



Effects of rhizospheres on the community composition of Collembola in a temperate forest



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ABSTRACT

Influence of living roots on the community composition of Collembola was investigated in a coniferous forest of *Chamaecyparis obtusa*. We conducted a buried pot experiment that constructed two different systems for carbon (C) availability. We used two types of pots with or without a *C. obtusa* seedling. The former pots were used to make a system with soil including living roots (i.e. a system based on root-derived and litter-derived C), while the latter was equivalent to soil system without living roots (i.e. a system based on litter-derived C). After 8 months, we harvested the pots and examined the collembolan community and environmental factors. The presence of living roots affected collembolan abundance and species-specific responses. These changes could be explained in terms of leaf biomass of the seedlings, indicating a possible linkage between above-ground productivity and collembolan community. Given the possibility that root-derived C is associated with aboveground plant mass, we speculate that collembolan community responds to root-derived C. The three dominant species, which are widespread in Japanese temperate forests, were more abundant in the presence of living roots. Moreover they were positively correlated with leaf biomass in the system with a seedling, again suggesting the fundamental importance of living roots for the organisation of the collembolan community.

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1. Introduction

Collembola are one of the most widespread and abundant groups of arthropods in terrestrial ecosystems (Hopkin, 1997). Their density reaches up to hundreds of thousands individuals per m² and species richness to 50–60 species in an ecosystem (Petersen and Luxton, 1982), but information about species biological characteristics (e.g., trophic role, or factors influencing abundance and distribution) is still scarce. Recent studies revealed that ecosystem functions of soil animals were affected largely by their species composition, rather than total abundance or species richness (Cragg and Bardgett, 2001; Eisenhauer et al., 2011; Heemsbergen et al., 2004). This could be because each species has distinct function derived

from species-specific characteristics, and thus, information about ecological species traits and the determinant factors of community composition is needed (Bardgett and Wardle, 2010; Van Straalen et al., 2008).

Studies on collembolan communities have been conducted primarily in connection with litter decomposition systems, because Collembola were considered to graze on humus or fungi involved in litter decomposition and contribute decomposition process (Filser, 2002). Those studies demonstrated that organic matter content or litter quality affected the collembolan community (Detsis, 2009; Fujii and Takeda, 2012; Hasegawa, 2002; Pflug and Wolters, 2001; Takeda, 1987). However, these effects were often weak and detected for only a few species, and effects of physical environmental factors other than food resources were often higher (Filser, 2002; Fujii and Takeda, 2012; Hasegawa, 2002; Pflug and Wolters, 2001).

On the other hand, collembolan abundance and distribution have been known to be affected by plant roots (Eo and Nakamoto, 2008; Wiggins and Curl, 1979; Wiggins et al., 1979). Recent studies using isotope tracers have demonstrated that Collembola can

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depend on carbon (C) derived from living roots (e.g., rhizodeposits, or microorganisms in rhizospheres) (Albers et al., 2006; Endlweber et al., 2009; Murray et al., 2009; Ostle et al., 2007; Pollierer et al., 2007). In addition, Collembola were often shown to depend largely on root-derived C as compared to litter-derived C (Pollierer et al., 2007; Endlweber et al., 2009). These indicate that living roots may also be an important factor in collembolan communities, although little is known about the role of roots in organisation of collembolan community. In particular, microarthropods living in humus or mineral layer that is poor in litter-derived C sources (i.e., euedaphic species) have been considered to have close relationships with plant roots, compared to epigeic or hemiedaphic species involved in litter decomposition processes (e.g., Faber, 1991). Distribution of microarthropods in deeper soil tends to be further influenced by plant roots (Hishi et al., 2008; Parmelee et al., 1993), and euedaphic species often contribute to functioning of plant roots (e.g., nutrient uptake; Faber, 1991).

We investigated whether living roots influenced collembolan communities and which species showed a response to the root system. We conducted a field manipulative experiment consisting of two systems: soil including living roots (i.e., a system based on root-derived and litter-derived C) and soil excluding living roots (i.e., a system based on litter-derived C). We set up the two systems by burying two types of pots, with or without a seedling, and compared responses of the collembolan community in the two systems. Plant roots can affect collembolan communities by altering soil chemistry or the physical environment (e.g., soil moisture), in addition to supplying organic carbon. Thus, we also measured various environmental factors involved in plant roots and examined the correlations between these factors and collembolan communities. We hypothesised that presence of living roots may affect euedaphic species, likely resulting in the community-level shift of collembolan distribution with the vertical profile of soil.

2. Materials and methods

2.1. Site description

The study was conducted in a natural forest of *Chamaecyparis obtusa* at the Kamigamo Experimental Forest Station of Kyoto University, about 12 km north of Kyoto City (35°04' N, 135°43' E). *C. obtusa* is a shade-tolerant conifer, which is extensively planted for timber production in Japan and often regenerates naturally in some secondary forests. The study site is located at the top of a gradual hill (200 m asl). The annual precipitation was 1401 mm and the mean temperature was 14.9 °C in 2009. The canopy layer of the forest consisted primarily of *C. obtusa*, and the understory vegetation consisted of the shrubs *Cleyera japonica*, *Eurya japonica*, *Lyonia ovalifolia* and *Rhododendron macrosepalum* and *C. obtusa* saplings. The soil humus form in this study site was a Moder with an organic layer (A_0) about 3–5 cm thick above a poorly developed A horizon 1–2 cm thick and a BC layer. In the A_0 layer, fine roots of *C. obtusa* were densely distributed and formed a root mat.

2.2. Experimental design

A buried pot field experiment was conducted to construct a system with soil including plant roots and a system with soil excluding plant roots in the forest floor. In April 2009, one first-year seedling of *C. obtusa* was transplanted individually into half of the pots (the shape of the pots is 8 cm of height, 9 cm of top diameter and 6 cm of bottom diameter with three 7-mm-diameter holes in the bottom). The treatment of plant presence was often used to represent root-derived C input in microcosm experiments (e.g., Eisenhauer and Reich, 2012). Mean initial above- and below-ground biomass of

these seedlings was 0.242 (± 0.014) g and 0.135 (± 0.012) g, respectively (standard errors are given in parentheses, $n=5$). The pots were filled with soil that had been collected in the study site and homogenised thorough mixing to equalise the abiotic conditions and faunal distributions, although such a homogenisation procedure reduces faunal abundance (Maraun et al., 2003). Indeed, there may have been some minor influences of the homogenisation procedure on collembolan community, however, we found no major difference in species composition between the present experiment and previous studies that we conducted in the same forest (Takeda, 1987; Fujii and Takeda, 2012). We constructed two soil layers in the pots: the bottom parts (height 3.5 cm from the bottom of the pots) were filled with 10 mm mesh-sieved mineral soil and the upper parts (height 4.5 cm) were filled with 2 mm mesh-sieved organic soil. No seedling was transplanted into the other half of the pots. Eight pairs of pots (with and without a seedling) were buried in a study plot of 15 m \times 10 m. Each pair of the pots was positioned randomly more than 1 m apart from other pairs. To minimise the difference in physical soil condition in pots between the two systems (with and without a seedling) such as soil temperature and moisture, which can be influenced by the shade of the above-ground part, pot surfaces were covered with 3-mm-mesh plastic sheets.

2.3. Measurements

After eight months (in December 2009), to allow time for the proliferation of several generations of Collembola at the site (Takeda, 1987), we harvested all the pots. The above-ground parts of first-year seedlings of *C. obtusa*, composed almost of leaves, were cut off from pots with a seedling and dried at 70 °C to constant mass and weighed. Fresh soil for chemical analyses on both systems was sampled with a stainless-steel core (inside diameter 13 mm). Six soil cores were taken from the middle points between centre and circumference in six radial directions. The six cores were pooled, sieved with 2-mm mesh and used for measurements of soil dissolved organic C (DOC) content and soil water content. To measure DOC, each 8-g soil sample was extracted with 40 mL deionised water for 1 h and the suspension was filtered through a 0.45- μ m PTFE membrane filter (Advantec, Tokyo, Japan) after filtration through a filter paper (No. 5C; Advantec). The dissolved C content of the suspension was determined using a total organic C analyser (TOC-VC analyser; Shimadzu, Kyoto, Japan). Each fresh soil (approximately 2–3 g) was weighed and oven-dried at 105 °C for 48 h to determine its water content. The water content of each soil subsample was calculated using the following formula: water content = (fresh weight of soil – dry weight of soil)/dry weight of soil \times 100 (%). The rest of the soils from the six cores were dried at 40 °C to constant mass. These dry soils were used for measurements of soil pH and total C and N contents. The soil pH was determined using a glass electrode in a soil–water suspension of each 10-g soil sample shaken with 25 mL of ion exchange water. Total C and N contents were measured by combustion (NC analyser SUMIGRAPH NC-900; Sumitomo Chemical Co., Osaka Japan). The other soil in the pots was used for extraction of soil microarthropods. Collembola were extracted using a Tullgren funnel at 35 °C for 10 days and were identified and counted at the species level under an optical microscope at a magnification of 400 \times . After the extraction of Collembola, plant roots were separated from the soil, dried at 70 °C to constant mass and weighed.

2.4. Statistical analyses

Wilcoxon's signed-rank test was used to compare the mean values of soil chemical properties between the systems (with and without a seedling; $n=8$). The effect of treatment (with a seedling)

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