PHOSPHATASE ACTIVITY OF SOIL

Samuel Alina Dora[*], Domuţa Cornel[**], Şandor Maria[**], Vuşcan Adrian[**]

[*]*University of Oradea, Department of Plant Biology, Oradea, Romania*
[**]*University of Oradea, Faculty of Environmental Protection, Oradea, Romania*
E-mail: samuelalina@rdslink.ro

Abstract
Phosphatase activities were determined in the 0-20, 20-40 and 40-60 cm layers of a preluvosoil submitted to a complex tillage (no-till and conventional tillage), crop rotation (2 and 6 crop rotations) and fertilisation [mineral(NP) fertilisation and farmyard-manuring] experiment. It was found that the activities decreased in the order: acid phosphatase activity > alkaline phosphatase activity. Each activity decreased with increasing sampling depth. No-till –in comparison with conventional tillage – resulted in significantly higher soil phosphatase activities in the 0-20 cm layer and in significantly lower activities in the deeper layers. The soil under maize or wheat was more phosphatase-active in the 6 than in the 2 crop rotation. In the 2 crop rotation higher soil phosphatase activities were recorded under wheat than under maize. Farmyard-manuring of maize – in comparison with its mineral fertilisation – led to a significant increase in each activity.

Key words: crop rotation, fertilisation, phosphatase activity, tillage

INTRODUCTION

The degradation of plant and animal matter, the release and binding of nutrients and trace elements, is one of the most important functions of soil organisms (Borza, 2008; Domuţa, 2008). The microorganisms are important for the enzymatic degradation of the complex organic substances to nutrients and for the release of nutrients and trace elements from the mineral soil fraction (Bandick and Dick, 1999; Dick et al., 1988; Dick et al., 1994).

The metabolic activity of soil microorganisms is essential for organic matter turnover (Campbell et al., 1986).

Special enzymes catalyze the organic matter turnover. The name phosphatase describes a group of enzymes that hydrolyzes esters as well as anhydrides of phosphoric acid. There are different phosphatases in soils: phosphomonoesterases, phosphodiesterases, phosphotriesterases, polyphosphatases and phosphoamidase.

The phosphomonoesterases differ in their substrate specificity and their pH optimum. One can thus differentiate between acid and alkaline phosphatases in the soil. The determination of phosphodiesterase, phosphotriesterase and polyphosphatese activities is rarely used in soil analysis. The importance of phosphatase for plant nutrition has repeatedly been pointed out (Deng and Tabatabai, 1997). In most soils, the organically bound P –fraction is higher than the inorganic. Phosphorus uptake by plants requires mineralization of the P-component by phosphatases to ortophosphosphate. Phosphatases are excreted by plant roots and by microorganisms. Microbial phosphatases dominate in soils (Balota et al., 2003; Canarutto et al., 1995; Clarholm and Rosengren-Brinck, 1995).

MATERIAL AND METHODS

The ploughed layer of the studied soil is of mellow loam texture, it has a pH value of 5.5, medium humus(2.32%)and P(22 ppm) contents, but it is rich in K (83 ppm).

The experiment started in 1992. The experimental field occupying 3.84 ha was divided into plots and subplots for comparative study of no-till and conventional tillage, rotations of 2 and 6 crops, and mineral (NP) fertilisation and farmyard-manuring.

The crops of the two rotations are wheat and maize. Each plot consisted of two subplots representing the no-till and conventional tillage variants. The plots were annually NP-fertilised at rates of 120 kg of N/ha and 90 kg of P/ha, excepting in each year, a maize plot (in the 6-crop rotation) which received farmyard (50t/ha) instead of mineral fertilisers. The plots (and subplots) were installed in three repetitions.

In October 2008 soil was sampled from all subplots. Sampling depths were 0-20, 20-40 and 40-60 cm. The soil samples were allowed to air-dry, then ground and passed through a 2 mm sieve and, finally, used for determination of phosphatase activities. Disodium phenylphosphosphate served as enzyme substrate. Two activities were measured: acid phosphatase activity in reaction mixtures to which acetate buffer (pH 5.0) was added and alkaline phosphatase activity in reaction mixtures treated with borax buffer (pH 9.4).

The reaction mixtures consisted of 2.5g soil, 2 ml toluene (antiseptic), 10 ml buffer solution and 10 ml 0.5% substrate solution. Reaction mixtures without soil or without substrate solution were the control. All reaction mixtures were incubated at 37^0C for 2 hours. After incubation, the phenol released from the substrate under the action of phosphatases was determined spectrophotometrically (at 614 nm) based on the colour reaction between phenol and 2,6-dibromoquinone-4-chloroimide (Őhlinger, 2006). Phosphatase activities are expressed in mg phenol/g soil/2 hours. The activity values were submitted to statistical evaluation by the two –way t-test (Sachs, 2002).

RESULTS AND DISCUSSIONS

Results of the statistical evaluation are summarised in Table 1.

Comparison of the two phosphatase activities measured. At the same soil depth (0-20, 20-40, or 40-60 cm) in both subplots under wheat and maize crop of both 2 and 6 crop rotations, the activities decreased in the order: acid phosphatase activity > alkaline phosphatase activity.

Variation of the two soil phosphatase activities in dependence of sampling depth. It is evident from Table 1 that each phosphatase activity decreased with sampling depth in both subplots under wheat and maize crops. In addition, Table 1 shows that the mean values of each of the two activities in both non-tilled and conventionally tilled subplots also decreased with increasing soil depth.

The effect of tillage practices on the phosphatase activities in soil. Each of the two phospatase activities determined was significantly higher (at least at $p< 0.01$) in the upper (0-20 cm) layer of the non-tilled subplots than in the same layer of the conventionally tilled subplots. The reverse was true (at least at $p< 0.02$) in the deeper (20-40 and 40-60 cm) layers. These findings are valid for subplots under each crop of both rotations.

The effect of crop rotations on the phosphatase activities in soil. For evaluation of this effect, the results obtained in the three soil layers analysed in the two subplots of each plot were considered together.

Soil phosphatase activities as affected by different crops in the same rotation

*The 2-crop rotation. Acid phosphatase activity measured in the wheat soil exceeded significantly (*p< 0.01) the coresponding activity recorded in the maize soil. Alkaline phosphatase activity is the same under wheat and maize crops.

The 6-crop rotation. Significant (p< 0.05 to p< 0.001) and unsignificant (p> 0.05 to p> 0.10) differences were registered in the soil phosphatase activities depending on the type of activity and the nature of crop.

Table 1

Significance of the differences between phosphatase activities in a preluvosoil submitted to different management practices

Management practices	**Soil enzymatic activity***	**Soil depth (cm)**	**Mean activity values in management practices**			**Significance of the differences**
			a	**b**	**a-b**	
1.	2.	3.	4.	5.	6.	7.
No-till(a) versus conventional tillage(b)	AcPA	0-20	0.296	0.272	0.024	0.002>p> 0.001
		20-40	0.178	0.202	-0.024	0.02>p> 0.01
		40-60	0.128	0.148	-0.020	0.01>p> 0.002
	AlkPA	0-20	0.256	0.218	0.038	0.01>p> 0.002
		20-40	0.155	0.178	-0.023	0.001>p> 0.0001
		40-60	0.060	0.080	-0.020	0.001>p> 0.0001
The same crop in the two rotations						
Maize in 2-crop rotation (a)versus maize in 6-crop rotation (b)	AcPA	0-60	0.17	0.185	-0.008	0.01>p> 0.002
	AlkPA	0-60	0.138	0.150	-0.012	0.0001>p
Wheat in 2-crop rotation(a) versus wheat in 6-crop rotation (b)	AcPA	0-60	0.194	0.227	-0.033	0.10>p> 0.05
	AlkPA	0-60	0.138	0.179	-0.041	0.002>p> 0.001
Different crops in the same rotation						
2-crop rotation						
Maize (a) versus wheat (b)	AcPA	0-60	0.177	0.194	-0.017	0.01>p> 0.002
	AlkPA	0-60	0.138	0.138	0.000	-
6-crop rotation						
Maize (a) versus wheat (b)	AcPA	0-60	0.185	0.227	-0.042	0.02>p> 0.01
	AlkPA	0-60	1.150	0.179	-0.029	0.01>p 0.002
Maize (a) versus maize (FYM)**(b)	AcPA	0-60	0.185	0.218	-0.033	0.001>p>0.0001
	AlkPA	0-60	0.150	0.181	-0.031	0.01>p> 0.002
Wheat (a) versus maize (FYM) (b)	AcPA	0-60	0.227	0.218	0.009	0.01>p> 0.002
	AlkPA	0-60	0.179	0.181	-0.002	0.02>p> 0.01

* AcPA – Acid phosphatase activity. AlkPA – Alkaline phosphatase activity.

** (FYM) – (farmyard-manured)

Based on these differences the following decreasing orders of the activities could be establised in the soil:

- acid phosphatase activity: wheat> soybean> maize (FYM) > oats+clover> maize plot 6 > maize plot 3;
- alkaline phosphatase activity: maize (FYM) > wheat >soybean >maize plot 6 > oats+clover >maize plot 3.

It is evident from these orders that each of the six plots presented either a maximum or a minimum value of the soil enzymatic activities.

Consequently, these orders do not make it possible to establish such an enzymatic hierarchy of the plots which takes into account each activity for each plot. For establishing such a hierarchy, we have applied the method suggested in (Brejea, 2009). Briefly, by taking the maximum mean value of each activity as 100% we have calculated the relative (percentage) activities. The sum of the relative activities is the enzymatic indicator which is considered as an index of the biological quality of the soil in a given plot. The higher the enzymatic indicator of soil quality, the higher the position of plots is in the hierarchy. Table 2 shows that the first three positions are occupied by those plots in phosphatase activities were the highest. Thus, position 1 was occupied by the farmyard-manured maize plot, whereas the minerally fertilised wheat plot and the minerally fertilised legumes (soybean and clover) were placed on the positions 2, 4 and 5, respectively. The minerally fertilised maize plot occupied the last position could be considered as the least enzyme-active soil.

Table 2

Enzymatic indicators of soil quality in plots of the 6–crop rotation

Position	Plot	Enzymatic indicator of soil quality
1	Farmyard-manured maize	293.0
2	Minerally fertilised (M.f.) wheat	257.8
3	M.f. maize (plot 6)	252.3
4	M.f. soybean	250.3
5	M.f. oats+clover mixtures	247.4
6	M.f. maize (plot 3)	240.1

Soil phosphatase activities as affected by fertilisation. The two maize plots in the 6-crop rotation could serve for comparing the effect of mineral (NP) fertilisation (plot 3) and farmyard-manuring (plot 6) on the soil phosphatase activities. Each activity was higher in the farmyard-manured maize plot than in the minerally fertilised maize plot. The differences were significant (at least at $p < 0.01$).

CONCLUSIONS

No-till in comparison with conventional tillage resulted in higher phosphatase activities in the 0-20 cm soil layer and in lower activities in the 20-40 and 40-60 cm soil layers.

The 6-crop rotation as compared to the 2-crop rotation led to higher phosphatase activities in the soil layers under maize or wheat. In the 2-crop rotation, the soil layers under wheat were more phosphatase-active than those under maize.

Farmyard-manuring in comparison with mineral (NP) fertilisation proved to be more efficient in increasing phosphatase activities in soil layers under maize in the 6-crop rotation.

REFERENCES

1. Balota E.L., A.C. Colozzi-Filho, D.S. Andrade, R.P. Dick, 2003, Microbial biomass in soils under different tillage and crop rotation systems, Biol. Fertil. Soils, 35, p. 300-306.
2. Bandick A.K., R.P. Dick, 1999, Field management effects on soil enzyme activities, Soil. Biol. Biochem., 31, p. 1471-1479.
3. Borza I.A., 2008, Study regarding the influence on water use efficiency in maize crop from Crişurilor Plain, An. Univ. Oradea, Fasc. Prot. Med., Vol.XIII, p. 20-26.
4. Brejea R., 2009, Tehnologii de protecţie sau refacere a solurilor, Ed. Univ. Oradea, p. 212-250.
5. Campbell C.A., M. Schnitzer, J.W.B. Stewart, V.O. Biederbeck, F. Selles, 1986, Effect of manure and P fertilizer on properties of a black chernozem in southern Saskatchewan, Can. J. Soil Sci., 66, p. 601-613.
6. Canarutto S., M. Mazzoncini, A. Perna, S. Cervelli, 1995, The effect of reduction of inputs on phosphatase activity, organic carbon content and water stability index in a corn cultivated soil, Fresenius Environ. Bull., 4, p. 291-296.
7. Clarholm M., M. Rosengren-Brinck, 1995, Phosphorus and nitrogen fertilization of a Norway spruce forest-effects on needle concentrations and acid phosphatase activity in the humus layer, Plant Soil, 175, p. 239-249.
8. Deng S.P., M.A. Tabatabai, 1997, Effect of tillage and residue management on enzyme activities in soils. III Phosphatases and arylsulfatase, Biol. Fertil. Soils, 24, p. 141-146.
9. Dick R.P., P.E. Rasmussen, E.A. Kerle, 1988, Influence of long-term residue management on soil enzyme activities in relation to soil chemical properties of a wheat-fallow system, Biol. Fertil. Soils, 6, p.159-164.
10. Dick R.P., , J.A. Sandor, N.S. Eash, 1994, Soil enzyme activities after 1500 years of terrace agriculture in the Colca Valley, Peru, Agric. Ecosyst. Environ., 50, p. 123-131.
11. Domuţa Cr., 2009, Irrigation opportunity in soybean in the moderate wet area of the Crişurilor Plain during 2006-2008, An. Univ. Oradea, Fasc. Prot. Med., Vol.XIII, p. 39-45.
12. Őhlinger R., 1996, Phosphomonoesterase activity with the substrate phenylphosphate. In: Schinner, F., Őhlinger, R., Kandeler, E., Margesin, R., (eds.) Methods in Soil Biology, Springer, Berlin, p. 210-213.
13. Sachs L., 2002, Der Statistik Test. In: Sachs, L. (ed) Angewandte Statistik Anwerdung statisticher Methoden, Springer, Berlin, p. 189-195.