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Mean age of carbon in fine roots from temperate forests and grasslands with different management

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Abstract

Fine roots are the most dynamic portion of a plant's root system and a major source of soil organic matter. By altering plant species diversity and composition, soil conditions and nutrient availability, and consequently belowground allocation and dynamics of root carbon (C) inputs, land-use and management changes may influence organic C storage in terrestrial ecosystems. In three German regions we measured fine root radiocarbon (^{14}C) content to estimate the mean time since C in root tissues was fixed from the atmosphere in 54 grassland and forest plots with different management and soil conditions. Although root biomass was on average greater in grasslands 5.1 ± 0.8 g (mean ± SE, $n = 27$) than in forests 3.1 ± 0.5 g ($n = 27$), the mean age of C in fine roots in forests averaged 11.3 ± 1.8 yr and was significantly older and more variable compared to grasslands 1.7 ± 0.4 yr. We further found that management affects the mean age of fine root C in temperate grasslands mediated by changes in plant species diversity and composition. Fine root mean C age is positively correlated to plant diversity ($r = 0.65$) and to the number of perennial species ($r = 0.77$). In temperate grasslands the mean age of fine root C is also influenced by the study region mainly driven by differences in soil characteristics and climate which reflect in plant composition variations, with averages of 0.7 ± 0.1 yr ($n = 9$) on mostly organic sandy soils in northern Germany and of 1.8 ± 0.3 yr ($n = 9$) and 2.6 ± 0.3 ($n = 9$) in more silty and clayey soils respectively in central and southern Germany. Our results indicate an internal redistribution of C in perennial species and suggest linkages between fine root C age and management in grasslands. These findings improve our ability to predict and model belowground C fluxes across broader spatial scales.

1 Introduction

In terrestrial ecosystems, fine plant roots (defined here as < 2 mm in diameter) play an important role in biogeochemical cycling (Brunner and Godbold, 2007). The flux of

C from plants to soil includes allocation to grow and maintain roots, and to support supply of C to the rhizosphere. Fine roots are considered to be the most dynamic part of the root system and to control the acquisition of water and nutrients from the soil (Lukac, 2012). However, estimating the amount of C allocated in fine roots and the turnover times of root systems are still challenges in ecology. Furthermore, our ability to directly observe and quantify roots in situ is limited (Majdi et al., 2005; Trumbore and Gaudinski, 2003)

Estimates of C allocation to root growth and maintenance have been based on the assumption, derived from the root standing stock and belowground C fluxes, that fine roots turn over approximately annually (Jackson et al., 1997). However, measurements of isotopes in fine roots demonstrated that both the ^{14}C age (Gaudinski et al., 2001) and the incorporation rate of a continuous ^{13}C label (Matamala et al., 2003) in fine root C were inconsistent with an annual turnover. The various observations can be reconciled by assuming that fine roots are not a single homogeneous pool (Gaudinski et al., 2010; Guo et al., 2008; Strand et al., 2008; Tierney and Fahey, 2002; Trumbore, 2009). To date, published fine root radiocarbon (^{14}C) data are mostly from forest ecosystems and little is known of how these observations apply to non-forested ecosystems, or a range of forest types (Fröberg, 2012; Gaudinski et al., 2010; Riley et al., 2009). Identifying predictors of fine root C age is of specific importance to develop indices of root dynamics and modelling efforts given the pressing need to improve our knowledge about belowground C fluxes (Ostle et al., 2009).

Management intensification and land-use changes are considered to be the main processes eroding species diversity (Laliberté et al., 2010). Changes in plant species can influence ecosystem nutrient dynamics by a variety of mechanisms including biomass production, decomposition and nutrient cycling (Hättenschwiler et al., 2005; Hobbie, 1992; Tilman et al., 1996). However, the mechanisms by which management-driven alterations of plant species diversity, and soil conditions including moisture, pH and nutrient availability can affect mean ages of fine root C and belowground C cycling remain unclear (Schmidt et al., 2011). For example, older roots may occur in less fertile sites because plants invest less in new tissues than plants in nutrient-rich sites (Chapin et al., 2002). Thus, differences in the mean age of root C between trees and grasses, or perennial and annual plants, could reflect differences in plant resource conservation and acquisition strategies between species or functional types. These in turn can reflect differences in root anatomy, chemistry and architecture, and the capacity for internal recycling of C in perennial root systems. Predicting ecosystem responses to environmental change requires understanding of root dynamics from a range of vegetation covers representing forests and grasslands across a large spatial scale and over a range of managements and soil types. To address some of these questions, plant species may be classified according to their ecological behavior (i.e. Ellenberg's ecological indicator values; Ellenberg et al., 2001), to summarize environmental factors like soil moisture and available nitrogen (N) content. However, those factors such as moisture and the available N content in the soil can be altered by management and change during the growing season.

Radiocarbon (^{14}C) measurements of roots are a useful measure for understanding belowground carbon fluxes (Majdi et al., 2005). ^{14}C is a radioactive isotope (half-life 5730 yr) which is naturally present in the atmosphere. Thermonuclear weapons explosions in the atmosphere also produce radiocarbon and during the 1950s and early 1960s nearly doubled the amount in the Northern Hemisphere atmosphere. Since the nuclear test ban treaty in 1963, atmospheric ^{14}C values have been declining through atmospheric mixing, incorporation into terrestrial and aquatic C pools and dilution by combustion of ^{14}C free fossil fuels. The documented time history of atmospheric "bomb" ^{14}C provides a global isotope tracer of the carbon cycle (Naegler and Levin, 2006; Randerson et al., 2002). Plants fixing atmospheric CO_2 record its ^{14}C signature, once data are corrected for mass-dependent isotopic fractionation (Stuiver and Polach, 1977). The precision of ^{14}C measurements using accelerator mass spectroscopy (AMS; ± 2–3 ‰) combined with the recent rate of ^{14}C decline of ~ 4–5 ‰ per year (Levin et al., 2010) enables us to use radiocarbon as a tool for determining the average time elapsed between C fixation and its incorporation into root tissues (Gaudinski et al., 2001).

Here we use ^{14}C to estimate root C age of fine roots samples in 27 grasslands and 27 forest plots with different management in three regions in Germany, with a steady state model implemented by Gaudinsky et al. (2001). Because part of the overall C age might reflect plant allocation of older carbon to the root system we use the term “fine root C age” instead of fine root age. Our main objective is to evaluate the differences in fine root C age in grassland and forest sites under diverse management, plant species diversity and soil properties. Moreover we are interested in understanding if the mean age of C in fine roots is related to fine root nutrient concentration and root biomass. We hypothesize that (i) in forest ecosystems the age of root carbon is older than in grasslands due to the greater ability of trees to use storage compounds and recycle C internally; (ii) in grasslands the management effect is reflected in the mean age of fine root C and is mediated by the total number of perennial species present; (iii) ^{14}C of fine roots differs between study regions mainly driven by differences in soil characteristics, i.e. nutrient contents, and climate.

2 Materials and methods

2.1 Study regions

The research was carried out in 54 plots distributed in three German regions, the so called Biodiversity Exploratories (Fischer et al., 2010) which comprise a variety of forests and grasslands managed with different intensities. The Schwäbische Alb (ALB) is situated in south-western Germany, the Hainich-Dün (HAI) in central Germany and the Schorfheide-Chorin (SCH) in north-eastern Germany. The three study regions differ in climate, altitude and soil characteristics (Table 1; for detail see Fischer et al., 2010).

In each region we selected 9 grassland and 9 forest plots to span a range of land-use intensities. In grasslands of the ALB and SCH we selected three plots of different land-uses: unfertilized pastures, fertilized meadows and mown pastures. The mown pastures were unfertilized in SCH and fertilized in ALB. In forests of the ALB and SCH we selected three European beech (*Fagus sylvatica* L.) dominated unmanaged stands, three European beech dominated age-class forests and three conifer plantations (Norway spruce (*Picea abies* (L.) H. Karst) in the ALB and Scots pine (*Pinus sylvestris* L.) in the SCH). For the HAI we followed a different plot-selection scheme, and selected 9 grassland and 9 forest plots following gradients of soil texture and land-use intensity. Land-use intensity in HAI grasslands was quantified as an land-use intensity index (LUI) summarizing the individual land-uses by summing up values for fertilization ($\text{kg}\,\text{N}\,\text{ha}^{-1}\,\text{yr}^{-1}$), mowing (times mown per year), and grazing intensities (livestock units per hectare per year) which have been normalized by the mean of the appropriate land-use type in order to standardize the scales (Blüthgen et al., 2012). Land-use and disturbance intensity in forests was calculated by combining values of stand density and diameter at breast height for a relatively unmanaged forest and different management schemes, in conjunction with selfthinning values (Luyssaert et al., 2011).

2.2 Soil and root sampling

Root sampling took place in early May 2011. On each plot we collected 14 mineral soil cores using a split tube sampler with a diameter of 5 cm. In the forest sites organic layers were collected and removed before coring; in grasslands, aboveground portions of plants were removed. Cores were taken along two transects which were selected always in the same relationship to the overall plot. The two transects were 20 m long in grasslands and 40 m long in forests. We then opened the core and cut a section representing a fixed sampling depth of 0 to 10 cm for further analysis. We prepared composite samples by mixing the material selected from the 14 cores. Roots were then removed from the composite sample, refrigerated at 4 °C and transported to the laboratory. Then, we removed the soil particles attached to the roots by cleaning them with distilled water in a 500 µm sieve and collected the fine roots with a diameter < 2 mm. Dead roots were removed from the < 2 mm samples based on qualitative visual characteristics, including color and breakability. Fine root samples were dried at

40 °C to constant weight in a force-air oven. The roots were stored in plastic bags at room temperature until analysis.

2.3 Biomass, C and N concentrations and C and N stocks of fine roots

We weighed the dry biomass of the fine roots after drying. Total C and N concentrations of ground fine root material were analysed using an elemental analyzer "Vario EL" (Elementar Analysensysteme GmbH, Hanau, Germany). Spruce needles (CRM 101 with 51.44 % C; 1.889 % N) were used as reference material. Root samples collected in the grasslands, which in our plots are characterized by higher pH values (5 to 7.5) than forests (3.0 to 6.0), were decalcified in order to avoid carbonate contamination. Carbon and nitrogen stocks of fine roots (0–10 cm depth) were calculated from the total carbon and nitrogen concentrations in the roots and from the dry biomass values of each sample.

2.4 Management data, vegetation survey and soil characteristics

In forests, land-use was verified with a systematic inventory of a circular sampling area of 500 m^2 (Fischer et al., 2010). In grasslands land-use intensity data was obtained from a questionnaire for all land users. Forests were not fertilized. In 2008, we recorded the vegetation in forests in spring and again in summer, and in grasslands only once in summer. Plots in grasslands were 4 m × 4 m and in forests 20 m × 20 m (in the center of each plot and concentric within the forest inventory circle). We identified all vascular plant species and estimated their percentage cover. To assess the diversity and correct cover value of vascular plant species per plot in forests, we combined the spring and summer records, always using the highest cover value of the particular species either in spring or in summer. We calculated the "Shannon Index" as a measure of plant species diversity. This index is based on an expression elaborated by Shannon for his mathematical theory of communication, where H corresponds to the entropy (Shannon, 1948) (Eq. 1):

$$H' = -\sum_{i=1}^{n} p_i \ln p_i \tag{1}$$

p_i is the percentage of the individuals represented by species i and is assessed by the quotient of number of individuals of species i (N_i) and the total number of individuals (N). Thus, the maximum diversity possible for N individuals occurs when each individual belongs to a different species. We further calculated the total number of annual and perennial species per plot. We used Ellenberg indicator values for “moisture” and “nitrogen” for each plot. These indicator values represent a measure of the realized ecological niche on an ordinal scale from 1 to 9 (Ellenberg et al., 2001) and are considered to be a valuable tool that can perform well (Schaffers and Sykora, 2000).

We also measured soil pH of the same combined mineral soil samples from which we collected the roots. Soil samples were air-dried and sieved to < 2 mm. Then, we calculated the mean of two pH measurements per soil sample which were analyzed in a 0.01 M $CaCl_2$ solution with a soil solution ratio of 1 : 2.5.

2.5 Radiocarbon measurements and root C mean age

We measured the ^{14}C content of the ground fine root samples at the accelerator mass spectrometry (AMS) facility in Jena, Germany (Steinhof et al., 2004). After combusting the samples, the resulting CO_2 was catalytically reduced to graphite at 625 °C by H_2 reduction. An aliquot of the CO_2 was used to determine the $\delta^{13}C$ of the sample. We express radiocarbon data as $\Delta^{14}C$ which is defined as the difference in parts per thousand (‰) between the $^{14}C/^{12}C$ ratio in the sample, corrected for mass dependent isotope fractionation to a common $\delta^{13}C$ value of −25 ‰, in comparison to an oxalic acid universal standard (Trumbore, 2009) (Eq. 2). The standard is corrected for decay between 1950 and the year of the measurement y, which for the present work was 2011.

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$$\Delta^{14}\mathrm{C}=\left[\frac{\left.\frac{^{14}\mathrm{C}}{^{12}\mathrm{C}}\right]_{\mathrm{sample},-25}}{0.95\left.\frac{^{14}\mathrm{C}}{^{12}\mathrm{C}}\right]_{\mathrm{Ox1},-19}\times\exp^{((y-1950)/8267)}}-1\right]\cdot 1000 \tag{2}$$

We further derived the mean C age of fine roots, which represents the time C was stored in the plant before being allocated for root growth, plus the root average lifespan. For our composite root samples we chose a steady state model implemented by Gaudinsky et al. (2001). This method includes the ^{14}C concentration of atmospheric $\mathrm{CO_2}$ over the past n yr, where n represents the average age of the root composite sample assuming that any variation in ages of the root mixture is normally distributed around the mean (Gaudinski et al., 2001). The equation we used is Eq. (3):

$$C_{(t)}\times R_{\mathrm{root}(t)}=I\times R_{\mathrm{atm}(t)}+C_{(t-1)}\times R_{\mathrm{root}(t-1)}\times(1-k-\lambda), \tag{3}$$

In this method C is the carbon stock of fine roots expressed in $\mathrm{g\,C\,m^{-2}}$, I is the input of C by new production of fine roots in $\mathrm{g\,C\,m^{-2}\,yr^{-1}}$, k is the multiplicative inverse of the mean age of fine root C age in $\mathrm{yr^{-1}}$ and t equals the time (yr) for which the calculation is being performed. R_{root} is calculated as $[\Delta^{14}\mathrm{C}_{\mathrm{root}}/1000+1]$ (Torn et al., 2009), λ is the radioactive decay constant for ^{14}C (1/8267 yr). For the time history of radiocarbon (R_{atm}) at the three sites, we used Levin et al. (2010), updated to I. Levin, personal communication, 2012. An example of the method for using ^{14}C values to estimate the mean age of C in fine roots is illustrated in Fig. 1. The atmospheric $^{14}\mathrm{CO_2}$ (R_{atm}) value for spring 2011 is equal to (1.034, or 34 ‰/1000 +1). Local release of fossil C can impact the mean ^{14}C signature at any or all of our sites, which would tend to reduce the R_{atm}, and can potentially lead to $\Delta^{14}\mathrm{C}_{\mathrm{root}}$ values lower than 34 ‰ (i.e. contemporary roots were formed in 2011 from freshly fixed photosynthetic products). The presence of fossil C signatures would serve to systematically underestimate the age of root C; since we did not encounter many data with Δ^{14}C below the current atmosphere, and mostly in the SCH grasslands, we do not think there is a large systematic bias in our results.

2.6 Statistics

We conducted all analyses with the R version 2.15.2 (R Development Core Team 2012). To examine statistical differences of biomass, C and N concentrations and stocks, ^{14}C content and mean C age of fine roots across the three study regions and between grasslands and forests we used analysis of variance (ANOVA) accompanied by Holm's test. We present data as means ± standard error. We also used linear least-squares to compare correlations among all variables. For the grassland sites we used the Cox proportional hazards test (Cox, 1972) to identify the risk ratio of different variables on fine root C mean age. We used linear regression to assess the effect of plant diversity, number of perennial species, root nutrient concentrations, Ellenberg indicator values for soil moisture and available N content on the mean age of fine roots. Prior to analysis we transformed data if necessary to meet assumptions of normality.

3 Results

3.1 Fine root biomass and C and N stocks

Fine root biomass was greater in grasslands than in the forests of HAI and SCH (Table 2), although this pattern was reversed in the ALB study region. In grasslands, fine root biomass was greatest in SCH, followed by HAI and ALB, while in the forests HAI had the highest biomass, followed by ALB and SCH. Carbon concentrations of roots were higher in the forest sites compared to the grassland sites for all study regions. Total C concentrations were slightly higher in SCH and ALB than in HAI for both grasslands and forests. Total fine root N concentrations did not differ significantly among forests and grasslands of the three study regions (Table 3). C and N stocks of roots reflected patterns of biomass (Table 2).

3.2 Fine root radiocarbon in grasslands and forests

Most ^{14}C values exceeded the contemporary atmospheric mean (34 ‰ in spring 2011), indicating that the fine root samples analysed in this study contained a detectable proportion of "bomb" carbon fixed from the atmosphere after 1964. In the forests, fine root mean ^{14}C values exceeded the contemporary atmospheric mean in all study regions by 53 to 58 ‰. In the grasslands, mean fine root ^{14}C values also exceeded the contemporary atmospheric mean by 6 ‰ in ALB and 2 ‰ in HAI, while in SCH the average root ^{14}C values were lower than atmospheric ^{14}C values for 2011 by 2 ‰.

Overall, the greatest difference in fine root Δ^{14}C values was between forests and grasslands, with higher and more variable Δ^{14}C in the forest sites, ranging from 135 ‰ to 50 ‰ (Table 4), than in grasslands with fine root ^{14}C values ranging from 49 ‰ to 26 ‰. Due to the major variation of fine root ^{14}C values, we did not encounter any significant effects of changes in plant diversity, management and root quality on the fine root carbon age in forests ($p < 0.05$).

The ^{14}C values of fine roots collected in the forest sites did not differ significantly between study regions although the variability in ALB was higher 88 ± 12 ‰ than in HAI 94 ± 6 ‰ and SCH 87 ± 7 ‰. This corresponds to mean ages of 5 to 30 yr in the ALB, 8 to 16 yr in the HAI and 6 to 16 yr in the SCH. In contrast, root ^{14}C values were less variable in grasslands and significant variations among the three study regions were detected (Fig. 2). The ^{14}C content of bulk live fine roots in the ALB grasslands had a mean value of 40 ± 2 ‰ indicating mean ages overall ranging from 1 to 5 yr, whereas the root samples from the HAI grasslands showed mean ^{14}C values of 36 ± 2 ‰ with ages ranging from 0 to 3 yr, and the SCH grasslands had the lowest ^{14}C values 32 ± 1 ‰ with ages of 0 to 1 yr.

3.3 Effect of plant diversity and number of perennial species on fine root C mean age in grasslands

In grasslands, plant species diversity (Shannon diversity) was positively correlated with the total number of perennial species (Table 5). Further, across all study regions the mean age of fine roots in grasslands increases with species diversity and with the number of perennial species ($P < 0.001$, Fig. 3). The unit risk ratios indicated that a 0.1 higher plant diversity resulted in a 9 % higher mean age of C in roots. In addition one perennial species per plot more resulted in a 12 % increase of fine root mean age (Table 6). Mean plant diversity was largest in ALB (2.3 ± 0.1), followed by HAI 2 ± 0.1 and SCH 1.6 ± 0.1. Also the average number of perennial species is higher in ALB 29 ± 4 followed by HAI 22 ± 3 and SCH 15 ± 1. Plant diversity ranged from 1.0 to 2.9 (mean: 2.0 ± 0.1) and the absolute number of perennial species present on plots ranged from 11 on sites characterized by higher soil available N content to 47 on sites characterized by lower soil available N content (mean: 22 ± 2), according to the Ellenberg indicator values. Both plant diversity and the number of perennial species also declined significantly with increasing values of Ellenberg indicators values for soil moisture content and root N concentrations ($P < 0.05$, Fig. 3). We found no strong relation between pH and biomass effect on ^{14}C age of fine roots for our grassland study sites. Fertilization, grazing and mowing activities as well as the LUI index did not directly influence fine root C mean age significantly ($P < 0.05$).

4 Discussion

4.1 Root ^{14}C age difference between grasslands and forests

Our findings of older ^{14}C in the forest fine roots than in the grassland fine roots by 9 yr on average supports our first hypothesis (Table 2). Overall the ages of fine root C were broadly consistent with earlier radiocarbon studies in forest mineral soils and in pastures (Gaudinski et al., 2001, 2010; Sah et al., 2012; Strand et al., 2008; Veldkamp, 1994). Older root C age in the forest sites may be due to higher contents of perennial root tissues or to the ability of tree species to use storage compounds and recycle C internally on a longer term compared to annual and perennial species in grasslands. Sah et al. (2012) recently concluded from their results that new live tree roots may use old carbon reserves for cellulose formation.

We further found a greater variability in ^{14}C of fine roots in forests than in grasslands. This could be attributed to major differences in root-branching and chemistry in fine tree roots compared to fine roots of herbaceous species growing in grasslands (Waisel et al., 2002). Guo et al. (2004) suggested that root age might be related to cellulose content in different root branching orders in trees. Cellulose was observed to increase from the most distal parts of the fine root system (first and second root orders) to the more proximal portions (fifth order roots) (Guo et al., 2004). Negative relations between fine root N concentrations and fine root lifespans have been also previously reported (Tjoelker et al., 2005). Furthermore, patterns of higher N and P concentrations in roots of lower order rather than higher order were observed in four tree species of temperate forests by Goebel et al. (2011). However, we did not see large differences in N content between roots of forests and grasslands, even though they differed markedly in age. Under the assumption that older mean ages of forests are due to longer turnover times rather than internal recycling of C, our observation of less root biomass in forests compared to grasslands suggests overall smaller root litter input to forest than grassland topsoils. We can however only refer to the upper 10 cm of the mineral soil and it is possible that in forests litter input to subsoils is larger than in grasslands (Jobbágy and Jackson, 2000).

4.2 Root ^{14}C age in grassland ecosystems

We confirmed our second hypothesis, by observing that the age of C in fine roots was greater on sites characterized by a higher plant diversity and total number of perennial species. This relates to the results from a number of ^{14}C pulse labeling studies by Veldkamp (1994) and Milchunas (1985) which demonstrated that C is recycled in perennial grass roots over a period of years. In contrast, annual plants are constructed almost entirely of contemporary atmospheric-derived C except what is inherited from their seed.

Although the number of fertilized plots was limited to 12 plots our results suggest a negative relation between the amount of fertilizer and the total number of perennial species present on the plots (Table 5, $p=0.06$), this in turn can reflect in variations of mean age of C in fine roots. Previous studies have already demonstrated the negative effects of fertilization on species richness (Socher et al., 2012; Stewart and Pullin, 2008; Zechmeister et al., 2003). Recently, a study by Socher et al. (2012) conducted in the same study regions has shown a loss of 19 % of the total number of vascular plant species on a grassland site with an annual fertilization input of 35 kg of N per hectare. Given the limited amount of grazed and mowed sites in our dataset, we did not find any significant effect of these management practices on root C age. We also did not find any significant correlation with soil pH, which was surprising because we expected that soil pH would vary according to different management schemes (Falkengren-Grerup et al., 2006; Birkhofer et al., 2012).

Addressing to our third hypothesis, we found that in grasslands the variation of ^{14}C age of fine roots among study regions is considerable, with older root C in ALB followed by HAI and SCH. We assume that the annual plant roots with lower than ^{14}C atmospheric values, which we mainly found in the SCH grasslands, might define the local atmospheric ^{14}C. As this effect was not great (2 ‰), we did not correct for the calculated ages. We further believe that regional differences are not due to different levels of contamination because there was no such trend in the adjacent forests. Nevertheless, we cannot exclude the possibility that these roots may have taken up extremely old carbon which was stored in the organic soils, i.e. amino acid C. The ability of herbaceous plant species to take up amino acids was demonstrated by Näsholm et al. (2000).

We did not find the same pattern in the forests, where the larger variability may overprint similar effects and induced management dissimilarities. We relate differences among study regions to variations in climate and soil properties like moisture and nutrient content, which may lead to changes in species diversity and total number of perennial species. Nutrient poor sites (i.e. ALB) could for example favour perennial species over annual species, reflecting a tradeoff between rapid acquisition of resources and conservation of resources (Tjoelker et al., 2005). Older root C age in nutrient poor sites may be explained by the necessity of plants to optimize the uptake of nutrients (Sah et al., 2012), for example by reducing carbon and nutrient expenses in fine root production.

The climate gradient across the three study regions could also reflect differences in fine root C mean age. As the mean temperatures are higher in SCH than in ALB and we collected samples in spring, plants in northern Germany may have grown a larger amount of newly grown roots than plants in southern Germany at the time of sampling. This might have resulted in a relatively larger contribution of young roots in SCH.

Furthermore, variations in perennial root tissue may also occur as a result of plant functional types or phenotypical variation in plants in reaction to different nutrient and energy limitations in the study regions, resulting in root mean age C changes. The negative correlation between root C mean age and root N content, which we found in grasslands for example, suggests that larger N contents of roots with faster turnover could be due to a greater contribution of lower order-roots. We also found that root biomass and related C stocks were greater in more fertile sites characterized by lower ^{14}C content in roots, indicating faster root turnover (i.e in SCH) than in less fertile sites showing higher ^{14}C content in roots (HAI, ALB). The largest fine root litter input to the soil therefore probably occurs in the more fertile sites with a large biomass of young roots. Whether this leads to larger soil organic carbon stocks depends on the decomposition rates.

5 Conclusions

Our observations of the mean age of fine root C from forest and grassland sites in Germany indicate that variations in the mean age of fine root C between tree vs herbaceous grassland species and annual vs perennial herbaceous species in grasslands can be associated to differences in root tissue lifetime or in resource acquisition and resource conservation strategies. Differences in plant resource acquisition and maintenance strategies in turn are reflected in the ability of recycling C internally. The mean age of root C in grasslands is affected by management-induced changes in soil moisture and available nutrients via alterations of plant species diversity and number of perennial species. This indicates that the C inputs of fine root litter to the soil are larger in more fertile sites or in sites with higher soil moisture, where we found a larger biomass of young roots. Therefore, plant diversity indices or other easy to measure parameters like the Ellenberg indicator values for “nitrogen” or “moisture” are potentially applicable by grassland ecologists and modelers to make some first speculations about the turnover time of root C in their field sites. They are therefore of particular importance for further understanding and descriptions of management influences on belowground processes to be included in climate and landscape models.

We suggest additional efforts to improve our knowledge of how the internal redistribution dynamics of C occur in perennial plant species and in roots belonging to different species and plant functional types. This would enable separation of the age of C into a recycling component and a newly grown component and allow estimation of the root lifetimes from ^{14}C data.

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Table 1. Main geographical and environmental characteristics of the three study regions.

	ALB	HAI	SCH
Location	SW Germany	Central Germany	NE Germany
Coordinates	48°26′ N 9°23′ E	51°9′ N 10°28′ E	53°0′ N 13°46′ E
Area	~ 422 km^2	~ 1300 km^2	~ 1300 km^2
Soil type forest	Cambisol (Eutric)	Luvisols	Cambisol (Dystric)
Soil type grasslands	Leptosol-Cambisol	Cambisol - Stagnosol	Histosol-Gleysol
Altitude a.s.l.	460–860 m	285–550 m	3–140 m
Annual mean temperature	6.0–7.0 °C	6.5–8.0 °C	8.0–8.5 °C
Annual mean precipitation	700–1000 mm	500–800 mm	500–600 mm

Table 2. Root biomass, C and N concentrations and C and N stocks in fine roots (< 2 mm) among different land-uses and study regions (mean ± SE).

Land use	Study region	*n*	Root biomass (g)	C conc. (%)	N conc. (%)	C stocks $(\mathrm{g\,C\,m^{-2}})$	N stocks $(\mathrm{g\,C\,m^{-2}})$
Forest	All plots	27	3.1 ± 0.5	47.2 ± 0.8	1.1 ± 0.03	51.1 ± 7.7	1.2 ± 0.2
	ALB	9	2.8 ± 0.7	48.8 ± 0.7	1.2 ± 0.1	39.7 ± 9.7	0.9 ± 0.2
	HAI	9	4.9 ± 1	42.8 ± 1.1	1.1 ± 0.04	79.9 ± 16.4	2.0 ± 0.4
	SCH	9	1.7 ± 0.4	49.9 ± 0.8	1.1 ± 0.1	33.8 ± 7.6	0.7 ± 7.6
Grasslands	All plots	27	5.1 ± 0.8	41.8 ± 0.5	1.2 ± 0.1	80.24 ± 13.4	2.4 ± 0.5
	ALB	9	1.7 ± 0.6	41.7 ± 0.7	0.9 ± 0.5	20.4 ± 3.4	0.4 ± 0.1
	HAI	9	6.2 ± 0.7	39.9 ± 1.1	1.1 ± 0.1	92.04 ± 11.9	2.4 ± 0.8
	SCH	9	7.4 ± 1.7	43.8 ± 0.4	1.6 ± 0.1	128.2 ± 29.2	4.5 ± 29.2

Table 3. ANOVA results of fine root biomass, C and N concentrations and stocks, to compare variance between study regions and differences between grasslands and forests (defined here as land-use).

	Study region			Land-use			Study region × Land-use		
Dependent variable	df	*F*	*P*	df	*F*	*P*	df	*F*	*P*
Root biomass	2	9.67	< 0.001	1	6.77	0.012	2	6.44	0.003
C concentrations	2	24.05	< 0.001	1	63.94	< 0.001	2	3.65	0.034
N concentrations	2	7.91	< 0.001	1	1.42	0.240	2	11.47	< 0.001
C stocks	2	13.42	< 0.001	1	3.52	0.001	2	6.80	0.002
N stocks	2	9.52	< 0.001	1	10.73	0.001	2	11.75	< 0.001

Table 4. Mean values of ^{14}C content and mean age of fine roots (< 2 mm) among land-use types and study regions (mean ± SE).

Land use	Study region	*n*	Fine root Δ^{14}C (‰)	Fine root mean age
Forest		27	90 ± 5	11.3 ± 1.8
	ALB	9	88 ± 12	11.9 ± 3.0
	HAI	9	94 ± 6	10.6 ± 0.9
	SCH	9	87 ± 7	10.5 ± 1.1
Grasslands		27	36 ± 1	1.7 ± 0.4
	ALB	9	40 ± 2	2.6 ± 0.3
	HAI	9	36 ± 2	1.8 ± 0.3
	SCH	9	32 ± 1	0.7 ± 0.1

Table 5. Pearson's correlation matrix between variables. Numbers in bold indicate a significant correlation ($P < 0.05$).

Trait	Plant diversity (S.I.)	Perennial species	Nitrogen (E.I.)	pH	Moisture (E.I.)	Fertilization	Root biomass	N conc. roots	C conc. roots
Root C mean age	**0.65**	**0.77**	−**0.72**	−0.20	−**0.73**	−0.33	−0.30	−**0.63**	−0.26
Plant diversity (S.I.)		**0.71**	−**0.51**	−0.19	−**0.72**	−0.25	**-0.42**	−**0.59**	−0.11
Perennial species			−**0.88**	−0.02	−**0.79**	−0.41	−0.16	−**0.64**	−0.06
Nitrogen (E.I.)				−0.06	**0.69**	0.31	−0.01	**0.54**	−0.06
pH					−0.09	0.28	**0.45**	0.24	−0.11
Mositure (E.I)						0.29	0.28	**0.60**	**0.38**
Fertilization							−0.18	0.07	−0.38
Root biomass								0.14	0.13
N conc. roots									0.24
C conc. roots									

E.I. = Ellenberg indicator values, S.I. = Shannon Index.

Table 6. Summary of proportional hazards fit. Risk ratios < 1.0 represent a positive effect on fine root mean age and risk ratios > 1.0 represent a negative effect. Results in bold indicate significant results ($P < 0.05$).

Factor	X^2	$P > X^2$	Risk ratio
Plant diversity (S.I.)	15.1	<**0.001**	0.088
Perennial species	20.8	<**0.001**	0.88
Nitrogen (E.I.)	23.1	<**0.001**	3.99
pH	0.78	0.38	–
Moisture (E.I.)	19.1	<**0.001**	3.11
Fertilization	1.34	0.24	–
Root biomass	2.1	0.14	–
N conc. roots	13.8	<**0.001**	7.3
C conc. roots	1.4	0.24	–

E.I. = Ellenberg indicator values, S.I. = Shannon Index.

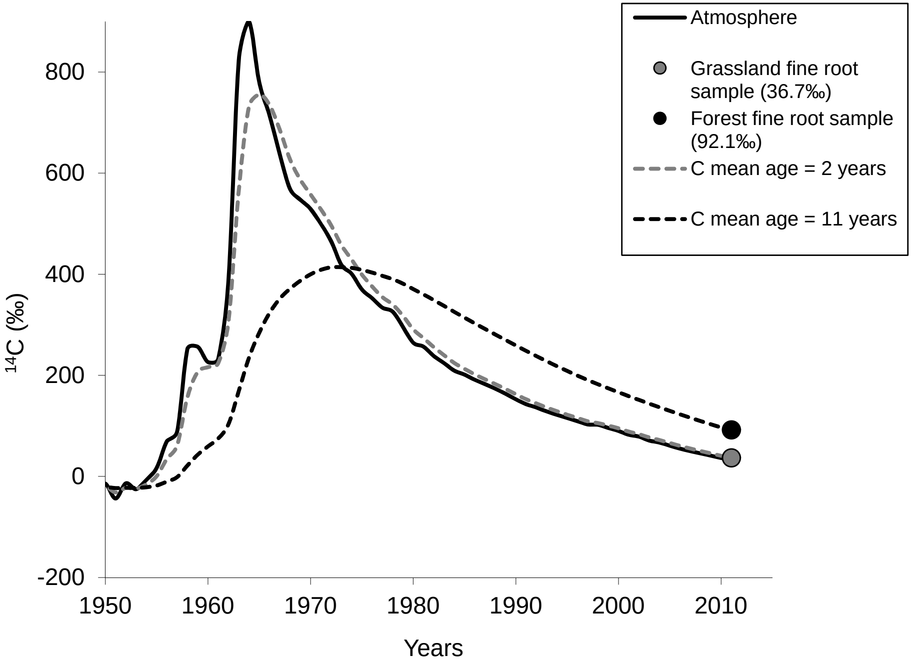

Fig. 1. Example of the model we used to determine mean age of fine root C from the ^{14}C signature of the fine roots (Gaudinski et al., 2001).

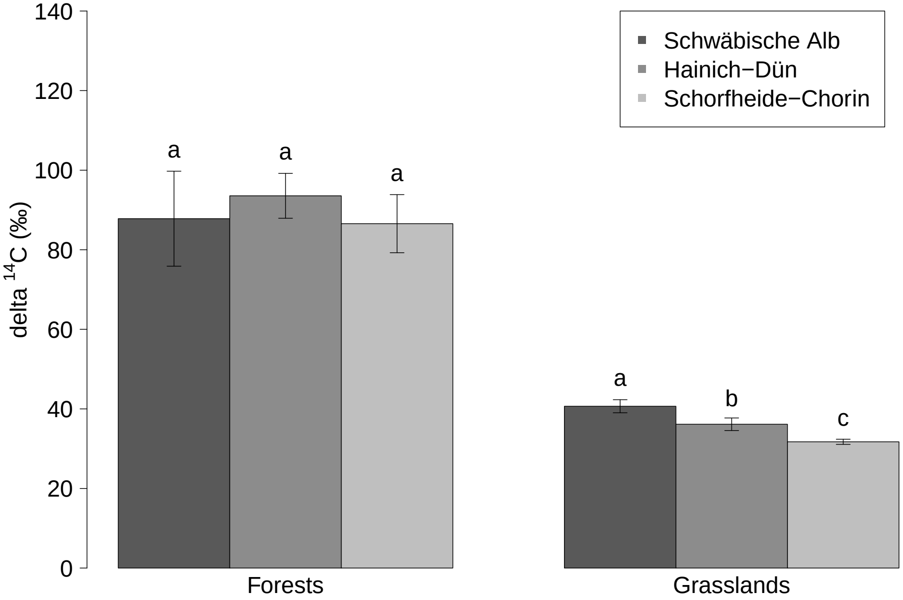

Fig. 2. Comparison of mean Δ^{14}C values of fine roots between grassland and forest sites across study regions ($p < 0.001$). The columns denoted with different letters are significantly different ($p < 0.05$). $n = 9$, error bars represent SE of means.

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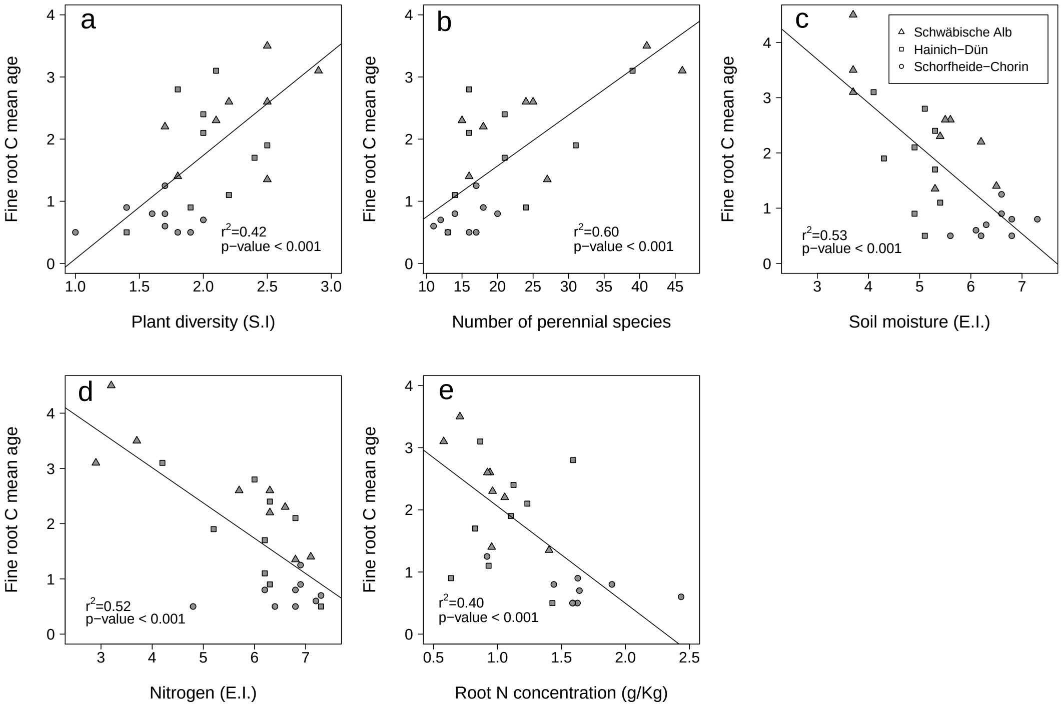

Fig. 3. Relationships between the mean age of fine roots and **(a)** plant diversity, **(b)** total number of perennial species, **(c)** Ellenberg indicator for moisture, **(d)** Ellenberg indicator for nitrogen, **(e)** fine root nitrogen concentration in the grasslands ($n = 27$).