SOIL GLYCOSIDASE ACTIVITIES AND WATER SOLUBLE ORGANIC CARBON UNDER DIFFERENT LAND USE TYPES

X. Z. Ma^{1,2}, L. J. Chen^{*1}, Z. H. Chen¹, Z.J. Wu¹, L.L. Zhang¹, Y.L. Zhang¹

¹Institute of Applied Ecology, Chinese Academy of Sciences, Shenyang 110016 (China)
²Institute of Soil Fertilizer and Environment Resource, Heilongjiang Academy of Agricultural Sciences, Harbin 150086, China.

* Corresponding author: <u>ljchenchina@hotmail.com</u>

ABSTRACT

The purpose of this study was to measure the effects of different land uses on soil glycosidase activities (α - and β -glucosidase, α - and β -galactosidase), water soluble organic carbon (WSOC) and their relationships. Glycosidase activities showed significant differences under different land use types, the highest one was woodland. β -glucosidase had the highest activity among the four glycosidases. The activities of these glycosidases decreased with increasing soil depth, being all significantly affected by change of soil depth. Except grassland, the four glycosidase activities intercorrelated each other. Woodland had the highest content of WSOC in the soil depth of 0-20 cm and at increasing soil depth, WSOC content decreased sharply under woodland and grassland. Glycosidase activities had positive and significant relationships with WSOC. Glycosidase activities and WSOC all had significant correlations with soil total organic carbon (TOC) and pH, which were sensitive to different land use types. We found that glycosidase activity indirectly impacts on nutrient recycling and energy flow in soil under different land use types.

Keywords: Glycosidase activities, Water soluble organic carbon, Land use, Soil depth.

INTRODUCTION

Different land use types not only had effects on soil structure, soil erosion and biodiversity (Crist *et al.*, 2000), but also on soil enzymes activities and soil nutrient cyclings (Gewin *et al.*, 1999; Islam and Weil, 2000; Acosta-Martínez *et al.*, 2003). Enzymes catalyze all biochemical reactions and are integral part of nutrient cycling in soil. Soil glycosidase is a group of hydrolases involved in the hydrolysis of soil glycosides, among which, α - and β -glucosidase and α - and β -galactosidase are the major members, widely distributed in nature (Eivazi and Tabatabai, 1990)

and playing an important role in the carbon cycle of soil ecosystem. βglucosidase was sensitive to different soil management (Deng and Tabatabai, 1996; 1999). Bandick and Dick, researches had studied the effects of and Tabatabai, 1996; tillage (Deng Curci et al., 1997), crop rotation (Bandick and Dick, 1999), fertilizer amendment (Mijangos et al., 2006; Melero et al., 2007; Sastre-Conde et al., 2007) on glycosidase activities, but less studies are present about the effects of different land use types on glycosidase activities. Among the components of soil carbon

storage, water-soluble organic carbon (WSOC) is the most dynamic C pool in soils. It is only a small proportion of the total organic matter in the soil, present in soil solution and passing a filter pore size of 0.45 µm (Herbert and Bertsch, 1995).

The WSOC could be used by microbes quickly, and it is a useful indicator reflecting the turnover rate of soil organic matter. Many researches had studied its response to fertilization and tillage management (Mazzarino *et al.*, 1993; Erich and Trusty, 1997; Campbell *et al.*, 1999; Chantigny *et al.*, 1999), but little is know about its dynamics under different land uses, and its relations with soil glycosidase activities.

The present investigation has been aimed at studying the effect of different land use types on soil glycosidase activities and WSOC. In addition, relationships between glycosidase activities and WSOC were also studied.

MATERIALS AND METHODS

Study site

Shenyang Experimental Station of Ecology is a member of Chinese Ecosystem Research Network (CERN) under Chinese Academy of Sciences, and locates in the Sujiatun District of Shenyang City, Northeast China. This station was established in 1990, with a total area of about 15 hm². Its soil is classified as aquic brown soil.

The mean annual temperature is 7.0-8.0°C, mean annual precipitation is 650-700 mm, and non-frost period is 147-164 days. Since its establishment, this station installed four types of land use, i.e., lowland for rice, upland for corn, grassland, and woodland for *Populus canadensis*.

Soil sampling

Soil samples were taken at the depths of 0-5, 5-10, 10-20, 20-30, 30-40 and 40-50 cm from each type of the lands in March 2004 by using a core sampler of 5 cm in diameter.

Four duplicates were installed for each type of the lands, and each sample was a composite of 5 cores. A portion of the samples was air-dried for physical and chemical analysis, and another portion was kept fresh for the determination of soil water soluble organic carbon content and enzyme activities.

Soil total organic carbon (TOC) and pH analysis

Soil pH was measured in soil: water suspension (1:2.5 ratio) with glass electrode (PSH-3C) (Lu, 2000); total organic carbon was determined with Liqui TOC analyzer (Elementar, German).

Determination of soil glycosidase activity

Glucosidase and galactosidase activities were determined as described by Eivazi Tabatabai (1988), glycosidase activities were determined with pnitrophenyl-glucopyranoside as substrate (50 mmol L⁻¹), with incubation at pH 6.0 (modified universal buffer, MUB) and 37°C. After 1 h, 0.5 M CaCl₂ and pH 12.0 MUB were added to precipitate humic molecules responsible for brown coloration and extract p-nitrophenol, respectively.

The amount of *p*-nitrophenol released by glycosidases was determined colorimetrically at 410 nm (extinction coefficient is 0.9998**). Glycosidase activities were expressed as mg *p*-nitrophenol kg⁻¹ soil h⁻¹.

Determination of soil water soluble organic carbon (WSOC)

Soil water soluble organic carbon (WSOC) was determined by shaking 50 g field-moist soil with 150 mL deionized water for 1 h (250 rpm), the suspension was centrifuged at 10000 rpm for 10 min, and the supernatant was collected with a 0.45 µm polycarbonate membrane filter under vacuum (Chantigny *et al.*, 1999). The WSOC content was determined by using Liqui TOC analyzer (Elementar, German).

Statistical analysis

The results were analyzed statistically adopting analysis of variance (ANOVA), which were performed using SPSS 11.0 statistical package. Means separation was using Fisher's least significant difference (LSD) test at p≤0.05.

RESULTS

Total organic C (TOC) content and pH value

TOC and pH value of the tested soils under different land use types are shown in Figure 1. There was the highest content of soil organic matter under woodland, followed by grassland, lowland and upland, in the order listed. The content of TOC decreased with soil depth increasing, sharply in the upper layer (0-10 cm) under woodland and grassland, then, changed gently. The pH value was smaller in upland than others, and was higher in the whole soil profiles except 0-5cm under grassland; it increased with soil depth increasing under upland, lowland and grassland, whereas, under woodland, it decreased in 0-20 cm, then increased below 20 cm.

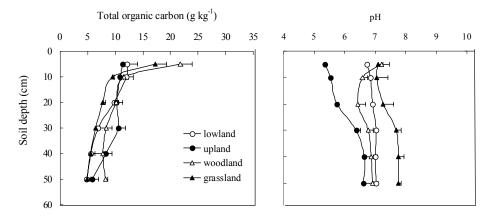


Figure 1. Total organic carbon and pH value under different land use types.

Soil glycosidase activities

The activities of glycosidases were all significantly affected by changes of soil depth and land use and β -glucosidase had the highest activity among them in the soil profiles under different land use types (Figure 2). Generally, they sharply

decreased by increasing the soil layer from 0-5 cm to 5-20 cm, but gently in deeper layers, displaying the same distribution patterns observed for other soil enzymes. Among the four types of land use, woodland had the highest

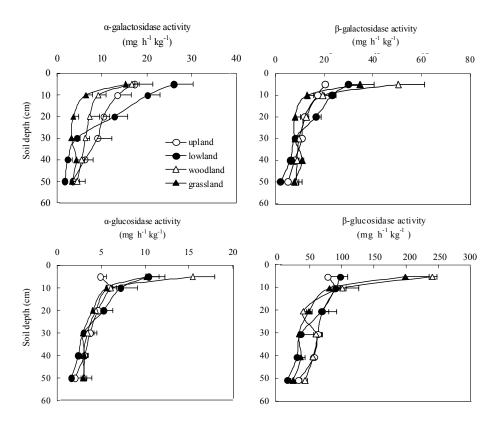


Figure 2. Glycosidase activities in soil profile under different land use types.

activities of α - and β -glucosidase and β -galactosidase in 0-10 cm soil layer, followed by grassland, lowland and upland, while lowland had a significantly higher α -galactosidase activity in this layer than other lands (Figure 2). The relationships between glycosidase activities and TOC, pH are shown in Table 1.

There were positive and significant relationships among glycosidases activities and TOC content (p \leq 0.001) under four land use types, well, there were negative and significant relationships with soil pH (p \leq 0.001) in soil profiles except in woodland. Meanwhile, except α -glucosidase, other three glycosidases activities had positive

relationships with soil pH (p≤0.05) in woodland. The vertical distribution of glycosidases activities had the close relationships with soil characteristics (TOC and pH) (Table 1). Linear regression analysis of the activities of the four enzymes showed that they were significantly intercorrelated in woodland, lowland and upland (Table 2). By contrast, there was no significant relationship between glucosidase and galactosidase in grassland (Table 2).

Soil water soluble organic carbon (WSOC)

Contents of soil water soluble organic carbon (WSOC) decreased sharply with soil depth increasing in woodland,

Table 1. Correlation coefficients between soil glycosidase activities and soil total organic carbon (TOC) and pH under different land use types.

		Enzyme activities				
Land use types	Soil characteristics	α-Glu	β-Glu	α-Gala	β-Gala	
Lowland	TOC	0.794***	0.899***	0.843***	0.817***	
	pН	-0.906***	-0.839***	-0.924***	-0.912***	
Upland	TOC	0.759***	0.778***	0.758***	0.756***	
	pН	-0.825***	-0.737***	-0.783***	-0.882***	
Grassland	TOC	0.943***	0.907***	0.901***	0.881***	
	pН	-0.535**	-0.475*	-0.462*	n.s.	
Woodland	TOC	0.966***	0.938***	0.882***	0.888***	
	pН	n.s.	0.464*	0.447*	0.470*	

(Glu=glucosidase Gal=galactosidase) (* p \leq 0.05, ** p \leq 0.01, *** p \leq 0.001, n.s. not significant)

grassland and lowland, but a smaller fluctuation in upland (Figure 3). Woodland had the highest WSOC content in 0-20 cm, well, the lowland, grassland and upland had lower content of WSOC. Four land use types [except for upland (n.s.)], there were significant and positive between correlations glycosidase activities and WSOC content (from r=0.66*** to r=0.90***) (Table 3). Results also showed that content of WSOC under lowland, grassland and woodland had significantly correlations with TOC ($p \le 0.01$), but not significant in upland; and there were negative and significant correlations with pH under lowland and grassland ($p \le 0.01$) (Table 4).

DISCUSSION

Total organic C (TOC) and pH value

Different land use types affected the input and output of soil organic matter directly; vegetation also had significant effect on soil organic matter. Woodland always had higher content of TOC than cropland, due to many fallings back to soil, lots of root distributed widely in soils of woodland and grassland, so they had much higher content of soil TOC in the upper soil layer. Besides, the cultivation accelerated the decomposition of soil organic matter, made it decrease sharply (Davidson, 1986). Different land use types not only affect the content and distribution of C in soil directly, also had effects on some microbial conditions, which had close relationships with formation and transformation of C, then affect nutrient distribution indirectly.

Soil glycosidase activities

Different land use types had different levels of soil fertility. These changing trends of glycosidases activities decreasing with soil depth have been reported in many researches (Eivazi and Tabatabai, 1990; Deng and Tabatabai, 1996; Taylor *et al.*, 2002), they were just like the tendency of organic C in soil profile, because these activities of

Table 2. Correlation coefficients (r) between activities of paired enzymes in soil samples under different land use types.

		Enzyme activities			
Land use types		β-Gala	α-Glu	β-Glu	
	α-Gala	0.994***	0.881*	0.887*	
Lowland	β-Gala		0.886*	0.889*	
	α-Glu			0.998***	
	α-Gala	0.993***	0.990***	0.980**	
Upland	β-Gala		0.990***	0.990***	
	α-Glu			0.961**	
	α-Gala	0.996***	n.s.	n.s.	
Grassland	β-Gala		n.s.	n.s.	
	α-Glu			0.982***	
	α-Gala	0.989***	0.988***	0.967**	
Woodland	β-Gala		0.996***	0.990***	
	α-Glu			0.988***	

 $(*p \le 0.05, **p \le 0.01, ***p \le 0.001, n.s. not significant)$

glycosidases in soil and content of organic C always had significant and positive relationships, which had been proved by several investigators (Eivazi and Tabatabai, 1988; Bandick and Dick, 1999; Marx *et al.*, 2005), our study also found the significant relationships between activities of glycosidases and TOC in the tested soils (Table 1).

Table 3. Relationships between soil glycosidase activities and water soluble organic carbon content.

-	Upland	Lowland	Grassland	Woodland
α-glucosidase	n.s.	0.68***	0.84***	0.77***
β-glucosidase	n.s.	0.68***	0.90***	0.73***
α-galactosidase	n.s.	0.66***	0.81***	0.73***
β-galactosidase	n.s.	0.71***	0.78***	0.67***

(** p<0.01, *** p<0.001, n.s. not significant)

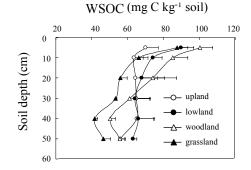


Figure 3. Water soluble organic carbon content in soil profile under different land use types.

Table 4. Correlation coefficients between WSOC and soil properties (TOC, pH) in soil profile under different land use types.

	тос	pН
Lowland	0.633**	-0.681***
Upland	n.s.	n.s.
Grassland	0.864***	-0.639**
Woodland	0.771***	n.s.

(** p<0.01, *** p<0.001, n.s. not significant)

In topsoil enzymes always had higher activities than other soil depth, the main reason maybe that there were higher content of soil organic matter and microbial biomass C, which would stimulate the activity of microorganism, and accelerate the rate of enzyme synthesize (Ekenler and Tabatabai, 2003). There were different enzyme activities under different land use types, due to the differences in organic C content among soils (Acosta-Martínez et al., 2007).

The β -glucosidase activity had close relationship with soil pH, which was conformed to other studies (Eivazi and Tabatabai, 1990; Wang and Lu, 2006), they found that β -glucosidase activity decreased with increasing pH from 4.3 to 7.4, 4.5 to 8.5 respectively, in this study it

decreased with pH increasing from 5.4 to 7.8. However, Deng and Tabatabai (1996) found the inconsistent relationship between β-glucosidase activity and soil pH, significant and positive correlation between them. The main reason maybe soil рН influenced microorganism, synthesis and secretion of enzymes, also the stability of enzymes (Wang and Lu, 2006). Besides, the differences in enzyme activities found in soil samples also may have been due to the difference in soil pH, because the rates of synthesis and release of these enzymes by soil microorganisms are related to soil pH (Deng and Tabatabai,

The α - and β -glucosidase and β galactosidase had higher activities under woodland than other land use types. In woodland, there were some kinds of litter fallings remained on or in the soils, higher content of organic matter, higher enzymes activities. Investigators also had found that enzymatic characteristic of soil was very sensitive, and could do as a potential quality index of soil system (Bandick and Dick, 1999). Activity of βglucosidase was the highest one in this study, which was conformed to former studiers (Eivazi and Tabatabai, 1988; Ekenler and Tabatabai, 2003), which meant that β-glucosidase was sensitive to changes of land use types. α - and β glucosidase and α-and β-galactosidase activities were significantly intercorrelated, suggest that glycosidase have similar origin and persistence in soil (Bandick and Dick, 1999; Acosta-Martínez et al., 2007). We may conclude that glucosidase and galactosidase have a different origin under grassland.

Soil water soluble organic carbon

Soil water soluble organic carbon was only fewer percents of soil total organic carbon, but as the active part. Land uses and management practices could affect soil properties, and also influence WSOC. Soil properties determine organic matter solubility. WSOC content in soil profile decreased with soil depth increasing, maybe due to close relationships between WSOC with soil total organic carbon, which showed similar trends in soil profiles. Generally, large numbers of soluble organic matter were eluviated from residues layer to mineral layer in forest soil, so that amount of WSOC in the topsoil was always higher than other soil depths.

In general, WSOC concentrations varied in different land uses, such as the forest soils, grassland soils, arable soils and so on, mostly due to different vegetation types (Delprat et al., 1997; Haynes, 2000). Besides, we also had learned that fertilization could affect the content of WSOC greatly (Zsolnay and Görlitz, 1994; Jensen et al., 1997; Martin-Olmedo and Rees, 1999). Chantigny (2003) found that content of WSOC decreased with increasing amount of nitrogenous fertilizer as a result, upland had higher content of WSOC than other land uses. Another factor was moisture, Christ and David (1996) found that the total amount of WSOC leaching from forest soil were increasing with times of leaching, so that the woodland and lowland had higher content of WSOC, the other possible reason would be the material returned to the soil by tree canopy, which contained more lignin and other recalcitrant compounds agricultural crop residues (Chantigny, 2003). The WSOC content in upland was lower than others, due to the reasons of fertilization and moisture (Christ and David, 1996), the biochemical and physical environment of upland were different from other land uses. In this study, there was significant and negative correlation between WSOC and soil pH in soil profile under lowland and

grassland ($p \le 0.01$), while not in upland and woodland.

CONCLUSIONES

The present results clearly showed that different land use types had profound impact on soil glycosidase activities and content of water soluble organic carbon (WSOC), which were all decreasing with soil depth increasing. Besides, β -glucosidase was most sensitive to different land uses. Close relationships among glycosidase activities, WSOC and total organic carbon (TOC) contents, meant that glycosidase activity would be indicators for changing in soil quality, which also indicated that effects of different land uses on soil biological activity are very important.

ACKNOWLEDGMENTS

This research was funded by Research and Demonstration of Agricultural Science (2007-3), Public Sector's Special Research of Ministry of Agriculture. The authors thank Shenyang Experimental Station of Ecology, which is a member of Chinese Ecosystem Research Network (CERN) under Chinese Academy of Sciences, for its support to collect soil samples.

REFERENCIAS

Acosta-Martínez, V., Cruz, L., Sotomayor-Ramírez, D., Pérez-Alegría L. 2007. Enzyme activities as affected by soil properties and land use in a tropical watershed. Appl. Soil Ecol. 35, 35-45.

Acosta-Martínez, V., Zobeck, T.M., Gil,l T.E., Kennedy A.C. 2003. Enzyme activities and microbial community structure in agricultural semiarid soils. Biol. Fertil. Soils 3, 216–227.

Bandick, A.K., Dick, R.P. 1999. Field management effects on soil enzyme activities. Soil Biol. Biochem. 31, 1471-1479.

Campbell, C.A., Biederbeck, V.O., Wen, G., Zentner, R.P., Schoenau, J., Hahn, D. 1999. Seasonal trends in selected soil biochemical attributes: effects of crop rotation in the semiarid prairie. Can. J. Soil Sci. 79, 73-84.

Chantigny, M.H. 2003. Dissolved and water-extractable organic matter in soils: a review on the influence of land use and management practices. Geoderma 113, 357-380.

Chantigny, M.H., Angers, D.A., PreÂvost, D., Simarda, R.R., Chalifour, F. 1999. Dynamics of soluble organic C and C mineralization in cultivated soils with varying N fertilization. Soil Biol. Biochem. 31, 543-550.

Christ, M.J., David, M.B. 1996. Dynamics of extractable organic carbon in spodosol forest floors. Soil Biol. Biochem. 28, 1171-1179.

Crist, P.J., Kohley, T.W., Oakleaf, J. 2000. Assessing land-use impacts on biodiversity using an expert systems tool. Landscape Ecol. 15(1), 47-62.

Curci, M., Pizzigallo, M.D.R., Crecchio, C., Mininni, R. 1997. Effect of conventional tillage on biochemical properties of soils. Biol. Fertil. Soils 25, 1-6.

Davidson, S. 1986. Cultivation and soil organic matter. Rural Res. 131, 13-18.

Delprat, L., Chassin, P., Linères, M., Jambert, C. 1997. Characterization of dissolved organic carbon in cleared forest soils converted to maize cultivation. Eur. J. Agron. 7, 201-210.

Deng, S.P., Tabatabai, M.A. 1996. Effect of tillage and residue management on enzyme activities in soils.II. Glycosidases. Biol. Fertil. Soils 22, 208-213.

Eivazi, F., Tabatabai, M.A. 1988. Glucosidases and galactosidase in soils. Soil Biol. Biochem. 20(5), 601-606.

Eivazi, F., Tabatabai, M.A. 1990. Factors affecting glucosidases and galactosidases in soils. Soil Biol. Biochem. 20(5), 601-606.

- **Ekenler, M., Tabatabai, M.A. 2003.** Effects of liming and tillage systems on microbial biomass and glycosidases in soils. Biol. Fertil. Soils 39, 51-61.
- **Erich, M.S., Trusty, G.M. 1997.** Chemical characterization of dissolved organic matter released by limed and unlimed forest soil horizons. Can. J. Soil Sci. 77, 405-413.
- Gewin, V.L., Kennedy, A.C., Veseth, R., Miller, B.C. 1999. Soil quality changes in eastern Washington with Conservation Reserve Program (CRP) take out. J. Soil Water Conserv. 54, 432-438.
- **Haynes, R.J. 2000.** Labile organic matter as an indicator of organic matter quality in arable and pastoral soils in New Zealand. Soil Biol. Biochem. 32, 211-219.
- Herbert, B.E., Bertsch, P.M. 1995. Characterization of dissolved and colloidal organic matter in soil solution: a review. In: W.W. McFee, Kelly, J.M., (eds). Carbon Forms and Functions in Forest Soils. Madison, WI: Soil Science Society of America, pp: 63-68.
- **Islam, K.R., Weil, R.R. 2000.** Land use effects on soil quality in a tropical forest ecosystem of Bangladesh. Agric. Ecosyst. Environ. 79(9), 9-16.
- Jensen, L.S., Mueller, T., Magid, J., Nielsen, N.E. 1997. Temporal variation of C and N mineralization, microbial biomass and extractable organic pools in soil after oilseed rape straw incorporation in the field. Soil Biol. Biochem. 29, 1043-1055.
- **Lu, R.K. 2000.** Methods of soil and agrochemistry analysis. Beijing: Chinese Agricultural Science and Technology Press (in Chinese)
- Martin-Olmedo, P., Rees, R.M. 1999. Short-term N availability in response to dissolved organic carbon from poultry manure, alone or in combination with cellulose. Biol. Fertil. Soils 29, 386-393.

- Marx, M.C., Kandeler, E., Wood, M. 2005. Exploring the enzymatic landscape: distribution and kinetics of hydrolytic enzymes in soil particle-size fractions. Soil Biol. Biochem. 37, 35-48.
- **Mazzarino, M.J., Szott, L., imenez, M.J. 1993.** Dynamics of soil total C and N, microbial biomass, and water soluble C in tropical agroecosystems. Soil Biol. Biochem. 25, 205-214.
- Melero, S., Madejon, E., Ruiz, J.C., Herencia, J.F. 2007. Chemical and biochemical properties of a clay soil under dryland agriculture system as affected by organic fertilization. Eur. J. Agron. 26 (3), 327-334
- Mijangos, I., Perez, R., Albizu, I., Garbisu, C. 2006. Effects of fertilization and tillage on soil biological parameters. Enzyme Micro. Tech. 40, 100-106.
- Sastre-Conde, I., Cabezas, J.G., Guerrero, A., Vicente, M.A., Lobo, M.C. 2007. Evaluation of the soil biological activity in a remediation soil assay using organic amendments and vegetal cover. Sci. Total Environ. 378, 205-208.
- **Taylor, J.P., Wilson, B., Mills, M.S. 2002.** Comparison of microbial numbers and enzymatic activities in surface soils and subsoils using various techniques. Soil Biol. Biochem. 34, 387-401.
- Wang, X.C., Lu, Q. 2006. Beta-glucosidase activity in paddy soils of the taihu lake region, China. Pedosphere 16(1), 118-124.
- **Zsolnay, A., GÖrlitz, H. 1994.** Water-extractable organic matter in arable soils: effects of drought and long-term fertilization. Soil Biol. Biochem. 26, 1257-1261.