Effect of land use on soil enzyme activities at karst area in Nanchuan, Chongqing, Southwest China

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ABSTRACT

The study of soil enzyme activities under different land use is of importance for exploration of the soil quality evolution and its evaluation during the revegetation at karst area in Nanchuan, Chongqing, China. Seven kinds of land use were chosen as subject, aiming at revealing the changes in soil enzyme activities through experimental and statistical analysis. The results showed that different land use significantly influenced the enzyme activities. Soil urease, invertase, catalase and amylase behaved the different change. A descending order of urease activity was artificial forest, natural forest, shrubbery, grassland, slope field into terrace, rock desertification and farmland. As to invertase and amylase, they have no clear change orderliness with land use change. Moreover, no significant change was observed in catalase activity and the lower value was found in natural forest. The above results cannot reflect the land use effect on the enzyme activities. According to the soil enzyme index (SEI), it can be seen that the SEI changed with an order of natural forest > artificial forest > grassland > shrubbery > farmland > slope field into terrace > rock desertification, which can objectively and fully evaluate the land use change in soil enzyme.

Keywords: karst; land use; soil enzyme index; soil urease; invertase; catalase; amylase

Soil enzymes play an essential role in catalyzing reactions necessary for organic matter decomposition and nutrient cycling in ecosystems (Taylor et al. 1989, Johansson et al. 2000). Agricultural management practices (e.g. crop rotation, mulching, burning, tillage and application of fertilizers and pesticides) have diverse effects on the various soil enzyme activities (Ajwa et al. 1999, Bandick and Dick 1999, Aon and Colaneri 2001, Xu et al. 2002). So, enzymatic activities are candidate 'sensors' of soil stress for management practices that may timely forewarn soil degradation (Bergstrom et al. 1998, Margesin et al. 2000).

China is one of the countries with the largest karst landscape, with an area of more than 2×10^6 km^2, accounting for 5% of carbonate coverage in the world (Yuan 1991). In the southwestern China, the coverage of karst landform is about 540×10^3 km^2, administratively including Guangxi, Guizhou, Yunnan, Sichuan, Hunan, Hubei and Chongqing. With rapid population growth and vulnerability of karst environment, soil erosion becomes so serious that it has caused the loss of surface soil in steep slopes and bareness of bedrock in large area, which directly threats further development of economy in SW China (Wang et al. 2004). To date, the significant relationship between soil enzymes and other soil characteristics was demonstrated involved in the species of enzymes and the environmental variables at the karst area (Zhang et al.

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2006), but the soil enzyme index (SEI) almost was not discussed, which may be a suitable method to understand the soil enzyme activity under the effect of the land use (Wang et al. 2009). So, the study of soil enzyme activity and its index in different land use is of importance for exploration of the soil quality evolution and its evaluation at karst area.

MATERIAL AND METHODS

Study area. Nanping town (106°56'15''–107°0'30''E, 29°05'30''–29°0'10''N) is located in southwest of Nanchuan county, Chongqing province with an area of 10 km^2. The study area is characterized with a humid subtropical monsoon climate. Seasons alternate between humid springs, scorching summers with intensive rainfall, dry autumns and cold humid winters. The mean annual temperature is about 16°C and the average annual precipitation is 1300 mm. The average annual sunshine is 1273 h and the global solar radiation is about 334 614 J/sq. The frost-free period is about 310 days. At this site, the local people have to depend on rainfall for drinking and living due to the deep buried groundwater and the developed karst fracture. Under the bad condition, the crops were irrigated by natural rainfall, so the rocky desertification is very serious with the 34% soil erosion area (Wei et al. 2010). The local people are almost under the living standard.

As a representative agricultural and rocky desertification county of karst area in China, more than 80% of the total area of Nanping town is used as cultivated land. The current fertilization and management style have prevailed for more than 50 years. In this area, single crops were replanted annually with continuous spring maize, sweet potato, as the prevailing cropping system.

Soil sampling. The 26 samples from 7 types of land use were collected randomly in 2008 and 2009 from Nanping town. At each sample plot, three adjacent soil cores (5 cm diameter) were collected at 0–20 cm depths, which were mixed as one soil sample. The soil samples were packaged on ice and shipped to the laboratory overnight, where the soils from each plot were sieved (2 mm), separated from rocks and roots, homogenized, and stored at 4°C until analysis.

Enzyme assays. Soil enzyme activities were assayed as described by Guan (1986). All enzyme activities were determined from air-dried samples in triplicate. The invertase activity was expressed as mg glucose released g/h. The catalase activity was measured using H_2O_2 as a substrate, shaked for 20 min and the filtrate was titrated with 0.1 mol/L $KMnO_4$. The catalase activity was expressed as mL 0.1 mol/L $KMnO_4$/g/h. The urease activity was determined using urea as substrate, and the soil mixture was incubated at 37°C for 24 h. The produced NH_4^+-N was determined by a colorimetric method, and urease activity was expressed as μg NH_4^+-N/g/h. The amylase activity was determined by measuring the amount of reducing sugars generated from starch under the catalysis of the amylase at 50°C for 30 min and the activity was expressed as mg glucose released g/h.

Statistical analysis. In order to fully reveal the change of soil enzyme activities under different land use, the soil enzyme index was calculated according to the linear weighted method based on the determination of enzyme activities (Wang et al. 2009). In order to eliminate the influence of different dimensional parameter upon factor loadings, the measured value of enzyme activities need to be standardized and is transformed using 0–1 mixed integer programming with the certainty. Then, the normalized difference index will be achieved. Because of the continuity change of soil factors, the membership degree function with continuous property is adopted to get the enzyme evaluation index. Then, according to the properties of positive and negative of the loading value from the principal component factor, the lift or drop type membership functions will be set, which prove that the enzyme evaluation index is corresponding to the vegetation effect. In our paper, the drop type membership function was selected to evaluate invertase and catalase (1), and the lift type membership function was selected to evaluate urease and amylase (2). The calculation formula of drop membership function and the lift membership functions are listed as follows:

$$\mathrm{SEI}\ (X_i) = (X_{ij} - X_{imin})/(X_{imax} - X_{imin}) \quad (1)$$

$$\mathrm{SEI}\ (X_i) = (X_{imax} - X_{ij})/(X_{imax} - X_{imin}) \quad (2)$$

Where: X_{ij} – soil enzyme activity; X_{imax} and X_{imin} – max and min activity of the soil enzyme.

Moreover, due to the different condition of the soil properties and the importance of active factors, the weighting coefficient was used in this article to conform the soil factors of all the indicators in the analysis system. Firstly, based on the transformation of membership matrix for realizing the principal component analysis, the contribution ratio and cumulative contribution ratio about every factor of principal component will be calculated. Secondly, according to the loading value of the principal component to calculate the size of each soil factor, their weight values are determined. That can be explained that based on the percentage of every communality in the total sum of communalities, the weight values are transformed using 0–1 mixed integer programming. The weight values of soil enzyme are calculated by:

$$W_i = C_i/C$$

Where: W_i – weight value of soil enzyme; C_i – communality, and C – total sum of communalities.

Based on the normalization of dimensional parameter and weight about soil factors, the soil enzyme index was calculated with the method of the weighted sums.

$$\text{SEI} = \sum_{i-1}^{n} W_i X \text{SEI}(X_i)$$

Where: W_i – weight values of soil enzyme; SEI (X_i) – membership function of soil enzyme.

One-way ANOVA tests were performed using SPSS computer language program to access the changes in soil enzyme activities with different land use.

The significance of differences among means was calculated using JMP version 5.0, SAS Institute Inc., Cary, USA. Different letters indicate significant differences in activity at $P < 0.05$.

RESULTS AND DISCUSSION

Influence of land use on enzyme activities. Soil enzyme activities can be used as potential indicators of nutrient cycling processes and fertility management (Böhme et al. 2005, Fließbach et al. 2007). Due to soil enzymes considered sensitive to disturbances in ecosystem (Zwikel et al. 2007), their activities were increased to different degrees by organic manure incorporation and gave a significant and positive relationship of enzyme activity with organic C and total N (Nayak et al. 2007). Conversely, the soil degradation process reduced C turnover and nutrient availability (Bhattacharyya et al. 2005, Gopal et al. 2007).

Our study shows that the effect of land use on soil enzyme activity is different (Figure 1). Urease, a key enzyme that regulated soil nitrogen transformation, comes mainly from plants and microbes, and plays an indispensable role in the nutrient cycling process (Kong et al. 2008a,b). From Figure 1, it can be seen that the urease activity in the artificial forest is quite higher than others. The high urease activities in artificial forest with the human maintenance may be the consequence of both microbial growth and stimulation of microbial activity by enhanced resource availability, as well as of changes in microbial community composition. In addition, the improvement of soil physical properties can make the soil environment more favorable for microbial life (Iovieno et al. 2009, Tejada et al. 2009), which resulted in significant supplement in soil nutrient transformation (Mandal et al. 2007, Nayak et al. 2007). Moreover, soil ureases as microbial products can accumulate in cell-free forms in the soil because they are highly resistant to environmental degradation (Zantua and Bremner 1977). Therefore, in other land use, the urease activities are followed by natural forest > shrubbery > grassland > slope field into terrace > rock desertification > farmland, which reflects that the urease activity is closely related with soil ecosystem stability and soil health (Kong et al. 2007, Liu et al. 2008). Moreover, in contrast with the significant differences of urease activity in natural forest and artificial forest, which responds to the obvious influence of anthropogenic disturbances in the forest community, there are no statistical differences in the fragile ecosystem of shrubbery, grassland, slope field into terrace, rock desertification and farmland (Figure 1).

The activities of invertase and amylase, related to the C cycling of the soil (Sardans et al. 2008), have no obvious interactions with the eco-environment in different land use. From Figure 1, the descending order of invertase activity was grassland, slope field into terrace, shrubbery, natural forest, artificial forest, farmland and rock desertification, and the descending order of amylase activity was natural forest, grassland, rock desertification, farmland, artificial forest, shrubbery and slope field into terrace. The reason is that in the karst area, the soil has its own particularities, such as calcium-rich and high alkalinity. The soil organic carbon is quite stable, which decreases the decomposition process of organic carbon and the activities of invertase and amylase (Li et al. 2014). Moreover, based on the statistical and analytical results, there are almost no significant differences of invertase and amylase in our research sites (Figure 1). So, it can be seen that the land use impact on soil enzyme

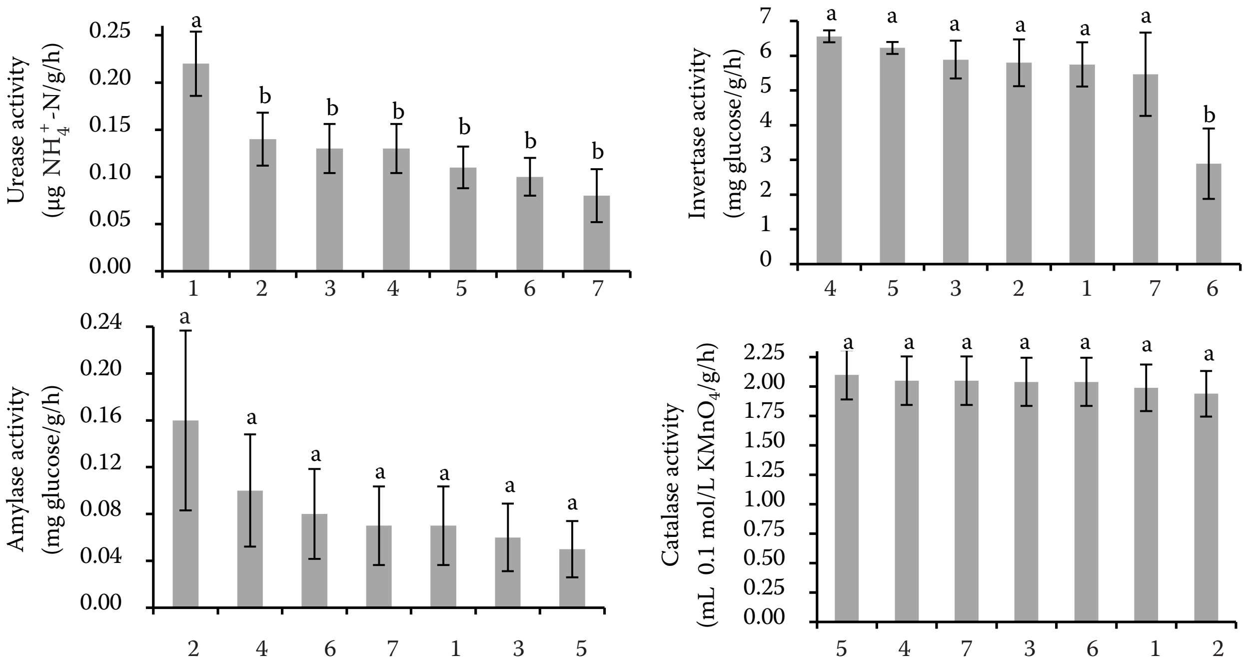

Figure 1. Change in soil enzyme activity of different land use. 1 – artifical forest; 2 – natural forest; 3 – shrubbery; 4 – grassland; 5 – slope field into terrace; 6 – rockdesertification; 7 – farmland

activities (invertase and amylase) vary under different types of soil and environmental conditions.

Catalase is an intracellular enzyme found in all aerobic bacteria and most facultative anaerobes, but absent in obligate anaerobes (Trasar-Cepeda et al. 2000). It is well known that the products of oxygen reduction, such as hydrogen peroxide, superoxide radical, and hydroxyl radical, can be highly toxic for cells and might damage cellular macromolecules. Catalase can split hydrogen peroxide into molecular oxygen and water and thus prevent cells from damage by reactive oxygen species (Yao et al. 2006). Although, it was one of the first isolated and purified enzymes, its physiological function and regulation are still poorly understood. Catalase activity may be related to the metabolic activity of aerobic organisms and has been used as an indicator of soil fertility (Trasar-Cepeda et al. 2007) and has a significant correlation with the content of organic carbon decreasing (Alef and Nannipieri 1995). In the karst area of China, the decomposition process of organic carbon is reduced. The invertase and amylase have no significant regular change under the effect of land use. Though, the karst ecosystem in southwestern China is fragile and the rock-desertification under the anthropogenic impact is serious (Yuan 1991), there are no obvious statistical differences in these land use types. Therefore, under the adverse environments, no significant change tendency was observed in catalase activity and the lower value was found in natural forest (Figure 1).

Invertase, urease and amylase, the hydrolytic enzymes responsible for macromolecules hydrolytic breakdown which is assumption for mineralization, are involved in C and N processes (Ajwa et al. 1999, Marx et al. 2001, Sardans et al. 2008). Ureases catalyze the hydrolysis of urea into ammonia and carbamate, which spontaneously decomposes to form carbon dioxide and a second molecule of ammonia ($CO(NH_2)_2 + H_2O \rightarrow 2\ NH_3 + CO_2$) and are essential in the chain of hydrolysis of amino compounds which are supplied to the soil from plants and to a lesser extent from animals and microorganisms (a molecule of ornithine combines with one molecule of ammonia and one of CO_2 to form citrulline; a second amino group is added to citrulline to form arginine, which is then hydrolyzed to yield urea, with regeneration of ornithine) (Sardans et al. 2008). Invertase and amylase can break down some carbohydrate polymers in order to liberate the nutrients of organic compounds through its role in the first phases of degradation of organic compounds that reduce the molecular size and produce smaller organic structure, and thus facilitate future microbe enzyme activities (Sardans et al. 2008). Therefore, it can be seen that the invertase, urease and amylase have the intimate relationship and the catalase

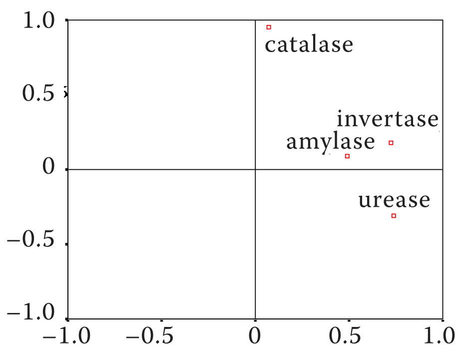

Figure 2. The component plot in rotated space of the soil enzyme

departures from the invertase, urease and amylase in the plot of quadrants at Figure 2. Moreover, due to the complex geological background at karst area, though the catalase can reduce the damage from dry and hot karst soil, it is not the main factor to reflect the pattern and magnitude of soil character. To sum up, if only single soil enzyme was used to estimate the land use type, we cannot acquire enough information to evaluate the environmental condition.

The soil enzyme index. From the above results, it can be found that the soil enzyme has the different variation under the effect of different land use. In order to overcome the one-sided results of the soil enzyme characteristics due to the land use change, especially without significant statistical differences, the soil enzyme index (SEI) was used to reflect the comprehensive and objective characters of the soil enzymes activities (Figure 3). It can be found that the SEI was significantly decreased from the natural forest, the artificial forest, the grassland, the shrubbery, the farmland, the slope field into terrace to the rock desertification. Due to the serious human disturbance, the farmland and the slope field into terrace have the low SEI and the SEI for farmland and slope field into terrace has no statistical differences, which reflects the land use type with the low land-cover, the single species, and the low metabolism rate. Under the impact of anthropogenic activities, the rocky desertification area has the serious problem of water and soil erosion, which can reduce the C turnover and nutrient availability. Then, the SEI in the rock desertification area is negative and the rock desertification has the significant statistical differences with other land use types. Compare to the artificial forest, as to those land use types in the different succession stages for natural forest, grassland and shrubbery, the SEI has the

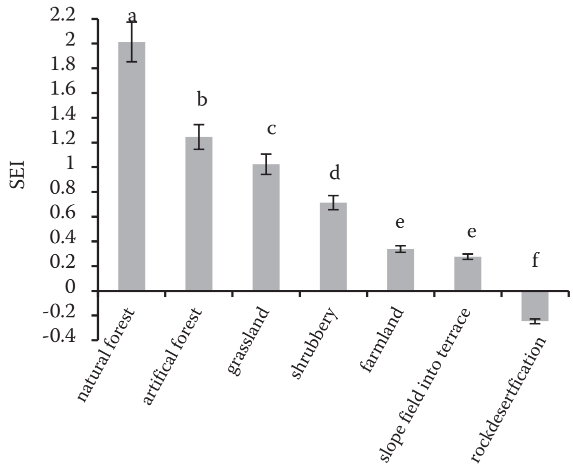

Figure 3. Change in soil enzyme index (SEI) of different land use

significant statistical differences, which reflects their ecological effects to land use (Figure 3). In summary, the soil enzyme index can be used as a suitable evaluation method to indicate the soil quality at karst area, especially when the limited experiments and data were achieved.

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