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The effects of N and P additions on soil microbial properties in paired stands of temperate secondary forests and adjacent larch plantations in Northeast China

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ABSTRACT

The conversion of secondary forests to larch plantations in Northeast China has resulted in a significant decline in soil available nitrogen (N) and phosphorus (P), and thus affects plant productivity and ecosystem functioning. Microbes play a key role in the recycling of soil nutrients; in turn, the availability of soil N and P can constrain microbial activity. However, there is little information on the relationships between available soil N and P and the microbial biomass and activity in larch plantation soil. We studied the responses of soil microbial respiration, microbial biomass and activity to N and P additions in a 120-day laboratory incubation experiment and assessed soil microbial properties in larch plantation soil by comparing them with the soil of an adjacent secondary forest. We found that the N-containing treatments (N and N + P) increased the concentrations of soil microbial biomass N and soluble organic N, whereas the same treatments did not affect microbial respiration and the activities of β-glucosidase, N-acetyl-β-glucosaminidase and acid phosphatase in the larch plantation. In addition, the concentration of microbial biomass P decreased with N addition in larch plantation soil. In contrast, N and N + P additions decreased microbial respiration, and N addition also decreased the activity of N-acetyl-β-glucosaminidase in the secondary forest soil. The P treatment did not affect microbial respiration in either larch plantation or secondary forest soils, while this treatment increased the activities of β-glucosidase and acid phosphatase in the secondary forest soil. These results suggested that microbial respiration was not limited by available P in either secondary forest or larch plantation soils, but microbial activity may have a greater P demand in secondary forest soil than in larch plantation soil. Overall, there was no evidence, at least in the present experiment, supporting the possibility that microbes suffered from N or P deficiency in larch plantation soil.

1. Introduction

Secondary forests, which regenerate naturally following disturbance of primary forests by human beings or by extreme natural causes, account for approximately 50% of the total forests in China (Chen et al., 1994). To meet the growing demands for timber and other forest products, extensive areas of secondary forests in northeast China have been replaced by plantations of predominantly larch (*Larix* spp.) species since the 1960s. For example, there are about 200,000 ha of larch plantations in Northeast China (Liu et al., 2007), forming the mosaic landscapes of plantation-secondary forests. However, these larch plantations have altered soil nutrient availability as a consequence of changes in quantity and quality of plant material entering the soil when compared with adjacent secondary forests (Yang et al., 2013). A previous study reported that declines in soil nutrient concentrations, such as available nitrogen (N) and labile organic phosphorus (P), occurred in larch plantations (Yang et al., 2010, 2012). Generally, N and P are the most common limiting elements in forest primary production and soil biological processes (Allen and Schlesinger, 2004; Elser et al., 2007; Liu et al., 2012). Many studies have shown that the availability of N and P directly controls the productivity of aboveground forests in temperate forest ecosystems (Sverdrup et al.,

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2006; Pregitzer et al., 2008; Thomas et al., 2010), but the nature and extent of the impact of nutrient limitation on soil microbial processes are still poorly understood (Jonasson et al., 1996; Esberg et al., 2010). Given the decline in soil available N and P, larch plantations have received much attention in the role for sustainable forest productivity. Therefore, it is essential to explore whether soil N and P availability in larch plantations can maintain or limit microbial biomass and activities, and to compare these with the secondary forest soil because the secondary forest can provide both timber and provision of ecosystem services with high available N and P in soil. These data will contribute to the understanding of soil nutrient status and may have important implications for the management of larch plantations within the secondary forests.

Microbes play a critical role in the recycling of soil nutrients and soil fertility (Gallardo and Schlesinger, 1994; Galicia and García-Oliva, 2004). Soil microbial biomass and activity are considered to be limited by the availability and quality of carbon (C) (Ilstedt and Singh, 2005; Demoling et al., 2007). However, microbial utilization of C might be constrained by the availability of N or P, or both (Allen and Schlesinger, 2004; Gnankambary et al., 2008; Esberg et al., 2010). N has been found to be a main limiting factor after C in the temperate forest soil (Joergensen and Scheu, 1999). Indeed, the addition of N to temperate forests did not result in consistent effects upon microbial activity (i.e. CO_2 evolution) because positive, negative, and neutral effects all have been reported (Fog, 1988; Arnebrant et al., 1996; Thirukkumaran and Parkinson, 2002; Wallenstein et al., 2006). For example, Wallenstein et al. (2006) reported that N fertilization decreased microbial biomass C and substrate-induced respiration in the forest soils of New England, but Thirukkumaran and Parkinson (2002) found that microbial respiration and substrate-induced respiration were unaffected by N addition in a Canadian Rocky Mountain pine forest soil. Although microbial respiration or microbial growth is usually N limited in temperate forest ecosystems, P limitation on soil microbial biomass has been demonstrated in at least two temperate forest soils (Scheu, 1990; Deforest et al., 2012). These opposing results show that the limiting effects of N, P, or both on microbial biomass and microbial respiration depended on soil characteristics and environmental factors.

Soil enzymes produced by microbes and their response to N or P addition have received considerable attention because these enzymes control nutrient and carbon cycling in soils (Olander and Vitousek, 2000; Zeglin et al., 2007). In general, enzyme production is energy intensive; hence, soil microbes only produce enzymes at the expense of microbial growth and metabolism if the available nutrients are scarce (Koch, 1985). When a nutrient is scarce, microbes can produce enzymes to mobilize resources from substrate sources. Once the concentrations of products increase sufficiently, enzyme synthesis becomes suppressed. Conversely, when a nutrient is abundant, microbes may shift their resources away from the synthesis of nutrient-acquiring enzymes. Thus, nutrient availability is often negatively correlated with the activity of nutrient-acquiring enzymes. This model appears to be supported by negative correlations between phosphatase activity and available P in soil (Olander and Vitousek, 2000; Allison and Vitousek, 2005). For instance, if inorganic P is scarce, then organic P becomes the most important P pool for soil microbes. While soil microbes can use organic P via several mechanisms, the most significant is the production of extracellular phosphatase enzymes. Therefore, producing these phosphatase enzymes is likely to be a substantial microbial resource investment that can maintain soil microbial growth.

The objective of this study was to assess the effects of N and P addition on microbial biomass and activity and thus, to determine whether soil microbes are N and P limited in larch plantation soil by using adjacent secondary forest soil for a comparison. We selected soil microbial properties relevant to soil C, N and P cycling as indicators, including soil microbial respiration and N mineralization, microbial biomass C, N and P, and the potential activities of exoglucanase, β-glucosidase, N-acetyl-β-glucosaminidase and acid phosphatase. Because the intermediate products of enzymatic reactions can provide additional information on soil microbial processes (Waldrop and Zak, 2006; Yao et al., 2009), we also measured soluble organic C and N. Four paired sites of larch plantation and adjacent secondary forest stands were randomly chosen in Northeast China. Thus, a better understanding of which soil nutrients may exert a constraint on microbial activity in larch plantation soil and comparing this effect with microbial activity in secondary forest soil can improve the theoretical basis of forest management oriented toward the recovery of soil fertility in temperate forest ecosystems.

2. Materials and methods

2.1. Study site and soil sampling

The experimental site was located in the Qingyuan Forest CERN (41°51′ N, 124°54′ E) of the Chinese Academy of Sciences in Liaoning Province, China. The soil is a typical brown forest soil classified as Udalfs according to the second edition of USDA soil taxonomy. The region has a continental monsoon climate with a humid and rainy summer and a cold and dry winter, a mean annual temperature of 4.7 °C, a minimum monthly temperature of −12.1 °C in January, and a maximum monthly temperature of 21.0 °C in July. Annual precipitation fluctuates between 700 and 850 mm, with >80% falling during June–August. The frost-free period lasts for 130 days on average, with an early frost in October and a late frost in April (Zhu et al., 2010).

The study site had been covered primarily by mixed broadleaf Korean pine forests before the 1930s and thereafter had been subjected to unregulated timber removal for decades. Massive controlled burns were used in the early 1950s for clearing out the original forest. Then, the study site gradually had turned into secondary forest that comprised a tree layer including *Fraxinus rhynchophylla*, *Juglans mandshurica*, *Phellodendron amurense*, *Quercus mongolica* and *Acer mono*; a shrub layer including *Acer mandshricum*, *Acer triflorun*, *Acer tegmentosum* and *Syringa amurensis*; and an herbaceous layer including *Cardamine leucantha*, *Allium monanthum*, *Arisaema amurense* and *Polygonatum involucratum*. In the 1960s, the natural secondary forest was partially cleared, and patches were converted to larch (*Larix gmelinii*) plantations that also contained a shrub layer including *A. tegmentosum*, *A. pseudo-sieboldianum*, *Schisandra chinensis*, *Syringa wolfi*, and *Acanthopanax senticosus* and an herbaceous layer including *C. leucantha*, *Rubia sylvatica*, and *Spuriopimpinella brachycarpa*. Because larch plantations were interspersed with secondary forests, our study site represented a mosaic plantation-secondary forest landscape. Given differences in tree species between secondary forests and larch plantations, the forest floor is likely different in nutrient content and thus affects microbial biomass and activity in soil. The experimental design in this study was limited by understanding the impacts of available N and P additions on microbial properties in the forest floor of two forest types.

Four independent and paired stands of secondary forests and adjacent larch plantations with different landforms (i.e., aspects and altitudes) were randomly chosen from separate watersheds in Qingyuan Forest CERN, with slopes <15° and altitudes ranging from 589 to 836 m above sea level. A 30 m × 30 m plot was established in each secondary forest and larch plantation stand for a total of eight