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Original article

Influence of soil faunal properties and understory fine root on soil organic carbon in a "mesh bag" approach

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ABSTRACT

The obstruction of fine mesh bag has been widely applied to study the functions of plant fine roots and soil fauna in ecosystems. However, plant fine roots and soil fauna can both be either excluded from the soil within fine mesh bags or admitted into the soil without mesh bags or within coarse mesh bags, because of their similar ranges of body width. As a result, both their effects could be influenced by fine mesh bags. In this study, we investigated the effects of soil fauna and understory fine roots on soil organic carbon, with a special type of fine mesh bags designed for obstructing the growth of understory fine roots into soil with free access of soil fauna. Results showed that both the biomass of understory fine roots and soil faunal diversity along with soil total carbon and microbial biomass carbon were significantly reduced by the fine mesh bags ($P < 0.01$). Boosting regression tree models showed that the understory fine roots contributed more in the variations of soil organic carbon than soil faunal properties. Faunal biodiversity represented by Pielou's evenness index was more important than taxa richness and individual abundances of taxa. Results suggest that understory fine roots may be more influential in soil organic carbon than faunal factors, and both their functions could be influenced by fine mesh bags in a mesh bag study. Evenness of individual abundances between taxa could be more important than taxa richness for the influence of soil fauna community on soil organic carbon.

1. Introduction

Soil fauna [1–5] and understory vegetation [6–8] are important functional components in forest, and their functions have been paid increasing attention. The importance of understory vegetation to soil respiration has been increasingly emphasized [9–12], and widely investigated by understory removal. However, the influence of understory vegetation on soil organic carbon [13,14], especially the effects of understory fine roots have not been well studied [15]. For studying the functions of understory fine roots, using micro-mesh bags (mesh < 45 μm) seems to be a suitable approach, because this method has been widely used to exclude tree fine roots from soil [16,17]. Similarly, an approach for studying the functions of soil fauna in soil processes is also using various fine mesh bags that can exclude different functional groups of soil fauna from the soil [18–22]. This method can be called field micro/meso-cosom experiment, in which soil medium is prepared through fetching upper soil in the field and defaunating it by deep-freezing [22,23] or heating [24], and so on. The defaunated soil is then wrapped by different meshes of nylon nets and replaced in the field for biotic recolonization.

Both the body widths of soil fauna and understory fine roots can range from scales of microns to centimetres [25]. The mesh bag approach for soil fauna involves the incubation of defaunated soil cores back in the field where the surrounding understory fine roots can also penetrate through coarse mesh bags into the soil cores. Consequently, soil fauna and understory fine roots may either be excluded from the soil cores within fine mesh bags or admitted into the soil cores without mesh bags or within coarse mesh bags.

When evaluating the influence of soil fauna or understory fine roots on soil organic carbon using the mesh bag approach, fine mesh bags possibly impose unexpected influence on the other factor. The effects of soil fauna and understory fine roots might become mixed and estimated inaccurately. Thus, the effects of soil fauna and understory fine roots on soil organic carbon in a mesh

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bag approach may need to be simultaneously investigated and compared. Two issues should be clarified during this investigation, namely, the relationships of soil fauna with understory fine roots, and the groups of soil faunal properties that should be stressed. Soil faunal properties such as individual abundances of taxa and biodiversity might be simultaneously influenced by mesh size. The functions of individual taxa, functional groups and biodiversity of soil fauna in soil processes have been highlighted [26–29]. Thus, we investigated the effects of soil faunal properties involving individual abundances of fauna taxa, faunal biodiversity and understory fine roots on soil organic carbon.

We designed a special type of fine mesh bags to exclude understory fine roots from soil with free access to soil fauna. We assumed that this type of fine mesh bags could exclude the understory fine roots out of the soil, whilst imposing little disturbance on soil faunal properties. The influence of fine mesh bags would be arbitrarily attributed to understory fine roots, provided that the fine mesh bags can significantly reduce the biomass of understory fine roots, and does not influence the soil faunal properties. Otherwise, the effects of soil fauna on soil organic carbon may not be considered undisturbed if the soil faunal properties are also significantly influenced by the fine mesh bags. In any case, we investigated the relative influence of soil faunal properties and the biomass of understory fine roots on soil organic carbon using boosting regression trees (BRT). BRT is useful for treating datasets that contain many explanatory variables, and automatically considers the interactions of predictor variables.

2. Materials and methods

2.1. Study site and experimental design

The study was conducted in Huitong National Research Station of Forest Ecosystem (26° 50′ N, 109° 36′ E; elevation 300–500 m). The climactic and topographical information of this area can be referred to Wang et al. [30]. The main forest types in the region are secondary evergreen broadleaf forests, *Cunninghamia lanceolata* plantations and *Pinus massoniana* plantations. The dominant tree species in the secondary evergreen broadleaf forests are *Castanopsis fargesii*, *Machilus pauhoi*, *Alniphyllum fortune*, and *Cyclobalanopsis glauca*, and the common understory species mainly include *Maesa japonica*, *Kadsura coccinea*, *Maesa perlarius* and *Millettia dielsiana*. The common understory species in *C. lanceolata* plantations mainly include *Mussaenda esquirolii*, *M. japonica*, *Urena procumbens*, *Actinidia fulvicoma*, *Maesa perlarius* and *Yua thomsoni*. Two secondary evergreen broadleaf forests (labelled as A and C) naturally regenerated from clear-cutting forestlands of *C. lanceolata* in 1996 and two *C. lanceolata* plantations (labelled as B and D) established in 1983 were selected for this study. The soils in sites A and B were both yellowish red soils (ultisols), whilst the soils in sites C and D were both yellow soils (inceptisols).

The experimental units were defaunated soil cores originating from the four study sites. Three experimental factors, namely, the type of mesh bags (fine- or coarse-mesh bags), litter addition (with or without leaf litter) and soil source (transferred or untransferred) were arranged in each site. The two types of mesh bags (fine- and coarse-mesh bags, both 13 cm in diameter and 35 cm high) were used to wrap the defaunated soils in the field (Fig. 1). The coarse mesh bags were closed at the top but open at the bottom, whereas the fine mesh bags were closed both at the top and bottom. The closed tops of the two types of mesh bags prohibited fallen leaves to lie on the soil. The coarse mesh bags were of 4 mm mesh throughout. The meshes of fine mesh bags were on the whole 0.038 mm, except that two rows of and a circle of 4×4 mm^2 holes were distributed evenly at the two lateral sides and the circumferential bottom, respectively (Fig. 1). The coarse mesh (4 mm) bags allowed the access of both the whole soil fauna and understory fine roots. Soil fauna ideally had higher mobility and discrimination than plant roots at all dimensions in the soil. Thus, we inferred that the fine mesh (0.038 mm) bags could allow the access of most of the soil fauna through 4×4 mm^2 holes, whilst excluding the understory fine roots from soil by the 0.038 mm meshes. The water permeability of fine meshes was also tested. The results showed that the fine meshes allowed the passage of water and gasses.

Defaunated soils originating from each site were prepared as follows. A rectangular area of approximately 30 m $\times$ 12 m was demarcated in each forest. Approximately 8 parallel transects 1.5 m apart were established, because of the irregular topography, and approximately 20 points were located 1 m apart along each transect. A total of 160 sampling points were marked and numbered in each site. The topsoil (17 cm or so in depth) was fetched on each point using a sampler with 13 cm inside diameter. The 160 holes dug during sampling were retained for the refilling of the defaunated soils. Plant roots were eliminated from the collected soils. The soils were defaunated by steam sterilization for 2 h, mixed sufficiently and air-dried.

Half of the 160 holes in each site were randomly selected to install 80 fine mesh bags, and the other half for 80 coarse mesh bags. Both halves of the two types of mesh bags fitted into the holes in sites A and B were randomly selected and refilled with the defaunated soil originating from site A (SoilA: untransferred soil in site A and transferred soil in site B). The left mesh bags were refilled with defaunated soil originating from site B (SoilB: transferred soil in site A and untransferred soil in site B). Another reciprocal soil transfer was similarly conducted between sites C and D. The soil cores formed in the mesh bags were approximately 17 cm high and 13 cm in diameter. Half of both transferred and untransferred soil cores in sites A and C were covered with 10 g of dry weight of fresh leaf litter of *C. fargesii* contained in nylon bags (4 mm mesh). The other half was not covered with litter but with plastic threads to simulate the physical effect of litter covering. The same litter manipulation was implemented on sites B and D with leaf litter of *C. lanceolata*. Thus, a total of 640 soil cores were established, that is, 4 sites $\times$ 2 types of mesh bags $\times$ 2 soil sources $\times$ 2 litter manipulations $\times$ 5 replicates $\times$ 4 times of sampling (Fig. 2). The effects of litter manipulation and soil transfer on soil organic carbon and soil fauna were not the focuses here and not addressed in this report. However, the two factors were involved in the following statistical tests to obtain reliable results. This field experiment was established from April to May in 2011.

2.2. Sampling and laboratory analysis

The 160 soil cores incubated in the field for the fourth sampling were sampled after 805 days for faunal extraction, measurement of soil organic carbon and the biomass of understory fine roots. Destructive soil sampling for faunal extraction was performed in each soil core using a sampler with 4.5 cm inside diameter from three layers (0–5, 5–10 and 10–15 cm) twice corresponding to two different extraction apparatuses. One apparatus was a Tullgren dry funnel, which mainly extracts microarthropods [31], whilst the other was a modified Baermann funnel, which mainly extracts hydrobionts, such as Nematode and Enchytraeidae. The fauna individuals were preserved in 70% alcohol for identification and count. Most specimens were identified to order level while the class was ascertained for Nematode and the family for Enchytraeidae. Abundance of each taxon was counted in each sample. After soil sampling for fauna had been conducted, the understory fine roots (width $\leq$ 2 mm) were collected by hand from the destroyed soil