



No synergistic effects of water and nitrogen addition on soil microbial communities and soil respiration in a temperate desert



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ABSTRACT

Soil microbial communities play an important role in regulating land–atmosphere CO₂ exchange in terrestrial ecosystems. However, their responses to climate change are unclear. We explored the effects of water and nitrogen addition and their interaction on soil microbes and the resulting impacts on soil carbon emissions in the Gurbantunggut Desert, northwestern China. A manipulative 30% increase in precipitation and 5 gN m⁻² year⁻¹ deposition alone and in combination was applied across three years from 2011 to 2013. Water addition significantly increased microbial biomass and respiration, metabolic quotient, and the utilization of carbohydrates, carboxylic acids and amino acids. Water addition did not change the microbial community composition. Nitrogen addition only significantly increased soil bacterial PLFAs, while exerting no significant impacts on soil fungal PLFAs, microbial respiration and soil respiration. Moreover, nitrogen addition had no significant impacts on microbial community composition. Water and nitrogen addition in combination did not generate synergistic effects on microbial communities and soil respiration. Across treatments in three years, soil respiration and microbial respiration were positively correlated with microbial total PLFAs, microbial carbon utilization profiles, while being independent of microbial community structure. This study suggests water addition can increase soil carbon emission through increasing microbial utilization of labile carbon, and water plus nitrogen addition had no synergistic effects on soil microbial communities and soil respiration, demonstrating nitrogen is not a limiting factor for soil microbial communities in the scenario of increasing precipitation in this desert ecosystem.

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1. Introduction

Soil microbial communities act as a profound carbon sink in soils, and are also a mediator of soil organic matter decomposition (Schimel et al., 2007). Because of the importance of soil microbial communities in soil carbon emission and nutrient dynamics, they are gaining increasing concern (Manzoni et al., 2012a; Delgado-Baquerizo et al., 2013; Serna-Chavez et al., 2013). However, studies exclusively focusing on desert ecosystems are not abundant, and results are not consistent, which greatly hampers our understanding of soil carbon dynamics in the scenario of climate change (Insam and Ranggner, 1997; Collins et al., 2008; Bell et al., 2014).

Desert ecosystems are characterized by low water and substrate availability to microbial organisms, and soil microbial activities are largely inhibited due to the low water availability and carbon supply (Austin et al., 2004). In some desert ecosystems, soil microbial biomass is under detectable at dry period, and can reach 15.3 mg N kg⁻¹ soil in the wet duration (Gallardo and Schlesinger, 1992, 1995). The varying

microbial biomass with soil moisture may exert profound impacts on soil carbon emission. Under drought, soil microbes must accumulate solutes to reduce the water potential in internal cells, which may lead to a decreased microbial respiration and soil carbon emission (Manzoni et al., 2012a; Taylor et al., 2012b). However, when moisture is available, soil microbes dispose osmolytes rapidly, which usually generates a pulse of CO₂ at the ecosystem level (Su et al., 2013). Besides of the microbial biomass, altered soil microbial community with water availability may also elicit the variation of soil carbon emission (Six et al., 2006; Schimel et al., 2007; Manzoni et al., 2012a) due to the contrasting carbon use efficiencies between different functional groups (Sinsabaugh et al., 2013).

Soil organic matter quantity can determine the degradation ability by soil microbes and subsequent carbon emission from the soil. The two most important elements affecting microbes in deserts are carbon and nitrogen, and studies are controversial in terms of their relative importance across ecosystems (Sinsabaugh et al., 2013). Similarly, manipulative experiments also showed contrasting responses of microbial biomass and respiration to altered soil substrates. For instance, 8 years of N addition of 3 g m⁻² year⁻¹ did not alter the microbial community composition, while significantly reducing microbial biomass by 18% in a

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temperate forest (DeForest et al., 2004). In contrast, N application of 2 mg of $\text{NH}_4\text{NO}_3\text{-N}$ g wet soil⁻¹ only exerted transitory negative effects on microbial respiration and biomass (Soderstrom et al., 1983). Despite that N addition can trigger soil acidification, which usually exerts negative effects on soil microbial biomass and activity, these studies suggest that nitrogen availability effects on soil microbial communities deserve further studies.

The Gurbantunggut Desert is a temperate desert and located in the center of the Eurasian Continent. Because of the high vegetation cover of shrub-steppe community, this desert provides many ecological and economic services to local inhabitants. It has been predicted that precipitation will be continuously increasing through 2030 in this region (Liu et al., 2010). Besides, atmospheric nitrogen deposits at a rate of 3.6 gN m⁻² year⁻¹ in this desert due to the intensive agricultural activities adjacent to the desert in the past half century (He et al., 2007). Increasing water and nitrogen availability in this desert ecosystem may alter soil microbial communities and thus change soil carbon emission. However, few studies have been aimed to understand their potential impacts (Zhou et al., 2012). To elucidate the dynamics of microbial responses and their implications for soil carbon emission in the context of increasing precipitation and nitrogen deposition, we established a field experiment with manipulations of a 30% increase in precipitation and 5 gN m⁻² year⁻¹ deposition in the Gurbantunggut Desert in 2011–2013. Our previous observations have shown that water addition can significantly promote soil moisture and plant growth, implying that water addition can alleviate the water limitation and increase respiratory substrates to soil microbes (Huang et al., 2015). Thus, our first hypothesis is water addition can stimulate microbial growth and respiratory activity, and therefore soil respiration will be increased. Nitrogen addition can increase soil inorganic nitrogen content, which can change some important elemental ratios, primarily soil C:N. Considering that the primary functional groups of soil microbes have contrasting stoichiometric ratios and carbon use efficiencies (Keiblinger et al., 2010), our second hypothesis is that N addition can change the microbial community structure and microbial respiratory activities, leading to a different soil respiration as compared to the control. Significant promotion of plant growth can be achieved under nitrogen addition when water availability is high, which is thereafter favorable for soil microbial growth, thus our third hypothesis is that concurrent additions of water and nitrogen have synergistic effects on soil microbial communities and soil respiration.

2. Materials and methods

2.1. Study site description

The experiments were conducted in the vicinity of the southeastern Gurbantunggut Desert, northwestern China (44°17'N, 87°56'E, 475 m a.s.l.). This region has a continental arid, temperate climate, with a hot, dry summer and cold winter. The annual mean temperature is 6.6 °C and the annual mean precipitation is 160 mm, in which 70% to 80% fall in April–September. The annual precipitation and mean air temperature were 167.4 mm and 6.4 °C, 102 mm and 6.8 °C, and 133.7 mm and 7.7 °C in 2011, 2012, and 2013, respectively (Fig. S1). The pan evaporation is 2000 mm. The soil great groups are Torripsamments, under the soil order of Entisols; and Haplocalcids, under the soil order of Aridisols (USDA Soil Taxonomy). The soil is silt loam texture, with 81.7% sand, 16.8% silt and 1.5% clay. Soil organic carbon content is 2.4 g kg⁻¹. Soil pH is 9.5. The plants are *Tamarix ramosissima*, *Haloxylon ammodendron*, *Haloxylon persicum*, *Alyssum linifolium*, *Leptaleum filifolium*, *Erodium oxyrrhynchum*, *Myosotis scorpioides*, *Eremurus indiensis*, *Salicornia brachiata*, and *Ceratocarpus arenarius*, with a cover of 30%. Soil surface was covered by biological soil crusts, which are an intimate association between soil particles and cyanobacteria, algae, microfungi, lichen and bryophytes in different proportions (Li, 2012). Its cover reaches 40% in the study site (Su et al., 2013).

2.2. Experiment design, sample collection, soil and plant properties measurements

Twenty-four 10 × 10 m plots were established and equally distributed in six blocks. Within each block, control (CK), water addition (W), nitrogen addition (N) and water plus nitrogen addition (WN) treatment were applied following a randomized block design. The distance between two adjacent plots was 10 m. In each W and WN treatments, precipitation increased 30%, according to predictions for northern China over the next 30 years (Liu et al., 2010). The 30% extra precipitation was collected using “rainfall collection pans”. The pans were constructed from galvanized iron sheets, with an area of 1.9 × 1 m, totaling 18 pans was installed for each plot, the total area of pans was equivalent to 30% of the area in the plot, corresponding to the added water of 50.2 mm, 30.6 mm, and 40.1 mm in 2011, 2012, and 2013, respectively. Each pan was erected at a slight angle, and the rainfall intercepted was collected in a bucket that was buried in the soil. Immediately after a rainfall event, the collected rain was evenly sprayed onto the plots in the early morning or late afternoon to prevent excessive evaporation. Moreover, given the ecological significance of snowmelt in our site (Fan et al., 2014), snow fallen in the pan was also evenly added in the corresponding plot in early spring. In the N and WN treatments, N was applied in liquid form, 1667-g NH_4NO_3 was diluted in 15-L distilled water and evenly sprayed (equal to 0.15 mm rainfall) onto the corresponding plots (10 × 10 m) in early-April and mid-July, corresponding to the rapid growth duration of spring ephemerals and summer annuals in the study site (Huang and Li, 2015), which aimed to eliminate N limit to plant growth. The same amount of distilled water was added in the CK and W treatments. N (5 gN m⁻² year⁻¹) application was based upon the real mean airborne N deposition rate (3.6 gN m⁻² year⁻¹) registered in Xinjiang, northern China over the past 10 years (He et al., 2007). All experimental design and instrument arrangements were applied in 2010. Moreover, before the application of water and nitrogen treatments, vegetation and soil nutrient concentrations (soil organic matter, soil total nitrogen, soil total phosphorus, soil potassium and inorganic nitrogen) all showed no statistically significant difference between treatments using block as a covariance in ANCOVA analysis, suggesting no heterogeneity among the blocks undergone four treatments.

Five soil cores (5 cm diameter, 0–5 cm depth) were collected from each plot to get a composite sample on August 10th in 2011, 2012 and 2013. In desert ecosystems, because soil microbial communities exhibit large variations by water availability, we collected soil samples at the day without rainfall occurred in the prior five consecutive days. After removing plant roots and large stones using a 2-mm sieve, soil samples were packed into a portable refrigerated box and transported to the laboratory for soil and microbial properties measurements.

Soil nitrate-N (NO_3^- -N) and ammonium-N (NH_4^+ -N) were extracted with 2 M KCl (Sala et al., 2012; Reichmann et al., 2013) and measured with Auto Analyzer 3 (AA3, BRAN-LUEBBE Ltd., Hamburg, Germany). Dissolved organic carbon (DOC) was extracted by adding 50 mL of 0.5 M K_2SO_4 to subsamples of 12.5 g homogenized soil, and by agitating it on an orbital shaker at 120 rpm for 1 h. The filtrate was analyzed using a TOC analyzer (multi N/C 3100, Jena, Germany). Soil pH was measured using a 1:5 dilution of soil:water (ISSCAS, 1978).

Soil respiration was measured with Li-Cor 840 portable photosynthesis systems (IRGA; LI-840, LiCor Inc., Lincoln, NE, USA) equipped with a chamber. The chamber dimension was 0.5 × 0.5 m at the base and 0.3 m in height. This method is widely used and provides valuable information that cannot be obtained easily in other ways (Risch and Frank, 2007). The chamber walls were constructed of clear polyethylene sheeting, and when measuring soil respiration, the chamber was covered with an opaque cloth. Two electric cooling fans were installed to circulate air in the chamber during measurements. Aluminum collars (ca. 3 cm deep) with a groove for chamber

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