Classification: Biological Sciences - Ecology

Title: Contrasting dynamics and trait controls in first-order root compared to leaf litter decomposition

Tao Sun[a,b,1], Sarah E. Hobbie[b], Björn Berg[c,d], Hongguang Zhang[e], Zhengwen Wang[a,1], and Stephan Hättenschwiler[f]

[a]Key Laboratory of Forest Ecology and Management, Institute of Applied Ecology, Chinese Academy of Sciences, Shenyang 110016, China;

[b]Department of Ecology, Evolution and Behavior, University of Minnesota, St. Paul, MN 55108, USA;

[c]Department of Forest Sciences, University of Helsinki, Helsinki 00014, Finland;

[d]Section of Biology, University of Gävle, SE-801 76 Gävle, Sweden;

[e]Laoshan Forest Station, Northeast Forestry University, Harbin 150040, China;

[f]Centre d'Ecologie Fonctionnelle et Evolutive (CEFE) UMR 5175, CNRS - Université de Montpellier - Université Paul-Valéry Montpellier - EPHE, 1919 Route de Mende, 34293 Montpellier, France

Keywords: Long-term decomposition, mycorrhizal fungi, root tips, plant-soil interactions, trait coordination

[1]To whom correspondence may be addressed: Tao Sun (Email: suntao28329@163.com, Tel.: +86-024-83970896, Fax.: +86-024-83970200) or Zhengwen Wang (E-mail: wangzw@iae.ac.cn, Tel.: +86-024-83970392, Fax.: +86-024-83970200)

Abstract

Decomposition is a key component of the global carbon (C) cycle, yet current ecosystem C models do not adequately represent the contributions of plant roots and their mycorrhizae to this process. The understanding of decomposition dynamics and their control by traits is particularly limited for the most distal first-order roots. Here we followed decomposition of first-order roots and leaf litter from 35 woody plant species differing in mycorrhizal type over six years in a Chinese temperate forest. First-order roots decomposed more slowly (k = 0.11 ± 0.01 yr^{-1}) than did leaf litter (0.35 ± 0.02 yr^{-1}), losing only 35% of initial mass on average after six years of exposure in the field. In contrast to leaf litter, non-lignin root C chemistry (non-structural carbohydrates, polyphenols) accounted for 82% of the large interspecific variation in first-order root decomposition. Leaf litter from ectomycorrhizal (EM) species decomposed more slowly than that from arbuscular mycorrhizal (AM) species, whereas first-order roots of EM species switched, after two years, from having slower to faster decomposition compared to those from AM species. The fundamentally different dynamics and control mechanisms of first-order root decomposition compared to those of leaf litter challenge current ecosystem C models, the recently suggested dichotomy between EM and AM plants, and the idea that common traits can predict decomposition across roots and leaves. Aspects of C chemistry unrelated to lignin or nitrogen, and not presently considered in decomposition models, controlled first-order root decomposition; thus, current paradigms of ecosystem C dynamics and model parameterization require revision.

Significance Statement

Decomposition of plant roots and associated fungal mutualists is a dominant process in ecosystem carbon cycles, yet is woefully understudied compared to decomposition of leaf litter, particularly for the finest order roots that have the highest turnover. In a field experiment, we compared decomposition of the finest, most distal roots and leaf litter among 35 co-occurring temperate forest species over six years. We found that decomposition rates of root tips were considerably lower than those of leaf litter and were controlled by non-lignin carbon compounds in contrast to lignin:nitrogen ratio control over leaf litter decomposition. Our study suggests that models of terrestrial carbon cycling based on aboveground patterns are inadequate to describe decomposition of the finest plant roots.

body

Introduction

Plant litter decomposition is a key process in the ecosystem carbon (C) cycle (1-4). Most of the conceptual advancements and mechanistic understanding of how litter quantity and chemistry affect C cycling are based in empirical evidence from hundreds of studies on leaf litter decomposing at the soil surface (1-3, 5, 6). This body of knowledge has converged to a paradigm of C:nitrogen (N) and lignin:N control over plant litter decomposition, and both variables are widely used in global C models (4, 7, 8). Much less is known about how roots decompose within the soil matrix (2, 3, 9-12, 13), and whether the litter traits that influence leaf litter similarly influence root decomposition, or how well coordinated these influential traits are across leaves and roots (10, 14, 15). Because root-derived C may dominate the soil C pool (16), these are critical knowledge gaps in the current understanding of decomposition dynamics, soil organic matter formation, and the robustness of leaf-derived litter quality traits in ecosystem C models.

Fine roots are the belowground plant organs with the highest production and turnover rates (17). Their residence time in soil can thus have a major impact on soil C balance. However, decomposition of fine roots is much less studied than that of leaf litter, with conflicting results on root trait control over decomposition (17, 18). For example, a meta-analysis showed that fine root C:N ratio and Ca concentration were the traits most closely linked to root decomposition rates globally (9). However, other studies observed that neither initial C:N, N concentration, or Ca concentration were correlated with fine root decomposition rates (10-12, 19). Such inconsistencies among past studies likely arose in part because of the methods used to study root decomposition. In most root decomposition studies, roots were separated into

diameter size classes, arbitrarily defining fine roots as those less than 2 mm in diameter (17, 18). “Fine roots” defined by the 2 mm diameter threshold include unknown, species-specific proportions of different root orders varying vastly in function, morphology and tissue chemistry (11, 12, 17-20). Such variability hinders the interpretation of interspecific comparisons of root traits, how traits relate to decomposition, and how interspecific differences in root decomposition compare to those of leaves.

As the primary interface with mycorrhiza, the most distal and finest first-order roots, or root tips function similarly across species to capture nutrients and water (21). Similar to leaves, the primary light and CO_2 capturing structures, first-order roots have high production and turnover rates (17, 22). Thus, they are particularly important for root decomposition dynamics; however, they are rarely distinguished from higher order roots using the predominant root diameter-based approach. By specifically considering first-order roots, a recent study showed a clear decoupling of the global organization of functional root traits from that of the leaf economics spectrum (23), in contrast to other studies that did not distinguish explicitly among root orders (24, 25). While the leaf economics spectrum identifies increasing leaf [N] (associated with increasing specific leaf area (SLA) and decreasing leaf life span) as the major axis of functional trait variation at a global scale (26), root diameter drives first-order root trait variation, with only a minor role for interspecific differences in [N] (23). The ecosystem consequence of these contrasting patterns in trait variation between leaves and first-order roots for decomposition is currently unknown, because of the extreme paucity of data on first-order root decomposition.

Plant and fungal tissues are difficult to separate in first-order roots, and they decompose as an entity within the soil matrix. Recent studies suggest that the type of

mycorrhizal association determines C and nutrient cycling to an important degree (27), and it was shown that forests dominated by arbuscular mycorrhizal (AM) and ectomycorrhizal (EM) plants may differ in their soil C stocks, but not in a consistent manner (28-31). This difference may partly result from distinct decomposition dynamics of roots colonized with EM fungi (32), because the intense hyphal layering around EM roots potentially modifies the overall quality more than the internal and less massive structures of AM. However, decomposition of first-order roots of EM compared to AM species has not been studied in detail across a wide variety of co-occurring plant species. The distinct nutrient acquisition strategies of EM and AM plants also are associated with differences in leaf litter quality, resulting in slower leaf litter decomposition of EM than AM tree species (27, 33, 34). How such differences relate to those of mycorrhizal root decomposition of the same species at relevant temporal scales of multiple years under field conditions is at present unknown. It is also unclear which first-order root traits would drive such differences and whether they mirror those that drive leaf litter decomposition. This uncertainty critically limits the understanding of the relative importance of root and leaf litter decomposition in ecosystem C dynamics and nutrient cycling and its predictability with ongoing global change and species range shifts.

Here we compared long-term (6 years) *in situ* decomposition dynamics of leaf litter and first-order roots (as opposed to a fixed diameter cutoff) across 35 co-occurring woody species of a temperate forest ecosystem (*Materials and Methods* and Table S1). We specifically accounted for mycorrhizal type and its impact on leaf and first-order root decomposition by including nearly equal numbers of EM and AM plant species (Table S1). By measuring a large number of leaf and first-order root traits (31 morphological and chemical traits), we tested the hypothesis that

decomposition of leaf litter and first-order roots are controlled by the same set of initial traits. Specifically, we expected that decomposition would proceed more rapidly with increasing initial N concentrations in both leaf litter and first-order roots (35-37). Our second major hypothesis was that both leaf litter and first-order roots produced by EM plant species would decompose more slowly than those produced by AM plant species.

Results and Discussion

With unprecedented taxonomic breadth and temporal scale, our study showed that the so far largely neglected finest and most short-lived roots of woody plants decomposed at substantially lower rates than leaves, and that these decomposition rates correlated with entirely different sets of traits in first-order roots and leaf litter. The role of mycorrhizal type differed significantly in leaf litter decomposition, but not in first-order root decomposition.

Slower first-order root than leaf litter decomposition. Across all 35 woody plant species we found an average 23% of leaf litter mass remaining after six years of decomposition in the field (Fig. 1). In contrast, a distinctly larger amount (65%) of initial first-order root mass remained on average (Fig. 1). A single-exponential decomposition model provided a better fit for first-order root mass remaining across the eight consecutive harvests than the double-exponential or asymptotic model. In contrast, for leaf litter decomposition, the asymptotic model was the best fit or was equally as good as the single-exponential model across all species, while the double-exponential model showed poorer fits. Species-specific decomposition rate constants (k) calculated from single-exponential model fits differed by a factor of 3.8 and of 3.4 in leaf litter and first-order roots, respectively (Fig. 2). The reported range

of *k*-values for leaf litter decomposition and its mean (0.34 ± 0.02 yr^{-1}) compared relatively well with those from European or American temperate forests sharing some of the same genera of woody plants (5, 38, 39). In contrast, the *k*-values of first-order root decomposition (0.11 ± 0.01 yr^{-1} on average across all species) were considerably lower than those found in earlier studies (9, 10). However, most previous work measured decomposition of bulk fine roots with a diameter < 2 mm. These roots typically contain several root orders varying strongly in structure, lifespan, physiological activity, and chemical composition (17). The very few existing studies comparing decomposition across different orders of fine roots found decreasing mass loss rates with decreasing root order (11, 12, 40, 41). The reported mean *k*-values from combined first- and second-order roots in these studies ranged between 0.011 and 0.10 covering roughly the lower half of the *k*-values reported here (Fig. 2).

Collectively, the evidence indicates that the most distal roots are the most slowly decomposing root fraction, despite their small size, short lifespan, and comparatively high nutrient concentrations (Table S2). When we fit an asymptotic decay model, the resulting asymptote indicated an average limit value for first-order root decomposition of 38% mass loss, compared to 85% for leaf litter decomposition. In other words, almost two-thirds of total first-order root biomass contributed to a fraction of very slowly decomposing organic matter. We estimate that roughly 39 g C m^{-2} enters this fraction in the top 10 cm of soil each year, based on a first-order root turnover rate of 1.37 yr^{-1} (calculated by the generalized model of fine root lifespan (18) and the model parameters measured in this study), standing crop of first-order root biomass of 98 g m^{-2} (top 10 cm of soil) at our study site, and a mean C concentration of 46.4%. In comparison, leaf litter may contribute roughly 23 g C m^{-2} yr^{-1}, based on an average annual leaf litter fall of 309 g m^{-2} at our study site and a mean leaf litter C

concentration of 49.5%. This illustrates the significance of first-order roots for the ecosystem C cycle. Based solely on mass loss data, however, it is difficult to infer how decomposition of fresh detritus translates into the formation of soil organic matter (SOM) and its longer-term persistence. Readily decomposed litter may be transformed via microbial uptake and production of residues into more stable soil SOM, whereas the mean residence time of more slowly decomposing litter once it becomes SOM likely depends on the potential for it to become physically or chemical protected (42, 43). If the potential for physical and chemical protection of root-derived C is high relative to leaf-derived C because of its immediate proximity to soil minerals, fungal hyphae, live roots, microbial polysaccharides, and other factors that promote sorption and aggregate formation, the differences in the slower mean residence time of root- vs. leaf-derived C could be accentuated once it becomes SOM. Although beyond the scope of the present study, it would be important to test this hypothesis in future experiments, for example by following the fate of root tip versus leaf litter C using a stable isotope approach (44).

Distinct traits control leaf litter and first-order root decomposition. With a detailed assessment of a total of 31 different leaf and root traits, we evaluated if and how these traits correlated with interspecific decomposition rates. Both leaf litter and first-order root traits varied considerably (Table S2). Leaf litter varied particularly widely in elemental ratios such as C:phosphorus (P) ratio which ranged 7.3-fold (Table S3). First-order roots varied most strongly in morphological and architectural traits, with for example an up to 5.9-fold difference in root diameter between the species with the smallest (*Lonicera praeflorens*, 0.09 mm) and the largest (*Phellodendron amurense*, 0.53 mm) diameter roots (Table S3). Overall, the trait differences among species were poorly coordinated, especially for first-order roots, as

indicated by the low variation explained by the first two axes of a principle components analysis of all traits (Fig. S1), and by relatively few significant pairwise correlations among traits (Tables S4).

In line with the wider decomposition literature and with our hypothesis, initial lignin:N ratio showed the tightest (negative) correlation with leaf litter decomposition rates among all the traits measured (Fig. 3, Fig. S2). Significant correlations were also found for C:N ratio (negative), SLA (positive), and the concentrations of N, Mg, Mn, water-soluble compounds and lignin (all positive except for lignin, Fig. 3, Fig. S2). The overall best multiple-trait model for predicting leaf litter decomposition according to the lowest AICc scores included initial concentrations of Mg and Mn, as well as lignin:N ratio, accounting for a total of 67% of the variation in *k*-values (Fig. 4). These initial quality traits have been reported to correlate alone or in combination with other traits multiple times in many studies across different ecosystems (5, 6, 35, 36, 45, 46) and support the paradigm of lignin:N ratio control over litter decomposition (5, 37, 38). On the other hand, N also correlated negatively with species-specific limit values of decomposition (i.e. the asymptote of the asymptotic decomposition model). This means that leaf litter with a low initial lignin:N ratio produced a higher fraction of slowly decomposing organic matter in the late stages of decomposition despite of a high *k*-value, consistent with a growing number of long-term decomposition studies (46-48).

In strong contrast, however, neither lignin:N ratio, C:N ratio, nor the concentrations of lignin, N, or of any other measured nutrients correlated with first-order root decomposition (Fig. 3 and Fig. S2). The very few previous decomposition studies that separated at least the two lowest orders of fine roots from the bulk of "< 2 mm fine roots" also observed no (12) or even negative correlations

(11, 40) with initial N concentrations. Our results with a much larger set of species expand on these previous studies, questioning the generality of N-associated trait control of decomposition when extended to the critically underrepresented low order roots. High root N concentrations may not stimulate decomposition because N was not limiting to microbial decomposers given the much narrower mean C:N ratio of 21.4 in first-order roots compared to that typically measured in leaf litter (35, 36). This value is actually very close to that measured in the surface soil of our studied forest (12.8) and is rather below the threshold of 20 to 30, above which microorganisms are thought to be N limited (49, 50), and towards which different leaf litter types tend to converge in their final stages of decomposition (38).

Nitrogen concentration also did not emerge as a major driver of trait variation in the first comprehensive analysis of exclusively first-order roots from 369 plant species (23). Instead, root diameter was the most important trait structuring interspecific variation (23). Despite great variation in root diameter and other morphological traits among the study species (Table S2), those traits did not explain any variation in *k*-values of decomposing first-order roots (Fig. S2). This result is surprising and points to a disconnect between traits selected for during evolutionary history and those relevant for afterlife effects on ecosystem functioning, at least among the species studied here. Whether this disconnect holds across more species and biomes is unknown.

Unexpectedly, other aspects of root C chemistry, besides lignin, correlated well with *k* of first-order root decomposition. For example, *k* increased with increasing concentrations of initial non-structural carbohydrates (NSC) and decreased with increasing concentrations of bound phenolics and condensed tannins (Fig. 3). Root N concentration did not correlate with initial C chemistry (Table S4), which allowed

separating the effects of root N and C quality on decomposition. Similarly, different aspects of root C chemistry were poorly correlated with one other (Table S4). The overall best multiple-trait model for predicting first-order root decomposition included initial concentrations of NSC, total phenolics, bound phenolics and CT, together accounting for 82% of the observed variation in decomposition (Fig. 4). The apparent strong effects of C-chemistry over first-order root decomposition, suggest that substrate C quality controls root microbial decomposers in the studied temperate forest, while microbial decomposers in the litter layer are rather controlled by N availability. These distinct controls between the soil and litter layer are in line with contrasting C versus nutrient limitations of soil and litter microbial communities suggested in recent studies (50, 51). Within the soil, microbial assimilation of labile NSC may provide the energy necessary for the production of enzymes, which then prime the degradation of more complex C compounds (52). On the other hand, bound phenolics were reported to crosslink lignins to cellulose, creating a structural barrier that limits substrate accessibility for microbes (53). Bound phenolics may occur at particularly high concentrations in first-order compared to higher order roots as was recently shown in the shrub species *Ardisia quinquegona* (54). Condensed tannins (CT) have previously been shown to negatively affect decomposition of leaf litter (45, 55), either through direct toxicity to decomposers or because of reduced nutritional quality of litter as a result of binding with dietary proteins, cell wall components or digestive enzymes (56). The mean CT concentration of 8.1% we measured here in first-order roots was much higher than that in leaf litter (1.8%, Table S2) and bulk fine roots (< 2 mm) measured in another study (57). Such high CT levels in root tips may be related to increased plant defense against herbivory in these nutrient-rich and soft tissue roots (58).

Our six-year study clearly showed that distinct traits control leaf litter and first-order root decomposition across the same 35 co-occurring species, with no trait overlap in the respective best multiple-trait models. Moreover, the traits predicting either leaf litter or first-order root decomposition were not correlated (Table S6), and *k*-values also showed no correlation between leaf litter and first-order roots (Fig. 5). Collectively, these findings do not support the existence of coordinated traits and decomposition between leaves and roots contrary to what has been suggested previously for predominantly herbaceous species (13, 59, 60). Our results are in line with the few experiments comparing leaf and root decomposition of tree species (10, 15), which may suggest that trees differ from herbaceous species, possibly due to the different structure of roots and mycorrhizal associations.

The role of mycorrhizal type as driver of decomposition. Leaf litter from AM plants had significantly higher N concentrations, and lower lignin concentrations, lignin:N and lignin:P ratios than that of EM plants, but none of the other leaf litter traits differed significantly between mycorrhizal types (Table S7). Four out of the 19 EM species were conifers, but the trait differences between AM and EM species were the same regardless whether or not gymnosperms were included in the analysis. Multivariate analysis also did not show any clustering of gymnosperms (Fig. S1). Accordingly, and in support of our initial hypothesis, the mean *k*-value of AM leaf litter (0.42 ± 0.03) was 62% higher than that of EM leaf litter (0.26 ± 0.02, $P < 0.001$; Fig. 2). This result is consistent with previous studies that also documented faster leaf litter decomposition in AM- than EM-species (27, 34, 39).

In contrast to leaf litter, first-order root chemistry did not differ between AM- and EM-species (Table S8). Mycorrhizal colonization rate and root length were the only first-order root traits that differed according to mycorrhizal type, with AM-plants

having lower mycorrhizal colonization rate and longer roots than EM-plants (Table S8). On the other hand, several first-order root traits differed between gymnosperms and angiosperms, with gymnosperms having coarser roots with lower specific root length (SRL), higher lignin concentration, lower concentrations of N and P, and higher lignin:N ratios. Species did not cluster according to their mycorrhizal type in the trait space of first-order roots, but the gymnosperm family Pinaceae was separated from other families (Figs. S1 and S3). Contrary to our initial hypothesis, the mean *k*-values of AM roots (0.12 ± 0.01) did not differ from those of EM roots (0.11 ± 0.01, $P = 0.15$; Fig. 2). Likewise, mycorrhizal colonization rate did not explain any variation in decomposition rates across all species ($r^2 < 0.01$, $P = 0.82$; Fig. S2). *k*-values did not differ among families, or between gymnosperms and angiosperms ($P = 0.95$, mean ± SE of gymnosperms and angiosperms were 0.09 ± 0.02, and 0.11 ± 0.01, respectively). Furthermore, phylogenetically independent contrasts suggested that root morphological traits (e.g, diameter, length, SRL, and mycorrhizal colonization) displayed a strong phylogenetic signal (Blomberg's *K* values in Table S9), in line with a recent global-scale analysis of first-order roots (23). By contrast, neither root chemical traits nor decomposition rates were influenced by evolutionary history (Table S9).

Nevertheless, after approximately two years of exposure, the initially slower EM-root decomposition switched to faster decomposition compared to AM-roots for the remaining four years (Fig. 1). Supplementary repeated measures analyses confirmed this change in dynamics with a significant interaction between time of exposure and mycorrhizal type on remaining litter mass ($P = 0.02$). This change in decomposition dynamics was not related to any of the measured initial root traits but may reflect changes during the process of decomposition such as the breaking up of

EM fungal sheaths improving microbial access and activity and leading to faster EM-root decomposition for example. On the other hand, the relative easily degradable chitin (61) in EM fungal mycelia perhaps can prime the decomposer community, accelerating root tissue decomposition in late stages. We also cannot rule out that other factors, such as a shift in the microbial decomposer community, caused the contrasting decomposition patters of EM and AM colonized roots early versus later in decomposition.

The relatively well-established dichotomy between EM and AM woody plants for leaf litter decomposition (27, 34, 39) seems not to generalize to first-order root decomposition, likely because of similar first-order root traits and a similar range of variation in root C chemistry between the two mycorrhizal types. The lack of correlation between mycorrhizal colonization rate and decomposition rate is in rough agreement with a recent study showing that mycorrhizal colonization either had no effects on fine root decomposition or increased root decomposition (62). It will be important to assess in future research whether our results from a Chinese temperate forest can be confirmed at other study sites and how they integrate with the general conceptual framework of different C and nutrient cycling in EM versus AM dominated ecosystems (27, 28).

Our results might have been influenced to some degree by the chosen methodology of using litterbags to assess first-order root decomposition. Using litter bags was necessary to compare decomposition among species and (1, 2) to follow the decomposition of first-order roots, as identifying and following the decomposition of first-order roots of 35 species in situ is not feasible using alternative methods such as intact cores (63). Nevertheless, we acknowledge that enclosing first-order root material within litterbags does not fully represent the conditions of naturally decaying

first-order roots because it disrupts the tight connections between the soil matrix, roots and extramatrical hyphae (13, 19, 63, 64). Such disruption could affect the mycorrhizal type-specific decomposition dynamics as EM root tips typically have much more extramatrical hyphae than AM root tips (13). Also, the mesh size of 50 µm for litterbags, necessary to avoid any ingrowth of living roots during the six years of field exposure (which would have compromised the assessment of mass loss dynamics), excluded meso- and macrofauna that contribute to decomposition potentially leading to underestimated decomposition rates in our study (3, 58). However, this should not have impacted the relative differences among species, or between first-order root and leaf litter decomposition, since we used the same mesh size for both materials. On the other hand, the use of living first-order roots instead of dead roots may have caused more rapid decomposition at least initially, because of different chemical characteristics with for example higher N and non-structural carbohydrate concentrations in live roots (13, 65). However, there is currently no adequate method to collect sufficient material of naturally dead or senescent roots that are not already decomposing. We suggest that new approaches to accurately study fine root decomposition *in situ* should be used to replace the traditional litterbags. A very promising approach was recently proposed by combining isotopic labeling with -omics techniques and imaging to precisely track the products of decomposition and study root decomposition *in situ* (13).

Conclusions

The data from this large comparative assessment of first-order root decomposition in a temperate forest ecosystem suggest that the smallest (a mean length and diameter of 4.4 mm and 0.24 mm in the studied 35 species, respectively) and most short-lived root fraction decomposes at much slower rates than leaf litter from the same species. Our

results further indicate that first-order roots do not mirror the mycorrhizal type-specific decomposition dynamics reported for leaf litter decomposition, a finding that needs integration into the predictive framework of biogeochemical cycling based on plant-mycorrhizal associations. Moreover, in later stage first-order root decomposition, the mycorrhizal pattern appears opposite to that observed for leaf litter decomposition between these two mycorrhizal types. Most importantly, in contrast to leaf litter, the large interspecific variation in first-order root decomposition cannot be predicted by the commonly used parameters like C:N or lignin:N ratio, but is predicted by C compounds of low abundance in root tissues. If confirmed for other types of ecosystems, the finding that slow first-order root decomposition is controlled by non-lignin C quality rather than lignin:N ratio changes the general understanding of ecosystem C cycling and suggests that models of the global C cycle need updating.

Materials and Methods

Experimental setup. The experiment was established in an old-growth and species-rich temperate forest in China. We used four permanent plots, each 50 × 50 m, that were set up in 2006 for studying the carbon balance of an old-growth forest. We chose 35 different woody species, mostly trees (28 species), but also a few shorter statured shrub species (seven species) that are all common in this type of temperate forest (Table S1). Besides selecting relatively abundant species, species were also selected to obtain equal representation of mycorrhizal type. Sixteen species are AM and 19 are EM.

For each individual tree or shrub, we established 1.5 × 1.5 m plots within 1 to 3 m distance from the trunk. From each plot we excavated the complete root system up to the first five orders of roots within the top 15 cm of soil in July 2008. To assure species identity we harvested only roots that could be traced back to the stem of each

405 target individual. For the identification of root order we used Strahler's stream
406 ordering system (18). All fifth-order root branches were then cut from the sixth-order,
407 larger diameter woody roots. The collected roots were put immediately on ice in a
408 cooler in the field, transported to the laboratory, and frozen at -20°C for later
409 processing. In the laboratory, we cut all of the most distal root-tips defined as
410 first-order roots (18). Although extremely time consuming, this procedure was critical
411 for a functionally meaningful comparison of the same root cohort across species (17).
412 All leaf litter and first-order roots were then oven-dried (60°C) until constant weight.
413 Both leaf litter and root bags were constructed using 50-μm mesh nylon tissue.
414 This mesh size allowed the passage of fungal hyphae but not of larger-sized soil
415 organisms, which can contribute significantly to decomposition, especially for leaf
416 litter on the soil surface (3). The use of larger mesh sizes for litterbags was not
417 possible because it would allow ingrowth of fine roots as well as loss of decomposing
418 first-order roots from litterbags. For the sake of comparison between leaf litter and
419 first-order root decomposition we kept the same mesh size, and thus, the same
420 decomposer community structure for both materials. Approximately 8 g of leaf litter
421 and 0.2 g of first-order roots for each species were then sealed into their respective
422 bags. For the decomposition of leaf litter, 32 bags per species were placed on the
423 common soil surface in each of the four permanent old-growth forest plots in October
424 2008. Four different leaf litter bags per species and per plot were harvested in May
425 2009, October 2009, May 2010, October 2010, and in October of each following year
426 (2011-2014), yielding a total of 4 bags$\times$8 harvests$\times$4 plots (n = 4) = 128 bags in
427 total for each species. Eight root litterbags of each species were buried horizontally at
428 10 cm soil depth in each of the four plots in May 2009. The location of the eight
429 litterbags per species was selected randomly in the center of each plot to allow

sequential harvest through time while not disturbing the remaining litterbags. One of these eight root litterbags per species and per plot was harvested in July 2009, in October of each year from 2009 to 2014, and in May 2015, yielding a total of 1 bag × 8 harvests × 4 plots (n = 4) = 32 bags in total for each species. Upon harvest, decomposed leaf litter and root samples were removed from the litterbags, rinsed, dried (65°C) and weighed. We also analyzed subsamples from each harvest for ash content to calculate mass loss on an ash free dry mass basis. See *SI Materials and Methods* for more details.

Statistical analyses. We fitted the proportion of remaining ash-free leaf litter or root dry mass against time using three different models and determined the best model based on Akaike's Information Criteria (AIC; Table S9). When the difference between the minimum AIC and the AIC of other candidate model(s) was less than three, we concluded that the model with the minimum AIC and the other model(s) were indistinguishable in their abilities to fit the data. The three models used were 1) single-exponential ($X = e^{-kt}$), 2) double-exponential ($X = Ce^{-k_1 t} + (1-C)e^{-k_2 t}$), and 3) asymptotic ($X = A + (1-A)e^{-k_a t}$) decomposition models, where X is the proportion of initial litter mass remaining at time t (in years). In the single-exponential model, k is the decay constant using nonlinear least-squares fitting. In the double-exponential model, C is the fraction of the initial litter mass that decays at a decomposition rate k_1, while the remaining fraction ($1-C$) decays at a rate k_2. In the asymptotic model, A is the fraction of the initial litter mass with a decomposition rate of zero (i.e., the asymptote), while the remaining fraction (1-A) decays with decomposition rate k_a. Additional details are available in *SI Materials and Methods*.

Acknowledgments. We acknowledge M. Luke McCormack and Richard P. Phillips

for helpful comments on previous versions of the manuscript. We also acknowledge Josep Padulles Cubino for helping to test the phylogenetic analyses. The funding for this research was supported by the State Key Program of China (2016YFA0600800 and 2016YFD0300904), Natural Science Foundation of China (31500361) and Key Research Program of Frontier Sciences, CAS.

1. Swift JM, Heal OW, Anderson JM (1979) *Decomposition in Terrestrial Ecosystems.* University of California Press, Berkeley, CA.
2. Prescott CE (2010) Litter decomposition: what controls it and how can we alter it to sequester more carbon in forest soils? *Biogeochemistry* 101:133–149.
3. Berg B, McClaugherty C (2014) Plant litter - Decomposition, Humus Formation, Carbon Sequestration. Springer Verlag, Heidelberg, Berlin. pp. 317.
4. Wieder WR, Boehnert J, Bonan GB (2014) Evaluating soil biogeochemistry parameterizations in Earth system models with observations. *Glob. Biogeochem. Cycles* 28:211–222.
5. Melillo JM, Aber JD, Muratore JF (1982) Nitrogen and lignin control of hardwood leaf litter decomposition dynamics. *Ecology* 63:621–626.
6. Coûteaux MM, et al. (1998) Chemical composition and carbon mineralisation potential of Scots pine needles at different stages of decomposition. *Soil Biol Biochem* 30:583–595.
7. Moorhead DL, CurrieWS, Rastetter EB, PartonWJ, Harmon ME (1999) Climate and litter quality controls on decomposition: an analysis of modeling approaches. *Glob. Biogeochem. Cycles* 13: 575–589.
8. Adair EC, et al. (2008) Simple three-pool model accurately describes patterns of long-term litter decomposition in diverse climates. *Glob Change Biol* 14:

2636–2660.

9. Silver WL, Miya RK (2001) Global patterns in root decomposition: comparisons of climate and litter quality effects. *Oecologia* 129:407–419.
10. Hobbie SE, Oleksyn J, Eissenstat DM, Reich PB (2010) Fine root decomposition rates do not mirror those of leaf litter among temperate tree species. *Oecologia* 162:505–513.
11. Goebel M, et al. (2011) Decomposition of the finest root branching orders: linking belowground dynamics to fine-root function and structure. *Ecol Monogr* 81:89–102.
12. Xiong Y, Fan P, Fu S, Zeng H, Guo D (2013) Slow decomposition and limited nitrogen release by lower order roots in eight Chinese temperate and subtropical trees. *Plant Soil* 363:19–31.
13. Beidler KV, Pritchard SG (2017) Maintaining connectivity: understanding the role of root order and mycelial networks in fine root decomposition of woody plants. *Plant Soil* 420(1-2):19–36.
14. Birouste M, Kazakou E, Blanchard A, Roumet C (2012) Plant traits and decomposition: are the relationships for roots comparable to those for leaves? *Ann Bot* 109:463–472
15. Ma C, Xiong, Y, Li L, Guo D (2016) Root and leaf decomposition become decoupled over time: implications for below- and above-ground relationships. *Funct Ecol* 30:1239–1246.
16. Clemmensen K, et al. (2013) Roots and associated fungi drive long-term carbon sequestration in boreal forest. *Science* 339:1615–1618.
17. McCormack ML, et al. (2015) Redefining fine roots improves understanding of below-ground contributions to terrestrial biosphere processes. *New Phytol* 207:

505–518.

18. Pregitzer KS, et al. (2002) Fine root architecture of nine North American trees. *Ecol Monogr* 72:293–309.
19. Sun T, et al. (2013) Further evidence for slow decomposition of very fine roots using two methods: litterbags and intact cores. *Plant Soil* 366, 633–646.
20. McCormack ML, et al. (2012) Predicting fine root lifespan from plant functional traits in temperate trees. *New Phytol* 195: 823–831.
21. Guo D, et al. (2008) Anatomical traits associated with absorption and mycorrhizal colonization are linked to root branch order in twenty-three Chinese temperate tree species. *New Phytol* 180:673–683.
22. Eissenstat DM, Wells C, Yanai R, Whitbeck J (2000) Research view: Building roots in a changing environment: Implications for root longevity. *New Phytol* 147(1):33–42.
23. Ma Z, et al. (2018) Evolutionary history resolves global organization of root functional traits. *Nature* 555:94–97.
24. Roumet C, et al. (2016) Root structure-function relationships in 74 species: evidence of a root economics spectrum related to carbon economy. *New Phytol* 210:815-826.
25. Valverde-Barrantes OJ, Freschet GT, Roumet C, Blackwood CB (2017) A worldview of root traits: the influence of ancestry, growth form, climate and mycorrhizal association on the functional trait variation of fine-root tissues in seed plants. *New Phytol* 215:1562-1573.
26. Wright IJ, et al. (2004) The worldwide leaf economics spectrum. *Nature* 428:821-827.
27. Phillips RP, Brzostek E, Midgley MG (2013) The mycorrhizal‐associated

528 nutrient economy: a new framework for predicting carbon–nutrient couplings in
529 temperate forests. *New Phytol* 199:41–51.
530 28. Averill C, Turner BL, Finzi AC (2014) Mycorrhiza-mediated competition
531 between plants and decomposers drives soil carbon storage. *Nature* 505:543–545.
532 29. Craig ME, et al. (2018) Tree mycorrhizal type predicts within‐site variability in
533 the storage and distribution of soil organic matter. *Glob Change Biol* DOI:
534 10.1111/gcb.14132.
535 30. Jo I, Potter KM, Domke GM, Fei S (2018) Dominant forest tree mycorrhizal type
536 mediates understory plant invasions. *Ecol Lett* 21(2):217-224.
537 31. Zhu K, et al. (2018) Association of ectomycorrhizal trees with high carbon‐to‐
538 nitrogen ratio soils across temperate forests is driven by smaller nitrogen not
539 larger carbon stocks. *J Ecol* 106(2):524-535.
540 32. Langley JA, Chapman SK, Hungate BA (2006) Ectomycorrhizal colonization
541 slows root decomposition: the post‐mortem fungal legacy. *Ecol Lett* 9:955–959.
542 33. Cornelissen JHC, Aerts R, Cerabolini B, Werger MJA, van der Heijden MGA
543 (2001) Carbon cycling traits of plant species are linked with mycorrhizal strategy.
544 *Oecologia* 129: 611–619.
545 34. Lin G, McCormack ML, Ma C, Guo D (2017) Similar below‐ground carbon
546 cycling dynamics but contrasting modes of nitrogen cycling between arbuscular
547 mycorrhizal and ectomycorrhizal forests. *New Phytol* 213:1440–1451.
548 35. Aerts R (1997) Climate, leaf litter chemistry and leaf litter decomposition in
549 terrestrial ecosystems: a triangular relationship. *Oikos* 79:439–449.
550 36. Cornwell WK, et al. (2008) Plant species traits are the predominant control on
551 litter decomposition rates within biomes worldwide. *Ecol Lett* 11:1065–1071.
552 37. Freschet GT, Aerts R, Cornelissen JH (2012) Multiple mechanisms for trait

553 effects on litter decomposition: moving beyond home-field advantage with a new
554 hypothesis. *J Ecol* 100:619–630.
555 38. Moore TR, et al. (1999) Litter decomposition rates in Canadian forests. *Glob*
556 *Change Biol* 5:75–82.
557 39. Hobbie SE, et al. (2006) Tree species effects on decomposition and forest floor
558 dynamics in a common garden. *Ecology* 87:2288–2297.
559 40. Fan P, Guo D (2010) Slow decomposition of lower order roots: a key mechanism
560 of root carbon and nutrient retention in the soil. *Oecologia* 163:509–515.
561 41. Sun T, Dong L, Zhang L et al (2016) Early stage fine-root decomposition and its
562 relationship with root order and soil depth in a *Larix Gmelinii* plantation. *Forests*
563 7:234.
564 42. Schmidt MWI, et al. (2011) Persistence of soil organic matter as an ecosystem
565 property. *Nature* 478:49–56.
566 43. Cotrufo MF, Wallenstein MD, Boot CM, Denef K, Paul E (2013) The Microbial
567 Efficiency-Matrix Stabilization (MEMS) framework integrates plant litter
568 decomposition with soil organic matter stabilization: do labile plant inputs form
569 stable soil organic matter? *Glob Change Biol* 19:988-995.
570 44. Cotrufo MF, et al. (2015) Formation of soil organic matter via biochemical and
571 physical pathways of litter mass loss. *Nat. Geoscience* 8:776-779.
572 45. Makkonen M, et al. (2012) Highly consistent effects of plant litter identity and
573 functional traits on decomposition across a latitudinal gradient. *Ecol Lett*
574 15:1033–1041.
575 46. Berg B (2014) Decomposition patterns for foliar litter–a theory for influencing
576 factors. *Soil Biol Biochem* 78:222–232.
577 47. Berg B (2000) Litter decomposition and organic matter turnover in northern

578 forest soils. *Forest Ecol Manag* 133:13–22.
579 48. Hobbie SE (2015) Plant species effects on nutrient cycling: revisiting litter
580 feedbacks. *Trends Ecol Evol* 30:357–363.
581 49. Kaye JP, Hart SC (1997) Competition for nitrogen between plants and soil
582 microorganisms. *Trends Ecol Evol* 12:139–143.
583 50. Fanin N, et al. (2011) Distinct microbial limitations in litter and underlying soil
584 revealed by carbon and nutrient fertilization in a tropical rainforest. *PLoS One* 7.
585 51. Mooshammer M, et al. (2014) Adjustment of microbial nitrogen use efficiency to
586 carbon:nitrogen imbalances regulates soil nitrogen cycling. *Nat Commun* 5.
587 52. Fontaine S, et al. (2007) Stability of organic carbon in deep soil layers controlled
588 by fresh carbon supply. *Nature* 450:277–280.
589 53. Austin AT, Ballaré CL (2010) Dual role of lignin in plant litter decomposition in
590 terrestrial ecosystems. *Proc Natl Acad Sci* 107:4618–4622.
591 54. Wang JJ, Tharayil N, Chow AT, Suseela V, Zeng H (2015) Phenolic profile
592 within the fine-root branching orders of an evergreen species highlights a
593 disconnect in root tissue quality predicted by elemental-and molecular-level
594 carbon composition. *New Phytol* 206:1261–1273.
595 55. Coq S, Souquet JM, Meudec E, Cheynier V, Hättenschwiler S (2010)
596 Interspecific variation in leaf litter tannins drives decomposition in a tropical rain
597 forest of French Guiana. *Ecology* 91:2080–2091.
598 56. Coulis M, Hättenschwiler S, Rapior S, Coq S (2009) The fate of condensed
599 tannins during litter consumption by soil animals. *Soil Biol Biochem*
600 41:2573–2578.
601 57. Xia M, Talhelm AF, Pregitzer KS (2015) Fine roots are the dominant source of
602 recalcitrant plant litter in sugar maple-dominated northern hardwood forests. *New*

603 *Phytol* 208:715–726.
604 58. Stevens GN, Jones RH (2006) Patterns in soil fertility and root herbivory interact
605 to influence fine-root dynamics. *Ecology* 87:616–624.
606 59. Liu GF, et al. (2010) Coordinated variation in leaf and root traits across multiple
607 spatial scales in Chinese semi-arid and arid ecosystems. *New Phytol* 188,
608 543–553.
609 60. Freschet GT, et al. (2013) Linking litter decomposition of above ‐ and below ‐
610 ground organs to plant–soil feedbacks worldwide. *J Ecol* 101: 943–952.
611 61. Fernandez CW, Koide RT (2012) The role of chitin in the decomposition of
612 ectomycorrhizal fungal litter. *Ecology* 93:24–28.
613 62. Koide RT, Fernandez CW, Peoples MS (2011) Can ectomycorrhizal colonization
614 of *Pinus resinosa* roots affect their decomposition? *New Phytol* 191(2):508-514.
615 63. Dornbush ME, Isenhart TM, Raich JW (2002) Quantifying fine root
616 decomposition: an alternative to buried litterbags. *Ecology* 83:2985–2990.
617 64. Li A, et al. (2015) Fine root decomposition, nutrientmobilization and fungal
618 communities in a pine forest ecosystem. *Soil Biol Biochem* 83:76–83.
619 65. Kunkle JM, Walters MB, Kobe RK (2009) Senescence-related changes in
620 nitrogen in fine roots:mass loss affects estimation. *Tree Physiol* 29:715–723.