Global Change Biology

Global Change Biology (2009) **15**, 2003–2019, doi: 10.1111/j.1365-2486.2009.01850.x

Sources of plant-derived carbon and stability of organic matter in soil: implications for global change

SUSAN E. CROW*† [1], KATE LAJTHA*, TIMOTHY R. FILLEY†, CHRISTOPHER W. SWANSTON‡[2], RICHARD D. BOWDEN§ and BRUCE A. CALDWELL*

*Department of Botany and Plant Pathology, Oregon State University, Corvallis, OR 97330, USA, †Department of Earth and Atmospheric Science and the Purdue Climate Change Research Center, Purdue University, West Lafayette, IN 47907, USA, ‡Center for Accelerator Mass Spectrometry, Lawrence Livermore National Laboratory, Livermore, CA 94551, USA, §Department of Environmental Science, Allegheny College, Meadville, PA 16335, USA

Abstract

Alterations in forest productivity and changes in the relative proportion of above- and belowground biomass may have nonlinear effects on soil organic matter (SOM) storage. To study the influence of plant litter inputs on SOM accumulation, the Detritus Input Removal and Transfer (DIRT) Experiment continuously alters above- and belowground plant inputs to soil by a combination of trenching, screening, and litter addition. Here, we used biogeochemical indicators [i.e., cupric oxide extractable lignin-derived phenols and suberin/cutin-derived substituted fatty acids (SFA)] to identify the dominant sources of plant biopolymers in SOM and various measures [i.e., soil density fractionation, laboratory incubation, and radiocarbon-based mean residence time (MRT)] to assess the stability of SOM in two contrasting forests within the DIRT Experiment: an aggrading deciduous forest and an old-growth coniferous forest. In the deciduous forest, removal of both above- and belowground inputs increased the total amount of SFA over threefold compared with the control, and shifted the SFA signature towards a root-dominated source. Concurrently, light fraction MRT increased by 101 years and C mineralization during incubation decreased compared with the control. Together, these data suggest that root-derived aliphatic compounds are a source of SOM with greater relative stability than leaf inputs at this site. In the coniferous forest, roots were an important source of soil lignin-derived phenols but needle-derived, rather than root-derived, aliphatic compounds were preferentially preserved in soil. Fresh wood additions elevated the amount of soil C recovered as light fraction material but also elevated mineralization during incubation compared with other DIRT treatments, suggesting that not all of the added soil C is directly stabilized. Aboveground needle litter additions, which are more N-rich than wood debris, resulted in accelerated mineralization of previously stored soil carbon. In summary, our work demonstrates that the dominant plant sources of SOM differed substantially between forest types. Furthermore, inputs to and losses from soil C pools likely will not be altered uniformly by changes in litter input rates.

Keywords: carbon, coniferous forest, cutin, deciduous forest, lignin, net primary productivity, soil organic matter, suberin

Received 21 July 2008; revised version received 3 December 2008 and accepted 15 December 2008

Correspondence: Present address: Susan E. Crow, [14]CHRONO Centre for Climate, the Environment, and Chronology, 42 Fitzwilliam St., Queen's University Belfast, Belfast BT9 6AX, UK, tel. + 44 28 9097 3085, fax: 44 28 9097 3897, e-mail: s.crow@qub.ac.uk

[1]Present address: [14]CHRONO Centre for Climate, the Environment, and Chronology, 42 Fitzwilliam St., Queen's University Belfast, Belfast BT9 6AX, UK

[2]Present address: USDA Forest Service, Northern Research Station, Houghton, MI 49931, USA

Introduction

The composition and input rate of plant litter to the forest floor varies in response to climate, land-use, and ecosystem disturbance (Jobbagy & Jackson, 2003). Litter is the primary source of soil organic matter (SOM) (Kögel-Knabner, 2002), which is the terrestrial biosphere's largest pool of organic carbon (C) and an

integral part of the global C cycle (Schimel, 1995). Small relative changes in the quality or quantity of litter inputs may alter the net accumulation or loss of soil C (Boone *et al*., 1998) and a growing body of research indicates that expected increases in atmospheric CO_2 and nitrogen (N) deposition will alter ecosystem net primary productivity (NPP) and allocation of resources between above- and belowground productivity in forests (Norby *et al*., 2005).

A complex and variable mixture of C-rich organic compounds, including polysaccharides (e.g., cellulose), aromatics (e.g., lignin and tannins), and aliphatics (e.g. waxes, suberin, cutin), comprises all plant litter. Lignin and most aliphatic compounds are among the more recalcitrant plant biopolymers, but the extent to which they are stabilized in soil is still unclear. Lignin is the second most abundant compound after cellulose, and is highly refractory during litter decay (Berg & Meentemeyer, 2002). However, it does not always appear as recalcitrant in soils as initially thought (e.g., Kiem & Kögel-Knabner, 2003; Dignac & Rumpel, 2006). Recent evidence shows that aliphatic plant compounds often accumulate in soil, thus contributing to stable SOM pools (e.g., Nierop, 1998; Lorenz *et al*., 2007).

The composition of above- and belowground plant tissue is substantially different (Kögel-Knabner, 2002) and the chemistry of specific compounds influences their ability to be decomposed and/or stabilized by association with soil minerals (Kleber *et al*., 2007). Various experiments have demonstrated a greater relative contribution of roots than shoots to soil C (Rasse *et al*., 2005), as well as the accumulation of root-derived suberin (e.g., Bull *et al*., 2000; Nierop *et al*., 2003; Filley *et al*., 2008a). Therefore, natural or anthropogenic changes in forest productivity and shifts between above- vs. below-ground allocation of resources have the potential to influence SOM formation and stabilization through both input rates and preferential stabilization of some tissues and compounds.

Few studies are designed to conclusively link detrital input quantity and quality (i.e. decomposability and relative amounts of C and N compounds) to SOM formation and stability. Inspired by an ongoing experiment initiated in 1957 in forest and grassland ecosystems at the University of Wisconsin Arboretum (Nielson & Hole, 1963), a growing network of long-term manipulative field experiments was established beginning in 1990 to examine effects of altering plant litter inputs on organic matter accumulation in soil. The central goal of this ongoing collaborative experiment, the Detritus Input Removal and Transfer (DIRT) Experiment, is to assess how rates and sources of plant inputs control the accumulation and dynamics of SOM and nutrients in different forest soils over decadal timescales (Nadelhoffer *et al*., 2004).

To examine the response of soil to sustained changes in plant litter input rate and source, we sought to (1) identify dominant sources of plant biopolymers, i.e., lignin-derived phenols and cutin/suberin-derived acids, in soils and (2) assess the stability of SOM from two contrasting forested sites: an aggrading deciduous forest and an old-growth coniferous forest that are part of the DIRT Experiment network. We used a combination of experimental field manipulations, key biogeochemical indicators of plant source, and measures of soil stability to identify important inputs to, and gauge the relative stability of, SOM in these sites to better understand the range of expected soil responses to altered rates and sources of plant litter across forest ecosystems.

Materials and methods

Site descriptions

The deciduous site (Bousson) is located in a nutrient-rich mixed deciduous forest in northwestern Pennsylvania, USA within the Bousson Experimental Forest (41°36′N, 80°3′W, 381 m) owned by Allegheny College. The site is dominated by black cherry (*Prunus seròtina* Ehrh.) and sugar maple (*Acer saccharum* Marsh.) in the overstory and by sugar maple saplings (<5 cm diameter at 1.35 m height) in the understory (~80-year-old stand). Of the aboveground biomass, 60% is black cherry and 28% is sugar maple (Bowden *et al*., 2000). American beech (*Fagus grandifolia* Ehrh.) and oaks (*Quercus* spp.) are secondary in the overstory. Litterfall is 2100 kg C ha^{-1} yr^{-1} (Bowden *et al*., 1993) and annual atmospheric N deposition for the site is ~ 13 kg N ha^{-1} yr^{-1} (Bowden *et al*., 2000). The climate is temperate with a 4 month growing season. Daily temperatures average −4 °C in January and 21 °C in August; snow cover is present for ~ 4 months annually and precipitation is spread evenly throughout the year (105 cm yr^{-1}). Soils are coarse loamy mixed superactive mesic Oxyaquic Fragiudalfs (Cambridge series) derived from glacial till overlying shale and sandstone (USDA-SCS, 1979) with a fragipan present at 60 cm. Soil pH in the top 6 cm of the A horizon is 4.0 (Bowden *et al*., 2000).

The coniferous site (H.J. Andrews) is located in a temperate coniferous rainforest within the H.J. Andrews Experimental Forest (44°15′N, 122°10′W, 531 m) in the Cascade Mountains of west-central Oregon, USA. The canopy is dominated by mixed old-growth Douglas-fir (*Pseudotsuga menziesii* Mirb.) and western hemlock (*Tsuga heterophylla* (Raf.) Sarg.). A major stand-replacing fire ca. 1500 AD was followed by establishment of

Douglas-fir, many of which are still present in the overstory. Coarse woody debris and moss cover extensive areas of the forest floor, as is typical of old-growth stands in this region. Litterfall is 600 kg C ha^{-1} yr^{-1} (Sulzman *et al*., 2005) and atmospheric N deposition in this region is low, ~1.6–2 kg N ha^{-1} yr^{-1} (Vanderbilt *et al*., 2003). The experimental plots are located at 726 m elevation on a relatively flat and stone-free bench compared with the surrounding hill slopes. The climate is Mediterranean, with dry summers and a cool wet season between October and March. Annual temperatures average 8.8 °C, and annual precipitation is 220 cm (Sulzman *et al*., 2005). Seventy percent of annual precipitation occurs during the wet season, mostly as rain. The soils are coarse loamy mixed mesic Andic Dystrudepts derived from volcanic parent material (Sollins *et al*., 2006). Soil pH is 5.2 at 0–10 cm (Yano *et al*., 2004).

Plant litter manipulation and sample collection

The Bousson DIRT plots were initiated in 1991, followed by the H.J. Andrews plots in 1997 (Nadelhoffer *et al*., 2004). The treatment plots are replicated three times at each site and consist of treatments that either add or remove above- and belowground litter inputs to the soil (see Table 1 for a description of treatments). Briefly, aboveground litter is excluded from the forest floor using screens, which are swept several times a year and transferred to the litter addition plots, and soil was trenched and root barriers placed to remove root inputs. At Bousson, plots are 3 × 3 m and at H.J. Andrews the plots are 10 × 15 m; all plots are within a 1 ha area at each site.

Table 1 Treatments and methods of the Detritus Input Removal and Transfer (DIRT) Experiment plots

Treatment	Method
Control (CTL)	Normal litter inputs are allowed
No Litter (NL)	Aboveground inputs are excluded from plots with netting*
Double Litter (DL)	Aboveground leaf/needle inputs are doubled by adding litter removed from No Litter plots
Double Wood (DW)	Aboveground wood inputs are doubled by adding large shredded wood pieces, both fresh and highly decayed, based on measured input rates of woody debris fall (H.J. Andrews only)
No Roots (NR)	Roots are excluded with impermeable plastic barriers extending from the soil surface to the top of the C horizon
No Inputs (NI)	Aboveground inputs are prevented as in No Litter plots; belowground inputs are prevented as in No Roots plots

*At Bousson litter is excluded primarily in fall during senescence, whereas at H.J. Andrews litter screens are in place throughout the year due to continuous litter fall.

Mineral soil (0–5 cm) was collected in June 2002 from H.J. Andrews (5 years after initiation) and in June 2003 from Bousson (12 years after initiation). Six 5 cm^2 cores were collected to a depth of 5 cm per treatment plot and composited, providing ~750 g of soil per plot. Each composite sample was sieved moist to remove material >2 mm. Mixed-species roots collected in the sieve were picked out with forceps, rinsed with distilled, deionized water, and air-dried. A subsample of the soil was removed and air-dried. The remaining sample was stored moist at 4 °C in tightly sealed plastic bags for up to 2 months until density fractionation (described later).

Mixed-species branches and freshly senesced leaf samples from the dominant tree species were collected from the forest floor in the autumn at Bousson. Fresh (i.e., green) leaves from the dominant tree species were collected from trees in the following spring. At H.J. Andrews, mixed Douglas fir and western hemlock needles were collected from litter traps placed on the forest floor. Fresh wood was obtained from the supply of wood chips retained for the Double Wood treatment and decayed wood was collected from the remnants of a downed log (decay class 4) on the forest floor near the DIRT plots. Air-dried plant litter and soil was ground with mortar and pestle until all material passed through a 250 μm sieve to assure heterogeneity.

Quantification of plant biopolymers

Alkaline cupric-oxide (CuO) oxidation was used to extract and quantify lignin-derived phenols (Hedges & Mann, 1979) and cutin/suberin-derived substituted fatty acids (SFA) (Goñi & Hedges, 1990a) from the dominant plant inputs and bulk soil at both DIRT sites. Owing to the labor-intensive nature of these analyses, a subset of the DIRT treatments were selected for analysis (Bousson: CTL, DL, and NI; H.J. Andrews: CTL, DL, DW, and NI; see Table 1 for treatment acronyms).

The CuO extractions utilized Monel reaction vessels (Prime Focus Inc. Seattle, WA, USA) and followed the original method of Hedges & Ertel (1982) with slight modifications (Dalzell *et al*., 2005). Ethyl vanillin and DL-12 hydroxystearic acid were added as internal recovery standards following the initial alkaline reaction and before the solvent extraction phase. Lignin-derived phenols were quantified by analysis of the trimethylsilane (TMS) derivatives of vanillyl (V)-based, (i.e., vanillin, acetovanillone, and vanillic acid); syringyl (S)-based (i.e., syringaldehyde, acetosyringone, and syringic

acid); cinnamyl (Ci)-based (i.e., *p*-hydroxycinnamic and ferulic acids) monomers using extracted-ion internal calibration curves. The TMS derivatives of two long-chain (C_{16} and C_{18}) SFA [i.e., 16-hydroxyhexadecanoic (ω-C_{16}), hexadecane-1,16-dioic (C_{16}DA)] were quantified using extracted-ion internal calibration curves and six long-chain SFA [i.e., 18-hydroxyoctadec-9-enoic (ω-$C_{18:1}$), 9,16&10,16-dihydroxyhexadecanoic (9&10, ω-C_{16}), 9-octadecene-1,18-dioic ($C_{18:1}$DA), 7&8-hydroxyhexadecane-1,16-dioic (7&8-C_{16}DA), 9,10,18-trihydroxyoctadec-12-enoic (9,10, ω-$C_{18:1}$), and 9,10,18-trihydroxyoctadecanoic(9,10, ω-C_{18})] were assessed by extracted ions based on similar proxy standard calibration curves.

A Hewlett-Packard (5971) quadrupole mass spectrometer interfaced to a 5890 series II gas chromatograph was used in the quantification of individual compounds by extracted-ion calibration curves. Derivatization of samples and GC-MS performance was verified by the addition of a 3,4 dimethoxybenzoic acid recovery standard immediately before derivatization. The abundance of lignin-derived phenols and SFA were determined relative to the ethyl vanillin standard. Duplicate CuO analyses were performed for all the plant litter samples and a subset of soil samples to assure consistency. Beyond the calibration curves and internal standards used, a standard peach leaf (NIST 1547) was used as a lab CuO reference material in each batch of analyses and to monitor the consistency of the reaction and GC performance on a biological substrate similar to our samples. Mean standard reproducibility for the analytical method was 2–5% for lignin phenols and 2–9% for SFA.

The concentration of C within the three classes of lignin-derived phenols (S-Lignin, V-Lignin, Ci-Lignin) and the total lignin-derived phenol concentration (SVCi-Lignin (S + V + Ci) were quantified in units of mg/100 mg OC (Hedges & Mann, 1979), i.e.,% OC. The ratios of the various lignin classes, e.g., S/V and Ci/V, can be used to interpret dominant sources of lignin (Hedges & Mann, 1979). For example, coniferous plants are concentrated in V-Lignin relative to S- and C-Lignin, deciduous plants have approximately equal concentrations of S- and V-Lignin, and grasses are concentrated in C-Lignin relative to S- and V-Lignin. Further, the ratios of acid and aldehyde monomers within the S and V classes [$Ac/Al_{(S)}$, $Ac/Al_{(V)}$] can be used as an index for the degree of lignin decay; elevated Ac/Al indicates greater decay state (Ertel & Hedges, 1984; Kögel, 1986).

The total C concentration within cutin/suberin-derived acid (ΣSFA) also was quantified in units of mg/100 mg OC. The relative abundances of specific SFA in root vs. leaf inputs have been used to distinguish between root and leaf sources of soil SFA (e.g., Kögel-Knabner *et al*., 1989; Riederer *et al*., 1993; Filley *et al*., 2008a; Crow *et al*., 2009). Cutin and suberin share many monomeric units; therefore, the compounds identified as leaf- and root-indicator acids are not exclusively from leaves or roots (but typically are ~ 90% exclusive) and ΣLeaf-indicator acids + ΣRoot-indicator acids does not equal ΣSFA. The classification of root-indicator acids (ΣRoot-indicator acids) and leaf-indicator acids (ΣLeaf-indicator acids) was made by identifying suites of SFA for each site such that ΣRoot-indicator acids/ΣSFA was greatest for roots and lowest for leaves, whereas ΣLeaf-indicator acids/ΣSFA was greatest for leaves and lowest for roots. The leaf-indicator and root-indicator acids are not the same for every site. Based on relative inputs rates and SFA concentration in above- vs. belowground inputs, and assuming that the leaf- and root-indicator acids decay at the same relative rate, we calculated the *expected* proportions of leaf- and root-indicator acids in SOM to account for the overlap in compounds among input sources and to guide our interpretations of the SOM plant biopolymer chemistry

$$\begin{aligned} EXP_{LA} &= (BG_{input(x)})(pBG_{LA}) \\ &\quad + (AG_{input(1-x)})(pAG_{LA}) \\ EXP_{RA} &= (BG_{input(x)})(pBG_{RA}) \\ &\quad + (AG_{input(1-x)})(pAG_{RA}) \\ EXP_{other} &= 1 - pSOM_{LA} - pSOM_{RA} \end{aligned}$$

where EXP_{LA} is the *expected* SOM ΣLeaf-indicator acids/ΣSFA, $BG_{input(x)}$ is the proportion of total inputs that are belowground (root) inputs, pBG_{LA} is the root ΣLeaf-indicator acids/ΣSFA, $AG_{input(1-x)}$ is the proportion of total inputs that are aboveground (leaf) inputs, pAG_{LA} is the leaf ΣLeaf-indicator acids/ΣSFA, EXP_{RA} is the *expected* SOM ΣRoot-indicator acids/ΣSFA, pBG_{RA} is the root ΣRoot-indicator acids/ΣSFA, pAG_{RA} is the leaf ΣRoot-indicator acids/ΣSFA, and EXP_{other} is the *expected* SOMΣNon-leaf/root indicator acids/ΣSFA.

Density fractionation

Soil was physically divided into two density fractions based on flotation in a 1.6 g cm^{-3} solution of sodium polytungstate (SPT; SOMETU, Van Nuys, CA). This method (Monnier *et al*., 1962; Greenland & Ford, 1964) separates soil into light and heavy recoverable fractions and a nonrecoverable fraction that is soluble in SPT (Crow *et al*., 2007).

Soil was added to the SPT solution in a 1/3 soil/SPT ratio by volume, shaken for 1 h, and allowed to settle gravimetrically for 48 h. Light fraction was aspirated and rinsed with distilled, deionized water on a pre-combusted Whatman GF/F (0.7 μm pore size). The cycle of shaking, settling, and aspiration was repeated until no light fraction remained floating, typically three

cycles. Heavy fraction material was rinsed by repeated centrifugation and resuspension in distilled, deionized water, typically four cycles were required (Crow *et al*., 2007).

Elemental analysis

Organic C (OC) and total N concentrations of bulk soil, density fractions, and plant tissues were determined by dry micro-Dumas combustion (NA1500 C/H/N Analyzer, Carlo Erba Strumentazione, Milan) at the Stable Isotope/Soil Biology Laboratory of the University of Georgia, Institute of Ecology.

Carbon mineralization

Bulk soil, light fraction, and heavy fraction from each plot at both sites were incubated for 1 year (dark conditions, 23 °C) in bench top filtration units (Falcon Filter, Becton Dickinson Labware) modified after Nadelhoffer (1990). With the goal of incubating the same amount of OC from each fraction, ~ 20 g of bulk soil, 6 g of light fraction, or 30 g of heavy fraction material were mixed with an equal amount of acid-washed sand for aeration (Swanston *et al*., 2002) and incubated. At the start of the incubation, the sand and soil mixture was rewetted by adding 10 mL of an inoculum solution prepared from fresh soil of the respective site shaken in distilled water for 1 h (1/10, soil/water by volume) and filtered with a Whatman GF/F (0.7 μm pore size). Moisture content was maintained by adding distilled, deionized water to each chamber weekly to maintain a constant weight over the incubation period.

CO_2 efflux for each chamber was measured on days 3, 5, 8, 12, 17, 26, 53, 151, 267, and 361 for H.J. Andrews and days 2, 5, 13, 20, 28, 64, 114, 208, 290, and 367 for Bousson. For each measurement, chamber headspace was purged with CO_2-free air and sealed for ~240 min while respired CO_2 accumulated. A 500 μL-calibrated syringe was used to mix the headspace gas several times before extracting a sample, which was immediately injected into a Hewlett Packard 5700A gas chromatograph (Palo Alto, CA, USA; now Agilent Technologies) fitted with a 2 m Poropak R 80/100 column and thermal conductivity detector. Respiration rate was plotted as concentration against time, and cumulative respiration was calculated for each substrate in SAS (SAS Institute, v. 9.1, Cary, NC, USA) by using PROC EXPAND to approximate the area under the curve using the trapezoidal method.

Mean residence time (MRT)

Radiocarbon concentration of the bulk soil and density fractions was determined at the Center for Accelerator Mass Spectrometry at Lawrence Livermore National Laboratory, CA with a Van de Graaff FN accelerator mass spectrometer. Samples were prepared for analysis by combustion of OC to CO_2 with CuO and powdered Ag in sealed evacuated tubes at 900 °C and subsequent reduction of the CO_2 onto iron powder in the presence of H_2 (Vogel *et al*., 1984). Radiocarbon data are expressed according to Stuiver & Polach (1977) as $\Delta^{14}C$, the deviation in parts per thousand from the absolute international standard activity ($^{14}C:{}^{12}C$ ratio of oxalic acid corrected for decay since 1950). The $\delta^{13}C$ values for each fraction was independently measured and used to adjust the $\Delta^{14}C$ values for mass-dependent fractionation.

MRT was calculated for the density fractions with a time-dependent steady-state model (Trumbore *et al*., 1995; Gaudinski *et al*., 2000). Three assumptions of the model are: (1) bulk C inputs equal loss in each pool at each time step; (2) the $\Delta^{14}C$ of inputs to the light and heavy fraction pools are equal to that of the atmosphere in the previous year, except for the NI treatment where inputs were set equal to the year of treatment initiation; and (3) the $\Delta^{14}C$ of inputs to the pool attributed to OM mobilized during fractionation was mass-weighted between the light and heavy fractions (Crow *et al*., 2007). Three chronologies of annual atmospheric $\Delta^{14}C$ beginning in calendar year 1511 were used in the model (Stuiver *et al*. 1998; Hua & Barbetti, 2004; Levin & Kromer, 2004).

At Bousson, two solutions to the modeled MRT for the light fraction were possible. The most-likely solution was determined based on further division of the light fraction into 6 N HCl acid hydrolysis residue (Paul *et al*., 2006) and an additional size separation of light fraction at 500 μm. The $\Delta^{14}C$ measured for the additional fractionations allowed superposition of the light fraction, small size fraction, and acid hydrolysis residue along the bomb-curve. Assuming that both the small size fraction and acid hydrolysis residue were effectively 'older' than the whole light fraction, this approach revealed the most likely model solution depending on whether further fractionation resulted in a radiocarbon value that was higher or lower on the bomb curve.

Statistical analysis

Comparisons of means for the DIRT treatments at each site were made using a completely randomized design, one-way ANOVA. To make comparisons between density fractions, a repeated measures ANOVA model was used. All ANOVA were completed using PROC MIXED in SAS v. 9.1 (SAS Institute Inc.). A Tukey–Kramer HSD *post hoc* test was used for comparison of means if a significant

P-value was found. Owing to low sample size and high natural heterogeneity of soil, significance for the contrasts was set at $P = 0.10$ and significant *P*-values were reported. If the data to be analyzed were in the form of a proportion or percent, an arcsine square-root transformation was used for statistical comparisons.

Results

Bousson: deciduous forest

Plant biopolymer composition of litter. The total yield of CuO-extractable plant biopolymers (i.e., SVCi-Lignin + ΣSFA) in forest floor litter and roots ranged from 9.4 mg/100 mg OC in sugar maple leaves to 15.7/100 mg OC in black cherry leaves (Table 2). Both S- and V-Lignin were abundant in all plant inputs, which is typical for angiosperm species (Hedges & Mann, 1979). Of the forest floor litter, mixed branches had the greatest SVCi-Lignin concentration and black cherry leaves had the lowest SVCi-Lignin concentration. During leaf senescence, the ratio of cinnamyl to vanillyl phenols (Ci/V) decreased and the ratio of syringyl to vanillyl phenols (S/V) increased for both black cherry and sugar maple leaves (Table 2). Branches had the greatest S/V, senescent black cherry had the greatest Ci/V, and roots had both the lowest S/V and Ci/V (Fig. 1a). $Ac/Al_{(V)}$ and $Ac/Al_{(S)}$ were consistent with previously reported values (Ertel & Hedges, 1984) (Table 2).

Total SFA yield ranged from 2.0/100 mg OC in branches to 13.4/100 mg OC in senescent black cherry leaves (Table 2). Roots were dominated by 9,10, ω-C_{18} (6.3/100 mg OC), ω-$C_{18:1}$ (0.7/100 mg OC), and C_{16}DA (0.5/100 mg OC), which together comprised 90% of total SFA yield. Senescent leaves from sugar maple and black cherry were dominated by a different suite of SFA than roots; 9&10, ω-C_{16} (14.4/100 mg OC) and 7&8-C_{16}DA (1.3/100 mg OC), which together comprised on average 84% of SFA yield for the leaves. These suites of compounds are henceforth referred to as the root-indicator acids and leaf-indicator acids for this site. The relative amount of leaf-indicator acids compared with root-indicator acids decreased during leaf senescence (Table 2), but leaf-indicator acids remained more than five times more concentrated than root-indicator acids for senescent leaves of both species.

Source of plant biopolymers in SOM. The DIRT treatments did not significantly change the ratios of the lignin-derived phenols classes in surface mineral soils (Table 3). However, the soil lignin-derived phenol composition, on average for the site, was most similar to that of roots and senescent sugar maple leaves (Fig. 1a). Soil Ci/V is elevated from that of roots and sugar maple leaves, indicating an additional input source of

Table 2 Summary of lignin-derived phenol and cutin/suberin-derived SFA yields (mg/100 mg OC) and parameters for the dominant plant inputs at Bousson and H.J. Andrews

	Bousson						*H.J. Andrews*			
	Fresh litter		Forest floor litter				Forest floor litter			
	Cherry	Maple	Mixed branches	Cherry	Maple	Mixed roots	Wood chips	Decayed wood	Mixed Needles	Mixed Roots
S-Lignin	0.4	0.2	7.7	1.1	1.1	1.2	1.9	0.2	0.1	0.2
V-Lignin	0.3	0.6	3.3	0.6	1.9	4.5	7.6	13.0	4.0	3.2
Ci-Lignin	1.1	0.8	0.7	0.6	0.4	0.7	2.0	0.2	0.8	0.3
SVCi-Lignin	1.8	1.7	11.7	2.3	3.3	6.5	11.4	13.4	4.9	3.7
Ci/V	3.3	1.3	0.2	0.9	0.2	0.2	0.26	0.02	0.2	0.1
S/V	1.2	0.3	2.3	1.7	0.6	0.3	0.25	0.02	0.04	0.1
$Ac/Al_{(S)}$	0.14	0.09	0.13	0.20	0.16	0.25	0.12	0.34	0.55	0.31
$Ac/Al_{(V)}$	0.19	0.12	0.17	0.21	0.19	0.25	0.16	0.32	0.26	0.22
Σ Leaf-indicator acids	5.8	3.9	0.4	10.5	5.2	0.2	0.1	0.1	26.4	0.4
Σ Root-indicator acids	0.2	0.2	1.3	1.9	0.7	7.5	1.2	0.02	0.04	2.8
Σ SFA	7.7	4.3	2.0	13.4	6.1	8.3	4.8	0.1	26.9	3.4

The reported values represent the mean of two analytical replicates.
S, syringyl; V, vanillyl; Ci, cinnamyl; Ac/Al, acid-to-aldehyde ratio; SFA, substituted fatty acids.

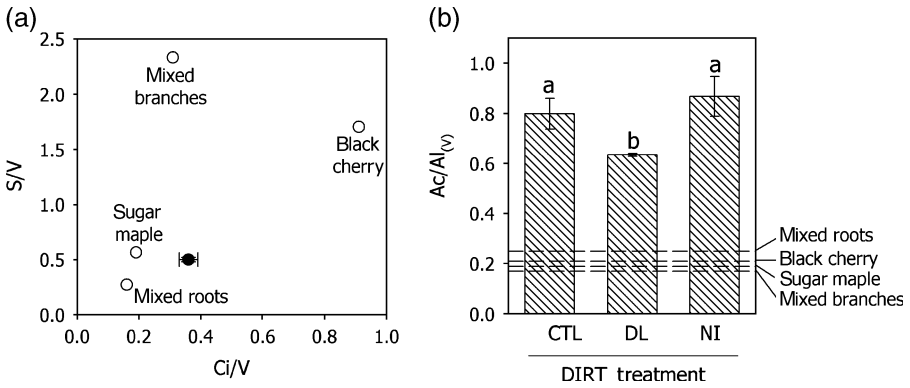

Fig. 1 Comparing ratios of lignin-derived phenol classes for bulk soil (solid symbol) and plant litter (open symbols) (a) and the acid-to-aldehyde ratio of V-Lignin for bulk soil (bars) and plant litter (dotted lines) (b) at the Bousson DIRT plots. Values are means ± 1 standard error; letters indicate significant differences among means for the treatments; if no differences existed, the site mean is shown; see Table 3 for *P*-values.

black cherry leaves. Although there were no significant differences in soil S-Lignin, V-Lignin, Ci-Lignin, or SVCi-Lignin concentration among the DIRT treatments (Table 3), $Ac/Al_{(V)}$ was significantly lower in the DL plots than the CTL and NI plots (Fig. 1b).

Total SFA yield was significantly greater in the NI plots compared with the CTL and DL plots (Fig. 2a). Although the DIRT treatments did not change the soil leaf-indicator acid yield, the soil root-indicator acid yield was significantly greater in the NI plots compared with the CTL and DL plots (Table 3). As a result, the proportion of total SFA that were root-indicators was greater ($P = 0.077$), and the proportion of total SFA that were leaf-indicators was less ($P = 0.083$), in soil from the NI plots compared with the CTL or DL plots (Fig. 2b).

Stability of SOM. The DIRT treatments did not significantly change bulk soil OC%, C/N ratio, and OC distribution among soil fractions. On average for all treatments, bulk soil OC concentration was 6.9 ± 0.5% and C/N ratio was 13.9 ± 0.5. The majority of soil OC was mineral-associated and thus recovered in the heavy fraction (site mean, 67.0 ± 2.2%). Approximately one-fifth (site mean, 20.8 ± 2.1%) of soil OC was solublized in the SPT solution and rinsed away during density fractionation as has been previously reported (Crow *et al.*, 2007). The remainder of soil OC was recovered within the light fraction (site mean, 12.2 ± 0.8%).

Carbon mineralization from the density fractions during one year incubation differed among the DIRT treatments, whereas C mineralization from bulk soil did not. There was a significant interactive effect of DIRT treatment and density fraction on cumulative respiration from the soil fractions ($P = 0.062$) (Fig. 3a). Respiration was significantly greater for light fraction from the DL plots than the light fraction from the NI plots and heavy fraction from all DIRT treatments. Respiration also was significantly greater for light fraction from the NL plots than the heavy fraction for the CTL, NI, and NR treatments and for light fraction from the CTL and NR plots than the heavy fraction for the NR plots. On average for all treatments, 4.8 ± 0.3 mg C/100 mg $OC_{initial}$ was mineralized from bulk soil during incubation.

The modeled MRT of OC within the soil light fraction changed as a result of altered plant inputs; light fraction MRT from the NI plots (185 ± 14 years) was significantly longer than the other DIRT treatments (78–113 years) (Table 4). Heavy fraction MRT did not differ significantly among the DIRT treatments (site mean, 251 ± 24 years). Within each treatment, the SPT soluble fraction (estimated by mass balance) had the shortest MRT of the soil fractions.

H.J. Andrews: coniferous forest

Plant biopolymer composition of litter. The total yield of CuO-extractable plant biopolymers (i.e., SVCi-Lignin + ΣSFA) in litter ranged from 7.1/100 mg OC in roots to 31.8/100 mg OC in needles (Table 2). All plant inputs were dominated by V-Lignin, which is typical for coniferous species (Hedges & Mann, 1979). Among the litter inputs, decayed wood had the greatest SVCi-Lignin concentration and mixed roots had the lowest SVCi-Lignin concentration. Needles had the greatest Ci-Lignin concentration. Both Ci/V and S/V decreased from fresh to decayed wood. This pattern was driven both by decreases in S- and Ci-Lignin, which are nearly absent in the decayed material, and an increase in V-Lignin concentration (Table 2). Decayed

Table 3 Summary of lignin-derived phenol and cutin/suberin-derived SFA yields (mg/100 mg OC) and parameters for the bulk soil at the Bousson and H.J. Andrews DIRT plots.

Site and DIRT Treatment	S-Lignin	V-Lignin	Ci-Lignin	SVCi-Lignin	Ci/V	S/V	$Ac/Al_{(S)}$	$Ac/Al_{(V)}$	Σ Leaf-indicator acids	Σ Root-indicator acids	Σ SFA
Bousson											
CTL	0.4 (0.0)	0.7 (0.1)	0.2 (0.0)	1.3 (0.1)	0.34 (0.02)	0.50 (0.01)	0.58 (0.04)^{a}	0.80 (0.06)^{a}	2.4 (0.6)	3.7 (0.6)^{b}	6.3 (1.2)^{b}
DL	0.4 (0.0)	0.8 (0.0)	0.3 (0.0)	1.5 (0.1)	0.32 (0.02)	0.51 (0.03)	0.48 (0.03)^{ab}	0.64 (0.01)^{ab}	2.7 (0.9)	2.8 (0.8)^{b}	5.8 (0.2)^{b}
NI	0.4 (0.0)	0.7 (0.0)	0.3 (0.0)	1.4 (0.1)	0.42 (0.06)	0.51 (0.04)	0.61 (0.04)^{a}	0.87 (0.08)^{a}	3.8 (0.3)	13.6 (0.6)^{a}	18.0 (0.7)^{a}
P-value	ns	ns	ns	ns	ns	ns	*0.087*	*0.070*	ns	*<0.001*	*<0.001*
Site mean	0.4 (0.0)	0.8 (0.0)	0.3 (0.0)	1.5 (0.1)	0.36 (0.03)	0.50 (0.02)			3.0 (0.4)		
H.J. Andrews											
CTL	0.2 (0.0)	3.2 (0.7)	0.4 (0.0)	3.8 (0.7)	0.13 (0.03)	0.06 (0.01)	0.55 (0.03)	0.45 (0.03)	3.5 (1.0)	0.1 (0.0)	4.2 (1.0)
DL	0.1 (0.0)	3.6 (1.0)	0.3 (0.1)	4.0 (1.0)	0.13 (0.07)	0.05 (0.02)	0.60 (0.30)	0.55 (0.02)	4.4 (1.5)	0.2 (0.1)	5.1 (1.7)
DW	0.1 (0.0)	5.4 (1.4)	0.4 (0.0)	6.0 (1.4)	0.10 (0.03)	0.03 (0.01)	0.60 (0.04)	0.50 (0.02)	3.7 (0.2)	0.1 (0.0)	4.3 (0.3)
NI	0.2 (0.0)	5.6 (1.5)	0.4 (0.0)	6.1 (1.5)	0.09 (0.03)	0.04 (0.02)	0.57 (0.30)	0.49 (0.05)	3.7 (1.0)	0.1 (0.0)	4.3 (1.1)
P-value	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
Site mean	0.2 (0.0)	4.4 (0.6)	0.4 (0.0)	5.0 (0.6)	0.11 (0.02)	0.04 (0.01)	0.58 (0.02)	0.49 (0.02)	3.8 (0.5)	0.1 (0.0)	4.5 (0.5)

The reported values are means of three replicate plots (one standard error). Significance was determined by one-way ANOVA followed by a Tukey–Kramer HSD *post hoc* test. Means followed by different letters in superscript were significantly different from each other at $P \leq 0.10$, *P*-value is reported. If no significant differences were detected among treatment means, the site mean and standard error was reported.

CTL, control; DL, double litter; DW, double wood; NI, no inputs; NL, no litter; NR, no roots; S, syringyl; V, vanillyl; Ci, cinnamyl; Ac/Al, acid-to-aldehyde ratio; SFA, substituted fatty acids; ns, nonsignificant.

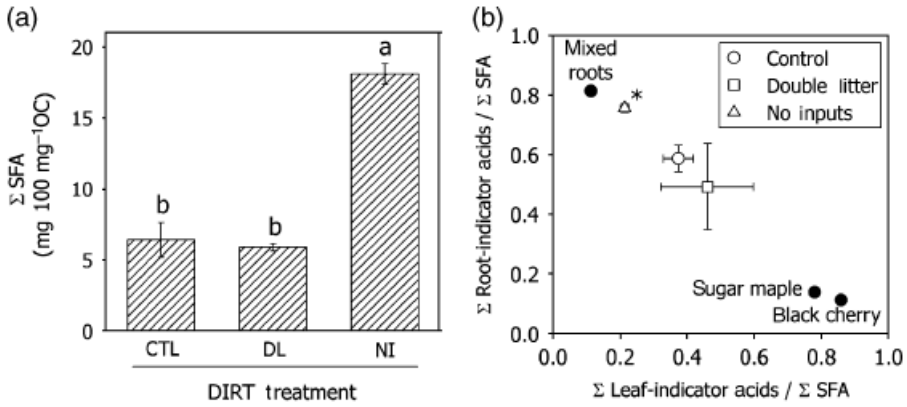

Fig. 2 Bulk soil SFA yield (a) and the relative proportion of root-indicator acids vs. leaf-indicator acids for bulk soil and plant litter (b) for the Bousson DIRT plots. Values are means ± 1 standard error; letters indicate significant differences among treatment mean ΣSFA values; *Significant differences between both variables for that treatment compared with the others; see Table 3 and text for *P*-values.

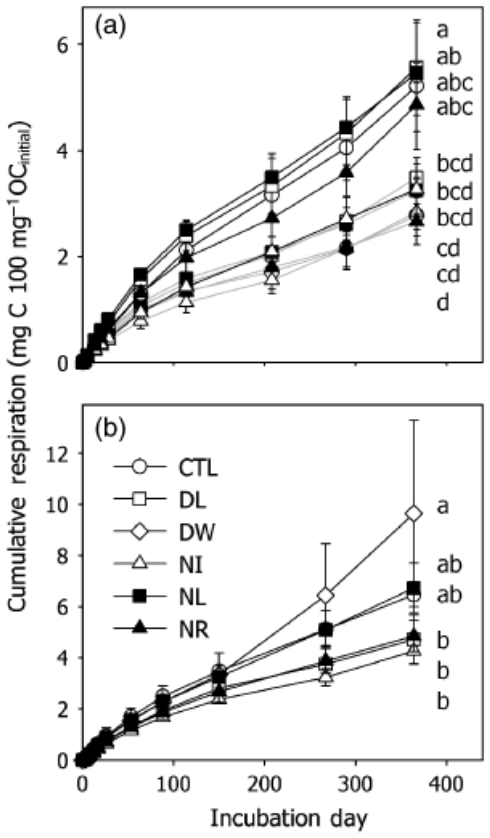

Fig. 3 Cumulative respiration during a 1-year incubation of soil light fraction (black lines) and heavy fraction (grey lines) from the Bousson DIRT plots (a) and bulk soil from the H.J. Andrews DIRT plots (b). Values are means ± 1 standard error; letters indicate significant differences among means, see text for *P*-values. Note that (a) and (b) are on different scales.

wood had the lowest Ci/V and S/V and fresh wood had the greatest Ci/V and S/V (Fig. 4a). Mixed roots had greater S/V than decayed wood and mixed Douglas-for/western hemlock needles had greater Ci/V than decayed wood. The $Ac/Al_{(S,V)}$ ratios were consistent with previously reported data (Ertel & Hedges, 1984) (Table 2).

Total SFA yield ranged from 0.1 mg/100 mg OC in decayed wood to 26.9/100 mg OC in needles (Table 2). Roots were dominated by ω-$C_{18:1}$ (2.2/100 mg OC) and $C_{18:1}$DA (0.6/100 mg OC), which together comprised 82% of total SFA yield. Douglas fir and western hemlock needles were dominated by a different suite of SFA than roots; 9&10, ω-C_{16} (20.5/100 mg OC) and 7&8-C_{16}DA (5.9/100 mg OC), which together comprised 98% of total SFA from needles. These suites of compounds are henceforth referred to as the root-indicator acids and leaf-indicator acids for this site. Fresh wood contained some root-indicator acids that likely were present in bark, which also shares some similar compounds with roots (Matzke & Riederer, 1991).

Source of plant biopolymers in SOM. The soil lignin-derived phenol composition (i.e., S/V vs. Ci/V) elucidated the sources of plant biopolymers in soil. The DIRT treatments did not significantly change the relative ratios of the lignin-derived phenols classes (Table 3). However, the soil lignin-derived phenol composition, on average for the site, was most similar to roots (Fig. 4a). There were no significant differences in the soil S-Lignin, V-Lignin, Ci-Lignin, SVCi-Lignin concentrations or $Ac/Al_{(S,V)}$ among the DIRT treatments (Table 3). Although not statistically significant, soil $Ac/Al_{(V)}$ was elevated for the DL plots than the CTL plots.

The DIRT treatments did not change the soil ΣSFA, ΣCutin acids, or ΣSuberin acids. On average for the site, 84% of soil SFA were leaf-indicator acids and 2% were root-indicator acids (Fig. 4b).

Table 4 Mean $\Delta^{14}C$ and modeled MRT of bulk soil and organic matter fractions for the Bousson and H.J. Andrews DIRT plots

Site and DIRT Treatment	$\Delta^{14}C$ (‰)			MRT (yr)		
	Bulk Soil	Light Fraction	Heavy Fraction	Light Fraction	Heavy Fraction	SPT Soluble
Bousson						
CTL	17.9 (5.0)	73.9 (9.7)	1.9 (4.0)	84 (13)[b]	300 (17)	29 (19)
DL	44.0 (19.6)	83.9 (9.8)	28.3 (6.4)	84 (18)[b]	233 (32)	56 (36)
NI	5.5 (4.2)	34.8 (6.5)	10.7 (4.1)	185 (14)[a]	280 (16)	146 (46)
NL	34.5 (6.8)	80.1 (13.2)	22.9 (9.1)	113 (16)[b]	236 (40)	28 (17)
NR	49.5 (7.0)	90.1 (13.8)	33.6 (11.5)	78 (5)[b]	208 (33)	1.1 (0.6)
P-value	na	na	na	*0.002*	ns	na
Site mean					251 (24)	
H.J. Andrews						
CTL	85.5 (15.4)	73.4 (20.9)	97.7 (11.4)	111 (23)	93 (12)	19 (10)
DL	76.7 (21.5)	63.4 (26.3)	86.3 (14.4)	132 (35)	107 (16)	12 (8)
DW	85.7 (14.2)	52.2 (16.8)	99.6 (14.0)	108 (13)	155 (77)	0.4 (0)
NI	59.4 (6.9)	27.2 (18.5)	65.6 (7.1)	217 (69)	134 (11)	10 (9)
NL	90.7 (14.1)	78.8 (23.3)	96.5 (10.9)	99 (15)	133 (34)	24 (7)
NR	69.8 (7.6)	49.1 (27.9)	73.8 (7.4)	115 (29)	212 (57)	87 (75)
P-value	na	na	na	ns	ns	ns
Site mean				131 (16)	139 (17)	25 (13)

The reported values are means of three replicate plots (1 standard error). When applicable, significance was determined by one-way ANOVA followed by a Tukey–Kramer HSD *post hoc* test. Means followed by different letters in superscript were significantly different from each other at $P \leq 0.10$, *P*-value is reported. If no significant differences were detected among treatment means, the site mean was reported.

MRT, mean residence time; CTL, control; DL, double litter; DW, double wood; NI, no inputs; NL, no litter; NR, no roots; ns, nonsignificant; na, not applicable.

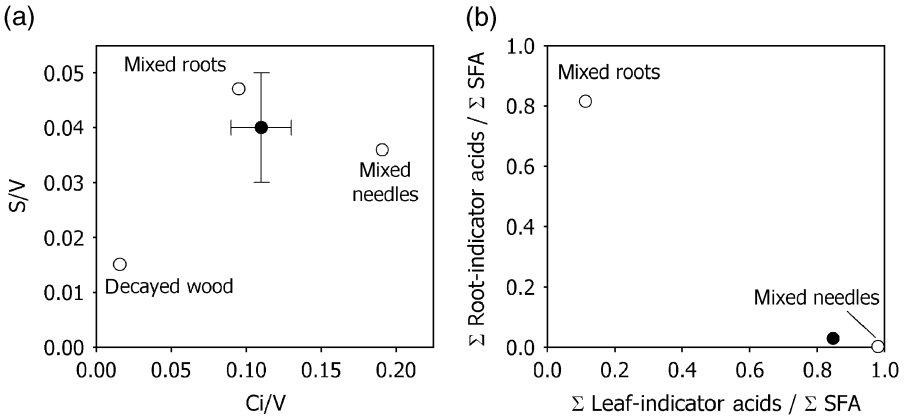

Fig. 4 Comparing ratios of lignin-derived phenol classes (a) and the relative proportion of root-indicator acids vs. leaf-indicator acids for bulk soil (solid symbols) and plant litter (open symbols) (b) at the H.J. Andrews DIRT plots. No significant differences existed among the DIRT treatments, values are site means ± 1 standard error.

Stability of SOM. The DIRT treatments did not significantly impact bulk soil OC% or C/N ratio, but did alter the relative amount of OC within the soil light fraction. On average for all treatments, the bulk soil OC concentration was 9.8 ± 1.4% and C/N ratio was 37.3 ± 2.0. The majority of soil OC was mineral-associated and thus recovered in the heavy fraction (site mean, 57.1 ± 1.7%). Nearly one-third (site mean, 29.3 ± 3.2%) of total OC was solublized in the SPT. The remainder of soil OC was recovered within the light fraction. The proportion of soil OC recovered in light fraction was significantly greater for the DW plots than the NI plots ($P = 0.097$) (Fig. 5).

Carbon mineralization from the bulk soil during 1-year incubation was sensitive to changes in plant inputs, whereas C mineralization from the density

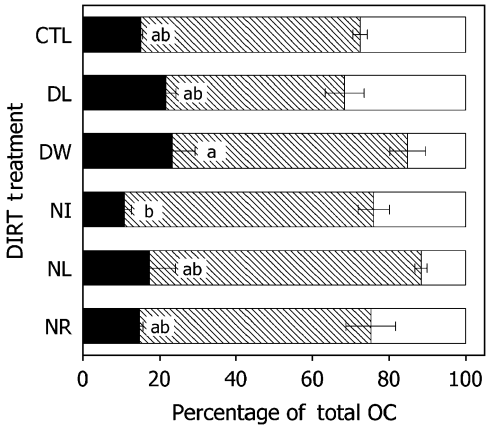

Fig. 5 Partitioning of bulk soil OC among the light fraction (black bars), heavy fraction (hatched bars), and SPT-soluble fraction (white bar) at the H.J. Andrews DIRT plots. Values are means ± 1 standard error; the soluble fraction was calculated by difference; letters indicate significant differences among means for the treatments, $P = 0.097$.

fractions was not. Cumulative respiration during incubation was greater for bulk soil from the DW plots than for DL, NI, and NR plots ($P = 0.089$) (Fig. 3b). On average for all treatments, cumulative respiration during 1-year incubation of light fraction was 3.2 ± 0.2 mg C/100 mg $OC_{initial}$ and of heavy fraction was 3.7 ± 0.2 mg C/100 mg $OC_{initial}$.

The estimated residence time of C within the soil fractions did not change as a result of altered plant inputs (Table 4). On average for all treatments, light fraction had an MRT of 130 ± 16 years, heavy fraction had an MRT of 139 ± 17 years, and SPT-soluble carbon had an MRT of 25 ± 13 years.

Discussion

Bousson: deciduous forest

Source and fate of plant biopolymers in soil. Both black cherry and sugar maple leaves had high concentration of Ci-Lignin that was lost during decomposition from fresh leaves to senescent leaves. Cinnamyl-based lignin phenols are thought to be bonded to lignin polymers and sugars by ester linkages that are readily hydrolyzed and preferentially lost during early stages of litter decomposition (Bahri *et al*., 2006). In contrast, S-Lignin, V-Lignin, and SFA concentrations increased during leaf senescence, indicating that other compounds such as carbohydrates and cellulose were preferentially lost whereas S- and V-Lignin and SFA were preserved (Table 2). The progressive oxidation of aldehyde to acid functional groups on the lignin-derived phenol, resulting in the increase in $Ac/Al_{(V,S)}$ occurred during leaf senescence in both black cherry and sugar maple, which is typical during decay (Kögel, 1986). Lignin-derived phenols in soil at Bousson were a combination of roots, sugar maple leaves, and black cherry leaves (Fig. 1a). Adding aboveground litter decreased the lignin decay state, indicating that fresh litter from the additions, which has a lower $Ac/Ad_{(V)}$ than soil, was being incorporated into bulk soil C over time (Fig. 1b).

Aboveground inputs are approximately three times greater than belowground inputs at Bousson: measured annual litter fall inputs at Bousson were the equivalent of $210\,g\,C\,m^{-2}\,yr^{-1}$ (Bowden *et al*., 1993) and Jackson *et al*. (1997) reported mean fine root (<2 mm) input values for 14 temperate deciduous forests globally of $65\,g\,C\,m^{-2}\,yr^{-1}$ in the top 5 cm of soil, assuming fine root turnover of 1 year. The average SFA yield for senescent black cherry and sugar maple leaves (9.8/100 mg OC) was similar to that for roots (8.3/100 mg OC) (Table 2). Thus, we assumed that aboveground litter input rates were three times greater than belowground inputs and that the SFA concentration was equal in our estimation of the *expected* proportions of leaf- and root-indicator acids in SOM. If decay rates for above- and belowground SFA were equal, we calculated that 32% of soil SFA would be comprised of the root-indicator acids, 62% would be comprised of the leaf-indicator acids, and 6% would be comprised of other acids. However, our results showed that 59% of soil SFA were root-indicators, 38% were leaf-indicators, and 3% were other, nonleaf/root root-indicators in the Control soil (Table 3).

Conflicting results have come from the few studies investigating the selective decay of leaf- and root-derived aliphatic compounds, (Goñi & Hedges, 1990c; Riederer *et al*., 1993; Nierop *et al*., 2003; Dignac & Rumpel, 2006). Two of the three root-indicator compounds identified at Bousson (9,10, ω-C_{18}, and C_{16}DA) were preferentially preserved over other plant biopolymers in soil particulate organic matter in an deciduous forest in eastern Maryland, USA, whereas the third compound (ω-$C_{18:1}$) was not (Crow *et al*., 2009). In the same study, both leaf-indicator compounds identified at Bousson (9&10, ω-C_{16}, and 7&8-C_{16}DA) were more concentrated in soil particulate matter compared with other plant biopolymers. As such, these compounds should be reasonable conservative proxies for both above- and belowground litter inputs and sources of potentially stabilized plant-derived C in soil at this site. This indicates that leaf inputs to the soil were not as high relative to the root inputs as the input rates to the forest floor suggested. Aboveground litter at this site is easily degradable, thus less likely to enter SOM, and other factors such as invertebrate consumption of leaf material on the forest floor has been shown to reduce

the abundance of leaf-indicator acids in soil and accumulating litter (Filley *et al*., 2008b; Crow *et al*., 2009) and could contribute to this observed difference in the expected and observed soil SFA composition.

Removing above- and belowground litter tripled the concentration of all extractable aliphatic compounds in soil (Fig. 2a) and shifted the signature towards a more dominant root source (Fig. 2b). Fresh inputs were halted as part of the DIRT Experiment, therefore, the observed increase in SFA concentration when plant inputs were removed was likely a result of preferential decomposition of other, less stabilized non-SFA compounds. In contrast, lignin-derived phenol concentration did not increase when plant inputs were removed (Table 3), indicating that these compounds were not similarly preserved. Estimates of fine root (<2 mm diameter) decomposition vary widely depending on the method used (Strand *et al*., 2008) and can range from 1.6 to 18 years for deciduous species (Gaudinski *et al*., 2001; Matamala *et al*., 2003; Yang *et al*., 2004). Because the soil was sampled 12 years posttrenching, the majority of dead roots likely had decomposed. Thus, the observed increase in the relative proportion of root-derived acids in soil was due (1) to the stabilization of root-derived OC from the trenched roots (approximately decadal scale) (2) the expression of accumulated root-derived material over time (century to millennial scale), or (3) a combination of both.

Soil C stability. Taken together, our analysis of soil chemistry, mineralization during incubation, and soil pool MRT suggest that roots were an important source of stabilized SOM at Bousson. The removal of all plant inputs changed not only the response of SOM to further decay (i.e., decreased respiration during incubation compared with the control) (Fig. 3a) but also the C dynamics *in situ* (i.e., increased MRT of the light fraction compared with the other treatments) (Table 4). In spite of the pulse of fine roots (in the 0–5 cm soil sampled) to SOM following trenching, the MRT of the light fraction increased in the No Inputs treatment by over 100 years compared with the control. A MRT of 185 years for the light fraction indicates a dominant organic matter source that is resistant to rapid loss.

Similar to our results, other studies have shown root-derived aliphatics to accumulate in soil to a greater extent than lignin- and leaf-derived aliphatic compounds in some ecosystems (Nierop *et al*., 2003; Rasse *et al*., 2005; Feng & Simpson, 2007). Aboveground plant inputs at Bousson are largely leaf litter from black cherry and sugar maple. Consistent with our plant biopolymer and soil stability analyses that indicated aboveground litter is the source of the most actively cycling C, litter from these species have a high nutrient content that contributes to rapid decomposition and release of nutrients into the soil (Van der Heyden *et al*., 2001; Lorenz *et al*., 2004). CO_2-enrichment experiments have shown large increases in root production in deciduous forests (Norby *et al*., 2004) and 5.6% increase in soil C overall (Jastrow *et al*., 2005), indicating that roots are likely to become increasingly important litter inputs in the future. Root chemistry did not change under elevated atmospheric CO_2 concentration and soil resource availability in a northern aspen and sugar maple dominated forest, suggesting that increases in root production, but not changes in root chemistry, may impact the soil C cycle in the future (King *et al*., 2005).

H.J. Andrews: coniferous forest

Source and fate of plant biopolymers in soil. Coarse woody debris, defined as wood pieces larger than 10 cm in diameter and 1 m in length, accounts for 74–81% of the aboveground detritus mass in the forests at the H.J. Andrews (Harmon *et al*., 1990). Coarse woody debris fragments over time and, particularly in old-growth forests, leaves a long legacy of high C litter on the forest floor. Despite the importance of woody debris on the forest floor, the soil lignin-derived phenol composition was most similar to roots (Fig. 4a). Although this would suggest that lignin derived from coarse woody debris was not important in the SOM pool, we note that the CuO method does not detect changes lignin chemistry associated with brown-rot fungi, which extensively demethylate and depolymerize lignin (upwards of 25% in decay studies) without significant side chain oxidation and are common in coniferous forests (Filley *et al*., 2002). This limitation may lead to an underestimate of the contribution of decayed wood to the soil lignin-derived phenol pool. Needles had the greatest concentration of Ci-Lignin, which is the most easily degraded class of lignin phenol. If needle-derived Ci-Lignin continues to preferentially degrade during processing in soil, then the contribution of needles to the soil lignin-derived phenol pool also may be underestimated by our analysis.

Roots were an important source of soil lignin-derived phenols, but the soil SFA largely did not appear to be of root-origin (Fig. 4b). Needle and root inputs are approximately equal at H.J. Andrews: measured annual litter inputs were the equivalent of $60\,g\,C\,m^{-2}\,yr^{-1}$ to the soil surface at H.J. Andrews (Sulzman *et al*., 2005) and Jackson *et al*. (1997) reported mean fine root (<2 mm) input values for 10 temperate coniferous forests globally of $68\,g\,C\,m^{-2}\,yr^{-1}$ in the top 5 cm of soil, assuming fine root turnover of 1 year. The SFA yield in needles (26.9/100 mg OC) was

approximately eight times greater than in roots (3.4/100 mg OC) (Table 2). Thus, we assumed that above- and belowground litter input rates were equal, but that the SFA concentration was eight times greater for aboveground litter than belowground in our estimation of the *expected* proportions of leaf- and root-indicator acids in SOM. Assuming decay rates for above- and belowground SFA were equal, we calculated that 9% of soil SFA would be comprised of the root-indicator acids, 89% would be comprised of the leaf-indicator acids, and 2% would be comprised of other acids. Our results were similar: 2% of soil SFA were root-indicators, 84% percent were leaf-indicators, and 14% were other, non cutin/root-indicator acids for the site (Table 3).

The lower than expected observed value for the contribution of root-indicator acids to soil SFA composition may be attributable to an underestimation of root inputs or the selective loss of the root-derived acids compared with needle-derived acids. Both of the root-indicator acids (ω-$C_{18:1}$ and $C_{18:1}$DA) have double bonds, which often are preferentially degraded more than other SFA without these substitutions (Goñi & Hedges, 1990c; Feng & Simpson, 2007). A previous study showed that the root-indicator acids predominant at H.J. Andrews were preferentially lost during decay and were not preserved in soil particulate organic matter (Crow *et al*., 2009).

The leaf-indicator acids comprised over one-quarter of the total OC in a mixed sample of needles. Needles from Douglas fir and western hemlock are similar in SFA composition (Goñi & Hedges, 1990c) and were dominated by 9&10, ω-C_{16} and 7&8-C_{16}DA, which are common in coniferous foliar tissue (Goñi & Hedges, 1990b) and were the same as the leaf-indicator acids we identified at Bousson. Although these compounds were found to be lost from Douglas fir and western hemlock needles during initial sedimentation, over time they stabilized relative to other plant biopolymers (Goñi & Hedges, 1990c), and are so abundant that they accumulated in soil particulate organic matter (Crow *et al*., 2009). Selective loss of some of the leaf-indicator acids relative to the non leaf/root-indicator acids may account for the slightly lower observed than expected value for the contribution of leaf-indicator acids to soil SFA composition. The DIRT treatments had no effect on the soil SFA composition, indicating that the high concentration of cutin in needles may have a long legacy of stabilized aliphatic soil C in this forest and help buffer soil C from environmental changes.

Soil C stability. The fate of C added to the soil through woody debris is complex and the net effect on soil C storage is unclear. Wood additions (which included both fresh chips and highly decayed wood) elevated the amount of light fraction C (Fig. 5) and increased the amount of easily respired (i.e., rapid-turnover) C in the bulk soil relative to the No Inputs treatment (Fig. 3b), but the link between these results was not straightforward. From a C turnover perspective, light fraction was not very different from heavy fraction at H.J. Andrews; both mineralization during incubation and MRT were similar for the density fractions. Sollins *et al*. (2006) sequentially fractionated soil collected near the DIRT plots into six fractions of increasing density and found that the MRT of OC in the lower densities ($<2.3\,g\,cm^{-3}$) was <210 years. Only in the fractions of higher density ($>2.3\,g\,cm^{-3}$) did the MRT of OC increase to >680 years, but these fractions comprised $<5\%$ of bulk soil OC. Although our method of fractionation did not separate out OC with different rates of turnover, our fractions are representative of the vast majority of soil OC at this site.

Because Sulzman *et al*. (2005) found that both added wood and needle litter increased rates of CO_2 efflux in field measurements compared with the Control, we expected the addition of needles to similarly result in increased cumulative respiration during incubation. However, mineralization during incubation was lower in the Double Needle treatment than the Double Wood treatment, and was equal to the amount from the root removal treatments (Fig. 3b). Priming *in situ*, as reported to occur by Sulzman *et al*. (2005), would presumably have released the more easily degraded C forms and left behind residues that were more resistant to microbial processing; this is the material that was incubated in the laboratory. We also found that the decay state of the soil SVCi-Lignin from the Double Litter treatment was greater than the Control indicating that it is more decomposed, although not significantly (Table 3). Both of these observations are consistent with the idea that added needles, which are more N-rich than woody debris, resulted in a priming effect on the soil microbial community and accelerated processing of some stored soil carbon (see Crow *et al*., in press for further discussion).

Synthesis: Implications for global change

Plant-derived compounds that are not lost during early stages of decomposition on the forest floor have the potential to be stabilized in soil via organo-mineral interactions, protection with soil aggregate structure, or continued chemical recalcitrance in the soil (Sollins *et al*., 1996; Lützow *et al*., 2006). Although the concept of inherent biochemical recalcitrance as a significant means of stabilizing soil OC is increasingly called into question (Marschner *et al*., 2008), the slower decay kinetics and physiochemical properties of some plant-

derived compounds are thought to enhance their longer term stabilization capacity once in the soil, depending on the edaphic properties (Filley *et al*., 2008a). For example, plant components such as lignin and some aliphatic-based compounds are considered recalcitrant during litter decay (Berg & Meentemeyer, 2002). Further, lignin and aliphatics are largely hydrophobic and therefore likely to be sorbed to mineral surfaces (Kleber *et al*., 2007) and originate directly from plant inputs that aid in the creation and maintenance of soil structure (Jastrow *et al*., 2007); all of which contribute to potential stabilization in soil. Thus, the balance between input rates and the preferential loss/preservation of plant-derived compound is important for understanding dominant SOM sources and the implications of future global changes in climate, land-use, and disturbance.

Owing largely to the high nutrient status of the soil at Bousson, biomass allocation is primarily aboveground and root respiration constitutes only 15% of total soil respiration (R. D. Bowden, unpublished data). Although aboveground litter input rates at Bousson are nearly three times those at H.J. Andrews, doubling inputs did not change soil C balance or stability in the top 5 cm of mineral soil. Our results suggest that, in forests such as Bousson, future increases in aboveground NPP are not likely to enhance long-term C storage because aboveground litter is rapidly decomposed and cycled through active soil pools. However, an increase in belowground productivity at Bousson would provide a direct input of C that our results collectively indicated are stabilized in soil. In contrast to aboveground productivity, projected increases in atmospheric CO_2 concentration and subsequent increases in belowground productivity may have a particularly strong positive impact on soil C stabilization in deciduous forests similar to those at Bousson.

At H.J. Andrews, wood-derived lignin may not be uniformly long-lived; but root-derived lignin may be more important than considered previously. Elevated temperature decreased the lignin concentration in fine roots of Douglas fir seedlings, which may lead to a reduction in the role of root-derived lignin phenols in soil over time (Chen *et al*., 2008). However, this shift in root chemistry did not change root decomposition (measured over 1–3 years) and so may not affect the overall stability of soil C. The majority of SFA in the top 5 cm of mineral soil were derived from needles at H.J. Andrews, which is in contrast to other studies and our results from Bousson. Because aliphatic compounds generally accumulate in soil (e.g., Baldock *et al*., 1997; Nierop, 1998; Kramer *et al*., 2003), a small, sustained increase in needle production in old-growth coniferous forests such as H.J. Andrews may increase stable soil C pool size and offset any positive soil priming effects induced by additional aboveground litter inputs. In fact, Feng *et al*. (2008) reported a significant increase in the abundance of cutin-derived compounds relative to other plant biopolymers in a temperate mixed maple and cedar forest in eastern Canada, due in part to soil priming and to increased leaf production, under experimental warming conditions.

Increased ecosystem production (due to increased plant productivity) clearly does not translate simply and directly into increased soil C storage at these two forested sites. Further, altered allocation between above- and belowground plant tissues may have different effects over different timescales on soil C storage depending on the forest type. Differences in the dominant sources of stabilized OM among forest ecosystems are likely to be important in determining changes in soil C sequestration, and hence must be considered in considering long-term changes in forest soil C dynamics.

Acknowledgements

We acknowledge Dr. Elizabeth Sulzman for her dedication to the DIRT Experiment and her years of mentoring, advice, and enthusiasm without which these ideas, and many more like them, may never have developed fully. She provided invaluable support during planning, fieldwork, and laboratory analysis of this work and we dedicate this paper to the memory of our friend and colleague.

We thank three anonymous reviewers and Thimo Klotzbuecher for helpful comments on early versions of the manuscript. This project was supported by the National Research Initiative of the USDA Cooperative State Research, Education and Extension Service, grant number 2002-35107-12249 and by National Science Foundation (NSF), Division of Environmental Biology (DEB) grant number 0087081. Support to H.J. Andrews Experimental Forest and to this project was provided by H. J. Andrews Long Term Ecological Research program, funded by NSF-DEB. This work was performed in part under the auspices of the US Department of Energy by University of California, Lawrence Livermore National Laboratory under Contract No. W-7405-Eng-48. Sam Reese and Nick Baldauf aided in the fieldwork at Bousson. Additional support for the biopolymer analysis was provided by National Science Foundation (NSF), Division of Environmental Biology (DEB-grant number 0316523) and we thank D. Gamblin of Purdue University for sharing his expertise and contributing to the CuO analysis. This is Purdue Climate Change Research Center paper #0831.

References

Bahri H, Dignac MF, Rumpel C, Rasse DP, Chenu C, Mariotti A (2006) Lignin turnover kinetics in an agricultural soil is monomer specific. *Soil Biology and Biochemistry*, **38**, 1977–1988.

Baldock JA, Oades JM, Nelson PN, Skene TM, Golchin A, Clarke P (1997) Assessing the extent of decomposition of natural

organic materials using solid-state ^{13}C NMR spectroscopy. *Australian Journal of Soil Research*, **35**, 1061–1083.

Berg B, Meentemeyer V (2002) Litter quality in a north European transect versus carbon storage potential. *Plant and Soil*, **242**, 83–92.

Boone RD, Nadelhoffer KJ, Canary JD, Kaye JP (1998) Roots exert a strong influence on the temperature sensitivity of soil respiration. *Nature*, **396**, 570–572.

Bowden RD, Nadelhoffer KJ, Boone RD, Melillo JM, Garrison JB (1993) Contributions of aboveground litter, belowground litter, and root respiration to total soil respiration in a temperate mixed hardwood forest. *Canadian Journal of Forest Research*, **23**, 1402–1407.

Bowden RD, Rullo G, Stevens GR, Steudler PA (2000) Soil fluxes of carbon dioxide, nitrous oxide, and methane at a productive temperate deciduous forest. *Journal of Environmental Quality*, **29**, 268–276.

Bull ID, Nott CJ, van Bergen PF, Poulton PR, Evershed RP (2000) Organic geochemical studies of soils from the Rothamsted Classical Experiments – VI. The occurrence and source of organic acids in an experimental grassland soil. *Soil Biology and Biochemistry*, **32**, 1367–1376.

Chen H, Rygiewicz PT, Johnson MG, Harmon ME, Tian H, Tang JW (2008) Chemistry and long-term decomposition of roots of Douglas-fir grown under elevated atmospheric carbon dioxide and warming conditions. *Journal of Environmental Quality*, **37**, 1327–1336.

Crow SE, Filley TR, McCormick M, Szlavecz K, Stott DE, Gamblin D, Conyers G (2009) Invasive earthworms and forest successional stage interact to impact plant litter inputs and particulate organic matter chemistry. *Biogeochemistry*, doi: 10.1007/s10533-008-9260-1.

Crow SE, Swanston CW, Lajtha K, Brooks JR, Keirstead H (2007) Density fractionation of forest soils: methodological questions and interpretation of incubation results and turnover time in an ecosystem context. *Biogeochemistry*, **85**, 69–90.

Dalzell BJ, Filley TR, Harbor JM (2005) Flood pulse influences on terrestrial organic matter export from an agricultural watershed. *Journal of Geophysical Research-Biogeosciences*, **110**, G02011, doi: 10.1029/2005JG000043.

Dignac MF, Rumpel C (2006) Relative distributions of phenol dimers and hydroxy acids in a cultivated soil and above ground maize tissue. *Organic Geochemistry*, **37**, 1634–1638.

Ertel JR, Hedges JI (1984) The lignin component of humic substances - distribution among soil and sedimentary Humic, Fulvic, and Base-Insoluble fractions. *Geochimica Et Cosmochimica Acta*, **48**, 2065–2074.

Feng XJ, Simpson AJ, Wilson KP, Williams DD, Simpson MJ (2008) Increased cuticular carbon sequestration and lignin oxidation in response to soil warming. *Nature Geoscience*, **1**, 836–839.

Feng XJ, Simpson MJ (2007) The distribution and degradation of biomarkers in Alberta grassland soil profiles. *Organic Geochemistry*, **38**, 1558–1570.

Filley TR, Boutton TW, Liao JD, Jastrow JD, Gamblin DE (2008a) Chemical changes to nonaggregated particulate soil organic matter following grassland-to-woodland transition in a subtropical savanna. *Journal of Geophysical Research*, **113**, G03009, doi: 10.1029/2007JG000564.

Filley TR, Cody GD, Goodell B, Jellison J, Noser C, Ostrofsky A (2002) Lignin demethylation and polysaccharide decomposition in spruce sapwood degraded by brown rot fungi. *Organic Geochemistry*, **33**, 111–124.

Filley TR, McCormick MK, Crow SE, Szlavecz K, Whigham DF, Johnston CT, van den Heuvel RN (2008b) Comparison of the chemical alteration trajectory of *Liriodendron tulipifera L.* leaf litter among forests with different earthworm abundance. *Journal of Geophysical Research*, **113**, G01027, doi: 10.1029/2007JG000542.

Gaudinski JB, Trumbore SE, Davidson EA, Cook AC, Markewitz D, Richter DD (2001) The age of fine-root carbon in three forests of the eastern United States measured by radiocarbon. *Oecologia*, **129**, 420–429.

Gaudinski JB, Trumbore SE, Davidson EA, Zheng SH (2000) Soil carbon cycling in a temperate forest: radiocarbon-based estimates of residence times, sequestration rates and partitioning of fluxes. *Biogeochemistry*, **51**, 33–69.

Goñi MA, Hedges JI (1990a) Cutin-derived Cuo Reaction-products from purified cuticles and tree leaves. *Geochimica Et Cosmochimica Acta*, **54**, 3065–3072.

Goñi MA, Hedges JI (1990b) Potential applications of Cutin-derived Cuo reaction-products for discriminating vascular plant sources in natural environments. *Geochimica Et Cosmochimica Acta*, **54**, 3073–3081.

Goñi MA, Hedges JI (1990c) The diagenetic behavior of Cutin acids in Buried Conifer needles and sediments from a coastal marine-environment. *Geochimica Et Cosmochimica Acta*, **54**, 3083–3093.

Greenland DJ, Ford GW (1964) Separation of partially humified organic materials by ultrasonic dispersion. *Eighth International Congress of Soil Science, Transactions*, **3**, 137–148.

Harmon ME, Baker GA, Spycher G, Greene SE (1990) Leaf litter decomposition in the Picea/Tsuga forests of Olympic National Park, Washington, U.S.A. *Forest Ecology and Management*, **31**, 55–66.

Hedges JI, Ertel JR (1982) Characterization of lignin by gas capillary chromatography of cupric oxide oxidation-products. *Analytical Chemistry*, **54**, 174–178.

Hedges JI, Mann DC (1979) Characterization of plant-tissues by their lignin oxidation-products. *Geochimica Et Cosmochimica Acta*, **43**, 1803–1807.

Hua Q, Barbetti M (2004) Review of tropospheric bomb ^{14}C data for C cycle modeling and age calibration purposes. *Radiocarbon*, **46**, 1273–1298.

Jackson RB, Mooney HA, Schulze ED (1997) A global budget for fine root biomass, surface area, and nutrient contents. *Proceedings of the National Academy of Sciences of the United States of America*, **94**, 7362–7366.

Jastrow JD, Amonette JE, Bailey VL (2007) Mechanisms controlling soil carbon turnover and their potential application for enhancing carbon sequestration. *Climatic Change*, **80**, 5–23.

Jastrow JD, Miller RM, Matamala R, Norby RJ, Boutton TW, Rice CW, Owensby CE (2005) Elevated atmospheric carbon dioxide increases soil carbon. *Global Change Biology*, **11**, 2057–2064.

Jobbagy EG, Jackson RB (2003) Patterns and mechanisms of soil acidification in the conversion of grasslands to forests. *Biogeochemistry*, **64**, 205–229.

Kögel I (1986) Estimation and decomposition pattern of the lignin component in forest humus layers. *Soil Biology and Biochemistry*, **18**, 589–594.

Kögel-Knabner I (2002) The macromolecular organic composition of plant and microbial residues as inputs to soil organic matter. *Soil Biology & Biochemistry*, **34**, 139–162.

Kögel-Knabner I, Ziegler F, Riederer M, Zech W (1989) Distribution and decomposition pattern of Cutin and Suberin in Forest Soils. *Zeitschrift Fur Pflanzenernahrung Und Bodenkunde*, **152**, 409–415.

Kiem R, Kögel-Knabner I (2003) Contribution of lignin and polysaccharides to the refractory carbon pool in C-depleted arable soils. *Soil Biology and Biochemistry*, **35**, 101–118.

King JS, Pregitzer KS, Zak DR, Holmes WE, Schmidt K (2005) Fine root chemistry and decomposition in model communities of north-temperate tree species show little response to elevated atmospheric CO2 and varying soil resource availability. *Oecologia*, **146**, 318–328.

Kleber M, Sollins P, Sutton R (2007) A conceptual model of organo-mineral interactions in soils: self-assembly of organic molecular fragments into zonal structures on mineral surfaces. *Biogeochemistry*, **85**, 9–24.

Kramer MG, Sollins P, Sletten RS, Swart PK (2003) N isotope fractionation and measures of organic matter alteration during decomposition. *Ecology*, **84**, 2021–2025.

Levin I, Kromer B (2004) The tropospheric (CO_2)-C-14 level in mid-latitudes of the Northern Hemisphere (1959-2003). *Radiocarbon*, **46**, 1261–1272.

Lorenz K, Lal R, Preston CM, Nierop KGJ (2007) Strengthening the soil organic carbon pool by increasing contributions from recalcitrant aliphatic bio(macro)molecules. *Geoderma*, **142**, 1–10.

Lorenz K, Preston CM, Krumrei S, Feger KH (2004) Decomposition of needle/leaf litter from Scots pine, black cherry, common oak and European beech at a conurbation forest site. *European Journal of Forest Research*, **123**, 177–188.

Lützow Mv, Kogel-Knabner I, Ekschmitt K, Matzner E, Guggenberger G, Marschner B, Flessa H (2006) Stabilization of organic matter in temperate soils: mechanisms and their relevance under different soil conditions - a review. *European Journal of Soil Science*, **57**, 426–445.

Marschner B, Brodowski S, Dreves A *et al*. (2008) How relevant is recalcitrance for the stabilization of organic matter in soils? *Journal of Plant Nutrition and Soil Science*, **171**, 91–110.

Matamala R, Gonzalez-Meler MA, Jastrow JD, Norby RJ, Schlesinger WH (2003) Impacts of fine root turnover on forest NPP and soil C sequestration potential. *Science*, **302**, 1385–1387.

Matzke K, Riederer M (1991) A comparative-study into the chemical constitution of Cutins and Suberins from Picea-Abies (L) Karst, Quercus-Robur L, and Fagus-Sylvatica L. *Planta*, **185**, 233–245.

Monnier G, Turc L, Jeanson-Luusinang C (1962) Une nouvelle méthode de fractionnement densimétrique par centrifugation des matières organiques du sol. *Annales Agronomiques*, **13**, 55–63.

Nadelhoffer KJ (1990) Microlysimeter for measuring nitrogen mineralization and microbial respiration in aerobic soil incubations. *Soil Science Society of America Journal*, **54**, 411–415.

Nadelhoffer KJ, Boone RD, Bowden RD *et al*. (2004) In: *Forests in Time: The Environmental Consequences of 1000 Years of Change in New England* (eds Foster D, Aber J), pp. 300–315. Yale University Press, New Haven, CT.

Nielson GA, Hole FD (1963) A study of the natural processes of incorporation of organic matter into soil in the University of Wisconsin Arboretum. *Wisconsin Academy of Sciences, Arts, and Letters*, **52**, 213–227.

Nierop KGJ (1998) Origin of aliphatic compounds in a forest soil. *Organic Geochemistry*, **29**, 1009–1016.

Nierop KGJ, Naafs DFW, Verstraten JM (2003) Occurrence and distribution of ester-bound lipids in Dutch coastal dune soils along a pH gradient. *Organic Geochemistry*, **34**, 719–729.

Norby RJ, DeLucia EH, Gielen B *et al*. (2005) Forest response to elevated CO_2 is conserved across a broad range of productivity. *Proceedings of the National Academy of Sciences*, **102**, 18052–18056.

Norby RJ, Ledford J, Reilly CD, Miller NE, O'Neill EG (2004) Fine-root production dominates response of a deciduous forest to atmospheric CO_2 enrichment. *Proceedings of the National Academy of Sciences*, **101**, 9689–9693.

Paul EA, Morris SJ, Conant RT, Plante AF (2006) Does the acid hydrolysis-incubation method measure meaningful soil organic carbon pools? *Soil Science Society of America Journal*, **70**, 1023–1035.

Rasse DP, Rumpel C, Dignac MF (2005) Is soil carbon mostly root carbon? Mechanisms for a specific stabilisation. *Plant and Soil*, **269**, 341–356.

Riederer M, Matzke K, Ziegler F, Kögel-Knabner I (1993) Occurrence, distribution and fate of the lipid plant biopolymers Cutin and Suberin in temperate forest soils. *Organic Geochemistry*, **20**, 1063–1076.

Schimel D (1995) Terrestrial ecosystems in the carbon cycle. *Global Change Biology*, **1**, 77–91.

Sollins P, Homann P, Caldwell BA (1996) Stabilization and destabilization of soil organic matter: mechanisms and controls. *Geoderma*, **74**, 65–105.

Sollins P, Swanston C, Kleber M *et al*. (2006) Organic C and N stabilization in a forest soil: evidence from sequential density fractionation. *Soil Biology and Biochemistry*, **38**, 3313–3324.

Strand AE, Pritchard SG, McCormack ML, Davis MA, Oren R (2008) Irreconcilable differences: fine-root life spans and soil carbon persistence. *Science*, **319**, 456–458.

Stuiver M, Polach HA (1977) Reporting of C-14 Data. *Radiocarbon*, **19**, 355–363.

Stuiver M, Reimer PJ, Braziunas TF (1998) High-precision radiocarbon age calibration for terrestrial and marine samples. *Radiocarbon*, **40**, 1127–1151.

Sulzman EW, Brant JB, Bowden RD, Lajtha K (2005) Contribution of aboveground litter, belowground litter, and rhizosphere respiration to total soil CO_2 efflux in an old growth coniferous forest. *Biogeochemistry*, **73**, 231–256.

Swanston CW, Caldwell BA, Homann PS, Ganio L, Sollins P (2002) Carbon dynamics during a long-term incubation of separate and recombined density fractions from seven forest soils. *Soil Biology and Biochemistry*, **34**, 1121–1130.

Trumbore SE, Davidson EA, Decamargo PB, Nepstad DC, Martinelli LA (1995) Belowground cycling of carbon in forests and pastures of Eastern Amazonia. *Global Biogeochemical Cycles*, **9**, 515–528.

USDA Soil Conservation Service (1979) *Crawford Country Soil Survey*. USDA-SCS, Washington, DC.

Van der Heyden D, Skelly J, Innes J, Hug C, Zhang J, Landolt W, Bleuler P (2001) Ozone exposure thresholds and foliar injury on forest plants in Switzerland. *Environmental Pollution*, **111**, 321–331.

Vanderbilt KL, Lajtha K, Swanson F (2003) Biogeochemistry of unpolluted forested watersheds in the Oregon Cascades: temporal patterns of precipitation and stream nitrogen fluxes. *Biogeochemistry*, **62**, 87–117.

Vogel JS, Southon JR, Nelson DE, Brown TA (1984) Performance of catalytically condensed carbon for use in accelerator mass-spectrometry. *Nuclear Instruments and Methods in Physics Research Section B-Beam Interactions with Materials and Atoms*, **233**, 289–293.

Yang YS, Chen GS, Lin P, Xie JS, Guo JF (2004) Fine root distribution, seasonal pattern and production in four plantations compared with a natural forest in Subtropical China. *Annals of Forest Science*, **61**, 617–627.

Yano Y, Lajtha K, Sollins P, Caldwell BA (2004) Chemical and seasonal controls on the dynamics of dissolved organic matter in a coniferous old-growth stand in the Pacific Northwest, USA. *Biogeochemistry*, **71**, 197–223.