JOURNAL OF FOREST SCIENCE, *61*, 2015 (12): 544–550

doi: 10.17221/101/2015-JFS

Acid phosphomonoesterase activity as affected by salicylic acid and its relation to selected biochemical characteristics in soils of Norway spruce stands

S. Kočvarová, J. Pecháček, K. Rejšek, V. Vranová

Faculty of Forestry and Wood Technology, Mendel University in Brno, Brno, Czech Republic

ABSTRACT: The main aim of the study was to determine extracellular acid phosphomonoesterase activity in soil with and without addition of salicylic acid. Some other important chemical and physical features of soil were also investigated including the content and seasonal dynamics of bioavailable phosphorus and water soluble carbohydrate contents. Soil samples were collected at a depth of 5 cm (H horizon) at the Rájec-Němčice Ecosystem Station (southern Moravia, Czech Republic) in three differently managed Norway spruce pure stands. No correlation was found between the acid phosphomonoesterase activity and phosphorus content. The addition of salicylic acid reduced the acid phosphomonoesterase activity compared to soil without any substrate added.

Keywords: phosphorus; carbohydrates; 2-hydroxybenzoic acid; *Picea abies*

Phytotoxicity, autotoxicity, general toxicity or antimicrobiality of substances entering soils naturally as a part of root exudates or as a result of decomposition processes certainly affect also the activity of soil enzymes. Phosphomonoesterases (PME) are important enzymes that are closely related to the phosphorus (P) cycle in the nature. Phosphorus that occurs in plant tissues in inorganic phosphate form is a significant controlling agent in photosynthesis and metabolism of carbohydrates. Phosphomonoesterases may have an important role due to phosphorus supply, especially in terrestrial ecosystems (Rejšek 1991; Garcia-Ruiz et al. 2000). The absence of phosphorus (Nieminen, Penttilas 2004) often stimulates the release of extracellular phosphomonoesterase from plant roots, fungi and other microorganisms (Šarapatka 2003). The enzymes such as phosphomonoesterases are able to hydrolyze phosphorus in its organic form to other forms, easily available to plants. The activity of phosphomonoesterases can be a very good biological or pollution indicator (Grierson Adams 2000; Holišová 2008; Rejšek et al. 2012). These enzymes often occur as linked to dormant cells, in unbroken dead cells or in their fragments. We can identify them as a part of soil aggregates held by physical or chemical forces.

In forest soils, carbohydrates are amongst the more rapidly degraded compounds of plant litter (Traversa et al. 2008). Vranová et al. (2013) reviewed comprehensively the sources, roles and the importance of soil carbohydrates. Considering carbohydrates in forest soils, Ussiri and Johnson (2003), Stevenson et al. (2004), Allard (2006) and Ring et al. (2015) provided data on the specificity of the environment, Zhang et al. (1999) and Jolivet et al. (2006) reported on carbohydrates and afforestation after clear-cutting; distinctive features of carbohydrate fractions in Norway spruce stands were pointed out by Rosenberg et al. (2003). Ball et al. (1996) demonstrated the effects of ecosystem management on carbohydrate quantity, quality and distribution in soils, and Ro-

Supported by Project No. 55/2013 of Mendel University in Brno.

vira and Vallejo (2002) as well as Bending et al. (2004) described carbohydrates as biochemical indicators of soil organic matter quality.

The possible phytotoxicity of salicylic acid (2-hydroxybenzoic acid) has been a phenomenon studied for a long time (Popova et al. 1997; Mauch et al. 2001). It enters the soil either via plant root exudates (Asao et al. 2003) or as a product of dead organic matter decomposition (Vranová et al. 2013). In soil, salicylic acid is a part of humic compounds (Cardoza et al. 2004). Salicylic acid then affects decomposition processes in soil as well as growth and development of plants. It is a known fact that the activity of this acid is specifically conditioned by its sorption to soil colloids; generally speaking, it depends on forest type, soil type, soil characteristic and soil depth. Salicylic acid plays a role in the regulation of plant oxidative stress (Radwan 2012) and thus also in the induction of plant defence mechanisms. If a plant is threatened by a pathogen or affected by a disease, salicylic acid will help it to resist the infection. The plant starts to accumulate salicylic acid to stop the infection (Guo et al. 2009; Delaney 2010). The acid is also important to balance osmotic and salt stress (Eraslan et al. 2007).

The cultivation of spruce in monocultures is associated with decreasing increment and site regeneration, caused, among other things, by the input of phytotoxic substances into soil. Spruce is a potential producer of a number of phytotoxic, toxic and antimicrobial substances including amino acids, phytohormones and terpenes as well as phytotoxic proteins and dipeptides. Produced by shallow-rooted Norway spruce, the effect of exuded salicylic acid can be studied in the surface H-horizons. Autotoxicity and phytotoxicity of soil cause slow regeneration of forest sites as the phytotoxic substances in soil induce imbalances in tree nutrition, growth inhibition and alteration of metabolic pathways. Phytotoxically active hormones in soil and surface humus obstruct seed germination and growth of young plants including the formation of mycorrhizal symbioses.

Firstly, the paper is aimed at characterization of differences between the PME activities of the tested forest stands and presentation of the relations to both water soluble carbohydrates and bioavailable phosphorus content. Secondly, the authors focused on the seasonal dynamics of both PME and bioavailable phosphorus contents and, thirdly, the interrelationships between the PME activities and the forest soils affected by the addition of salicylic acid when the interactions of salicylic acid and PME were analysed in an organic horizon. For such goals, several hypotheses were tested. Firstly, the authors tested a negative correlation between the PME activity and bioavailable phosphorus content. Secondly, the authors tested a negative correlation between the addition of salicylic acid and the PME activity when the enzyme activities relate to forest management practices and forest stand age.

MATERIAL AND METHODS

Study site. Research was conducted at the Rájec-Němčice Ecosystem Station, situated near Němčice (about 3 km north), the Czech Republic (49°26'31''N, 16°41'30''E), with H horizon thickness of about 6 cm. From the viewpoint of timber production and richness the site is acidic, affected by Norway spruce monocultures. Typical of this environment is the fir-beech forest altitudinal zone (5^{th} FAZ) with forest site 5S1 [*Abieto-Fagetum mesotrophicum* with *Oxalis acetosella* (4AB3 – *Fageta quercino abietina*, the alliance *Luzulo-Fagion*, the association *Luzulo-Fagetum*)] situated at an altitude of 600–660 m a.s.l. The average annual temperature in 2007–2009 was 9.24°C and annual precipitation reached 717 mm (Menšík 2007, 2009). The study plots were established in 1968 by the Institute of Forest Ecology, Mendel University in Brno, in the framework of the UNESCO International Biological Program (IBP) and the UNESCO program "Man and the Biosphere" (MAB) (Fabiánek et al. 2009). The experiment covers three plots with different silvicultural management. Three pure Norway spruce stands with different silvicultural management (Rosík et al. 2013) were selected: 108-years-old mature stand (MSS), 36-years-old stand thinned from above (STFA) and 36-years-old stand thinned from below (STFB). The major soil type is classified as Dystric Cambisol (IUSS Working Group WRB 2006), Eumoder humus form (AFES 1998), on the felsic granodiorite where pH/H_2O in A-horizon is 3.8–4, and pH/H_2O in O horizon is 3.8 (Menšík 2010).

Sampling and biochemical analyses. Analyses were focused on enzymatic activity in H horizon and finding a relationship between enzymatic activity, bioavailable P content according to Egnér et al. (1960), and amount of water soluble carbohydrates. Measurements were carried out at monthly intervals for six months over the growing season May–October 2013. From each of the selected plots, three subsamples were randomly collected and homogenized into one. In total, nine (homogenized) samples were collected for this purpose, three from each study plot. For detection of the acid PME activity, fresh soil was incubated in succinate-borate buffer, pH 4.8 with *p*-nitrophenyl phosphate as (*p*-NPP) a substrate. The sample was placed into a 100-ml Erlenmeyer flask and 12 ml of the p-NPP solution was added. The reaction mixture was shaken and kept at 37°C for 1 hour. The absorbance was set to 410 nm. The activity of acid PME was calculated per 1 g of dry soil. The analysis of PME activity after addition of salicylic acid was performed in the same way, however, with addition of 1 mg salicylic acid before incubation. For detection of water soluble carbohydrates, fresh soil with demineralized water was shaken for 20 min until the soil solution was obtained. After that, the samples were incubated at room temperature for 10 min with 4 ml anthrone reagent and with absorbance set to 625 nm.

Statistical data analysis. Several data analyses were performed in order to determine the relations between the measured parameters. Our presumption was that we will be able to confirm a correlation between them. Statistical data processing was done by R statistical software and STATISTICA 11 (SPSS, Tulsa, USA).

RESULTS AND DISCUSSION

The bar graph (Fig. 1) shows the dynamics of acid PME activity in soil samples without added salicylic acid throughout the whole vegetation season. The highest activity was measured in samples collected during June and October. Fig. 1 is based on ANOVA for all cumulative differences in all three stands between individual months and the presented mean values demonstrate the statistically significant differences (the highest PME activity determined in June) only for two cases: (*i*) between June and May, and (*ii*) between June and September. The overall lower values were measured in STFB. Noticeably higher PME activity was observed in spring and autumn. The absence of correlation between PME activity and phosphorus content is demonstrated by the phosphorus regression line.

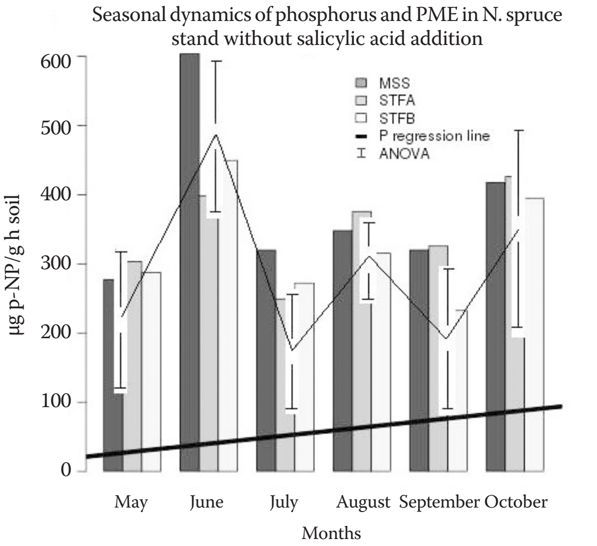

Fig 1. Comparison of the PME mean value without salicylic acid addition and regression line of phosphorus in individual stands during the vegetation season

Fig. 2 shows progression in the case of soil with added salicylic acid. We can see the lowest PME activity on all study plots in June. Generally, the largest difference between the measured values was recorded in July, when the mature stand had twice higher PME activity compared to the young stand thinned from below. With statistical significance and regardless of the type of thinning and stand age, June and August reached the lowest reaction on all study plots. Extracellular acid phosphomonoesterase showed the highest activity in

Table 1. Pearson correlation test – monthly progression 2013

	MSS			STFA			STFB		
	PME vs. P	PME vs. CH	PME vs. DM	PME vs.P	PME vs. CH	PME vs. DM	PME vs. P	PME vs. CH	PME vs. DM
May	NA	–0.5448	–0.7462	NA	0.2160	0.8707	NA	–0.2551	0.8511
Jun	–0.9563	0.9151	–0.9965	0.8414	0.2799	–0.9039	0.7222	–0.3698	–0.5690
Jul	0.7356	0.3564	–0.5643	–0.3121	–0.9748	0.4246	0.9548	0.2113	0.8099
Aug	0.5940	–0.0551	–0.2290	0.7751	0.6164	–0.9959	0.9820	–0.0908	–0.3400
Sep	NA	0.2450	–0.8979	NA	–0.8606	0.0607	NA	0.9473	–0.9379
Oct	–0.2010	0.9606	–0.3416	–0.1038	–0.8166	0.3338	–0.4312	–0.2489	0.9217
GS	–0.7411	0.1933	–0.5873	0.0523	–0.0228	–0.2632	–0.2658	0.0296	–0.4658

MSS – 108-years-old mature stand, STFA – 36-years-old stand thinned from above, PME – acid phosphomonoesterase, NA – data not available, CH – carbohydrates, DM – dry mass, STFB – 36-years-old stand thinned from below, GS – growing season

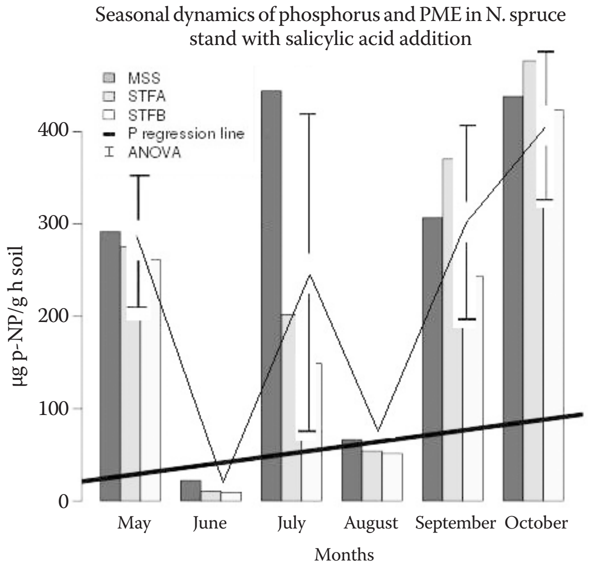

Fig. 2. Comparison of the PME mean value with added salicylic acid and regression line of phosphorus in individual stands during the vegetation season

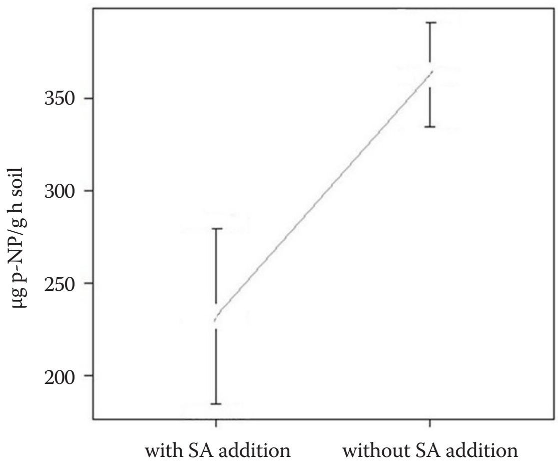

Fig. 3 ANOVA – statistical significance of the effect of salicylic acid addition on PME activity (pooled data from all three study plots and all months included)

all three stands during October. The total effect of salicylic acid addition was that it reduced PME activity (Figs 1 and 2) and induced its greater fluctuations during the vegetation season compared to the experiment without any added substrate. A phosphorus linear model (Figs 1 and 2) shows an increasing trend, but there is no statistically significant correlation between P forms and phosphomonoesterase (Table 1).

Fig. 3 demonstrates that the addition of salicylic acid significantly decreases the PME activity, based on a comparison of the data obtained from the three study plots tested. According to ANOVA performed with pooled data from all three study plots and all months included, there is a statistically significant difference in the PME activity between the soil without added salicylic acid and the soil with salicylic acid addition (Fig. 3).

Table 1 shows the individual parameters in relation to acid PME and development during the growing season 2013 (from May to October), where the last line expresses the correlation for which the data for the whole growing season were pooled. Based on the annual summary, the correlation between the studied parameters has not been proven.

Experiments were situated in Norway spruce monocultures with different age and type of management. Based on the data from all the study plots for the entire length of the vegetation period, both thinned young stands manifested higher PME activity than the mature stand. It seems that the values correlate the most in June in the MSS. Table 2 compares the mean values, standard deviations and standard errors in all three Norway spruce stands: the PME activity is compared with each of the other measured parameters.

As regards the phytotoxic effect of salicylic acid, it has been confirmed that it shows statistically significant seasonal dynamics with the lowest values measured in June. There are no statistically detectable differences between three Norway spruce stands in the content of bioavailable phosphorus in their soils tested. In the case of carbohydrates, the differences were statistically detectable only in July and September. The addition of phytotoxic substance had a higher (statistically non-significant) effect on PME activity in the mature stand; the strongest effect of salicylic acid was observed

Table 2. Standard error and standard deviation of measured parameters in Norway spruce stands during 2013

	PME	Phosphorus	Carbohydrates	Dry mass
MSS				
Mean	381.2324	59.8658	242.1617	0.5338
SD	123.0225	18.9094	71.3550	0.1245
SE	28.9967	5.4587	16.8185	0.0293
STFA				
Mean	346.4079	51.1460	275.9380	0.5721
SD	74.9915	33.1708	72.6097	0.1035
SE	17.6757	9.5756	17.1143	0.0244
STFB				
Mean	325.2815	107.7585	288.1192	0.5679
SD	93.5482	97.0166	71.4874	0.1007
SE	22.0495	28.0063	16.8498	0.0237

MSS – 108-years-old mature stand, STFA – 36-years-old stand thinned from above, STFB – 36-years-old stand thinned from below, SD – standard deviation, SE – standard error in all three stands in July and September. The phytotoxic effect of salicylic acid on PME in the mature stand was statistically non-significantly lower compared to the young stands. Fig. 2 proves that both young stands showed the lower PME activity. The addition of salicylic acid is always reflected in a decrease of PME activity; however, the obtained results did not confirm a statistically significant trend of this relation.

Most of the studies focused on enzymatic activity were conducted in top horizons, because of the high microbial biomass concentration (Herold et al. 2013). The presented results show a comparison between the basal PME and its change after salicylic acid addition, and proved a negative effect of the salicylic acid addition on the enzymatic activity. This acid is able to inhibit some enzymatic activity (Chen et al. 2003). Sahu et al. (2010) discovered that a low concentration of salicylic acid stimulates phosphate uptake by plants. Organic acids act as desorbents of acid phosphomonoesterase (Zhang et al. 2003) from mineral colloids.

Kunito et al. (2012) were concerned with both PME activity and interrelationships between PME activity and soil microorganisms. They demonstrated that PME generally shows higher activity at low pH (4.0–5.0). In our study, with the soil pH of 6.5, no significant correlation was found between enzymatic activity and bioavailable P content. Kunito et al. (2012) also observed that phosphorus limitation in microorganisms leads to high phosphomonoesterase activity in acid forest soils. Keefer (2000) stated that optimum P availability for Norway spruce is at pH between 5.0 and 6.0. Our results demonstrate that more acid forest soils have a lower optimum pH and higher activity of phosphomonoesterase.

Salicylic acid decreases enzymatic activity (Wang et al. 2009). The monthly oscillation was proved by Criquet et al. (2004). The obtained results have shown a similar character of the seasonal dynamics of PME activity. From the biological point of view, it is possible to compare the soil under mature and young stands. Silvicultural treatments may affect the quantitative parameters of soil biological activity but not directly its qualitative parameters (Kočvarová 2014).

CONCLUSIONS

The seasonal dynamics of phosphorus content and PME activity in spruce monocultures differing in age and type of thinning were determined at monthly intervals. Salicylic acid, as the added substance, displays a verifiable effect on the rate of enzymatic activity. This effect is negative – for all the study plots, this study confirmed significant differences in PME activity with the addition of salicylic acid during the vegetation season. The linear curve of phosphorus content shows a rising trend but no correlation with the PME activity has been proved. There was neither evidence nor any statistically significant correlation between the activity of PME and both water soluble carbohydrates and bioavailable phosphorus content.

References

AFES (1995): Référentiel pédologique. Principaux sols d'Europe. Paris, INRA: 332.

Allard B. (2006): A comparative study on the chemical composition of humic acids from forest soil, agricultural soil and lignite deposit: bound lipid, carbohydrate and amino acid distributions. Geoderma, 130: 77–96.

Asao T., Hasegawa K., Sueda Y., Tomita K., Taniguchi K., Hosoki T., Pramanik M.H.R., Matsui Y. (2003): Autotoxicity of root exudates from taro. Scientia Horticulturae, 97: 389–396.

Ball B.C., Cheshire M.V., Robertson E.A.G., Hunter E.A. (1996): Carbohydrate composition in relation to structural stability, compatibility and plasticity of two soils in a long-term experiment. Soil and Tillage Research, 39: 143–160.

Bending G.D., Turner M.K., Rayns F., Marx M.C., Wood M. (2004): Microbial and biochemical soil quality indicators and their potential for differentiating areas under contrasting agricultural management regimes. Soil Biology and Biochemistry, 36: 1785–1792.

Chen Q.X., Liu X.D., Huang H. (2003): Inactivation kinetics of mushroom tyrosinase in the dimethyl sulfoxide solution. Biochemistry (Moscow), 68: 644–649.

Criquet S., Ferreb E., Farnet A.M., Le Petit J. (2004): Annual dynamics of phosphatase activities in an evergreen oak litter: influence of biotic and abiotic factors. Soil Biology and Biochemistry, 36: 1111–1118.

Egnér H., Riehm H., Domingo W.R. (1960) Untersuchungen über die chemische Bodenanalyse als Grundlage für die Beurteilung des Nährstoffzustandes der Böden. II. Chemische Extraktionsmethoden zur Phosphor- und Kaliumbestimmung. Kungl-Lantbrukshögsk Annalls, 26: 199–215.

Eraslan F., Inal A., Gunes A., Alpaslan M. (2007): Impact of exogenous salicylic acid on growth, antioxidant activity and physiology of carrot plants subjected to combined salinity and boron toxicity. Scientia Horticulturae, 113: 120–128.

Garcıa-Ruiz R., Hernández I., Lucena J., Niell F.X. (2000): Significance of phosphomonoesterase activity in the regeneration of phosphorus in a meso-eutrophic, P-limited reservoir. Soil Biology and Biochemistry, 32: 1953–1964.

Grierson P.F., Adams M.A. (2000): Plant species affect acid phosphatase, ergosterol and microbial P in a Jarrah (*Eucalyptus marginata* Donn ex Sm.) forest in south-western Australia. Soil Biology and Biochemistry, 32: 1817–1827.

Guo T., Wang M., Shen H. (2009): Progress of studies on effect of AMF inoculation on vegetable quality. China Vegetables, 6: 1–6.

Herold N., Dietrich T., Grigsby W.J., Franich R.A., Winkler A., Buchelt B., Pfriem A. (2013): Effect of maleic anhydride content and ethanol dilution on the polymerization of furfuryl alcohol in wood veneer studied by differential scanning calorimetry. BioResources, 8: 1064–1075.

Holišová P. (2008): Aktivita kyselé fosfomonoesterázy v půdě rozdílně obhospodařovaných lučních a lesních ekosystémů Moravskoslezských Beskyd. [Master Thesis.] Brno, Mendel University in Brno: 57.

IUSS Working Group WRB (2006): World Reference Base for Soil Resources 2006. World Soil Resources Reports No 103. Rome, FAO: 145.

Jolivet C., Angers D.A., Chantigny M.H., Andreux F., Arrouays D. (2006): Carbohydrate dynamics in particle-size fractions of sandy spodosols following forest conversion to maize cropping. Soil Biology and Biochemistry, 38: 2834–2842.

Keefer R.F. (2000): Handbook of Soils for Landscape Architects. New York, Oxford University Press: 272.

Kočvarová S. (2014): Activity of Extracellular Acid Phosphomonoesterase of the Soil and the Roots in Differently Managed Norway Spruce Stands. [Master Thesis.] Brno, Mendel University in Brno: 63.

Kunito T., Tobitani T., Moro H., Toda H. (2012): Phosphorus limitation in microorganisms leads to high phosphomonoesterase activity in acid forest soils. Pedobiologia, 55: 263–270.

Mauch F., Mauch-Mani B., Gaille C., Kull B., Haas D., Reimmann C. (2001): Manipulation of salicylate content in *Arabidopsis thaliana* by the expression of an engineered bacterial salicylate synthase. The Plant Journal, 25: 67–77.

Menšík L. (2007): Ekologické aspekty transformace smrkových monokultur. [Master Thesis.] Brno, Mendel University in Brno: 103. (in Czech)

Menšík L. (2010): Frakcionace humusových látek lesních půd. [Ph.D. Thesis.] Brno, Mendel University in Brno: 210.

Menšík L., Fabiánek T., Tesař V., Kulhavý J. (2009): Humus conditions and stand characteristics of artificially established young stands in the process of the transformation of spruce monocultures. Journal of Forest Science, 55: 215–223.

Nieminen M., Penttilä T. (2004): Inorganic and organic phosphorus fractions in peat from drained mires in northern Finland. Silva Fennica, 38: 243–251.

Popova L., Pancheva T., Uzunova A. (1997): Salicylic acid: Properties, biosynthesis and physiological role. Bulgarian Journal of Plant Physiology, 23: 85–93.

Radwan D. (2012): Salicylic acid induced alleviation of oxidative stress caused by clethodim in maize (*Zea mays* L.) leaves. Pest Biochemical Physiology, 102: 182–188.

Rejšek K. (1991): Acid phosphomonoesterase activity of ectomycorrhizal roots in Norway spruce pure stands exposed to pollution. Soil Biology and Biochemistry, 23: 667–671.

Rejšek K., Vranová V., Pavelka M., Formánek P. (2012): Acid phosphomonoesterase (E.C. 3.1.3.2) location in soil. Journal of Plant Nutrition and Soil Science, 175: 196–211.

Ring E., Högbom L., Nohrstedt H.Ö., Jacobson S. (2015): Soil and soil-water chemistry below different amounts of logging residues at two harvested forest sites in Sweden. Silva Fennica, 49. Article ID 1265.

Rosenberg W., Nierop K.G.J., Knicker H., De Jager P.A., Kreutzer K., Weiss T. (2003): Liming effects on the chemical composition of the organic surface layer of a mature Norway spruce stand (*Picea abies* [L.] Karst.), Soil Biology and Biochemistry, 35: 155–165.

Rosík J., Fabiánek T., Marková I. (2013): Soil CO_2 efflux in young Norway spruce stands with different silviculture practices. Acta Universitatis Agriculturae et Silviculturae Mendelianae Brunensis, 61: 1845–1851.

Rovira P., Vallejo V.R. (2002): Labile and recalcitrant pools of carbon and nitrogen in organic matter decomposing at different depths in soil: an acid hydrolysis approach. Geoderma, 107: 109–141.

Sahu G.K., Kar M., Sabat S.C. (2010): Alteration in phosphate uptake potential of wheat plants co-cultivated with salicylic acid. Journal of Plant Physiology, 167: 326–328.

Stevenson B.A., Sparling G.P., Schipper L.A., Degens B.P., Duncan L.C. (2004): Pasture and forest soil microbial communities show distinct patterns in their catabolic respiration responses at a landscape scale. Soil Biology and Biochemistry, 36: 49–55.

Šarapatka B. (2003): Phosphatase Activities (ACP, ALP) in Agroecosystem Soils. [Doctoral Thesis.] Uppsala, Swedish University of Agricultural Sciences: 113.

Traversa A., D'Orazio V., Senesi N. (2008): Properties of dissolved organic matter in forest soils: influence of different plant covering. Forest Ecology and Management, 256: 2018–2028.

Vranová V., Rejšek K., Formánek. P. (2013): Aliphatic, cyclic and aromatic organic acids, vitamins and carbohydrates in soil: a review. The Scientific World Journal, No. 524239, 1–15.

Vranová V., Rejšek K., Formánek P. (2013): Proteolytic activity in soil: A review. Applied Soil Ecology, 70: 23–32.

Ussiri D.A.N., Johnson C.E. (2003): Characterization of organic matter in a northern hardwood forest soil by ^{13}C NMR spectroscopy and chemical methods. Geoderma, 111: 123–149.

Wang Y.Q., Feechan A., Yun B.W., Shafiei R., Hofmann A., Taylor P., Xue P., Yang F.Q., Xie Z.S., Pallas J.A., Chu C.C., Loake G.J. (2009): S-nitrosylation of AtSABP3 antagonizes the expression of plant immunity. Journal of Biological and Chemistry, 284: 2131–2137.

Zhang X., Amelung W., Yuan Y., Samson-Liebig S., Brown L., Zech W. (1999): Land-use effects on amino sugars in particle size fractions of an Argiudoll. Applied Soil Ecology, 11: 271–275.

Zhang G.P., Zeng F., Chen S., Ying M., Wu F. (2003): Changes of organic acid exudation and rhizosphere pH in rice plants under chromium stress. Environmental Pollution, 155: 284–289.

Received for publication October 29, 2015
Accepted after corrections November 18, 2015

Corresponding author:

doc. Ing. Valerie Vranová, Ph.D., Mendel University in Brno, Faculty of Forestry and Wood Technology, Department of Geology and Soil Science, Zemědělská 3, 613 00, Brno, Czech Republic; e-mail: vranova@mendelu.cz