments, statistical analysis of soil nutrients, soil moisture, bulk density, and tree seedling growth (change in height and root collar diameter, respectively), was completed using one-way Analysis of Variance (ANOVA). Prior to conducting the ANOVA, linear mixed effects models that included block as a random effect were utilized to determine if the block had a significant effect on the analysis. When block was found to be non-significant, the term was dropped and an ANOVA was utilized to compare treatment effects for the variables. Prior to conducting ANOVA, data were checked for homogeneity of variance using Bartlett tests and residuals were visually assessed for normality using Q-Q plots. When ANOVA was significant ($p < 0.05$), post-hoc Tukey's honest significant difference tests determined differences among treatments with $p < 0.05$ indicating significance. Linear mixed effects models (including block as a random effect) were utilized (R package lme4 and LmerTest) to compare treatment effects on mean annual soil moisture as preliminary analysis revealed that there was a significant block effect (Bates et al. 2015; Kuznetsova et al. 2017).

Analysis of treatment effects (explanatory variables) on vegetation cover (response variables) was based on multivariate generalized linear models (GLM) using the mvabund package in R to take into account multiple comparisons, potential correlations between species, and non-normal data. Mvabund fits a separate GLM to each species allowing for resampling-based hypothesis testing with greater power than distance-based ordination methods (Warton, Wright, and Wang 2012). In the analysis a negative binomial residual distribution was assumed, in agreement with graphical analysis of residual patterns. Probability integral transform residual bootstrap resampling (PIT trap sampling) was utilized to calculate P-values, as recommended by Wang et al. 2012. Species that were observed in less than four plots were excluded from analyses. Preliminary analyses indicated a significant block effect on plant community composition, so block was included as a covariate in GLM analyses. Redundancy Analysis (RDA) was utilized to graphically describe multivariate treatment effects on community composition.

Path analysis completed using the lavaan package in R was used to investigate causal relationships between the treatment effects on both the competing vegetation and white spruce seedling growth (Rosseel, 2012). Principal Components Analysis (PCA) was used to obtain composite variables to quantify total ion-exchange-resin measurements for soil nutrients (B, Mg, NH_4, NO_3, K, P, S, Mn, and Ca) and for total toxic metals plus micronutrients commonly at excess concentrations (Al, Pb, Zn, Fe, Cd, and Cu). Partial regressions established the strengths of the observed relationships. The structure of the path model examined assumes that biochar applications can potentially influence tree growth through direct effects of total nutrients, total toxic metals plus micronutrients commonly at excess concentrations, and through vegetation competition, evaluated as total percent cover of competing vegetation; the latter was itself a function of soil nutrient and toxic metals content.

Results

Effects of biochar amendments on soil properties

There were significant differences among treatments in availability of Ca, Cu, Zn, B, S, and Pb eight weeks after biochar application (Table 2). The majority of the observed differences in available nutrients were a result of enhanced availability of micronutrients and metals in soil amended with high-carbon wood ash.

Pairwise comparisons indicated that high-carbon wood ash treatments had significantly greater availability of the following compared to both the control and the poplar biochar: Cu, Zn, B, SO_4^{2-}, and Pb and significantly greater availability of Ca compared to the control (Table 2). Differences among treatments for soil NO_3-, NH_4^+, Mg, K, PO_4^{3-}, Mn, and Al availability were not significant.

Three years following biochar amendments, there were significant differences in soil pH among the three treatments (F(2,14) = 8.22, *P* = 0.004) (Figure 1). High-carbon wood ash amendment caused significant increases in soil pH compared to both the poplar biochar (*P* = 0.030) and control (*P* = 0.003); pH values for the poplar biochar treatment were slightly higher than the control, though the difference was not significant. There were no significant differences among treatments with regard to soil moisture or bulk density ($P > 0.05$). There was some limited evidence of elevated soil C and N in the poplar biochar treatments compared to both the high-carbon wood ash and the control though the relationship was not quite significant for carbon (F(2, 16) = 2.74, *P* = 0.094) and not significant for nitrogen.

Average soil temperature in the high-carbon wood ash biochar plots were higher than the poplar biochar plots and the controls in September, October, and in April, May, June, July and August of the following year, though the difference was only significant during the month in which the experiment was installed, in September 2014 (F(2,16) = 5.125, *P* = 0.0194); there was also a significant block effect (F(3,14) = 4.938, *P* = 0.033), with the upslope block showing slightly higher soil temperatures. In September 2014, both biochar treatments exhibited higher soil temperatures than the control, though only the difference between the high-carbon wood ash treatment and control was significantly different (*P* = 0.016) (Figure 2). Temperature differences among treatments were not statistically significant in later months.

Tree growth responses

Tree survivorship averaged 66% over the 3 years of the study, and did not vary significantly among treatments; growth effects were evaluated for the surviving trees only. The change in root collar diameter (RCD) of planted white spruce over three years was significantly different among treatment types (F(2,22) = 4.916, *P* = 0.017) (Figure 3). Trees showed significantly reduced RCD growth in plots amended with the high-carbon wood ash biochar compared to the control (*P* = 0.018). Change in height over three years was not significantly different among treatment types, (*P* > 0.05) (Figure 3). Path analysis showed that RCD growth was significantly affected by soil metals content (*P* = 0.024), while soil nutrient content path coefficient was only marginally significant (*P* = 0.090), and percent cover of competing vegetation did not have a significant effect on RCD growth (*P* > 0.05).

Vegetation community responses

A total of 29 non-tree plant species were identified in the plots. Total vegetation cover did not differ significantly with treatment (based on a linear mixed model analysis), but there was a trend toward higher cover in the poplar biochar treatment (mean cover values were 63.4±5.9% (SE), 75.5±4.4%, and 69.5±4.2% for the control, poplar biochar, and high-carbon wood ash biochar treatments, respectively). Generalized multiple linear model results indicate that there were significant responses of plant community composition to treatment (F(2,54) = 74.7, P = 0.006), and also significant differences between blocks (upslope vs. lower slope) (F(1,53) = 58.84, P = 0.004). Table 4 summarizes the observed species-specific responses and Figure 5 depicts responses for the most common species. Both biochars significantly increased cover of *Solidago canadensis* and *Vicia cracca*. The poplar biochar also significantly increased cover of *Rubus idaeus* compared to the controls. High-carbon wood ash biochar significantly decreased cover of *Symphyotrichum cordifolium* (compared to both the poplar biochar and control), and there was a notable (but not significant) decrease in Salix *bebbiana*. High-carbon wood ash biochar significantly increased cover of *Ranunculus acris* compared to both the poplar biochar and the control. RDA biplots (Fig. 6) show a clear separation of the control plots vs. the biochar-amended plots, with more overlap between the latter; these patterns are attributable mainly to the changes in abundances of common species noted in Figure 5. Linear mixed model results indicate that species richness (number of species per plot, pooling the 3 replicate sub-plot samples) was not significantly different among treatments. Path analysis results did not detect significant nutrient and metals content effects on non-tree vegetation percent cover (P > 0.05) (Figure 4).

Discussion

The results of our study indicate that high-carbon wood ash and poplar biochar soil amendments can significantly increase soil pH and alter species composition of the ground vegetation plant community under field conditions, with large species-specific differences in responses to biochar type. Poplar biochar and high-carbon wood ash biochar soil amendments did not enhance growth of the planted white spruce seedlings compared to controls. Indeed, the high-carbon wood ash had significant negative effects on spruce seedling height growth. Path analysis results suggest that this response is due to the presence of metals and micronutrients found in excess in the high-carbon wood ash biochar, though it should be kept in mind that path analysis is based on correlation structure rather than manipulations of individual variables.

Tree Seedlings

We observed non-significant effects of biochar amendments on seedling height growth and significant negative effects of high-carbon wood ash biochar amendments on seedling root collar diameter growth. The enhanced cover of *Rubus idaeus* in poplar-biochar-amended plots suggests possible competition effects, consistent with prior studies. For example, in a 5-year study in a clear-cut boreal mixedwood site in Quebec, Jobidon (2000) found that white spruce growth was adversely affected even by moderate levels of competing vegetation cover. However, in the present study path analysis revealed that the negative effects of the heavy metals present in the high-carbon wood ash biochar had a greater effect on seedling growth than competition. In fact the path coefficient between total understory cover and tree RCD, while not significant, was positive. A meta-analysis (Thomas and Gale 2015) suggests that angiosperm trees are generally much more responsive to biochar applications than conifers. Recent results also mostly support this pattern (e.g., Staples and Van Rees 2001; Pluchon et al. 2014; Sarauer and Coleman 2018). Our finding of small or negative effects of biochar addition on white spruce performance is thus consistent with prior work.

Soil properties

The concentration of heavy metals present in both biochars utilized in this study are well below the International Biochar Initiative's Maximum Allowed Thresholds (IBIMAT) for As, Cd, Co, Cu, Pb, Mo, Ni, and Zn (International Biochar Initiative November 2015). However, the negative effects of the high-carbon wood ash biochar on white spruce seedling growth suggest that the IBIMAT's thresholds may not be stringent enough to limit the potentially negative effects of heavy metals on tree seedlings subject to high-carbon wood ash amendment. Indeed, when compared to the results of a meta-analysis examining common concentrations of nutrients and metals in wood ash, that found no significant effect of wood ash on heavy metal contamination of the soil, the high-carbon wood ash utilized in this study was above the median reported value for Al, Fe, Zn, Cu and Cd (Augusto, Bakker, and Meredieu 2008). In contrast, the poplar biochar had heavy metal concentrations that were well below the median and not within the range of those commonly reported for wood ash (Augusto, Bakker, and Meredieu 2008). Ion exchange resin data indicated that both Cu and Zn availability in the high-carbon-wood-ash-biochar-amended plots was significantly higher than both the control and the poplar biochar, while availability of Al, Fe, and Cd was not, despite higher concentrations of the latter in the high-carbon wood ash.

A prior study in a Scots pine (*Pinus sylvestris* L.) stand utilizing the same application rate examined here (5 t/ha), found that heavy metal contamination from wood ash soil amendment was limited for some heavy metals (Pb, Cu, Cr, Cd) as a result of binding in insoluble forms to organic matter and the alkalizing effect of the biochar that reduced heavy metal solubility; however there was significant downward transport of Zn two years following treatment, suggesting that Zn is not immobilized in this manner (Ozolincius and Varnagiryte 2005). While Zn is an essential plant micronutrient and prior studies have found spruce to be tolerant to lower levels of Zn (3 – 33 ppm), higher levels of Zn (>100 ppm) have resulted in significant decreases in spruce seedling shoot, root and primary needle length, and increased seedling desiccation (Ćurguz, et al. 2012). Given the high concentration of Zn in the high-carbon wood ash (511 ppm), and significantly enhanced availability of Zn in the soil observed in the present study, it is likely that Zn toxicity contributed to the negative effects observed. Further study is recommended in this area in order to improve existing Zn guidelines for biochar certification.

Soil temperature responses indicated a short-term warming effect of the biochar treatments on the soils during the first month following treatment. However, this effect was no longer significant after the first month over the 15 months in which temperature was measured. These results indicate biochar impacts on surface albedo and potential negative feedback effects and implications for enhanced soil CO_2 flux, as highlighted in Genesio et al. 2012, were highly transient in this case.

Plant community composition

There have been very few prior field studies examining biochar effects on plant community composition, and the few results reported have been mixed. Biederman et al. (2017) examined community change in a restoration trial in a tallgrass prairie system in western Iowa, USA, finding relatively small effects of biochar amendment on total cover and no detectable change in community composition over 5 years. In contrast, in a grassland restoration study in the Netherlands, van de Voorde et al. (2014) found dramatic effects of biochar amendment on species composition over one growing season, with leguminous plants increasing 3-fold in abundance with a concomitant increase in N fixation. In a lodgepole pine (*Pinus contorta* Douglas) forest in Colorado, Rhoades et al. (2017) found no detectable effects of biochar addition on total forb cover or cover of fireweed (*Chamerion angustifolia*), but did not assess other species. Here we found that both studied biochars significantly enhanced cover of *Solidago canadensis* and *Vicia cracca.* Poplar biochar significantly increased cover of the ruderal species *Rubus ideaus* compared to the control, and high-carbon wood ash biochar significantly decreased cover of *Symphyotrichum cordifolium* and increased cover of *Rununculus acris* compared to the other two treatments.

We hypothesized that the biochars would favor nutriphilious species, as in prior studies (van de Voorde et al. 2014, Kuttner and Thomas 2017; Liu et al. 2013); and indeed cover of all of the nitrogen-fixing species identified in this study (*Alnus incana* (L.) Moench*, Trifolium pratens* L.*, Trifolium repens* L. *and Vicia cracca*), was enhanced in biochar amended sites compared to controls, though only the *Vicia cracca* exhibited a significant response. This result is consistent with prior studies in which biochar application resulted in significantly enhanced growth of legumes (including *V. cracca*), a result that was largely attributed to increased soil K availability in biochar-amended plots (van de Voorde et al. 2014). Enhanced soil available K is likely a significant contributor to the enhanced legume cover observed here as both biochars had significantly higher soil available K compared to the control. Potassium is a known mediator of root nodulation, and has been linked to enhanced legume competitiveness in the past (Sangakkara et al. 1996). Indeed, in a greenhouse experiment examining the effects of enhanced K availability on legume competitiveness, increases in K availability resulting from either direct K fertilization or biochar amendment resulted in a beneficial effect under N-limited conditions (Oram et al. 2014). *Solidago canadensis* cover was also significantly enhanced by both biochars. It is possible that this species has a competitive advantage when there is limited soil inorganic N, as this species is known to utilize amino acids as a source of N (Yu et al. 2016), and we observed reduced availability of inorganic N forms. Prior studies have found biochar sorption and immobilization of inorganic N, in particular ammonium (Nguyen et al. 2017, Gale et al. 2017). In this study, *Rubus idaeus* had increased abundances in both biochar treatments. *Rubus idaeus* is an aggressive early colonizer of disturbed sites with a well-known capacity to effectively outcompete conifer crop species, including white spruce (Bell and Pitt 2007).

Increased cover of *Ranunculus acris* observed in the high-carbon-wood-ash-biochar-amended sites is consistent with prior work that found biochar amendment led to a shift from grasses to forbs, possibly related to higher nutrient retention capacity of forb species in nutrient-poor environments (Schimmelpfennig et al. 2015). The significantly decreased cover of *Symphyotrichum cordifolium* in the high-carbon-wood-ash-biochar-amended sites was unexpected: prior studies examining soil amendments on mine spoils have found that this species favored growth in high pH environments and had the ability to adapt to widely varying nutrient availability (Voeller, Zamora, and Harsh 1998). A recent survey of early-successional plant species responses to a hardwood biochar found negative responses prevalent among the most N-demanding species with the highest photosynthetic rates (Gale et al. 2017).

All three treatments in our study experienced a “regeneration failure”: i.e., conversion from a white spruce plantation on bare topsoil to a dense, continuous cover of mixed native pioneer and non-native ‘weed’ species in a period of three years. This result is consistent with prior studies of regenerating stands in which high herbaceous cover negatively affected regeneration of conifer trees (Gartner et al. 2014). A 2005 review on the role of vegetation management in global forest productivity found that the elimination of competing vegetation commonly resulted in 2-10-fold increases in tree growth (Wagner et al. 2005). The presence of a rich seed bank consisting of both non-native and pioneer species was not anticipated as part of this study. Indeed, invasive species tend to be limited in boreal forests due to the lack of propagule pressure, limited disturbance, and cold winters (Langor et al. 2014). However, the proximity of our study site to areas of both active and historical agricultural use appears to have provided a sufficient seed supply, while the clearcut harvest and site preparation provided open areas of full sunlight that these species require to thrive. In the pre-harvest white-spruce-dominated forest, the closed canopy likely limited invasive plant establishment. Had the non-native species present in our site been allelopathic, it is possible that the biochar additions may have facilitated enhanced competitiveness as hypothesized as biochar can sorb phenolics from allelopathic species, enhancing survivorship in boreal ecosystems (Wardle et al. 1998). Use of biochar in boreal forests where competitors are known to be allelopathic may be of greater benefit and should be investigated further.

Conclusions and implications for management

This study highlights large species-specific responses to biochar soil amendments. Increases in highly competitive species such as *Rubus idaeus* following biochar addition suggest the potential for soil amendments to exacerbate competitive effects; however, our results indicate that direct negative effects of toxic elements in some biochars may be of greater concern.

Investigation of biochar effects on soil function and nutrient cycling is intuitive, in recognition of the potential contribution to carbon sequestration and importance of fire in boreal ecosystems. Although the high-carbon wood ash utilized in this study meets the standards outlined by the Canadian Food Inspection Agency (CFIA) under the Federal Fertilizer Act, our results suggest that the thresholds outlined by the CFIA for some metals and micronutrients (e.g., S, B, Ca, Cu, Zn, Pb) may not be robust enough to limit unintended adverse effects on forest regeneration. Further research is recommended in this area in order to elucidate species-specific impacts and toxicity thresholds. Further research in the area of vegetation management in boreal forest regeneration in response to changing management objectives and climate warming is also recommended. Despite the unintentional effects of soil amendments on boreal forest regeneration in this study, biochar remains a promising option as a supplementary feature of existing NDE management in the boreal forest.

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Table 1: Properties of poplar biochar and high-carbon wood ash used in experiments. Values are listed ± standard error (SE). ND indicates values below detection threshold.

Attribute	**Poplar biochar**	**High-carbon wood ash biochar**
Peak Pyrolysis Temperature	350°C	1100°C
Ash (%)	12.34±2.27	78.3±0.46
Moisture (%)	60.4±4.28	5.56±0.63
Volatile matter (%)	23.37±1.10	6.07±0.10
Electrical Conductivity (µS)	202 ± 20.95	711 ± 7.4
pH	7.5 ± 0.08	8.6 ± 0.01
Bulk density (g cm^{-3})	15.8 ± 0.68	38.70 ± 0.15
Elemental Composition		
C (%)	71.1 ± 0.1	22.0 ± 0.3
N (%)	0.405 ± 0.004	0.245 ± 0.0.006
Al (%)	ND	3.45±0.13
Ca (%)	0.61±0.02	10.0±0.24
Fe (%)	0.13±0.005	1.51±0.08
K (%)	0.26±0.005	1.81±0.03
Na (%)	0.005±0	1.37±0.06
P (%)	0.04±0	0.14±0.02
S (%)	ND	ND
Mg (%)	0.07±0.003	1.23±0.04
Ti (%)	ND	1.83±0.03
Ag (ppm)	0.23±0.03	0.3±0
As (ppm)	ND	29.0±1.5
Au (ppm)	ND	ND
Ba (ppm)	135.0±2.3	905.7±10.65
Be (ppm)	ND	ND
Bi (ppm)	ND	ND
Cd (ppm)	1.2±0.06	3.36±0.2
Ce (ppm)	ND	17.3±0.7
Co (ppm)	0.5±0	6.8±0.3
Cr (ppm)	6.3±0.7	100.6±3.8
Cu (ppm)	6.4±0.8	145.3±6.1
Cs (ppm)	ND	0.63±0.03
Hf (ppm)	ND	0.2±0.1
La (ppm)	0.3±0	6.83±0.3

Li (ppm)	ND	8.2±0.05
Nb (ppm)	0.6±0.4	0.1±0
Ni (ppm)	1.1±0.09	25.4±3.5
Rb (ppm)	12.5±0.3	47.8±0.8
Pb (ppm)	1.23±0.6	60.4±3.4
Mn (ppm)	127.6±20.0	2106.7±61.2
Mo (ppm)	0.23±0.07	10.2±4.6
Sb (ppm)	ND	0.4±0.2
Sc (ppm)	ND	4±0
Sn (ppm)	0.43±0.13	0.47±0.14
Sr (ppm)	65.0±0.58	405.0±4.5
Ta (ppm)	ND	ND
Th (ppm)	ND	1.83±0.03
Tl (ppm)	ND	0.22±0
U (ppm)	ND	0.53±0.03
V (ppm)	ND	28.6±1.7
W (ppm)	ND	0.7±0.26
Y (ppm)	ND	5.3±0.09
Zn (ppm)	109.7±1.3	511.3±23.3
Zr (ppm)	0.25±0.05	14.0±3.1

Table 2: Nutrient and metal ion availability by treatment (control (C), poplar biochar (P) and high-carbon wood ash (W)) measured by ion exchange resins (PRS probes) in the first 8 weeks following soil amendment treatments. Values are expressed in units of as PRS™ probe supply rate (μg 10 cm^{-2} 56 d^{-1}). Means denoted by a different letter indicate significant differences between treatments (Tukeys HSD, $p < 0.05$).

Ion	C	P	W	Df	*F*-statistic	*p* value
Nitrate	0.2080	0.1780	0.0490	2, 35	2.82	0.073
Ammonium	0.0133	0.0085	0.0053	2, 35	2.40	0.105
Calcium	3.4459^{b}	3.8873^{a,b}	4.5615^{a}	2, 35	5.81	**0.006**
Magnesium	0.6074	0.5459	0.6173	2, 35	0.82	0.447
Potassium	0.2587	0.2986	0.3453	2, 35	0.46	0.633
Phosphate	0.0206	0.0197	0.0144	2, 35	0.49	0.616
Iron	0.0889	0.1323	0.1087	2, 35	0.24	0.785
Manganese	0.0332	0.0305	0.0372	2, 35	0.07	0.934
Copper	0.0003^{b}	0.0003^{b}	0.0015^{a}	2, 35	22.39	**<0.001**
Zinc	0.0043^{b}	0.0048^{b}	0.0091^{a}	2, 35	10.25	**<0.001**
Boron	<0.0001^{b}	<0.0001^{b}	0.0002	2, 35	5.04	**0.010**
Sulphur	0.0438^{b}	0.0358^{b}	0.5838^{a}	2, 35	60.22	**<0.001**
Lead	0.0004^{b}	0.0003^{b}	0.0008^{a}	2, 35	5.54	**0.008**
Aluminum	0.0348	0.0326	0.0385	2, 35	0.36	0.698
Cadmium	<0.0001	<0.0001	<0.0001	2, 35	0.46	0.629

Table 3: Effects of biochar amendments on soil properties. Means denoted by a different letter indicate significant differences between treatments (Tukey's HSD, $p < 0.05$).

Property	C	P	W	Df	*F*-statistic	*p* value
pH	4.34^a	4.57^a	5.13^b	2, 14	8.22	**0.004**
Bulk density (g/cm^2)	0.632	0.506	0.620	2, 13	1.405	0.282
Total C (mg/g)	72.0	77.8	119.7	2, 16	2.74	0.094
Total N (mg/g)	4.14	4.51	6.00	2, 16	2.52	0.111

Table 4: Mean percent cover of non-tree understory plants and treatments, control (C), poplar biochar (P) and high-carbon wood ash (W). Tests for species-specific cover responses to biochar soil amendments and block effects (in third growing season following treatment) are based on a generalized linear model analysis with a negative binomial residual distribution.

	Percent Cover (%)			**P-value**	
Species*	**C**	**P**	**W**	**Treatment**	**Block**
Rubus ideaus L.	5.8	23.2	14.4	**0.012**	0.205
Ranunculus acris L.	1.2	1.0	4.4	**0.018**	0.790
Fragaria vesca L.	6.4	3.2	4.9	0.346	0.949
Fraxinus nigra Marshall	3.4	1.4	5.5	0.206	0.504
Solidago canadensis L.	1.2	6.5	7.2	**0.059**	**0.036**
Lonicera canadensis Bartram	2.5	0	2.8	0.219	0.684
Betula papyrifera Marshall	0.8	0.5	0.8	0.858	0.104
Salix spp. L.	10.3	4.4	10.8	0.143	0.352
Salix bebbiana Sarg.	4.3	2.2	0.8	0.260	0.354
Trifolium pratense L.	1.1	0.7	0.3	0.612	**0.006**
Cirsium spp. Mill.	0.27	2.4	0.5	0.147	0.464
Taraxacum officinale (L.) Weber ex F.H. Wigg	1.5	0.9	3.0	0.231	**0.001**
Symphyotrichum cordifolium (L.) G.L.Nesom	10.7	14.6	3.0	**0.050**	0.064
Trifolium repens L.	0.3	1.7	2.7	0.281	0.091
Lupinus polyphyllus Lindl.	0.4	0.7	0.9	0.675	0.273
Leucanthemum vulgare Lam.	0.3	0.9	0.6	0.599	0.401
Viburnum acerifolium L.	1.8	1.4	0	0.067	0.208
Vicia cracca L.	0	4.1	4.5	**0.011**	0.720
Sambucus racemosa L.	1.4	0.5	0.8	0.815	0.445

* Additional uncommon species not included in analyses include *Caprifoliaceae* spp., *Ostrya virginiana* K. Koch, *Populus* deltoides W. Bartram ex Marshall, *Polygonum* spp., *Osmorhiza claytonii* (Michx.) C.B. Clark, *Cornus* spp., *Alnus incana* (L.) Moench, *Symphoricarpos albus* (L.) S.F. Blake, *Rubus odoratus* (L.), *Plantago major* (L.) and *Anemone canadensis* (L.).

Figure Captions

Figure 1: Soil pH (uppermost 15 cm of mineral soil) by treatment; means are plotted ± 1 SE. Data are separated by treatment: white bars are the control, black bars are poplar biochar, and grey bars are wood ash biochar. Means denoted by a different letter indicate significant differences between treatments ($p < 0.05$). Sample size is $n = 7$ per treatment.

Figure 2: Average soil temperature (°C) for the month of September 2014 (immediately following biochar amendments); means are plotted ± 1 SE. Data is separated by treatment: white bars are the control, black bars are poplar biochar and grey bars are wood ash biochar. Means denoted by a different letter indicate significant differences between treatments ($p < 0.05$). Sample size is $n = 7$ per treatment.

Figure 3: Change in white spruce (*Picea glauca*) root collar diameter (RCD) (a), and height (b), by treatment after three years; means are plotted ± 1 SE. Data are plotted by treatment: white bars are the control, black bars are poplar biochar and grey bars are wood ash biochar. Means denoted by a different letter indicate significant differences between treatments ($p < 0.05$). Sample size is $n = 7$ per treatment.

Figure 4: Path analysis linking white spruce growth, quantified as the change in Root Collar Diameter (RCD) over three years, to total percent cover of non-tree understory vegetation, substrate nutrient levels (the first PCA axis for ion exchange resin flux values for B, Mg, NH_4, NO_3, K, P, S, Mn, and Ca), and substrate toxic metals levels (the first PCA axis for ion exchange resin flux values for Al, Pb, Zn, Fe, Cd, and Cu). Solid lines indicate statistically significant paths ($P < 0.05$) except for the path between RCD and nutrient content ($P < 0.1$). Dashed lines indicate non-significant paths ($P > 0.05$).

Figure 5: Percent cover of *Rubeus idaeus, Solidago Canadensis, Vicia cracca, Symphoricarpos albus, Salix bebbiana* and *Rununculus acris* after three years; means are plotted ± 1 SE. Data are separated by treatment: white bars are the control, black bars are poplar biochar and grey bars are wood ash biochar. Means denoted by a different letter indicate significant differences between treatments ($p < 0.05$).

Figure 6: Redundancy analysis (RDA) biplot based on total non-tree species cover, with treatment as the explanatory variable. Replicate plots are illustrated as the terminal points of "spider diagrams", with the centroid corresponding to the treatment weighted average: control plots is shown in green, poplar biochar in red, and high-carbon wood ash in purple.

Figure 1

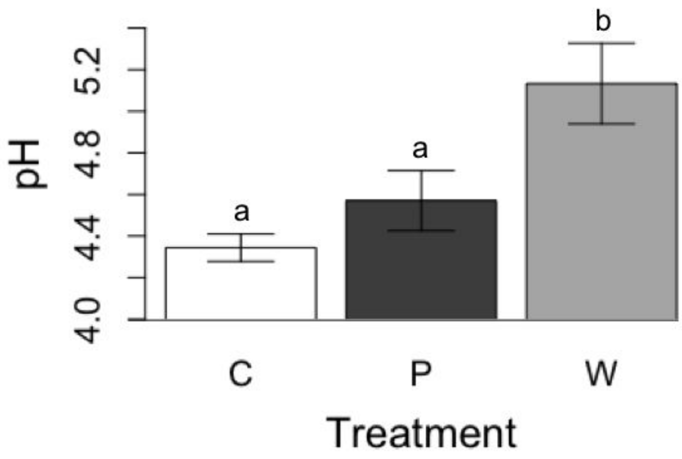


Figure 2

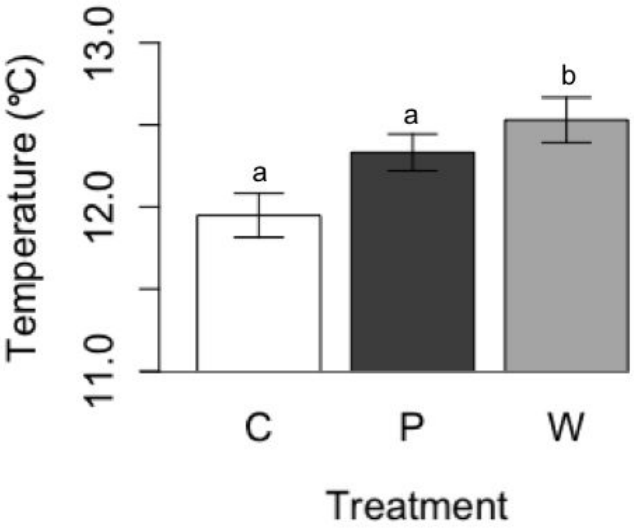


Figure 3

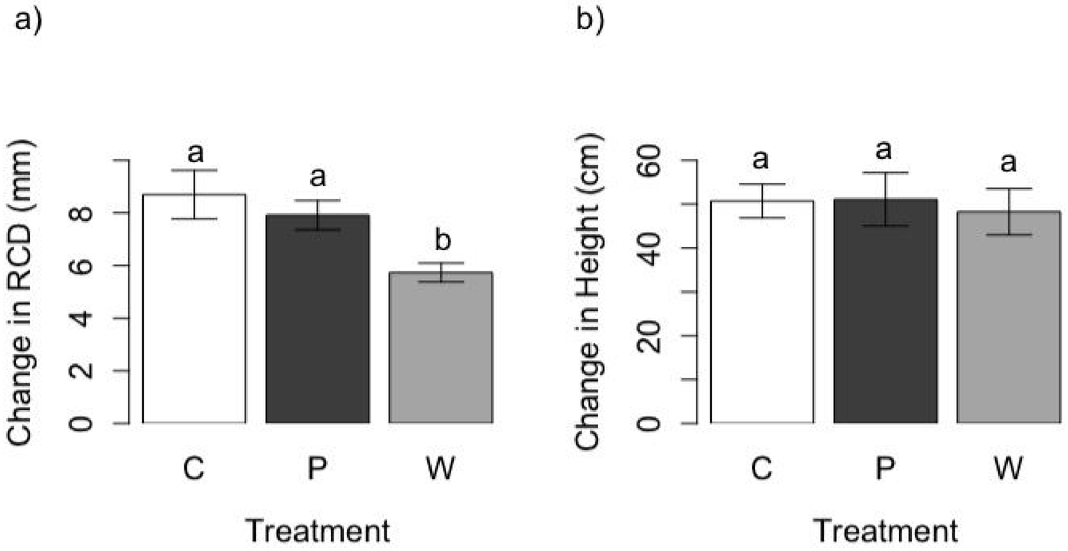

a)
Change in RCD (mm)
0
2
4
6
8
a
a
b
C
P
W
Treatment
b)
Change in Height (cm)
0
20
40
60
a
a
a
C
P
W
Treatment

Figure 4

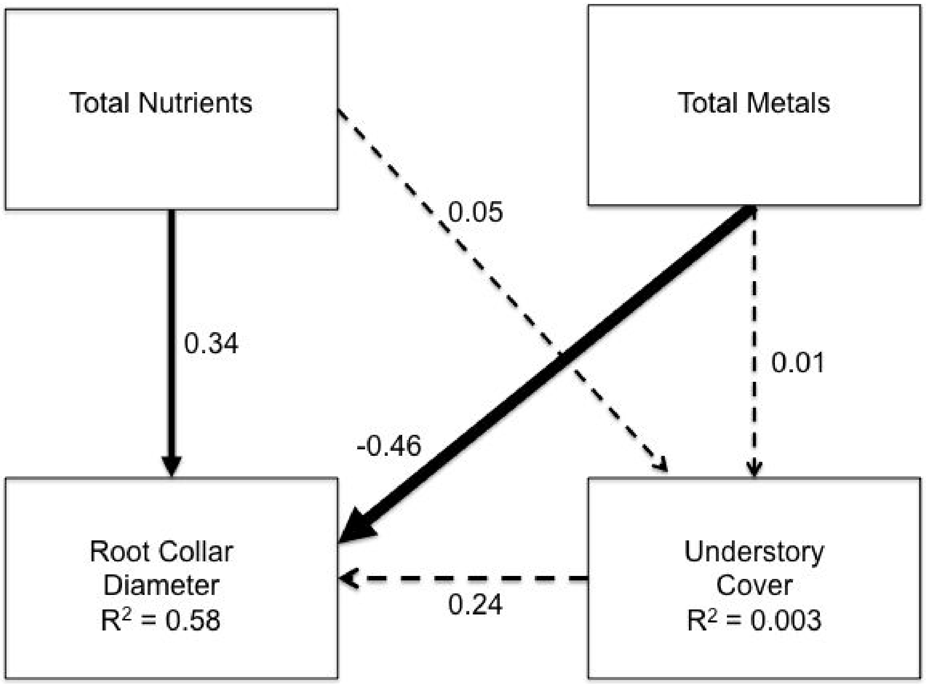

Total Nutrients
Total Metals
0.05
0.34
0.01
-0.46
Root Collar Diameter $R^2 = 0.58$
0.24
Understory Cover $R^2 = 0.003$

Figure 5

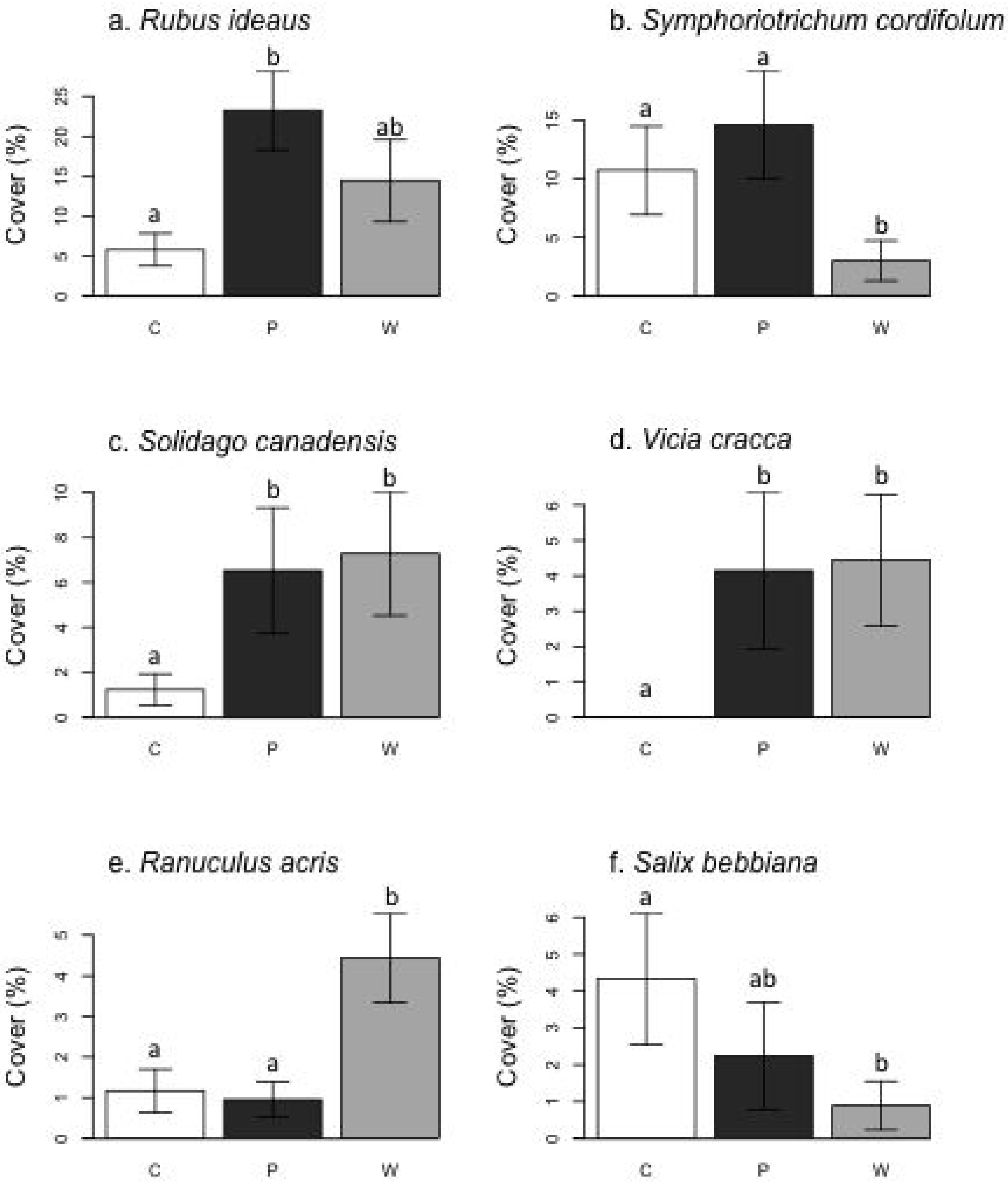
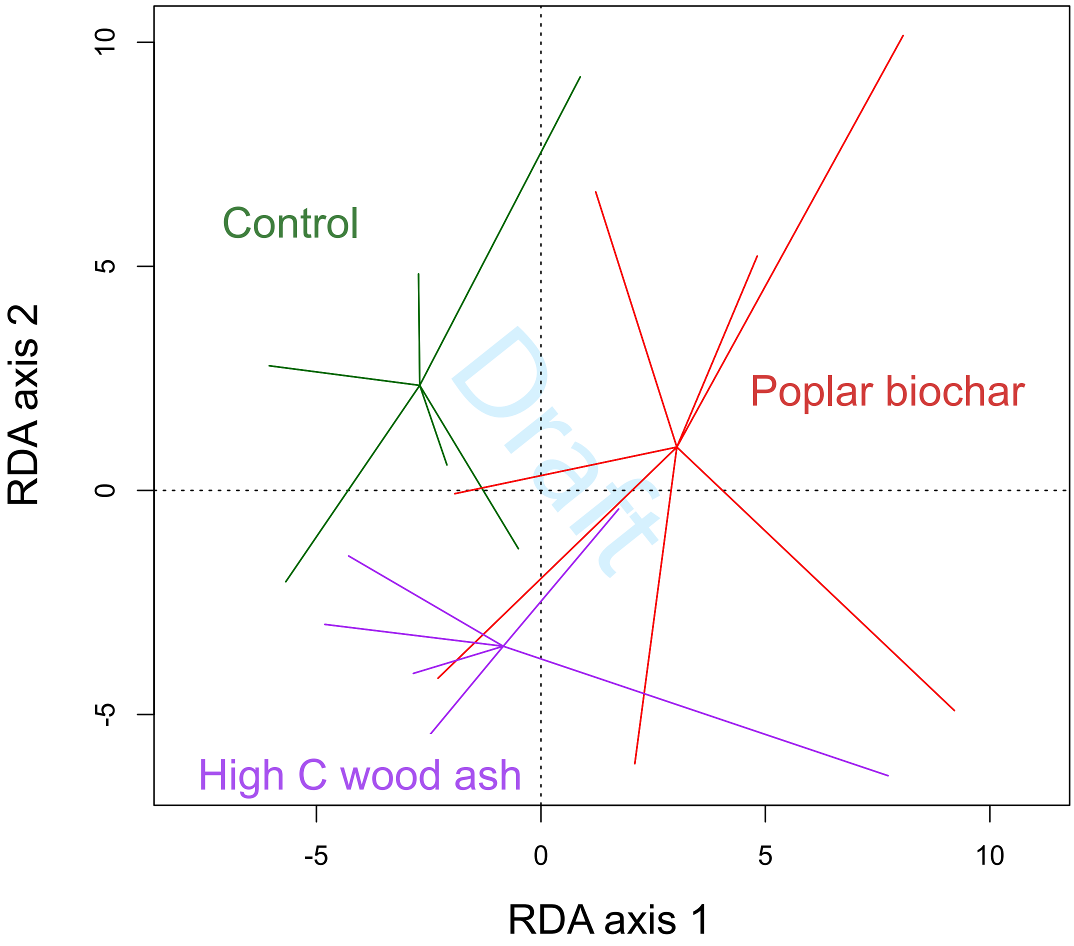

a. Rubus ideaus
Cover (%)
0
5
10
15
20
25
a
b
ab
C
P
W
b. Symphoriotrichum cordifolum
Cover (%)
0
5
10
15
a
a
b
C
P
W
c. Solidago canadensis
Cover (%)
0
2
4
6
8
10
a
b
b
C
P
W
d. Vicia cracca
Cover (%)
0
1
2
3
4
5
6
a
b
b
C
P
W
e. Ranuculus acris
Cover (%)
0
1
2
3
4
5
a
a
b
C
P
W
f. Salix bebbiana
Cover (%)
0
1
2
3
4
5
6
a
ab
b
C
P
W

Figure 6


Control
Poplar biochar
High C wood ash
RDA axis 1
RDA axis 2
-5
0
5
10
-5
0
5
10