



Response of soil enzyme activity to long-term restoration of desertified land[☆]



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ABSTRACT

Low extracellular enzyme activity in desert soil can be recovered during the succession of re-vegetation, especially in soils forming under shrubs (microsite soil), which closely reflects desert restoration conditions. However, not much is known about the restoration of soil enzyme activity at these microsites. By using the space-for-time substitution method, soils on moving sand dunes that had been stabilized at different dates over a fifty year period at the southeastern fringe of the Tengger Desert were selected to investigate the enzyme activities in the surface soil crust and three other soil depths at microsites to demonstrate the evolution of enzymatic activity at different stages from bare soil to complex vegetation over a fifty year sequence. The results showed that organic C and total and available N, P, and enzyme activities (dehydrogenase, catalase, α - and β -glucosidase, protease, and phosphatase) were progressively enhanced in each microsite soil in the 50-year chronosequence and had effect down to 35 cm depth. Soil enzyme activities of the crust and the 0–5 cm soil layer were higher than in deeper soil layers. The observed increase over time of the values of the measured soil properties, such as organic C, total and available N, was much larger in the crust and the 0–5 cm soil layer in comparison to the deeper layers. The improvement of desert soil quality indicated that desertification can be mitigated to a certain extent under human controls.

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1. Introduction

Intensive cultivation and overgrazing lead to the loss of vegetative cover, followed by the loss of most topsoil. Millions of hectares of cultivated land have been lost in this way (Wang et al., 2012). Re-vegetation has been reported as one effective method to control soil erosion and restore a healthy ecosystem in the desert region (Li et al., 2003). Soil nutrient levels increased during the restoration process and were used as an indication of successful restoration (Cao et al., 2008; Chen and Duan, 2009; Tongway et al., 2003). He et al. (2009) reported that the storage of C and N in bulk soil and soil particle-size fractions increased during a soil restoration time sequence measured at 3, 8, 20, 24, and 28 years from planting. However, soil microbial properties, such as soil enzyme activities, which are the driving force in nutrient cycling, are more sensitive than physical and chemical indicators (Tabatabai, 1994). Soil enzyme levels were closely related to crust development,

nutrient transformation, soil formation on stabilized dunes, and soil fertility in rehabilitated sandy soils (Consuelo and Teodoro, 2002). Sampling of sites 5, 10, and 23 years after replanting showed a progression of increasing leaf deposition, net primary productivity and electrical conductivity, and consequently elevated nutrient content and microbial biomass (Cao et al., 2008). Over time, vegetation restoration improved activities of polyphenol oxidase, dehydrogenase urease, protease, and phosphomonoesterase (Cao et al., 2008; Consuelo and Teodoro, 2002; Dick et al., 1996; Nannipieri et al., 2002). Blank (2002) noted increased activities of N-mineralizing enzymes at inter-shrub areas (microsites) after restoration. Re-planting of sandy soils has resulted in the restoration of stable vegetation to combat desertification and has significantly improved physical, chemical, and biological soil properties (Bolling and Walker, 2000; Cao et al., 2008; Ffolliott et al., 1995; Zhang et al., 2008; Zuo et al., 2009).

The progressive response of soil enzymes after replanting can indicate the degree of success of desert restoration projects (Cao et al., 2008). Plant restoration enhanced physical, chemical and microbial properties from top-soil to deeper layers (Cao et al., 2008), meaning that sandy soil recovered biological activity from the surface downwards. Tongway et al. (2003) demonstrated that the enhancement trend was most apparent in the top 1 cm of soil rather than deeper layers. Usually, enzyme activities decrease with increasing soil depth

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(Blank, 2002). Crusts develop slowly from barren or disturbed soils and restoration of desertified sands is a long process, during which natural succession resulted in the disappearance of some replanted species at a later stage (Thompson et al., 2006). Soil nutrients and biological properties in soils beneath shrubs were higher than those between shrubs in dry soils (Jackson et al., 1988; Sarig and Steinberger, 1994; Schlesinger et al., 1996). Sarig et al. (1994) considered that nutrient heterogeneity was mainly controlled by the presence of plant cover as such, not species forming that cover. Thompson et al. (2006) documented that microsites can reflect degree of desert restoration, yet much remains unknown about microsite soil.

There have been many studies about specific areas under the canopy of specific species, and most studies have concentrated on the effects of the shrub on the chemical and physical properties of the soil as well as some enzyme activities (Cao et al., 2008; Zuo et al., 2009). Such studies cannot accurately reflect the situation over a large region. Little information has been reported about the progressive changes in soil rehabilitation projects during periods as long as 50 years. The objectives of the present study were 1) to determine the rate of change in various chemical and biochemical soil properties during sand land rehabilitation, including nutrient contents and activities of dehydrogenase, catalase, α - and β -glucosidase, protease, and phosphatase, and 2) to identify the rates of change of soil properties in inter-shrub areas over a span of 50 years, when sand-fixing was first used on desertified land.

2. Materials and methods

2.1. Site description

Field investigation and sample collection were conducted at the Shapotou Desert Research and Experiment Station (SDRES) of the Chinese Academy of Sciences (CAS) (37°32'N, 105°02'E, 1300 masl). The station is located in the Shapotou–Hongwei area at the southeastern edge of the fourth largest desert in China (Tengger Desert with 36,000 km² area), near the semi-arid agro-pastoral transitional zone of Northwest China. The Shapotou region has evolved complex, stable, productive, rain-fed, artificial, and natural ecosystems capable of reversing desertification from the simple, artificial vegetation system in the arid desert region (Chen and Duan, 2009; Duan et al., 2004; Li et al., 2004; SDRES, CAS, 1991; Zhang et al., 2004). Shapotou is referred to as one of the most successful models for desert control and ecological restoration in the arid desert region of China and probably of the world (Li et al., 2010).

The SDRES area (37°27'N, 104°57'E) is at an elevation of 1339 m a.m.s.l., in the steppe desert zone and can also be described as an ecotone between desert and oasis (Li et al., 2010). The annual mean temperature is 10.0 °C, the mean low temperature is –6.9 °C in January, and the high is 24.3 °C in July. The mean annual precipitation was 186 mm, and approximately 80% of this occurs between May and September. The annual potential evaporation is approximately 2900 mm. The mean annual wind velocity is 2.9 m/s. The growing season ranges from 150 to 180 days between the last frost in mid-April and the first frost in late September. The primary soils in the SDRES area are Orthic Sierozem and Typic Psammaquent (Chen et al., 1998; FAO/UNESCO, 1974). The dominant vegetation species are annual plants (*Agriophyllum squarrosum* Moq. and *Hedysarum scoparium* Fisch) and shallow-rooting shrubs (*Artemisia ordosica*), and natural vegetation has approximately 1% surface coverage (Li et al., 2004).

The fields investigated in this study lie in the artificially re-vegetated desert area (Li et al., 2002). The non-irrigated vegetation protective system was established in the 1950s. Initially, barriers to sand movement were established using a matrix of straw ropes in a checkerboard pattern. The straw checkerboards held in place almost 99% of the quantity of sand previously transported over the mobile dunes. Shrubs were planted in the checkerboards in different years (1956, 1964, 1975, and 1987) and the successful revegetation over this time period has attracted the attention of many scientists who now seek to study the

ecological patterns and processes. The stabilized sand surface provided better conditions for all plants and promoted airborne dust deposition onto the surface, which led to the formation of soil bio-crust (Li et al., 2002). The mobile dune-dominated landscape has been turned into a stable, complex desert ecosystem and the evolution of micro-habitats has been promoted (Wang et al., 2006).

2.2. Experimental design and soil sampling

Soil samples were collected from different sites representing different time sequences from 20 m × 20 m plots in August of 2007. Soil samples (superficial crust plus three subsurface layers at depths of 0–5 cm, 5–15 cm, and 15–35 cm) were collected with four replicates from five sites including one shifting sand site and the dunes restored in 1956, 1964, 1975, and 1987. (i.e., dunes stabilized and replanted for 50, 43, 32, and 20 years). A shifting sand site was chosen outside the stabilized area, where the main landscape type was high and dense reticulate chains of barchan dunes. There were no soil crusts and plants in mobile dunes (Li et al., 2014). The replanting dates were chosen to examine the effect of time on sand soil development. These sites were restored with very similar treatments by planting seedlings of the same shrub species with the same density in similar straw checkerboards. The selected sequence represents the different stages of transformation from the relatively simple primary sand-binding vegetation into a functional ecosystem with a complex structure and composition (Li et al., 2002). Previous research papers have noted that in addition to the surface crust, there have also been differentiations of the subsurface sands into separate horizons (Duan et al., 2004). As a result, we set three depths down to 35 cm.

All samples were gently mixed, sieved through a 2 mm screen to remove the root material and other debris, and stored in polyethylene bags. Half of each sample was air-dried and stored at room temperature for analysis of chemical and physical soil properties. The other half of each sample was kept field-moist in a cooler at 4 °C for analysis within two weeks to allow determination of soil biological properties.

2.3. Measurements

2.3.1. Soil chemical properties

All methods for pH, total C, N, and P and available N, P, and soil moisture were described in detail in Zhang et al. (2010). Briefly, the pH of soil samples was measured with a suspension of soil in distilled water (1:2.5) by a glass electrode. Total C and N were determined using an Elementar Vario EL analyzer (Matejovic, 1995), and total P was determined by the H₂SO₄–HClO₄ digestion method (Kuo, 1996). Available P was determined using the Olsen method with NaHCO₃ as an extractant (Kuo, 1996), and available N was extracted with KCl (2 mol·L⁻¹) and analyzed by one Continued Flow Analysis (CFA) (Miller and Keeney, 1982).

2.3.2. Soil enzyme activities

Soil enzyme activities were measured with colorimetric determination methods described in Zhang et al. (2010). Briefly, the soil dehydrogenase activity was measured by using triphenyltetrazolium chloride (TTC) (Sigma-Aldrich Inc., US) as the substrate (3:100 substrate:water, w/v) and expressed as $\mu\text{g TPF g}^{-1}$ soil 24 h (Tabatabai, 1994). The catalase activity was determined by spectrophotometry via the measurement of hydrogen peroxide breakdown (Trasar-Cepeda, 1999). The protease activity was determined using casein (Sigma-Aldrich Inc., US) as the substrate (Ladd and Butler, 1972). The phosphatase, α -D-glucosidase, and β -D-glucosidase activities were measured by colorimetric determination of the released *p*-nitrophenol (Tabatabai, 1994), with sodium *p*-nitrophenyl phosphate (Seebio Biotech Inc., China), *p*-nitrophenyl α -D-glucoside (J&K China Chemical Ltd.,) and *p*-nitrophenyl β -D-galactoside (Sigma-Aldrich Inc.) as substrates. For phosphatase, α -D-glucosidase, and β -D-glucosidase activity measurements, controls were also included, in which substrates were added after the soil sample incubation and prior to analysis.

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