

# **Decoupling of nitrogen and phosphorus release from fine and coarse roots during seven years of decomposition**

Jun Pan<sup>1,2</sup>, Rui Wang<sup>3</sup>, Craig R. See<sup>4</sup>, Junwei Luan<sup>5</sup>, Jing Wang<sup>6</sup>, Fan Liu<sup>7</sup>, Xiankui Quan<sup>1,2</sup>, Hongyang Chen<sup>1,2</sup>, Xingchang Wang<sup>1,2,8</sup>, Chuankuan Wang<sup>1,2,8</sup>

<sup>1</sup> Center for Ecological Research, Northeast Forestry University, Harbin 150040, China

<sup>2</sup> Key Laboratory of Sustainable Forest Ecosystem Management – Ministry of Education, Northeast Forestry University, Harbin 150040, China

<sup>3</sup> Key Laboratory of Biochemistry and Molecular Biology in Universities of Shandong Province, Weifang University, Weifang 261061, China

<sup>4</sup> Center for Ecosystem Science and Society, Northern Arizona University, Flagstaff, AZ, 86001, U.S.A.

<sup>5</sup> Institute of Resources and Environment, Key Laboratory of National Forestry and Grassland Administration/Beijing for Bamboo & Rattan Science and Technology, International Centre for Bamboo and Rattan, Beijing, 100102, China

<sup>6</sup> Institute of Geographic Sciences and Natural Resources Research, Chinese Academy of Sciences, Beijing 100101, China

<sup>7</sup> Key Laboratory of Agricultural Water Resources, Center for Agricultural Resources Research, Institute of Genetics and Developmental Biology, Chinese Academy of Sciences, Shijiazhuang

050021, China

<sup>8</sup> Corresponding Author. E-mail: [xewang\\_cer@nefu.edu.cn](mailto:xewang_cer@nefu.edu.cn); [wangck-cf@nefu.edu.cn](mailto:wangck-cf@nefu.edu.cn)

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Should the manuscript be accepted, the data supporting the results will be archived in an appropriate public repository (Dryad, Figshare or Hal) and the data DOI will be included at the end of the article.

### **Authors contributions**

XW designed the experiment, JP and XW collected data, JP, FL, and XW performed data analyses.

JP wrote the first draft of the manuscript, and all authors contributed substantially to revisions.

## **Abstract**

Belowground litter decomposition represents an important source of the limiting nutrients nitrogen (N) and phosphorus (P) to forest soils. However, the rates and drivers of nutrient immobilization and release from root litter remain poorly understood. To address this gap, we conducted a seven-year field decomposition experiment using roots from three species, across five diameter classes (up to 20 mm) in a temperate forest. All roots released P to the surrounding soil within the first year of decomposition but immobilized N for much longer, particularly coarse roots. Long-term N release was mainly related to initial nutrient stoichiometry of the substrate, whereas P release was better predicted by substrate carbon (C) quality and decomposition rate. Initial root nutrients well predicted the difference between long-term N versus P release. Our results highlight the fact that N and P dynamics should be considered separately when modeling nutrient release during root decomposition.

## **KEYWORDS**

Root decomposition; Root diameter; Nutrient release; Nutrient immobilization; Initial root quality; Nitrogen; Phosphorus

## **INTRODUCTION**

Root litter decomposition represents a critical entry point of carbon (C), nitrogen (N), and phosphorus (P) into soil systems (e.g., Li et al. 2015; Sariyildiz 2015), but the long-term dynamics of

these processes remain poorly understood relative to leaf litter decomposition (Freschet et al. 2013; Keller et al. 2021). The traits controlling rates of mass loss and nutrient release of belowground tissues do not necessarily mirror those of aboveground tissues (Sun et al. 2018), even within species (Hobbie et al. 2010; See et al. 2019). Critically, the duration of most root decomposition studies is less than 3 years (diameter < 2 mm, See et al. 2019), which is shorter than the half-life of root litter in most systems (Parton et al. 2007). Furthermore, long-term studies of decay dynamics have focused exclusively on fine roots (diameter < 2 mm), but coarser roots constitute a larger biomass pool (Fahey et al. 2005), decompose more slowly (Zhang & Wang 2015), and differ from fine roots in both C chemistry and N and P concentrations. While fine roots are often a thought to be net source of nutrients to soil during the early stages of decomposition, estimates vary considerably across studies (e.g., Li et al. 2015; Zhang & Wang 2015). Thus, the controls over long-term N and P release from decomposing roots and their relationship to root diameter remain highly uncertain.

Ecosystem models often assume that long-term nutrient release from decomposing roots is proportional to mass (C) loss rates (e.g., Sulman et al. 2017), but empirical evidence for this is scarce. The best support comes from a 10-year global litterbag study in which N release was well correlated with mass loss in the fine roots of three species (Parton et al. 2007). However, long-term rates of N release from coarse roots have not been estimated using the litterbag approach, and studies of excavated roots from logging chronosequences suggest that N release from woody roots may be

decoupled from mass loss during the early stages of decay, and remain net N sinks for decades (Fahey et al. 1991; Chen et al. 2001). To our knowledge there have been no litterbag studies that have directly measured late-stage P dynamics from decomposing roots of any size. A better understanding of the relationship between long-term rates of mass loss, N release, and P release across root diameters is therefore necessary to further both conceptual understanding and modeling of forest nutrient cycles.

Previous research on the early stages of root decomposition suggest that the relationship between diameter class and nutrient release differs for N and P. Nitrogen release generally decreases with increasing root diameter (Goebel et al. 2011; Zhuang et al. 2018), while P release appears to be more consistent (Jing et al. 2019; Pang et al. 2022), but may be higher during the early stages of decay (Gang et al. 2019). The mechanism for this variation in nutrient dynamics with diameter size is due in large part to differences in substrate chemistry. Nutrient release tends to increase with the initial substrate concentration (Chen et al. 2002; Cusack et al. 2009; Hobbie et al. 2010) because microbial communities immobilize limiting nutrients until they have met their stoichiometric requirements. Consequently, the decomposition of higher C:N substrates can sometimes require additional import of N by the decomposer community, leading to a net immobilization of N in the decomposing root which can last for years (Parton et al. 2007). Thus, the controls over nutrient immobilization and release throughout the decomposition process are likely driven by changing

tradeoffs between the energy and nutrient demands of the decomposer community (Gill et al. 2021).

Root C chemistry is an important controlling factor of root decomposition (Berg et al. 2000; Silver & Miya 2001; Zhang et al. 2008; Fujii & Takeda 2010; Bakker et al. 2011; Jiang et al. 2021; Song et al. 2021), and its effects can be broadly categorized into those of structural and non-structural components. Structural C makes up the majority of plant C and it's dominant form influences decomposition rates, with carbohydrates degraded hydrolytically (i.e., cellulose, hemicellulose) decomposing faster than those requiring less energetically unfavorable oxidative processes. As a result, initial lignin (an acid unhydrolyzable compound) commonly predicts the rate of decomposition worldwide (Zhang et al. 2008; Harmon et al. 2009; See et al. 2019; Guo et al. 2021). Non-structural C fractions such as phenolics (Sun et al. 2018), non-structural carbohydrates (NSC), and other secondary compounds also influence decomposition rate in a variety of ways, but their effects on nutrient release are less understood. A comprehensive understanding of how substrate C composition affects the long-term trajectory of mass loss and nutrient release from root litter is still lacking.

In addition to C chemistry, nutrient limitation of decomposer communities plays an important and complex role in determining decomposition rate and nutrient retention (Zhou & Wang 2016). Low substrate N concentrations can suppress microbial activity, reducing decomposition rate (Parton et al. 2007; Hobbie et al. 2010; Goebel et al. 2011; See et al. 2019). Conversely, N limitation can

enhance the decomposition of more recalcitrant substrates (i.e., N-mining), and alleviation of N limitation can inhibit ligninase activity and promote the accumulation of organic matter (Hobbie 2015; Wang et al. 2019; Yang et al. 2019). Higher substrate N and P concentrations consistently lead faster decay rates during the early-stages of fine root decomposition worldwide (See et al. 2019; Jiang et al. 2021), but effects on later stages of decomposition and nutrient release remain largely unexplored. In addition, there is increasing recognition for the role of other rock-derived nutrients in regulating litter decomposition, including magnesium (Mg; Goebel et al. 2011; Wambsganss et al. 2022), calcium (Ca; See et al. 2019), and silicon (Si; Schaller & Struyf 2013). A better understanding of how these less-reported elements interact to affect long-term rates of decomposition and nutrient release is therefore needed.

Here we present the results of a seven-year decomposition experiment conducted across roots of five diameter classes, from three temperate tree species in Northeast China. We explore patterns of short-term and long-term rates of decomposition and nutrient release, and identify the initial substrate characteristics that best predict these rates. We hypothesized that: (1) Nutrient release (as a percent of the initial pool) would be higher from lower diameter roots, (2) Decomposition rate and nutrient release would be higher from species with higher initial concentrations, and (3) The decomposition rate would predict nutrient release during both short-term and long-term stages.

## **MATERIALS AND METHODS**

**Site description**

This study was conducted at the Maoershan Forest Ecosystem Research Station in northeastern China (N45°25', E127°40'). The climate is continental monsoonal, with a warm-humid summer and a dry-cold winter. The mean annual precipitation is 726 mm, and the mean annual temperature is 2.1 °C during the period of 2008-2019 (Sun et al. 2021). The forest is a 70-yr old broadleaved deciduous forest, with a mean elevation of 400 m and a mean slope of 9°. The soils are classified as Alfisols (Eutroboralfs) based on the United States Soil Taxonomy.

**Field experiments**

Three dominant tree species from the site, *Betula platyphylla* (BP), *Ulmus davidiana* var. *japonica* (UJ), and *Fraxinus mandshurica* (FM), were selected to study the effects of initial root quality on decomposition of roots with different diameters. Roots from three to five mature individuals of each species were excavated in late May 2012 from the 0-20 cm soil layer, and immediately taken back to the lab for processing. The roots were cleaned with tap water, dead roots were removed, and then divided into five diameter classes: < 1 mm, 1-2 mm, 2-5 mm, 5-10 mm, 10-20 mm. A sample of 2-5 g air-dried roots was placed into each 0.1 mm mesh litter-bag, and masses were later corrected to oven-dried (65 °C) mass using species- and diameter-specific correction factors. There were 720 bags in total: 60 bags for each of the < 1 mm, 1-2 mm, and 2-5 mm diameter classes, and 30 bags each of the 5-10 mm and 10-20 mm diameter classes in a species. Five individual trees were



repeated for each species. Litter-bags were buried on June 3rd 2012 to 10 cm depth underneath five replicate trees of each species, with species corresponding to the species of root contained in the litterbag (Lin et al. 2020). After burying the bags, the litter layer was restored as much as possible to minimize disturbance.

Litterbags were retrieved on eight occasions during the first two years of incubation to effectively capture dynamics during the early stages of decomposition. To characterize later stage dynamics, we retrieved one set of litterbags each in the 3rd, 5th and 7th years of incubation. This resulted in a total of 11 litterbag collections, representing incubation times of 1, 2, 3, 4, 5, 11, 13, 15, 27, 49, and 87 months. In total, 666 of 720 litter bags were retrieved, and the rest were lost or destroyed by wild boar (*Sus scrofa* L.). Upon removal from the field, all roots were cleaned, oven-dried it at 65 °C, massed, and analyzed for C, N and P concentrations.

**Root chemistry measurements**

Additional root samples of each species and diameter class were collected on May 29st, 2021 to measure the initial root quality. We characterized each sample for concentrations of C, N, P, Ca, Mg, Si, soluble sugars, starch, hemicellulose, cellulose, lignin, and total phenolics. The total C concentration was determined by the dry combustion method (Multi N/C 2100 S, Analytik Jena, Jena, Germany). The N and P concentrations were determined by sulphuric acid/hydrogen peroxide digestion, followed by sodium salicylate/sodium hypochlorite colorimetric determination for N and

by ammonium molybdate/ascorbic acid colorimetric determination for P using continuous flow analysis (AA3, SEAL Analytical, Germany). Ca and Mg concentrations were determined by flame spectrophotometer with an Atomic Absorption Spectrophotometer (TAS-990, Pgeneral, China), Si concentration was obtained by mass method after digesting with nitric acid/perchloric acid. Soluble sugars, starch and non-structural carbohydrates (NSC, sum of soluble sugars and starch) concentrations were determined by the modified phenol sulfuric acid method (Zhang et al. 2014). Hemicellulose, cellulose, and lignin concentrations were determined by high pressure liquid chromatography (UltiMate 3000, Thermo, USA). Total phenolics concentration was determined by the Folin phenol colorimetry. Two or three repetitions were carried out in each sample to reduce the operating error. For the C quality, structural carbohydrates indicated cellulose and lignin, decomposable carbohydrates were NSC and hemicellulose, resistant carbohydrates represented cellulose, lignin, and total phenolics.

### **Data analysis**

The Mauchly's Test of Sphericity showed that there was a significant temporal autocorrelation of mass and nutrient remaining rates, thus the two-factor repeated measurement analysis of variance of tree species and diameter and their interaction was used to test the effect of decomposition time, diameter, tree species and their interaction on nutrient remaining. Duncan post-hoc analysis was used to test significant differences in decomposition rate, nutrient release, and initial root quality

between root diameter within species, between species for the same root diameter.

We fit the proportion of mass remaining to three exponential decay models and determined the best fit among them both using Akaike Information Criterion (AIC) and considering the biological meanings of decomposition rates.

Single exponential model (Olson 1963):

$$M = e^{-kt} \quad (1)$$

Double exponential model (Hobbie et al. 2012):

$$M = Ce^{-k_1t} + (1 - C)e^{-k_2t} \quad (2)$$

Asymptotic model (Hobbie et al. 2012):

$$M = A + (1 - A)e^{-k_at} \quad (3)$$

where  $M$  is the proportion of initial mass remaining at time  $t$  (in years), and  $k$  is the decomposition rate in single exponential model. In the double exponential model,  $C$  is the fraction of the initial mass that decomposes with decomposition rate  $k_1$ , while the remaining fraction  $(1 - C)$  decomposes with rate  $k_2$ . In the asymptotic model,  $A$  is the fraction of the initial mass with a decomposition rate of zero (i.e., the asymptote), while the remaining fraction  $(1 - A)$  decomposes with rate  $k_a$ .

While the two pool models generally fit better based on AIC, we ultimately chose the single exponential model for comparisons with nutrient release, because the  $k_1$  based on the double exponential model and  $k$  from the asymptotic model were too large (Table S1). We thus calculated

separate single exponential  $k$  values to represent the short-term (0 – 15 months) and long-term (i.e., 87-months' decomposition) decomposition stages (Pan et al. 2021). For each of these time periods, we calculated rates of nutrient release, where the nutrient remaining ( $L$ , %) was calculated as:

$$L = M_t C_t / M_0 C_0 \times 100\% \quad (4)$$

and the nutrient release ( $L_t$ , %) was calculated as:

$$L_t = 1 - L \quad (5)$$

where  $M_t$  is residual dry mass at time  $t$  (yr),  $M_0$  is initial dry mass,  $C_t$  is the nutrient concentration at time  $t$ , and  $C_0$  is the initial nutrient concentration. The root nutrient release is the mass of nutrient lost by the roots over a time period compared with the initial nutrient mass of the roots (Parton et al. 2007; Xiong et al. 2012; Freschet et al. 2021).

The similarities and differences in root decomposition rate and nutrient release were analyzed using principal component analysis and linear regression. Two-way ANOVA was used to determine whether the initial root quality was significantly influenced by tree species and diameter. Multi-methods were used to analyze the relationship between initial root quality and root decomposition parameters at different angles. Pearson correlation analysis was used to detect the relationship between initial root quality, decomposition rate, and nutrient release for the two stages of decomposition. We used linear regression with forward selection (based on the significance of the partial regression sum of squares) to find the best-fitting models for predicting root decomposition

rate and nutrient release based on initial root quality parameters. The regression procedures were conducted using the “relaimpo” package in R (Groemping 2006).

## RESULTS

### Short-term and long-term patterns of nutrient release

Nutrient release varied considerably across root diameter and tree species (Figures 1, 2, S1, and S2). Short-term N and P release systematically decreased with increasing root diameter (with the exception of P release from BP), while C release generally increased. Nitrogen was immobilized during the early stages of decomposition across all species and diameters, except in the smallest diameter class ( $< 1$  mm) of roots (Figure 1c). In contrast, C and P were generally released during this time (Figure 1b, d). Patterns in decomposition and nutrient release also varied by species and differed by element. Short-term C release was significantly higher for FM than the other two species in coarse roots (Figure 1b), and N release was also highest from fine roots of this species (Figure 1c). In contrast, short-term P release was generally highest from UJ in fine roots, while this species generally had the lowest rates of N release.

After seven years of decomposition, we observed a wide range in net source-sink dynamics across species and diameter classes, with a much wider range observed for N than P. In general, long-term decomposition rate, C loss, and P release in coarse roots was higher than in fine roots. Nitrogen release roughly decreased with root diameter and varied strongly among species. Most

diameter classes released N during the later stages of decomposition, but the coarsest roots of BP and UJ still immobilized N at the end of the experiment. The greatest N release was observed in < 1 mm FM roots (40.1% of initial N), while the coarsest roots of UJ had still immobilized 25.3% of initial N at the end of the experiment. In contrast, all roots released P, but net release still varied by a factor of three, with coarse roots of FM (5-10 mm diameter) releasing 69.6% of initial P, and UJ releasing 15.4% of initial P. Long-term nutrient release of FM was generally higher than that of the other two species (Figure 2).

Principal component analysis showed that short-term N and P release diverged from long-term decomposition rate (Figures 3 and S3). In general, long-term C and P release were similar to long-term decomposition rate (Figure 3), and C and P release were significantly positively correlated with long-term decomposition rate (Figure S3). Notably, rates of long-term N release were unrelated to mass loss, C loss, and P release.

### **Chemical drivers of short-term and long-term nutrient release**

Our analysis of initial substrate chemistry revealed that the strongest predictors of short-term decomposition rates and C losses differed from those driving nutrient release (Figures 4 and S4). Short-term C release, P release, and decomposition rate were largely driven by C fractions and structural components (i.e. Si), while short-term N release was directly related to N, Mg, and total C concentrations. The initial substrate effects on long-term root decomposition and C losses largely

mirrored those predicting short-term rates (Figures 5, S4, and S5). In contrast, none of the predictors of short-term N and P release remained predictors in the long-term model. Surprisingly, there were no significant relationships between long-term root decomposition rate and lignin. Higher initial N and P concentrations increased N but not C and P release.

The contributions of initial root quality to decomposition rate and nutrient release diverged during short-term and long-term stages (Figure 4, Table S5). Based on the multiple linear regression, initial root quality explained more than 80% of the variation in short-term root decomposition parameters. The major contributors of short-term decomposition rate, C release, and P release, were C quality and their ratios, but the primary predictors of N release generally nutrient concentrations. Surprisingly, initial Si concentrations predicted 54% of the variability in short-term P release. The major contributors of long-term decomposition rate, C and P releases were C quality and their ratios, while variability in N release was mainly driven by initial nutrient concentrations (Figure 4). For example, the contributions of soluble sugars:lignin to long-term P release and decomposition rate explanations were up to 60% and 56%, respectively. The top predictors of long-term C and N release were cellulose:phenolics (46% of variability) and P (29% of variability), respectively.

Sensitivities of decomposition during the short-term and long-term to the first two major contribution factors were shown in Figures S6 and S7. Short-term root decomposition parameters (except short-term N release) were consistently more sensitive to and increased with increasing the

first principal factor, and decreased with the increase of the second principal factor. Short-term N release was positively correlated with the initial concentration of C and N. However, long-term decomposition rate and long-term N release increased with increasing the first principal factor (soluble sugars:lignin and P, respectively), and decreased with increasing the second principal factor (phenolics and Ca, respectively), and were more sensitive to the first principal factor. Long-term C release increased with increasing cellulose:phenolics (more sensitive) and NSC:phenolics, and P release increased as soluble sugars:lignin and C:NSC (more sensitive) increase, respectively.

The relative difference in N release and P release at the end of the study was well related to initial and final nutrient concentrations (Figure 5). Initial N and N:P were good single predictors of the difference from N release and P release during long-term stage. And the difference between long-term N and P release well predicted the final N:P in root litter with a linear model. The multiple linear regression revealed that initial C:N, N:P and resistant carbohydrates concentration together explained 89% of the variation (Figure S8). However, the difference between N and P release during short-term stage was exclusively related to structural carbohydrates ( $R^2 = 0.39$ , Figure S9).

## **DISCUSSION**

### **Nitrogen and phosphorus dynamics during root decomposition**

Our results represent the most detailed study of the long-term N and P dynamics in decomposing roots to date, and highlight the fact that N release is decoupled from P release during all stages of



decomposition. These findings have clear implications for forest nutrient cycling, as root necromass represents a significant nutrient pool in soils, and soil N and P availability commonly co-limit productivity in terrestrial ecosystems (Elser et al. 2007; Harpole et al. 2011). Importantly, we found opposite relationships between root diameter and nutrient retention for these two elements, with greater proportional P release from coarse roots (diameter  $\geq 2$  mm), and greater proportional N release from fine roots (diameter  $\leq 2$  mm). Furthermore, our data strongly suggest that while long-term rates of P release during decomposition can be predicted as a function of mass loss, N release cannot (Figure S3), challenging the assumption that these fluxes are linearly related to decomposition (Parton et al. 2007; Cusack et al. 2009). These contrasting long-term dynamics are not explicitly represented in ecosystem models but are likely important, because the ability of ecosystems to redistribute stocks of N and P in synchrony between above and belowground pools in response to plant demand will be critical to determining future C sequestration under global change (e.g., Rastetter et al. 2022).

Short-term patterns of N and P release were correlated with mass loss in our dataset, consistent with previous work, but the magnitude of these fluxes varied by species and root size. Rates of N release decreased with root diameter and varied widely among species (Usman et al. 2000; Lin et al. 2010; Jani et al. 2015; Xu et al. 2022). Only the finest diameter ( $< 1$  mm) roots immediately released N during the first 15 months of decomposition (Figure 1; Parton et al. 2007; Sun et al. 2013).

Conversely, while short-term rates of P release also differed across diameter and species, there was considerably less variation, and all roots were net source of P to soil within the first 15 months (with the exception of the coarsest FM roots), suggesting that root necromass may be a more important long-term sink for N than P in soils. Similar to the relationships observed across diameters, N release also varied more across species than P, though we also found (smaller) interspecific differences in P release (between UJ and FM) that corroborate a previous study of these species (Zhuang et al. 2018). These interspecific differences support the idea that fine scale variation in rhizosphere species composition may have considerable afterlife effects on localized N and P availability (Tong et al. 2012; Freschet et al. 2013), ultimately leading to plant-driven feedbacks on rates of stand-scale nutrient cycling (Hobbie 2015). The fact that species differed in their relative rates of N and P release warrants further investigation (Figure S10). If within-species N:P stoichiometry of nutrient release is decoupled from with the N:P stoichiometry of annual plant demand, it could provide a previously overlooked reinforcing mechanism for maintaining community-level N and P colimitation (Bloom 1985).

**Effects of initial substrate quality on nitrogen, phosphorus, and carbon release**

Our results strongly suggest that differences in initial C chemistry and nutrient stoichiometry were the underlying mechanism for the observed differences in nutrient release among species and diameter class (Figures S11 – S16). Critically, the main drivers of nutrient release differed between

the early and late stages of decay. Initial concentrations of N and P predicted the release of these elements reasonably well during the first 15 months of decomposition, but P release was better predicted by initial Si and C chemistry than by initial P (Figure 4). Moreover, initial P concentrations were completely unrelated to the rate of P released at the end of the study (Figure S5), which was instead a function of initial C chemistry (i.e., the ratio of soluble sugars to lignin, and initial non-structural carbohydrates; Figure 4). Similarly, although long-term rates of N release were correlated with initial N, the best model for predicting N release did not include initial substrate N. Instead, N retention was best explained by initial concentrations of the other nutrients (P, Mg, Ca), and labile C fractions. Given that most roots had still retained over half of initial N and P after seven years, our results suggest that N and P cycles cannot be considered in isolation from other elements.

Our findings clearly demonstrate that rates of N and P release during root decomposition are not directly proportional, and vary widely across species and diameters. Critically however, long-term dynamics appear to be relatively predictable based on initial substrate chemistry. The differences in long-term N and P release were well predicted by initial C:N:P stoichiometry (Figures 5 and S8). In contrast, short-term differences were less predictable, but were correlated with initial structural carbohydrates (Figure S9). The effect of initial N (Figure 5) was overshadowed by the indirect effect of initial P and C quality during long-term decay. Taken together, our results indicate that the effects of root litter inputs on soil N and P availability could potentially be modeled based on

initial root traits, but further research is needed.

While our results are ultimately correlational, they strongly suggest multiple element control over long-term nutrient release, likely due to a combination of decomposer nutrient demands and nutrient-related stabilization of the decomposing substrates. The N immobilization we observed in most roots may be explained by root N concentrations being lower than the N concentration of the surrounding soil (6.3 vs 7.6 mg/g), facilitating the transfer of N from soil into the decomposing substrate. Previous nutrient manipulations (Zhou & Wang 2016) and foliar dynamics (Wang et al. 2022) suggest that our site it is not strongly N limited. Intriguingly, P was the most important initial nutrient controlling the long-term N immobilization, perhaps suggesting that the rate of N investment by the decomposer community may be in part controlled by P demand. This is further supported by the strong negative relationship between root N:P and C losses during the early stages of decomposition (Figure S4). In contrast, Ca had a significant inhibitory effect on N release, which might be caused by Ca stabilization of the cell wall in cortex parenchyma cells, which tend to have high N concentrations (Fujii & Takeda 2010). This may be a more likely explanation than Ca limitation, as Ca is not thought to be limiting to decomposer communities in this region (Gao et al. 2022). Mechanisms for the relationship between Si and P release are less understood, but Si enables plants to mitigate P stress (Tombour et al. 2020), so may reflect the correlations between initial substrate P and Si.

Beyond nutrient release, this experiment also represents one of the longest-running studies of C losses across root diameters and species during root decomposition. Our findings support the emerging idea that lignin alone is not sufficient to predict long-term C dynamics, and the effects of lignin have been previously shown to weaken over time (Figures. 4, S4 and S5; Harmon et al. 2009), and with increasing diameter class (Zhang & Wang 2015). Our results highlight the importance of negative effects of phenolics (Sun et al. 2018) in combination with the positive effects of more labile C fractions (i.e., soluble sugars, NSC, cellulose; Harmon et al. 2009; Fan & Guo 2010; Sun et al. 2013; Wang et al. 2019) in controlling the long-term trajectory of C losses from root litter. Nevertheless, the greater C losses from coarser roots were likely due in part to lower acid-insoluble compounds (mainly composed of lignin; Figure S12e; Harmon et al. 2009; Fan & Guo 2010; Sun et al. 2013; Cotrufo et al. 2015; Wang et al. 2019). However, coarser roots also had much lower concentrations of phenolics, which emerged as a more important control over C losses with time (Figure 4), suggesting that C lost during the early stages was primarily composed of relatively labile materials. Importantly, the variation in initial lignin concentrations across species was considerably lower than the variation in phenolics and labile C fractions (Figures S11 and S12), likely driving the patterns we observed. A better understanding of this variability will be critical to elucidating the role of root traits in long-term soil C sequestration, as more than half of initial root C remained at the end of the study.

Beyond influencing the residence time of C retention in decomposing litter pools, substrate chemistry differences across species and diameter classes likely impact the formation of new stabilized C in surrounding soil. Recent evidence suggests that arbuscular mycorrhizal roots decompose faster than ectomycorrhizal roots (See et al. 2019), and these more labile litter inputs lead may result in more efficient C transfer to mineral associated soil pools with longer residence time (Cotrufo et al. 2015; Craig et al. 2022). Our long-term C loss patterns support the idea that arbuscular mycorrhiza species (FM) lose C more rapidly than ectotrophic mycorrhiza species (UJ, BP), and suggests that these differences amplify with time, and with increasing diameter size (Figures 1 and 2). Although the proportion of soil minerals in direct contact with decomposing root litter is likely lower for coarse roots than fine roots (due to a smaller lower surface area to volume ratio), the redistribution C from litter into mineral soils via fungal hyphae is common (Frey et al. 2003; See et al. 2022). Thus, relatively low concentrations of lignins and phenolics and relatively high concentrations of carbohydrates in coarse roots likely represents a labile source of C for microbial transfer to mineral-associated C pools.

#### **DECLARATION OF COMPETING INTEREST**

The authors declare that they have no conflicts of interest.

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## References

- Bakker, M.A., Carreño-Rocabado, G. & Poorter, L. (2011). Leaf economics traits predict litter decomposition of tropical plants and differ among land use types. *Functional Ecology*, 25, 473–483.
- Berg, B., Johansson, M.-B. & Meentemeyer, V. (2000). Litter decomposition in a transect of Norway spruce forests: substrate quality and climate control. *Canadian Journal of Forest Research*, 30, 1136–1147.
- Bloom, A. (1985). Resource Limitation in Plants--An Economic Analogy. *Annual Review of Ecology and Systematics*, 16, 363–392.
- Chen, H., Harmon, M.E. & Griffiths, R.P. (2001). Decomposition and nitrogen release from decomposing woody roots in coniferous forests of the Pacific Northwest: a chronosequence approach. *Canadian Journal of Forest Research*, 31, 246–260.
- Chen, H., Harmon, M.E., Sexton, J. & Fasth, B. (2002). Fine root decomposition and N dynamics in coniferous forests of the Pacific Northwest of USA. *Canadian Journal of Forest Research*, 32, 320–331.
- Cotrufo, M.F., Soong, J.L., Horton, A.J., Campbell, E.E., Haddix, Michelle L., Wall, D.H. et al. (2015). Formation of soil organic matter via biochemical and physical pathways of litter mass loss. *Nature Geoscience*, 8, 776–779.



381 Craig, M.E., Geyer, K.M., Beidler, K.V., Brzostek, E.R., Frey, S.D., Grandy, A.S. et al. (2022).  
382 Fast-decaying plant litter enhances soil carbon in temperate forests but not through microbial  
383 physiological traits. *Nature Communications*, 13, 1229.

384 Cusack, D.F., Chou, W.W., Yang, W.H., Harmon, M.E. & Silver, W.L. (2009). Controls on long-term  
385 root and leaf litter decomposition in neotropical forests. *Global Change Biology*, 15,  
386 1339–1355.

387 Elser, J.J., Bracken, M.E., Cleland, E.E., Gruner, D.S., Harpole, W.S., Hillebrand, H. et al. (2007).  
388 Global analysis of nitrogen and phosphorus limitation of primary producers in freshwater,  
389 marine and terrestrial ecosystems. *Ecology Letters*, 10, 1135–1142.

390 Fahey, T.J., Siccama, T.G., Driscoll, C.T., Likens, G.E., Campbell, J., Johnson, C.E. et al. (2005).  
391 The Biogeochemistry of Carbon at Hubbard Brook. *Biogeochemistry*, 75, 109–176.

392 Fahey, T.J., Stevens, P.A., Hornung, M. & Rowland, P. (1991). Decomposition and Nutrient Release  
393 from Logging Residue Following Conventional Harvest of Sitka Spruce in North Wales.  
394 *Forestry*, 64, 289–301.

395 Fan, P. & Guo, D. (2010). Slow decomposition of lower order roots: a key mechanism of root  
396 carbon and nutrient retention in the soil. *Oecologia*, 163, 509–515.

397 Freschet, G.T., Cornwell, W.K., Wardle, D.A., Elumeeva, T.G., Liu, W., Jackson, B.G. et al. (2013).  
398 Linking litter decomposition of above- and below-ground organs to plant-soil feedbacks

399 worldwide. *Journal of Ecology*, 101, 943–952.

400 Freschet, G.T., Pages, L., Iversen, C.M., Comas, L.H., Rewald, B., Roumet, C. et al. (2021). A  
401 starting guide to root ecology: strengthening ecological concepts and standardising root  
402 classification, sampling, processing and trait measurements. *New Phytologist*, 232,  
403 973–1122.

404 Frey, S.D., Six, J. & Elliott, E.T. (2003). Reciprocal transfer of carbon and nitrogen by decomposer  
405 fungi at the soil–litter interface. *Soil Biology and Biochemistry*, 35, 1001–1004.

406 Fujii, S. & Takeda, H. (2010). Dominant effects of litter substrate quality on the difference between  
407 leaf and root decomposition process above- and belowground. *Soil Biology and*  
408 *Biochemistry*, 42, 2224–2230.

409 Gang, Q., Chang, S.X., Lin, G., Zhao, Q., Mao, B. & Zeng, D.-H. (2019). Exogenous and  
410 endogenous nitrogen differentially affect the decomposition of fine roots of different  
411 diameter classes of Mongolian pine in semi-arid northeast China. *Plant and Soil*, 436,  
412 109–122.

413 Gao, D., Bai, E., Wang, S., Zong, S., Liu, Z., Fan, X. et al. (2022). Three-dimensional mapping of  
414 carbon, nitrogen, and phosphorus in soil microbial biomass and their stoichiometry at the  
415 global scale. *Global Change Biology*, 28, 6728–6740.

416 Gill, A.L., Schilling, J. & Hobbie, S.E. (2021). Experimental nitrogen fertilisation globally

417 accelerates, then slows decomposition of leaf litter. *Ecology Letters*, 24, 802–811.

418 Goebel, M., Hobbie, S.E., Bulaj, B., Zadworny, M., Archibald, D., Oleksyn, J. et al. (2011).

419 Decomposition of the finest root branching orders: linking belowground dynamics to

420 fine-root function and structure. *Ecological Monographs*, 81, 89–102.

421 Groemping, U. (2006). Relative Importance for Linear Regression in R: The Package relaimpo.

422 *Journal of Statistical Software*, 17, 1–27.

423 Guo, L., Deng, M., Yang, S., Liu, W., Wang, X., Wang, J. et al. (2021). The coordination between

424 leaf and fine root decomposition and the difference in their controlling factors. *Global*

425 *Ecology and Biogeography*, 30, 2286–2296.

426 Harmon, M.E., Silver, W.L., Fasth, B., Chen, H., Burke, I.C., Parton, W.J. et al. (2009). Long-term

427 patterns of mass loss during the decomposition of leaf and fine root litter: an intersite

428 comparison. *Global Change Biology*, 15, 1320–1338.

429 Harpole, W.S., Ngai, J.T., Cleland, E.E., Seabloom, E.W., Borer, E.T., Bracken, M.E. et al. (2011).

430 Nutrient co-limitation of primary producer communities. *Ecology Letters*, 14, 852–862.

431 Hobbie, S.E. (2015). Plant species effects on nutrient cycling: revisiting litter feedbacks. *Trends in*

432 *Ecology and Evolution*, 30, 357–363.

433 Hobbie, S.E., Eddy, W.C., Buyarski, C.R., Adair, E.C., Ogdahl, M.L. & Weisenhorn, P. (2012).

434 Response of decomposing litter and its microbial community to multiple forms of nitrogen

435 enrichment. *Ecological Monographs*, 82, 389–405.

436 Hobbie, S.E., Oleksyn, J., Eissenstat, D.M. & Reich, P.B. (2010). Fine root decomposition rates do  
437 not mirror those of leaf litter among temperate tree species. *Oecologia*, 162, 505–513.

438 Jani, A.D., Grossman, J.M., Smyth, T.J. & Hu, S. (2015). Influence of soil inorganic nitrogen and  
439 root diameter size on legume cover crop root decomposition and nitrogen release. *Plant and*  
440 *Soil*, 393, 57–68.

441 Jiang, L., Wang, H., Li, S., Fu, X., Dai, X., Yan, H. et al. (2021). Mycorrhizal and environmental  
442 controls over root trait-decomposition linkage of woody trees. *New Phytologist*, 229,  
443 284–295.

444 Jing, H., Zhang, P., Li, J., Yao, X., Liu, G. & Wang, G. (2019). Effect of nitrogen addition on the  
445 decomposition and release of compounds from fine roots with different diameters: the  
446 importance of initial substrate chemistry. *Plant and Soil*, 438, 281–296.

447 Keller, A.B., Brzostek, E.R., Craig, M.E., Fisher, J.B. & Phillips, R.P. (2021). Root-derived inputs  
448 are major contributors to soil carbon in temperate forests, but vary by mycorrhizal type.  
449 *Ecology Letters*, 24, 626–635.

450 Li, A., Fahey, T.J., Pawlowska, T.E., Fisk, M.C. & Burtis, J. (2015). Fine root decomposition,  
451 nutrient mobilization and fungal communities in a pine forest ecosystem. *Soil Biology and*  
452 *Biochemistry*, 83, 76–83.

453 Lin, C., Yang, Y., Guo, J., Chen, G. & Xie, J. (2010). Fine root decomposition of evergreen  
 454 broadleaved and coniferous tree species in mid-subtropical China: dynamics of dry mass,  
 455 nutrient and organic fractions. *Plant and Soil*, 338, 311–327.

456 Lin, D., Dou, P., Yang, G., Qian, S., Wang, H., Zhao, L. et al. (2020). Home-field advantage of litter  
 457 decomposition differs between leaves and fine roots. *New Phytologist*, 227, 995–1000.

458 Olson, J.S. (1963). Energy storage and the balance of producers and decomposers in ecological  
 459 systems. *Ecology*, 44, 322–331.

460 Pan, J., Wang, C. & Wang, X. (2021). Seven-year decomposition rates of roots with different  
 461 diameters for three temperate broadleaf tree species. *Acta Ecologica Sinica*, 41, 5166–5174.

462 Pang, Y., Tian, J., Lv, X., Wang, R., Wang, D. & Zhang, F. (2022). Contrasting dynamics and factor  
 463 controls in leaf compared with different-diameter fine root litter decomposition in secondary  
 464 forests in the Qinling Mountains after 5 years of whole-tree harvesting. *Science of the Total*  
 465 *Environment*, 838, 156194.

466 Parton, W., Silver, W.L., Burke, I.C., Grassens, L., Harmon, M.E., Currie, W.S. et al. (2007).  
 467 Global-scale similarities in nitrogen release patterns during long-term decomposition.  
 468 *Science*, 315, 361–364.

469 Rastetter, E.B., Kwiatkowski, B.L., Kicklighter, D.W., Barker Plotkin, A., Genet, H., Nippert, J.B. et  
 470 al. (2022). N and P constrain C in ecosystems under climate change: Role of nutrient

471 redistribution, accumulation, and stoichiometry. *Ecological Applications*, 32, e2684.

472 Sariyildiz, T. (2015). Effects of tree species and topography on fine and small root decomposition  
473 rates of three common tree species (*Alnus glutinosa*, *Picea orientalis* and *Pinus sylvestris*) in  
474 Turkey. *Forest Ecology and Management*, 335, 71–86.

475 Schaller, J. & Struyf, E. (2013). Silicon controls microbial decay and nutrient release of grass litter  
476 during aquatic decomposition. *Hydrobiologia*, 709, 201–212.

477 See, C.R., Keller, A.B., Hobbie, S.E., Kennedy, P.G., Weber, P.K. & Pett-Ridge, J. (2022). Hyphae  
478 move matter and microbes to mineral microsites: Integrating the hyphosphere into  
479 conceptual models of soil organic matter stabilization. *Global Change Biology*, 28,  
480 2527–2540.

481 See, C.R., McCormack, M.L., Hobbie, S.E., Flores-Moreno, H., Silver, W.L. & Kennedy, P.G.  
482 (2019). Global patterns in fine root decomposition: climate, chemistry, mycorrhizal  
483 association and woodiness. *Ecology Letters*, 22, 946–953.

484 Silver, W.L. & Miya, R.K. (2001). Global patterns in root decomposition: comparisons of climate  
485 and litter quality effects. *Oecologia*, 129, 407–419.

486 Song, S., Hu, X., Zhu, J., Zheng, T., Zhang, F., Ji, C. et al. (2021). The decomposition rates of leaf  
487 litter and fine root and their temperature sensitivities are influenced differently by biotic  
488 factors. *Plant and Soil*, 461, 603–616.

489 Sulman, B.N., Brzostek, E.R., Medici, C., Shevliakova, E., Menge, D.N.L. & Phillips, R.P. (2017).  
490 Feedbacks between plant N demand and rhizosphere priming depend on type of mycorrhizal  
491 association. *Ecology Letters*, 20, 1043–1053.

492 Sun, T., Hobbie, S.E., Berg, B., Zhang, H., Wang, Q., Wang, Z. et al. (2018). Contrasting dynamics  
493 and trait controls in first-order root compared with leaf litter decomposition. *Proceedings of*  
494 *the National Academy of Sciences*, 115, 10392–10397.

495 Sun, T., Mao, Z. & Han, Y. (2013). Slow decomposition of very fine roots and some factors  
496 controlling the process: a 4-year experiment in four temperate tree species. *Plant and Soil*,  
497 372, 445–458.

498 Sun, X., Liu, F., Zhang, Q., Li, Y., Zhang, L., Wang, J. et al. (2021). Biotic and climatic controls on  
499 the interannual variation in canopy litterfall of a deciduous broad-leaved forest. *Agricultural*  
500 *and Forest Meteorology*, 307.

501 Tombeur, F.d., Turner, B.L., Laliberté, E., Lambers, H., Mahy, G., Faucon, M.-P. et al. (2020). Plants  
502 sustain the terrestrial silicon cycle during ecosystem retrogression. *Science*, 369, 1245–1248.

503 Tong, J., Xiang, W., Liu, C., Lei, P., Tian, D., Deng, X. et al. (2012). Tree species effects on fine root  
504 decomposition and nitrogen release in subtropical forests in southern China. *Plant Ecology*  
505 *& Diversity*, 5, 323–331.

506 Usman, S., Singh, S.P., Rawat, Y.S. & Bargali, S.S. (2000). Fine root decomposition and nitrogen

507 mineralisation patterns in *Quercus leucotrichophora* and *Pinus roxburghii* forests in central  
508 Himalaya. *Forest Ecology and Management*, 131, 191–199.

509 Wambsganss, J., Freschet, G.T., Beyer, F., Bauhus, J. & Scherer-Lorenzen, M. (2022). Tree Diversity,  
510 Initial Litter Quality, and Site Conditions Drive Early-Stage Fine-Root Decomposition in  
511 European Forests. *Ecosystems*, 25 1493–1509.

512 Wang, P., Liu, X., Mou, P., Guo, J. & Li, S. (2019). Root order and initial moisture status influenced  
513 root decomposition in a subtropical tree species *Liquidambar formosana*. *Plant and Soil*, 443,  
514 539–548.

515 Wang, X., Song, H., Liu, F., Quan, X. & Wang, C. (2022). Timing of leaf fall and changes in litter  
516 nutrient concentration compromise estimates of nutrient fluxes and nutrient resorption  
517 efficiency. *Forest Ecology and Management*, 513.

518 Xiong, Y., Fan, P., Fu, S., Zeng, H. & Guo, D. (2012). Slow decomposition and limited nitrogen  
519 release by lower order roots in eight Chinese temperate and subtropical trees. *Plant and Soil*,  
520 363, 19–31.

521 Xu, Y., Huang, R., Zhou, B. & Ge, X. (2022). Fine-Root Decomposition and Nutrient Return in  
522 Moso Bamboo (*Phyllostachys pubescens* J.Houz.) Plantations in Southeast China. *Front*  
523 *Plant Sci*, 13, 735359.

524 Yang, S., Cheng, R., Xiao, W., Shen, Y., Wang, L., Guo, Y. et al. (2019). Heterogeneity in



525           Decomposition Rates and Nutrient Release in Fine-Root Architecture of *Pinus massoniana*  
526           in the Three Gorges Reservoir Area. *Forests*, 11, 14.

527   Zhang, D., Hui, D., Luo, Y. & Zhou, G. (2008). Rates of litter decomposition in terrestrial  
528           ecosystems: global patterns and controlling factors. *Journal of Plant Ecology*, 1, 85–93.

529   Zhang, H., Wang, C. & Wang, X. (2014). Spatial variations in non-structural carbohydrates in stems  
530           of twelve temperate tree species. *Trees*, 28, 77–89.

531   Zhang, X. & Wang, W. (2015). The decomposition of fine and coarse roots: their global patterns and  
532           controlling factors. *Scientific Reports*, 5, 1–10.

533   Zhou, Z. & Wang, C. (2016). Responses and regulation mechanisms of microbial decomposers to  
534           substrate carbon, nitrogen, and phosphorus stoichiometry. *Chinese Journal of Plant Ecology*,  
535           49, 620–630.

536   Zhuang, L., Yang, W., Wu, F., Tan, B., Zhang, L., Yang, K. et al. (2018). Diameter-related variations  
537           in root decomposition of three common subalpine tree species in southwestern China.  
538           *Geoderma*, 311, 1–8.

539

## Figure captions

**FIGURE 1** Short-term decomposition rate ( $k$ ) and nutrient release in different root diameter classes of three species. BP, *Betula platyphylla*; UJ, *Ulmus davidiana* var. *japonica*; FM, *Fraxinus mandshurica*. Different lower-case letters indicate significant differences between root diameter within species; different capital letters indicate significant difference between species within root diameter ( $P < 0.05$ ). Short-term decomposition rate ( $k$ ) based on Eq. (1).

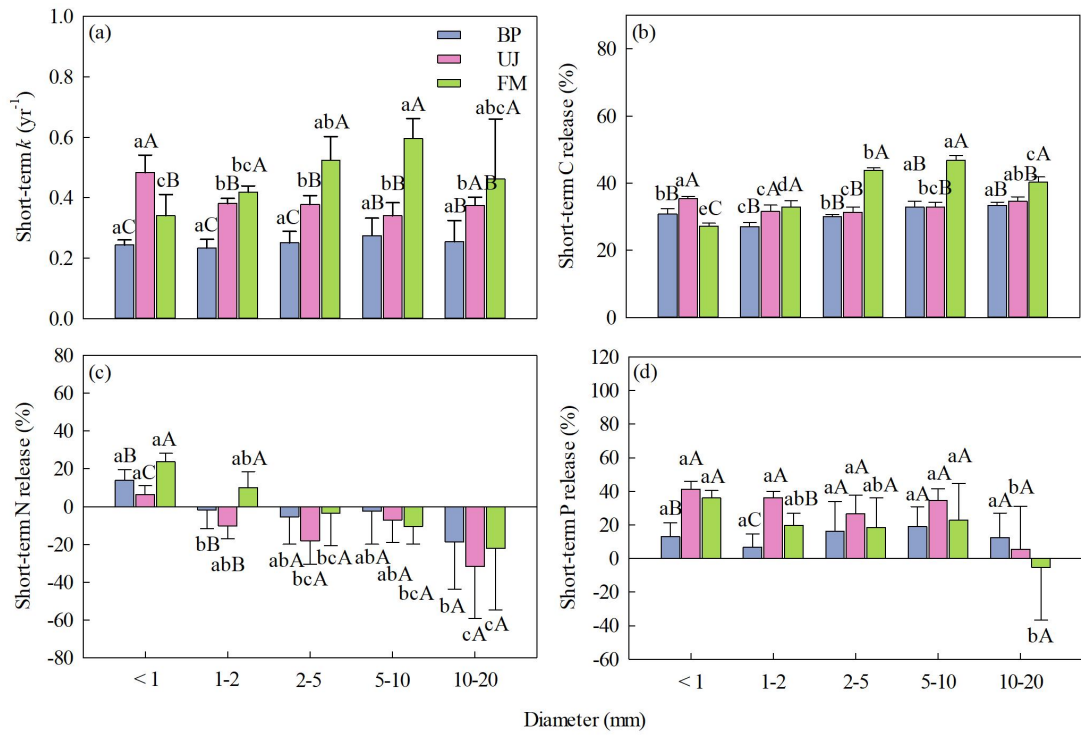
**FIGURE 2** Long-term decomposition rate ( $k$ ) and nutrient release in different root diameter classes of three species. BP, *Betula platyphylla*; UJ, *Ulmus davidiana* var. *japonica*; FM, *Fraxinus mandshurica*. Different low-case letters indicate significant differences between root diameter within species; different capital letters indicate significant difference between species within root diameter ( $P < 0.05$ ). Long-term decomposition rate ( $k$ ) based on Eq. (1).

**FIGURE 3** The first two principal components of the PCA for decomposition rates and nutrient release showing their similarities and differences.  $Lk$ , Long-term decomposition rate; LCR, Long-term C release; LNR, Long-term N release; LPR, Long-term P release;  $Sk$ , Short-term decomposition rate; SCR, Short-term C release; SNR, Short-term N release; SPR, Short-term P release. BP, *Betula platyphylla*; UJ, *Ulmus davidiana* var. *japonica*; FM, *Fraxinus mandshurica*.

**FIGURE 4** The relative importance (%) of the main contributors of the initial root quality to short-term and long-term decomposition constant ( $k$ ) and nutrient release rates.  $Lk$ , Long-term decomposition constant; LCR, Long-term C release rate; LNR,

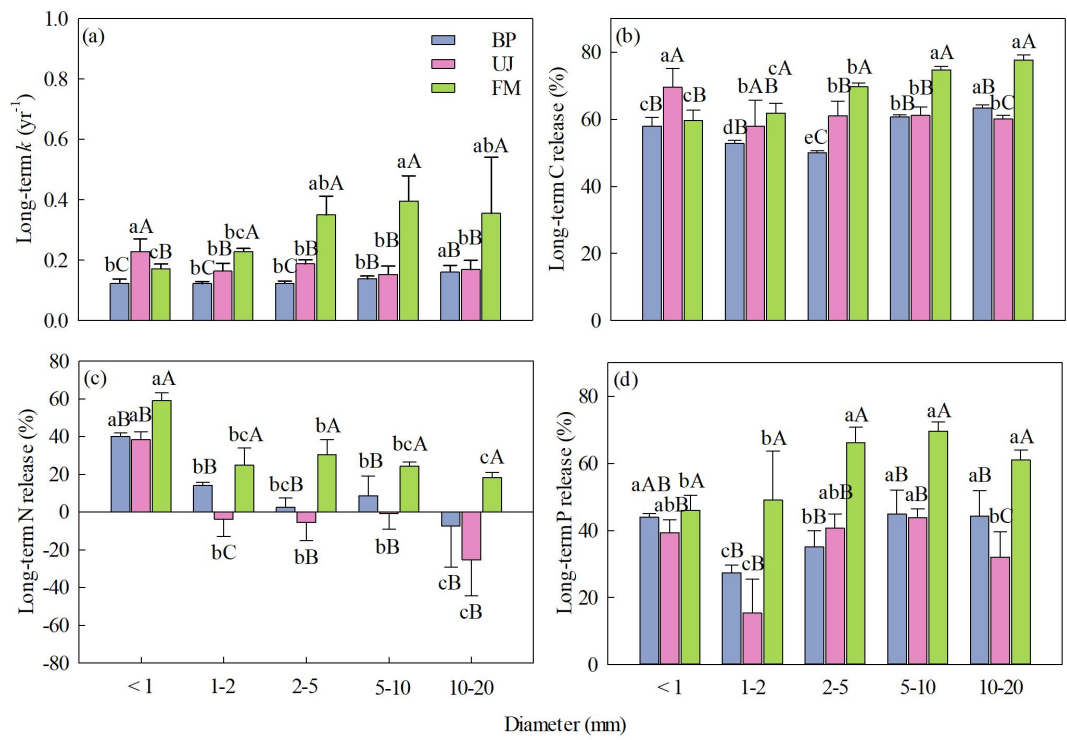
23 Long-term N release rate; LPR, Long-term P release rate;  $S_k$ , Short-term  
24 decomposition constant; SCR, Short-term C release rate; SNR, Short-term N release  
25 rate; SPR, Short-term P release rate. SS, Soluble sugars; NSC, Non-structural  
26 carbohydrates; Hemi, Hemicellulose; Cell, Cellulose; Lign, Lignin; Phe, Phenolics; C,  
27 Carbon; P, Phosphorus; Ca, Calcium; Mg, Magnesium; Si, Silicon.

28 **FIGURE 5** The relationship of the difference from long-term N release and P release  
29 with initial and final nutrients in roots. Each point within a species represents a  
30 diameter class.



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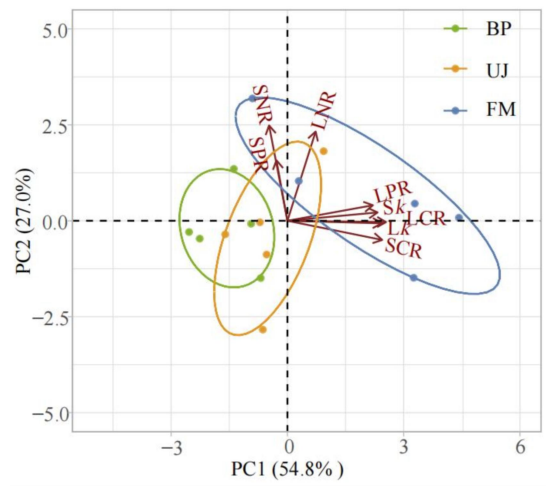
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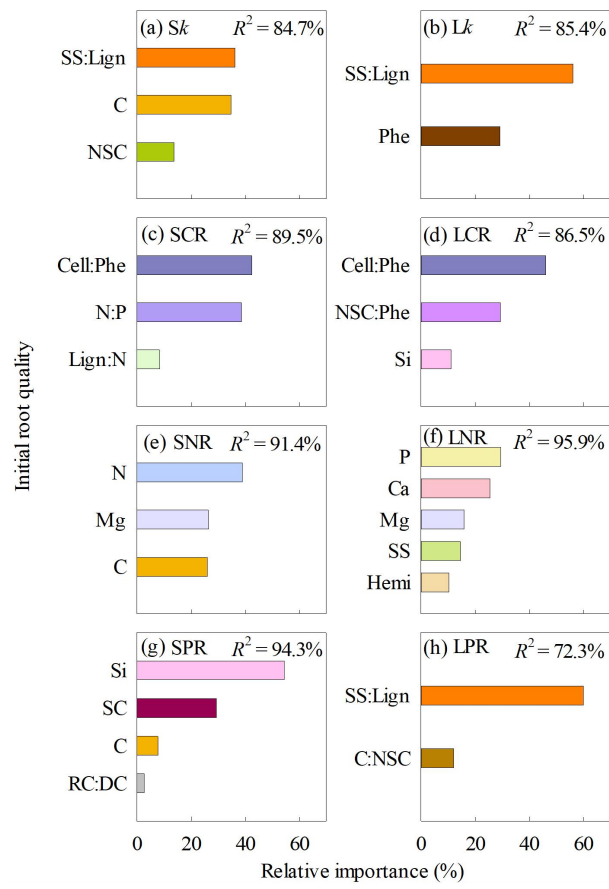
37     FIGURE 3



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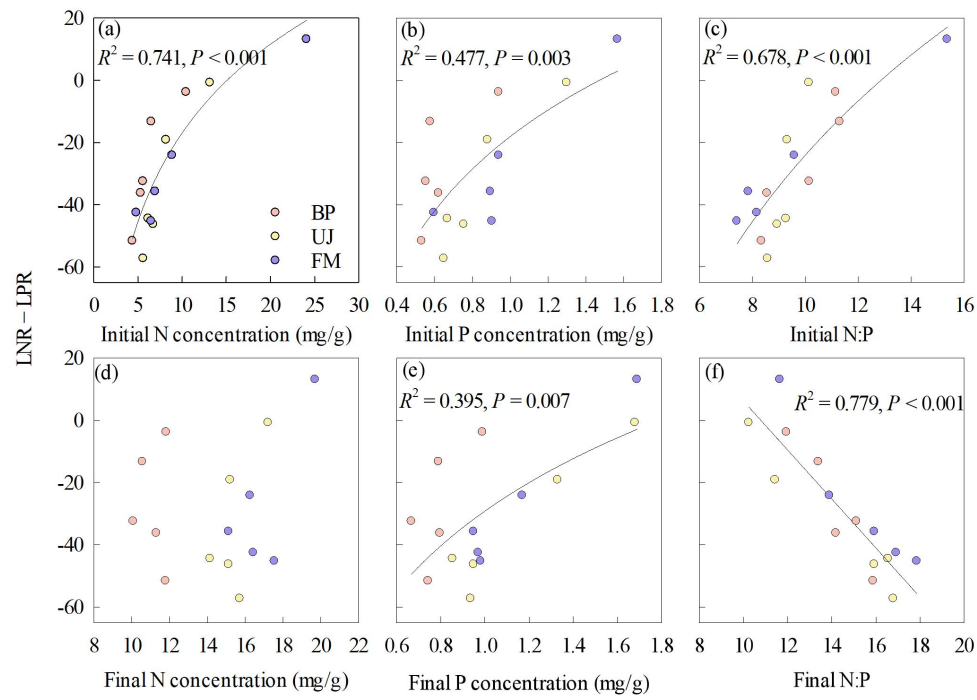
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40      **FIGURE 4**



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## **Supporting Information**

**Table S1** Decomposition parameters obtained from fitting double exponential, single exponential, and asymptotic model and its Akaike Information Criterion (AIC) for roots with various diameter of three species

**Table S2** Repeated measurements analysis of variance for carbon, nitrogen, phosphorous concentrations for the three species

**Table S3** Repeated measurements analysis of variance for carbon, nitrogen, phosphorous remaining for the three species

**Table S4** Results of two-way analysis of variance for initial root quality for the three tree species

**Table S5** Forward multiple linear regressions between the initial root quality and root decomposition rate ( $k$ ), nutrient release

**FIGURE S1** Carbon, nitrogen and phosphorous concentrations dynamic in different root diameter classes of three species during the seven years' decomposition

**FIGURE S2** Carbon, nitrogen and phosphorous dynamics in different root diameter classes of three species during the seven years' decomposition

**FIGURE S3** Linear regression of long-term and short-term decomposition rates ( $k$ ) against long-term and short-term nutrient release for the three species

**FIGURE S4** Correlations between initial root quality and short-term decomposition rate ( $k$ ),

nutrient release for the three species.

**FIGURE S5** Correlations between initial root quality and long-term decomposition rate ( $k$ ), nutrient release of the three tree species.

**FIGURE S6** Sensitivities of short-term decomposition rate ( $k$ ) and nutrient release to their two major contribution factors.

**FIGURE S7** Sensitivities of long-term decomposition rate ( $k$ ) and nutrient release to their two major contribution factors.

**FIGURE S8** The relative importance (%) of the main contributors of the initial root quality to the difference between long-term N release and long-term P release

**FIGURE S9** Linear regression of the main contributors of the initial root quality structural carbohydrates (SC) to the difference between short-term N release and short-term P release

**FIGURE S10** Linear regression of short-term and long-term N release rate (%) against short-term and long-term P release rate (%) for the three species.

**FIGURE S11** Comparison of initial decomposable carbon fractions in roots with different diameters for the three species

**FIGURE S12** Comparison of initial resistant carbon fractions in roots with different diameters for the three species

**FIGURE S13** Comparison of initial element concentrations in roots with different diameters for the

three species

**FIGURE S14** The ratio of initial elements and carbon quality to phenolics

**FIGURE S15** The ratio between initial carbon, nitrogen, phosphorus, and NSC

**FIGURE S16** The ratio of initial nitrogen, phosphorus, and carbon quality to carbon quality among

root diameters for the three species

**FIGURE S17** Comparison of carbon concentration in roots with different diameters for the three

species at different periods

**Table S1** Decomposition parameters obtained from fitting double exponential, single exponential, and asymptotic model and its Akaike Information Criterion (AIC) for roots with various diameter of three species. C, the fraction of the initial mass that decomposes with decomposition rate  $k_1$ ;  $k_1$ ,  $k_2$ , double exponential model decomposition rate;  $k$ , single exponential model decomposition rate; A, asymptote;  $k_a$ , asymptotic model decomposition rate.

Species	Diameter	double exponential				single exponential		asymptotic		
		C	$k_1$	$k_2$	AIC	$k$	AIC	A	$k_a$	AIC
BP	<1	0.154	4.556	0.067	-47.417	0.123	-22.445	0.582	0.707	-36.385
BP	1-2	0.148	4.845	0.068	-52.681	0.122	-23.376	0.579	0.669	-36.901
BP	2-5	0.186	3.016	0.057	-50.010	0.123	-21.356	0.596	0.803	-39.273
BP	5-10	0.158	7.019	0.089	-27.587	0.138	-10.639	0.483	0.596	-14.338
BP	10-20	0.123	5.872	0.119	-22.296	0.161	-14.319	0.391	0.453	-21.483
UJ	<1	0.290	14.000	0.068	-48.504	0.228	-4.046	0.595	5.027	-26.035

UJ	1-2	0.269	5.718	0.047	-47.522	0.164	-8.789	0.625	2.690	-33.880
UJ	2-5	0.238	6.401	0.076	-54.968	0.187	-11.987	0.559	1.477	-28.519
UJ	5-10	0.220	6.115	0.080	-32.140	0.152	-6.776	0.504	0.939	-14.285
UJ	10-20	0.243	5.776	0.083	-23.326	0.169	-5.560	0.482	1.007	-13.340
FM	<1	0.219	10.210	0.073	-36.667	0.172	-10.582	0.588	1.655	-23.158
FM	1-2	0.256	7.336	0.090	-47.602	0.228	-10.133	0.522	1.585	-24.227
FM	2-5	0.310	4.668	0.125	-41.146	0.350	-11.602	0.394	1.201	-27.486
FM	5-10	0.347	5.635	0.140	-22.642	0.396	-3.514	0.317	1.175	-12.299
FM	10-20	0.716	0.734	0.030	-19.192	0.355	-10.697	0.233	0.680	-21.144

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**Table S2** Repeated measurements analysis of variance for root carbon, nitrogen, phosphorous concentrations for the three tree species

Sources of variation	C concentration		N concentration		P concentration	
	<i>F</i> -value	<i>P</i> -value	<i>F</i> -value	<i>P</i> -value	<i>F</i> -value	<i>P</i> -value
Time (T)	131.672	< 0.001	88.338	< 0.001	5.372	< 0.001
Diameter (D)	23.186	< 0.001	1031.806	< 0.001	212.726	< 0.001
Species (S)	335.384	< 0.001	654.597	< 0.001	692.23	< 0.001
D×S	4.023	0.010	199.577	< 0.001	19.985	< 0.001
D×T	10.807	< 0.001	12.569	< 0.001	4.312	< 0.001
S×T	12.042	< 0.001	3.290	< 0.001	9.113	< 0.001
D×S×T	0.946	0.500	4.126	< 0.001	3.066	< 0.001

**Table S3** Repeated measurements analysis of variance for root carbon, nitrogen, phosphorous remaining for the three tree species

Sources of variation	C remaining		N remaining		P remaining	
	<i>F</i> -value	<i>P</i> -value	<i>F</i> -value	<i>P</i> -value	<i>F</i> -value	<i>P</i> -value
Time (T)	3521.558	< 0.001	24.504	< 0.001	45.287	< 0.001
Diameter (D)	37.739	< 0.001	143.014	< 0.001	101.501	< 0.001
Species (S)	661.988	< 0.001	87.048	< 0.001	158.783	< 0.001
D×S	284.199	< 0.001	1.523	0.227	32.597	< 0.001
D×T	10.812	< 0.001	6.986	< 0.001	4.445	< 0.001
S×T	22.930	< 0.001	7.135	< 0.001	14.649	< 0.001
D×S×T	10.294	< 0.001	1.714	0.038	4.929	< 0.001

**Table S4** Results of two-way analysis of variance for initial root quality for the three species. SS, Soluble sugars; Sta, Starch; NSC, Non-structural carbohydrates; Hemi, Hemicellulose; DC, Decomposable carbohydrates; Cell, Cellulose; Lign, Lignin; SC, Structural carbohydrates; Phe, Phenolics; RC, Resistant carbohydrates; C, Carbon; N, Nitrogen; P, Phosphorus; Ca, Calcium; Mg, Magnesium; Si, Silicon.

Initial quality	root	Species (S)		Diameter (D)		S×D	
		<i>F</i>	<i>P</i>	<i>F</i>	<i>P</i>	<i>F</i>	<i>P</i>
Soluble sugars		45.382	< 0.001	1.409	0.242	11.053	< 0.001
Starch		42.629	< 0.001	1.358	0.260	3.806	0.001
NSC		39.210	< 0.001	1.952	0.114	11.569	< 0.001
Hemicellulose		40.796	< 0.001	21.435	< 0.001	14.989	< 0.001
DC		24.251	< 0.001	12.206	< 0.001	13.044	< 0.001
Cellulose		18.236	< 0.001	21.552	< 0.001	2.726	0.015
Lignin		6.964	0.002	10.829	< 0.001	2.837	0.012
SC		18.490	< 0.001	4.776	0.003	4.481	< 0.001
Phenolics		272.000	< 0.001	47.663	< 0.001	10.059	< 0.001
RC		21.297	< 0.001	4.322	0.005	4.801	< 0.001
C		148.640	< 0.001	3.636	0.011	2.362	0.029
N		125.931	< 0.001	416.969	< 0.001	65.449	< 0.001
P		60.383	< 0.001	85.396	< 0.001	5.159	< 0.001
Ca		285.398	< 0.001	19.489	< 0.001	26.449	< 0.001
Mg		251.585	< 0.001	72.458	< 0.001	3.736	0.001

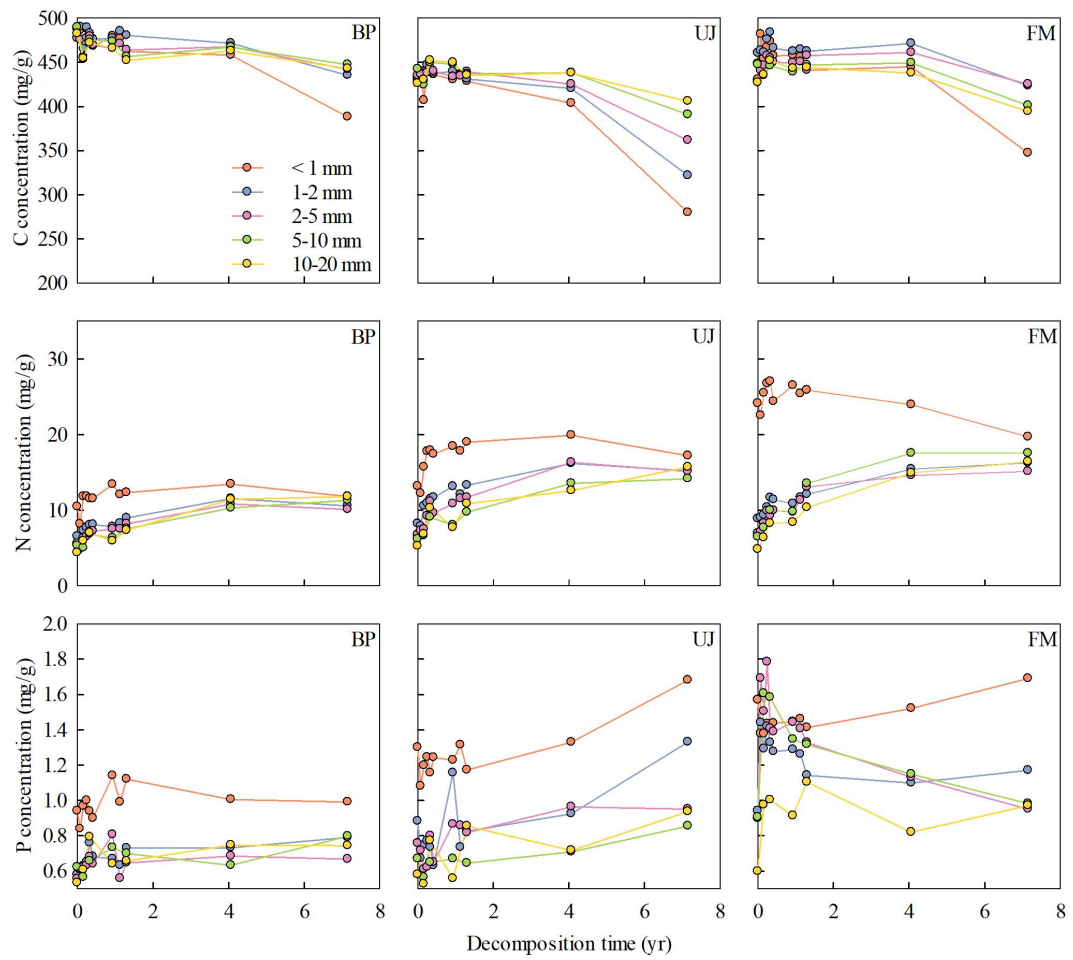


Initial	root	Species (S)		Diameter (D)		S×D	
quality		<i>F</i>	<i>P</i>	<i>F</i>	<i>P</i>	<i>F</i>	<i>P</i>
Si		67.200	< 0.001	40.591	< 0.001	4.076	0.002
SS:Phe		1185.90	< 0.001	102.828	< 0.001	37.666	< 0.001
		2					
NSC:Phe		528.961	< 0.001	83.790	< 0.001	5.769	< 0.001
N:Phe		420.043	< 0.001	132.292	< 0.001	74.668	< 0.001
C:Phe		412.422	< 0.001	65.547	< 0.001	3.488	0.002
Cell:Phe		210.614	< 0.001	74.574	< 0.001	10.261	< 0.001
C:N		72.875	< 0.001	128.209	< 0.001	1.618	0.140
N:P		3.697	0.031	52.208	< 0.001	17.983	< 0.001
C:P		70.244	< 0.001	37.785	< 0.001	1.762	0.104
NSC:N		9.487	< 0.001	76.905	< 0.001	15.512	< 0.001
NSC:P		7.062	0.002	24.677	< 0.001	9.418	< 0.001
C:NSC		69.286	< 0.001	4.514	0.003	12.993	< 0.001
Cell:N		23.760	< 0.001	69.393	< 0.001	2.915	0.010
Lign:N		34.077	< 0.001	57.879	< 0.001	3.118	0.007
Lign:P		32.836	< 0.001	14.686	< 0.001	2.767	0.014
SS:Lign		44.612	< 0.001	5.107	0.002	5.433	< 0.001
SC:NSC		36.911	< 0.001	3.351	0.017	5.215	< 0.001
RC:DC		19.311	< 0.001	14.555	< 0.001	9.778	< 0.001

**Table S5** Forward multiple linear regressions between the initial root quality and root decomposition rate ( $k$ ), nutrient release.  $Lk$ , Long-term decomposition rate; LCR, Long-term C release; LNR, Long-term N release; LPR, Long-term P release;  $Sk$ , Short-term decomposition rate; SCR, Short-term C release; SNR, Short-term N release; SPR, Short-term P release. SS, Soluble sugars; NSC, Non-structural carbohydrates; Hemi, Hemicellulose; DC, Decomposable carbohydrates; Cell, Cellulose; Lign, Lignin; SC, Structural carbohydrates; Phe, Phenolics; RC, Resistant carbohydrates; C, Carbon; N, Nitrogen; P, Phosphorus; Ca, Calcium; Mg, Magnesium; Si, Silicon.

Variable	$R^2$	Regression
$Lk$	0.796	$1.333 \times \text{SS:Lign} - 0.008 \times \text{Phe} + 0.006$
LCR	0.558	$1.474 \times \text{Cell:Phe} + 0.451 \times \text{Si} - 209.751 \times \text{NSC:Phe} + 503.878$
LNR	0.770	$52.789 \times \text{P} - 5.775 \times \text{Ca} - 0.767 \times \text{Hemi} + 0.602 \times \text{SS} - 28.939 \times \text{Mg} + 37.163$
LPR	0.492	$347.430 \times \text{SS:Lign} + 6.996 \times \text{C:NSC} - 61.705$
$Sk$	0.563	$2.328 \times \text{SS:Lign} - 0.003 \times \text{C} - 0.004 \times \text{NSC} + 1.478$
SCR	0.474	$0.331 \times \text{Cell:Phe} - 2.082 \times \text{N:P} - 0.170 \times \text{Lign:N} + 58.095$
SNR	0.698	$1.360 \times \text{N} + 0.476 \times \text{C} + 33.255 \times \text{Mg} - 270.928$
SPR	0.756	$1.132 \times \text{Si} - 0.254 \times \text{SC} + 0.237 \times \text{C} - 6.096 \times \text{RC:DC} + 25.107$





**FIGURE S1** Carbon, nitrogen and phosphorous concentrations dynamic in different root diameter classes of three species during the seven years' decomposition. BP, *Betula platyphylla*; UJ, *Ulmus davidiana* var. *japonica*; FM, *Fraxinus mandshurica*.

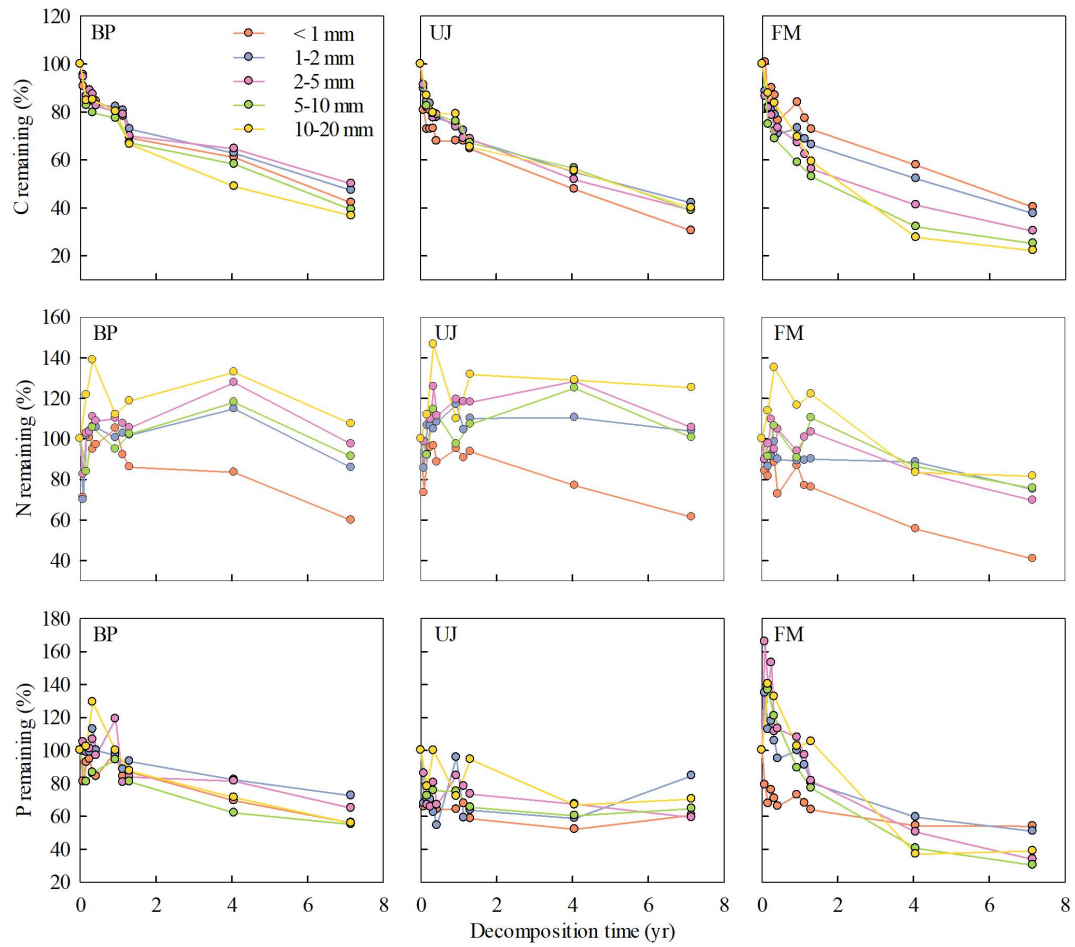
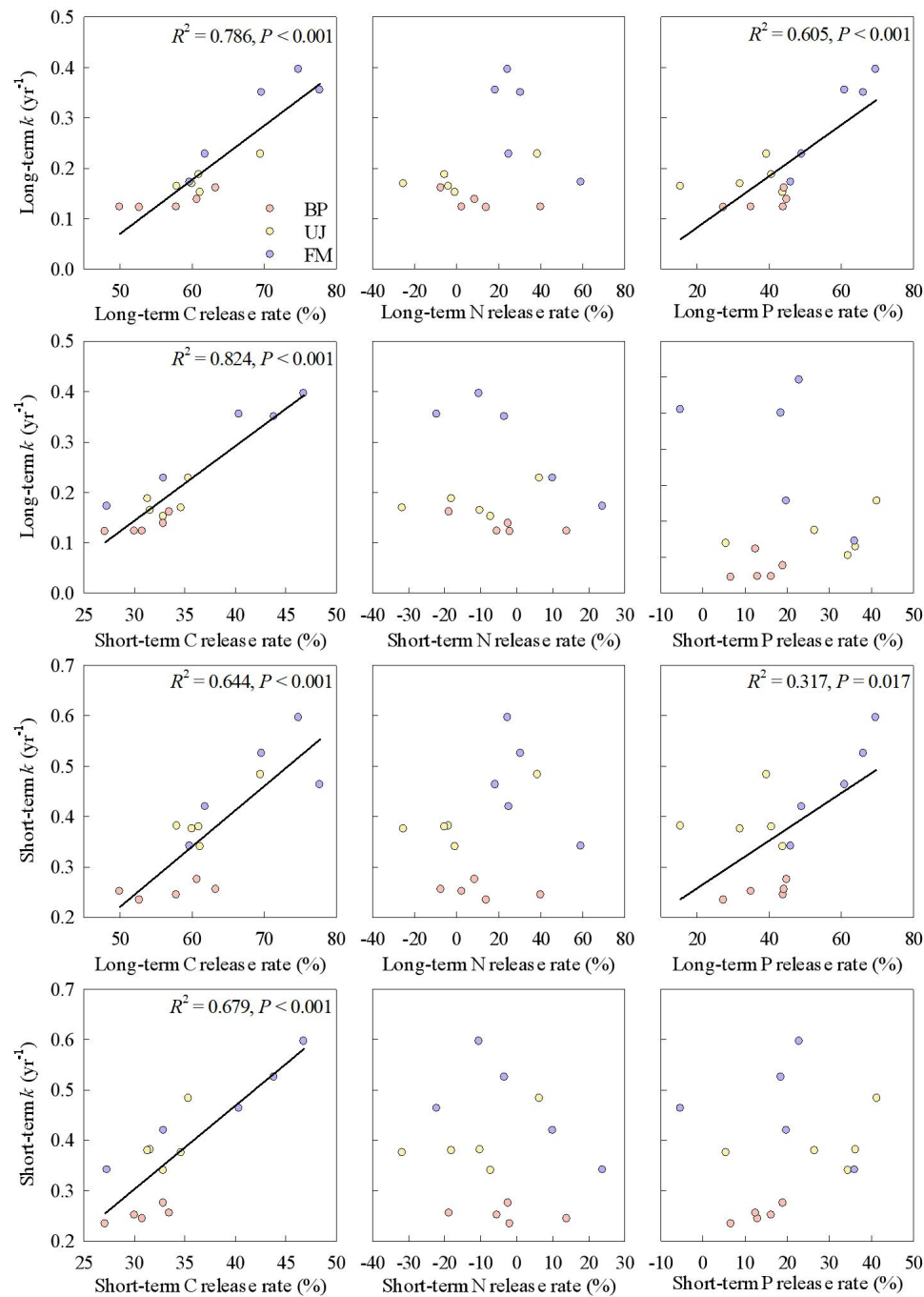


FIGURE S2 Carbon, nitrogen and phosphorous dynamics in different root diameter classes of three species during the seven years' decomposition. BP, *Betula platyphylla*; UJ, *Ulmus davidiana* var. *japonica*; FM, *Fraxinus mandshurica*.



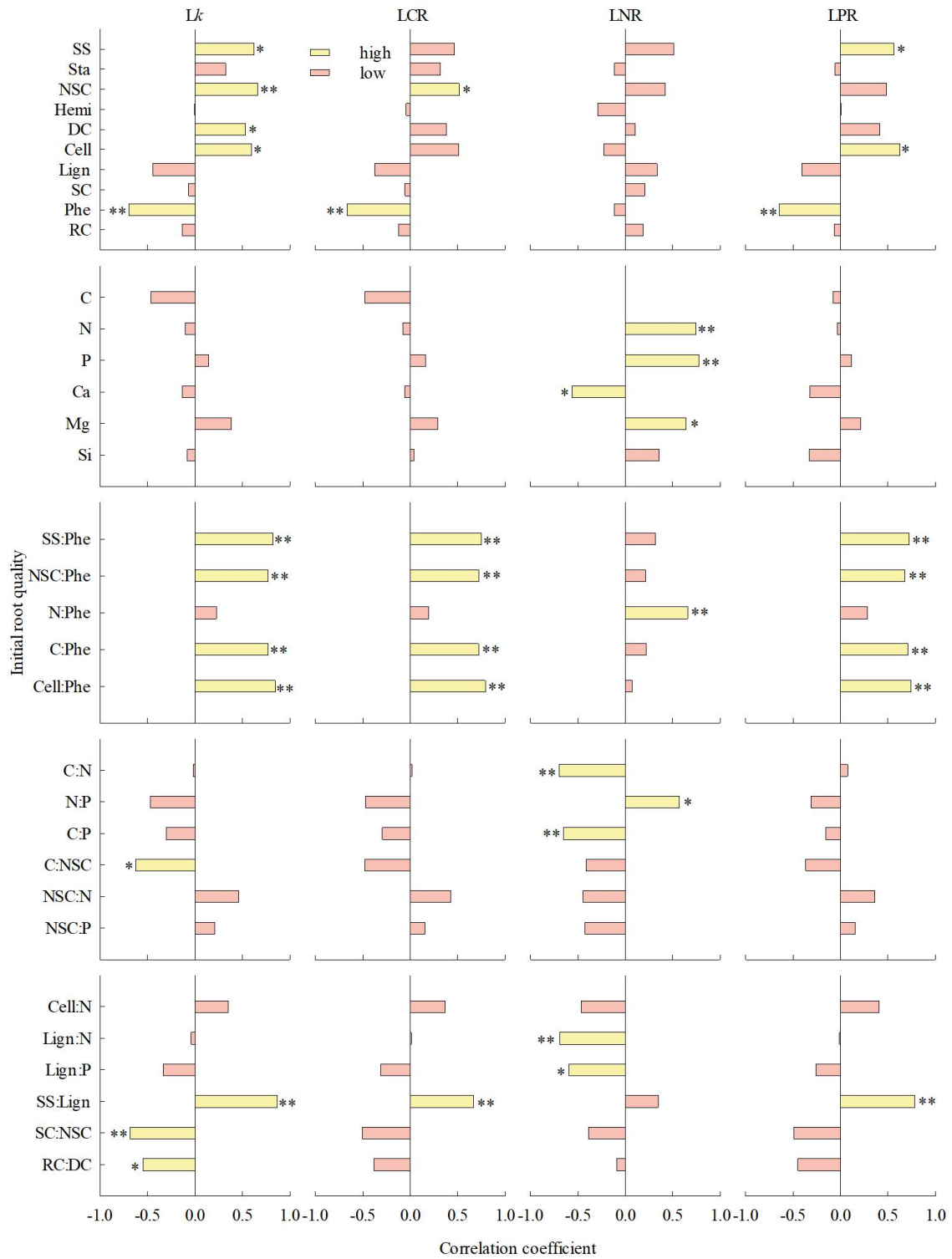
**FIGURE S3** Linear regression of long-term and short-term decomposition rates ( $k$ ) against long-term and short-term nutrient release for the three species. Each point within a species represents a different diameter class. Linear regression showed that long-term C and P release but not N release were significantly positively correlated with long-term decomposition rate ( $P < 0.05$ ), short-term C release but not N and P release was significantly positively correlated with decomposition rate ( $P < 0.05$ ).

BP, *Betula platyphylla*; UJ, *Ulmus davidiana* var. *japonica*; FM, *Fraxinus mandshurica*.



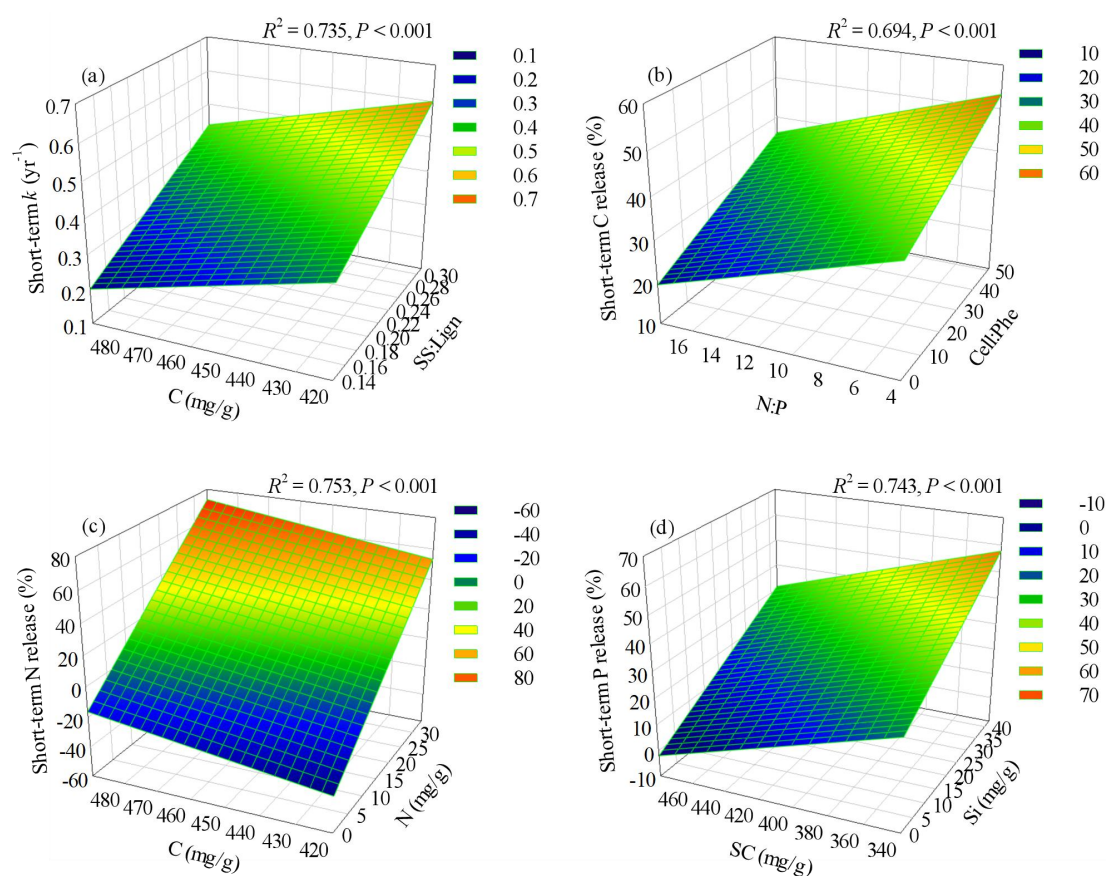
**FIGURE S4** Correlations between initial root quality and short-term decomposition rate ( $k$ ), nutrient release for the three species.  $Sk$ , Short-term decomposition rate;  $SCR$ , Short-term C release;  $SNR$ , Short-term N release;  $SPR$ , Short-term P release.  $SS$ , Soluble sugars;  $Sta$ , Starch;  $NSC$ , Non-structural carbohydrates;  $Hemi$ , Hemicellulose;  $DC$ , Decomposable carbohydrates ( $NSC$ ,  $Hemi$ );  $Cell$ , Cellulose;  $Lign$ , Lignin;  $SC$ , Structural carbohydrates ( $Cell$ ,  $Lign$ );  $Phe$ , Phenolics;  $RC$ , Resistant carbohydrates ( $Cell$ ,  $Lign$ ,  $Phe$ );  $C$ , Carbon;  $N$ , Nitrogen;  $P$ , Phosphorus;  $Ca$ , Calcium;  $Mg$ , Magnesium;  $Si$ , Silicon. \* represents  $P < 0.05$ , and \*\* represents  $P < 0.001$ . The same below.



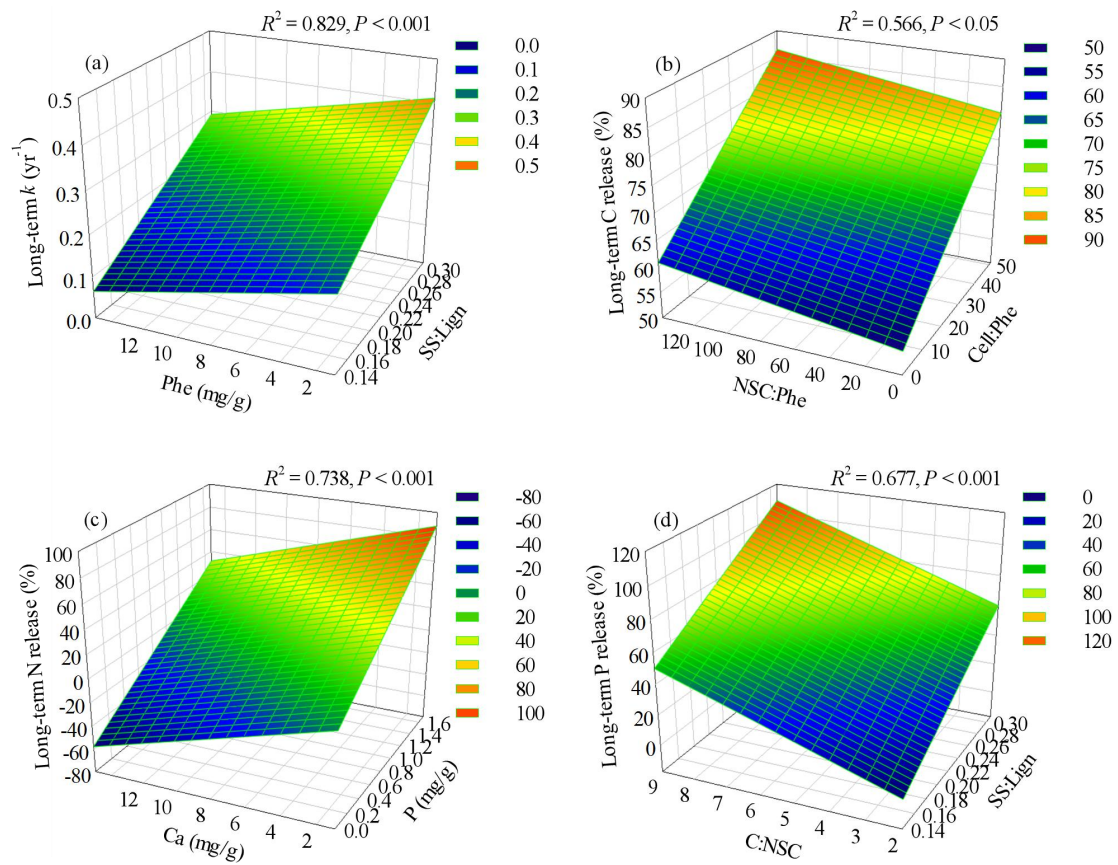


**FIGURE S5** Correlations between initial root quality and long-term decomposition rate ( $k$ ), nutrient release of the three tree species.  $Lk$ , Long-term decomposition rate; LCR, Long-term C release; LNR, Long-term N release; LPR, Long-term P release. SS,

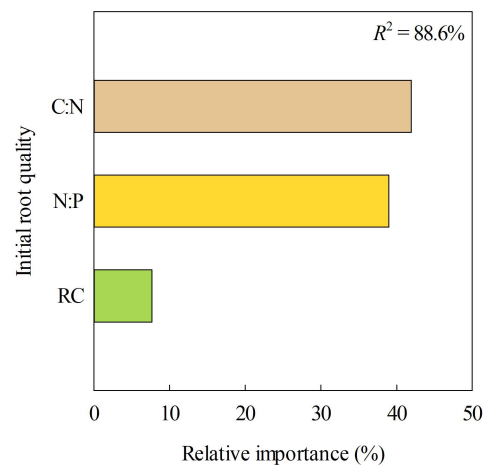
Soluble sugars; Sta, Starch; NSC, Non-structural carbohydrates; Hemi, Hemicellulose; DC, Decomposable carbohydrates (NSC, Hemi); Cell, Cellulose; Lign, Lignin; SC, Structural carbohydrates (Cell, Lign); Phe, Phenolics; RC, Resistant carbohydrates (Cell, Lign, Phe); C, Carbon; N, Nitrogen; P, Phosphorus; Ca, Calcium; Mg, Magnesium; Si, Silicon.



**FIGURE S6** Sensitivities of short-term decomposition rate ( $k$ ) and nutrient release to their two major contribution factors. The other significant factors were not considered. SS, Soluble sugars; Cell, Cellulose; Lign, Lignin; SC, Structural carbohydrates (Cell, Lign); Phe, Phenolics; C, Carbon; N, Nitrogen; P, Phosphorus; Si, Silicon.

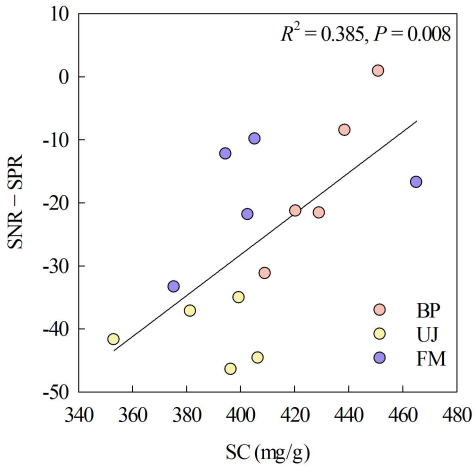


**FIGURE S7** Sensitivities of long-term decomposition rate ( $k$ ) and nutrient release to their two major contribution factors. The other significant factors were not considered. SS, Soluble sugars; NSC, Non-structural carbohydrates; Cell, Cellulose; Lign, Lignin; Phe, Phenolics; C, Carbon; P, Phosphorus; Ca, Calcium.

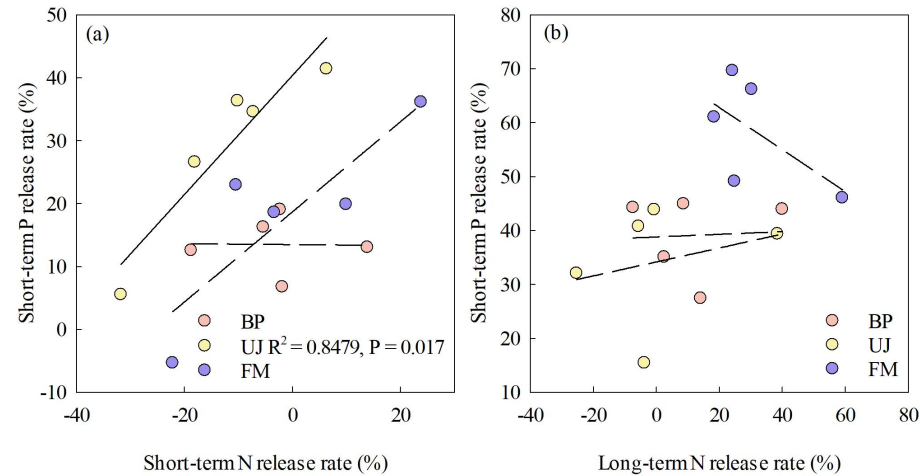


**FIGURE S8** The relative importance (%) of the main contributors of the initial root

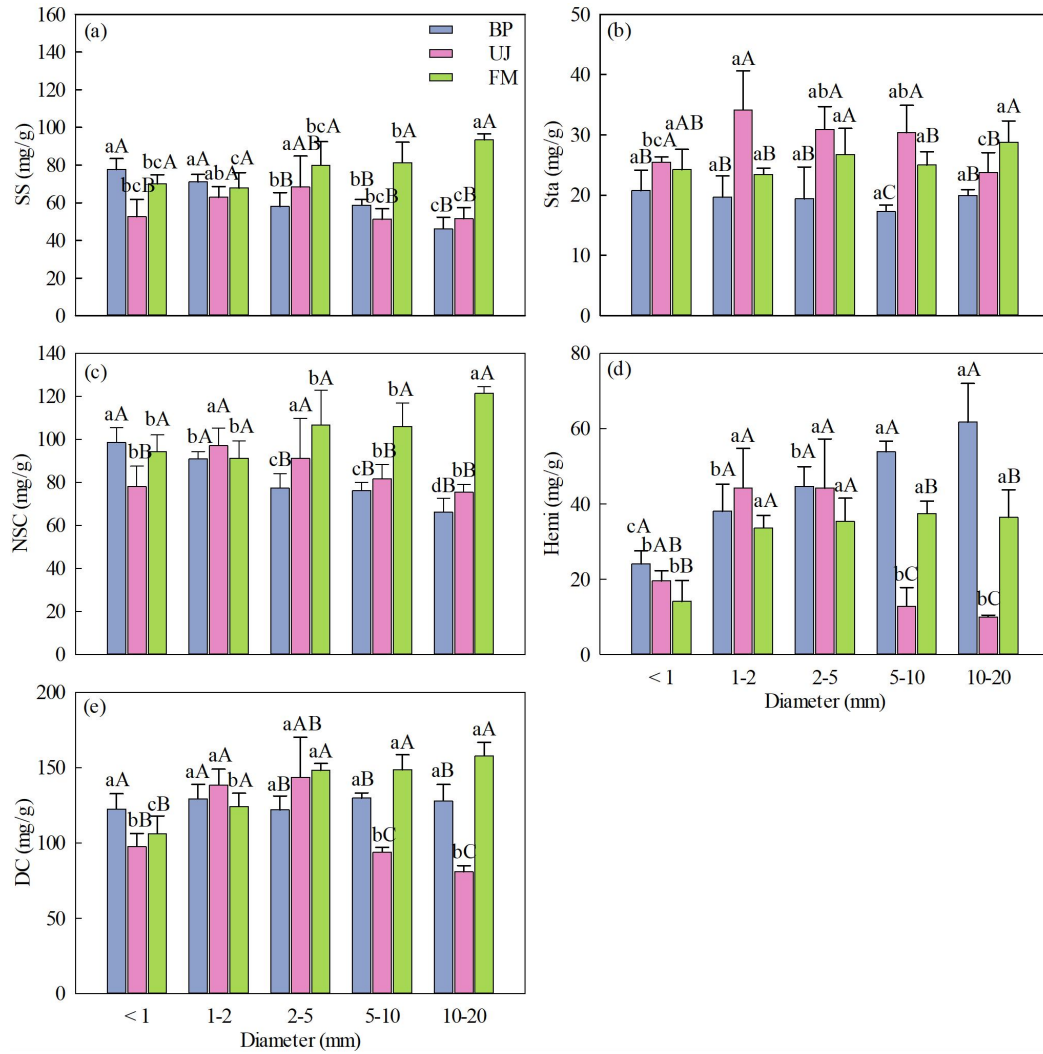
quality to the difference between long-term N and P release from the same species and diameter class. C: Carbon, n: nitrogen, P: phosphorous; RC: Resistant carbohydrates.



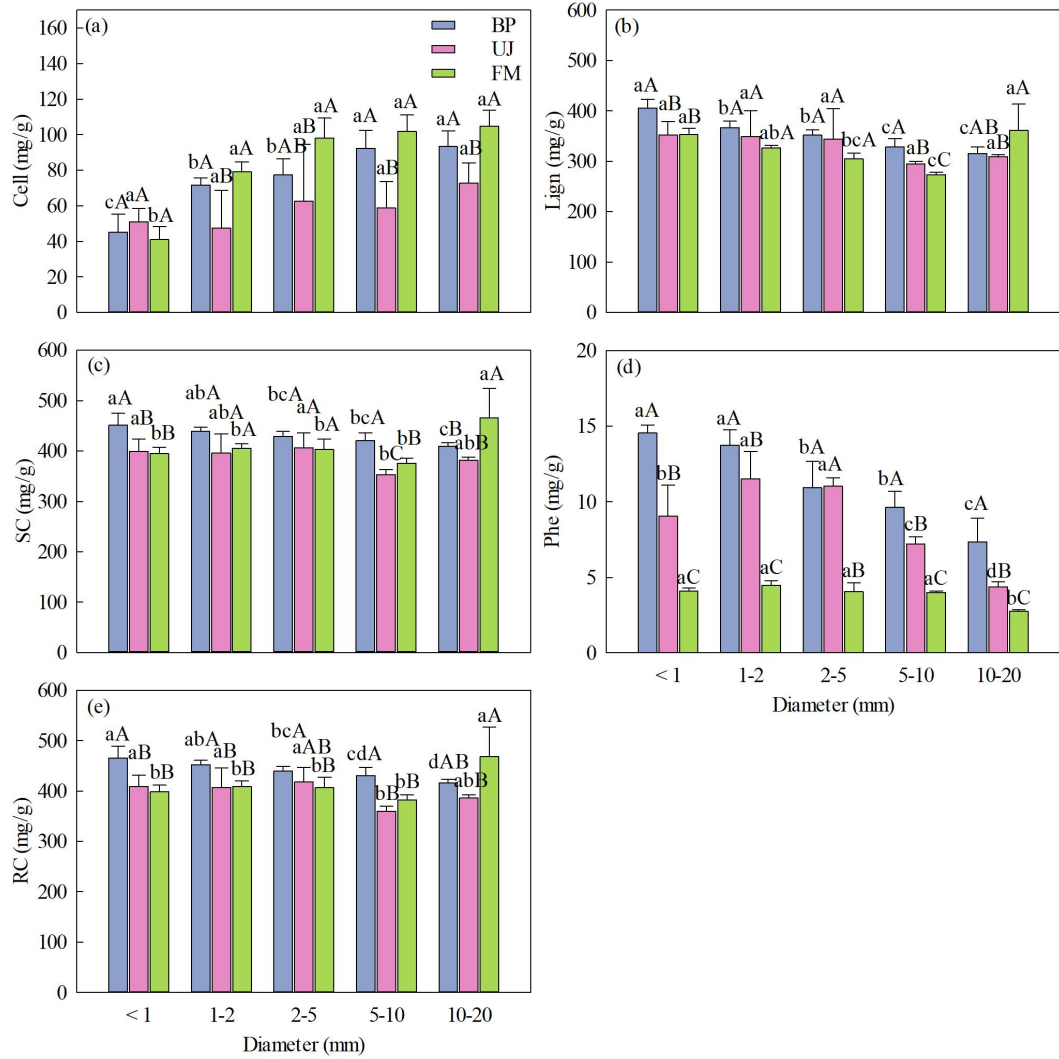
**FIGURE S9** Linear regression of the main contributor of the initial root quality structural carbohydrates (SC) to the difference between short-term N release and short-term P release.



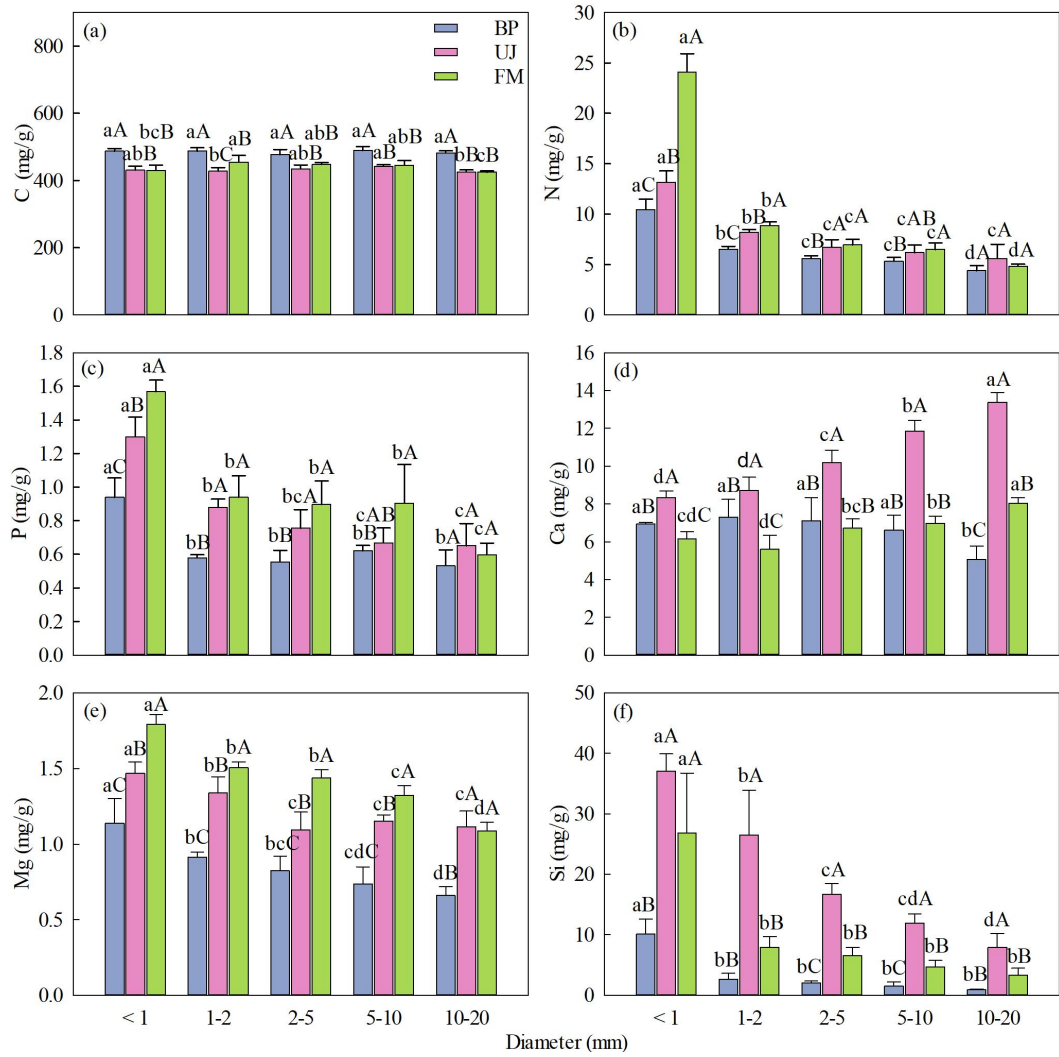
**FIGURE S10** Linear regression of short-term and long-term N release rate (%) against short-term and long-term P release rate (%) for the three species.



**FIGURE S11** Comparison of initial decomposable carbon fractions in roots with different diameters for the three species. BP, *Betula platyphylla*, UJ, *Ulmus davidiana* var. *japonica*, FM, *Fraxinus mandshurica*. SS, Soluble sugars; Sta, Starch; NSC, Non-structural carbohydrates; Hemi, Hemicellulose; DC, Decomposable carbohydrates. Different low-case letters indicate significant differences between root diameter with in species; Different capital letters indicate significant difference between species with in root diameters ( $P < 0.05$ ),  $n = 5$ . The same below.



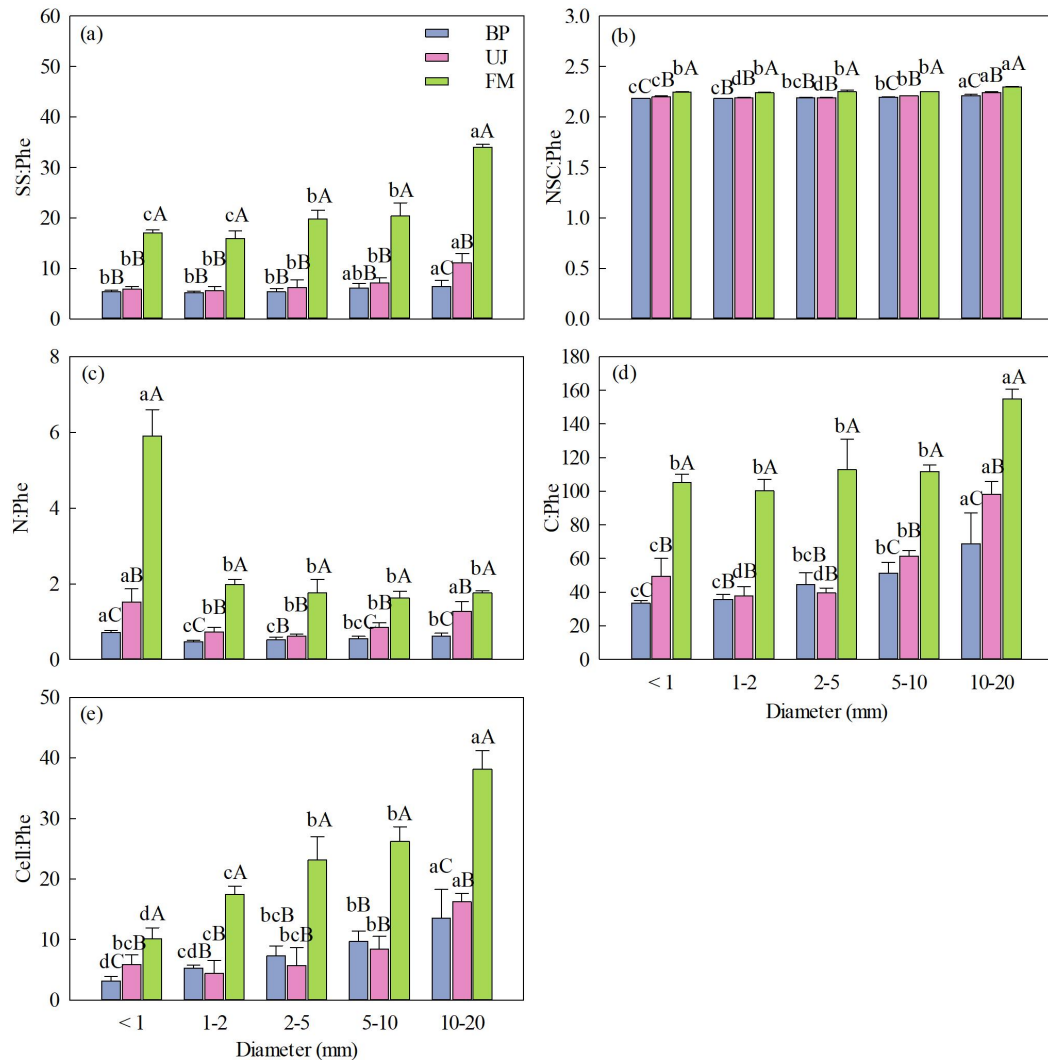
**FIGURE S12** Comparison of initial resistant carbon fractions in roots with different diameters for the three species. BP, *Betula platyphylla*, UJ, *Ulmus davidiana* var. *japonica*, FM, *Fraxinus mandshurica*. Cell, Cellulose; Lign, Lignin; SC, Structural carbohydrates; Phe, Phenolics; RC, Resistant carbohydrates.



**FIGURE S13** Comparison of initial element concentrations in roots with different diameters for the three species

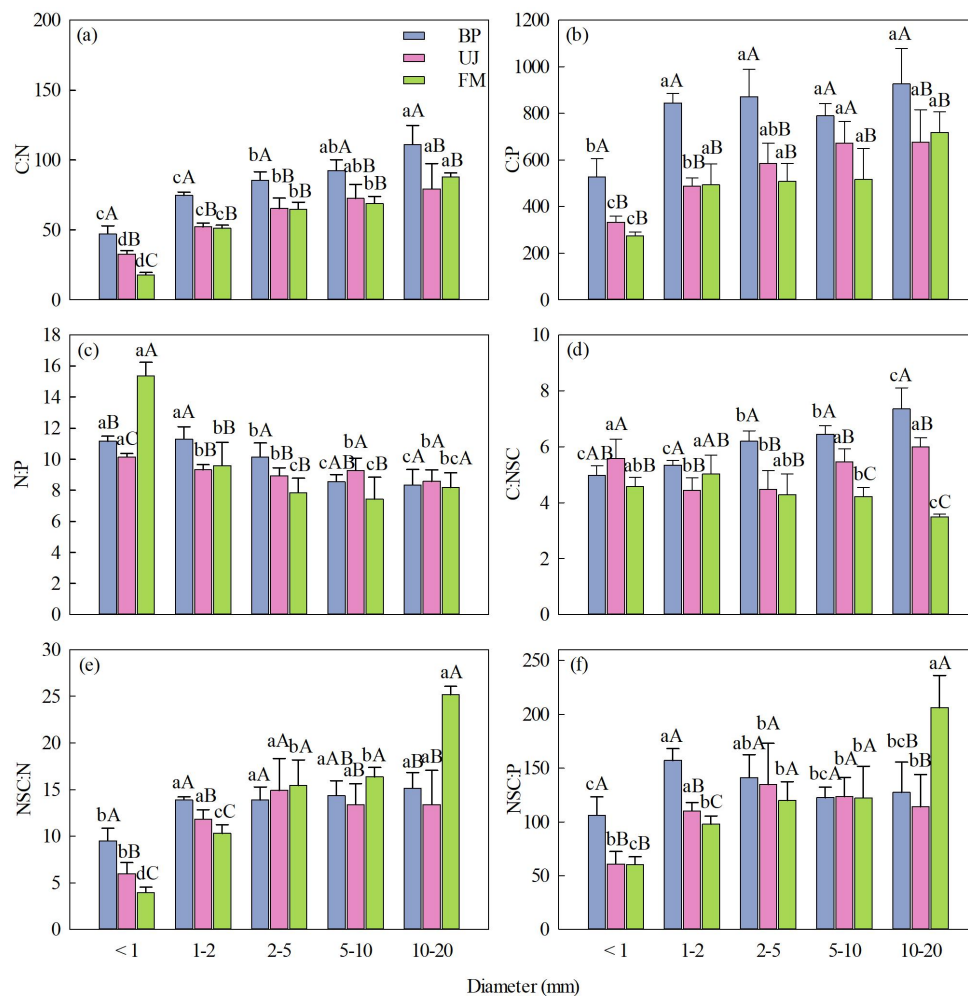
BP, *Betula platyphylla*, UJ, *Ulmus davidiana* var. *japonica*, FM, *Fraxinus mandshurica*. C, Carbon; N, Nitrogen; P, Phosphorus; Ca, Calcium; Mg, Magnesium; Si, Silicon.



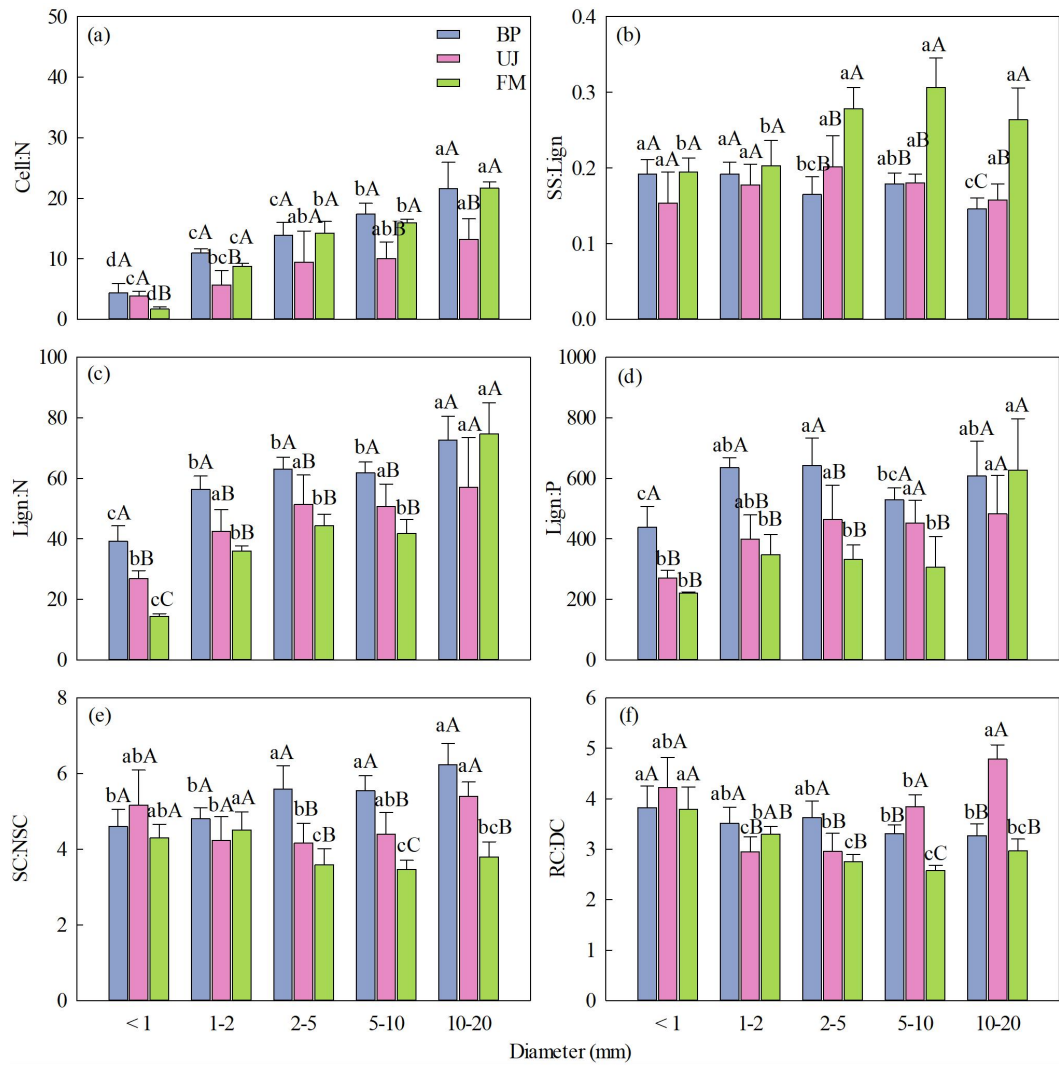


**FIGURE S14** The ratio of initial elements and carbon quality to phenolics. BP, *Betula platyphylla*, UJ, *Ulmus davidiana* var. *japonica*, FM, *Fraxinus mandshurica*. SS, Soluble sugars; NSC, Non-structural carbohydrates; Cell, Cellulose; Phe, Phenolics; C, Carbon; N, Nitrogen.

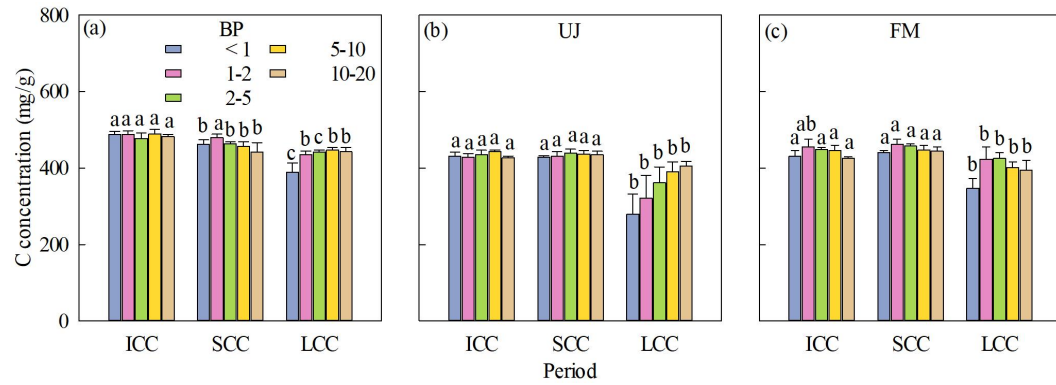




**FIGURE S15** The ratio between initial carbon, nitrogen, phosphorus, and NSC. BP, *Betula platyphylla*, UJ, *Ulmus davidiana* var. *japonica*, FM, *Fraxinus mandshurica*. NSC, Non-structural carbohydrates; C, Carbon; N, Nitrogen; P, Phosphorus.



**FIGURE S16** The ratio of initial nitrogen, phosphorus, and carbon quality to carbon quality among root diameters of the three species. BP, *Betula platyphylla*, UJ, *Ulmus davidiana* var. *japonica*, FM, *Fraxinus mandshurica*. SS, Soluble sugars; NSC, Non-structural carbohydrates; DC, Decomposable carbohydrates; Cell, Cellulose; Lign, Lignin; SC, Structural carbohydrates; RC, Resistant carbohydrates; N, Nitrogen; P, Phosphorus.



**FIGURE S17** Comparison of carbon concentration in roots with different diameters for the three species at different periods. The change of C concentration in the root decomposition was small, and the change of short-term decomposition C concentration diameter of *Betula platyphylla* and *Fraxinus mandshurica* roots was not significant. BP, *Betula platyphylla*; UJ, *Ulmus davidiana* var. *japonica*; FM, *Fraxinus mandshurica*. ICC, initial carbon concentration; SCC, short-term carbon concentration; LCC, long-term carbon concentration.