Applied Soil Ecology 107 (2016) 368–380

Contents lists available at ScienceDirect

Applied Soil Ecology

journal homepage: www.elsevier.com/locate/apsoil

Soil microbial responses to wood ash addition and forest fire in managed Ontario forests

Genevieve L. Noyce[a,b,*], Roberta Fulthorpe[b], Adam Gorgolewski[c], Paul Hazlett[d], Honghi Tran[e], Nathan Basiliko[f,g]

[a] Department of Geography, University of Toronto, 100 St. George St, Toronto, ON M5S 3G3, Canada
[b] Department of Physical and Environmental Sciences, University of Toronto Scarborough, 1065 Military Trail, Toronto, ON M1C 1A4, Canada
[c] Faculty of Forestry, University of Toronto, 33 Willcocks St, Toronto, ON M5S 3B3, Canada
[d] Great Lakes Forestry Centre, Canadian Forest Service, 1219 Queen St E, Sault Ste. Marie, ON P6A 2E5, Canada
[e] Department of Chemical Engineering and Applied Chemistry, University of Toronto, 200 College St, Toronto, ON M5S 3E5, Canada
[f] Department of Biology and the Vale Living with Lakes Centre, Laurentian University, 840 Ramsey Lake Rd, Sudbury, ON P3E 6H5, Canada
[g] Department of Geography, University of Toronto Mississauga, 3359 Mississauga Rd, Mississauga, ON L5L 1C6, Canada

ARTICLE INFO

Article history:
Received 17 March 2016
Received in revised form 11 July 2016
Accepted 12 July 2016
Available online xxx

Keywords:
Wood ash
Microbial communities
Bacteria
Fungi
Amplicon sequencing
Forest soil amendments

ABSTRACT

Wood ash is typically applied to soils to counteract acidification and base cation depletion, but the effects of this ash on the organic layer microbial community are rarely studied. We analyzed microbial responses to ash addition in two field-scale experiments in Ontario, Canada. One experiment was in a young boreal forest soil and the other was in an uneven-aged hardwood north-temperate forest soil. We also investigated the effects of a natural forest fire near the boreal experiment. In both cases, ash addition had no effect on overall microbial biomass and respiration, but increased the phylogenetic diversity of bacterial communities and the relative abundance of *Bacteroidetes* taxa, though effects on other bacterial taxa were site-specific. Eukaryotic effects also varied by experiment; at the boreal site, ash increased eukaryotic phylogenetic diversity and decreased the fungi:bacteria ribosomal marker ratio, but at the temperate site ash decreased eukaryotic diversity and did not affect the fungi:bacteria ratio. There was limited additional effect on the boreal soil microbial community of increasing ash addition from 0.7 to 5.7 t ha^{-1}, as determined by T-RFLP analysis, though soil pH in both experiments increased with higher addition rates. At the temperate site, ash effects were consistently stronger for fly ash than for bottom ash. In both experiments, only 14 unique bacterial taxa were found after ash addition, and the strongest driver of overall community composition was the forest type, not ash treatment. In contrast, the soil microbial community observed at the forest fire site was clearly different. Overall, these results indicate that wood ash addition has only minimal effects on the composition of the soil microbial community in sites across two distinct global forest biomes.

Published by Elsevier B.V.

1. Introduction

Wood ash is a byproduct of the biomass combustion widely carried out in the forestry industry that also contains the macro- and micro-nutrients required for plant growth, with the exception of nitrogen (N). In North America, unlike in parts of Europe and South America, the majority of this ash is required to be disposed of in landfills (Elliott and Mahmood, 2006), but using it as a soil amendment instead can replace nutrients removed during harvest. If ash addition changes soil properties, this in turn can alter the resident soil microbial community. Generally, wood ash increases soil pH and base saturation in forested ecosystem soils (Bååth and Arnebrant, 1994; Fritze et al., 1994, 2001; Perkiömäki et al., 2003), as well as water-holding capacity (Etiégni et al., 1991). These effects typically persist beyond the first year after ash addition (Fransman and Nihlgård, 1995; Saarsalmi et al., 2001; Jacobson et al., 2004), so ash can have long-term microbial effects. Soil pH is a particularly strong driver of soil bacterial diversity, as well as the overall fungi:bacteria ratio; fungi usually dominate the soil microbial community in acidic soils, whereas bacterial abundance is highest when the soil pH is near neutral (Strickland and Rousk, 2010). However, ash from woody mill wastes such as bark can also be enriched in heavy metals that can have negative microbial

* Correspondence to: Smithsonian Environmental Research Center, PO Box 28, Edgewater, MD 21037, USA.
E-mail address: noyceg@si.edu (G.L. Noyce).

http://dx.doi.org/10.1016/j.apsoil.2016.07.006
0929-1393/Published by Elsevier B.V.

effects (Kandeler et al., 1996; Müller et al., 2002; Singh et al., 2014). Though the microbial role in managed forest ecosystems is not completely understood, characterizing microbial responses is crucial for understanding if ash addition increases the overall health and productivity of the forest (Zak et al., 2003; van der Heijden et al., 2008).

In previous experiments, ash addition typically altered bacterial community composition, although effects on fungal community composition and overall microbial biomass were varied (Bååth et al., 1995; Mahmood et al., 2003; Perkiömäki et al., 2003; Perkiömäki and Fritze, 2005; Lupwayi et al., 2009). These effects were more pronounced in the top 5 cm of the mineral soil (Augusto et al., 2008; Lupwayi et al., 2009) and were apparent on both short and long time-scales (Fritze et al., 1994; Saarsalmi et al., 2012), but very few studies have investigated ash effects in the organic soil horizons, which are an important zone of nutrient cycling and microbial habitat in many forest ecosystems, including those on Canada's granitic Precambrian Shield. While there have been ongoing ash-addition experiments in northern European and South American forests, the variable microbial responses make it challenging to extrapolate to other forest types and few field-scale ash addition studies have been previously conducted in Canada. As with any soil amendment, variations in both the ash itself and soil characteristics at the experimental site alter overall ecosystem effects (Pitman, 2006; Rosenberg et al., 2010; Reid and Watmough, 2014). In addition, sustainable forest management policy in Ontario is predicated on silvicultural practices that emulate natural disturbances. As forest fires are the dominant natural disturbance in the boreal forest region and release ash and char, we wanted to determine if the effects of manual ash addition on the soil microbial community were broadly comparable to residual fire effects.

Here, we evaluated the responses of the organic layer and mineral soil microbial communities in two, distinct, ash-addition field trials. Though these two trials were not replicates of the same experimental design, both were established to assess effects of long-term, field-scale ash amendment in managed forested ecosystems in Ontario, Canada. Experiment 1, Island Lake, was a boreal softwood forest with a clear-cutting silvicultural system where four addition rates of bottom ash (0.7–5.7 t ha^{-1}) were applied. Experiment 2, Haliburton Forest, was a temperate hardwood forest with a partial-harvest silvicultural system where three addition rates of fly and bottom ash (1–8 t ha^{-1}) were applied. We used these two experiments to test the following hypotheses: (1) Ash addition will increase soil pH and concentrations of important base cations and consequently increase the relative abundance and diversity of soil bacteria compared to fungi; (2) The response of the soil microbial community will be strongest at the highest rate of ash application in each experiment; and (3) The response of the microbial community to ash addition in the boreal forest of Experiment 1 will be broadly similar to the response of the microbial community after forest fire.

2. Methods

2.1. Ash chemistry

All ash chemical analyses were conducted at the Soil and Plant Analytical Lab at the Great Lakes Forestry Center (Sault Ste. Marie, Ontario, Canada). Total C and N content of the applied ashes were determined using an NCS combustion analyzer (Vario EL III, Elementar Americas Inc, USA). After high temperature microwave acid digestion, an inductively coupled argon plasma (ICAP) spectrometer (Varian Analytical Instruments, USA) was used to measure P, S, K, Ca, Mg, Fe, Cu, Mn, Al, Na, S, As, Cd, Co, Mo, Ni, Pb, Se, and Sr, following EPA standard method 3052. Ash pH was determined in a slurry of distilled water.

2.2. Experiment 1: site description, experimental design, and soil sampling

The Island Lake Biomass Harvest Experiment is located in the Boreal forest region of northeastern Ontario (47.4°N, 83.4°W) (Kwiaton et al., 2014). The forest was a 40-year-old jack pine (*Pinus banksiana*) stand that was full-tree harvested during December 2010 and January 2011. As the harvest was designated to produce chips for bioenergy production, all traditionally non-merchantable trees were harvested in addition to sawlogs. Jack pine seedlings were replanted in May 2012. The soil is characterized by sandy, acidic Cambisols developed on glaciofluvial deposits with a Fibrimor forest floor averaging 10 cm. Soil chemistry is described in Table 1. Mean annual temperature (1981–2010) is 2.0 °C and mean annual precipitation (1981–2010) is 809 mm, of which 545 mm is rain and the rest is snow (Environment Canada, 2016). The average length of frost-free period is 93 days and the average growing degree days (GDD) above 10 °C is 719 (Environment Canada, 2016).

An incomplete block design was applied to 16 25 × 25 m plots at Island Lake in October 2011. The four treatment levels were set in relation to harvest removals of Ca; bottom ash was applied at 0.7, 1.4, 2.8 or 5.7 t dry ash ha^{-1}, which was the equivalent of adding 50, 100, 200, or 400 kg Ca ha^{-1} (Supplementary Table S1). An estimated 190 kg Ca ha^{-1} was removed during harvest. These bottom ash treatments are referred to as Ash 50, Ash 100, Ash 200, and Ash 400. There were four replicates of each treatment and all ash was evenly distributed on the soil surface by hand. The applied ash was generated from an older style (pinhole grate stoker boiler) thermal electricity-generating facility burning lumber mill residues. This ash had a high concentration of C, a pH of 10.2, and relatively low heavy metal concentrations. Detailed ash chemistry is provided in Table 2. An additional four plots with no added ash were used as controls. Soil cores were collected in June 2013, which was 20 months after ash application. Eight 5-cm soil cores were collected per plot, divided into the organic (O) horizon and the top 10 cm of the mineral soil (A and B horizons), pooled per plot, and frozen until analysis.

Six 5-cm soil cores were also collected in June 2013 from each of five transects along the site of a forest fire approximately 55 km northwest of Island Lake, following the same procedure. This 35-year old jack pine plantation boreal forest burned from May 27 to August 10, 2010, so these samples represented the soil chemical and microbial conditions 37 months after wildfire initiation. In total, the wildfire burned 2050 ha, killed all of the aboveground herbaceous vegetation, and partially consumed the Fibrimor forest floor horizons at the sampling site.

Table 1
Study site characteristics before ash addition. Values represent average characteristics in the top 20 cm of the soil, including the O, A, and B horizons.

	Island Lake[a]	Haliburton Forest[b]
pH (H_2O)	4.7	4.08
C (g kg^{-1})	87	78
N (g kg^{-1})	2.3	4.6
P (g kg^{-1})	0.05	0.7
K (g kg^{-1})	0.27	19.7
Ca (g kg^{-1})	0.48	17.7
Mg (g kg^{-1})	0.08	4.5

[a] Kwiaton et al. (2014).
[b] Pugliese et al. (2014).