Dynamics of decadally cycling carbon in subsurface soils

Jun Koarashi,[1,2] William C. Hockaday,[3,4] Caroline A. Masiello,[3] and Susan E. Trumbore[1,5]

[1]   Subsurface horizons contain more than half of the global soil carbon (C), yet the dynamics of this C remains poorly understood. We estimated the amount of decadally cycling subsurface C ($\sim$20 to 60 cm depth) from the incorporation of 'bomb' radiocarbon (^{14}C) using samples taken over 50 years from grassland and forest soils in the Sierra Nevada Mountains, California. The radiocarbon content of all organic matter fractions (roots, low-density (LF), high-density (HF), and non-oxidizable HF) increased from the pre- to post-bomb samples, indicating $\sim$1–6 kgC m^{-2}, or about half of the subsoil C, consists of C fixed since 1963. Low-density (LF-C) represented <24% (grassland) to 40–55% (forest) of the subsurface C and represented a mixture of post-bomb C and varying amounts of pre-1950 charcoal, identified using ^{13}C-NMR spectroscopy. The ^{14}C content of HF-C increased rapidly from 1992 to 2009, indicating a significant time lag (>20 years) for the arrival of 'bomb' ^{14}C to this fraction. A two-pool (fast-cycling and passive) model including >20 year time lag showed that 28–73% of the subsoil mineral-associated C had turnover times of 10–95 years. Microbially respired C was enriched in bomb ^{14}C compared to both LF and HF fractions in 2009. Overall, we estimate that C fluxes through decadally cycling pools in the subsurface are equivalent to 1–9% (grassland) to 10–54% (forest) of the surface litterfall at these sites. Our results demonstrate the importance of decadally cycling C for ecosystem C balance, and that a lagged response of the large subsurface C stores to changes in environmental conditions is possible.

1.   Introduction

[2]   Soils contain large stocks of organic carbon (OC) that may be vulnerable to destabilization with changes in environmental conditions. Recent work using coupled carbon cycle-climate models suggests that the soil feedback to climate change could lead to a doubling of the projected warming by the end of this century [*Jenkinson et al.*, 1991; *Cox et al.*, 2000; *Jones et al.*, 2003]. While these experiments illustrate the potential sensitivity of climate change projections to soil C dynamics, there is a great deal of mechanistic uncertainty that underlies simulations of soil responses to climate.

[3]   Most studies and models of soil C and its response to climate and land-use changes have focused on the upper 20–30 cm of soil (surface horizons, nominally the A or Ap horizon in most soils). This is understandable because higher C concentrations and greater microbial activity are usually found in these horizons than in the underlying subsurface (normally, B) horizons. However, soil organic carbon (SOC) stored in the subsurface horizons has received increasing interest in recent years [*Rumpel and Kögel-Knabner*, 2011], mainly for two reasons. First, soil C inventory studies have shown that a large fraction (>50%) of total soil C is located in the subsurface horizons in a wide range of ecosystems [*Batjes*, 1996; *Jobbágy and Jackson*, 2000]. Second, recent observations show incorporation of 'bomb' ^{14}C produced by atmospheric weapons testing in subsurface organic matter [*Trumbore et al.*, 1995; *Richter et al.*, 1999; *Baisden et al.*, 2002a; *Baisden and Parfitt*, 2007], in apparent contradiction with the previous assumptions based on bulk ^{14}C ages of centuries to millennia that C in these horizons was very stable [e.g., *Scharpenseel et al.*, 1989; *Paul et al.*, 1997]. These findings suggest that a fraction of the C in subsurface horizons is dynamic on decadal timescales, and that C cycling associated with this pool can be significant at the global scale. Moreover, increasing evidence that the mechanisms regulating SOC stability vary with depth [*Fierer et al.*, 2003; *Fontaine et al.*, 2007; *Salomé et al.*, 2010] suggest that responses to changes in environmental conditions will differ for surface and subsurface soil C.

[4]   While surface horizons and the early stages of plant litter decomposition dominate C fluxes from soils [*Davidson et al.*, 2006], the longer term (i.e., over the next century)

[1]Department of Earth System Science, University of California, Irvine, California, USA.
[2]Nuclear Science and Engineering Directorate, Japan Atomic Energy Agency, Tokai-mura, Japan.
[3]Department of Earth Science, Rice University, Houston, Texas, USA.
[4]Department of Geology, Baylor University, Waco, Texas, USA.
[5]Max Planck Institute for Biogeochemistry, Jena, Germany.

Corresponding author: J. Koarashi, Nuclear Science and Engineering Directorate, Japan Atomic Energy Agency, Tokai-mura, Ibaraki 319-1195, Japan. (koarashi.jun@jaea.go.jp)

Table 1. Carbon Inventory, Mineralogical Properties, and C Mineralization for the Sierra Transect Subsurface Soils

Soil Series[a]	Fallbrook	Musick	Shaver
Elevation (m)	470	1240	1780
MAT (°C)	17.8	11.1	8.5
MAP (cm)	31	91	101
Vegetation zone	Annual grasses with scattered oaks	Mixed conifer	Mixed conifer
Soil classification	Typic Xerochrept	Ultic Haploxeralf	Pachic Xerumbrept
C in 1958–62 (kgC m^{-2})[b]	3.8 (0.6, 3.2) in 10–112 cm	9.9 (4.0, 5.9) in 18–114 cm	5.8 (3.2, 2.7) in 15–155 cm
C in 1992 (kgC m^{-2})[b]	2.6 (0.2, 2.4) in 12–75 cm	8.8 (4.4, 4.5) in 19–85 cm	5.3 (2.9, 2.5) in 20–100 cm
C in 2005 (kgC m^{-2})[b]	0.5 (0.1, 0.4) in 12–30 cm	6.3 (2.6, 3.7) in 21–65 cm	4.5 (2.5, 2.0) in 20–74 cm
Clay (%)[c]	NA	16–27	6–8
Al$_o$ (g kg^{-1})[c]	NA	0.8–0.9	3.9–5.3
Fe$_d$ (g kg^{-1})[c]	NA	9.3–14.4	4.3–4.6
C mineralization (gC kg^{-1} soil C)[d]	NA	2.8 (0.6)	13.6 (0.7)
C in O+A horizons (kgC m^{-2})[e]	2.5	5.8	7.4

[a]Locations of sites (latitude, longitude): Fallbrook, 36°43′N, 119°17′W; Musick, 37°1′N, 119°16′W; and Shaver, 37°2′N,119°11′W [*Trumbore et al.*, 1996]. Note that the locations of the 2001 sample collections were: Musick, 37°04′N, 119°22′W, 1400 m above sea level; and Shaver, 37°03′N, 119°09′W, 1800 m above sea level, respectively.

[b]Numbers in parenthesis indicate C inventory as LF (low-density <2.0 g cm^{-3}; left) and as HF (high-density >2.0 g cm^{-3}; right), respectively.

[c]Data are for 29–96 cm for Musick and 18–102 cm for Shaver, respectively, from *Dahlgren et al.* [1997]. Al$_o$: Acid oxalate extractable Al, Fe$_d$: Citrate-dithionite extractable Fe.

[d]The amount of C mineralized from <4 mm-sieved, root-free soil samples during the 55-day incubation (see text). Number in parenthesis indicates the standard deviation (N = 3).

[e]Data from *Trumbore et al.* [1996].

responses of soils to environmental change will largely be determined by the dynamics of decadally cycling SOC pools that are stabilized by a range of physical and biological mechanisms. It is generally acknowledged that increased stability with soil depth reflects fundamental differences in the mechanisms of soil organic matter (SOM) stabilization. With depth, increases in the relative amount of mineral-associated C (high density fraction) compared to C relatively free of mineral particles (low density fraction) may interfere with degradation of the SOM by soil microbes [*Sollins et al.*, 1996; *Torn et al.*, 1997; *Baldock and Skjemstad*, 2000; *Six et al.*, 2002; *Masiello et al.*, 2004; *Rasmussen et al.*, 2005; *Swanston et al.*, 2005; *Salomé et al.*, 2010].

[5] Despite the critical role of the decadally cycling C in the global C balance, our understanding of the dynamics, and even the size and properties of this C in subsurface horizons remains extremely fragmentary [*Rumpel and Kögel-Knabner*, 2011]. *Trumbore et al.* [1996] used only two sampling points in time: 1958 and 1992 for studying the C dynamics in the surface (O + A) soil horizons. By following the evolution of bomb radiocarbon for an additional decade and specifically focusing on the subsurface horizons, we are able to examine a previously untractable problem – the turnover time of slower-cycling C. Here, we address the following questions about the C dynamics in subsurface soil horizons: (1) To what extent does subsurface SOC participate in decadal C cycling? (2) What is the dynamics of the decadally cycling C pool in subsurface soils? (3) What is the role of soil minerals in controlling the decadal C cycling? (4) What is the lag time for the atmospheric C to become incorporated into mineral-associated SOC pool in subsurface soils?

[6] To answer these questions, we determined ^{14}C signatures of a range of soil fractions (density-separated fractions, size-separated low-density fractions, chemically oxidized high-density fractions, and fine roots) in subsurface horizons collected at several points in time over the past half century from one grassland and two forest soils in the western Sierra Nevada, California. We used changes in ^{14}C signatures

in different SOC fractions over the period since peak nuclear weapons testing ended in 1963 to estimate the amount and turnover time of the subsurface soil C that interacts with C fixed from the atmosphere over the last 50 years. The structural chemistry of the relatively mineral-free low-density fractions was determined to explore the reason for the overall old ^{14}C age of the LF fractions in subsurface soils. We also performed laboratory incubations to provide evidence that the primary source of CO_2 evolved from the subsurface soils is dominated by SOC fixed recently from the atmosphere.

2. Materials and Methods

2.1. Study Sites

[7] Soil samples were collected from well-drained soils along an elevation transect of the western slope of the Sierra Nevada, in central California. The climate, vegetation, soil type, and soil carbon stocks for each site are summarized in Table 1. The sites have also been well-studied for soil properties and processes [*Dahlgren et al.*, 1997] and for the C dynamics in the surface (O + A) soil horizons [*Trumbore et al.*, 1996; *Rasmussen et al.*, 2006]. Here, we emphasize changes in radiocarbon in the B horizons (>20 cm depth), which were not previously published, and include information from soil incubations. Briefly, the soils are formed from granodiorite at higher (>1000 m) elevation and tonalite at lower elevation, and vary from Alfisols to Inceptisols with increasing elevation [*Trumbore et al.*, 1996]. The soils are all >60% sand, with maximum clay content and citrate-dithionite extractable Fe at the mid-elevation (Musick) site [*Dahlgren et al.*, 1997]. Of particular interest is the observation of a sharp change in a number of factors such as soil color, pH, type of clay and amount of secondary Fe, between the Musick and the Shaver soil series, which is hypothesized to be associated with the present-day average effective winter snowline (~1600 m) [*Dahlgren et al.*, 1997]. Mean annual temperature decreases linearly with elevation, from 17.8°C at 470 m to 8.5°C at 1780 m [*Trumbore et al.*, 1996]. Precipitation is 31 cm yr^{-1} at 470 m, and ~90–100 cm yr^{-1}

at 1200 to 1800 m. Vegetation is mixed conifer at the Musick and Shaver sites, and annual grasses with scattered oak trees at the Fallbrook site in the Sierra foothills [*Trumbore et al.*, 1996].

2.2. Sample Collection

[8] Sampling locations for each series have been visited several times over the past half-century [*Trumbore et al.*, 1996]. We obtained soil samples that had been collected throughout the profiles (A and B horizons) in 1958–1962, 1992, and 2005, from archives maintained by the Division of Ecosystem Sciences, University of California, Berkeley, and the Department of Earth System Science, University of California, Irvine [*Trumbore et al.*, 1996]. We also obtained samples of the topmost subsurface horizon collected in 2001 about 4 and 10 km away from the Musick and Shaver sites, respectively, from archives maintained by the Department of Soil, Water and Environmental Science, University of Arizona [*Rasmussen et al.*, 2006] (see also Table 1 legend). For the original reoccupation of sites in 1992, we attempted to locate the site used for the original soil pits using site descriptions, and are confident that we were within 100 m of the locations where the archived samples were collected [*Trumbore et al.*, 1996]. We also matched the soil profile descriptions as accurately as possible, so that horizon boundaries were close to those in the original 1950s soil descriptions. Sites in 2005 and 2009 were located based on the 1992 soil pit locations, and in all cases were more clearly co-located; these sites have also been more frequently visited as part of ongoing studies [e.g., *Wang et al.*, 1999, 2000]. In each case, a soil pit was dug, and the face with horizon boundaries and soil description most closely matching previous descriptions was sampled. Samples represented a depth interval corresponding to the horizon descriptions; for B horizons, the 1992 and 2005 horizons were subsampled with depth intervals of 10–12 cm.

[9] We revisited the Musick and Shaver sites in 2009, and collected subsurface horizons (from depths of 55–70 cm and 40–60 cm for the Musick and Shaver sites, respectively) specifically for incubation studies. A portion of the 2009 soil samples was dried at 50°C and sieved (<2 mm) in the same manner as for the archived soil samples, while the remaining soil samples were stored moist at 7°C in tightly sealed plastic bags for about four months until the start of incubations.

2.3. C inventory in Subsurface Soil

[10] Bulk densities determined from the oven-dried weight of known volume samples were available for all horizons of the 1992 soil profile [*Trumbore et al.*, 1996]. The C inventory in a given subsurface soil, $C_{inventory}$ (gC cm^{-2}), is calculated as

$$C_{inventory} = \sum BD \cdot C_{mf} \cdot d \cdot (1 - g) \qquad (1)$$

where BD is the bulk density (g cm^{-3}), C_{mf} is the mass fraction of C in soil material (gC g^{-1} soil), d is the thickness of the horizon (cm), and g is the mass fraction of stones and gravel greater than 2 mm in size (g g^{-1}). Bulk densities of the pre-bomb and 2005 soil profiles were not available, and were estimated based on the 1992 bulk density data.

## 2.4. Soil Fractionation

[11] We physically separated bulk (dried, <2 mm) soil into two density fractions: low-density fraction (LF: <2.0 g cm^{-3})

and high-density fraction (HF: >2.0 g cm^{-3}), based on flotation in a 2.0 g cm^{-3} solution of sodium polytungstate [*Trumbore et al.*, 1995, 1996]. Filters used for separation were pre-combusted quartz filters for the pre-bomb and 1992 samples, paper filters for the 2005 samples, and pre-combusted glass fiber filters for the 2001 and 2009 samples. Dissolved organic carbon lost to sodium polytungstate solution and DI water during the fractionation was not recovered. The C loss was estimated as <10% of the total soil C for the 2009 Musick samples and ∼20% for the 2009 Shaver samples, which is similar to the reported proportions [*Baisden et al.*, 2002b; *Crow et al.*, 2007; *Castanha et al.*, 2008].

[12] Visible fine roots (mixed live + dead) and charred material in the LF were removed by hand prior to carbon isotope analysis. The fine-root samples (<0.5 mm in diameter) were processed using either an acid/alkali/acid procedure to remove easily hydrolyzable carbon such as starches that may post-date formation of root structural C [*Gaudinski et al.*, 2001], or an ultrasonic washing in DI water to remove adhering soil particles. We further applied size separation by sieving to some of the LF (for the 1992 and 2005 samples) to get three size fractions: <63, 63–500 and 500–2000 μm in diameter.

[13] Four HF samples obtained from the pre-bomb and 2009 profiles at the Musick and Shaver sites were subjected to chemical oxidation in an effort to distinguish pools with faster and slower cycling timescales within the HF fractions. Briefly, 1 g of HF sample was wetted with 10 ml DI water for 10 min, and 30 ml of 10% hydrogen peroxide (H$_2$O$_2$) was added [*Helfrich et al.*, 2007]. The oxidation was conducted at 24°C for 24 h. After the oxidation, the suspension was centrifuged for 15 min and the supernatant was decanted. The residue was rinsed repeatedly with 40 ml DI water and freeze-dried. We also applied stronger oxidation to two HF samples obtained from 43 to 65 cm depth of the 2005 Musick soil and 40–59 cm depth of the 1992 Shaver soil. The oxidation conditions were 2–3 days × 3 times at 50°C and 3 days × 1 time at 24°C for the Musick and Shaver samples, respectively.

2.5. Carbon Isotope Analysis

[14] The soil fractions (homogenized by grinding) and fine roots were combusted in evacuated quartz tubes with CuO wires for 2 h at 900°C. The resulting CO$_2$ was cryogenically purified and measured manometrically to allow determination of weight percentage of C of the samples. An aliquot of the purified CO$_2$ was transferred to a He-filled septum-caped vial, and analyzed for δ^{13}C with an isotope ratio mass spectrometer (Thermo Finnigan Delta Plus, USA) [*Czimczik and Trumbore*, 2007; *Xu et al.*, 2007]. Roughly 0.1 to 1 mgC of CO$_2$ was sealed into an evacuated Pyrex tube, and reduced to graphite on iron powder [*Xu et al.*, 2007]. Radiocarbon was measured on graphite targets with the use of accelerator mass spectrometry (AMS) at the W.M. Keck AMS facility at University of California, Irvine [*Southon and Santos*, 2004]. We report ^{14}C data as Δ^{14}C, the per mil (‰) deviation of the ^{14}C/^{12}C ratio of the sample from that of an oxalic acid standard in 1950, with correction for mass-dependent fractionation to a common δ^{13}C of −25‰ [*Stuiver and Polach*, 1977]. Positive Δ^{14}C values indicate the presence of bomb-^{14}C, whereas negative values indicate that on average the carbon has resided in soil long enough to reflect the radioactive decay of cosmogenic ^{14}C. The
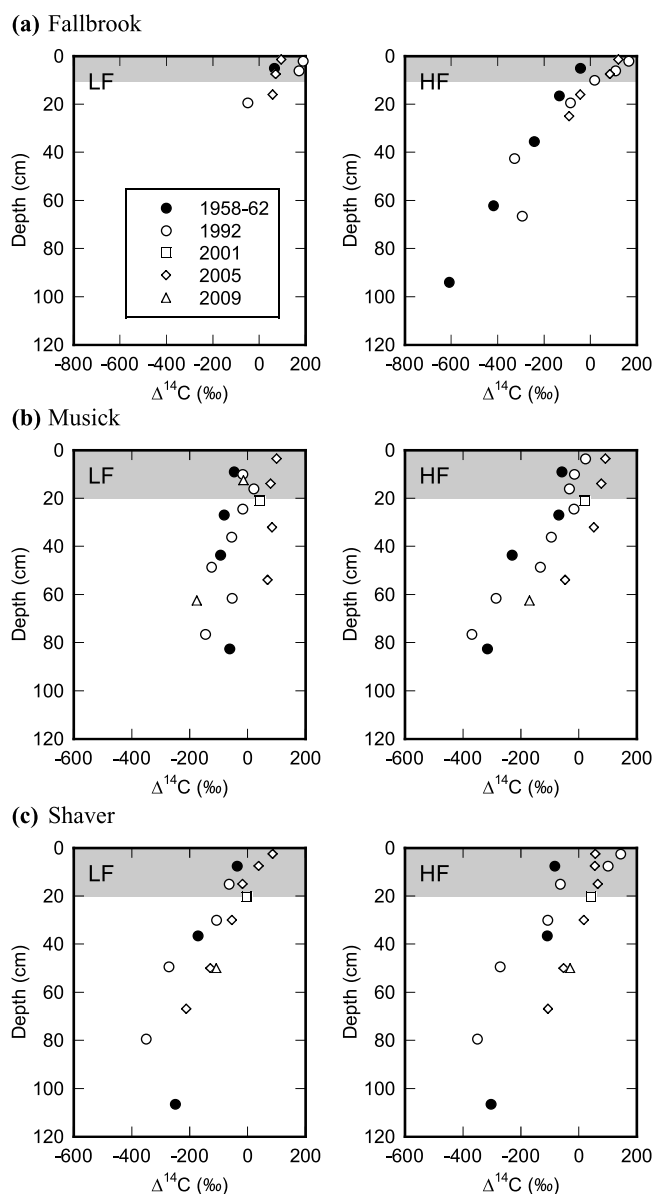
Figure 1. [14]C signatures of (left) LF and (right) HF organic carbon in subsurface soil horizons at the (a) Fallbrook, (b) Musick, and (c) Shaver sites. Shaded areas indicate A horizons.

analytical uncertainty in the $\Delta^{14}C$ values was normally less than 3‰, based on repeated measurements of secondary standard materials [Xu et al., 2007].

2.6. Solid-State ^{13}C NMR Analysis for Size-Separated LFs

[15] Nuclear magnetic resonance (NMR) experiments were conducted for ten size-separated LF samples from the 1992 and 2005 Musick soils and from the 2005 Shaver soils. Of the ten, six were from the subsurface soil horizons, and the remaining four were from the A horizon for comparison.

[16] The NMR spectra were obtained on a 200 MHz Bruker Advance spectrometer with a solid-state 4 mm dual frequency magic angle spinning (MAS) probe, located at the Rice University shared equipment facility. Approximately

40–120 mg of homogenized, size-separated LF samples were packed in a 4 mm (outside diameter) NMR rotor with a Kel-F cap, and spun at an MAS frequency of 5 kHz. The soil C functional group distribution was measured by a cross polarization pulse sequence with a contact time of 1 ms and a recycle delay time of 2.5 s. Approximately 20,000–35,000 acquisitions (scans) were collected for each sample. The data collected were treated with an exponential appodization function and Fourier-transformed with 50–75 Hz line broadening to obtain a spectrum to which we applied manual phase and baseline corrections. We integrated the peak areas according to spectral regions specified in Baldock et al. [2004].

[17] The integrated peak areas were mathematically corrected for spinning sidebands of aromatic, phenolic and amide/carboxyl C. The corrected peak areas were then used in a molecular mixing model developed by Baldock et al. [2004] to calculate the mass percentage of carbohydrate, protein, lignin, lipid, carbonyl, and charcoal in the light fraction (<2 g cm^{-3}) organic matter.

2.7. Incubations

[18] The subsurface soil samples collected at the Musick and Shaver sites in 2009 were sieved (4 mm) while still moist and remaining visible roots were removed by hand. The soil samples (65 g dry weight equivalent, N = 3) were placed in cups, and moisture content of the soils was adjusted to ∼50% water holding capacity (WHC) by adding DI water (100% WHC was determined as the gravimetric water content of water-saturated soil that had been allowed to drain over 6 h in a funnel with filter.). The cups were then placed into 0.5-L Mason jars, and the jars were closed with lids equipped with gas sampling ports. After one day pre-incubation at laboratory temperature (∼24°C), we added 1 ml of DI water into the soils (moisture content increased to 53–54%WHC) and then mixed to remove CO_2 accumulated in soil pores. The jars were closed and the headspace was flushed completely with CO_2-free air, and the soils were incubated for 55 days at laboratory temperature (∼24°C). Soil moisture was maintained during the incubation by the addition of DI water to the bottom of the jars to maintain 100% relative humidity.

[19] The headspace was sampled several times in the 55 day interval to determine CO_2 evolution rates and estimate the time required to build up sufficient C for the radiocarbon determination. CO_2 concentration was determined on a 2 ml subsample using a Licor CO_2 analyzer (LI-6262 CO_2/H_2O Analyzer, Licor) [Davidson and Trumbore, 1995]. On day 55, we transferred the headspace air into a pre-evacuated 0.5 L stainless steel container. We cryogenically purified CO_2 in the collected air, and measured its carbon isotope signatures (^{13}C and ^{14}C) as described above [Czimczik and Trumbore, 2007].

2.8. Modeling Mineral-Associated C Dynamics Using [14]C Measurements

[20] To quantify the dynamics of subsurface HF-C, we focused on the 10–30 cm depth interval of the Fallbrook soil and the 20–60 cm depth interval of the Musick and Shaver soils, respectively, because our data below these depths especially in the 2005 soil profiles were limited (see Figure 1). As not all depth increments sampled were the same for each time point, we estimated the mean $\Delta^{14}C$ values for the target depth intervals, as follows. We calculated the cumulative C inventory from soil surface down to

Table 2. Dynamics of Subsurface Mineral-Associated Soil Carbon Fractions Derived From ^{14}C Measurements and a Two-Pool Model

Site	Depth Interval (cm)	HF-C Stock (kgC m^{-2})	Lag Time (yr)	Turnover Time of Passive C (yr)	Turnover Time of Fast-Cycling C (yr)	Proportion Fast-Cycling C	Predicted C Mineralization Rate (gC m^{-2} yr^{-1})
Fallbrook	10–30	0.49–1.30	25	3400^a–6700	24–62	0.49–0.67	5.3–29.9
			30	2200–6700	16–55	0.28–0.67	6.0–23.1
			35	2200–6700	14–43	0.28–0.67	7.6–26.4
			40	3400^a–6700	16–25	0.48–0.67	13.1–39.3
Musick	20–60	2.60–3.60	30	4400–6700	14–47	0.55–0.73	40.6–141.8
			35	4400–6700	13–37	0.55–0.73	51.5–152.6
			40	4400–6700	10–22	0.60–0.73	86.3–216.4
Shaver	20–60	1.20–1.69	0	3400	42–81	0.61–0.67	10.1–26.0
			5	2200–3400	29–95	0.46–0.69	8.9–27.2
			10	2200–3400	30–95	0.46–0.69	8.9–26.4
			15	2200–3400	27–94	0.45–0.69	8.9–28.6
			20	2200–3400	24–91	0.45–0.68	9.2–32.1
			25	2200–3400	24–80	0.45–0.68	10.3–32.1
			30	2200–3400	21–70	0.44–0.67	11.8–35.8
			35	2200–3400	23–52	0.49–0.67	15.6–36.3
			40	2200–3400	17–31	0.54–0.67	26.9–54.1

^{a}Turnover time of 3400 yr instead of 2200 yr was used for passive C pool when the model shows no result for explaining the observed ^{14}C change.

the depth by using the C inventory data for the sampled layers, and fitted a linear or a logarithmic function to the cumulative C inventory versus depth data for the depths covering the target depth intervals. ^{14}C inventory for the sampled layers was also calculated as C × Δ^{14}C from the observed ^{14}C value and the estimated C inventory. Then, we fitted a slope to the cumulative ^{14}C inventory versus depth data, and estimated the mean Δ^{14}C values for the target depth intervals by dividing ^{14}C inventory in the intervals by C inventory there. We also estimated the uncertainty in the Δ^{14}C values based on 95% confidence interval as defined by the linear regression.

[21] For the target depth intervals, we compared the observed change in Δ^{14}C values with the change predicted with a time-dependent, two-pool model [*Trumbore et al.*, 1996; *Baisden et al.*, 2002b; *Torn et al.*, 2009]. The model consists of a fast-cycling C pool with decadal turnover times, and a passive C pool with millennial turnover times, both of which are homogeneous pools with a first-order decay behavior. For each year (t) from 1900 through 2009, we calculated the fraction modern carbon of the HF, $F_{HF(t)}$, equal to $\Delta^{14}C_{(t)}/1000 + 1$, as

$$F_{HT(t)} = P_f\left[F_{f(t-1)}\left(1 - k_f - \lambda\right) + k_f F_{atm}(t - \tau)\right] \\ + \left(1 - P_f\right)\left[F_{p(t-1)}\left(1 - k_p - \lambda\right) + k_p F_{atm}(t - \tau)\right] \quad (2)$$

where P_f is the proportion of the fast-cycling C pool, $F_{f(t-1)}$ and $F_{p(t-1)}$ are the fraction modern carbon in the fast-cycling and passive C pools in the previous 1-yr time step, k_f and k_p are decomposition rate constants for the fast-cycling and

passive C pools (yr^{-1}), and λ is the radioactive decay constant for ^{14}C (1.21 × 10^{-4} yr^{-1}). $F_{atm}(t - \tau)$ tracks the fraction modern of carbon inputs to the HF fraction, and is derived from the atmospheric ^{14}C record ($F_{atm}(t)$) of *Levin and Kromer* [2004], supplemented with more recent data by personal communication from *I. Levin* of Heidelberg University.

[22] To account for the time elapsed between production of fresh plant tissues and the appearance of that C in the HF fraction, we apply a time lag, τ (yr), ranging from 0 to 40 years (5 years interval, see the fourth column of Table 2). The time lag could reflect a range of processes, including long-lived root inputs or downward DOC transport, which add C to deeper layers. The Δ^{14}C of the HF in year t is then calculated as $1000 \times (F_{HF(t)} - 1)$, and the turnover times of the fast-cycling C pool is as k^{-1}.

[23] We estimated the passive pool Δ^{14}C values from the measured radiocarbon signatures of the residues following chemical oxidation of the HF fraction. The Δ^{14}C values of the residue after moderate oxidation for the pre-bomb samples were −351‰ for 51–114 cm in the Musick pre-bomb soil and −215‰ for 15–58 cm in the Shaver soil (Table 3), which correspond to turnover times of ~4400 yr and ~2200 yr, respectively, for the assumed steady state reservoirs [*Torn et al.*, 2009]. These estimates represent the minimum turnover time for the passive pool. The ^{14}C signature of moderate oxidation residues increase from the pre-bomb to 2009 samples, indicating that the oxidation residue consists of a mix of faster cycling C (that incorporated bomb ^{14}C) and even slower cycling C.

Table 3. ^{14}C Signatures of HF-C Retained and Oxidized by Hydrogen Peroxide Treatment

Site	Year	Depth (cm)	Oxidation Condition^a	Retained HF-C (%)	Oxidized HF-C (%)	Δ^{14}C (‰)		
						Untreated	Retained	Oxidized
Musick	1958	51–114	Moderate	51	49	−315	−351	−278
	2009	55–70	Moderate	49	51	−169	−225	−116
	2005	43–65	Strong A	4	96	−46	−452	−27
Shaver	1959	15–58	Moderate	45	55	−108	−215	−19
	2009	40–60	Moderate	42	58	−30	−157	+61
	1992	40–59	Strong B	36	64	−147	−294	−64

^{a}Moderate: 1 day at 24°C, Strong A: 2–3 days × 3 times at 50°C, and Strong B: 3 days × 1 time at 24°C.

Table 4. Chemical Characterization of Size-Separated LF Carbon[a]

Year	Site	Depth (cm)	Particle Size (μm)	Carbohydrate (%)	Lipid (%)	Lignin (%)	Char (%)	Protein and Carbonyl (%)	Alkyl: O-Alkyl
1992	Musick	13–19	<63	19	14	23	22	23	0.99
		13–19	500–2000	28	7	24	22	20	0.44
		19–30	<63	19	15	24	20	22	1.01
		19–30	500–2000	28	11	32	16	12	0.57
2005	Musick	21–43	<63	25	17	23	12	23	1.00
		21–43	63–500	27	15	28	11	20	0.84
2005	Shaver	10–20	<63	25	18	13	19	25	1.04
		10–20	500–2000	17	6	12	41	25	0.47
		40–60	<63	20	17	16	28	19	0.97
		40–60	500–2000	25	7	8	43	17	0.44

[a]Values determined by solid-state cross-polarization ^{13}C NMR and the molecular mixing model [*Baldock et al.*, 2004].

A stronger oxidation treatment of the 2009 soils in the target depths of the B horizons removed more HF-C (96 and 64%) than the moderate oxidation (49–58%), and the retained HF-C were more depleted in ^{14}C (−452‰ and −294‰, respectively) (Table 3). We estimated the maximum turnover time for the passive pool in the target depths using the Δ^{14}C values of residues from the stronger oxidation, ~6700 yr and ~3400 yr for the Musick and Shaver soils, respectively (see the fifth column of Table 2). For the Fallbrook site, the minimum and maximum turnover times from the other sites (2200 and 6700 yr), were assumed to bracket the passive pool turnover time. The C mineralization flux from the HF-C pool in the modeled layer (in gC m^{-2} yr^{-1}) is calculated as [$k_f P_f + k_p (1 - P_f)] \times C_{HF}$, where C_{HF} is the steady state C inventory of HF in the layer (gC m^{-2}).

3. Results

3.1. C Inventory in the Subsurface Soil

[24] The C inventory in the subsurface soils ranged from 0.5 to 9.9 kgC m^{-2} (Table 1). Although the total depth interval of subsurface soil layers collected from pits differed among the pre-bomb, 1992, and 2005 soil profiles, it was clear that more C was stored in the higher elevation forest soils than in the grassland soil. The LF-C was a large fraction (40–55%) of the C in the subsurface soils at the forest sites, but was a minor fraction (<24%) at the grassland site (Table 1, numbers in parentheses). The maximum C storage was found both in the LF and HF at the mid-elevation Musick site where the highest clay and citrate-dithionite extractable Fe (Fe$_d$) content were also observed [*Dahlgren et al.*, 1997]. Despite decreased C concentration with depth (data not shown), at all sites the C inventory in the subsurface soils was roughly comparable to that in the O and surface (A) horizons combined [*Trumbore et al.*, 1996].

3.2. ^{14}C Signatures of LF-C

[25] The LF-C showed Δ^{14}C values <0‰ at all subsurface depths in the pre-bomb soil profiles (filled symbols in non-shaded areas in Figure 1), and generally remained depleted in ^{14}C even in the post-bomb profiles (open symbols in Figure 1). Some exceptions showing positive Δ^{14}C values were found at the upper parts of the subsurface soils in the Fallbrook and Musick sites. Note again that positive Δ^{14}C values indicate the presence of bomb-^{14}C (young, decades-old C) and negative, lower Δ^{14}C values indicate older

carbon. The LF Δ^{14}C value increased in the upper parts of the B horizon between pre- and post-bomb soils, but did not necessarily increase in the deeper layers.

3.3. ^{14}C Signatures of HF-C

[26] Overall, the Δ^{14}C values of the HF-C decreased with depth (Figure 1). Like the LF-C, all HF-C showed negative Δ^{14}C value (filled symbols in non-shaded areas) in the pre-bomb soil profiles. The ^{14}C signature in nearly all the HF-C increased between the pre- and post-bomb subsurface soils (open symbols). The trends of decreasing ^{14}C with depth and of increasing ^{14}C between points in time spanning the bomb-^{14}C period both appeared more clearly in HF-C than in LF-C. In general, the Δ^{14}C value of the HF-C in the subsurface soil layers was equal to or lower (i.e., older) than that of the LF-C at the Fallbrook and Musick site, but was in contrast higher (younger) at the Shaver site. Positive Δ^{14}C values were observed even in the HF-C in the upper parts of the post-bomb subsurface soils at all sites.

### 3.4. Structural Chemistry and ^{14}C Signatures of Size-Separated LF-C

[27] The ^{13}C NMR analyses showed clear differences in the molecular components making up LF-C between the two forested sites (Table 4; see also Figure 2 for examples of NMR spectra). The Shaver LF-C has more charcoal C and less lignin C than the Musick soil, especially in larger size fraction. The smaller size fractions (<63 micron) generally had more lipid C and less carbohydrate C than larger size fractions of low density material (Table 4). Alkyl:O-alkyl ratios (often used as a measure of degree of decomposition [*Baldock et al.*, 1997]) increase with depth (higher ratios) and decrease with particle size (Table 4). The exception is the Shaver 2005 soil, where chemical composition remains unchanged with depth in both size fractions, and where relatively 'fresh' detritus appears to be mixed with large quantities of charcoal.

[28] The ^{14}C measurements of the size-separated LF fractions differed for the two forest sites. The Shaver LFs got older with increasing size, but the Musick LFs got younger (Figure 3). A strong negative correlation (Figure 3; $r^2 = 0.79$, $p < 0.001$) was found between the Δ^{14}C values and charcoal C content of the LFs, irrespective of the sample origin (site, depth, and size class). The LF with the lowest charcoal contents had positive Δ^{14}C values, indicating that the majority of this carbon was fixed in the decades since 1960.
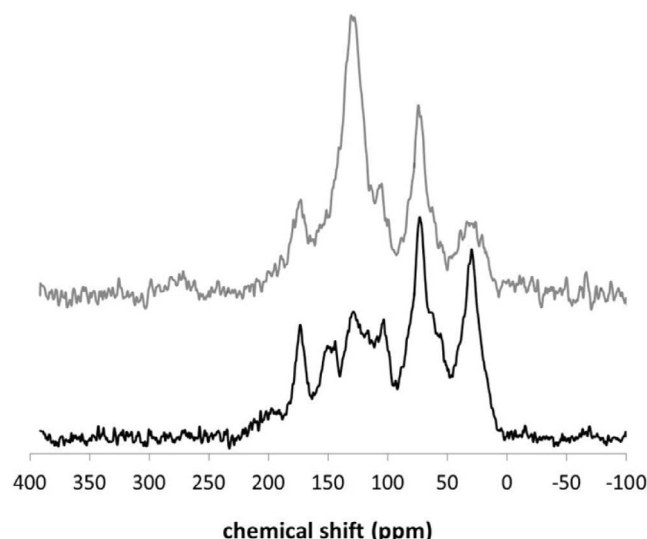
Figure 2. Carbon-13 NMR (CPMAS) spectra of the low density fractions (500–2000 μm in diameter) from the Musick 21–43 cm (black) and Shaver 40–60 cm (gray) soils in 2005. Spectra are scaled to equivalent magnitude to allow quantitative comparison of peak.

Using the linear relationship in Figure 3, we estimate end-members of charcoal and non-charcoal to have Δ^{14}C values of −577.9‰ and +149.9‰, respectively. This suggests that non-charcoal LF-C is of recent origin (fixed since 1963). The radiocarbon age of charcoal LF-C is estimated to be ~7000 yr BP, though this mean age probably includes younger and older components. If we assume that pre-1960 charred materials made up 20–40% of the Shaver soil LF-C, and 10–20% of the Musick LF-C, the amount of decadally cycling LF-C in the subsurface horizon can be estimated to be 1.5–2.6 kgC m^{-2} (Shaver) or 2.1–4.0 kgC m^{-2} (Musick). Inferred charcoal inventories in the subsoil range from 0.3 up to 1.3 kgC m^{-2}.
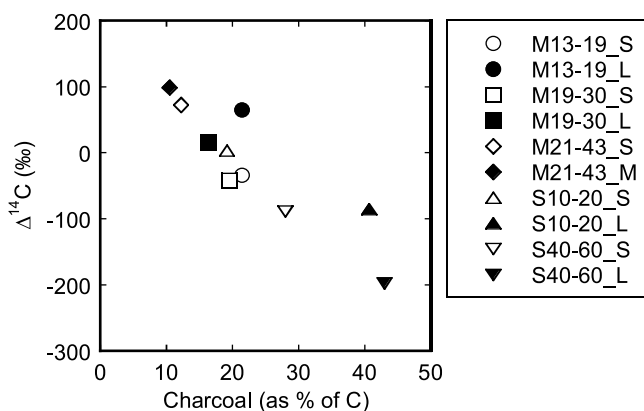
Figure 3. Relationship between Δ^{14}C and charcoal content of size-separated LF carbon. Letters M and S in the sample names represent Musick and Shaver sites, respectively, numbers represent depth intervals where the samples were collected, and S, M and L after the numbers represent size classes of <63, 63–500, and 500–2000 μm, respectively.

3.5. ^{14}C Signatures of Fine Roots

[29] The Δ^{14}C values of fine roots (mixed live + dead, <0.5 mm in diameter) collected in the subsurface horizons of the post-bomb soil profiles were higher by 3–171‰ than concurrent atmospheric ^{14}CO$_2$ values (Figure 4). Overall, the Δ^{14}C values decreased with time, approximately in parallel with the atmospheric ^{14}CO$_2$ trend (Figure 4). No consistent trend of root Δ^{14}C values with depth was observed.

3.6. Oxidation of HF-C

[30] Moderate oxidation (1 day at 24°C) for the pre-bomb and 2009 soil samples removed 49–58% of the total HF-C, more from the Shaver soils than from the Musick soils (Table 3; amount of HF-C is given in Table 1). The Δ^{14}C values of the HF-C resistant to oxidation (retained HF-C) were always lower than those of the starting material (HF-C), indicating that the oxidization treatment removed a portion of HF-C with higher Δ^{14}C values. Using mass balance to estimate the Δ^{14}C of oxidized C resulted in a value of +61‰ for the oxidized HF-C in the Shaver soil and −116‰ in the Musick soil. Both the retained and oxidized HF-C fractions increased in ^{14}C between the pre-bomb and 2009 soils.

[31] Stronger oxidation (2–3 days × 3 times at 50°C or 3 days × 1 time at 24°C) removed more C (96% and 64%) from the Musick and Shaver's HF than the moderate oxidation. Although the post-bomb soil samples were used for the stronger oxidation, the retained HF-C was more depleted in ^{14}C than that after the moderate oxidation for the pre-bomb soil samples. We used the Δ^{14}C value of this fraction to estimate passive pool C turnover time for the two-pool model.

3.7. ^{14}C-derived Dynamics of Subsurface HF-C

[32] The observed Δ^{14}C values for the target subsurface layers showed a similar pattern of little or no increase between pre-1960 and 1992, followed by a marked increase from 1992 to 2005 (Figure 5). This increase cannot be explained without the introduction of a time lag (τ) for inputs arriving to the HF at these soil depths; the arrival of the 'bomb' ^{14}C to the subsurface mineral-associated soil HF has been delayed by several decades.

[33] A range of adjustable model parameters (minimum or maximum age of passive fraction, turnover time of the fast-
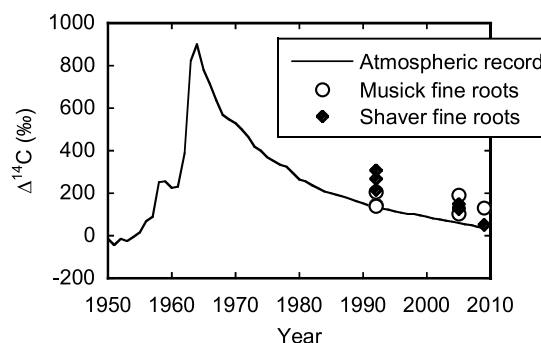
Figure 4. ^{14}C signatures of fine roots picked from the LF fraction separated from subsurface soil horizons at two forest sites, compared with atmospheric ^{14}C record. The roots are a mixture of live and dead material.
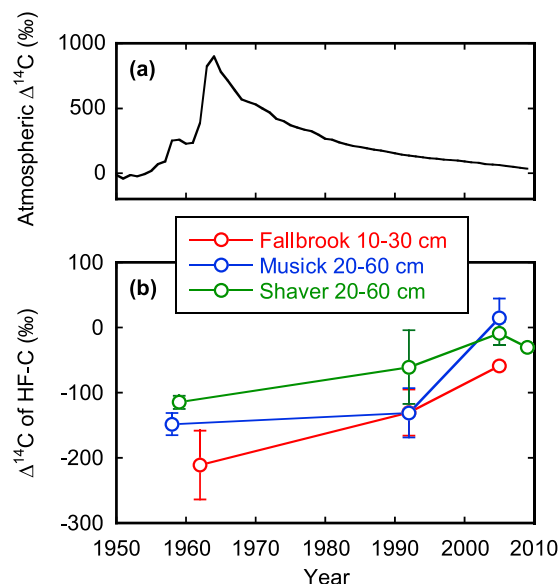
Figure 5. ^{14}C signatures of (a) atmospheric CO_2 and (b) mineral-associated soil fraction in a subsurface soil layer (10–30 cm for Fallbrook site or 20–60 cm for Musick and Shaver sites), estimated from depth profile data for C and ^{14}C. Errors are estimated based on 95% confidence interval for the slope of the linear regression analysis. The Shaver 2009 point is for 40–60 cm.

cycling C pool, and time lag between C fixation and input to the HF) could reproduce the observed changes in ^{14}C for the subsurface target layer (Table 2 and Figure 6). Only a few of these are shown in Figure 6 for simplicity. For the Shaver soil, the estimated error in 1992 is large enough that it is possible to explain the results with no time lag. However, for the other two sites the model could not explain the observed Δ^{14}C changes unless a minimum of 20–25 years time lag was included, regardless of the passive C turnover times we assigned (Table 2).

[34] The model results confirm that a significant proportion (28–73%) of the mineral-associated C turns over on the timescales of decades in the subsurface soil layer (Table 2).

The fastest turnover (10–47 years) and the largest proportion (55–73%) of decadally cycling HF-C were found in the Musick soil with the highest SOC, clay and Fe_d contents (Table 1). Given the much larger overall stock of HF-C in the Musick soil (2.6–3.6 kgC m^{-2}), the actual amount of decadally cycling C in the layer (1.4–2.6 kgC m^{-2}) was significantly greater compared to that in the Shaver soil (44–69% of 1.2–1.7 kgC m^{-2}, or 0.5–1.2 kgC m^{-2}), and the Fallbrook soil (28–67% of 0.5–1.3 kgC m^{-2}, or 0.1–0.9 kgC m^{-2}). There are tradeoffs within the model structure as assumed. For example, assuming a longer lag time in C inputs causes shorter turnover times in the faster-cycling C pool. Overall, assuming longer turnover times of the passive C pool yielded longer turnover times and a larger proportion of the faster-cycling C pool.

[35] At steady state, the model estimates both the amount and Δ^{14}C signature of the decomposing C. These ranged from 5 to 216 gC m^{-2} yr^{-1} (Table 2), with larger C fluxes estimated for models having longer lag time and shorter passive pool turnover times. The estimated C mineralization rate was higher in the Musick soil than the other soils because of the shorter turnover and larger size of the faster-cycling C pool.

3.8. C Mineralization in Subsurface Soils and C Isotopic Signatures of the Evolved CO_2

[36] The rate of C mineralization over the 55-day laboratory incubation of the recent, root-free subsurface was about five times higher in the Shaver soil than in the Musick soil (Table 1), although the Shaver soil had less soil C, and lower clay and Fe_d concentrations. The CO_2 evolved from the subsurface soil samples had positive Δ^{14}C values (i.e., mostly C fixed in the last decades) for both the Musick and Shaver sites (see vertical axes in Figure 7). The Δ^{14}C values of the respired CO_2 (+39 and +92‰ for the Musick and Shaver soils, respectively) were similar to those of fine roots collected from the soil samples, and were generally higher than those of the soil fractions. Using a one-pool model (equation (2)) these correspond to average transit times for C in the subsurface of ~200 years (Musick) and 10 or 90 years (Shaver), respectively. However, these are clearly averaging sources of younger and older C (Figure 7).
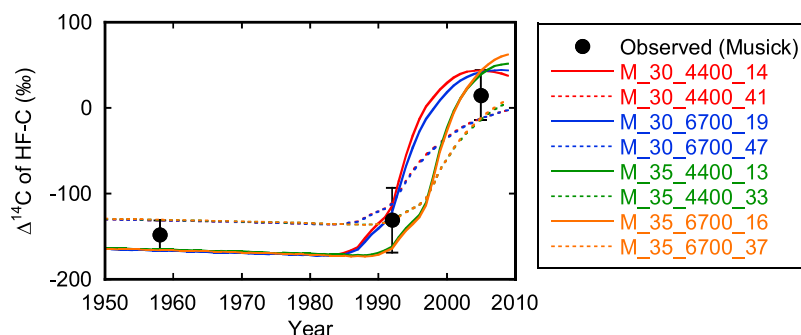
Figure 6. Observed and model-predicted changes in Δ^{14}C values of mineral-associated soil fractions in 20–60 cm layer of the Musick soil. The name of model predictions indicates: [site (Musick)]_[lag time for inputs (yr)]_[turnover time of passive C (yr)]_[turnover time of fast-cycling C (yr)]. Of all the model predictions that explain the observation, only two predictions that represented the shortest and longest turnover times of fast-cycling C for each lag time (30 or 35 years) and passive C turnover (4400 or 6700 years) case are shown in the figure. For all model results, see Table 2.
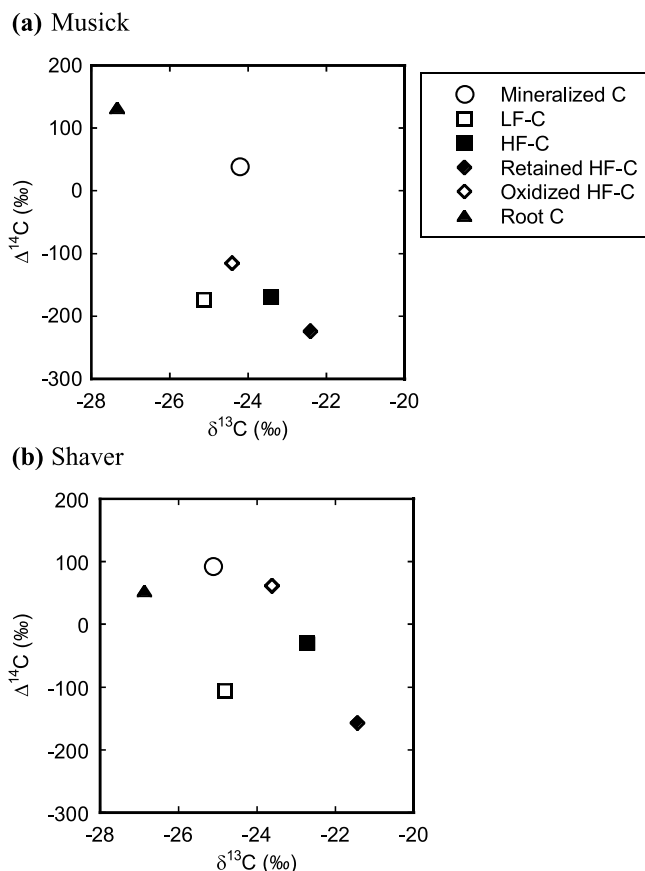
(a) Musick

(b) Shaver

Figure 7. Carbon isotopic signature of CO_2 evolved and soil fractions obtained from the 2009 (a) Musick and (b) Shaver subsurface soil samples.

[37] Our incubation was root free, but should represent contributions from decomposition of both LF and HF organic matter fractions. The estimated fluxes from modeling the HF-C pool represent 45–174% and 4–16% of the C mineralization rate in the laboratory incubations for Musick and Shaver soils, respectively. Radiocarbon signatures predicted from the model of HF-C dynamics with lagged inputs were high (88–289‰ and 163–410‰ for Musick and Shaver, respectively) compared to the values measured in the incubations. Even the model without time lags showed higher $\Delta^{14}C$ values (98–145‰ for the Shaver soil) for the mineralized C in 2009.

[38] The ^{13}C signature of the evolved CO_2 was roughly intermediate between the ^{13}C-depleted fine root fraction and the ^{13}C-enriched HF-C fraction (Figure 7), though ^{13}C data may be affected by microbial fractionation during decomposition and do not necessarily record the balance of potential substrates.

4. Discussion

4.1. Amount and Nature of Decadally Cycling Subsurface C

[39] Three observations provide evidence for the significant amounts of decadally cycling C in subsurface soils. First, the ^{14}C signatures of organic carbon in all soil fractions increased during the bomb-^{14}C period (Figure 1), indicating significant addition of C fixed in the last 50 years although the mean age of bulk SOM was hundreds (Shaver) to thousands (Musick) of years. While the radiocarbon signatures of LF-C were complicated by the incorporation of varying amounts of pre-1960 charcoal (Figure 3), subtraction of the charcoal component indicates that up to 1.5–4.0 kgC m^{-2} in the subsoils of the forest sites consists of C with post-1960 origin.

[40] Modeling the dynamics of the HF pool implies that a surprisingly large proportion (44–73%, or 0.5–2.6 kgC m^{-2}) of the mineral-associated SOC can turn over decadally in the forest subsurface soils. For the Shaver soil, this is substantiated by the oxidation treatment of the 2009 samples, which removed ∼58% of the HF-C with $\Delta^{14}C$ of +68‰. (Oxidation in the Musick 2009 soil removed younger material that still had a strong contribution from pre-bomb $\Delta^{14}C$). Hence a total of at least 2.0 up to 6.6 kgC m^{-2} of the (root-free) subsurface soil C is in forms that contain post-1960 carbon in the forest soils. In the grassland site, we estimate that 0.2–1.5 kgC m^{-2} of the subsurface (10–30 cm) C was fixed in the last 50 years. For comparison, the O+A horizons of these soils, normally considered the most dynamic, contain 2.5 to 7.4 kgC m^{-2} (Table 1). While we cannot estimate turnover times for the fast-cycling fraction of the LF-C because of charcoal contamination, the fast-cycling fraction of HF-C carbon turnover times ranged from 10 to 95 years. To match the time trend of ^{14}C requires that the turnover times determined using the model are shorter, with a time lag of 20–40 years for C inputs to enter the fast-cycling pool in all soils.

[41] Second, laboratory incubations of root-free subsurface soil samples from the two forest sites (55–70 cm for Musick and 40–60 cm for Shaver) showed significant rates of C mineralization (Table 1), indicating that metabolically active microbes exist at these depths and that a fraction of the subsoil organic matter is decomposable to the microbes. Comparison of the radiocarbon signature of respired CO_2 with fractionated SOM sources from the incubated soil (Figure 7) indicated that the age of C being respired was a mixture of younger and older C with an average age of decades up to 200 years. However, the ^{14}C signatures of the C observed in incubations (which integrate HF and LF sources) were consistently overestimated using models that reproduced the HF ^{14}C time history. Possible explanations include: (1) much of the respired C could be derived from LF rather than HF fractions; (2) the potentially episodic nature of C inputs to the subsurface soil, meaning the steady state model is not adequate for describing timescales of more rapid decomposition; (3) spatial (as opposed to temporal) heterogeneity, including preferential flow pathways and plant-rooting behavior, results in heterogeneous distribution of active SOC pool in deeper parts of the soil profiles [*Sanderman et al.*, 2008; *Chabbi et al.*, 2009]; (4) the possible presence of very ^{14}C-depleted, finely disseminated charcoal in the HF fraction combined with spatial variability which could make us overestimate the turnover time of HF [*Golchin et al.*, 1994a; *Glaser et al.*, 2000; *Brodowski et al.*, 2005; *Rasmussen et al.*, 2005], or (5) artifacts such as physical disturbance of soil structure associated with laboratory conditions mean incubation fluxes do not map well to in situ field conditions [*Ewing et al.*, 2006]. Regardless of which explanation may be correct, the fact that the respired CO_2 was

dominated by C fixed since the bomb period provides further evidence of fast-cycling C in the subsoil horizons.

[42] The third piece of evidence for a decadally cycling C pool in the subsoil comes from the need to use decadal time lags for C inputs to model the time course of HF radiocarbon in deeper soil layers. The $\Delta^{14}C$ values of fine roots (mixed live + dead, <0.5 mm in diameter) collected from the subsurface layers in the post-bomb soil profiles were higher by 3–171‰ (mean ages of 1–20 years) than C fixed in the year they were collected. These results are consistent with other studies using ^{14}C signatures to infer fine root lifespans of 3–18 years in other temperate [*Gaudinski et al.*, 2001] and tropical [*Trumbore et al.*, 2006] forests, and support the use of lagged C inputs in the model. However, this fine root lifespan (1–20 years) alone is not enough to explain the delayed (>20 years) increase in radiocarbon of the HF fraction in subsurface layers after 1992 (Figure 5). The source of this additional time lag could include coarse roots (though these are likely small contributors to overall inputs) or reflect the time required for C to pass through a microbial pool before being incorporated into the HF, or the time required for downward transport of carbon from overlying horizons, or the occlusion of SOM by aggregation [*von Lützow et al.*, 2006], or the fixation of high ^{14}C-CO_2 derived from decomposition and/or plant root respiration by CO_2 fixing bacteria in soils [*Miltner et al.*, 2005].

[43] A major limitation of this study is the large error introduced because of limited replication. For these archived soil samples we have no options, but clearly in the future and as radiocarbon measurements become more accessible more efforts should be made to establish the magnitude of spatial variability; we have assumed rather large errors in our analysis instead. Given that the same pattern of ^{14}C increase between 1992 and 2005 occurred in all three sites (Figure 5), and the results of other researchers (see next section) we think this is a robust pattern that should stimulate further research in the dynamic nature of subsurface carbon.

4.2. Factors Controlling Subsoil C Dynamics

[44] A study of the C dynamics in O and A horizons for the same elevation transect [*Trumbore et al.*, 1996] demonstrated that C cycling in surface horizons slowed as a function of elevation. In contrast, we observe no consistent trend with elevation in the overall amount or inferred turnover time of fast-cycling C in subsurface horizons for the same soils. The age of fine roots at the Fallbrook site, consisting mostly of annual grasses, is considerably less than those for woody roots found in the high-elevation forests. However, the time lag required to model delivery of C inputs to the subsurface HF fraction is >20 years at the Fallbrook site and the turnover times we deduce for the fast-cycling HF-C are fastest at the mid-elevation forest site (Musick).

[45] Low-density carbon (LF) is commonly characterized as proxy for the fast-cycling carbon in soils, as it consists of relatively fresh plant material not associated with minerals [e.g., *Golchin et al.*, 1994b]. Although SOM at greater depths has increased mineral association, interestingly the LF is still a large fraction (40–55%) of the C in these coarse-textured subsoils at the two forest sites (Table 1). The apparent ^{14}C age of this LF-C increases dramatically with depth (Figure 1), up to ~3500 yr BP. However, the chemical structures of the LF-C, analyzed by ^{13}C NMR, were similar

in the surface and subsurface layers. For example, carbohydrate, generally considered to be degraded at a faster rate than other organic compounds [*Golchin et al.*, 1994a], remains a substantial component (17–28% of the total C) in the subsurface layers (Table 4). Given the lack of change in chemical nature of the LF-C with depth, mechanisms to allow for its older age include physical protection by aggregate formation or stabilization on mineral surfaces, processes that make organic materials less accessible to soil microbes and enzymes [*Golchin et al.*, 1994a; *Sollins et al.*, 1996; *Baisden et al.*, 2002b; *Six et al.*, 2004; *Rasmussen et al.*, 2005].

[46] Alternatively, charcoal seems to be the primary factor affecting the apparent ^{14}C age of LF-C, irrespective of LF's size class and depth in the soil profiles (Figure 3). The amounts of charcoal C inferred from the amount of LF and its char content are as much as 1.0 kgC m^{-2} in the Shaver forest subsurface horizons, where the apparent age of LF-C exceeded that of HF-C in the same depth interval. Although the older LF-C with higher charcoal content that we found seems to contradict recent findings [*Ohlson et al.*, 2009] that soil pools of charcoal C are less refractory (ages of hundreds of years) than previously thought (thousands of years), the differences in fire processes and in fauna between our site and that of *Ohlson et al.* [2009] may explain the charcoal age offset. Lower charcoal production rates have been found in fire-prone boreal regions [*Czimczik et al.*, 2003]. It has also been hypothesized that what char is made in boreal regions is more vulnerable to loss in recurrent fires because boreal permafrost soils are less hospitable to the burrowing organisms necessary to move organic carbon to soil horizons below burn depths [*Czimczik and Masiello*, 2007]. The age of charred material we infer from the linear mixing model (Figure 3) has an end-member ^{14}C age for char of about 7000 years, implying great stability for char under the conditions in our ecosystems.

[47] The biggest differences in the dynamics of subsurface C cycling are found between the two forest sites. The Musick soil, with higher clay and Fe_d content, contains more C (including decadally cycling C); it has the fastest modeled turnover time for the HF-C fast-cycling pool, but the oldest passive pool. There is clear evidence that the association of organic compounds with minerals slows their decomposition in soils [*Torn et al.*, 1997; *Baldock and Skjemstad*, 2000; *Six et al.*, 2002; *Masiello et al.*, 2004; *Rasmussen et al.*, 2005], and this has been thought to be an important mechanism for the long-term stabilization of soil C particularly in biologically less active deeper soil horizons. However, the incorporation of bomb-^{14}C into the HF-C we observed suggests at least some of the carbon stabilization processes controlled by minerals operate on decadal timescales at significant depths [*Kleber et al.*, 2007].

[48] What can be the reason for the long time lags we must infer for C entering the subsoil, with bomb ^{14}C taking decades to be detected, then rising rapidly in subsurface soils? The two possible sources of time-lagged or aged carbon to the subsurface soil are in situ production by roots, and downward transport of dissolved or particulate carbon from overlying layers. A number of studies using labeled plant litter suggest that fine roots are the major source of subsoil organic matter [*Rasse et al.*, 2005]. However, none of these studies has been carried on long enough to detect downward

transport that may take decades. In our study, the ^{14}C signature of the HF-C increases after 1992, while that of the fine root C declines or stays the same (Figure 4). Hence fine roots cannot be the direct source of the time lag – either there is a lagged transfer from live root to dead root to HF-C [*Trumbore et al.*, 2006], or coarse roots with longer lifespans are an important source of HF-C, or there is a delayed transfer of C from above [*Fröberg et al.*, 2007]. An alternative hypothesis could be fixation of soil-CO_2 [*Miltner et al.*, 2005] that includes the decomposition time lags, but that signal would be diluted by root respiration that includes a more recent component.

[49] The difference in ^{13}C signatures between the HF-C (^{13}C-enriched) and fine roots (^{13}C-depleted) at depth (Figure 7) suggests that the HF-C is dominated by strongly microbially processed materials rather than by partially transformed root materials. This is consistent with the finding of *Trumbore et al.* [2006] that a portion of the decomposing roots in a subsurface tropical soil is cycled through SOM pools with decadal turnover time. *Jobbágy and Jackson* [2000] found that SOC distributes more deeply than root biomass. Potential explanations for this inconsistency include: decreasing SOC turnover with depth, resulting in higher C accumulations per unit of C input in deep soil layers; increasing root turnover with depth, resulting in higher C inputs in deep soil layers; and downward transport of SOC from upper to lower layers [*Jobbágy and Jackson*, 2000]. While our results do not resolve the question of the sources of C to the subsurface horizons, they do demonstrate that the process operating is one that introduces a time lag at our sites.

[50] In contrast to our study, *Richter et al.* [1999] in a study of an aggrading temperate forest (Calhoun forest, South Carolina) demonstrated a time lag of less than a decade before the bomb ^{14}C increase was observed in subsurface soils (>15 up to 60 cm depth). However, the Calhoun forest was aggrading (planted in 1957) whereas our forests are mature, so that the subsurface in Calhoun could be reflecting the expansion of root populations. South Carolina also experiences higher precipitation than our study sites, and therefore possibly different rates of DOC transport to deeper horizons.

4.3. How Important Are Subsurface Pools in Ecosystem C Cycling?

[51] Increases in subsurface soil ^{14}C due to incorporation of bomb radiocarbon have been found in other studies comparing archived and contemporary samples in California and New Zealand temperate grasslands [*Baisden et al.*, 2002a; *Baisden and Parfitt*, 2007] and in a South Carolina re-establishing forest [*Richter et al.*, 1999]. High Δ^{14}C in subsoil CO_2 has been found in eastern Amazonian tropical forests [*Trumbore et al.*, 1995], where the presence of deep roots drives a deep soil carbon cycle. Together, these studies suggest that subsurface soil C cycling can be significant on decadal timescales.

[52] We estimate C mineralization fluxes of 5–39 and 9–216 gC m^{-2} yr^{-1} for the grassland and forest subsurface soils, respectively. These fluxes correspond to ∼3% and 3–16% of the total soil respiration, or 1–9% and 10–54% of the annual surface litter C input at the Fallbrook (grassland) and Musick (forest) sites, respectively [*Wang*

et al., 2000]. Our estimates agree with previous studies in temperate grassland soils [*Baisden et al.*, 2002a; *Baisden and Parfitt*, 2007] that ∼10–40% of total SOC below 50 cm depth turns over on decadal timescales, producing a C flux of ∼23 gC m^{-2} yr^{-1}. If we assume that these estimates can be generalized to temperate grassland and deciduous forest biomes globally (9×10^{12} and 7×10^{12} m^2, respectively [*Jobbágy and Jackson*, 2000]), then the natural cycling of subsurface SOC can be 0.1–1.9 PgC yr^{-1}. This amounts to up to 1.6% of annual gross terrestrial primary production (GPP), currently estimated at 120 PgC yr^{-1} [*International Panel on Climate Change*, 2007].

[53] Together, studies of subsurface C cycling have several implications for how soils may respond to global environmental change. First, the amount of decadally cycling C in soils is significantly larger than expected from studies of the A horizons alone. Current global change models based on the top 20–30 cm of soil may therefore significantly underpredict the response of soil C to climate or land-use change. Second, the dynamics of decadally cycling C in subsurface soil layers may be largely controlled by interactions with soil minerals, which contrasts with stronger climate control in surface soil layers [*Trumbore et al.*, 1996; *Adair et al.*, 2008]. We do not know how rapidly C associated with soil minerals, and minerals themselves, can respond to climate change [*Trumbore*, 2009]. However, the abrupt changes in soil properties within a small elevation distance (Musick versus Shaver) suggest the potential for some soil mineralogical components and thereby their interactions with SOM to adjust relatively rapidly to new climate conditions. The interactions also influence the dynamics of C not associated with minerals through indirect controls on factors such as vegetation and hydrology [*Torn et al.*, 2005; *Reich et al.*, 2005]. Third, destabilization of large C stores in subsurface horizons is possible with changes in vegetation, (root) litter quality and inputs, nutrient availability, and DOC behavior, and as a result could have significant impacts on global C cycling [*Schwendenmann and Veldkamp*, 2006].

[54] The long time lags between C fixation and incorporation into the decadal-cycling SOC pool in subsurface horizons suggest a lagged response of deep soil C to climate and land-use changes that would be difficult to detect without the bomb-^{14}C tracer. Future studies should look for patterns in the amount and dynamics of subsurface C as a function of mineralogy and climate, and especially need to identify the role of roots versus dissolved transport as the source of time lags to explain the timing of the arrival of bomb ^{14}C in the subsurface horizons.

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