Methodological uncertainty in estimating carbon turnover times of soil fractions

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ABSTRACT

Improving predictions of soil organic carbon (SOC) dynamics by multi-compartment models requires validation of turnover times of different SOC pools. Techniques such as laboratory incubation and isotope analysis have been adopted to estimate C turnover times, yet no studies have systematically compared these techniques and assessed the uncertainties associated with them. Here, we tested whether C turnover times of soil fractions were biased by methodology, and how this changed across soil particle sizes and ecosystems. We identified 52 studies that quantified C turnover times in different soil particles fractionated either according to aggregate size (*e.g.*, macro- versus micro-aggregates) or according to soil texture (*e.g.*, sand versus silt versus clay). C turnover times of these soil fractions were estimated by one of three methods: laboratory incubation (16 studies), $\delta^{13}C$ shift due to C_3–C_4 vegetation change (25 studies), and ^{14}C dating (19 studies). All methods showed that C turnover times of soil fractions generally increase with decreasing soil particle size. However, estimates of C turnover times within soil fractions differed significantly among methods, with incubation estimating the shortest turnover times and ^{14}C the longest. The short C turnover times estimated by incubation are likely due to optimal environmental conditions for microbial decomposition existing in these studies, which is often a poor representation of field conditions. The ^{13}C method can only be used when documenting a successive C_3 versus C_4 vegetation shift. C turnover times estimated by ^{14}C were systematically higher than those estimated by ^{13}C, especially for fine soil fractions (*i.e.*, silt and clay). Overall, our findings highlight methodological uncertainties in estimating C turnover times of soil fractions, and correction factors should be explored to account for methodological bias when C turnover times estimated from different methods are used to parameterize soil C models.

1. Introduction

Uncertainty in predicting carbon–climate feedbacks largely stems from poor representation of soil organic carbon (SOC) pools. This is an important consideration as SOC is the largest C pool in terrestrial ecosystems and perturbation of it strongly modulates climate change (Todd-Brown et al., 2013; Koven et al., 2015; Luo et al., 2016). SOC is heterogeneous in terms of composition, structure, location, and stabilization mechanism (Stevenson, 1994; Sollins et al., 1996; Schmidt et al., 2011; Lehmann and Kleber 2015). Conventional soil C models classify SOC into multiple conceptual pools with different turnover times based on their resistance to microbial decomposition (Jenkinson and Rayner, 1977; Parton et al., 1987). A growing body of research calls for mechanistic representations of SOC processes in Earth System Models, such as protection by physical isolation and mineral sorption (Sulman et al., 2014; Wieder et al., 2014; Tang and Riley, 2015). Therefore, attention should be paid to physically fractionated SOC fractions which are measurable and could represent soil organic matter (SOM) protection mechanisms (Christensen, 1996; von Lützow et al., 2007; Schmidt et al., 2011). Quantifying C turnover times of these soil

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fractions is important for models which integrate explicit mineral protection processes. Until now there has been no consensus on the turnover times of various measurable SOC fractions, due to various methodologies being used to estimate C turnover times.

There are three commonly used methods for assessing SOC turnover times: the laboratory incubation (Christensen, 1987), shifts in natural ^{13}C abundance after C_3–C_4 vegetation change (Balesdent et al., 1987), and ^{14}C dating (O'Brien and Stout, 1978; Trumbore, 2000). The laboratory incubation directly quantifies biological decomposition of isolated soil fractions under controlled optimal conditions. This method is easy to conduct and has been widely used. In contrast, the ^{13}C and ^{14}C methods trace C isotopes during decomposition and stabilization processes to estimate C turnover times (O'Brien and Stout, 1978; Balesdent et al., 1990). The ^{13}C method can only be used in studies where there are $\delta^{13}C$ shifts after years of successive C_3–C_4 vegetation change and requires careful C inventory measurements of disturbed and undisturbed soils (Balesdent et al., 1987; Zhang et al., 2015). The ^{14}C dating method assumes that SOC fractions are at equilibrium between input and decay, and that all the C inputs to soils enter the system at the same time or are constant (Trumbore, 1993; Bruun et al., 2005). These assumptions are often not met in reality and soil ^{14}C is expensive to measure. Due to these differences in methodology, the three methods likely generate different estimates of SOC turnover times. For instance, the turnover times of mineral associated organic matter (MOM) at 0–10 cm depth has been reported to be 8–43 years using the laboratory incubation method (Rabbi et al., 2014), 53–63 years using the ^{13}C abundance after C_3–C_4 vegetation change (Dalal et al., 2013; Liang et al., 2014), and 52–381 years when using ^{14}C dating (Budge et al., 2011).

Bulk soil can be separated into soil fractions using the physical, chemical, density, and combined fractionation methods, among which the physical fractionation is able to generate soil fractions with distinct C turnover times (Christensen, 2001; Mikutta et al., 2006; von Lützow et al., 2007). Variation in C turnover times results from different SOC protection mechanisms associated with soil particles as well as inconsistent methods used to estimate C turnover times (Bird et al., 2002; Tan et al., 2013; Yonekura et al., 2013; Beniston et al., 2014). Physically fractionated soil particles are often obtained according to soil aggregate size or soil texture. According to soil aggregates size, C in macro-aggregates (i.e., coarse organic matter, COM) turns over fast, while C in the micro-aggregates (i.e., fine organic matter, FOM) and MOM is supposed to represent C that is primarily protected by physical isolation and mineral matrix, respectively (Six et al., 1998; Baldock and Skjemstad, 2000; von Lützow et al., 2007). According to soil texture, C in the sand fraction has a short turnover time and C associated with the silt and clay fractions is considered as mineral associated OM in models (Parton et al., 1987; Beniston et al., 2014; Tang and Riley, 2015; Wieder et al., 2014). However, we still do not know whether different classifications to separate soil fractions can differentiate their C turnover times.

By synthesizing published studies, we compared C turnover times of physically fractionated soil particles (i.e., COM − FOM − MOM or sand − silt − clay) across ecosystems. We aimed to test whether C turnover times of soil fractions estimated using the laboratory incubation, ^{13}C, and ^{14}C were different, and how this changed with soil particle size and ecosystems. We predicted that C turnover times estimated using the laboratory incubation would be shorter than those using the C isotope methods, and that C turnover times based on soil fractions would increase with decreasing particle size.

2. Material & methods

2.1. Data sources

We searched the literature to find information that included: (1) at least one of the following physically fractionated soil particles as study materials: macro-aggregates (coarse organic matter, COM, 250–2000 μm), micro-aggregates (fine organic matter, FOM, 20/53/63–250 μm), MOM (<20/53/63 μm), sand (20/53/63–2000 μm), silt (2–20/53/63 μm), and clay (<2 μm), and (2) CO_2 flux measured multiple times over the time course of laboratory incubations, or C turnover rates or times assessed based on the $\delta^{13}C$ difference after years of successive C_3–C_4 vegetation change, or mean residence times estimated based on $\Delta^{14}C$ activity. Detailed information of the selected studies can be found in Table 1 and the supplementary materials (Supplementary Material Table S1). We extracted information on 537 soil fractions from 52 studies around the world (Fig. 1). For all the studies identified, we also gathered the information regarding soil fraction classification used, the coordinates, climate, soil depth, soil type, vegetation at soil sampling sites, and the mass proportion and organic C concentration or content of each soil fraction (Supplementary Material Table S1).

2.2. Carbon turnover estimate

For the studies using laboratory incubations to estimate C turnover time, we generated a sub-dataset that included the following data for each soil fraction: the date of measurement, initial organic C concentration or content, and CO_2 respiration rate or cumulative CO_2 respiration at each time point. We used the two-pool rather than one-pool exponential decomposition model to estimate C turnover times of soil fractions, because C in soil fractions is not homogeneous and so the two-pool model could more accurately describe decomposition than the one-pool model (Derrien and Amelung, 2011). For comparison, we converted values of cumulative CO_2 respiration from the original unit (mg CO_2-C g^{-1} sample) to mg CO_2-C per gram of initial organic C concentration of a sample.

$$C_t = f_l \times \left(1 - e^{-k_l \times t}\right) + (1 - f_l) \times \left(1 - e^{-k_s \times t}\right) \quad (1)$$

was used to estimate C turnover times of soil fractions, where C_t is the cumulative CO_2 respired, f_l is the proportion of labile SOC pool, and k_l and k_s are the decomposition constants of labile and stable SOC pools. The turnover times of labile (τ_l) and stable (τ_s) SOC are the reciprocal of k_l and k_s, respectively. Given that stable SOC accounts for a large proportion of total SOC and τ_l is similar for the studied soil fractions from a variety of ecosystems, using τ_s instead of τ_l is much more representative to characterize C turnover of the entire SOC. Therefore, τ_s values of soil fractions were used to compare whether the three methods provide different C turnover times values. Parameters in the two-pool model were estimated using probabilistic inversion approach (Xu et al., 2006; Weng and Luo, 2011), which was performed using the Metropolis-Hastings (M-H) algorithm − a Markov Chain Monte Carlo (MCMC) technique (Metropolis et al., 1953; Hastings, 1970). Rationale and details about this technique can be seen in Schädel et al. (2013).

For the studies using $\delta^{13}C$ after C_3–C_4 vegetation change to estimate C turnover time, we collected the data of turnover time (τ, year) or decomposition constant (k, year^{-1}) for all of the six soil fractions (i.e., COM-FOM-MOM and sand-silt-clay). In the studies where neither k nor τ were reported, we calculated k using Equation (2) or (3) according to the data available in selected studies.

Table 1
Study sites and data distribution of C turnover times of soil fractions estimated by the laboratory incubation, $\delta^{13}C$ after successive C_3-C_4 vegetation change, and the ^{14}C dating methods. COM: coarse organic matter, 250–2000 μm; FOM: fine organic matter, 20/53/63–250 μm; MOM, mineral associated organic matter, <20/53/63 μm; sand: 20/53/63–2000 μm; silt: 2–20/53/63 μm; clay: <2 μm.

Methods	Fraction	Location (latitude, longitude)	Sample size	C turnover times			
				Mean	Min	Max	CV
Incubation	COM	−30.47−64.87°, −147.72−151.65°	28	8.6	0.5	51	140%
	FOM	−30.47−54.20°, −114.13−151.65°	31	31.5	3.6	342	227%
	MOM	−30.47−64.87°, −147.72−151.65°	37	30.9	3.8	662	347%
	Sand	−40.38−56.08°, −82.73−175.6°	16	6.8	1.6	38	130%
	Silt	−40.38−56.08°, −82.73−175.6°	16	17.9	4.7	123	159%
	Clay	−40.38−56.08°, −82.73−175.6°	16	24.0	6.2	122	118%
^{13}C	COM	−28.67−51.87°, −98.20−153.33°	50	54.1	3.0	278	108%
	FOM	−24.81−51.87°, −98.20−149.8°	38	83.9	6.0	429	133%
	MOM	−24.81−48.36°, 4.40−33.10°	36	210.0	29.0	3124	260%
	Sand	−22.72−48.35°, −88.31−13.18°	15	35.4	9.0	80	62%
	Silt	−10.50−51.87°, −75.35−117.93°	28	116.8	8.0	677	117%
	Clay	−22.72−51.87°, −75.35−117.93°	28	125.4	24.0	357	67%
^{14}C	COM	−30.44−68.10°, −115.30−152.69°	30	286.8	23.0	1265	114%
	FOM	−34.32−68.10°, −115.30−149.98°	28	646.3	42.0	2585	116%
	MOM	−34.32−64.47°, −75.35−149.98°	40	1179.7	26.0	6905	133%
	Sand						
	Silt	34.18−68.10°, −117.77−117.65°	37	415.0	18.0	1660	92%
	Clay	−29.37−68.10°, −117.77−117.65°	55	708.7	8.0	4745	131%
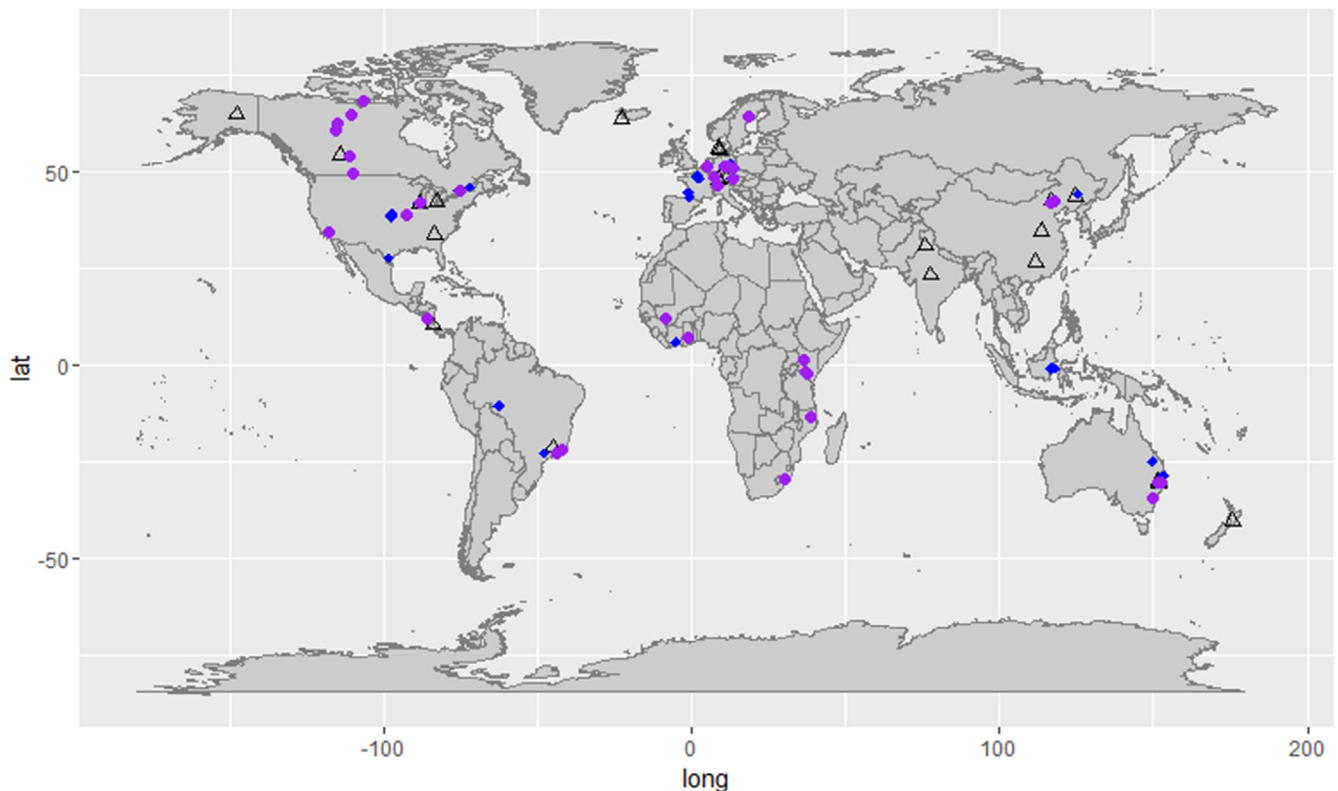
Fig. 1. Geographic locations of soil sampling sites to determine C turnover times of soil fractions. Triangles represent sampling sites for the laboratory incubation, circles for the ^{13}C method, and diamonds for the ^{14}C method.

$$k = \frac{-\ln(\text{proportion of old C})}{\text{period of C3} - \text{C4 vegetation change}} \quad (2)$$

$$A_t = A_0 \times e^{-k \times t} \quad (3)$$

where A_0 is the initial SOC stock of soil fraction, and A_t is old C stock of soil fraction at time t in years since C_3-C_4 vegetation change

(Balesdent et al., 1987).

Logarithmic transformation of Equation (3) is essentially the same as Equation (2). But when studies only measure $\delta^{13}C$ twice before and after the C_3-C_4 vegetation change, we necessarily used Equation (2) to calculate k. When studies measure $\delta^{13}C$ multiple times after the C_3-C_4 vegetation change, k was assessed using Equation (3), due to higher confidence in estimates obtained with this equation. This is due to the fact that calculations of k using Equation (2) overestimates when using $\delta^{13}C$ measured at an early

stage after the C_3–C_4 vegetation change, and underestimates when measuring $\delta^{13}C$ at a late stage after the vegetation change (Skjemstad et al., 1990; Liao et al., 2006; Derrien and Amelung, 2011). Calculating k according to Equation (2) is the two-point ^{13}C method, and the calculation according to Equation (3) is the multi-point ^{13}C method. According to these two calculation methods, we separated studies that report k or τ values to two groups, to test whether these two calculations generate different k estimates.

For studies that use ^{14}C dating techniques, there are also two distinct approaches to estimate C turnover times of soil fractions - the conventional ^{14}C model and the bomb ^{14}C model. The conventional ^{14}C method assumes that all C atoms in a sample entered soils at the same time and the measured SOC fraction is in steady state between input and decay (Talma and Vogel, 1993; Bruun et al., 2005), and calculates C turnover time (τ) by

$$\tau = \frac{1}{\lambda} \ln\left(\frac{A_{abs}}{A_t}\right) \qquad (4)$$

λ is the decay rate constant of ^{14}C, and A_{abs} is defined as 95% of the activity in 1950 of an oxalic acid standard, A_t is the ^{14}C activity of soil sample. But the assumptions in the conventional ^{14}C dating are mostly untrue for modern soils except for buried paleosols. Meanwhile, the bomb ^{14}C model uses the natural decay of atmospheric ^{14}C activity generated in the 1950s and 1960s bomb tests to estimate C turnover times (O'Brien and Stout, 1978; Trumbore, 1993; Rabbi et al., 2013). This model assumes that SOC decomposition follows the first order law and is at steady state, where C turnover time is described by

$$^{14}C_t = {}^{14}Catm_{t-lag} \times k + {}^{14}C_{t-1} \times (1 - k - \lambda) \qquad (5)$$

where $^{14}C_t$ and $^{14}C_{t-1}$ are the $\Delta^{14}C$ activities at years t and t-1, $^{14}Catm_{t-lag}$ is the $\Delta^{14}C$ of the atmosphere, k is the decomposition constant, and λ is the ^{14}C decay constant. Here, we grouped studies into those that used the conventional ^{14}C model or those that used the bomb ^{14}C model, aiming to find whether these two methods provide different C turnover times of soil fractions.

2.3. Statistical analysis

Multiple comparison was used to examine whether the laboratory incubation, ^{13}C, and ^{14}C methods generated different C turnover times for each of the six soil fractions, and to test whether C turnover times estimated by the same method are significantly different among COM, FOM, and MOM, and among the sand, silt, and clay fractions. In the multiple comparison to examine whether C turnover times estimated by the three methods were different, C turnover estimated by the two-point and multi-point ^{13}C methods were compiled, but only the estimates by the bomb ^{14}C model were used, since the two-point and multi-point ^{13}C methods did not generate significantly different estimates, but the conventional ^{14}C dating showed remarkably longer C turnover times compared to the bomb ^{14}C model. Since C turnover times were not normally distributed, a Mann-Whitney rank test was used for the multiple comparison by using the nparcomp R package (Konietschke et al., 2015). All differences were tested at the significance level of 0.05.

3. Results

Carbon turnover times differed with soil fractions and method used (Fig. 2). When bulk soils were separated into the COM, FOM, and MOM fractions, C turnover times estimated by the ^{13}C and ^{14}C methods were significantly longer than those using the laboratory incubation, but the estimates by the former two methods showed
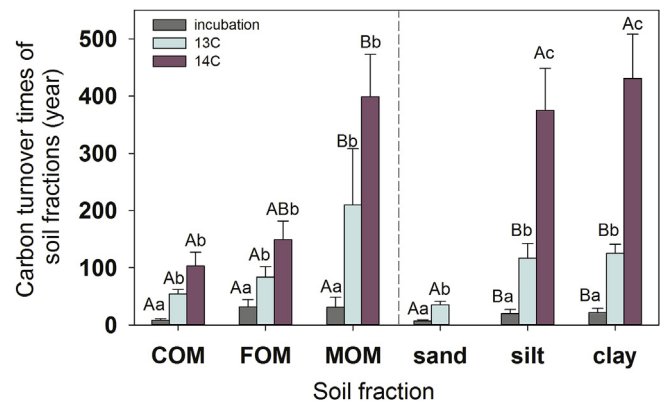
Fig. 2. Carbon turnover times of soil fractions estimated by the laboratory incubation and the ^{13}C and ^{14}C methods. COM: coarse organic matter, 250–2000 μm; FOM: fine organic matter, 20/53/63–250 μm; MOM, mineral associated organic matter, <20/53/63 μm; sand: 20/53/63–2000 μm; silt: 2–20/53/63 μm; clay: <2 μm. Data are mean ± SE. Different uppercase letters indicate that C turnover times estimated by the same method significantly differ among soil fractions, and different lowercase letters mean significantly different C turnover times among methods.

no significant difference (Fig. 2). The results of the laboratory incubation show that turnover times of stable SOC pool (mean ± SE) were significantly longer in the FOM (31.5 ± 12.9 yr) and MOM (30.9 ± 17.6 yr) fractions than in the COM fraction (8.6 ± 2.3 yr). C turnover times were significantly longer in the MOM (31.5 ± 12.9 yr) fraction than in the FOM and COM fractions when using the ^{13}C and ^{14}C methods (Fig. 2). When bulk soils were separated to the sand, silt, and clay fractions, C turnover times estimated by the three methods were significantly different from each other, following the order: incubation < ^{13}C < ^{14}C (Fig. 2). Regardless of estimate methods, C turnover times of the silt and clay fractions were similar to each other, both of which were significantly longer than those of the sand fraction (Fig. 2).

Using the ^{13}C after C_3–C_4 vegetation change, t-test results show that the multi-point and two-point calculation methods generated similar C turnover times of soil fractions, although estimates by the two-point method tended to be slightly lower (Fig. 3). Using the ^{14}C dating method, the two calculation methods (i.e., ^{14}C conventional and ^{14}C bomb) provided significantly different values of C turnover times of soil fractions (Fig. 4). C turnover times of small soil particles (i.e., FOM-MOM and silt-clay) estimated by the $\Delta^{14}C$
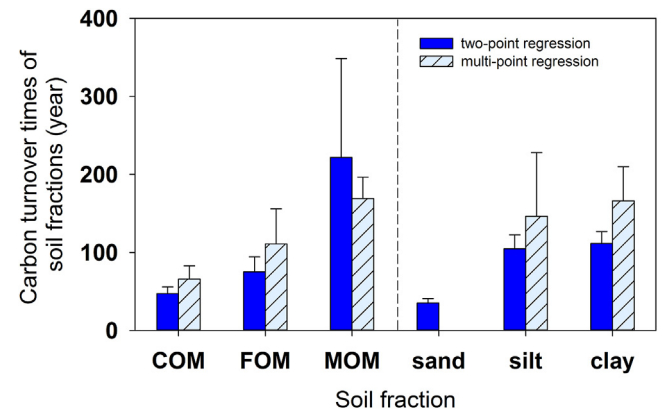
Fig. 3. Carbon turnover times of soil fractions estimated by using the two-point and multi-point calculations based on $\delta^{13}C$ shift after C_3–C_4 vegetation change. COM: coarse organic matter, 250–2000 μm; FOM: fine organic matter, 20/53/63–250 μm; MOM, mineral associated organic matter, <20/53/63 μm; sand: 20/53/63–2000 μm; silt: 2–20/53/63 μm; clay: <2 μm. Data are mean ± SE.
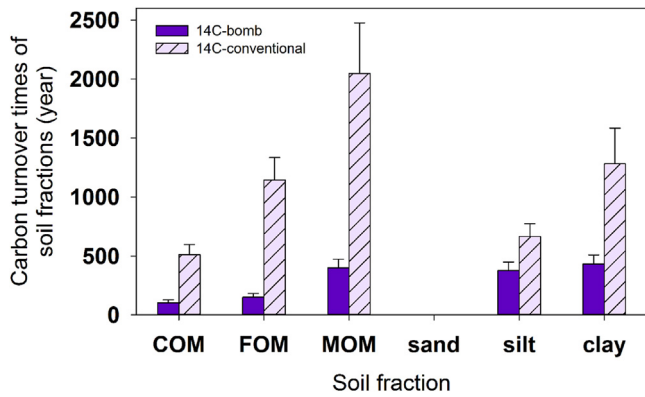
Fig. 4. Carbon turnover times of soil fractions estimated by using the conventional ^{14}C model and the bomb ^{14}C model. COM: coarse organic matter, 250–2000 μm; FOM: fine organic matter, 20/53/63–250 μm; MOM, mineral associated organic matter, <20/53/63 μm; sand: 20/53/63–2000 μm; silt: 2–20/53/63 μm; clay: <2 μm. Data are mean ± SE. * means values are statistically different between estimate methods.

conventional method were 665–2047 years, compared to 149–431 years estimated by using the Δ^{14}C bomb model (Fig. 4).

4. Discussion

Estimations of C turnover times of measurable soil fractions are important for incorporation into newly emerging soil C models that explicitly include interactions between organic matter and soil minerals. Our study shows that C turnover times of physically fractionated soil particles generally increase with decreasing particle size, following the order: COM ≈ FOM < MOM and sand < silt ≈ clay (Fig. 2), suggesting that fine soil fractions (i.e., FOM-MOM and silt-clay) allow a higher organic C preservation. These results agree with the reported range of C turnover times of these soil fractions: 3–203 years for the COM fraction, 1.2–374 years for the FOM fraction, 63–125 years for the MOM fraction, 8–1660 years for the silt fraction, and 33–4409 years for the clay fraction (Feller and Beare, 1997; Six et al., 2002; von Lützow et al., 2007). Although other studies have addressed C turnover times across soil fractions (Christensen, 1987; Feller and Beare, 1997; Bird et al., 2002; Six et al., 2002; von Lützow et al., 2007; Rabbi et al., 2014), this study has the advantage of including a large sample size for each of the six soil fractions and for each C turnover estimate method (Fig. 1; Table 1). Moreover, these soil fractions are from a wide variety of ecosystem types and span a substantial latitudinal gradient (68.10° N to 40.38° S (Fig. 1)).

Although the patterns of how C turnover times change with soil particle size is similar regardless of the estimate methods, the laboratory incubation, the ^{13}C method, and the ^{14}C dating method provide different mean values of C turnover times of soil fractions (Fig. 2), in the order: incubation < ^{13}C < ^{14}C. This difference highlights methodological uncertainties in estimating C turnover times of soil fractions. Special attention should be paid when parameterizing soil C turnover times to simulate SOC dynamics. Short C turnover times estimated via laboratory incubation might be due to microbial decomposition rates at optimal temperature and moisture, unrealistic under climatic limitations present in natural systems. Sieving and rewetting soils, that routinely occurs before incubation, has been found to increase C mineralization (Fierer and Schimel, 2002; Miller et al., 2005), and thus could lead to the underestimate of C turnover times. In contrast, the ^{13}C and ^{14}C methods estimates SOC in the field where the climate likely constrains microbial decomposition. Another reason for short C turnover times of soil fractions estimated by the laboratory incubation

could be that soils used in incubations are often from top soil layers, and C in shallow soils has shorter turnover times than deeper soils (Rumpel et al., 2002; Mathieu et al., 2015). Additionally, the fractionation procedure may redistribute C in different soil particles and accelerate C decomposition (Christensen, 1987; Parfitt and Salt, 2001; Benbi et al., 2014). So, C turnover times of soil particles estimated by the ^{13}C and ^{14}C methods are likely more representative of actual values in field. However, laboratory incubations are still useful to elucidate how factors other than climate might affect C turnover.

When using the ^{13}C method to estimate C turnover times of soil fractions, the two-point and the multi-point calculations generate similar values, although the former method estimates were slightly shorter C turnover times than the latter (Fig. 3). This finding demonstrates that the repeatability is high when using the ^{13}C method to estimate C turnover times of soil fractions (Fig. 2). The multi-point ^{13}C method is recommended to calculate soil C turnover, because it generates results with higher confidence. Derrien and Amelung (2011) also found that multiple-time measurements of δ^{13}C are better for estimating C turnover times, because this method can assess C turnover times at both steady and non-steady states while the two time-point measurements cannot. This study suggests that when multiple-time point measurements of δ^{13}C of soil fractions are not available, two time-point measurements can be used as a substitute to give reasonable estimates of C turnover times.

Using Δ^{14}C to estimate C turnover times of soil fractions, caution should be exercised concerning the calculation approach used. We found that C turnover times of all studied soil fractions estimated by the conventional ^{14}C model were 4–5 folds longer than those by the bomb ^{14}C model (Fig. 4), and were also longer than the values used in current multi-compartment soil C models (Jenkinson and Rayner, 1977; Parton et al., 1987). Thus, C turnover times of soil fractions estimated by the bomb ^{14}C model are recommended when simulating SOC dynamics by using multi-compartment models. This estimate divergence probably results from different assumptions of these two models. The conventional ^{14}C model assumes that C in different soil fractions are formed directly from external C sources with the age of zero, but C in some soil particles (e.g., the silt and clay sized particles) may be formed from the transfer of C in coarse soil particles with the age older than zero (Trumbore, 1993; Bruun et al., 2005). The bomb ^{14}C model that considers continuous C inputs to soils is more realistic, because it uses abundant ^{14}C derived from the 1950s bomb test as a tracer and the numerical solution to estimate C turnover times are more accurate when compared to the conventional ^{14}C method (Trumbore, 1993; Bruun et al., 2005). However, the steady state assumption may underestimate the turnover times of SOC fractions which need a long time to reach equilibrium (Bruun et al., 2005).

Even using the same estimate method, C turnover times of the same soil fraction still vary greatly (Fig. 2). This is likely because soils come from a variety of environments, where climate, vegetation, microbial community, and soil mineralogy and depth likely influence C turnover times in soils. Among these factors, soil depth is important in impacting C turnover time of soil fractions. We observed that at the same site, C turnover times of a given soil fraction generally increase with depth, regardless of the estimate method used (Skjemstad et al., 1990; Schöning and Kögel-Knabner, 2006; Yonekura et al., 2013; Dalal et al., 2013; Beniston et al., 2014; Liang et al., 2014). This finding is consistent with other studies (Rumpel et al., 2002; Rumpel and Kögel-Knabner, 2011; Mathieu et al., 2015). We did not observe longer C turnover times of soil fractions at high latitude than at low latitude. It is likely that local environments at studied sites, such as SOM chemistry and soil properties, cause large variations of C turnover times of soil

fractions, which masks the influences of latitude and associated climate on C turnover times. The reason might also be that there is not sufficient data on C turnover times along a latitude gradient to generalize patterns of how it changes with climate.

Although our study has used soil fractions from locations worldwide to estimate C turnover times by the laboratory incubation, the δ^{13}C after C_3–C_4 vegetation change, and the ^{14}C dating (Fig. 1), we are aware that these soil fractions used for estimation by these three methods are not the same or well paired. So, we cannot attribute the variations of C turnover times of soil fractions solely to different estimation methods. Other factors, such as temperature, precipitation, soil depth, and soil texture, that has been found to influence C turnover times of bulk soil might impact C turnover times of soil fractions as well (Carvalhais et al., 2014; Mathieu et al., 2015; Xu et al., 2016). To parameterize soil C turnover times in the multi-compartment models, we highly recommend studies that assess C turnover times of the same physically fractionated soil particles by using different methods.

This synthesis study compared C turnover times of physically fractionated soil fractions estimated using three methods: (1) laboratory incubation, (2) δ^{13}C after C_3–C_4 vegetation change, and (3) ^{14}C dating. We found that estimated C turnover times of soil fractions differed significantly among methods. We suggested that the relatively fast soil C turnover time found by the incubation studies under optimal environmental conditions are likely an overestimate of C turnover rates under field conditions, as soil moisture and temperature are not always at optimum levels in nature. Estimates derived from δ^{13}C and Δ^{14}C are likely closer to actual C turnover rates found in the field. However, the ^{13}C method can only be used when there are detectable changes in δ^{13}C after years of successive C_3 versus C_4 vegetation change, and ^{14}C dating could more accurately estimate C turnover of soil fractions when soils are under steady-state conditions or ^{14}C inputs derived from atmosphere and vegetation are well documented. It is noticeable that when using the ^{14}C dating method the presence of black C in soils could bias C turnover times of coarse organic matter, which is considered to be labile and has short turnover times (Baisden et al., 2002; Leifeld, 2008; Leifeld et al., 2015). Overall, these findings suggest that consideration should be given to methodological differences when using C turnover data to inform and parameterize soil C models.

Acknowledgements

Appendix A. Supplementary data

Supplementary data related to this article can be found at http://dx.doi.org/10.1016/j.soilbio.2016.06.003.

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