# Soil Microbial Functional Diversity in Native Mixed-Korean Pine Forests

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Abstract:Experiments were carried out on a serial of original korean pine forest compared to adjacent artificial forest. The experimental designed five plots (HD, HZ, HY, LY and BH) of 800 m<sup>2</sup>.Nevertheless, changes caused by the artificial application to the soils, leading to major adverse effects on the microbial community as compared to the untreated soils of original korean pine forest. Using the model, the integrated eco-environmental stability index (ESI) of study area in five forest types were computed. According to the numerical results, the stability of the three original korean pine mixed forest types treatments (HD. ΗZ and HY) demonstrated significantly higher (P < 0.05) compared with both the artificial pure forest types (LY and BH).

Keywords: Native Mixed-Korean Pine Forests; Microbial Diversity; Biolog;

# 1. Introduction

Aboveground and belowground components of terrestrial ecosystems are opment process. A change in the composition of a plant community leads to a change in litter quality, which alters the local nutrient cycling process and soil conditions; the changed soil conditions may in turn drive a further change in plant community composition. Those two processes taken together form a plant-soil feedback (PSF), a major driver of plant community dynamics and nutrient cycling<sup>[1,2]</sup>. plant–soil feedback (PSF) determines the structure of a plant community and nutrient cycling in terrestrial implicitly dependent on each other<sup>[3]</sup>. A plant community and local soil conditions are understood as an outcome of the plant-soil codevelecosystems. Microbial communities exhibit distinct compositions <sup>[4-6]</sup>and/or functions<sup>[7]</sup>, depending

on the litter quality and plant species with which they are associated with. It has been hypothesized that flexibility in the community composition and function of microbial decomposers either reinforces<sup>[8]</sup> or weakens<sup>[9]</sup> plant control over nutrient mineralization. If the dominant species favors nutrient-rich sites and produces a quickly decomposing litter, then the accelerated nutrient cycling maintains a competitive advantage, preventing competitor invasion and enhancing species dominance<sup>[10-12]</sup>

# 2. Materials and Methods

## 2.1 Study Area

Liangshui Nature Reserve (47°10'50"N, 128°53'20"E), a preserve which is located in Yichun city, Heilongjiang province of China. It had the efficacy of protecting korean pine broad-leaved mixed forest ecological system. The area is 6394 hm<sup>2</sup>, its temperate type belongs to the continental climate. In this area, the latitude is higher, average annual temperature is only -0.3°C while the annual average minimum and maximum temperatures are -6.6°C and 7.5°C respectively. It belongs to the northern temperate needle mongolica forest area. The zonal vegetation is mixed forest with needle and broad leaves , which is mainly Korean pine (Abbreviation broad-leaved Korean pine forest).

Eight sampling locations surrounding each original korean pine mixed forest types (type: HY: korean spruce & korean pine forest; HZ: oak & korean pine forest; HD: linden-korean pine forest) in Liangshui Nature Reserve, which were chosen based on previous studies conducted in this site. Two adjacent artificial forest types (type: BH: birch pure forest; LY: larch pure forest) was chosen to represent background.

#### 2.2 Vegetation Sampling

The above-ground plant species inventory was carried out between July and August of 2010. In order to avoid a possible edge effect, eight square sampling plots  $(25 \times 25 \text{ m}^2)$  were delimited according to the method of the species/area curve, demarcated approximately in the centre of each stand. The vegetation divided into three height classes, 0-0.5 m (herbaceous or small shrubs), 0.5-2 m (shrubs) and >2 m (tall shrubs or trees). In each sampling plot, trees ( $10 \times 10 \text{ m}^2$ ), shrubs ( $5 \times 5 \text{ m}^2$ ) and herbaceous  $(2 \times 2m^2)$  was destructively measured in plots placed in each facies whose datum within the  $10 \times 10$  m<sup>2</sup> plot was used for comparing with below-ground communities. Detailed measurements were made of stand and site characteristics on these plots: Each plant recorded was characterized by its species name, canopy dimensions, number and trunk circumference; habitat factors: longitude and latitude, altitude, topography, slope, soil type, etc

#### 2.3 Soil Sampling

Soil samples were taken at five different sites in the differen vegetation type. Soil samples of surface soil, 0-10 cm-layer soil and 10-20 cm-layer soil were collected and bagged into sterile plastic bag in July of 2010. All samples were stored in 4°C to determine microbial community diversity.

#### 2.4 Soil Microbial Analysis And Diversity Calculations

Biolog ECO microtiter plates (Biolog, Hayward, CA, USA) were used to assess microbial functional diversity as described previously. The slurry was diluted to  $10^{-3}$ , and used to inoculate Biolog Eco plates with 150 uL per well. Plates were incubated at 25°C in darkness. Carbon substrate utilization was measured every 24 h for 240 h as quantified by absorbance at a wavelength of 590 nm using an automatic plate reader (Biolog Microstation Elx808BLG, BIO-TEK Instruments Inc., USA). Overall colour development expressed as average well colour development (AWCD). was calculated as the mean of the blanked absorbance values for all the 31 wells per reading time. AWCD= $\sum (C-R)/N$  where C is colour production with each well, R is the absorbance value of the plate's blank well, and

N is the number of substrates (ECO plates, N=31). Three replicates per treatment and sampling time were performed. Kinetics of AWCD were used to determine the speed and the level of development of the bacterial communities using the 31 provided substrates. Moreover, the absorbance value of each well at 72 h of incubation was then divided by the AWCD in order to normalize the values and to minimize the influence of inoculum density between plates. These data from 72 h were used to calculate the functional diversity using Shannon's functional diversity index: Shannon's index (H) =  $-\Sigma$  [Pi\*Ln Pi], where Pi is the ratio of the blanked absorbance value of each well to the sum of absorbance values of all wells. Simpson's index (D) =  $\sum_{i=1}^{N} [ni(ni-1)/N(N-1)]$ 

McIntosh's index (U) =  $\sqrt{\sum n^2}$  where ni is the ratio of the blanked absorbance value of each well (i.e., C-R), where N is the sum of absorbance values of all wells (i.e.,  $\Sigma(C-R)$ ).

#### 3. Results

The treatment groups (Fig. 7)showed similar trends during the three soil layers (suface, 0-10cm, and 10-20cm). The simpson's index of the suface soil for the treatments was maximum. The treatments of 0-10cm soil layers appeared to show a decrease in volume of simpson's diversity. The simpson's index of the 10-20cm soil is minimum.

Both the simpson's diversity index for the soil of suface, 0-10cm, and 10-20cm layers was significantly higher (P<0.05) in the original Korean pine mixed forest types (HD, HZ, and HY) than the artificial pure forest types (LY, BH) at each sampling date. The simpson's diversity for the soil of the 10-20cm layer was lower (P<0.05) in the artificial pure forest types (LY, BH) than the original oak and korean pine mixed forest types (HZ) at each sampling date, but higher than the others original Korean pine mixed forest types (HD, HY), although discrepancy were not significant (Fig. 1).

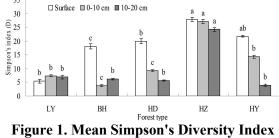


figure 1. Mean Simpson's Diversity Index for the Soil of Suface

0-10cm, and 10-20cm layers among the different forest types. ( $\pm$ S.E.) Means with different letters are significantly different (by Fisher PLSD test, at P < 0:05).

Both the mean shannon's diversity index and meintosh's evenness index of the three soil layers were similar among the treatments (Fig. 2; Fig. 3). The shannon's index and meintosh's index of the suface soil layer for the treatments was maximum. The treatments of 0-10cm and 10-20cm soil layers appeared to show a approach in volume of shannon's and meintosh's indexes.

Species diversity and evenness of the three soil layers were similar (shannon's index and mcintosh's index) among the treatments. Both the artificial pure forest types (LY and BH) demonstrated significantly lower (P<0.05) level compared with the original oak and korean pine mixed forest types (HZ).

Mean species diversity and evenness of the surface soil layer was higher (shannon's index and mcintosh's index) among the HD and HY treatments compared with both the artificial pure forest types( LY and BH ). The treatments of 0-10cm and 10-20cm soil layers appeared to show a approach in volume of shannon's and Mcintosh's indexes.

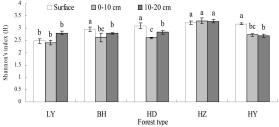
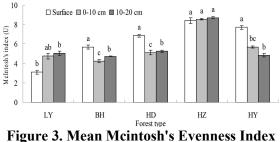


Figure 2. Mean Shannon's Diversity Index for the Soil of Suface



for the Soil of Suface

## 4. Conclusions

This study focused on an idea about Vegetation types influence the soil microbial community functional diversity through difference of plant diversity and catabolic diversity. At the meantime, the SPCA method is used to determine the variables and their weights. A numerical evaluation model is developed to analyze eco-environmental problem in mountainous region. From the study, we draw the following conclusions:

Eco-environmental stability in study area apparently showed that the original Korean pine mixed forest types (HD, HZ, and HY) were similar,and significantly higher than the artificial pure forest types (LY, BH).

The study of the effects of vegetation type on soil microorganisms has implications for ecosystem restoration in the north of China. The results indicated it is urgent that, besides the improvement and reinforcement of compensation mechanism construction, the work of eco-environmental recovering and rebuilding should be carried out according to enhancing eco-environmental stability of the regionalization.

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