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Soil microbial characteristics and the influencing factors in subtropical forests

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ABSTRACT

Soil microorganisms play an important role in the soil formation, nutrient cycling, and regulating plant productivity. Bacteria, fungi and actinomycetes are main microbial groups being responsible for the soil ecosystems. However, responses of soil microbial communities to litter characteristics and soil properties at the regional scale are poorly understood. Litter characteristics, soil properties and soil microbial communities were measured in coniferous forests and broadleaved forests at three natural forests (Dinghu Mountain National Field Forest Ecosystem Research Station (DHS), Ailao Mountain National Field Forest Ecosystem Research Station (ALS), Daming Mountain Nature Reserve Station (DMS)) and a plantation (Heshan National Field Forest Ecosystem Research Station (HSZ)) ecosystems in order to explore the effects of litter characteristics on soil microbial community structure. In our study, the soil microbial communities were analyzed by dilution-plate method. Furthermore, we analyzed the determined factors of soil microbial community structure in the forest ecosystems. The results showed that season, forest type and sampling site influenced the soil microorganisms and community structure. There were the obvious seasonal dynamics of soil microorganisms in subtropical region, China. The soil microbial counts and the ratio of fungi and bacteria in wet season were higher than those in dry season. The counts of bacteria and total microorganisms in coniferous forests were less than those in broadleaved forests. Moreover, the ratios of fungi and bacteria were greater in coniferous forests than that in broadleaved forests. Similarly, the counts of bacteria and total microorganisms in plantations were less than those in natural forests. Besides, the fungi in the two forests showed the different trends in dry season and wet season. Soil fungi in natural forests were higher than that in plantation in dry season, but it showed the opposite pattern in the wet season. The ratio of fungi and bacteria was significantly higher in plantation than that in natural forests. Linear regression results showed that variations of fungi, actinomycetes and total microorganisms were primarily explained by the litter quality in coniferous forests, but the variations of fungi and actinomycetes were explained by the litter storages in broadleaved forests. Redundancy analysis (RDA) results showed that soil and litter properties explained 79.7% of total variation in microbial community structure. Furthermore, the variations of microbial community structure were explained 19.1%, 16.7%, 16.6% and 11.6% by soil total nitrogen (STN), soil organic carbon (SOC), soil water content (SWC) and the ratio of soil organic carbon and the nitrogen (soil C:N), respectively. Overall, our findings will facilitate a better understanding of soil microbial characteristics in subtropical forests.

1. Introduction

Soil microorganisms were the important components of the soil ecosystems. Bacteria, fungi and actinomycetes were the three main groups of soil microorganisms which played significant roles in the soil ecological processes, such as the decomposition of the organic matter, mineralization of the nutrient and transformation [1,2]. In addition, soil microorganisms were the stock (fixed process) and source (mineralization process) of the active nutrient [3]. The numbers of soil microorganisms were closely related to the ecological environment [4]. Zhang reported that the diversification in numbers of bacteria, fungi and actinomycetes was one important indicator of the microbial activity in forest soils [5]. Soil bacteria (B) with the largest numbers and widely distributing in soil microorganisms decomposed the cellulose-based material in neutral and slightly alkaline environment. Soil fungi (F) were the active participants in the decomposition of soil carbohydrates, fiber, pectin and lignin, which were particularly important in many acidic forest soils [6]. The ratio of fungi to bacteria was of great importance to evaluate the soil fertility and soil health [7]. Soil microbial community with the higher ratio of fungi to bacteria (F: B) drives the further mineralization of soil endogenous carbon substrate, and soil ecosystems with higher F:B were more persistent and stable [8].

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Currently, using any methods are not exhaustive understanding of the soil microorganisms due to the extremely complex microbial composition. Dilution-plate counting as the traditional method did not completely respond to the quantities and structure of soil microorganisms, but this method was easy to operate. We can determine the numbers and morphology of soil microbial colonies by separation and purification. Therefore, the traditional dilution-plate method played an important role in the research of microorganisms [9].

Soil microbial characteristics and community structure in forest soils had been widely studied. The previous studies showed that the numbers of microorganisms were higher in the natural forests than those in the plantations [10]. Soil microbial community structure was very sensitive to the change of environmental conditions [11]. The management measures (such as afforestation and tending) changed the soil structure and environment, which would affect the soil microbial community structure [12]. In addition, soil pH and the ratio of carbon to nitrogen also influenced the soil microbial community structure [13]. It was reported that the annual precipitation and soil pH were the most important factors influencing the zonal distribution of soil microorganisms [14]. It was noteworthy that many previous studies were conducted at local scale (site) to investigate the soil microbial community in forest ecosystems, which was a relatively small scale for study. However, the high variability at a local scale made it difficult to extrapolate the pattern of soil microbial community to the regional scale. Therefore, soil microbial community and the related influencing factors in forest ecosystems at larger scale are still uncertain and need to be further clarified. In the present study, two main types of subtropical vegetation, monsoon evergreen broadleaved forests and coniferous forests were chosen to study the soil microbial characteristics and the influencing factors. Our objectives are to 1) reveal the soil microbial characteristics in the two types of subtropical forests, China; and 2) elucidate the factors affecting the soil microbial characteristic in subtropical forests. Our study would provide the scientific basis for the research of forest ecosystems at regional scale.

2. Materials and methods

2.1. Site description

The experimental sites were located at Ailao Mountain Site (ALS), Daming Mountain Site (DMS), Dinghu Mountain Site (DHS) and Heshan Station (HSZ), which were all in the subtropical region of China. The ALS, DMS, DHS and HSZ were situated near the Tropic of Cancer across the longitude of 100–115 °E in Yunnan, Guangxi, and Guangdong provinces, respectively. These stations were all at a typical subtropical zone with the alternating climate, and the obvious wet season and dry season.

ALS was located at the central part of Yunnan Province (24°32′ N, 102°01′ E), with 2400–2600 m altitude and the subtropical monsoon climate. The average annual temperature at ALS was 11.3 °C, the hottest temperature was 16.4 °C and the coldest temperature was 5.4 °C. The average annual sunshine hours, the annual rainfall, and the annual evaporation capacity were 2131.6 h, 1931 mm and 1485.9 mm, respectively. The soil was a typical yellow brown with pH 4.4–4.9. Zonal vegetation was dominated by middle-mountain moist evergreen broad-leaved forests, which was the type of mountain vertical vegetation in subtropical evergreen broad-leaved forests [15]. We chose the broad-leaved forests in the central reserve area and the coniferous forests with 700–1800 m located at the west slope. The dominant species in broad-leaved forests were *Schima noronhae*, *Manglietia insignis* and *Machilus viridis* and the dominant species in coniferous forests was *Pinus kesiya* [16].

DMS was located at Nanning City, northeast of Guangxi Province (23°27′ N, 108°22′ E). This area with an average elevation of 1200 m, at the Tropic of Cancer center, was subtropical humid mountainous monsoon climate, and had the characteristics of marine and mountain climate. The average annual temperature was 15.1 °C, the hottest temperature was 21.9 °C and the coldest temperature was 5.8 °C. The average annual sunshine was 1295.4 h and the average annual rainfall was 2630 mm [17]. Soil texture was mainly clay loam and loamy clay with the pH value in the 3.9–4.7 [18]. Zonal vegetation in this area is middle mountain monsoon evergreen broad-leaved forests. We chose the broadleaved forests dominated by *Quercus griffithii* species and coniferous forests dominated by the *Pinus massoniana* species.

DHS was located at the center of Guangdong Province (23°10′ N, 112°33′ E), with the altitude 10–1000 m. This area was a typical south subtropical humid monsoon climate. The average annual temperature was 21.4 °C, the hottest temperature was 28.0 °C and the coldest temperature was 12.6 °C. The annual sunshine was 1433 h, the average annual rainfall was 2630 mm and the average annual evaporation was 1115 mm. The soil was latosolic red and serious natural acidification with pH 4.1–4.9 [19–21]. Zonal vegetation in this area was the monsoon evergreen broad-leaved forests. We chose the broadleaved forests dominated by the *Castanopsis chinensis*, *Schima superba*, and *Cryptocarya chinensis* species and the coniferous forests with *P. massoniana*.

HSZ was located at the central area of Guangdong Province (22°34′ N, 112°54′ E), in low hilly ground district, ridge flat round and gentle slope with the average elevation 80 m. This area was subtropics monsoon climate. The average annual temperature was 21.7 °C, the hottest temperature was 29.2 °C and the coldest temperature was 12.6 °C. The annual sunshine was 1798 h, the average annual rainfall was 1800 mm and the average annual evaporation was 1638.8 mm. The regional soil was latosolic red soil and soil texture was mainly clay loam and loamy clay with the pH value in the 4.2–4.8 [22]. Different types of plantation were planted in 1981 to study the restoration mechanisms and methods of degraded vegetation in the hilly slopes. We chose the *S. superba* and *P. massoniana* plantations, which had larger planting area and wider distribution in subtropical region.

2.2. Experimental design and sampling

Three pairs of broad-leaved and coniferous forests were chosen from each of the four sites, totaling to 12 paired forest comparisons. Soil samples were taken from plots (20 m $*$ 20 m) in each forest. There were 24 plots in total and each plot consisted of five subplots (1 m $*$ 1 m) for soil sampling. The surface litter of each subplot was removed before taking soil samples. In the dry and wet seasons of 2011, eight soil cores (5 cm in diameter) were randomly taken from each subplot at a depth of 0–15 cm to form a composite sample. The total number of samples for each of the nine forest pairs was 240: namely, 2 forests $*$ 3 plots $*$ 5 subplots $*$ 4 sites $*$ 2 seasons. The fresh soil samples were divided into two portions. One portion was used to analyze the soil microbial colonies and community by the method of dilution plate [23,24]. The other portion of soil samples and litter was transported to the laboratory for further analysis.

Soil moisture content (SMC) was measured by oven-drying for 48 h at 105 °C, and soil pH was determined in 1:2.5 (w/v) soil solutions. The soil organic carbon (SOC) and litter organic C (LOC) were determined by dichromate oxidation and soil total nitrogen (STN) and litter total nitrogen (LTN) were measured based on Kjeldahl's method [25]. The results of soil and litter properties were shown in Tables 1 and 2 (the data in Tables 1 and 2 were just the background information, and some results were included in other unpublished manuscript).

2.3. Data statistics

Excel 2010 (Microsoft Office 2010), SPSS16.0 (SPSS, Inc., Chicago, IL) and Canoco 4.5(Ithaca, NY, USA) were used in the data processing and statistical analysis. Results in our study were shown as the mean value, and the statistical significance was accepted at a $P < 0.05$ level. Independent sample T-tests were used to compare the differences in soil microbial numbers and community between the broadleaved and coniferous forests, and between the natural forests and plantation. In addition, One-way ANOVA and LSD were used to compare the differences among the four sites, and Three-way ANOVA was used to