

Enzyme activities of urban soils under different land use in the Shenzhen city, China

Z.J. Shi^{1,2}, Y. Lu³, Z.G. Xu², S.L. Fu¹

¹South China Botanical Garden, CAS, Guangzhou, P.R. China

²Shenzhen Institute of Landscape Gardening, Shenzhen, P.R. China

³College of Natural Resources and Environmental Science, South China Agricultural University, Guangzhou, P.R. China

ABSTRACT

Urbanization has drastically changed soil properties, and an assessment of these changes is essential for soil management and soil health. The activities of urease, acid phosphatase, invertase and catalase, soil organic matter, pH, electrical conductivity (EC) and clay (< 0.01 mm) content of urban soils under two land-uses in the central built-up area of the Shenzhen city were investigated, and multivariate analysis was used to study the relationship between soil enzymes and soil physical-chemical properties. The results showed that invertase activity in roadside soil was significantly higher than that in urban park soil, whereas catalase activity was significantly higher in urban park soil. Soil organic matter had significant positive correlation with activities of invertase, urease and acid phosphatase but not with catalase. Soil pH had a significant negative direct effect on urease and acid phosphatase activity, but the effect was counteracted by positive indirect effect of soil organic matter. Soil EC had a positive direct effect on activities of catalase and there was a significant correlation between soil EC and soil catalase activities. Soil organic matter, soil pH and EC were the major factors influencing activities of soil enzymes.

Keywords: invertase; urease; phosphatase; catalase; urban soil

Soil enzymes play essential roles in soil processes such as nutrient cycling and energy transformation by catalyzing numerous chemical, physical and biological reactions. Soil enzymes are sensitive to variations induced by natural and anthropogenic disturbances (Gupta et al. 1988, Dick 1997). Vegetation, soil type, land use history and soil management strategy affect soil enzymes (Green and Oleksyszyn 2002, Wyszowska et al. 2005). Many studies have suggested that soil enzymes can be used as indices of soil contamination, soil fertility and soil health (Masciandaro et al. 1998, Saviozzi et al. 2001).

Previous studies demonstrated the significant relationship between soil enzymes and other soil characteristics, but the relationship is largely dependent on the species of enzymes and the environmental variables (Bergstrom et al. 1998). In contrast, some studies suggest that there is no relationship between soil enzymes and soil nutrients (Sakorn 1987). Urbanization has drastically altered soil properties and processes (Green and Oleksyszyn 2002). The objective of this study

was to assess the impact of land use on urban soil enzymes based on a systematic survey on soil properties in the Shenzhen city, and to illustrate if soil enzyme activities can be used as indices for fertility and health of urban soils.

MATERIAL AND METHODS

Site description

The Shenzhen city is located at 113°46'–114°37'E, 22°27'–22°52'N, south of Guangdong province of China. The city has the total area of 1 952 km² and the urban build-up area of 516 km² with 48% green space. The climate is subtropical monsoonal with abundant heat and moisture. Mean annual temperature and relative humidity is around 23.7°C and 72.3%, respectively. The original zonal vegetation in the area is monsoon evergreen broad-leaf forest. The dominant soil types are Plinthosols in the hill area and Anthrosols in the alluvial plain and basin according to the WRB soil classification

(IUSS Working Group RB 2006). For urban soils, average soil bulk density is 1.55 g/cm^3 , and the content of soil gravel ($> 2 \text{ mm}$) is approximately 30% in 0–20 cm soil depth (Lu et al. 2005). Urban parks and roadsides green belts are two major land use types of green space in the build-up urban area; their soil physical-chemical characteristics were described by Shi et al. (2004).

Soil sampling and analysis

Fifty sites were selected for soil sampling in nine urban parks and seven roadside green belts in the built-up urban area of the Shenzhen city in April 2004. Twenty-five samples were taken from urban parks and another twenty-five from roadside green belts. Composite topsoil samples (0–20 cm) were obtained by mixing sub-samples from six random points within 2 m^2 in each sampling site by using a 7.5-cm diameter hand auger. Soil samples were air-dried for 72 h at room temperature ($22\text{--}28^\circ\text{C}$), and crushed to pass through a 2-mm sieve. All samples were stored at $22\text{--}25^\circ\text{C}$ temperature for two months until laboratory analysis was conducted.

Soil physical and chemical properties were determined according to routine methods, i.e. soil organic matter (OM) was measured by wet oxidation, total nitrogen (TN) by the Kjeldahl method, alkali-hydrolysable nitrogen (AN) by the NaOH hydrolysable method, total phosphorus (TP) by colorimetric method after digested with hydrofluoric and perchloric acid, available phosphorus (AP) by the Olsen method, and soil clay ($< 0.01 \text{ mm}$) content (PC, based on the Kachinsky's classification system) by the hydrometer method, soil pH and electrical conductivity (EC) were measured in 1:2.5 and 1:5 (w/v) ratio of soil to distilled water suspension, respectively (Lu 1999). Soil enzyme activities were assayed in triplicate air-dried samples as described by Guan (1986). Briefly, urease activity was determined using urea as substrate, and the soil mixture was incubated at 37°C for 24 h, the produced $\text{NH}_3\text{-N}$ was determined by a colorimetric method, and urease activity was expressed as $\mu\text{g NH}_3\text{-N/g/h}$. Invertase activity was determined using sucrose as a substrate and incubation at 37°C for 24 h, measuring the produced glucose with the colorimetric method, and invertase activity was expressed as $\mu\text{g glucose/g/h}$. Acid phosphatase activity was measured using sodium phenolphthalein phosphate as a substrate, incubation at 37°C for 24 h, and the liberated phenol

was determined colorimetrically, acid phosphatase activity was expressed as $\mu\text{g phenol/g/h}$; catalase activity was measured using H_2O_2 as a substrate, shaken for 20 min and the filtrate was titrated with 0.1 mol/l KMnO_4 , catalase activity was expressed as $\text{ml } 0.1 \text{ mol/l KMnO}_4/\text{g/h}$.

Statistical analysis

The data from soil analyses were subjected to a two tailed *t*-test to test the statistical significance of soil physical-chemical properties and soil enzyme activities between park soil and roadside soil; the relative importance of direct and indirect effects of measured soil physical-chemical properties on the activity of four enzymes was determined by path coefficient analysis. Stepwise multiple regression analysis was employed to model the quantitative relationship between an enzyme activity and soil physical-chemical properties. All the statistical analyses were performed using SAS 9.0 software.

RESULTS AND DISCUSSION

Influence of land use types on enzyme activities

There was no significant difference between urban park soil and roadside soils for soil organic matter, total N, alkali-hydrolysable N and EC. Soil pH, soil total P and available P were significantly higher in urban park soils than in roadside soil but the content of clay was the opposite. Invertase activity was greater in roadside soils while catalase activity was greater in urban park soils. Land use type had no significant impact on the activity of soil urease and phosphatase (Table 1).

Invertase activity

Correlation analysis showed that soil invertase activity was significantly correlated with soil organic matter but not with other soil physical-chemical variables (Table 2). Path analysis showed that soil organic matter had a positive direct effect, whereas total N had a negative direct effect on invertase activity (Table 3). The indirect effects of soil organic matter on invertase activity through other variables were mostly negative but the effects were marginal, which reduced the direct effect in

Table 1. Characteristics of soils collected from urban parks (PS) and roadside (RS)

	Average value		SD		CV (%)	
	PS	RS	PS	RS	PS	RS
OM (g/kg)	15.80	14.88	6.86	4.99	43.41	33.56
TN (g/kg)	0.50	0.47	0.19	0.14	37.14	30.64
TP (g/kg)	0.23*	0.19	0.09	0.08	40.61	42.44
AN (mg/kg)	30.94	32.26	8.72	10.11	28.19	31.35
AP (mg/kg)	18.95**	6.31	13.16	4.33	69.45	68.62
pH	6.89**	6.35	0.37	0.51	5.31	8.02
EC (ms/cm)	1.37	1.15	0.66	0.43	48.2	37.19
PC (%)	15.44**	18.96	6.49	4.56	42.06	24.05
Invertase activity (μg glucose/g/h)	260.67**	360.12	177.12	184.59	67.95	51.26
Urease activity (μg $\text{NH}_3\text{-N}$ /g/h)	14.18	14.12	6.29	3.71	44.32	26.5
Phosphatase activity (μg phenol/g/h)	71.42	60.25	49.82	34.48	69.76	57.22
Catalase activity (ml 0.1 mol/l KMnO_4 /g/h)	6.45**	5.18	2.20	1.93	34.1	37.34

OM – organic matter; TN – total nitrogen; TP – total phosphorus; AN – alkali-hydrolysable nitrogen; AP – available phosphorus; EC – electrical conductivity; PC – soil clay content

*, **significant at 5% and 1% level

general. Soil total P, alkali-hydrolysable N and clay had a positive indirect effect on invertase via soil organic matter. Soil available P, pH, and EC did not show any direct or indirect effect on invertase. Based on these results we could conclude that soil organic matter has the most significant and positive effect on invertase. Stepwise regression analysis (Table 4) showed that soil total N has a significantly negative effect on invertase activity, which may indicate that invertase activity increased by soil organic matter but was inhibited by soil total N.

Urease activity

Soil urease activity was closely and positively correlated to soil total N, total P, alkali-hydrolysable N and physical clay as showed in Table 2. Path analysis showed that soil organic matter and alkali-hydrolysable N had a positive direct effect on urease activity and the effects were strengthened by all other indirect factors (Table 3). While total N had a positive indirect effect via soil organic matter and was strengthened by other factors. Total P and clay showed a closely positive relationship with an

Table 2. Simple correlation coefficients of soil properties

	X1	X2	X3	X4	X5	X6	X7	X8	X9	X10	X11	X12
X1	1.000	0.756**	0.602**	0.376**	0.211	0.229	−0.043	0.456**	0.448**	0.599**	0.444**	−0.029
X2		1.000	0.502**	0.453**	0.019	0.236	−0.050	0.358**	0.177	0.554**	0.334*	−0.145
X3			1.000	0.299*	0.562**	0.330*	−0.070	0.281*	0.016	0.402**	0.420**	−0.045
X4				1.000	0.036	−0.078	−0.273	0.367*	0.076	0.540**	0.384**	−0.210
X5					1.000	0.258	−0.043	−0.039	−0.105	0.064	0.565**	0.238
X6						1.000	−0.032	−0.045	−0.120	−0.059	−0.153	0.124
X7							1.000	−0.364**	0.068	−0.038	−0.241	0.311*
X8								1.000	0.236	0.342*	0.302*	−0.152
X9									1.000	0.295*	0.014	−0.254
X10										1.000	0.415**	0.042
X11											1.000	0.092
X12												1.000

*, **significant at 5% and 1% level

X_i ($i = 1-12$) stands for soil OM, TN, TP, AN, AP, pH, EC, PC (see Table 1), invertase activity, urease activity, acid phosphatase activity, catalase activity

Table 3. Path coefficients of soil physical-chemical properties on soil enzymes activity

		X1	X2	X3	X4	X5	X6	X7	X8	Sum
Invertase activity	X1	<u>0.912**</u>	-0.263	-0.149	-0.015	-0.023	-0.029	-0.003	0.019	0.448**
	X2	0.689**	-0.348*	-0.125	-0.018	-0.002	-0.031	-0.003	0.015	0.177
	X3	0.549**	-0.175	-0.248	-0.012	-0.063	-0.043	-0.005	0.012	0.016
	X4	0.344*	-0.158	-0.074	-0.039	-0.004	0.010	-0.018	0.016	0.076
	X5	0.192	-0.007	-0.139	-0.001	-0.112	-0.034	-0.003	-0.002	-0.105
	X6	0.209	-0.082	-0.082	0.003	-0.029	-0.135	-0.002	-0.002	-0.120
	X7	-0.038	0.017	0.017	0.011	0.005	0.004	<u>0.067</u>	-0.015	0.068
	X8	0.416**	-0.125	-0.070	-0.014	0.004	0.006	-0.024	0.042	0.236
Urease activity	X1	<u>0.378**</u>	0.086	0.062	0.122	-0.009	-0.037	-0.003	-0.002	0.599**
	X2	0.285*	0.113	0.052	0.146	-0.001	-0.039	-0.004	-0.002	0.554**
	X3	0.227	0.057	0.104	0.097	-0.024	-0.053	-0.005	-0.001	0.398**
	X4	0.142	0.051	0.031	<u>0.323*</u>	-0.002	0.013	-0.019	-0.002	0.540**
	X5	0.080	0.002	0.058	0.012	-0.042	-0.042	-0.003	0.000	0.062
	X6	0.083	0.026	0.033	-0.025	-0.011	-0.166	-0.002	0.000	-0.059
	X7	-0.016	-0.006	-0.007	-0.088	0.002	0.006	0.070	0.002	-0.038
	X8	0.173	0.041	0.028	0.119	0.002	0.007	-0.026	-0.004	0.341*
Phosphatase activity	X1	0.211	0.183	-0.101	0.046	0.146	-0.081	0.007	0.034	0.443**
	X2	0.159	0.242	-0.084	0.055	0.013	-0.086	0.008	0.026	0.333*
	X3	0.127	0.122	-0.167	0.037	0.390**	-0.119	0.011	0.021	0.420**
	X4	0.079	0.110	-0.050	0.122	0.025	0.028	0.043	0.027	0.383**
	X5	0.044	0.005	-0.094	0.004	<u>0.694**</u>	-0.093	0.007	-0.003	0.565**
	X6	0.046	0.056	-0.054	-0.009	0.174	<u>-0.369**</u>	0.006	-0.004	-0.153
	X7	-0.009	-0.012	0.012	-0.033	-0.030	0.014	-0.157	-0.027	-0.241
	X8	0.096	0.087	-0.047	0.045	-0.027	0.018	0.057	0.073	0.302*
Catalase activity	X1	0.137	-0.120	-0.175	-0.024	0.076	0.029	-0.013	0.032	-0.059
	X2	0.102	0.120	-0.147	-0.028	0.007	0.030	-0.016	0.025	0.218
	X3	0.082	-0.060	<u>-0.290*</u>	-0.019	0.202	0.042	-0.022	0.019	-0.045
	X4	0.052	-0.054	-0.087	-0.063	0.013	-0.010	-0.087	0.025	-0.210
	X5	0.029	-0.002	-0.164	-0.002	<u>0.361*</u>	0.034	-0.014	-0.003	0.239
	X6	0.030	-0.028	-0.096	0.005	0.093	0.129	-0.012	-0.003	0.124
	X7	-0.006	0.006	0.020	0.017	-0.016	-0.004	0.319*	-0.025	0.311*
	X8	0.062	-0.043	-0.082	-0.023	-0.014	-0.006	-0.116	0.069	-0.152

*, **significant at 5% and 1% level

The data underlined are direct path coefficients, data in Sum column are correlation coefficients, and the rest data are indirect path coefficients

X_i ($i = 1-8$) stands for soil OM, TN, TP, AN, AP, pH, EC, PC (see Table 1)

Table 4. Summary of stepwise multiple regression analysis of soil enzymes

Regression equations	R-square
Invertase activity = $242.166 + 17.782x_1 - 8.724x_2$	0.449
Urease activity = $1.715 + 0.396x_1 - 0.199x_2$	0.474
Phosphatase activity = $197.357 + 1.566x_1 - 0.732x_2 - 7.11x_3$	0.636
Catalase activity = $1.437 + 0.040x_3$	0.199

X_i ($i = 1-3$) stands for soil OM, TN, TP (see Table 1) respectively; (0.15 level, $n = 50$)

accumulated path coefficient though all factors had a rather small effect. Soil available P, pH and EC had no effect on urease activity. The effects of soil organic matter and total N on urease activity were similar to their effects on invertase activity, indicating that soil organic matter increased and total N inhibited urease activity in soils (Table 4).

Phosphatase activity

Though soil organic matter, total N, alkali-hydrolysable N and clay did not show any significant direct or indirect effect on phosphatase activity by path analysis (Table 3), simple correlation analysis showed that phosphatase was closely and positively correlated with total N, total P, alkali-hydrolysable N and clay (Table 2). Soil available P and total P showed a direct or indirect positive effect via available P; the direct effect was enhanced while the indirect effect was reduced by other soil properties. Soil pH had a significant negative direct effect, but it was counteracted by other soil properties. The stepwise regression model showed that phosphatase activity was quantitatively restricted by soil total N and total P but increased by soil organic matter (Table 4).

Catalase activity

Soil catalase activity was closely related to soil EC, but had no significant relationship with other physical-chemical properties as shown in Table 2. Total P showed a significant positive effect and available P showed a negative direct effect on catalase, respectively; however, these effects were all counteracted by other soil properties (Table 3). Though soil EC had a direct and positive effect on and significant relationship with catalase activity (Table 3), the stepwise regression model showed that soil EC and other soil physical-chemical properties except for soil total P were insignificant in reflecting quantitative relationship with catalase activity (Table 4).

It was reported that soil enzyme activity is sensitive in discriminating soil qualities among soil management treatment effects (Saviozzi et al. 2001, Sadowsky et al. 2006). Soil enzyme activity is an effective indicator of soil quality resulting from environmental stress or management practices (Dick 1997). Our results are consistent with other reports showing that soil invertase, as an effective indicator of soil quality, varies with land uses (Green and Oleksyszyn 2002, Liu et al. 2006). Nevertheless, urease and phosphatase in urban soils had no response to land use change. Catalase activity in urban soils showed a distinct difference between park and roadside soils, which disagreed with the previous findings that catalase did not differ under different land uses (Liu et al. 2006). Our results showed that invertase, urease and phosphatase activities are higher than those in barren wasteland soil but lower than those in farmland soil with good fertility as reported by Wang et al. (2003) and Qiu et al. (2004).

Wang et al. (2003) found that the sensitivity of three soil enzymes to environmental stress was in the order of urease > invertase > catalase. Soil invertase significantly and positively correlated with soil organic C, soil biological and chemical properties. Urease and phosphatase activities could be used as indicators for soil contaminated by Cd, Ni and Cu (Moreno et al. 2003, Wyszowska et al. 2005), and soil fertility (Qiu et al. 2004). Soil urease activity is also closely related with soil salinity in saline soils (Cookson 1999) and can be used as an index of soil degeneration and reclamation (Wang et al. 2003). Soil phosphatase activity was often used as an index for organic phosphorus mineralization and biological activity and was inhibited by soil contamination of Cu, Cd, and Ni (Moreno et al. 2003, Wyszowska et al. 2005). Soil phosphatase activity accompanied with soil urease activity can be a useful compound index for soil fertility (Qiu et al. 2004). Soil catalase activity is sensitive to soil biological factors (Asmar et al. 1992), is closely related with soil major nutrient elements (Rodriguez-Kabana and Truelove 1982), and inhibited by soil contamination of Pb and Cd

(Liu et al. 2007). All reports mentioned above mainly concerned about soil contamination, and the knowledge on enzymes activities related to soil fertility in urbanized anthropogenic soils is still scarce. In general, our results were consistent with these findings, and showed that invertase, urease and phosphatase activities are sensitive to the change of soil organic matter and soil total N. Soil catalase activity can be estimated through soil total P determination and can reflect soil salinity to a certain extent.

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Corresponding author:

Zheng Jun Shi, Ph.D., Shenzhen Institute of Landscape Gardening, Shenzhen 518003, P.R. China
phone: + 86 75 525 401 239, fax: + 86 75 525 011 758, e-mail: shizj1976@yahoo.com.cn
