Wood ash effect on chemical and microbiological properties of topsoil in a Norway spruce stand one year after the treatment

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Abstract

Gömöryová, E., Tóthová, S., Pichler, V., Homolák, M., Kriššák, V., Gömöry, D., 2016. Wood ash effect on chemical and microbiological properties of topsoil in a Norway spruce stand one year after the treatment. *Folia Oecologica*, 43: 156–163.

Changes of soil chemical and microbial characteristics in the forest floor and in the A-horizon caused by the application of wood ash (WA) on the soil surface were studied one year after the WA application. Soil sampling was performed at three plots in a 40-year-old Norway spruce stand in Central Slovakia – at the control plot (CP) and at the plots with WA application in the spring (P1) and the autumn (P2) 2010. Soil samples were taken from the litter (L), fermentation (F), humic (H) forest floor horizons and from the A-horizon (the depth of 0−10 cm). In soil samples chemical attributes (pH, C and N concentration, extractable C and base cations concentration) as well as microbial characteristics (microbial biomass C, basal respiration, catalase activity, richness and diversity of soil microbial functional groups) were analysed. WA application was reflected in the changes of chemical properties (soil reaction, C, N and Ca^{2+} concentration) only in the forest floor horizons but not in the A-horizon one year after the treatment. No significant differences between plots were found in microbial characteristics throughout the topsoil horizons. The most distinct differences in decreased C and N content compared to control plot were observed in the H-horizon. Soil acidity differed especially in the F-horizon, where the increase of pH-H_2O from 4.76 to 6.85 at P1 was recorded.

Key words

forest floor, forest soils, soil properties, wood ash

Introduction

In the last two decades, increasing number of studies on the effect of wood ash application on soil has appeared (Bååth et al., 1995; Demeyer et al., 2001; Liiri et al., 2002; Zimmermann and Frey, 2002; Jokinen et al., 2006; Lupwayi et al., 2009; Perruci et al., 2008; Saarsalmi et al., 2012). Wood ash (WA) is used as a liming agent in agriculture and forestry and its application on the soil has become a convenient way to recycle nutrient elements (Demeyer et al., 2001; Pitman, 2006).

The impact of WA application on soil can vary depending on the properties of WA (plant type used for burning, nature of the burn process, form of the ash), the conditions at the application site as well as other factors (Demeyer et al., 2001; Pitman, 2006). It is generally known that WA exhibits a high alkalinity and neutralizing capacity (Demeyer et al., 2001; Yusiharni and Gilkes, 2012) as a result of a large proportion of bases, especially Ca, Mg and K (Ozolinčius et al., 2005). Therefore, WA is recommended to be spread in forests to decrease soil acidification and nutrient deficiencies, and thus help to sustain long-term forest productivity in areas where acid deposition is high or whole-tree harvest has been practised (Meiwes, 1995; Jacobson, 2003). On the other side, WA may contain high concen-

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trations of heavy metals due to human activities, which can negatively affect plants and soil organisms. Especially fly ash as the lightest component can contain high concentrations of Cd, Cu, Cr, Pb and As, and therefore should not be used as a fertilizer (Pitman, 2006).

Addition of WA to soil can affect many soil properties, e.g. alkali metals content, soil salinity, soil structure, field capacity and soil aeration (Rumpf et al., 2001; Perruci et al., 2008). Especially the effect of WA on soil chemical properties is well documented (Meiwes, 1995; Saarsalmi, 2001; Ozolinčius, 2006). However, soil properties also directly and/or indirectly affect soil organisms including microorganisms and thus changes in the biomass, activity and composition of the microbial community are expected after WA application. The reports on the effects of WA on soil organisms are sometimes conflicting; WA may cause both increased and decreased microbial biomass, activity, community structure etc. (Bååth et al., 1995; Demeyer et al., 2001; Zimmermann and Frey; 2002, Bjørk et al.; 2010).

Studies about the impact of WA on soil mostly focus on the forest floor (surface organic horizon, the O-horizon) and the mineral top horizon (the A-horizon). Forest floor (FF) is an important component of forest ecosystems as it is a source of large amounts of nutrients and water for plants and soil organisms. It provides also habitat for many soil organisms and plant roots as well as a substrate for seedling establishment in forest ecosystems (Greiffenhagen et al., 2006; Šnajdr et al., 2008; Menšík et al., 2009). FF distinctly influences not only the rate of natural forest regeneration, but also the herb layer and adult trees, as their fine roots occur abundantly in this surface organic layer (Vance and Nadkarni, 1990).

The studies usually focus on the FF as a whole. However, FF is sometimes very thick especially in coniferous stands, and can be formed of several layers, which differ not only in morphology but also in their physical and chemical properties, water and temperature regimes etc. (Blume et al., 2010; Titeux and Delvaux, 2010; Gömöryová et al., 2013). This results in different ecological conditions for soil microorganisms and plant roots. In spite of this, studies dealing with the responses of soil properties, including microbiological attributes on WA addition in different FF layers are scarce. The aim of this study was thus to evaluate the influence of WA application on the changes in chemical and biological soil properties in particular layers of forest floor and the underlying mineral A-horizon in a field experiment. Such research is expected to improve our understanding of the processes undergoing in particular horizons after the treatments and the potential availability of nutrients for living organisms.

Material and methods

Study of the FF horizon changes after the WA application was performed in a 40-year-old Norway spruce (*Picea abies* (L.) Karst.) stand located in the central part of Slovakia (48º 35,006`N, 19º 36,283`E) at the altitude of 825 m asl. The mean temperature is 5 °C and annual precipitation 800 mm. The dominant soil type is Dystric Cambisol with a loam texture. In the forest floor, litter (L), fermentation (F) and humus (H) horizons with average thickness of 1.0, 1.5 and 1.5 cm, respectively, could be recognized.

Three plots with an area of 400 m^2 each situated on a very gently 5° slope oriented to S were established by the researchers from the National Forest Centre in Zvolen with the primary aim to monitor the impact of wood ash on the growth of spruce stand. The design of plots resulted from this primary objective:

- CP – control plot, without WA amendment
- P1 – WA (5 t ha^{-1}) was applied on the soil surface in the spring 2010
- P2 – WA (5 t ha^{-1}) was applied on the soil surface 6 months later in the autumn 2010.

Wood ash comes from the heating station in Hriňová (5 km from the study plot) which used chemically untreated wood (both deciduous and coniferous). WA not pelleted was carefully spread handly on the FF surface at P1 and P2. WA was not mixed with FF and plot was not irrigated to ensure conditions similar to those in usual forest management practice. All three plots were located next to each other to minimize the effects of soil spatial variability. As the slope is very gentle there was no risk of spreading ash onto the control plot.

At each plot soil sampling was done at three sampling points representing the vertex of an equilateral triangle with the distance of 15 m from the centre of triangle to each vertex. At each sampling point soil samples were taken from the distinct FF horizons (L, F, H) using a 0.25 × 0.25 m frame and the A-horizon from the depth of 0–10 cm using knife and scoop in the autumn 2011. Samples from distinct plot were not mixed together but each sample was analysed separately.

In the laboratory, each soil sample was divided into two parts. One part was stored in field-moist condition at 4 °C prior to microbial analyses. The other part used for chemical analyses was air-dried.

In air-dried samples, soil acidity, total carbon (C), total nitrogen (N), extractable carbon (C_{ext}) and base cations (Ca^{2+}, Mg^{2+}, K^+) concentration were determined. Soil reaction (pH-H_2O) was measured potentiometrically in water suspension (10 g samples from the L, F, H horizons and/or 20 g soil from the A-horizon plus 50 ml distilled water) after 24 h. VarioMacro CNS Analyser (CNS Version: Elementar, Germany) was used to determine soil C and N concentration. The extractable C was measured in 0.5 M K_2SO_4 extract and quantified by the oxidation with $K_2Cr_2O_7$/H_2SO_4 and subsequent titration with $Fe(NH_4)_2(SO_4)_2$. Exchangeable Ca^{2+}, Mg^{2+} and K^+ were estimated in 0.15 M NH_4Cl extract using atomic absorption spectrometry (GBC Avanta AAS).

In fresh soil samples, soil water content and sample dry weight were estimated gravimetrically by ovendrying soil at 105 °C for 24 h. Microbial biomass carbon (C_{mic}) was assessed according to ISLAM and WEIL (1998) using the microwave-irradiation procedure. After irradiation by microwaves, soil samples were extracted with 0.5 M K_2SO_4. C content in the extract was quantified the same way as C_{ext}. The same procedure was done with a non-irradiated sample. The microbial biomass carbon was then determined as $C_{mic} = (C_{irradiated} - C_{non\text{-}irradiated})/K_{ME}$, whereby extraction efficiency factor $K_{ME} = 0.213$.

Basal soil respiration (Resp) was measured by estimating the amount of CO_2 evolved during incubation of soil sample in a closed jar for 24 h (ALEF, 1991). CO_2 absorbed in a 0.05 M NaOH was determined by the titration with 0.05 M HCl using the phenolphthalein indicator after the precipitation of carbonates by $BaCl_2$. Catalase activity (Acat) was measured 10 min after 3% H_2O_2 was added to soil sample. The measurement is based on the volume of discharged oxygen based on the method of Khaziev (1976). The community-level metabolic profiles of soil microbial community were estimated using BIOLOG EcoPlates (INSAM, 1997). Briefly, fresh soil was resuspended in 0.9 % NaCl and supernatant was diluted $1:10^3$–10^4 depending on the microbial biomass. 150 ml of extract were incubated in microtitration plates at 37 °C during 7 days. Absorbance at 590 nm was recorded using the Sunrise Microplate Reader (Tecan, Salzburg, Austria). The richness (Richn) of the soil microbial community was assessed as the number of substrates with a non-zero response. For the estimation of functional diversity of the microbial community, Hill's diversity index (Div) was calculated (HILL, 1973).

Statistical analyses were done using the statistical package SAS/STAT (SAS, 2010). The variability of the soil characteristics between treatments and horizons was evaluated using two-way analysis variance; both horizon and treatment were considered fixed-effect factors. Pairwise differences of the means between the treatments were tested by Duncan´s test.

Results

One year after the treatment, the effect of WA application was reflected only in some soil properties (Table 1). Significant differences were found in soil reaction, C, N and Ca concentration. No changes were observed in microbial characteristics between the treatments. Almost all measured soil characteristics except for richness and diversity of microbial functional groups differed significantly between horizons. Treatment × horizon interactions were also significant in the case of soil reaction, N concentration and functional diversity.

Table 1. Analyses of variance of soil characteristics (significance of *F*-tests)

Factor	d.f.	pH	C	N	C/N	C_{ext}	Ca	Mg	K
Treatment	2	***	**	***	ns	ns	*	ns	ns
Horizon	3	***	***	***	***	***	***	***	***
Treatment *horizon	6	**	ns	*	ns	ns	ns	ns	ns
Error	24								

Table 1. Analyses of variance of soil characteristics (significance of *F*-tests) – continued

Factor	d.f.	C_{mic}	Resp	Acat	Richn	Div
Treatment	2	ns	ns	ns	ns	ns
Horizon	3	***	***	***	ns	ns
Treatment *horizon	6	ns	ns	ns	ns	*
Error	24					

Significance levels: ***P < 0.001, **P 0.001–0.01, *P 0.01–0.05; ns, non-significant.

C_{ext}, extractable carbon; C_{mic}, microbial biomass carbon; Resp, basal respiration; Acat, catalase activity; Richn, richness of soil microbial functional groups; Div, diversity of soil microbial functional groups.

WA application on the soil surface led to the decrease of acidity at the P1 and P2 in comparison to the CP (Table 2); the treated plots exhibit higher pH by approximately one unit of pH. Decreased acidity is observed in all horizons; however, the most distinct differences are found in F-horizon where pH increased from 4.76 to 6.85 at P1 (Fig. 1a). While at the CP plot pH decreases with the depth of FF profile, the treated plots do not exhibit the same pattern. One year after the WA application the highest pH is seen in the F-horizon and the smallest in the A-horizon.

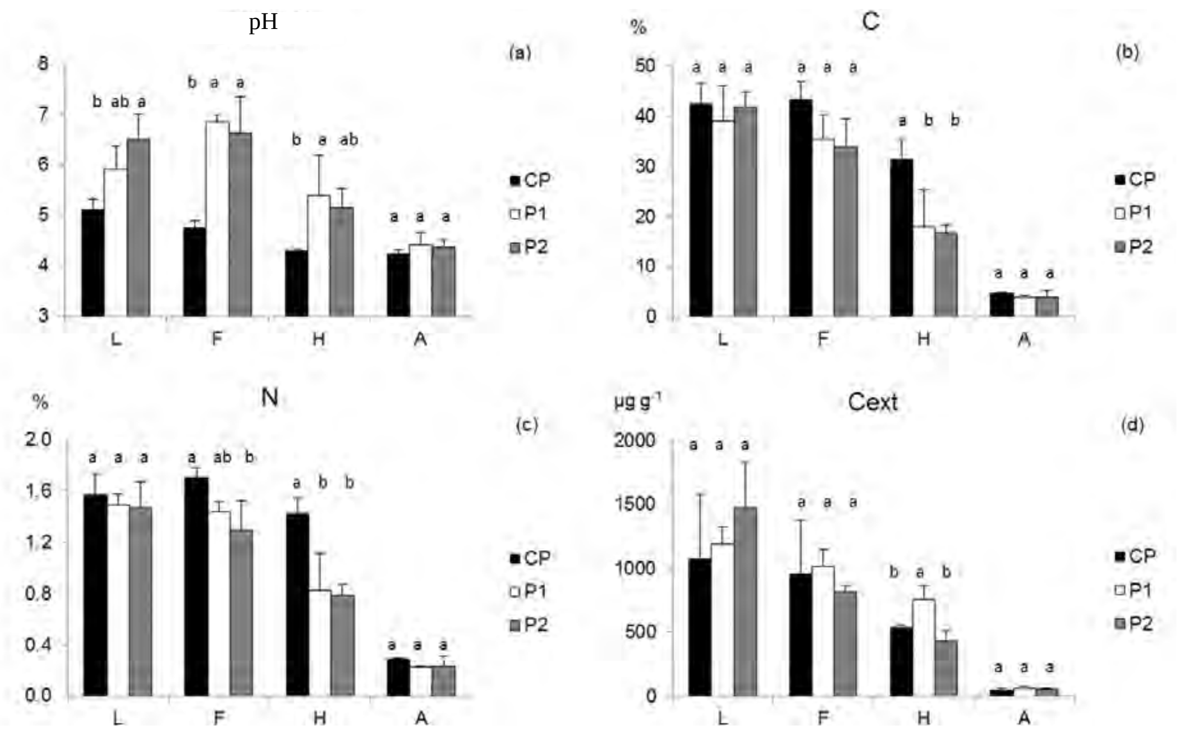
Table 2. Duncan's pairwise tests of the differences of means between treatments across horizons (treatments with the same capital letters do not differ significantly)

Treatment	pH	C	N	C/N	C_{ext}	Ca	Mg	K
		%	%		μg g^{-1}	mg kg^{-1}	mg kg^{-1}	mg kg^{-1}
CP	4.60 B	30.5 B	1.24 A	21.9 A	650 A	906 B	132.7 A	72.9 A
P1	5.64 A	24.1 A	0.99 B	23.5 A	755 A	1,176 A	124.9 A	74.9 A
P2	5.67 A	24.1 A	0.94 C	21.9 A	693 A	1,236 A	149.9 A	88.8 A

Table 2. Duncan's pairwise tests of the differences of means between treatments across horizons (treatments with the same capital letters do not differ significantly) – continued

Treatment	C_{mic}	Resp	Acat	Richn	Div
	mg g^{-1}	μg CO_2 $g^{-1}h^{-1}$	ml O_2 g^{-1} min^{-1}		
CP	12.66 A	98.4 A	5.16 A	7.5 A	4.4 A
P1	7.84 B	82.0 A	5.02 A	6.4 A	4.2 A
P2	10.76 AB	86.5 A	5.28 A	7.1 A	4.7 A

C_{ext}, extractable carbon; C_{mic}, microbial biomass carbon; Resp, basal respiration; Acat, catalase activity; Richn, richness of soil microbial functional groups; Div, diversity of soil microbial functional groups.

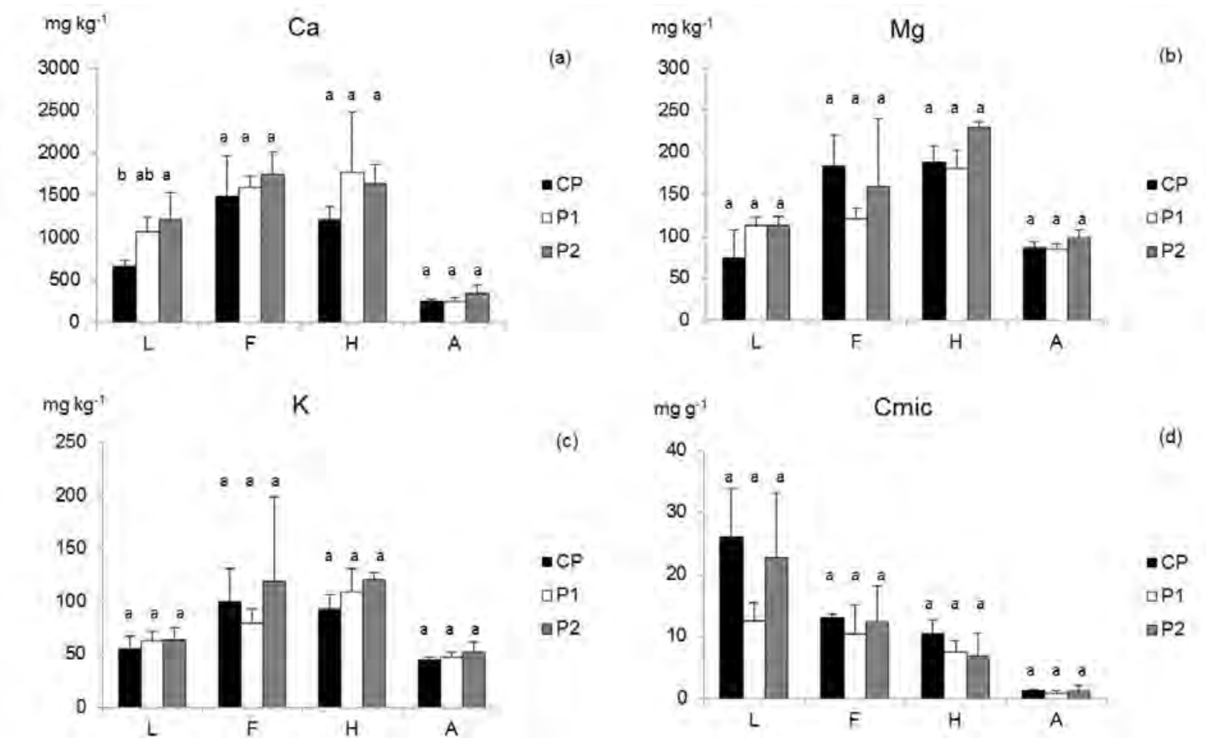


C_{ext}, extractable carbon.

Fig 1. Means and standard deviations of soil acidity (a), carbon (b), nitrogen (c) and extractable carbon concentration (d) sorted by treatments (CP, control plot; P1, plot with wood ash addition in spring 2010; P2, plot with wood ash addition in autumn 2010) in distinct soil horizons (L, F, H, A).

Application of WA caused a significant decrease in C and especially N concentration in the FF horizons of the P1 and P2 (Table 2). The differences in C and N concentration between CP on one side and treated plots on the other side increase down the horizons profile with maximal difference in the H-horizon where C and N concentration at P1 and P2 represents the half of that concentration at CP (Fig. 1b and c). The highest C_{ext} concentration occurs in L-horizon and with a depth its amount decreases. The differences between treatments are non-significant due to high within-treatment variability (Table 2, Fig 1d).

Among the base cations only the Ca concentration showed a significant difference between the treatments with higher values at the plots with WA application; in the case of Mg and K no pattern could be identified (Table 2, Fig. 2a–c). The highest concentration of base cations was found in the F and H horizons at all plots.



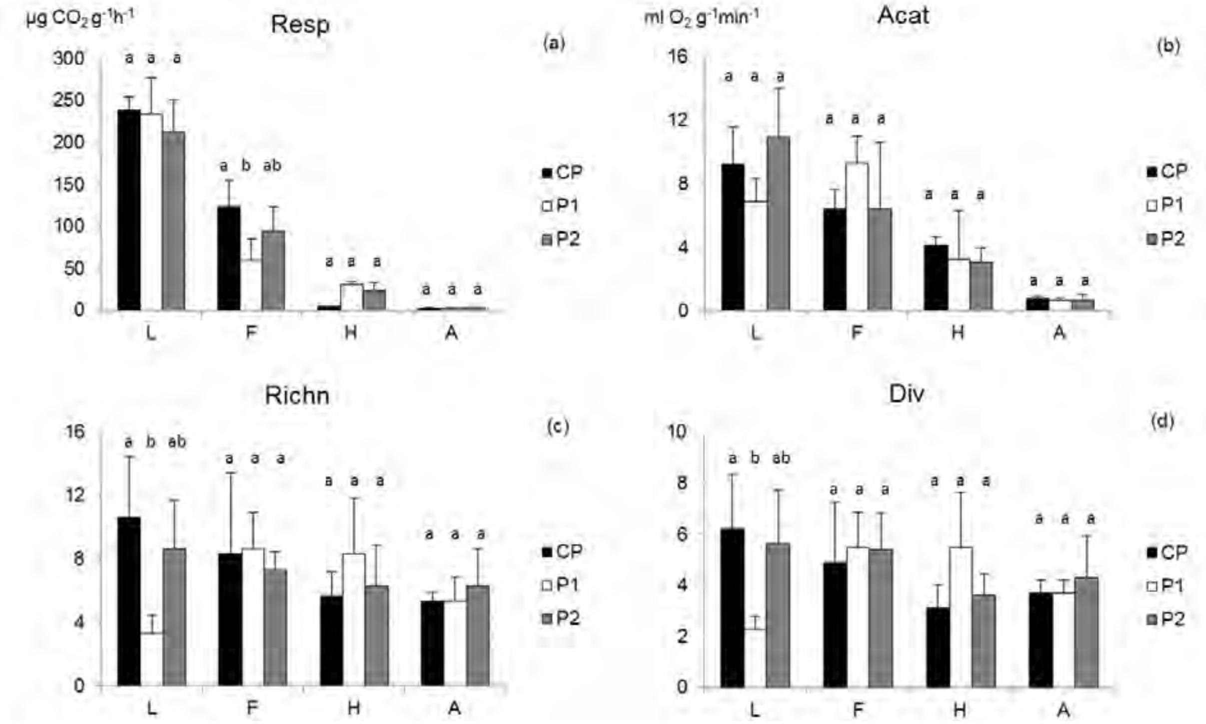
C_{mic}, microbial biomass carbon.

Fig 2. Means and standard deviations of calcium (a), magnesium (b) and potassium concentration (c) and microbial biomass carbon (d) sorted by treatments (CP, control plot; P1, plot with wood ash application in spring 2010; P2, plot with wood ash application in autumn 2010) in distinct soil horizons (L, F, H, A).

All microbial characteristics exhibited high within-treatment variability (Fig. 2d, 3a–d). While microbial biomass C, basal respiration and catalase activity decreased down the soil horizons profile, characteristics of microbial community structure did not differ significantly between horizons. No pattern in microbial characteristics between treatments throughout the FF and A- horizons was identified.

Discussion

It is known that WA application leads to the decrease of soil acidity due to the input of large base cations amount contained in the WA. The extent of such changes varies distinctly depending, for instance, on the WA dose, time after the treatment or depth of the soil. Ozolinčius et al. (2006) found that pH increased from 3.45 to 6.15, which means 2.7 pH units 4 months after WA application in FF, Levula et al. (2000) observed a pH increase up to 2 pH units 5 months after WA application of 5 t ha^{-1}, Saarsalmi et al. (2001) found a decrease of soil acidity by 0.2–2.4 pH units in FF 1–19 years after the WA addition. In our study treated plots showed decrease in soil acidity against the CP. The most distinct difference by 2 pH units was observed in the F-horizon. As already mentioned, FF is not homogeneous and its thickness can vary distinctly. Changes in pH throughout the topsoil can thus be influenced by the properties of distinct FF horizons on one side and the water amount coming into the soil on the other side. The fact that water infiltrated and percolating through the FF profile gradually depletes in nutrients top layers and enriches the lower horizons is reflected also in pH values as illustrated in the Fig. 1a. While the differences in soil reaction between the L- and F-horizon are small one year after the treatment (CP2), half year later the L-horizon is already more acid than the underlying F-horizon (CP1). However, in the A-horizon no changes have been observed yet. Ozolinčius et al. (2006); Saarsalmi et al. (2001) showed that the WA effect on pH in the mineral soil was less pronounced in short-term and appeared much later than in forest floor. Distinct changes in mineral horizons are often observed several years after the treatment. They found that mineral soil layers (below 10 cm depth) showed very little change at



Resp, basal respiration; Acat, catalase activity; Richn, richness of soil microbial functional groups; Div, diversity of soil microbial functional groups.

Fig 3. Means and standard deviations of basal respiration (a), catalase activity (b), richness (c) and diversity (d) of soil microbial functional groups sorted by treatments (CP, control plot; P1, plot with wood ash application in spring 2010; P2, plot with wood ash application in autumn 2010) in distinct soil horizons (L, F, H, A).

7 years after ash application, but after 16 years, the increased pH values at depth indicated a slow downward transfer of activity from the topsoil over time. However, the thickness of the FF layer has a significant role as a thin O-horizon has lower capacity to hold cations applied in ash than a thick horizon.

WA is a direct source of the major elements such as P, Ca, Mg and K in soils. Among base cations only Ca concentration differed significantly between treatments at our plots, which was in agreement with Ozolinčius et al. (2006), who found that in the case of loose ash, leaching of base cations had mostly occurred during the first year after WA addition. It can be associated with the solubility of particular base cations. According to Khanna et al. (1994) the major elements can be divided into three groups based on the solubility: K (dissolves very quickly) – Ca and Mg – P (remains relatively insoluble); magnesium is more soluble than calcium (Eriksson, 1998). As K and Mg are more soluble than Ca they can be leached more rapidly from the topsoil (Ozolinčius et al., 2006).

Beside the decreased soil acidity and increased base cations content, WA application can be also reflected in the changes of decomposition rate and consequently C and N content (Demeyer et al., 2001; Maljanen et al., 2014). There are many contradictory results regarding organic matter content after the WA application. Saarsalmi et al. (2001) did not find any changes in the C and N content either in organic or mineral horizons 7 years after the treatment. On the other side, Demeyer et al. (2001) stated that WA could lead to a decrease of total C and N because of the increase in the solubility of organic C and nitrification. Contradictory results can be caused by different time of soil sampling after the treatment and also by different C and N content in soil before the WA addition. According to Rosenberg et al. (2010), WA application onto the soils rich on N can significantly influence C and N cycle while at N-poor sites the presence of WA was not reflected in the C and N content and processes associated with it. In our study, WA application onto the soil surface provoked a decrease in C and N content in distinct FF horizons. Increased differences against the control plot were found down the FF profile from L- to H-horizon; in the A-horizon the difference was again minimal. Increase of pH and the base cation content down the FF profile led to increased mineralisation of organic matter and thus C and N losses. According to Pitman (2006), WA application generally has little direct impact on N availability due to its low levels in the raw material (<0.1 per cent). However, N availability often increases indirectly as a result of ash application due to a rise in soil pH, base cation content and consequent N-mineralization.

We did not find any changes in microbial characteristics between plots. Similarly, Zimmermann and Frey (2002) found that while the effect of WA on chemical properties was long-term, in the case of microbial characteristics the effect was only short-term. On the other side, Rosenberg et al. (2010) observed increased CO_2 evolution still 12 years after the WA application. In a number of studies decrease as well as increase in microbial biomass, respiration, N-mineralisation, differences in the fungi:bacteria ratio were observed (Bååth at al., 1995, Demeyer at al., 2001; Aronsson and Ekelund, 2004; Jokinen et al., 2006; Ozolincius et al., 2006; Perruci et al., 2008). The observed results documented the complexity of microbial processes and their different responses to the changed environment.

Conclusions

Wood ash application on the soil surface in a 40-year-old spruce stand led to a decrease in soil acidity and C and N concentration in particular forest floor horizons but no significant changes were found in the underlying mineral A-horizon still one year after the treatment. The extent of the changes in distinct horizons was not uniform. The most pronounced WA effect on pH was observed in the F-horizon; in the case of C and N concentration the most distinct differences were found in the H-horizon. Unlike chemical properties, no responses of microbial community to the WA addition were observed in the FF and A-horizons. Six-month temporal shift in the WA application did not lead to significant differences in soil characteristic between the treated plots. Changes in soil chemical properties throughout the forest floor profile are vertically differentiated, which needs to be considered when the processes in forest soils and habitat changes for soil organisms and plant roots occupying these particular layers are studied.

Acknowledgement

We thank T. Dvorská and Ž. Brnáková for laboratory assistance. This study was financed by the projects VEGA 1/0761/14, APVV-15-0176 and the Centre of Excellence „Wood ash application in forestry and evaluation of its effects“, ITMS: 26220220016, supported by the Research & Development Operational Programme funded by the ERDF.

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Received February 17, 2016
Accepted March 23, 2016