International Symposium "Root Research and Applications"
RootRAP, 2–4 September 2009, Boku – Vienna, Austria

Variation in fine root traits by branch order within a *Chamaecyparis obtusa* stand

Yasuhiro Hirano[1], Tomoe Ikeda[2], Naoki Makita[2],
Takeo Mizoguchi[1] and Yoichi Kanazawa[2]

[1] Kansai Research Center, Forestry and Forest Products Research Institute (FFPRI), 68 Nagai-Kyutaro, Momoyama, Fushimi, Kyoto 612-0855, Japan
[2] Graduate School of Agricultural Science, Kobe University, Kobe 657-8501, Japan
Contact: Yasuhiro Hirano, Phone: +81-75-611-1201, Fax: +81-75-611-1207,
E-Mail: yhirano@affrc.go.jp

ABSTRACT

Recent studies have revealed that both morphological and physiological variability exist within fine roots <2.0 mm in diameter and that an approach based on branch order should replace the arbitrary diameter approach to partition tree roots into homogenous units. Fine root traits by branch order have been clarified in several tree species, but variation in the traits of branch order roots to soil nitrogen (N) status had not yet been examined. Moreover, few studies have compared root traits by branch order to those by diameter class. The objective of this study was to clarify the relationships in root traits between branch order and diameter class and the variability in the root traits by branch order within a stand of *Chamaecyparis obtusa*, especially in relation to soil N status. In a *C. obtusa* stand (30 m × 60 m) on Mt. Hiei-zan in western Japan, 180 fine root systems were collected at a depth of 10 cm in July 2008. In total, 11,222 individual roots were dissected by branch order, with distal roots numbered as first-order roots. Roots that were <0.5 mm in diameter accounted for 72% of the total length among the first three root orders. The length of individual roots varied highly, by up to fivefold, within the stand, while variation in diameter was consistently low. The soil N concentration was positively correlated with the specific root length (SRL) of first-order roots and N concentrations of first- and second-order roots, but not with that of third-order roots or with the diameter and length of individual roots. We conclude that <0.5 mm diameter roots include mainly first- and second-order roots but also some third-order roots, and that roots <0.5 mm in diameter have different responses to soil N status in this tree species.

KEYWORDS: branch order, *Chamaecyparis obtusa*, root nitrogen, root tissue density (RTD), soil nitrogen, specific root length (SRL)

1. INTRODUCTION

Fine root dynamics is one of the least understood aspects of plant science, although it contributes significantly to the carbon cycle in forest ecosystems (e.g., Strand et al. 2008). Fine roots of forest trees have usually been defined based on an arbitrary diameter such as 2.0 mm and recognized as homogenous functional units (Pregitzer et al. 2002). Classifying roots by diameter class, however, often ignores relationships among root position, form, and function (Pregitzer et al. 2002). Recent studies have revealed that both morphological and physiological variability exist within roots <2 mm in diameter and that an approach based on branch order should replace the arbitrary diameter approach to partition tree roots into homogenous units (Guo et al. 2008a). Root traits by branch order have been clarified in several tree species, and in general, the most distal roots have thinner diameters with higher specific root lengths and nitrogen (N) concentrations (Wang et al. 2006). The variation in traits in relation to soil N status within one forest stand, however, had not yet been examined and few studies have compared roots classified by branch order to those classified by diameter class.

The objectives of this study were to clarify both the relationships in fine root traits between branch order and diameter class and the variability in root traits by branch order within a stand of *Chamaecyparis obtusa*, one of the most important plantation species in Japan, especially with regard to soil nitrogen N status.

2. MATERIALS AND METHODS

The study was undertaken in a 90-year-old *C. obtusa* stand at Mt. Hiei-zan, Ohtsu, Shiga, western Japan. The mean height and stem diameter of *C. obtusa* were 21 m and 38 cm, respectively, and the stand density was 333/ha. The soil is an Inceptisol derived from Palaezoic-Mesozoic sedimentary rock. The mean annual temperature and precipitation are 11^{o}C and 1500 mm, respectively. We established a 30 × 60 m plot and divided this into 18 subplots (10 × 10 m).

Ten intact segments of the distal portion of the branching fine root system and soil samples for N analysis in the upper 10-cm soil profile were carefully collected in each subplot in July 2008. Individual roots were dissected by order, with distal roots numbered as first-order roots according to Pregitzer et al. (2002). Individual roots from a segment were scanned immediately after dissection. The total length and mean root diameter were determined using WinRHIZO Pro 2007a software (Regent Instruments, Quebec, Canada). After scanning, the segments were dried, weighed and finely ground for N analysis. The specific root length (SRL; m/g) and root tissue density (RTD; g/cm^3) were calculated. The root branching ratio (Rb) was defined in terms of the number of daughter roots branching from a parent root (Wang et al. 2006). The total N concentration of roots and soils was measured with an NC analyzer (Sumigraph NC-900; Shimadzu, Kyoto, Japan).

To evaluate the root biomass and traits by different diameter classes, two 10-cm-long soil cores were sampled using a metal auger with an inner diameter of 5 cm in each plot during October 2008. Only live roots of *C. obtusa* were classified into the three classes by diameter: 0-0.5 mm, 0.5-1.0 mm and 1.0-2.0 mm. The same traits of roots as those for branch order were measured.

3. RESULTS AND DISCUSSION

3.1. Roots traits by branch order in intact segments

Lateral roots <0.5 mm in diameter accounted for 72% of the total length among the first three root orders. The mean diameters of first-, second-, and third-order roots were 0.45 mm, 0.47 mm, and 0.56 mm, respectively. The SRLs of the first-, second-, and third-order roots were 27.4, 23.3, and 15.4 m/g, respectively (Figure 1A). The mean length and diameter of individual roots increased but the SRL and N concentration decreased significantly with order. These trends were consistent with traits of other tree species described by Pregitzer et al. (2002) and Guo et al. (2008b). The length of individual roots varied highly, up to fivehold within the stand, while the variation in diameter was consistently low.

In *C. obtusa*, the first- and second-order roots had similar RTDs (0.25 and 0.26 g/cm^3, respectively) and the RTD (0.30 g/cm^3) of third-order roots was significantly higher than those of the first and second orders. The root branching ratio (Rb) between the second and first orders was 3.3 and lower than that between the third and second orders (6.5). The ratio in this species fell within the ranges of those in North American tree species (Pregitzer et al. 2002). The difference in the mean root diameter and length in *C. obtusa* between the first and second orders was lower than that between the second and third orders. These results suggest that the first- and second-order roots might have development stages similar to those of absorptive primary roots without a cork layer (Hishi & Takeda 2006).

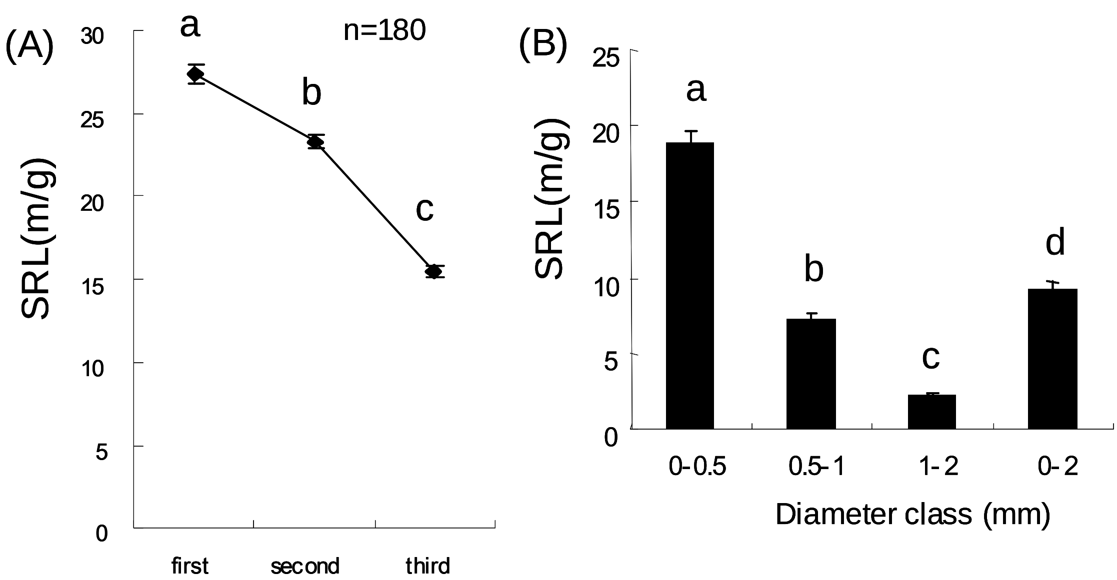

Figure 1. Specific root length (SRL; m/g) of the first-, second-, and third-order roots (A) and of roots in different diameter classes (B) in *Chamaecyparis obtusa* trees.

3.2. Roots traits by different diameter classes in coring

The mean biomass of fine roots <2.0 mm in diameter in the upper 10 cm of soils was 128 ± 10 g/m^2 (mean ± S.E.), and that of <0.5-mm-diameter roots was 63 g/m^2, which accounted for 49% of the total fine root biomass <2.0 mm. The SRLs in the diameter classes of roots <0.5 mm, 0.5-1.0 mm and 1.0-2.0 mm were 19.0, 7.3, and 2.4 m/g, respectively (Figure 1B). The RTDs of <0.5 mm and 0.5-1.0 mm roots were similar (0.30 and 0.34 g/cm^3, respectively), but significantly lower than that of 1.0-2.0-mm-diameter roots (0.39 g/cm^3). These results support the suggestion that roots <2.0 mm-diameter-roots, which have often been defined as "fine roots," have different functions. Specifically, the biomass of roots with diameters of <0.5 mm was half that of <2.0 mm-diameter-roots and had a higher SRL, corresponding to those of the first to third orders (Figure 1), which suggests that they have higher physiological activity, e.g., root respiration. Comparing the root traits by branch order, roots <0.5 mm in diameter included mainly first- and second-order roots but also some third-order roots.

3.3. Relationships among root traits by branch order and soil N status

The soil N concentration was positively correlated with the SRL of the first-order roots and with N concentrations of the first and second orders, but not with that of third-order roots or with the mean diameter and length of individual roots. The correlations between soil N and RTD of the first- to third-order roots was highly negatively significant.

We conclude that roots <0.5 mm in diameter have different responses to soil N status, depending on the position of an individual root on the branching root system, and that the responses of first-order roots are most sensitive to soil N status. The classification of fine roots by branch order provides significant insights into the linkage between root position and function (Wang et al. 2006, Guo et al. 2008b), but the disadvantage, in practice, is that the method is labor-intensive in contrast to the assessment of diameter classes. Our results comparing root traits both by branch order and diameter class suggest that detailed classification of fine roots <2.0 mm in diameter is needed, and that the classification of roots <0.5 mm in diameter, which include three root orders, is recommended to elucidate the high physiological activity in this tree species.

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