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Influences of N deposition on soil microbial respiration and its temperature sensitivity depend on N type in a temperate forest

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ABSTRACT

Knowledge on temperature sensitivity (Q_{10}) of soil microbial respiration is crucial to improving the accuracy in predicting soil organic carbon (C) dynamics in climate–C models. However, the responses of soil microbial respiration and its Q_{10} to nitrogen (N) deposition, particularly different N types, remain unclear. Therefore, we incubated surface soils collected from a temperate forest receiving simulated N deposition under 15 °C and 25 °C for 150 days to examine the effects of different types of N deposition on soil microbial respiration and its Q_{10} and reveal their underlying mechanisms. A mixture of inorganic and organic N had the highest suppression on soil microbial respiration, followed by organic N and inorganic N. This suggested that the suppression effect of atmospheric N deposition on microbial respiration was underestimated by previous studies based on single inorganic N. Q_{10} values in all soils ranged from 1.96 to 2.76 with a mean of 2.41 at the end of incubation. Inorganic N significantly increased the averaged Q_{10} values, suggesting that inorganic N caused soil microbial respiration to become more sensitive to climate warming than organic N. Across the incubation period, Q_{10} values exhibited substantial temporal variation, which depended on the N type. Soil microbial respiration was negatively controlled by NO_3^--N and bacteria:fungi and gram-positive:gram-negative bacteria ratios. However, Q_{10} was positively controlled by soil NH_4^+-N. Our results highlighted the effects of inorganic and organic N deposition on microbial respiration and its potential mechanisms and implied the necessity of considering the N type when predicting soil C cycling and dynamics in increasing N deposition scenario.

1. Introduction

Soil microbial respiration is a key process controlling the size of the soil organic carbon (SOC) storage and C loss from terrestrial ecosystems to the atmosphere (Bond-Lamberty and Thompson, 2010). A small change in the rate of soil microbial respiration may have a large effect on the net C flux and climate dynamics because of the large SOC stored in terrestrial soil systems (Bonan, 2008). Soil microbial respiration is primarily controlled by temperature, and its response to warming is called temperature sensitivity (Q_{10}). Q_{10} is an important mechanism for the possible feedback between the C cycle in terrestrial ecosystem and climate change (Davidson and Janssens, 2006). Furthermore, a small variation in Q_{10} can cause a large deviation in estimating CO_2 release from the soil into the atmosphere (Xu and Qi, 2001). Therefore, understanding the patterns and controlling the mechanism of soil microbial respiration and its Q_{10} under global warming is important to improve the accuracy of predicting changes in global C cycles and its feedback to climate change.

In the past several decades, many works on microbial respiration have been conducted to investigate the patterns and controlling factors of soil microbial respiration via laboratory incubation and field measurement (Thirukkumaran and Parkinson, 2000; Weand et al., 2010; Wang et al., 2017a). Previous studies demonstrated that soil microbial respiration is closely affected by environmental factors such as temperature, microbial community composition, and nutrient availability, especially in C-rich, N-limited temperate forest ecosystems (Janssens et al., 2010; Coucheney et al., 2013; Du et al., 2014). These environmental factors may be changed by N deposition (Ramirez et al., 2010; Wang et al., 2017b) and consequently influence soil microbial respiration. These pioneering studies have substantially improved our

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understanding of soil microbial respiration and its response to increasing N deposition. However, the direction and magnitude of the N deposition effect in different studies is controversial (Conde et al., 2005; Janssens et al., 2010; Tu et al., 2013; Wang et al., 2017a). This inconsistency suggests that the responses of soil microbial respiration to N deposition vary among ecosystems and soil types, which calls for further investigation across wide ecosystems. Furthermore, this inconsistency may be attributed to differences in N types used in various experiments.

In the recent decade, Q_{10} of soil microbial respiration has gained increasing attention because of its significance in regulating soil C cycling (Davidson and Janssens, 2006; Conant et al., 2008). Some incubation studies illustrated that soils with high substrate quality have low Q_{10} (Fierer et al., 2005; Conant et al., 2008; Craine et al., 2010; Liu et al., 2017). This supports the C quality–temperature hypothesis that Q_{10} increases with increasing biochemical recalcitrance of soil organic matter. The shift in soil microbial community composition caused by N deposition also affects Q_{10} (Thiessen et al., 2013; Karhu et al., 2014). For instance, Karhu et al. (2014) found that the microbial community level response to temperature is more often enhanced than reduced in mid- to long-term (90 days) Q_{10}. These studies to some extent advanced our understanding of the mechanisms that regulate Q_{10}. However, the effects of N deposition on Q_{10} in various experiments are inconsistent (Coucheney et al., 2013; Liu et al., 2016; Wang et al., 2017a). This inconsistency will increase the uncertainty of predicting the response of soil C cycle to global environmental change. More importantly, most previous studies used inorganic or organic N as lone N resources (e.g., Sinsabaugh et al., 2002; Wang et al., 2017a), which may not actually reflect the effects of atmospheric N deposition on soil microbial respiration and its Q_{10} because it contains inorganic and organic N components (Cornell, 2011). However, in temperate forest ecosystems, the mechanism on how inorganic and organic N deposition affects soil microbial respiration and its response to global warming are still unclear. Without this knowledge, our understanding of the drivers of soil CO_2 emission is incomplete.

Therefore, taking the advantage of a long-term N deposition experiment with different types of N deposition in a temperate forest, we conducted an incubation experiment using soils receiving N for a long period of time to assess how inorganic and organic N affect soil microbial respiration and its Q_{10}. We also investigated the underlying mechanisms regulating responses of soil microbial respiration and its Q_{10} to N deposition. In this study, we provided the first investigation to explore the response of soil microbial respiration and its Q_{10} to different types of N deposition in a temperate forest. Here, we aimed to address 1) how N types (organic vs. inorganic) affect soil microbial respiration and its Q_{10} and whether N has an effect on Q_{10} in terms of temporal variation; and 2) which soil chemical and microbial variables are the key factors controlling soil microbial respiration and its Q_{10} under N deposition.

2. Material and methods

2.1. Site description

This work was conducted in a mature temperate forest dominated by *Larix gmelinii*, locating at Laoshan Forest Research Station of Northeast Forestry University in Heilongjiang Province, northeastern China (127°34′E, 45°20′N). The site has a continental temperate monsoon climate, with a strong monsoon windy spring, a warm and humid summer, and a dry and cold winter. Annual precipitation ranges from 600 to 800 mm, most of which falls in July and August. The mean annual air temperature is 2.7 °C, and average air temperature is -19.6 °C in January and 20.9 °C in July. The parent material at the site is granite bedrock and the soil is well-drained Hap-Boric Luvisols (dark brown forest soil in Chinese Soil Taxonomic System) with rich C.

2.2. Design of long-term simulated experiment

In this long-term simulated N deposition experiment, twelve 10×20 m^2 plots were randomly established in April 2010. This experiment has 4 treatments with three replicates: control (CT), inorganic N (IN) deposition, organic N (ON) deposition and their mixture (MN) deposition. Ammonium nitrate was chosen as the inorganic N; while organic N was the equal mixture of urea and glycine (Cornell, 2011), and mixture of inorganic and organic N was used 7:3, which was mostly equal to the average ratio in the atmospheric N deposition (Cornell, 2011). There was a buffer zone of 15 m between any two plots. From May 2010, each plot started to be treated with various N solutions or deionized water. During the field experiment, each plot was sprayed with N solutions at a rate of $10\ g\ N\ m^{-2}\ yr^{-1}$, and control treatment involved addition of only deionized water. N was applied monthly in six equal applications of $1.667\ g\ N\ m^{-2}$ from May to October. In each application, the N fertilizer was dissolved in 50 L of deionized water, and the solution was sprayed with a portable sprayer at the early month. The detail about the experiment was described in Wang et al. (2017b).

2.3. Soil sampling and analysis

In this study, top mineral soils (0–10 cm depth) in each plot were sampled in September 2015. Eight soil cores were randomly collected in each plot using a metal corer with a 5 cm diameter and then mixed into a composite sample. Before sampling, litter on the surface was completely removed. These fresh soil samples were stored in sealed bags and transported immediately to the laboratory and then passed through a 2 mm mesh to eliminate residual organic matter (i.e., decomposed leave litter and roots). Some of each sample was used for incubation to determine soil microbial respiration. The rest of each sample was used for soil chemical and microbial property determination.

Soil organic C and total N concentrations of soil samples were measured using a C/N analyzer (Elementar, Germany). Soil total P was measured colorimetrically, whereas soil mineral N (sum of ammonium and nitrate N) was extracted using 2 M KCL solution and determined by colorimetry. Available soil P was analyzed colorimetrically through molybdate blue method after the soil was extracted with 1 M NH_4F solution. Soil pH was determined using a pH meter in a 1:2.5 (weight:volume) mixture of soil and deionized H_2O.

Soil microbial community composition was measured by phospholipid fatty acid (PLFA) analysis. PLFAs were extracted from the frozen-dried soil according to the method described by White and Ringelberg et al., 1998. Qualitative and quantitative fatty acid analyses were performed with an Agilent 6890 GC and the MIDI Sherlock Microbial Identification System (MIDI Inc., Newark, DE, USA). Fatty acids were quantified by calibration against standard solutions of FAME 19:0 (Matreya Inc., State College, PA, USA). Total bacterial content was derived as the sum of i15:0, a15:0, 15:0, i16:0, 16:1ω7c, 16:1ω9c, 16:0, a17:0, i17:0, cy17:0, 17:0, 18:0, cy19:0, and 20:0 PLFAs (Hill et al., 2000). PLFAs (i15:0, a15:0, i16:0, i17:0, and a17:0) were used as markers for Gram-positive bacteria, whereas PLFAs (16:1ω7c, 16:1ω9c, cy17:0, and cy19:0) were used as markers for Gram-negative bacteria. The 18:1ω9(c,t), 18:1ω7c, and 18:2ω9,12c PLFAs were used as markers for fungi. The 20:4ω6c and 20:2ω6c PLFAs were used as markers for protozoa. The ratios of cyclopropyl (17:0cy + 19:0cy) to monoenoic fatty acids (16:1ω7c + 18:1ω7c) (Cyc/Mon) and total saturated (15:0 + 16:0 + 17:0 + 18:0 + 20:0) to total monounsaturated fatty acids (16:1ω7c + 16:1ω9c + 18:1ω9c + 18:1ω7c) (Sat/Mono) have been used as indicators of nutritional stress in bacterial communities (Bossio and Scow, 1998; Fierer et al., 2003).

2.4. Soil microbial respiration measurement

We quantified the magnitude of CO_2 release from the top soils over an 150-day incubation period. For each soil sample, two microcosms