Presence of understory dwarf bamboo determines ecosystem fine root production in a cool-temperate forest in northern Japan

Sanae Yanagawa[a†], Karibu Fukuzawa[b]*, Kentaro Takagi[c], Hideaki Shibata[d], Fuyuki Satoh[d]

[a]Graduate School of Environmental Science, Hokkaido University, N9W9, Kita-ku, Sapporo, Hokkaido 060-0809, Japan; [b]Nakagawa Experimental Forest, Field Science Center for Northern Biosphere, Hokkaido University, 483 Otoineppu, Otoineppu, Hokkaido 098-2501, Japan; [c]Teshio Experimental Forest, Field Science Center for Northern Biosphere, Hokkaido University, Toikanbetsu, Horonobe, Hokkaido 098-2943, Japan; [d]Field Science Center for Northern Biosphere, Hokkaido University, N9W9, Kita-ku, Sapporo, Hokkaido 060-0809, Japan; [†]Sanae Yanagawa is deceased.

*Corresponding author: Karibu Fukuzawa, Nakagawa Experimental Forest, Field Science Center for Northern Biosphere, Hokkaido University, 483 Otoineppu, Otoineppu, Hokkaido 098-2501, Japan, caribu@fsc.hokudai.ac.jp

ORCiD

Karibu Fukuzawa 0000-0002-1490-2406

Kentaro Takagi 0000-0002-1321-2841

Hideaki Shibata 0000-0002-8968-3594

Presence of understory dwarf bamboo determines ecosystem fine root production in a cool-temperate forest in northern Japan

Abstract

Fine root biomass (FRB) and production (FRP) are crucial in forest carbon and nutrient cycling, but the factors controlling FRB and FRP are not well understood. Here, we examined FRB, FRP, aboveground environmental and stand factors, and soil environmental factors in four stands in a forest covered with dense understory vegetation of dwarf bamboo, *Sasa senanensis* (hereafter, Sasa). The four stands had different tree species composition and included a primary forest (PF), secondary forest (SF), conifer plantation (CP), and Sasa area (SA). We quantified the FRB and FRP of trees and Sasa separately using the ingrowth core method. Total FRP was higher in stands with substantial presence of Sasa (99–130 g m^{-2} yr^{-1}) than in CP with scarce Sasa (69 g m^{-2} yr^{-1}). Despite being occupied by Sasa alone, SA had high FRP, suggesting that the presence of Sasa regardless of trees is a key determinant of ecosystem FRP. Tree FRB increased with increasing tree aboveground biomass, tree density, or basal area at breast height, but Sasa FRB and total FRB decreased. Total FRP was also lower at higher values of these aboveground stand factors. In Sasa, specific root length was significantly higher, and root tissue density was significantly lower, than in trees, indicating the capacity of Sasa for explosive growth. Positive correlations between Sasa FRB or FRP and soil inorganic N or ammonium contents (i.e., N availability) were detected. We conclude that Sasa is important in determining FRB and FRP in this northern forest with understory vegetation.

Keywords: fine root biomass, species diversity, *Sasa senanensis*, tree aboveground biomass, soil nitrogen availability

Introduction

Forest is a huge carbon (C) sink in terrestrial ecosystems, accounting for 80% of aboveground C and 40% of belowground C (Dixon et al. 1994). Although fine root biomass (FRB) accounts for only 5% of total forest biomass (Vogt et al. 1996), fine root production (FRP) accounts for up to 76% of total net primary production (NPP) in forests (Gower et al. 1996), implying that fine roots are essential for the transfer of C and nutrients from vegetation to soil. In addition, uptake of water and nutrients by fine roots is crucial for plant survival and biogeochemical cycles (e.g., nitrogen (N) leaching). Therefore, fine root dynamics influences ecosystem functioning and services of forests (e.g., C sequestration and nutrient retention).

Multiple environmental and stand factors affect FRB, FRP, and root turnover rate. For example, FRB is higher in warm than in cool biomes, such as boreal forests, at a global scale (Vogt et al. 1996). Positive relationships have been demonstrated between FRB, FRP, and turnover rate, and air temperature and precipitation in boreal forests (Yuan and Chen 2010), and between turnover rate and air temperature on a global scale (Gill and Jackson 2000). FRB, FRP, or both are controlled by soil environmental factors, namely pH, and N and phosphorus (P) contents (Godbold et al. 2003; Yuan and Chen 2010), or N availability (i.e., inorganic N content: Aber et al. 1985; Nadelhoffer 2000). They are also controlled by stand factors, namely stand age (Yuan and Chen 2010) and basal area at breast height (BA, Finér et al. 2011a, b). Chen et al. (2004) demonstrated a positive relationship between FRB and BA, suggesting that the former can be predicted from the latter. Positive relationships have been reported between FRB and FRP (Finér et al. 2011b) and between FRP and root turnover rate (McCormack et al. 2014). However, understanding the relationship among these three parameters is insufficient because FRB is influenced not only by FRP but also by root turnover rate (Aber et al. 1989), and reports on FRP and turnover rate are scarce. Fine root dynamics depends on climatic, aboveground stand, and soil environmental factors. Site-scale analysis allows us to disregard climatic factors and focus on stand factors.

Although high plant species diversity increases aboveground NPP in grasslands (Tilman et al. 1996), the relationship between species diversity and FRP is not well understood. In forest ecosystems, some studies revealed FRP increases with increasing diversity of tree species (e.g., Brassard et al. 2013). However, reports of the species diversity–FRP relationship in forest ecosystems are scarce and more studies in diverse ecosystems are necessary.

Root traits affect foraging for soil nutrients. Thin roots with great surface area in contact with soil are advantageous for nutrient foraging, but such roots are ephemeral because of their vulnerable structure (Eissenstat and Yanai 1997). Specific root length (SRL: root length per unit weight) is an indicator of this trade-off: high-SRL (thin) roots are advantageous for nutrient foraging, whereas low-SRL (thick) roots have structural and maintenance advantages (Eissenstat and Yanai 1997; Ostonen et al. 2007). McCormack et al. (2012) demonstrated an inverse relationship between SRL and root lifespan and attributed the short lifespan of thin roots (high SRL) to low C investment in such roots. Root tissue density (RTD: root weight per unit volume) is another useful root trait indicator: roots with low RTD have an ability to explore for nutrients and are productive but short-lived (Ryser 1996). Measurements of root traits and those of FRB and FRP would provide useful insight into the mechanisms of fine root dynamics.

Fine root phenology is also important because it explains the detailed mechanism of fine root dynamics and influences water and nutrient dynamics in the soil. Tierney et al. (2003) suggested that environmental and endogenous factors affect the timing of FRP. Species-specific patterns have been demonstrated in a pot experiment (Makoto et al. 2020) and by literature analysis of ecosystem observation (Abramoff and Finzi 2015). However, information on the timing of FRP of distinct plant species or forms in the same fields is scarce.

Some cool-temperate or boreal forests have understory vegetation (e.g., herbs, shrubs). Reportedly, FRB and FRP cannot be predicted well unless understory vegetation roots are considered (Finér et al. 2011a, b); understory vegetation increases total FRB (Finér et al. 2007; Helmisaari et al. 2007; Hansson et al. 2013). However, the role of understory vegetation in fine root dynamics (e.g., FRB, FRP, turnover rate, root phenology) is not clear in the majority of studies because separating roots of understory vegetation from tree roots is difficult and labour intensive. It is possible that this role depends on understory species.

On Hokkaido Island, northern Japan, dwarf bamboo, *Sasa* spp. (hereafter Sasa; Gramineae) covers 89% of the forest area (Toyooka 1983). *Sasa senanensis* allocates half of biomass to belowground parts (Fukuzawa et al. 2015) and its FRB accounts for 59%–88% of the total FRB in a cool-temperate forest in northern Hokkaido (Fukuzawa et al. 2013). However, it is unclear whether total FRB or FRP and the proportion of Sasa roots to total roots change depending on stand type (e.g., tree species, tree aboveground biomass, BA, tree density, and soil environment). Such information could facilitate general understanding of the fine root dynamics in various tree–Sasa ecosystems in northern Japan. Furthermore, the FRP and the temporal patterns of production and turnover of Sasa and tree roots have not been identified. Root traits would influence fine root dynamics, however the differences in root traits between Sasa and trees in forests have not been clarified.

In the present study, we chose four stands covered with understory vegetation: three stands with different tree species composition and a Sasa area without trees. To predict the ecosystem FRB and FRP from the aboveground stand characteristics (tree density, BA, tree aboveground biomass, canopy openness), and soil environment, and clarify the contribution of Sasa to total FRB and FRP in tree–Sasa ecosystems, we quantified the FRB and FRP of trees and Sasa separately and investigated the relationship between FRBs or FRPs of trees, Sasa, and total and the stand characteristics. To understand the behaviour of the roots of trees and Sasa, we investigated seasonal changes and annual values of FRB and FRP and root traits of each plant form in four stand types. We hypothesized that (1) understory Sasa contributes to the total FRB and FRP and influences the relationship between them and tree aboveground stand factors (tree density, BA, tree aboveground biomass); (2) root traits (i.e., SRL and RTD) differ between trees and Sasa; (3) timing of FRP differs among stands as a reflection of different plant composition; and (4) FRB and FRP correlate with soil environmental factors, especially with soil N availability.

Materials and Methods

Study site

We established study plots in four stands in the Teshio Experimental Forest, Hokkaido University (45°03′N, 142°06′E) in northern Hokkaido, Japan. The stands are located on a flat ridge (70–80 m a.s.l.) within 1 km of each other. The selected representative stands were (1) conifer plantation (CP) of mature *Abies sachalinensis*, (2) primary forest stand (PF) dominated by *Quercus crispula* and composed of multiple broadleaved and conifer species, (3) secondary forest stand (SF) dominated by 69-year-old *Betula* *platyphylla*, and (4) Sasa area (SA) completely dominated by *S. senanensis* except for scarce young trees (Table S1). In 2005–2014, the mean annual air temperature was 5.7 °C and the total annual precipitation was 1190 mm at the meteorological station ca. 16 km south-west of the site (Teshio Experimental Forest); 30% of total annual precipitation fell as snow during November to April. The bedrock is Cretaceous sedimentary rock and the dominant soil is a Gleyic Cambisol (FAO, 1990).

In each stand, we randomly selected five individual target trees and established a plot for tree surveys (circles [10-m radius] around each target tree: type I) and a plot for fine root dynamics and aboveground and belowground environment (ca. 5 × 5 m: type II) with a centre 2 m away from each target tree (also within the type I plot) during July−August 2013. The target tree species were *A. sachalinensis* in CP, *Q. crispula* in PF, and *B. platyphylla* in SF. In SA, we randomly selected five plots in the Sasa vegetation community (ca. 1.5–1.7 m height).

Stand structure and aboveground biomass

We determined the overstory tree density and tree aboveground biomass in each plot in April 2014. We counted the trees to obtain tree density (trees ha^{-1}) and measured the diameter at breast height (1.3 m, DBH) of all individual trees in each type I plot. Then we calculated BA (m^2 ha^{-1}) as the sum of the basal areas at breast height of individual trees. To estimate tree aboveground biomass, we used the following allometric equation obtained from 22 individual trees with a wide DBH distribution (*Q. crispula*, *B. ermanii*, and *A. sachalinensis*) in the Teshio Experimental Forest (Takagi et al. 2010).

$$\ln Y = a \ln X + b \quad (1)$$

where X is DBH (cm), Y is aboveground biomass (kg), and a and b are constants (a = 2.428, $b = -2.282$, $r^2 = 0.994$). To evaluate the aboveground biomass of Sasa, we harvested its aboveground parts including culms and leaves in 50 cm × 50 cm quadrats in each plot in September 2014 after current-year leaves had completely expanded. We dried the culms and leaves separately (70 °C, 48 h) and weighed them.

Aboveground and belowground environments

We determined canopy openness, an indicator of light availability for the understory layer, in the centre of each type II plot; at 1.5 m above the ground, we took photographs in the zenith direction using a camera with a fish-eye converter (E4500 & FC-E8 0.21x, Nikon Corp., Tokyo, Japan; shutter speed, 1/250; aperture value, 2.6) in 2013 (September and November) and in 2014 (May and July) in the absence of direct solar radiation, and used the CanopOn2 software (URL: http://takenaka-akio.org/etc/canopon2/). We converted the hemispherical photos into black-and-white images and calculated the proportion of white area to estimate canopy openness. We measured soil temperature at 5-cm depth at two randomly selected plots for each stand at 1-h intervals from November 2013 to September 2014 using a thermometer with a data logger (UA-001-64, Onset Computer Corp., Bourne, MA, USA). We measured the mean soil volumetric water content of the surface 15 cm of soil using a time-domain reflectometer (TRIME-FM, IMKO GmbH Inc., Ettlingen, Germany) in September and November 2013. We measured soil gravimetric water content in collected soil (see below for the sampling method) by weighing soil before and after oven-drying (105 °C, 24 h) in September 2014. We also measured the thickness of the Oe/Oa layer at the points of soil environmental measurements in 2013 (August and November) and in 2014 (May and July).

Fine root biomass, production, and turnover

To measure FRB, we used in situ core sampling at the points of soil environmental measurements in each type II plot (one core at each time point) in 2013 (26 August and 6 November) and 2014 (21 May and 2 July). We removed the fresh litter (L) layer and collected the Oe/Oa layer and the 0–10 cm surface soil by auger (inner diameter: 4.4 cm). In each plot, sequential collection points were more than 20 cm apart from each other.

To measure FRP, we used in situ ingrowth cores (diameter: 4.4 cm, depth: 10 cm, 152 cm^3, lateral face: 4-mm polyethylene mesh). We collected soil to a depth of 10 cm at the representative point in each stand and sieved the soil through a 4-mm mesh to remove roots, used it to fill the ingrowth cores, and installed them into the 10-cm deep hole created by the FRB measurement in the soil. To measure FRP in the Oe/Oa layer, we put humus-filled ingrowth cores prepared similarly to those filled with soil on the installed soil-filled ingrowth cores. We established the ingrowth cores in both the Oe/Oa and soil layers (one core for each time period, each layer, and each plot) during 26 August–30 October 2013, 6 November 2013–14 May 2014, 21 May–1 July 2014, and 2 July–16 September 2014 to identify seasonal trends. To calculate annual FRP, we summed the FRP of each observation interval. We calculated fine root turnover (yr^{-1}) from FRP ($g\ m^{-2}\ yr^{-1}$) and FRB ($g\ m^{-2}$) according to the following equation (Dahlman and Kucera 1965; Gill and Jackson 2000):

$$\text{Fine root turnover} = \text{Annual FRP/annual mean FRB} \qquad (2)$$

Annual mean FRB was the temporal mean value calculated from the four collection times. For both FRB and FRP, we separated roots from soil by washing. We sieved soil through a 2-mm mesh and additionally used a 0.5-mm sieve attached below as a backup. We distinguished Sasa roots from tree roots by their light colour and branching style (Fig. S1). We selected roots <2 mm in diameter. We captured the images of the roots from each plot spread in a water-filled transparent acrylic box and measured total root length and root volume with a WinRHIZO root image analysis system (REG 2009, Regent Instruments Inc., Quebec, Canada) attached to a scanner (V700 Photo, Epson, Suwa, Japan). After imaging, we dried the roots (70 °C, 48 h) and weighed them. We calculated SRL (m g^{-1}) and RTD (g cm^{-3}) from the length, volume, and weight of roots in each plot.

Soil chemical properties

We determined soil C and N content, N availability, and soil environmental factors in the centre of each plot. We collected cores of the 0–10 cm surface soil layer after removing the Oe/Oa layer in September 2014 using an auger and removed gravel, roots, and coarse organic debris by sieving through a 2-mm mesh. To evaluate soil N availability, we extracted soil with KCl (fresh soil:2N KCl = 1:10, w/v; shaking for 1 h) and filtered the suspension (No. 5C, Advantec Inc., Tokyo, Japan). We measured nitrate and ammonium concentrations in the extract colorimetrically using a flow-injection N analyser (AACS-4, BL-TEC Inc., Osaka, Japan), calculated their contents per weight of dry soil, and added them to obtain inorganic N content. We extracted soil with water (dry soil:deionized water = 1:2.5, shaking for 1 h) and determined the pH of the suspension with a glass electrode (MM-60, TOA-DKK Inc., Tokyo, Japan). We ground oven-dried soil with an automated mortar (AMM-140D, Nitto Kagaku Co., Ltd., Nagoya, Japan) and analysed C and N contents with a CHNS/O analyser (PE2400II, Perkin Elmer Inc., Waltham, MA, USA).

Statistical analysis

We used two-way ANOVA to analyse the effects of stand type, season and their interaction on FRB, fine root length (FRL), and weight- and length-based FRP of total vegetation. We also used two-way ANOVA to analyse the effects of stand type, plant form (trees or Sasa), and their interaction on fine root turnover rate. We used the Tukey HSD test for multiple comparisons of FRB and FRL among stands and seasons. We used one-way ANOVA to analyse the effect of stand type on tree and Sasa aboveground biomass, the annual weight-based FRP of total vegetation, volumetric soil water content, soil ammonium and inorganic N contents, and soil pH. Before one-way ANOVA, we performed a square-root transformation for tree and Sasa aboveground biomass to ensure variance homogeneity. We used the Tukey HSD test after one-way ANOVA to compare stands. Because of the non-normal distribution or non-homogeneity of variance even after transformation, we used the Kruskal–Wallis test to analyse the effect of stand type on the annual length-based FRP of total vegetation, BA, tree density, Sasa culm density, canopy openness, Oe/Oa layer thickness, gravimetric soil water content, soil total C and N contents, soil C/N, and soil nitrate content, and then the Steele–Dwass test for the comparison among stands. We used three-way ANOVA to analyse the effects of plant form, soil layer, and stand type and their interactions on SRL and RTD. We conducted polynomial regression analysis using the least-squares method to identify the relationships between aboveground stand factors and FRB (mean of four seasons) or weight-based annual FRP in each plot. We assumed first- and second-order linear models and selected them on the basis of the Akaike information criterion (AIC). We applied a general linear mixed model (GLMM) using the maximum-likelihood method with a gamma error distribution and a log link to analyse the effects of soil properties on FRB and FRP of trees, Sasa, and total. We specified each soil property as a fixed effect and site (stand) as a random effect. We determined the significance of each fixed effect using analysis of deviance (type II test). In GLMM, we omitted soil temperature data from the analysis because of limited replication. We conducted correlation analysis for the relationship between stand mean FRB, FRP, and stand mean for each soil property. All statistical analyses were performed in R software (Version 4.0.3; R Core Team 2020).

Results

Aboveground stand factors and environment

The BA, tree density, and tree aboveground biomass were significantly higher in CP than in PF and SA (Table S2). These three parameters were significantly lower in SA than in the other stands. Sasa culm density and its aboveground biomass were significantly higher in SA than in CP, but no significant difference was found between SA and PF or SF (Table S2). Canopy openness was significantly lowest in CP and was 100% in SA (Table S2, Fig. S2).

Soil environmental factors

The thickness of the Oe/Oa layer was significantly lower in CP than in the other stands (Table S2). Mean annual soil temperature was lowest in CP, followed by SA, PF, and

SF (Table S2), with especially low temperature from December to April in CP (Fig. S3). In SF, soil volumetric water content was significantly lower than in the other stands, but soil gravimetric water content tended to be high (Table S2). Soil total C content and C/N ratio were significantly higher in SF than in the other stands, whereas soil total N content was not significantly different among stands (Table S2). Soil ammonium content was significantly higher in SA and PF than in CP (Table S2). Soil inorganic N content was significantly higher in SA and SF than in CP. Soil nitrate content was significantly higher in SF and SA than in PF, but the absolute values and ranges were smaller than those of ammonium, indicating that ammonium is the dominant form of inorganic N at the study site. Soil pH was significantly higher in SA than in CP and SF, and in PF than in SF (Table S2).

Fine root biomass and length

FRB differed significantly among stands, but not among seasons, and no interaction effect was observed (Table S3a). FRB was significantly higher in PF than in SF and CP and was significantly lower in CP than in the other stands (Fig. S4a). The average proportion of Sasa FRB to total FRB was extremely low in CP (8%), extremely high in SA (95%), and intermediate in PF and SF (Fig. S5a), indicating that CP and SA are composed of almost *A. sachalinensis* and Sasa, respectively. Despite the absence of trees in SA, its total FRB was similar to those of PF and SF and was significantly higher than that of CP (Fig. S4a).

FRL was significantly affected by both stand and season, with no significant interaction effect (Table S3b). FRL was significantly higher in SA than in PF and CP and was significantly lower in CP than in the other stands (Fig. S4b). FRL was significantly higher in early July 2014 than in late August and November 2013 (Fig. S4b).

Fine root production and turnover rate

Weight-based FRP was significantly affected by stand and season, with significant stand × season interaction (Table S3c). It was high in summer (July–September) in each stand, but the seasonal changes (whether FRP just peaked in summer or was continuously high during spring to summer [May–September]) depended on stands (Fig. S6a). Most FRP was found in soil, whereas FRP in the Oe/Oa layer was considerably lower (Fig. S6a), perhaps because of a methodological limitation of the use of ingrowth cores for estimating FRP in the Oe/Oa layer, such as dry layer or roots in the Oe/Oa layer originating from those in soil. The proportion of Sasa in weight-based FRP (Fig. S7a) was similar to that of FRB (Fig. S5). Length-based FRP was also significantly affected by stand and season, with significant stand × season interaction (Table S3d), and the trends among stands and seasons were similar to those of weight-based FRP (Fig. S6b). The proportion of Sasa in length-based FRP was similar to or slightly higher than that of weight-based FRP (Fig. S7b). Seasonal trends of weight- or length-based FRP of trees and Sasa were similar to those of the total FRP in PF and SF, where trees and Sasa co-exist (Fig. S6).

Weight-based annual FRP was significantly lower in CP than in SF and PF (Fig. 1a). Length-based annual FRP was significantly lower in CP than in the other stands (Fig. 1b). Fine root turnover rate was not significantly affected by stand, plant form, or their interaction (Table S4). We found positive relationships between FRB and FRP of trees, Sasa, and total (Fig. 2).

Root traits

The SRL was affected by plant form, layer (Oe/Oa layer vs. soil layer), and stand (Table S5a). The plant × layer, plant × stand, layer × stand, and plant × layer × stand interactions were significant. The SRL was significantly higher in Sasa than in trees, and in the Oe/Oa layer than in the soil layer (Table S5a, Fig. S8a). RTD was significantly affected by plant form, layer, and stand, and the plant × layer and plant × stand interactions were significant (Table S5b). RTD was significantly lower in Sasa than in trees, and in the Oe/Oa layer than in the soil layer (Table S5b, Fig. S8b).

Relationship between fine root dynamics and aboveground structure and environmental conditions

Tree FRB increased with increasing tree aboveground biomass, tree density, and BA (Fig. 3a–c), whereas Sasa and total FRB significantly decreased (Fig. 3a–c). On the other hand, Sasa and total FRB were high at high canopy openness (Fig. 3d) and Sasa FRB had a significant positive relationship with Sasa aboveground biomass (Fig. S9a), indicating that understory light affects not only Sasa aboveground biomass but also Sasa FRB.

Trends of the relationships between FRPs of trees and Sasa and aboveground stand factors were similar to those of FRB, but total FRP was highest in the middle range of the aboveground stand factors (Fig. 4a–c). Sasa FRP was high at higher canopy openness, but there was a significant negative relationship between tree FRP and canopy openness (Fig. 4d).

In GLMM analysis, soil pH significantly negatively affected total and Sasa FRP, soil ammonium content significantly positively affected tree FRB and FRP, Oe/Oa layer thickness significantly positively affected tree FRB, and Soil C and N contents significantly positively affected total and tree FRP (Table S6). Other soil properties did not significantly affect total, Sasa, or tree FRB or FRP. We found significant or marginal positive correlations between the mean values of soil inorganic N or ammonium contents and Sasa FRB or FRP (Fig. S10a, b, k, l) and marginal positive correlations between Oe/Oa layer thickness or soil temperature and total FRP (Fig. S10o, q). We also found tendencies of positive correlations with $|r| \geq 0.7$ between the mean values of soil inorganic N or ammonium contents and total FRB (Fig. S10a, b), between Oe/Oa thickness or soil temperature and total FRB (Fig. S10e, g), between soil pH and Sasa FRB or FRP (Fig. 10d, n), and tendencies of negative correlations between the mean values of soil inorganic N or ammonium contents and tree FRP (Fig. S10k, l) and between soil pH or soil water content and tree FRP (Fig. S10n, p).

Discussion

Understory vegetation changes the relationship between FRB and stand factors

Tree FRB and FRP increased with tree aboveground biomass, tree density, and BA (Figs. 3, 4). These trends of tree FRB agree with previous studies (Karizumi 1974; Finér et al. 2011a; Zhou et al. 2018). In contrast, total FRB had negative relationships with these parameters because of a strong negative relationship between them and Sasa FRB (Fig. 3). Sasa FRB was positively correlated with Sasa aboveground biomass, which was dependent on understory light (Table S2; Fig. S9a). Finér et al. (2011a) demonstrated that aboveground stand factors (e.g., tree density, BA, tree aboveground biomass) explained tree FRB but not total FRB, which included the FRB of understory vegetation, indicating that understory vegetation weakens the relationship between FRB and aboveground stand factors. On the other hand, the present study showed a negative relationship between total FRB and the aboveground stand factors in the presence of understory Sasa, because inverse relationships between trees and Sasa complement each other in this forest ecosystem (Fig. S9b, c; Fukuzawa et al. 2007, 2013). The regression lines of total FRP against tree aboveground biomass, BA, and tree density had maxima at around the intermediate values of these factors and were asymmetric (Fig. 4), indicating that total FRP was higher at lower values of these factors. However, total FRP did not differ significantly among PF, SF, and SA (Fig. 1). In this study, a significant positive relationship was observed between FRB and FRP (Fig. 2). Similar turnover rates among stands and plant forms (Tables S5, S6) also support this relationship. Using a global dataset, Finér et al. (2011b) showed that FRP is explained by FRB, and the present study confirms their result.

Understory vegetation determines ecosystem FRP

Weight-based annual total FRP was higher in stands with a substantial presence of Sasa than in the stand composed of a single tree species with scarce Sasa (CP), and FRP in SA, with negligible trees, was as high as that in PF and SF, where trees and Sasa co-exist (Fig. 1). These results suggest that the presence of Sasa, regardless of the presence or absence of trees, is a key determinant of ecosystem FRP. A large contribution of Sasa to total FRB (up to 57%), FRL (up to 75%), or FRP (59% and 72% for weight-based and length-based FRP, respectively) in stands with trees (Figs. 1, S5, S7) agrees with the reports of 71% and 59%–88% Sasa contribution in this forest (Fukuzawa et al. 2007, 2013), which may be the upper limit of the contribution of understory vegetation worldwide, and a report by Helmisaari et al. (2007) that showed a contribution of up to

50% by understory vegetation to total FRB in boreal forests in northern Finland. Seasonal mean FRB (891 g m^{-2}) was higher in a 60-cm soil profile in a forest covered with Sasa (Fukuzawa et al. 2013) than global mean FRB estimates (526–776 g m^{-2}) for the whole rooting depths in any of boreal, temperate, or tropical forests (Finér et al. 2011a).

An increase in FRP with increasing tree species diversity has been reported in a boreal forest in Canada (Brassard et al. 2009, 2013; Ma and Chen 2017). Meta-analysis also revealed an increase in FRP with increasing species diversity in multiple ecosystems including grasslands and forests (Ma and Chen 2016). In contrast, the positive effect of species diversity is small in young plantations (Domisch et al. 2015; Ma and Chen 2016). Brassard et al. (2013) suggested greater soil volume filling by a mixture of species with species-specific spatial and temporal patterns of root placement and proliferation, i.e., niche differentiation, as a cause of the increase in FRP with increasing tree species diversity. Ma and Chen (2017) also proposed that FRP can increase as a result of horizontal soil volume filling. Alternative mechanisms include the lack of pathogen-constrained root growth, which are typical in monocultures (de Kroon et al. 2012) and sampling effect, which results in an apparently greater probability of dominance by highly productive species with increasing species diversity (Wardle 1999). In the present study, the presence of Sasa increased total FRP in forest stands (PF and SF) in comparison with the single-tree species stand (CP) by adding Sasa FRP to the stable tree FRP (Fig. 1). This addition may be attributed to the complete cover of the land surface by Sasa. However, similar vertical root distributions of trees and Sasa (Fukuzawa et al. 2007) suggest that they would use the same vertical niche. On the other hand, the present study did not reveal higher FRP in mixtures of trees and Sasa

(PF and SF) than in SA (Fig. 1, Table S1); thus, we conclude that these mixtures do not always promote FRP. Then why was FRP in SA high despite its almost single-species composition?

SRL was significantly higher in Sasa than in trees (Fig. S8, Table S5), although we did not separate tree roots by species. Eissenstat (1991) highlighted the positive relationship of SRL with FRP and turnover rate, suggesting that roots with higher SRL are more productive. RTD was significantly lower in Sasa than in trees (Fig. S8, Table S5). Although RTD is often negatively correlated with SRL (Withington et al. 2006), Kramer-Walter et al. (2016) proposed that RTD is independent from SRL and that species with low RTD are fast-growing highly productive species. Our data on SRL and RTD indicate that Sasa grows faster and is more productive than trees, which is related to its ability of foraging for water and nutrients. This feature of root traits in Sasa is consistent with that of graminoids in a global dataset (Freschet et al. 2017), although we detected no difference in root turnover rate between trees and Sasa (Table S4). We speculate that Sasa, which has a capacity for explosive growth reflected in these root traits, increases FRP in SA, where nutrients, water, and light conditions are favourable because of the occupation by Sasa alone. This perspective would be important for evaluation of the fine root dynamics and ecosystem functioning in forests with a mixture of tree and grass species, such as cool-temperate forests in northern Japan.

A limitation of this study is that only a 10-cm-deep surface soil layer was analysed, although fine roots are also distributed in deeper layers. Fukuzawa et al. (2007) have surveyed a 60-cm soil profile in this forest and reported that 60% of fine roots are concentrated in a 15-cm-deep soil layer. They also revealed similar vertical patterns of the fine root biomass of the trees and Sasa as described above. These results imply that the fine root dynamics of the surface soil layer represents that of the whole soil layer (although the absolute FRP value may be underestimated) and suggest that the fine root dynamics of the whole soil layer can be extrapolated from the data on the interaction between trees and Sasa obtained in the present study.

Relationship between fine root dynamics and soil properties

The relationships between FRB or FRP and soil N availability (i.e., inorganic N content) in previous studies are controversial. Some studies reported negative relationships between FRB and soil N availability or N content in forests and suggested that plants can take up enough nutrients with fewer roots in fertile soil (Nadelhoffer 2000; Yuan and Chen 2010), whereas other studies suggested that roots often proliferate within nutrient (including N)-rich patches (Pregitzer et al. 1993; Hodge 2004). FRP is considered to increase with increasing soil N availability (Aber et al. 1989; Nadelhoffer 2000; Yuan and Chen 2012). However, meta-analysis of northern boreal or temperate forests showed an unclear effect of soil N availability on FRB and FRP (Brassard et al. 2009). In the present study, soil ammonium content significantly positively affected tree FRB and FRP after eliminating the effect of stand type (Table S6). Comparison of the stand means revealed positive correlations between Sasa FRB or FRP and soil inorganic N or ammonium contents (Fig. S10a, b, k, l). No significant relationships were found between FRB or FRP and soil nitrate content (Fig. S10c, m) because of low variation of the latter among stands and its low contribution to inorganic N at the study site. The above positive correlations are consistent with the reports of Pregitzer et al. (1993) and Hodge (2004).

Generally, soil N availability increases with nitrate leaching after disturbances such as the forest clear-cutting that created SA (Bormann and Likens 1994). However, Fukuzawa et al. (2006, 2015) showed that Sasa FRB increases and compensates for the reduction of tree roots immediately after selective or clear-cutting and mitigates nitrate leaching from the cleared site. Watanabe et al. (2016) reported a positive prompt response of Sasa aboveground biomass to N fertilization in forest. Favourable nutrients, water, and light without competition would enhance the above- and belowground growth of Sasa due to its ability to respond quickly to disturbances, as mentioned above. We cannot determine whether increased soil N availability would be maintained for the long term since it was increased by the disturbance or could be attributed to high productivity or a specific interaction in the rhizosphere (e.g., mycorrhizal colonization and root exudates) of this species. In the future, long-term changes in N dynamics in Sasa area after disturbances, and mechanisms of root and nutrient cycling should be clarified to characterize the ecosystem functioning in northern forests with mosaic structure including the Sasa areas (Inoue et al. 2017). Nevertheless, the present study revealed higher FRP and N availability in SA than in the tree stands.

Soil pH significantly negatively affected total and Sasa FRP (Table S6), but Sasa FRB and FRP tended to increase with increasing soil pH, whereas tree FRB and FRP tended to decrease in the comparisons among stands (sites) (Fig. S10d, n). The reason for this discrepancy between within-site and between-site relationships is unclear, but it might have been caused by a large variation among sites (Table S2). High soil pH stimulates root growth in various crop plants (Haynes, 1982) and is associated with high FRB in boreal forests (Yuan and Chen, 2010). A negative effect of excess aluminium ion on root growth at low pH is considered as a primary factor in relation to soil acidification (Hirano et al. 2007). In contrast, Godbold et al. (2003) reported high root growth, especially in the organic layer, at an acidic site in German Norway spruce forests. The opposite responses of tree FRB or FRP and those of Sasa to soil pH in the present study indicate that the effect of soil pH on root growth is not uniform. On the other hand, the positive relationship of Sasa FRB and FRP and soil pH is likely attributable to the cation-rich litter supply from Sasa, which prevents soil acidification in Japanese forests (Takamatsu et al. 1997).

Seasonal pattern of FRB and FRP

FRB did not significantly differ among seasons (Table S3a; Fig. S4a). These data agree with many previous studies (Aber et al. 1985; Yuan and Chen 2010). The absence of a trend may be caused by larger spatial heterogeneity during destructive root sampling than temporal variations (Fukuzawa et al. 2013). On the other hand, FRL was significantly affected by seasons, with high values in summer, despite destructive root sampling (Table S3b; Fig. S4b); these data agree with the seasonal pattern of root length in minirhizotron studies (Noguchi et al. 2005; Fukuzawa et al. 2013).

FRP was affected by season, with a significant stand × season interaction (Table S3c, d). FRP was high in PF and SA in spring and summer (May–September), but in summer (July–September) in SF and CP (Fig. S6). In SF, the lowest soil volumetric water content and the highest gravimetric water content indicated low soil bulk density. In addition, soil C content in SF was high, therefore high soil permeability may suppress root growth during the dry spring–early summer. The root growth pattern in CP is that of *A. sachalinensis* with a small contribution from Sasa. Abramoff and Finzi (2015) suggested that root growth in conifer species peaks later than in deciduous species. On the other hand, our data for SA suggest that a longer photosynthesis period in open sites enables Sasa to extend the root growth period (Fukuzawa et al. 2021).

Conclusion

The present study demonstrated that the total FRB and FRP do not parallel aboveground stand factors and the presence of Sasa regardless of the presence of trees is a key determinant of ecosystem FRP in a northern cool-temperate forest with dense understory vegetation. Lower Sasa FRP in the presence of trees suggests the competition effect for Sasa. The SRL was significantly higher in Sasa than in trees, and RTD was significantly lower, indicating that Sasa is a fast-growing highly productive species. We conclude that the roots of Sasa which has a capacity for explosive growth significantly affect fine root dynamics in forest ecosystems with dense understory vegetation. Our findings will be important for evaluation and prediction of biogeochemical cycling and ecosystem functioning in forests with understory vegetation.

Acknowledgments

We thank the staff of Teshio Experimental Forest, Hokkaido University, for their help in field surveys; the staff of North Research Office, Hokkaido University, for their help in chemical analysis; and members of the Course in Field Forest Science, Graduate School of Environmental Science, Hokkaido University, for their help in field surveys and valuable comments. We also thank Dr Toshiya Yoshida (Hokkaido University) for his help in the measurement of canopy openness and valuable comments on statistical analysis. This study was supported in part by JSPS KAKENHI (JP17K07830). This

research contributes to the Japan Long Term Ecological Research Network (JaLTER) and International LTER Network (ILTER).

Declaration of interest statement

Conflict of interest: The authors declare that they have no conflicts of interest.

Availability of data and material All data are available at https://db.cger.nies.go.jp/JaLTER/metacat/metacat/JaLTER-Hokkaido-Kita.1146.3/jalter.

References

Aber JD, Melillo JM, Nadelhoffer KJ, McClaugherty CA, Pastor J. 1985. Fine root turnover in forest ecosystems in relation to quality and form of nitrogen availability: a comparison of two methods. Oecologia 66:317-321.

Aber JD., Nadelhoffer KJ, Steudler P, Melillo JM. 1989. Nitrogen saturation in northern forest ecosystems. Bioscience 39:378-386.

Abramoff RZ, Finzi AC. 2015. Are above- and below-ground phenology in sync? New Phytol 205:1054-1061.

Bormann FH, Likens GE. 1994. Pattern and Process in a Forested Ecosystem. New York (NY): Springer-Verlag.

Brassard BW, Chen HYH, Bergeron Y. 2009. Influence of environmental variability on root dynamics in northern forests. Crit Rev Plant Sci 28:179-197.

Brassard BW, Chen, HYH, Cavard X, Laganiere J, Reich PB, Bergeron Y, Yuan ZY. 2013. Tree species diversity increases fine root productivity through increased soil volume filling. J Ecol 101:210-219.

Chen W, Zhang Q, Cihlar J, Bauhus J, Price DT. 2004. Estimating fine root biomass and production of boreal and cool temperate forests using aboveground measurements: A new approach. Plant Soil 265:31-46.

Dahlman RC, Kucera CL. 1965. Root productivity and turnover in native prairie.

Ecology 46:84 89.

de Kroon H, Hendriks M, Van Ruijven J, Ravenek J, Padilla FM, Jongejans E, Visser EJW, Mommer L. 2012. Root responses to nutrients and soil biota: drivers of species coexistence and ecosystem productivity. J Ecol 100:6-15.

Dixon RK, Brown S, Houghton RA, Solomon AM, Trexler MC, Wisniewski J. 1994. Carbon pools and flux of global forest ecosystems. Science 263:185-190.

Domisch T, Finér L, Dawud SM, Vesterdal L, Raulund-Rasmussen K. 2015. Does species richness affect fine root biomass and production in young forest plantations? Oecologia 177:581-594.

Eissenstat DM. 1991. On the relationship between specific root length and the rate of root proliferation: a field study using citrus rootstocks. New Phytol 118:63-68.

Eissenstat DM, Yanai RD. 1997. The ecology of root lifespan. Adv Ecol Res 27:1-60.

Finér L, Helmisaari HS, Lõhmus K, Majdi H, Brunner I, Børja I, Eldhuset T, Godbold D, Grebenc T, Konopka B, et al. 2007. Variation in fine root biomass of three European tree species: Beech (*Fagus sylvatica* L.), Norway spruce (*Picea abies* L. Karst.), and Scots pine (*Pinus sylvestris* L.). Plant Biosyst 141:394-405.

Finér L, Ohashi M, Noguchi K, Hirano Y. 2011a. Factors causing variation in fine root biomass in forest ecosystems. For Ecol Manag 261:265-277.

Finér L, Ohashi M, Noguchi K, Hirano Y. 2011b. Fine root production and turnover in forest ecosystems in relation to stand and environmental characteristics. For Ecol Manag 262:2008-2023.

[FAO] Food and Agriculture Organization of the United Nations, 1990. Soil map of the world. Revised legend. Reprinted with corrections 1990. World Soil Resources Reports 60. FAO-UNESCO-ISRIC, Rome.

Freschet GT, Valverde-Barrantes OJ, Tucker CM, Craine JM, McCormack ML, Violle C, Fort F, Blackwood CB, Urban-Mead KR, Iversen CM, et al. 2017. Climate, soil and plant functional types as drivers of global fine-root trait variation. J Ecol 105:1182-1196.

Fukuzawa K, Shibata H, Takagi K, Nomura M, Kurima N, Fukazawa T, Satoh F, Sasa K. 2006. Effects of clear-cutting on nitrogen leaching and fine root dynamics in a cool-temperate forested watershed in northern Japan. For Ecol Manag 225:257-261.

Fukuzawa K, Shibata H, Takagi K, Satoh F, Koike T, Sasa K. 2007. Vertical distribution and seasonal pattern of fine root dynamics in a cool-temperate forest in northern Japan: implication of the understory vegetation, Sasa dwarf bamboo. Ecol Res 22:485–495.

Fukuzawa K, Shibata H, Takagi K, Satoh F, Koike T, Sasa K. 2013. Temporal variation in fine root biomass, production and mortality in a cool temperate forest covered with dense understory vegetation in northern Japan. For Ecol Manag 310:700-710.

Fukuzawa K, Shibata H, Takagi K, Satoh F, Koike T, Sasa K. 2015. Roles of dominant understory Sasa bamboo in carbon and nitrogen dynamics following canopy tree removal in a cool-temperate forest in northern Japan. Plant Species Biol 30:104-115.

Fukuzawa K, Tateno R, Ugawa S, Watanabe T, Hosokawa N, Imada S, Shibata H. 2021. Timing of forest fine root production advances with reduced snow cover in northern Japan: implications for climate-induced change in understory and overstory competition. Oecologia 196:263-273.

Gill RA, Jackson RB. 2000. Global patterns of fine root turnover for terrestrial ecosystems. New Phytol 147:13-31.

Godbold DL, Fritz HW, Jentschke G, Meesenburg H, Rademacher P. 2003. Root turnover and root necromass accumulation of Norway spruce (*Picea abies*) are affected by soil acidity. Tree Physiol 23:915-921.

Gower ST, Pongracic, S, Landsberg JJ. 1996. A Global Trend in Belowground Carbon Allocation: Can We Use the Relationship at Smaller Scales? Ecology 77:1750-1755.

Hansson K, Helmisaari H-S, Sah SP, Lange H. 2013. Fine root production and turnover of tree and understorey vegetation in Scots pine, silver birch and Norway spruce stands in SW Sweden. For Ecol Manag 309:58-65.

Haynes RJ. 1982. Effects of liming on phosphate availability in acid soils. Plant Soil 68:289-308.

Helmisaari HS, Derome J, Nojd P, Kukkola M. 2007. Fine root biomass in relation to site and stand characteristics in Norway spruce and Scots pine stands. Tree Physiol 27:1493-1504.

Hirano Y, Mizoguchi T, Brunner I. 2007. Root parameters of forest trees as sensitive indicators of acidifying pollutants: a review of research of Japanese forest trees. J

For Res 12:134-142.
Hodge A. 2004. The plastic plant: root responses to heterogeneous supplies of nutrients. New Phytol 162:9-24.
Inoue T, Fukuzawa K, Watanabe T, Yoshida T, Shibata H. 2017. Spatial pattern of soil nitrogen availability and its relationship to stand structure in a coniferous-broadleaved mixed forest with a dense dwarf bamboo understory in northern Japan. Ecol Res 32:227-241.
Karizumi N. 1974. The mechanism and function of tree root in the process of forest production II Root biomass and distribution in stands. Bull Gov For Exp Sta 267:1-88.
Kramer-Walter KR, Bellingham PJ, Millar TR, Smissen RD, Richardson SJ, Laughlin DC. 2016. Root traits are multidimensional: specific root length is independent from root tissue density and the plant economic spectrum. J Ecol 104:1299-131.
Ma Z, Chen HYH. 2016. Effects of species diversity on fine root productivity in diverse ecosystems: a global meta-analysis. Glob Ecol Biogeogr 25:1387-1396.
Ma ZL, Chen HYH. 2017. Effects of species diversity on fine root productivity increase with stand development and associated mechanisms in a boreal forest. J Ecol 105:237-245.
Makoto K, Wilson SD, Sato T, Blume-Werry G, Cornelissen JHC. 2020. Synchronous and asynchronous root and shoot phenology in temperate woody seedlings. OIKOS 129:643-650.
McCormack ML, Adams TS, Smithwick EAH, Eissenstat DM. 2012. Predicting fine root lifespan from plant functional traits in temperate trees. New Phytol 195:823-831.
McCormack ML, Adams TS, Smithwick EAH, Eissenstat DM. 2014. Variability in root production, phenology, and turnover rate among 12 temperate tree species. Ecology 95:2224-2235.
Nadelhoffer KJ. 2000. The potential effects of nitrogen deposition on fine root production in forest ecosystems. New Phytol 147:131-139.
Noguchi K, Sakata T, Mizoguchi T, Takahashi M. 2005. Estimating the production and mortality of fine roots in a Japanese cedar (*Cryptomeria japonica* D. Don) plantation using a minirhizotron technique. J For Res 10:435-441.

Ostonen I, Puttsepp U, Biel C, Alberton O, Bakker MR, Lohmus K, Majdi H, Metcalfe D, Olsthoorn AFM, Pronk A, et al. 2007. Specific root length as an indicator of environmental change. Plant Biosyst 141:426-442.

Pregitzer KS, Hendrick RL, Fogel R. 1993. The demography of fine roots in response to patches of water and nitrogen. New Phytol 125:575-580.

R Core Team. 2020. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. https://www.r-project.org/

Ryser, P 1996. The importance of tissue density for growth and life span of leaves and roots: a comparison of five ecologically contrasting grasses. Funct Ecol 10:717–723.

Takagi K, Kotsuka C, Fukuzawa K, Kayama M, Makoto K, Watanabe T, Nomura M, Fukazawa T, Takahashi H, Hojyo H et al. 2010. Allometric Relationships and Carbon and Nitrogen Contents for Three Major Tree Species (*Quercus crispula*, *Betula ermanii*, and *Abies sachalinensis*) in Northern Hokkaido, Japan. Eurasian J For Res 13:1-7.

Takamatsu T, Kohno T, Ishida K, Sase H, Yoshida T, Morishita T. 1997. Role of dwarf bamboo (Sasa) community in retaining basic cations in soil and preventing soil acidification in mountainous areas of Japan. Plant Soil 192:167-179.

Tierney GL, Fahey TJ, Groffman PM, Hardy JP, Fitzhugh RD, Driscoll CT, Yavitt JB. 2003. Environmental control of fine root dynamics in a northern hardwood forest. Glob Change Biol 9:670-679.

Teshio Experimental Forest, Micrometeorology data (2002-): https://db.cger.nies.go.jp/JaLTER/metacat/metacat/JaLTER-Hokkaido-Kita.23.13/jalter

Tilman D, Wedin D, Knops J. 1996. Productivity and sustainability influenced by biodiversity in grassland ecosystems. Nature 379:718-720.

Toyooka H. 1983. Sasa growing in Hokkaido as biomass resources. Bamboo J 1:22–24 (in Japanese).

Vogt KA, Vogt DJ, Palmiotto PA, Boon P, O'Hara J, Asbjornsen H. 1996. Review of root dynamics in forest ecosystems grouped by climate, climatic forest type and species. Plant Soil 187:159-219.

Wardle DA 1999. Is "sampling effect" a problem for experiments investigating biodiversity-ecosystem function relationships? OIKOS 87:403-407.

Watanabe T, Fukuzawa K, Shibata H. 2016. Short-term response of Sasa dwarf bamboo to a change of soil nitrogen fertility in a forest ecosystem in northern Hokkaido, Japan. Plants 5:19.

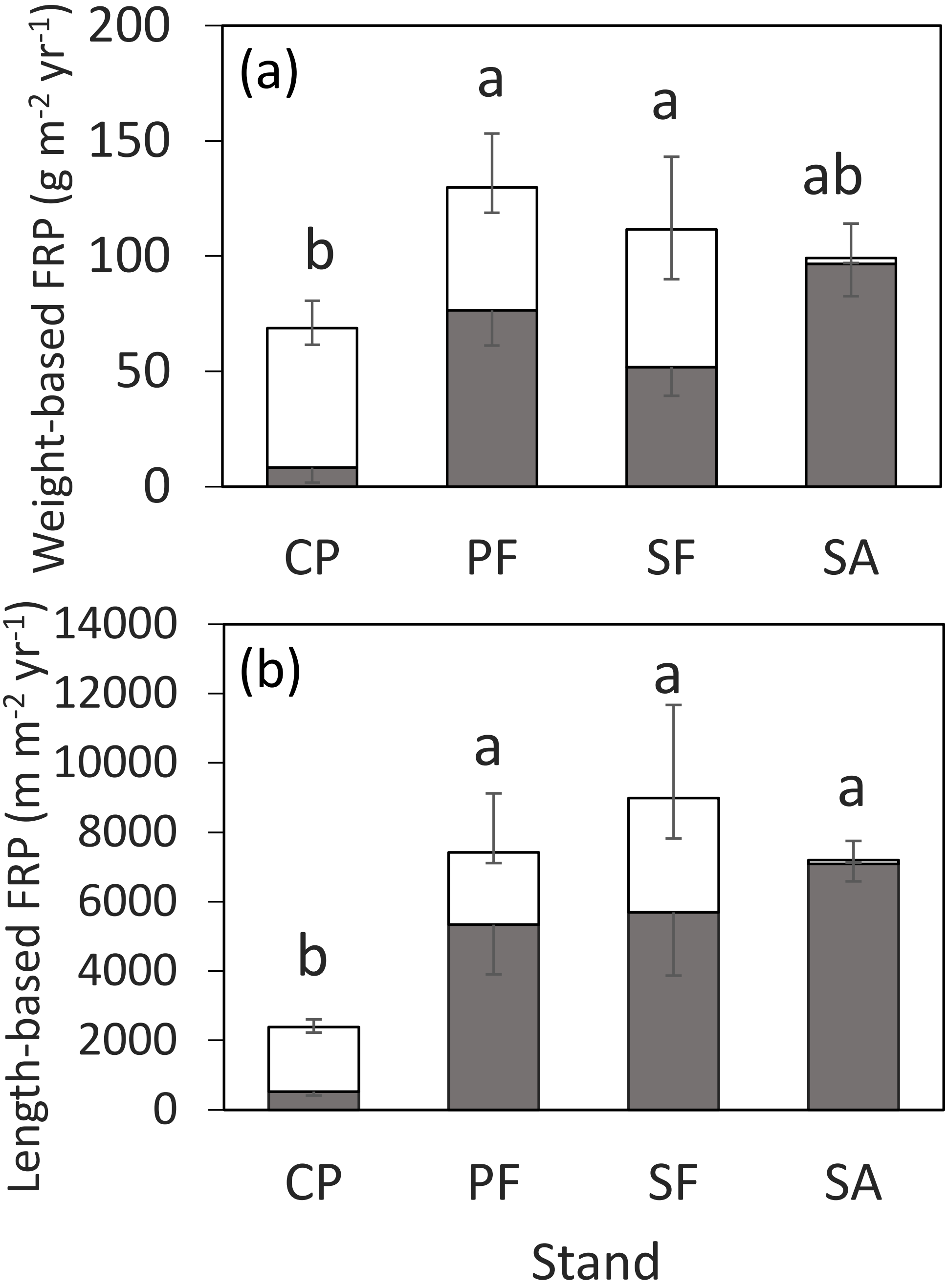
Withington JM, Reich PB, Oleksyn J, Eissenstat DM. 2006. Comparisons of structure and life span in roots and leaves among temperate trees. Ecol Monogr 76:381-397.

Yuan ZY, Chen HYH. 2010. Fine root Biomass, Production, Turnover Rates, and Nutrient Contents in Boreal Forest Ecosystems in Relation to Species, Climate, Fertility, and Stand Age: Literature Review and Meta-Analyses. Crit Rev Plant Sci 29:204-221.

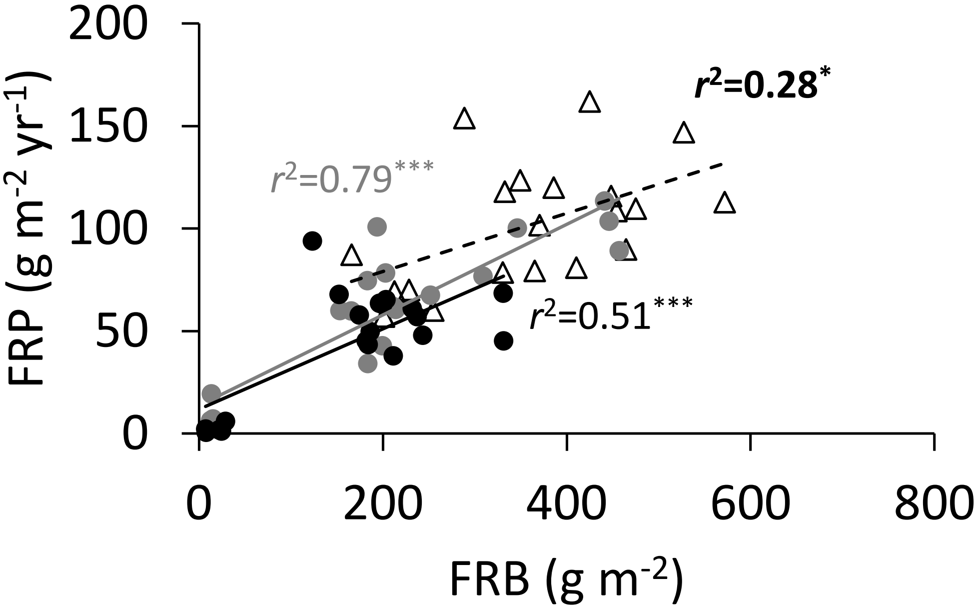
Yuan ZY, Chen HYH. 2012. A global analysis of fine root production as affected by soil nitrogen and phosphorus. Proc Royal Soc B: Biol Sci 279:3796-3802.

Zhou G, Meng S, Yu J, Zhou H, Liu Q. 2018. Quantitative relationships between fine roots and stand characteristics. Eur J For Res 137:385-399.

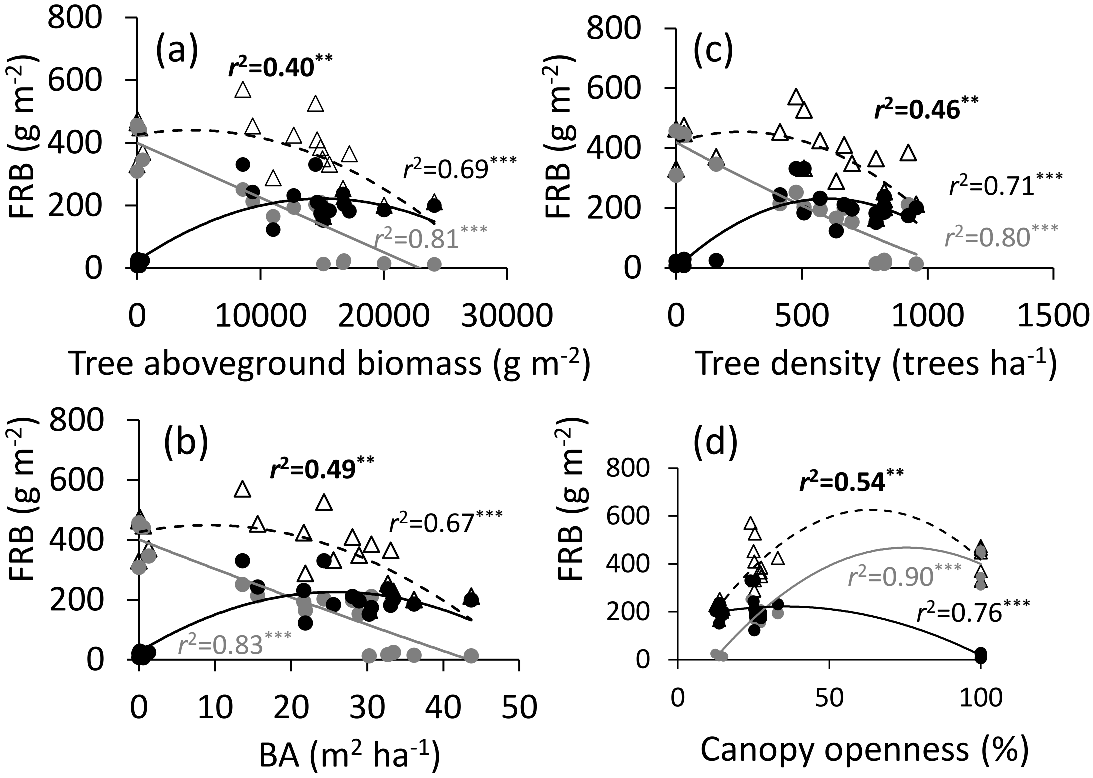
Figures



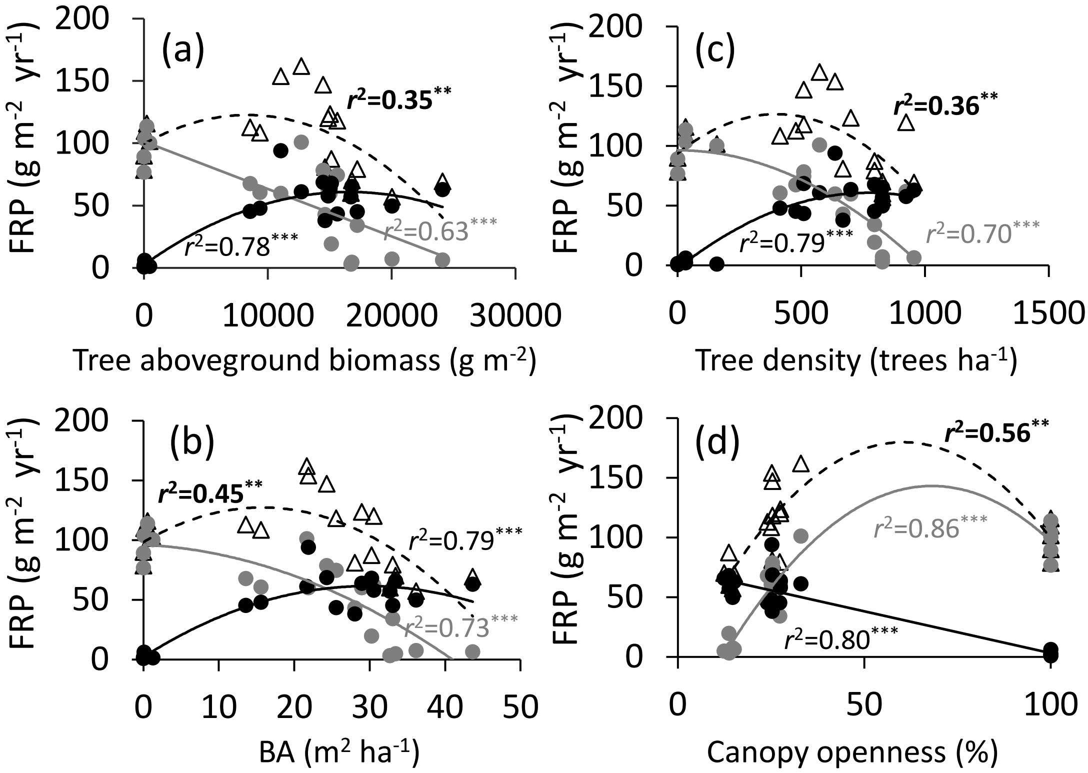
Yanagawa et al. Fig. 1



Yanagawa et al. Fig. 2



Yanagawa et al. Fig. 3



Yanagawa et al. Fig. 4

Figure captions

Figure 1. Annual weight-based (a) and length-based (b) fine root production (FRP) in each stand. Grey, Sasa; white, trees. Positive and negative error bars denote SD of total and of each plant form, respectively (n = 5). Total value is the sum of Oe/Oa and soil layers. Lowercase letters represent significant differences among stands ($P < 0.05$). CP, conifer plantation; PF, primary forest stand; SF, secondary forest stand; SA, Sasa area.

Figure 2. Relationships between fine root biomass (FRB) and fine root production (FRP) of trees, Sasa, and total. Trees, black circles and solid line; Sasa, grey circles and solid line; total, triangles and dashed line. The r^2 values are indicated in the corresponding shades (trees and Sasa) and in bold (total). *** $P < 0.001$; * $P < 0.05$.

Figure 3. Relationships between fine root biomass (FRB) and aboveground stand characteristics: tree aboveground biomass (a), tree density (b), basal area (BA) (c), and canopy openness (d). Trees, black circles and solid line; Sasa, grey circles and solid line; total, triangles and dashed line. The r^2 values are indicated in the corresponding shades (trees and Sasa) and in bold (total). Straight and curved lines denote the selected first-order or second-order linear models, respectively. *** $P < 0.001$; ** $P < 0.01$.

Figure 4. Relationships between fine root production (FRP) and aboveground stand characteristics: tree aboveground biomass (a), tree density (b), basal area (BA) (c), canopy openness (d). Trees, black circles and solid line; Sasa, grey circles and solid line; total, triangles and dashed line. The r^2 values are indicated in the corresponding shades (trees and Sasa) and in bold (total). Straight and curved lines denote the selected first-order or

second-order linear models, respectively. *** $P < 0.001$; ** $P < 0.01$; * $P < 0.05$.

Presense of understory dwarf bamboo determines ecosystem fine root production in a cool-temperate forest in northern Japan

Sanae Yanagawa, Karibu Fukuzawa*, Kentaro Takagi, Hideaki Shibata, Fuyuki Satoh

Journal of Forest Research, Online Resource

*Corresponding author:

Karibu Fukuzawa

Nakagawa Experimental Forest, Field Science Center for Northern Biosphere, Hokkaido University, 483 Otoineppu, Otoineppu, Hokkaido 098-2501, Japan, E-mail: caribu@fsc.hokudai.ac.jp; Tel: +81-1656-5-3216; Fax: +81-1656-5-3218

(a)

(b)

Figure S1. Photographs of Sasa (*Sasa senanensis*) (a) and tree (b) roots.

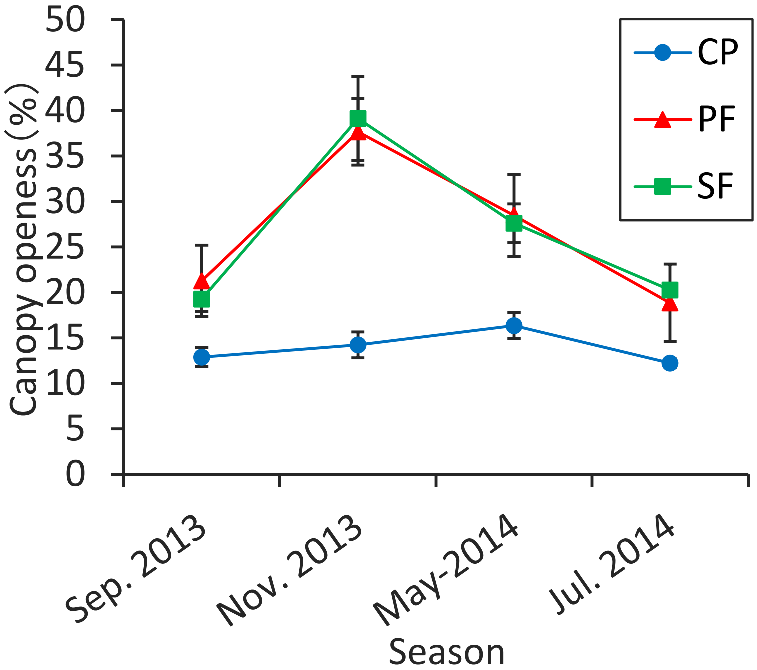


Figure S2. Canopy openness in each stand in four seasons. In the Sasa area, canopy openness was 100% at all times and the data are not shown. Error bars denote SD (n = 5). CP, conifer plantation; PF, primary forest; SF, secondary forest.

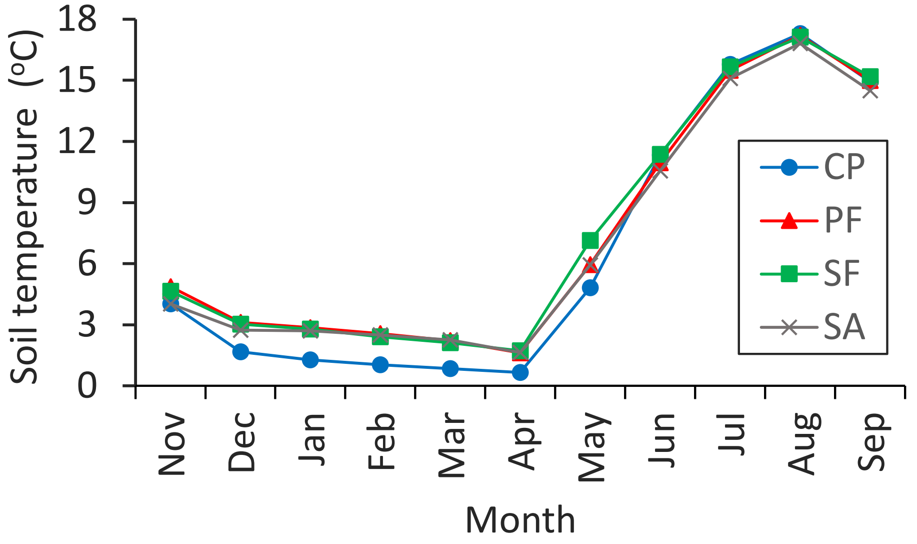


Figure S3. Mean monthly soil temperature in each stand from November 2013 to September 2014. CP, conifer plantation; PF, primary forest; SF, secondary forest; SA, Sasa area.

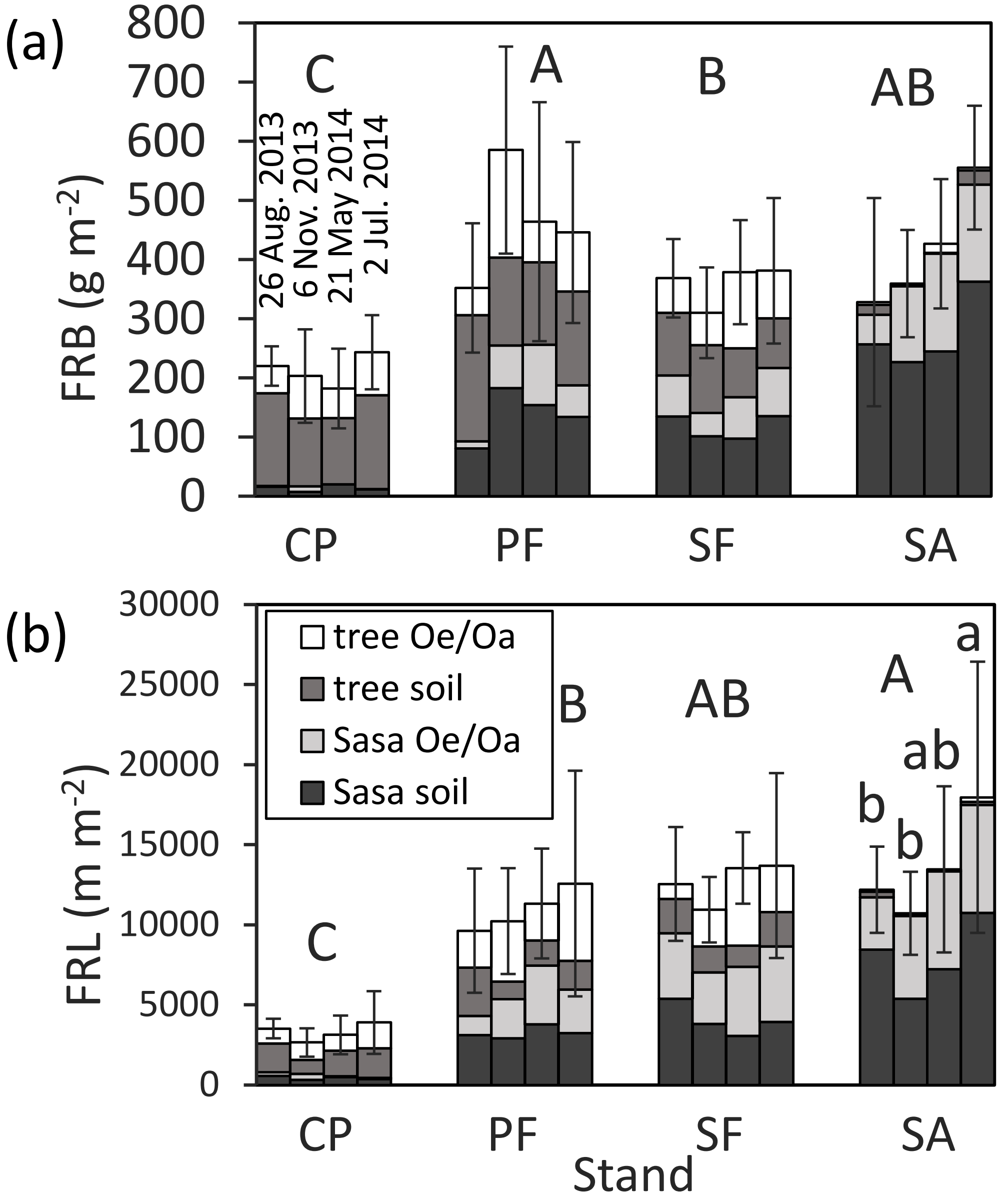


Figure S4. Seasonal changes in fine root biomass (FRB) (a) and fine root length (FRL) (b) in each stand. Error bars denote SD of total FRB or FRL (n = 5). Soil depth was 0–10 cm. Different capital and lowercase letters indicate significant differences ($P < 0.05$) among stands and seasons in the whole dataset, respectively. CP, conifer plantation; PF, primary forest; SF, secondary forest, SA, Sasa area.

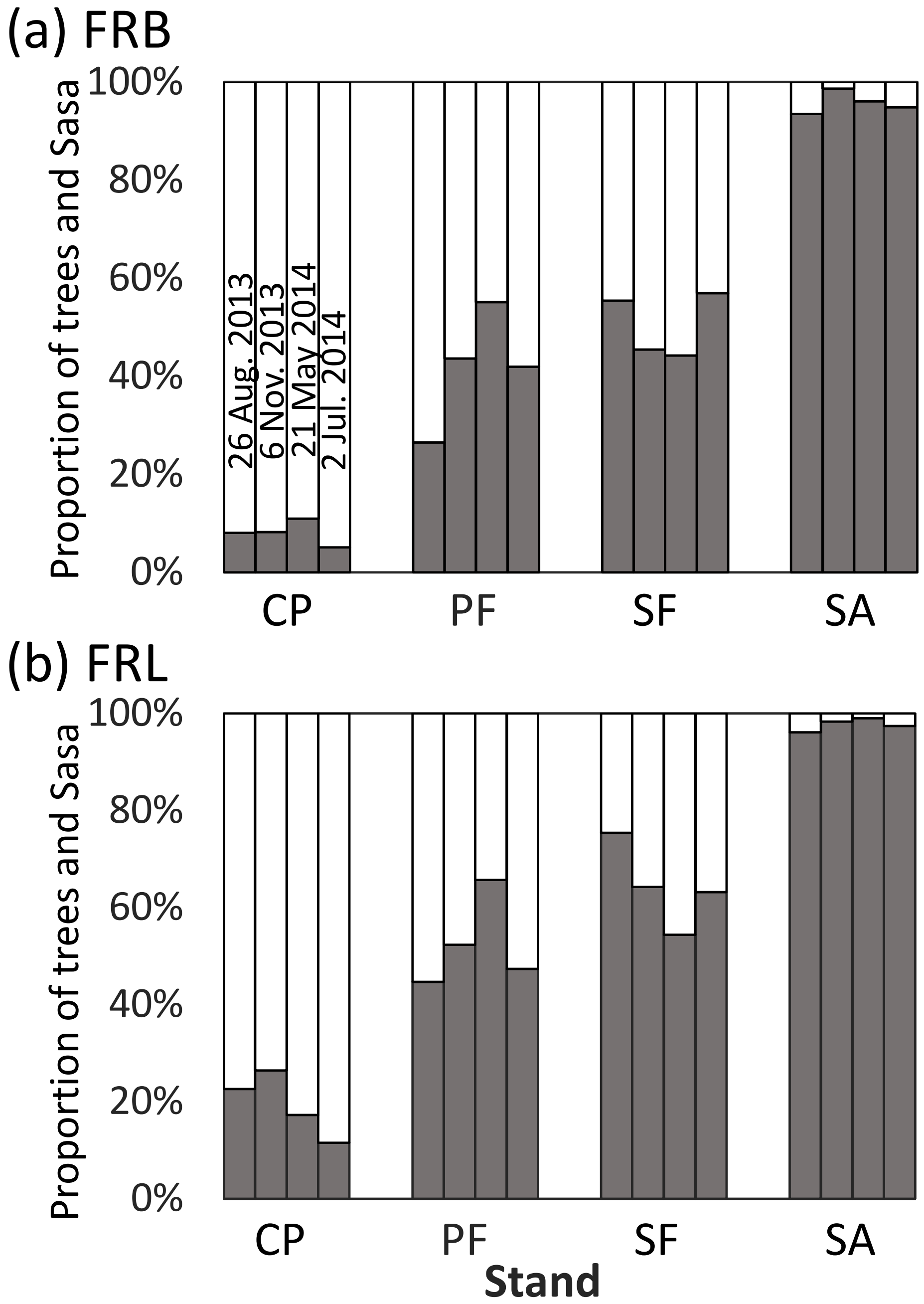


Figure S5. Proportion of trees and Sasa roots in fine root biomass (FRB) (a) and fine-root length (FRL) (b) for all layers (Oe/Oa and soil). Grey, Sasa; white, trees. CP, conifer plantation; PF, primary forest; SF, secondary forest, SA, Sasa area.

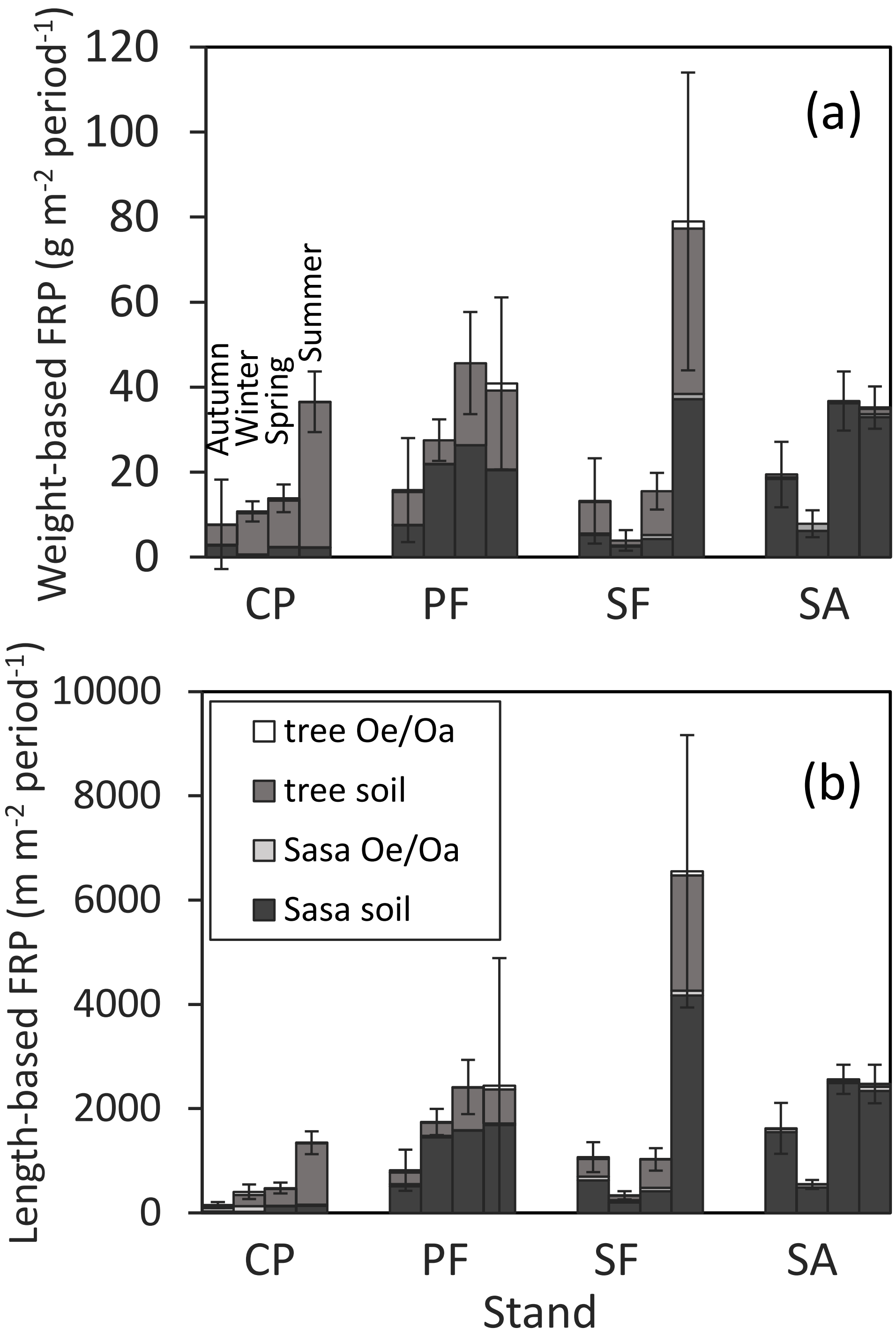


Figure S6. Seasonal changes in weight-based (a) and length-based (b) fine root production (FRP) in each stand. Error bars denote SD of total FRPs (n = 5). Soil depth was 0–10 cm. CP, conifer plantation; PF, primary forest; SF, secondary forest, SA, Sasa area. Autumn, 26 August–30 October 2013; Winter, 6 November 2013–14 May 2014; Spring, 21 May–1 July 2014; Summer, 2 July–16 September 2014.

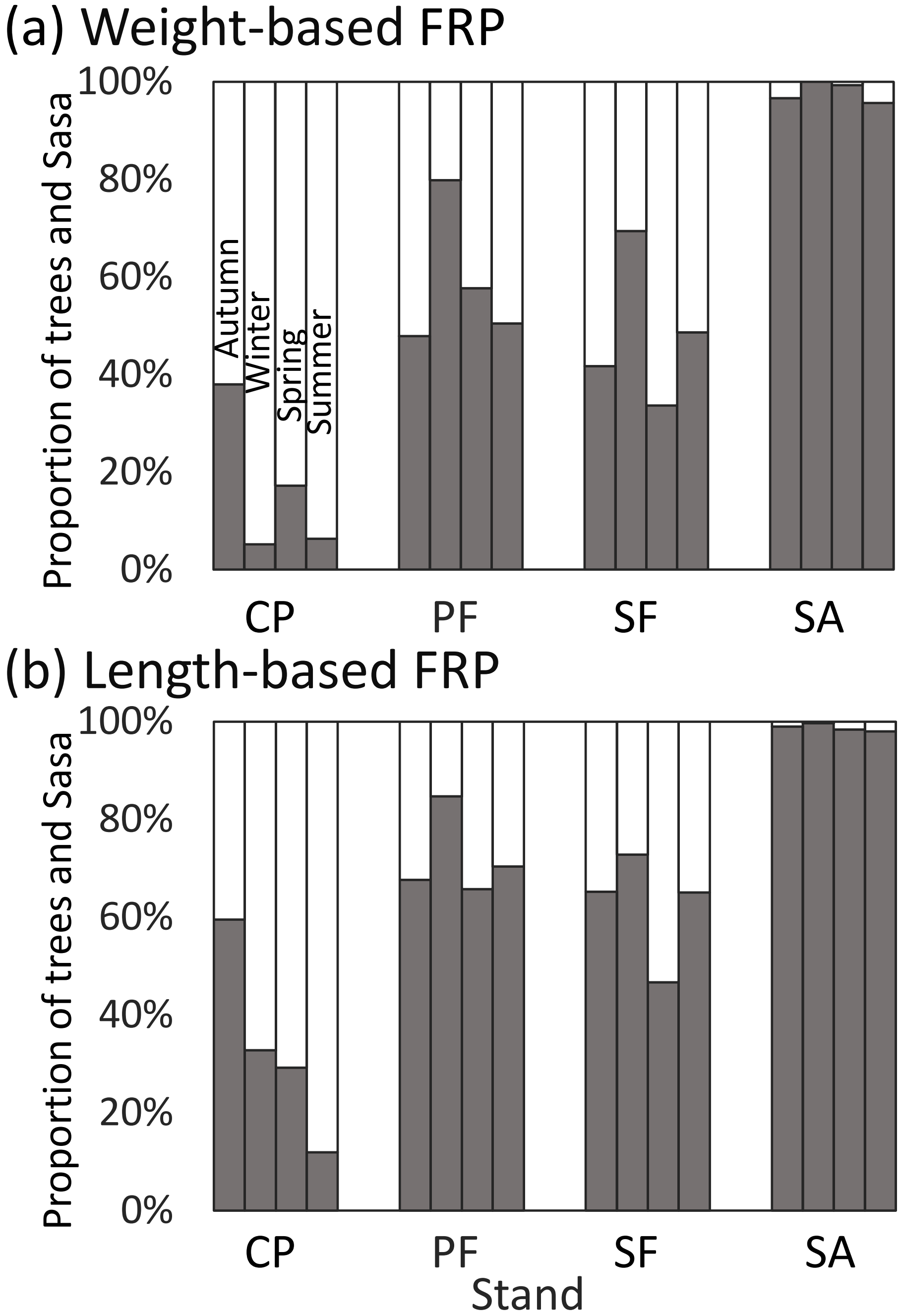


Figure S7. Proportion of trees and Sasa roots in weight-based (a) and length-based (b) fine root production (FRP) for all layers (Oe/Oa and soil). Grey, Sasa; white, trees. CP, conifer plantation; PF, primary forest; SF, secondary forest, SA, Sasa area. Autumn, 26 August–30 October 2013; Winter, 6 November 2013–14 May 2014; Spring, 21 May–1 July 2014; Summer, 2 July–16 September 2014.

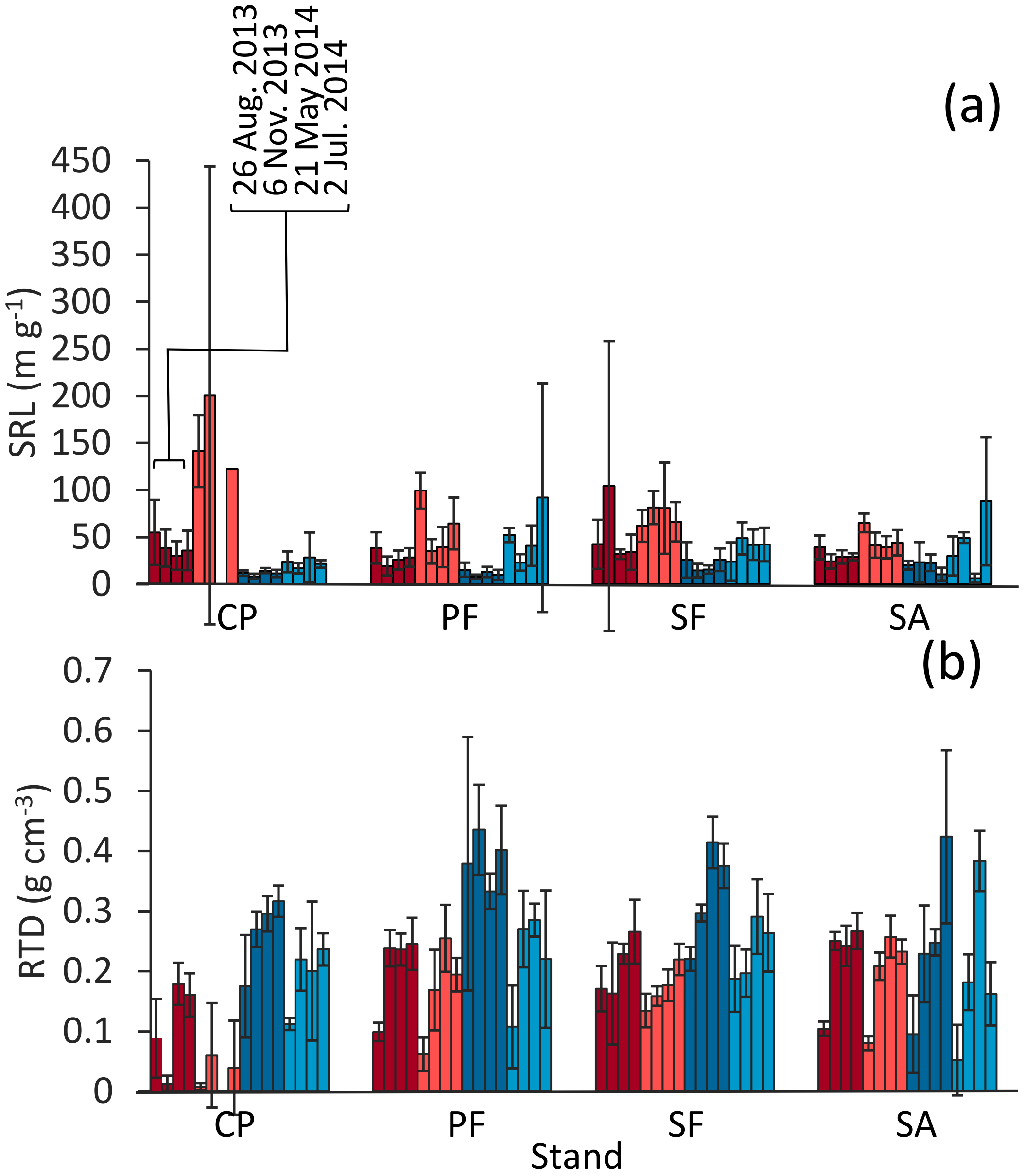


Figure S8. Root traits of trees and Sasa in soil and Oe/Oa layers in four seasons. Specific root length (SRL) (a) and root tissue density (RTD) (b). Error bars denote SD (*n* = 5). CP, conifer plantation; PF, primary forest stand; SF, secondary forest stand; SA, Sasa area.

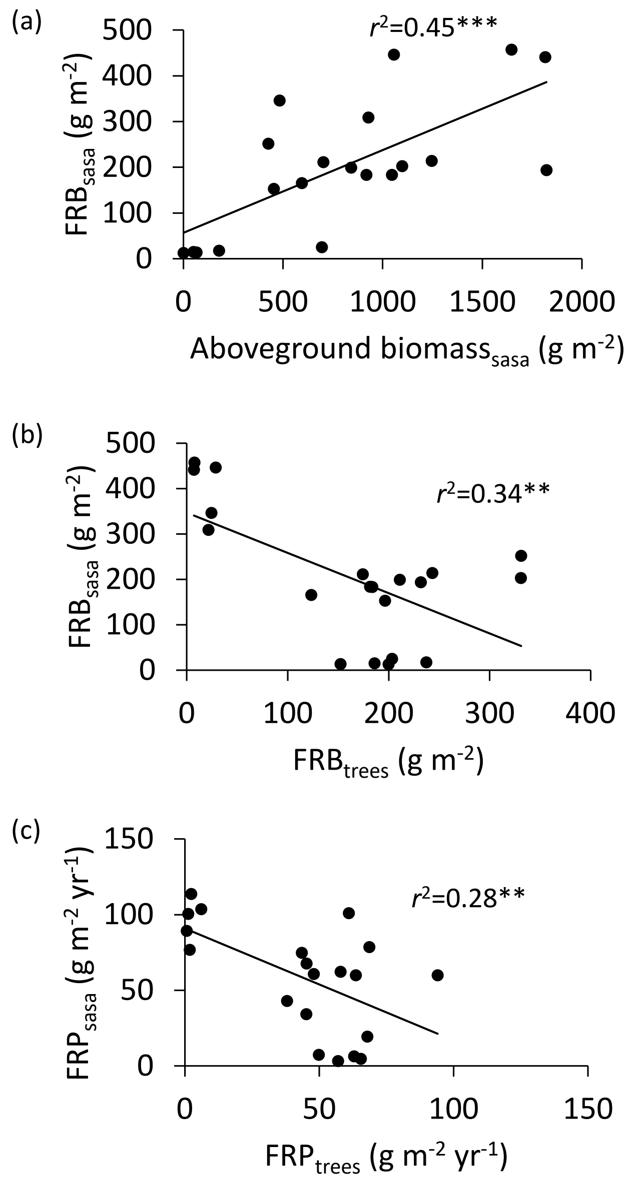


Figure S9. Relationships between Sasa aboveground biomass and Sasa fine root biomass (FRB$_{sasa}$) (a), between tree fine root biomass (FRB$_{trees}$) and FRB$_{sasa}$ (b), and between tree fine root production (FRP$_{trees}$) and Sasa fine root production (FRP$_{sasa}$) (c). ***$P < 0.001$; **$P < 0.01$.

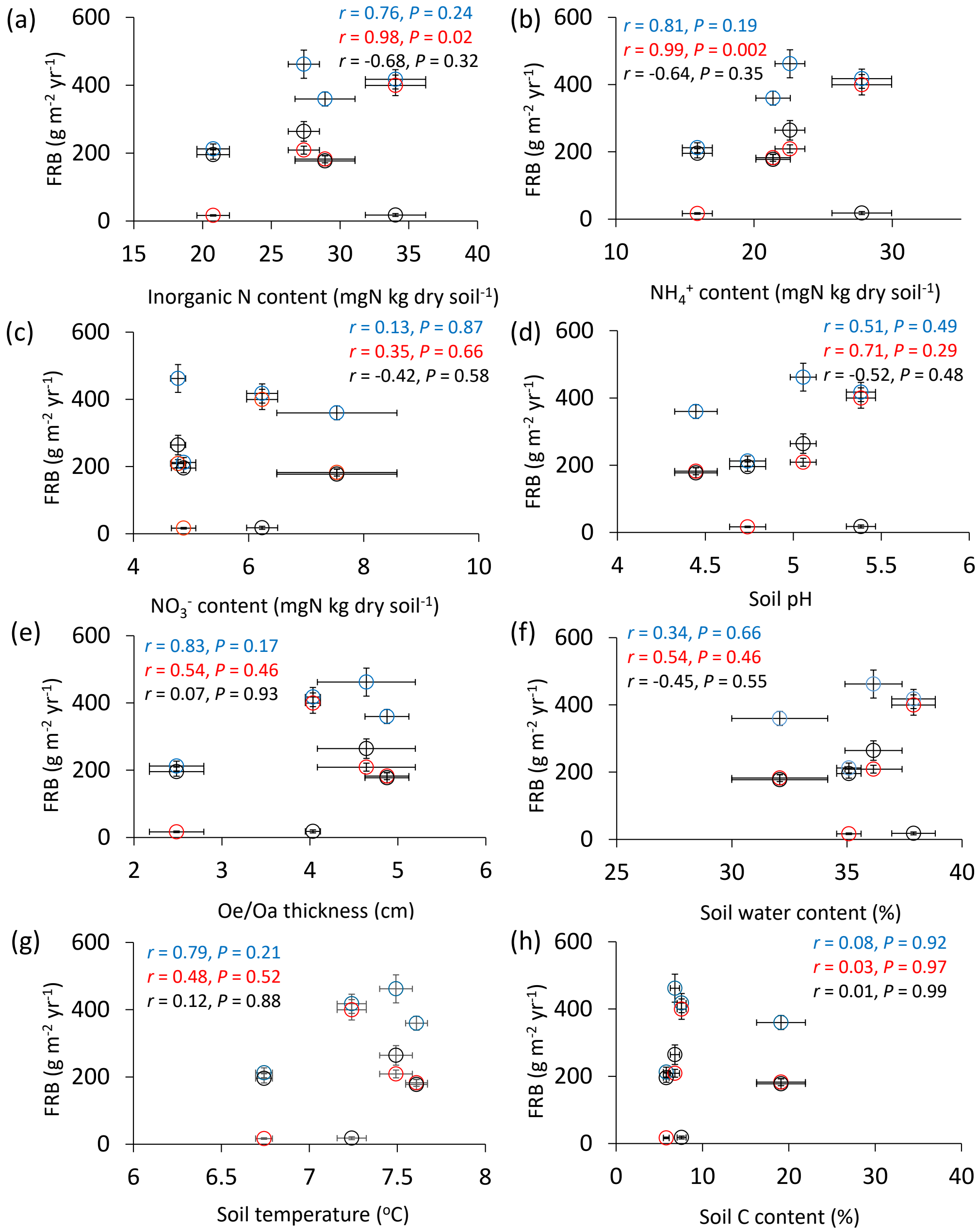


Figure S10. Relationships between site mean fine root biomass (FRB) and production (FRP) and site mean soil environmental factors: soil inorganic N content (a, k), soil ammonium content (b, l), soil nitrate content (c, m), soil pH (d, n), Oe/Oa layer thickness (e, o), soil water content (f, p), soil temperature (g, q), soil C content (h, r), soil N content (i, s), and soil C/N (j, t). Trees, black circles; Sasa, red circles; total, blue circles. Vertical and horizontal bars denote standard errors (n = 5). The r values and P values from correlation analysis for trees, Sasa, and total are indicated in the corresponding shades.

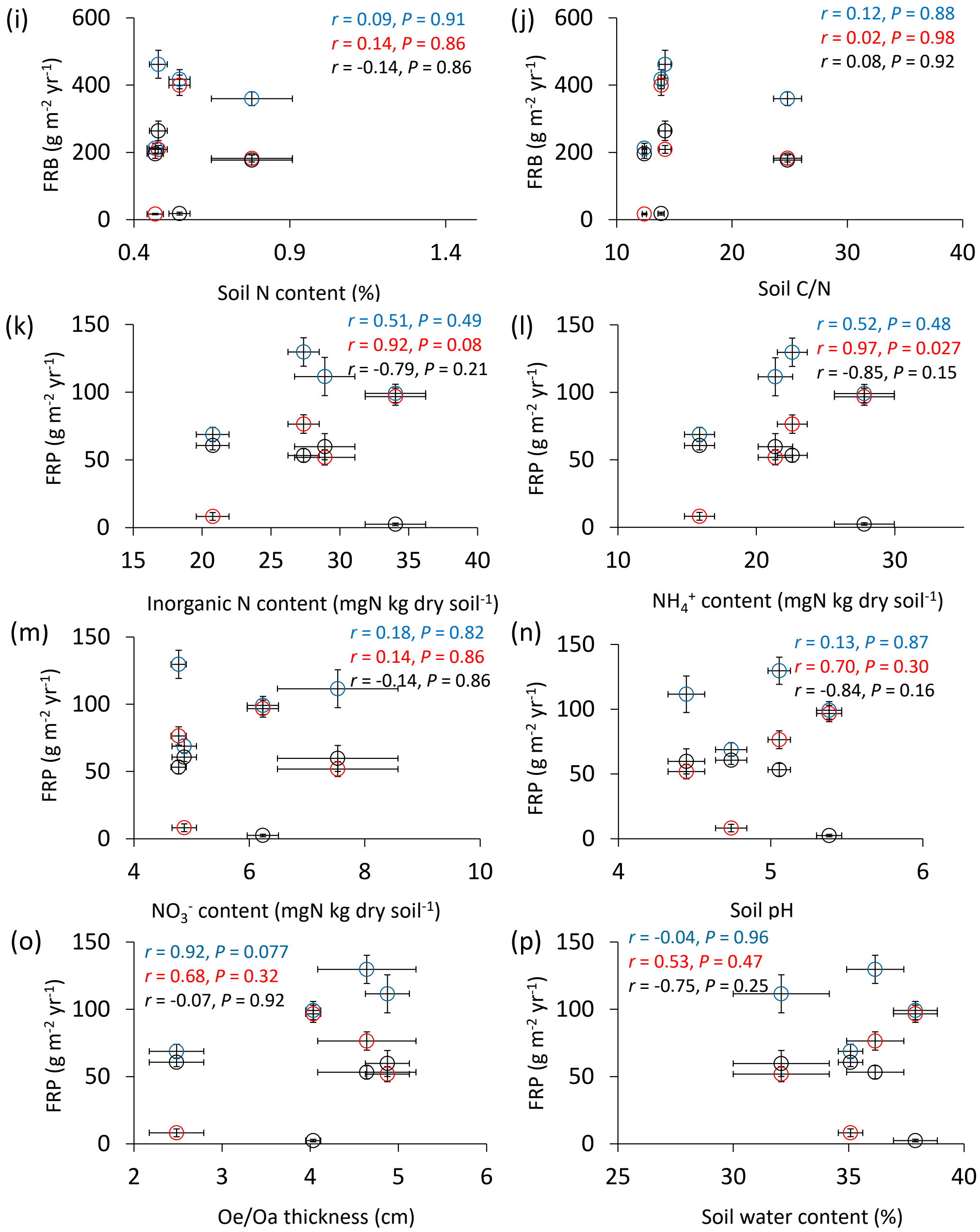


Figure S10. (Continued)

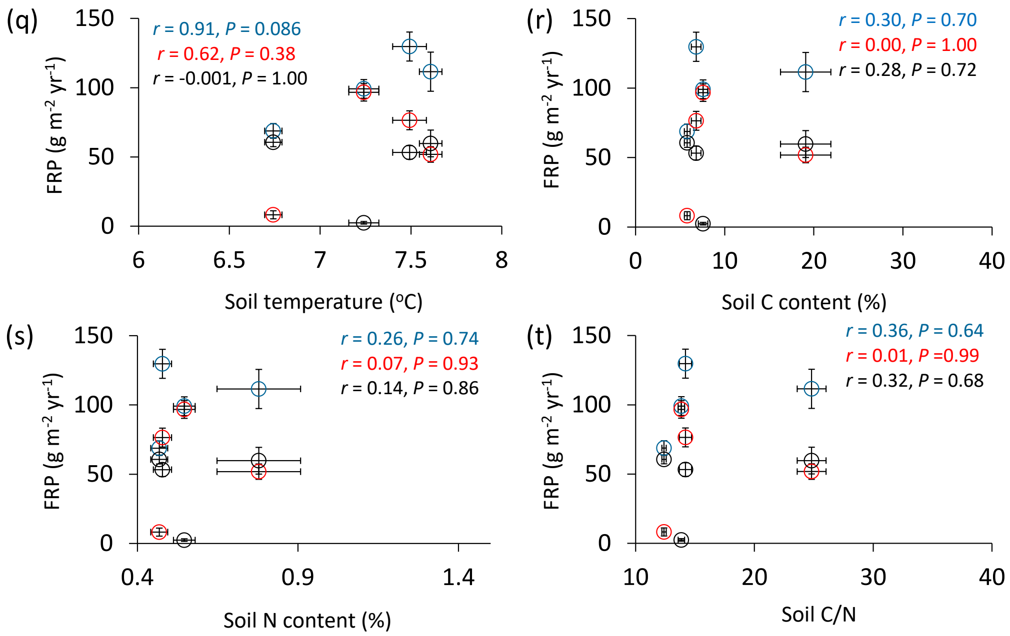


Figure S10. (Continued)

Table S1. Overview of stand characteristics.

Stand type	Abbreviation	Tree species composition	Stand history	Stand age (yrs)
Conifer plantation	CP	*Abies sachalinensis*	1929 forest fire (in part) 1975 soil scarification planting	39
Primary conifer-broadleaf mixed forest	PF	*Quercus crispula* *Magnolia obovata* *Alnus hirsuta* *Tilia japonica* *Acer pictum* *Phellodendron amurense* *Sorbus commixta* *Abies sachalinensis* *Betula ermanii* *Prunus ssiori*	1975 tree cutting (only for wind-fallen trees) 2001 selective tree cutting (in part)	>100
Secondary forest	SF	*Betula platyphylla* *Betula ermanii*	1929 forest fire 1945 forest fire	69
Sasa area	SA	*Phellodendron amurense* *Hydrangea paniculata*	2003 clear-cutting	10

Table S2. Aboveground stand characteristics and soil environmental factors in each stand.

Stand	CP	PF	SF	SA
Aboveground stand characteristics				
BA (m^2 ha^{-1})	35.2 ± 5.2 ^{a}	20.2 ± 5.3 ^{b}	28.5 ± 4.2 ^{ab}	0.39 ± 0.54 ^{c}
Tree density (trees ha^{-1})	847 ± 62 ^{a}	497 ± 58 ^{b}	745 ± 116 ^{a}	45 ± 66 ^{c}
Tree aboveground biomass (g m^{-2})	18549 ± 3584 ^{a}	12141 ± 3086 ^{b}	14551 ± 2224 ^{ab}	144 ± 199 ^{c}
Culm density of Sasa (culms m^{-2})	12 ± 11 ^{b}	48 ± 23 ^{ab}	58.4 ± 13 ^{a}	85.6 ± 41 ^{a}
Sasa aboveground biomass(g m^{-2})	440 ± 688 ^{b}	2410 ± 1218 ^{a}	1274 ± 435 ^{ab}	2344 ± 752 ^{a}
Canopy openness (%)	13.9 ± 1.0 ^{c}	26.6 ± 3.7 ^{b}	26.6 ± 1.2 ^{b}	100 ± 0 ^{a}
Soil environmental factors				
Oe/Oa layer thickness (cm)	2.5 ± 0.7 ^{b}	4.6 ± 1.2 ^{a}	4.9 ± 0.6 ^{a}	4.0 ± 0.2 ^{a}
Mean annual soil temperature (^{o}C)	6.7	7.5	7.6	7.2
Soil water content (volume) (%)	35.8 ± 1.3 ^{a}	35.0 ± 3.0 ^{a}	23.6 ± 3.8 ^{b}	37.6 ± 1.4 ^{a}
Soil water content (weight) (%)	33.6 ± 1.6 ^{a}	38.4 ± 7.0 ^{a}	49.0 ± 10.8 ^{a}	38.4 ± 4.1 ^{a}
Soil total C content (%)	5.8 ± 0.7 ^{b}	6.8 ± 1.2 ^{b}	19.1 ± 6.3 ^{a}	7.6 ± 1.2 ^{b}
Soil total N content (%)	0.47 ± 0.06 ^{a}	0.48 ± 0.06 ^{a}	0.78 ± 0.29 ^{a}	0.55 ± 0.08 ^{a}
Soil C/N	12.4 ± 0.4 ^{c}	14.2 ± 1.1 ^{bc}	24.8 ± 2.7 ^{a}	13.9 ± 0.6 ^{b}
NO_3^- (mgN kg dry $soil^{-1}$)	4.9 ± 0.5 ^{ab}	4.8 ± 0.29 ^{b}	7.5 ± 2.3 ^{a}	6.2 ± 0.6 ^{a}
NH_4^+ (mgN kg dry $soil^{-1}$)	15.9 ± 2.4 ^{c}	22.6 ± 2.4 ^{ab}	21.4 ± 2.8 ^{bc}	27.8 ± 4.8 ^{a}
Inorg-N (mgN kg dry $soil^{-1}$)	20.8 ± 2.7 ^{b}	27.4 ± 2.5 ^{ab}	28.9 ± 4.9 ^{a}	34.0 ± 4.9 ^{a}
Soil pH (H_2O)	4.7 ± 0.2 ^{bc}	5.1 ± 0.2 ^{ab}	4.5 ± 0.3 ^{c}	5.4 ± 0.2 ^{a}

Mean ± SD (n = 5). Different lowercase letters denote significant difference among stands (P < 0.05).

CP, conifer plantation; PF, primary conifer-broadleaf mixed forest; SF, secondary forest; SA, Sasa area.

BA, sum of basal tree area at breast height; NO_3^-, soil NO_3^- content; NH_4^+, soil NH_4^+ content; Inorg-N, soil inorganic N content.

Table S3. Two-way ANOVA of the effects of stand type (stand), season, and their interaction on (a) fine root biomass (FRB), (b) fine root length (FRL), (c) weight-based fine root production (FRP) and (d) length-based FRP.

Factor	*df*	*F-value*	*P-value*
(a) FRB			
stand	3	17.4	***
season	3	1.96	
stand×season	9	1.94	
(b) FRL			
stand	3	56.2	***
season	3	5.49	**
stand×season	9	1.08	
(c) Weight-based FRP			
stand	3	5.47	**
season	3	35.9	***
stand×season	9	7.23	***
(d) Length-based FRP			
stand	3	15.4	***
season	3	37.6	***
stand×season	9	12.8	***

***P < 0.001; **P < 0.01.

Table S4. Fine root turnover rate (yr^{-1}) of trees and Sasa at 0–10 cm soil depth in each stand with the results of two-way ANOVA for the effects of stand type (stand), plant form (plant), and their interaction on fine root turnover.

Stand	Trees	Sasa
CP	0.45 (0.08)	0.56 (0.51)
SF	0.62 (0.30)	0.42 (0.12)
PF	0.32 (0.08)	0.57 (0.15)
SA	0.28 (0.27)	0.35 (0.04)

Factor	*df*	*F-value*	*P-value*
stand	3	1.45	ns
plant	1	0.60	ns
stand×plant	3	1.45	ns

Mean with SD (n = 5) in parentheses. CP, conifer plantation; PF, primary forest stand; SF, secondary forest stand; SA, Sasa area. ns, not significant ($P \geq 0.05$).

Table S5. Results of three-way ANOVA for the effects of plant form (plant), soil layer (layer), stand type (stand), and their interaction on specific root length (SRL) and root tissue density (RTD).

Factor	*df*	*F-value*	*P-value*
(a) SRL			
plant	1	44.9	***
layer	1	53.3	***
stand	3	4.21	**
plant×layer	1	5.40	*
plant×stand	3	11.9	***
layer×stand	3	4.54	**
plant×layer×stand	3	8.41	***
(b) RTD			
plant	1	73.8	***
layer	1	39.0	***
stand	3	12.8	***
plant×layer	1	6.54	*
plant×stand	3	8.03	***
layer×stand	3	2.12	
plant×layer×stand	3	2.12	

***P < 0.001; **P < 0.01; *P < 0.05.

Table S6. Analysis of deviance for soil properties after GLMM for total, Sasa, and tree fine root biomass (FRB) and fine root production (FRP).

Factor	FRB_{total}	FRB_{Sasa}	FRB_{trees}		FRP_{total}		FRP_{Sasa}		FRP_{trees}	
Inorg N content	ns	ns	ns		ns		ns		ns	
NH_4^+ content	ns	ns	*	+	ns		ns		***	+
NO_3^- content	ns	ns	ns		ns		ns		ns	
Soil pH	ns	ns	ns		*	–	**	–	ns	
Oe/Oa layer thickness	ns	ns	***	+	ns		ns		ns	
Soil water content	ns	ns	ns		ns		ns		ns	
Soil C content	ns	ns	ns		**	+	ns		*	+
Soil N content	ns	ns	ns		**	+	ns		**	+
Soil C/N	ns	ns	ns		ns		ns		ns	

***P < 0.001; **P < 0.01; *P < 0.05; ns, not significant

GLMM, Generalized linear mixed model.

Estimated direction in GLMM: + positive; – negative.