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Research article

Enzyme activity and microbial biomass availability in artificial soils on rock-cut slopes restored with outside soil spray seeding (OSSS): Influence of topography and season

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A B S T R A C T

Large-scale railway construction has resulted in large areas of bare-cut-slope, and outside soil spray seeding (OSSS), a frequently used technique, has been adopted for slope restoration for many years. However, compared with natural slope soils, the quality of artificial soils on rock-cut slopes is low. Enzyme activity and microbial biomass are the main indices used for estimating soil quality; thus, our objective was to explore the influence of slope position, slope aspect, and season on two important factors that positively influence the plant growth capability in artificial soil. Further, we suggest modifications of the proportions of OSSS ingredients, not only to manage cut slopes more economically but also to provide a new framework for managing desertification.

We chose a bare-cut-slope that had been restored five years ago near the Suiyu Railway (Chongqing-Suining), in Sichuan Province, China, as our study plot. Soil samples were collected at a depth of 10 cm. We conclude that natural slopes exhibited higher urease, sucrase, and catalase activity and higher microbial biomass than cut slopes. The protease and polyphenoloxidase enzyme activities and the microbial biomass were higher on the cut slopes in the months of October and January, with the highest protease activity in October, and the highest polyphenoloxidase activity in January. The enzyme activity and microbial biomass were always lower on lower slopes, with the exception of polyphenoloxidase activity. The slope aspect influenced soil enzyme activity, resulting in higher activity on north-facing slopes than on south-facing slopes. These results provided scientific support for artificial revegetation methods in an ecological context.

1. Introduction

China is a mountainous country, with hilly regions and rugged plateaus covering over 50% of the total land area. Sichuan Province is characterized by excessive mountains and plateaus, which cover over 60% of the area in the region. The altitude exceeds 1000 m, and hills and rugged plateaus cover 94.7% of the area in the province (Chen et al., 2009; Ma et al., 1993). In recent decades, the necessity for transport has led to numerous linear construction projects, which have created large expanses of rock-cut slopes. These features have not only caused numerous ecological problems but also threatened the safety of highway and railway transportation (Hazzard, 1998; Sutejo and Gofar, 2015).

Soil properties of cut slopes undergo extensive changes due to destruction of the original topsoil and vegetation. Bare rock-cut slopes also result in a loss of biodiversity and can lead to slope collapse (Andrés et al., 2010; Arnaez et al., 2011). An awareness of this environment is needed to solve this problem. Outside soil spray seeding (OSSS) is a useful technology that consists of spraying

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artificial soil mixtures onto cut slopes to a depth of 10 cm to supply root anchorage and nutrient sources for new vegetation (Hai-Yan, 2008; Gao et al., 2016). New vegetation forms after several years on the rock fragments on the artificial soil slopes (Ai et al., 2012; Marumoto and Kohno, 2017). Many experiments have been conducted to evaluate the relation between artificial soil structures and the potential for revegetation (Chen et al., 2016). However, one potential problem with these studies is that the soil quality of cut-slopes is always lower compared to soil quality of natural slopes. It is necessary for us to evaluate the effectiveness of this method and modify its process.

Previous studies on the artificial soil of cut slopes mainly focused on optimizing artificial soils by evaluating the composition of artificial soils (Gao et al., 2007; Chen et al., 2015, 2016) and the enrichment of heavy metals in the soils (Liu et al., 2009; Chen et al., 2014; Brookes, 1995). Soil nutrients are closely related to soil enzyme activity (Sihi et al., 2017; Freeman et al., 2001a). Soil enzymes can increase the rate of the breakdown of macro-organisms in soil to make them available for plants and microorganisms (Li et al., 2016; Cheng et al., 2017). The soil microbial biomass is a primary factor that influences soil enzyme activity (Li et al., 2017; Lu et al., 2017). As an active part of the soil organic matter and source of available nutrients, the microbial biomass builds up with the increased accumulation of organic matter during soil development (Kaiser et al., 1992; Diaz-Ravina et al., 1993; Hassink, 1994). However, there are no published reports regarding the differences in the enzyme activity and microbial biomass of artificial soils on cut slopes for different topographies and seasons. Previous studies of soil enzymes and soil microbial biomass have mainly focused on the soils of cropland (Mahmood et al., 1997; Zhang et al., 2017) and of dry tropical agro-ecosystems (Sugihara et al., 2010; Ren et al., 2017). The effects of these factors on railway cut slopes have not yet been explored.

In our paper, we explored the spatial and seasonal variation in enzyme activity and investigated the links between these variables based on previous studies, with the purpose of providing a scientific knowledge base for guiding ecological restoration. Therefore, the objectives of this study were as follows: (1) to assess the seasonal fluctuation in soil enzyme activity and microbial biomass C, N, and P, on a railway rock slope protected by an OSSS artificial soil mixture compared with a natural slope; (2) to investigate the responses of the microbial biomass and enzyme activity to different slope positions on a railway rock slope protected by an OSSS artificial soil mixture and on a natural slope; and (3) to describe the relationship between soil enzyme activity and microbial biomass.

2. Materials and methods

2.1. Study site and sample collection

The experiment site is located in Suining, Sichuan Province, China in a hilly region near the Suining railway station (lat. 30°32′ N, long. 105°32′ E) with a gradient of approximately 45°. The slope at our study site had been restored five years ago, along with the infrastructure of the Chongqing-Suining Railway. The mean annual temperature in this area is 17.4 °C. The hottest month is August, with an average temperature of 27.2 °C and an extreme maximum temperature 39.3 °C. The coldest month is January. In this month, the average temperature is 6.5 °C and the extreme minimum temperature is −3.8 °C. The area is within a typical humid subtropical monsoonal climate and has an average annual precipitation of approximately 927.6 mm. Its average annual wind velocity is 0.7 m/s. The bottom layer of the bedrock is limestone, and the top layer is mudstone covered by purplish soil (Fig. 1).

We selected three similar slopes with different ecological context for this study:

a) Rock-cut slope (RS): protected by an OSSS artificial soil mixture containing rock fragments.
b) Natural slope (NS): original slope without human disturbance.
c) Agricultural slope (AS): used for farming.

The experiment conducted in this study was performed in January, April, July and October, with four replications using a randomized sampling strategy. The spray thickness of artificial soil was 10 cm therefore, soil samples were collected from a depth of 10 cm at three different slope positions (upper slope, US; middle slope, MS; and lower slope, LS) for the estimation of soil enzyme activities and soil microbial biomass. Four replicate soil samples were collected at each of the slope position at points separated by a distance of 100 m. Stones and plant roots were carefully removed following the removal of the surface organic materials, and each moist composite field soil sample was mixed, homogenized and sieved through a 2 mm mesh screen. The microbial biomass analysis was performed using moist field subsamples stored at 4 °C in a refrigerator.

2.2. Soil enzyme analysis

Five enzymes were tested in our study: urease, sucrase, catalase, protease and polyphenoloxidase. The urease activity was calculated using the methodology in Kandeler and Gerber (1988), the activity of soil sucrase was assayed by ammonium molybdate colorimetry (Guan et al., 1984), the protease activity was tested by the method of Duly and Nannipieri (1998), the catalase activity was tested using the method of Aebi (1984), and the polyphenoloxidase activity was determined according to the methodology in Perucci et al. (2000) and expressed as mg purpurogallin g^{-1} soil per 2 h.

2.3. Determination of soil microbial biomass

The soil microbial biomass C and N, were estimated using the chloroform fumigation extraction methods. Microbial biomass carbon (MBC) was determined by using the potassium dichromate titration method (Vance et al., 1987) and was calculated by the following equation:

$$MBC = Ec/KEc \quad (1)$$

where Ec is the difference between C extracted from unfumigated and not fumigated soils and KEc is 0.38, accounting for C in the microbial cell walls that is not released by the chloroform.

Ninhydrin N released from the microbial cells was determined colorimetrically at 570 nm (Joergensen and Brookes, 1990).

Microbial biomass nitrogen (MBN) was calculated by using the following equation:

$$MBN = En \times 5.0 \quad (2)$$

where En is the difference between N extracted from fumigated and unfumigated soils.

Microbial biomass phosphorus (MBP) was calculated as follows (Brookes et al., 1982):

$$MBP = EPi/(Kp \times RPi) \quad (3)$$

where EPi is the difference between P extracted from fumigated and unfumigated soils, Kp is 0.4, accounting for the extraction efficiency of microbial cells, and RPi is the proportion of added Pi recovered from each spiked unfumigated soil sample.