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Effects of rhizosphere interactions of grass interspecies on the soil microbial properties during the natural succession in the Loess Plateau

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A R T I C L E I N F O

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A B S T R A C T

Natural vegetation succession is a process of interactions between plants and various environmental factors. The dynamics of soil microbes during successional stages have been intensively explored in the past two decades. The effect of rhizosphere interactions of interspecies on microbial properties, however, has received less attention. During the early stage of succession on a Loess Plateau soil from China, we investigated microbial properties in the rhizospheres of three dominant grass species: *Artemisia capillaris*, *Artemisia sacrorum* and *Stipa bungeana*. Experiments with monocultured and polycultured potted plants were designed to determine the effects of rhizosphere interactions on plant growth and soil microbial properties. The results showed that root biomass, shoot biomass, plant height, microbial biomass carbon (C), microbial biomass nitrogen (N), invertase and urease activities, Gram-negative and Gram-positive bacteria were significantly higher for *A. capillaris* than for *A. sacrorum* and *S. bungeana* in both monocultures and polycultures. Lower root biomass, shoot biomass, plant height, microbial biomass C, microbial biomass N, invertase and alkaline phosphatase activities, and levels of microbial phospholipid-derived fatty acids in polycultures relative to the monocultures indicated that rhizosphere interactions led to lower plant growth and soil microbial activities. Our study suggests that rhizosphere interactions had a significant effect on the microbial properties of plant rhizospheres. The advantage of *A. capillaris* over *A. sacrorum* and *S. bungeana* in growth characteristics and rhizosphere microbial conditions is likely responsible for the dominance of *A. capillaris* in the early stage of succession in abandoned cropland on the Loess Plateau.

1. Introduction

The deterioration of natural ecosystems has accelerated during the last century due to increasing human activity and the extensive use of natural resources [1]. Natural recovery without further anthropogenic disturbance has been assumed to be the most effective way to restore disturbed soil ecosystems, because native plant species are better able to adapt to poor and changeable environmental conditions [2]. Natural succession is generally characterized by an initial dominance of some species, subsequent co-occurrence with other species and substitution by another dominant species with time. Finally, the ecosystem develops to a stable community of species adapted to the biotic and abiotic conditions. Vegetation succession is a slow ecological process and is completely dependent on the interactions between vegetation and the various environmental factors [3].

The rhizosphere is the soil region influenced by plant roots and characterized by high microbial activity [4]. Rhizosphere microbial activities are critical for the establishment of vegetation and soil nutrient cycling [5,6]. For instance, microbial biomass, as one of the living component of soil organic matter, responds rapidly to changes in the soil environment because of this high turnover rate [7]. Basal respiration is widely used as an indicator of microbial activity. Invertase is a type of hydrolase enzyme involved in the decomposition of organic matter and releases reducing sugars as end products of the carbon (C) cycle [8]. Alkaline phosphatase participates in soil phosphorus (P) cycling and transforms organic P into inorganic P, which is then available to plants. Urease has a vital role in soil nitrogen (N) cycling [9]. Given these important functions, there is considerable interest in understanding the linkage between microbial performance, soil variables and plant growth. As an available approach for determining microbial community composition, analysis of phospholipid fatty acids (PLFAs) uses the lipids of microbial membranes as biomarkers for specific groups of microorganisms [10]. Rapid changes in soil microbial community structure can be effectively detected by changes in PLFA patterns. In general, microorganisms play an important role in the re-establishment of vegetation and long-term ecosystem stability; however, recent evaluation of the success of revegetation by natural succession has been limited to visual inspection of aboveground indicators and to

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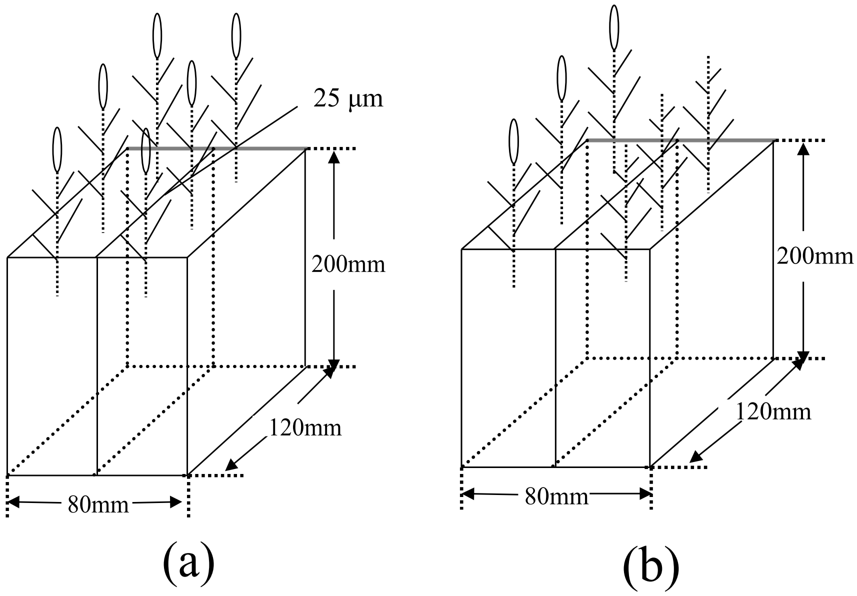

Fig. 1. Diagram of the rhizobox. (a) monoculture; (b) polyculture.

monitoring of soil parameters. Little attention has been given to microbial rhizosphere interactions between plants, and lack of such knowledge may lead to an incomplete understanding of ecosystem recovery.

The Loess Plateau of China has experienced a significant change in land use during the past 50 years. Historically, the native vegetation was destroyed to meet the food supply needs of an expanding population, resulting in severe soil erosion and land degradation. An ambitious conservation project, Grain for Green, was implemented by the Chinese government in 1999 to control erosion and restore soil quality, and large areas of sloping cropland have been restored to grassland or forest. The sloping cropland was abandoned for natural recovery without further human disturbance as an important measure of restoration. This natural restoration has led to an increase in vegetation cover as the abandoned fields were naturally recolonized by surrounding vegetation [11]. Intensive studies have indicated that this type of conversion can greatly improve soil aggregate characteristics [12,13]; C, N and P supply [14,15]; enzyme activities [16]; and microbial compositions [11] of the abandoned sloping cropland. These studies, however, mainly focused on the outcome of natural succession, such as the effect of recovery on vegetation communities and soil conditions, but the causes of the successional order and the interactions of microbial communities of plants remain unknown. Such information is important for a better understanding of natural succession and of the interactions among plant communities and for appropriately managing the ecological environment.

Our recent studies have shown that *Artemisia capillaris*, *Artemisia sacrorum* and *Stipa bungeana* are the three dominant species in communities during the early stage of succession on abandoned cropland of the Loess Plateau [17]. Notably, in the initial successional stage (1–10 years), *A. capillaris* was dominant, and *A. sacrorum* and *S. bungeana* were subdominant. The present study investigated the microbial properties of *A. capillaris, A. sacrorum* and *S. bungeana* rhizospheres in experiments with potted plants. We hypothesized that (i) rhizosphere interactions of interspecies could lead to a great variation in rhizosphere microbial properties and (ii) the *A. capillaris* rhizosphere would have higher levels of microbial biomass, enzyme activities and PLFAs compared with *A. sacrorum* and *S. bungeana*.

2. Materials and methods

2.1. Soils

Pot experiments were conducted in a greenhouse at the State Key Laboratory of Soil Erosion and Dryland Farming on the Loess Plateau, Northwest A&F University, China. The soil for the experiments was collected from the Ansai Research Station of Soil and Water Conservation in the Dunshan watershed, Shaanxi Province, on the northern Loess Plateau (109°19′23″E, 36°51′30″N). All soils were taken from the surface layer (0–20 cm) of abandoned cropland that had been allowed to experience natural succession for five years. The soils were air-dried, ground and sieved through a 2 mm plastic mesh. The chemical properties of soil were as follows (means of three determinations ± standard deviations): organic C, 3.24 ± 0.24 g kg^{-1}; total N, 0.29 ± 0.02 g kg^{-1}; total P, 0.51 ± 0.02 g kg^{-1}; available P, 1.17 ± 0.09 mg kg^{-1}; available N, 43.79 ± 3.61 mg kg^{-1}; and pH, 8.55 ± 0.14.

2.2. Experimental design

Rhizoboxes are increasingly used to investigate the properties of rhizospheres, because they avoid the overestimation of microbial biomass common to the root-shaking method and overcome the disadvantage of removing soluble material by washing roots [18,19]. We constructed rhizoboxes to investigate the properties of the rhizospheres of monocultured and polycultured plants. Three plant species common in the early stage of natural succession of abandoned cropland on the Loess Plateau (*A. capillaris*, *A. sacrorum* and *S. bungeana*) were planted in rhizoboxes as either monocultures or polycultures.

2.2.1. Monoculture experiment

The dimensions of monoculture rhizoboxes were 120 × 80 × 200 (length × width × height, mm) (Fig. 1a). The boxes were divided into left and right rhizosphere zones (40 mm in width) separated by 25-µm mesh nylon cloth. This design separated the soil zones and successfully prevented root hairs from entering the adjacent soil zones but permitted the transfer of soil microfauna and root exudates between the zones. Three species were respectively monocultured: *A. capillaris*, *A. sacrorum* and *S. bungeana*. A pot experiment without planting was conducted as a reference. Each rhizobox was filled with 3 kg of treated soil. Seeds of each species were thoroughly rinsed with water, and germinated on filter paper. After seeds were germinated for 24 h at 28 °C in darkness,