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Effects of tree species transition on soil microbial biomass and community structure in subtropical China

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Large-scale land use changes have remarkably influenced the global carbon (C) and nitrogen (N) cycling. Soil microorganisms are known to be the key drivers of these processes and act as susceptive indicators of changes in ecosystem functioning due to land use changes. In forest ecosystems, differences in the stability and turnover of soil C and N pools are mainly associated with the variation in the above- and belowground litter/root inputs to the soil from the tree species. However, the impact of soil C and N pool differences caused by tree species on soil microbial community structure has not been fully investigated in subtropical China. This study aimed to assess the effects of tree species conversion from a coniferous to broad-leaved plantation on the soil microbial biomass and community structure associated with C and N transformations within the plant-soil system. The microbial biomass and composition (reflected by 28 phospholipid fatty acid profiles), soil C and N pools in the top soils, and C and N contents of certain litter and fine root profiles were measured 19 years after the reforestation of Chinese fir (Cunninghamia lanceolata) woodland with the same species or a native broadleaf species Mytilaria laosensis. The results suggested that soil microbial biomass was significantly higher in the M. laosensis than in the C. lanceolata plantations, and non-metric multidimensional scaling ordination plots showed distinct patterns of soil microbial community structure between these two species. Soil microbial biomass showed negative correlations with litter N or mineral N content, i.e., ammonium N (NH $_4^+$ -N) and nitrate N (NO $_3^-$ -N), but was positively correlated with soil C content and litter C: N ratio. Further, there were negative correlations between soil microbial biomass C and mineral N pools. These results indicated that tree species transition from M. laosensis to C. lanceolata might have improved the soil labile C and N pools and their availability, leading to an increase in the soil microbial biomass. Redundancy analysis conducted to elucidate the relationships between the microbial community and C or N parameters also showed that the soil C: N ratio, soil total N, and NH $_4^+$ -N might be the major factors influencing the soil microbial community. However, soil microbial diversity and richness were not significantly altered by the tree species transition. These results suggested that the potential process rates mediated by litter-derived C and N availabilities might not always be accompanied by a remarkable response from community diversity, but might affect microbial biomass. In conclusion, long-term tree species transition from coniferous to broad-leaved plantations significantly improved soil C and N pools and their availabilities, thereby increasing the soil microbial biomass and changing the composition of in situ soil microbial community. Previous events (e.g., land use history) might have considerable long-lasting impacts on soil microbial diversity and richness than the contemporary environment variables caused by the tree species transition 19 years after reforestation.

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1. Introduction

Land use change is one of the vital factors that could affect the global carbon (C) and nitrogen (N) cycling, as well as mitigate global climate change [\[1\]](#page--1-0). As one of the most important kind of land use changes, forest conversion could greatly affect the C and N balances in soils due to

Corresponding author. E-mail address: zhiqunhuang@hotmail.com (Z. Huang). changes in tree species and forest types [\[2](#page--1-0)–4]. Since the 1950s, under the extensive national forestry policy, large areas of Chinese fir (Cunninghamia lanceolata, abbreviated as C. lanceolata) were planted to increase the economic benefits. However, the ever-increasing coniferous plantations failed to maintain the ecological benefits, resulting in decreased species diversity and decreased and soil fertility, as well as degraded ecological functions and other adverse consequences [5–[7\].](#page--1-0) The efficiency of forest ecological function is closely related to the soil fertility, while the stability of soil organic matter and nutrient

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effectiveness strongly affects soil fertility. Previous studies in the present study area revealed significantly increased soil C and N stocks after tree species transition from Chinese fir to native broad-leaved species [\[8\].](#page--1-0) In this context, investigations on the impacting factors that control over changes in soil organic matter, C and N pools following tree species conversions still remain to be an urgent issue for ecologists.

In forest ecosystems, tree species control over the stability, turnover and availability of soil C and N pools through altering litter and root debris inputs [\[9\].](#page--1-0) Some studies have found significant differences in soil mineral N under different tree species and this, to some extent, may influence the physiological and metabolic processes of soil microorganisms [10–[12\].](#page--1-0) As an important medium in regulating the forest structure, service and function, soil microorganism drives soil C and N cycling through directly participating in the decomposition of soil organic matter, heterotrophic respiration and N-immobilization process [\[13\]](#page--1-0). Thus the activities of microbes play vital roles in material cycling and ecological function, and microbial mediated biogeochemical cycling in plant-soil ecosystems has been a salient topic in studies of forest ecosystems [\[14,15\].](#page--1-0) Soil microbial biomass and diversity are sensitive indicators of soil vitality, and their metabolic processes are influenced by interactive effects of plant, environment and substrate properties [\[16,17\].](#page--1-0) As a result, they responded differently to different environments and metabolic substrates. For example, soil microbial biomass and community composition are closely related to compounds and availability of soil organic matter [\[18,19\]](#page--1-0). However, a recent study suggested that the impacts of silvicultural practices and climate change on soil organic matter is relatively slow, among which the labile C and N pools are more sensitive to disturbances than the total soil organic C and N [\[20\]](#page--1-0). This mainly because the inputs of fresh organic matter could stimulate the rapid turnover of labile C and N pools rather than the recalcitrant parts [\[21\]](#page--1-0). On the other hand, changes in soil organic C and N can influence soil microbial community structure via various pathways. In this regard, the response of soil microbes at community level to large-scale reforestation and its impacting factors are still not clear, especially after the 1990s, when many reforestation experiments were established using different tree species. Investigations on such changes of soil microbial community caused by these experiments are also in urgent needed.

After reforestation of Chinese fir woodland with itself or a native broadleaf species, Mytilaria laosensis (abbreviated as M. laosensis) for 19 years, we revisited this two plantations and determined the relationships between soil microbial community and C and N properties in soil, litter and fine root profiles. In the paradigm of plant-soil interaction, we explored the changes and impacting factors caused by the tree species conversion from needles to broad leaf, so as to provide some implications for future forest management and silvicultural practice.

2. Materials and methods

This study was conducted at Xiayang forest farm (26°48′N, 117°58′E, 229–246 m mean elevation), Southeast Wuyi Mountain. The climate is humid subtropical monsoon climate with a mean annual precipitation of 1644 mm and an average temperature of 20.0 °C. The relative humidity is 75.2% and the mean annual evapotranspiration is 1370 mm. The soils are derived from the underlying gneiss and schist bedrock and are classified as sandy clay loam Ferric Acrisol soils (FAO/UNESCO classification). In April 1993, after harvesting in a second rotation plantation of C. lanceolata, two species were then planted (2500 stems ha⁻¹) in the eight well established plots (20×30 m) as pure forest plantations with four plots of C. lanceolata seedlings and four plots of M. laosensis seedlings. The plots were separated by >10 buffer tree rows.

In July 2011, recent sites characteristics and several soil chemistry properties were investigated 19 years after reforestation, as compiled in Table 1. In May 2012, intact soil samples were taken from the top soil (0–5 cm depth) at 10 random locations with Diagonal-line sampling method for each plot. Briefly, PVC tubes were carefully driven into the

Table 1

The comparison of sites characteristics and soil labile C and N variables between M. laosensis and C. lanceolata plantations.

Different letters in the same row indicate significant difference at $P < 0.05$ level between the two species, and the data stand for mean and standard deviation.

soil to avoid major disturbances from human and stem flows. The soils were sieved through a 2 mm sieve, and then thoroughly mixed prior to subsequent analysis. For each plot, ten soil cores were pooled and shipped with ice bags. Upon arrival at the laboratory, some of the samples were stored in a 4 °C fridge for soil biochemical analysis and the others were freeze-dried for microbial community analysis. Phospholipid fatty acid (PLFA) analysis was used to characterize microbial community composition following the general methods of White et al. [\[22\]](#page--1-0) and modification from Bossio and Scow [\[23\].](#page--1-0) The relative abundance of each PLFA (n mol g^{-1} soil) were calculated to represent the community composition. PLFA biomarkers were grouped as Gram-positive bacteria (i15:0, a15:0, i16:1 G, i16:0, i17:0, a17:0, 15:0 3OH and 16:1 2OH), Gram-negative bacteria (cy17:0, 18:1 ω7c, 18:1 ω5c and cy19:0), actinomycetes (10Me 16:0, 10Me 17:0, 10Me 18:0 and 11Me 18:1 ω7c), fungi (18: 1 ω9c and 18:2 ω6c) and arbuscular mycorrhizal fungi (VAM fungi, 16:1ω5c) [\[23\].](#page--1-0) Other lipids such as 14:0, 15:0, 16:0, 17:0, 18:0, 19:0, 16:0 N, 20:1 ω9c and 20:4 ω6,9c were included for community analysis.

Microbial biomass carbon (MBC), microbial biomass nitrogen (MBN), dissolved organic carbon (DOC), dissolved organic nitrogen (DON), ammonium (NH $_4^+$ -N) and nitrate (NO₃-N) nitrogen in soil were determined. In brief, MBC and MBN was measured by the chloroform fumigation extraction method using an EC factor of 0.38 and 0.45 [\[24\]](#page--1-0), respectively. Field moist samples (10 g oven-dry equivalent) by shaking with 40 ml 0.5 M L⁻¹ K₂SO₄ for 30 min, followed by centrifuging at 2000 rpm for 20 min. Similarly, DOC and DON were extracted with 10 g soil and 40 ml deionized water, and mineral N (NH $_4^+$ -N and NO₃ $-$ N) were extracted with 10 g soil and 40 ml 2 M L^{-1} KCl, following the same shaking and centrifuging procedures above. The supernatant was filtered through Whatman 42# filter paper. The organic C and N concentrations were determined using a SHIMADZU TOC-VCPH/CPN analyzer (filtered through a 0.45 μm filter membrane prior to analysis) and mineral N was determined using an automated ion analyzer (Skalar $San⁺⁺$, Netherlands).

3. Statistical analysis

Pearson correlation coefficients were used to describe the correlations between microbial indices and plant and soil C and N properties (SPSS 19.0 for Windows). For spatial ordinations, Non-metric Multidimensional Scaling (NMDS) was performed in R 3.1.1 based on a Bray-Curtis dissimilarity matrix using the "vegan" package. The relative abundances of each lipid biomarker were converted into a dissimilarity matrix with the Bray distance before ordination. Canoco 4.5 were also used to conduct Detrended Correspondence Analysis (DCA)

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