



Isotopic characterization of sequestration and transformation of plant residue carbon in relation to soil aggregation dynamics



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ABSTRACT

Soil aggregates play a key role in preserving soil organic carbon (SOC). However, the mechanisms controlling aggregate formation and SOC distribution in different sizes of aggregates remain unclear. Here, we studied the dynamics of aggregate formation and associated SOC transferring among different aggregate fractions using an isotopic tracer technique (¹⁴C-labeled wheat residues). The year-long incubation results demonstrate that the wheat residue carbon applied to soil (Typic Hapludoll) was initially mainly stored in the pores between microaggregates that were agglomerated to form macroaggregates (>250 μm). The wheat carbon started to transfer to the inside of microaggregates (250–53 μm) after six months of the incubation. The newly formed humic acid carbon (HA¹⁴C) and fulvic acid carbon (FA¹⁴C) in the macroaggregates were 57–90% and 60–84% more than in microaggregates (250–53 μm), respectively. Later, the sequestered SOC was decomposed or chemically transformed, resulting in a decrease in the ratio of HA¹⁴C/FA¹⁴C in macroaggregates and an increase in the silt/clay fraction (<53 μm). This result indicated that the SOC in the macroaggregates was vulnerable to degradation due to less protection by soil macrostructure, while it experienced a slow degradation due to strong surface adsorption or pore protection in soil microstructure. This study suggests that macroaggregates considerably control SOC turnover and thereby their stability considerably influences how much newly introduced organic carbon could be sequestered into stable carbon pools like microaggregates. The turnover dynamics of macroaggregates provided insights into the potential of humic carbon formation that facilitates long-term carbon preservation in soil.

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1. Introduction

Content and chemical composition of soil organic matter (SOM) are influenced by management and amendment practices (Medina et al., 2015). In general, intensive cropping with conventional soil tillage causes SOM mineralization and depletion, whereas conservative practices, such as reduced tillage, organic fertilization, and residue incorporation, increase the content and change the quality of SOM (Simonetti et al., 2012). In many soils, the dynamics of soil structure development is closely related to the cycling of SOM. SOM is known to have a strong relationship with aggregate formation and stabilization (Six et al., 2002). The influences of particulate and humified organic materials, plant-derived polysaccharides, fungal hyphae, roots, microorganisms,

and microbial exudates on the formation of soil aggregates have been documented extensively (Lee et al., 2009; Sodhi et al., 2009; Bravo-Garza et al., 2010; Lugato et al., 2010; Majumder et al., 2010). The effects of organic inputs are larger on the formation of macroaggregates than microaggregates. The portion of macroaggregates increases with organic fertilization (Gryze et al., 2005; Yang et al., 2007; Lee et al., 2009; Sodhi et al., 2009; Lugato et al., 2010; Lucas et al., 2014). According to the hierarchical theory (Tisdall and Oades, 1982; Oades and Waters, 1991), water-stable microaggregates (250–53 μm) and silt/clay (<53 μm) are bound together to form macroaggregates (>250 μm) by organic compounds of different origin and stability. Microaggregates are usually stabilized by persistent binding agents (such as humic substance and organo-mineral complex), whereas the stability of macroaggregates is affected by the fate of transient, young organic materials (such as microbial- and plant-derived polysaccharides) or temporary binding agents (such as fungal hyphae and roots) (Lugato et al., 2010). As a result, larger aggregates contain more soil

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organic carbon (SOC) under no-tillage and application of crop residues (Choudhury et al., 2014; Liu et al., 2014).

Soil aggregates are structural units within soil that control the dynamics of SOM and nutrient cycling (Six et al., 2004). Physicochemical protection offered by aggregate structures and mineral surfaces is critical for building and maintaining soil carbon and nitrogen stocks (O'Brien and Jastrow, 2013). The decomposition of plant biopolymer residues in agricultural soils is mostly mediated by microorganisms, whose activities are subject to soil structure and associated water conditions. A part of the organic carbon input that is not completely decomposed to CO_2 are transformed into humic substances through the humification process—a biochemical protection mechanism (Sarkhot et al., 2007; Simonetti et al., 2012). Humic substances are considered to be recalcitrant due to their chemical composition with richness in aromatic and aliphatic structures (Simonetti et al., 2012). In view of the close linkage between aggregate structure and carbon turnover, aggregate fractionation is regularly used to identify the carbon sequestration process (Blanco-Canqui and Lal, 2004; Six et al., 2004; O'Brien and Jastrow, 2013). Overall, soil aggregates is suitable places, where SOC could be preserved and humified (Chaney and Swift, 1986; Fortun et al., 1989; Piccolo et al., 1997; Albert et al., 2005). However, it is unclear how the turnover dynamics of macroaggregates determine the transfer and/or sequestration of fresh organic carbon into microaggregates.

In this study, we hypothesize that the interaction between fresh organic carbon and aggregates involve a two-way selective action. On one hand, protection (or access cutoff) of organic carbon introduced into soil varies with the sizes of native and restructured aggregates, altering the degradation of fresh organic matter, ratio of SOC to humic carbon, and constitution of humic substances in different aggregate fractions. On the other hand, the differences in chemical properties and constituents of organic substances determine the stability of each aggregate fraction and associated resistance to microbial decomposition of the sequestered organic matter. Such a two-way selective process (or feedback effect) eventually reaches a balanced status after a certain length of time, which is subject to agricultural managements, such as fertilizer application, irrigation, and tillage. This study aimed to clarify such kind of selective mechanism through isotopic analysis of the formation of humic acid (HA) and fulvic acid (FA) from wheat residues in water-stable aggregates. The study provides theoretical insights into carbon sequestration through agricultural practices.

2. Materials and methods

2.1. Soil and sampling

A total of nine soil cores (7 cm inner diameter and 20 cm height) was randomly collected from the top soil layer (0–20 cm) in a long-term agricultural experimental station, which is located in Gongzhuling City, Jilin Province, China (43°31'N, 124°49'E). The local climate was temperate, humid continental with mean annual temperature of 5.6 °C and mean annual rainfall of 570 mm. The soil is classified into Typic Hapludoll with a loamy clay texture (39% sand, 30% silt, and 31% clay) as measured according to the method described by Gee and Bauder (1986). The soil secondary clay minerals are rich in illite and montmorillonite. The sampled soil was air-dried, grounded, and passed a sieve (2 mm mesh pore) for incubation experiments and measurement of soil basic properties. Each of the three replicate incubation experiments used a mixture of soils from three cores. On average, the soil contained 10.91 g organic carbon kg^{-1} dry soil, 1.15 g total nitrogen kg^{-1} dry soil, 109.40 mg available phosphorus kg^{-1} dry soil, 42.84 mg available potassium kg^{-1} dry soil, 114.00 mg hydrolysable nitrogen kg^{-1} dry soil, and a bulk density of 1.20 g cm^{-3} (Bremner, 1996).

2.2. Soil incubation

^{14}C -labeled wheat residues used in the incubation were obtained with courtesy from Sweden Agricultural University. The plant material contains ^{14}C with a specific activity of $3.70 \times 10^3 \text{ kBq g C}^{-1}$ as well as 470.32 g organic carbon, 6.40 g total nitrogen, 25.42 g humic-like acid, and 34.27 g fulvic-like acid kg^{-1} plant material. During the incubation, the water content of experimental soil (sieved through 2 mm) was remained with deionized water at a certain level representing field conditions. After equilibrium with water for seven days, the wet soil (equivalent to 200 g dry soil) was weighed and thoroughly mixed with 1.00 g ^{14}C -labeled wheat residues (sieved through 0.25 mm), 8.00 g non-labeled ^{12}C wheat residues (for maintaining microbial activity), and 0.05 g $(\text{NH}_4)_2\text{SO}_4$ (for adjusting C/N ratio to 25:1 to support microbial activity). The treated soil was then transferred into a 250 mL glass beaker under a moisture condition of 70% field capacity (equivalent to water content 25.67% in mass, a dominant water condition in the field site). The beaker was placed in a 2 L jar containing a CO_2 trap (10 mL 1 M NaOH). Water was added to the jar to maintain a moisture-saturated atmosphere. The soil was then incubated in the dark at 25 °C for 360 days. Each of the triplicate incubation experiments had three duplicate containers that allowed sampling for three times on the 60th, 180th, and 360th days, respectively. After taken out, soil samples were oven-dried at 40 °C to terminate microbial activity. The CO_2 traps were replaced with fresh ones daily in the first week, every two days in the second week, every three days in the third week, and weekly in the remaining time (Majumder and Kuzyakov, 2010).

2.3. Water-stable aggregate fractionation

Water-stable aggregates were isolated according to the method of Cambardella and Elliot (1993). One hundred grams of air-dried soil, which was sampled from the incubation containers, were submerged in deionized water at a soil-to-water ratio of 1:80 for five minutes at room temperature (22 °C) on a set of sieves consisting of 2000 μm , 250 μm , and 53 μm sieves. After aspirating away any floating litter, the >2000 μm , 2000–250 μm , and 250–53 μm aggregates were extracted from the sampled soil by moving the sieves three centimeters up and down for 30 times per minute in two minutes. Soil aggregates retained on corresponding sieves were then backwashed into pre-weighed containers. Silt and clay particles (<53 μm) were collected after the suspension had settled for 72 h. All samples were finally oven-dried at 50 °C until reaching a constant weight prior to weighing and storage in closed glass jars for SOC analysis.

2.4. Soil chemical analysis

Humic substances were extracted with 0.1 M mixture of NaOH and $\text{Na}_4\text{P}_2\text{O}_7$ from five grams of sieved soil or aggregate fractions at 70 ± 2 °C under oscillation for one hour. Then, 0.5 M H_2SO_4 was added to the extract to regulate pH to 1.0–1.5. Organic acids in the solution was taken as fulvic acid (FA) and precipitate as humic acid (HA). The HA was then filtered out for analysis, and the FA was analyzed directly as aqueous solution.

^{14}C activity (DPM) (including $^{14}\text{CO}_2$ trapped in NaOH, HA^{14}C , and FA^{14}C) was determined by liquid scintillation counting (Tri-carb2800) with addition of 2.5 mL scintillation cocktail to 0.5 mL aliquot of NaOH after a 24-hour decay of chemiluminescence. Radioactivity of residual ^{14}C in soil was obtained by subtraction.

The ^{14}C activity of the plant residues was determined after dry combustion of one gram of sample within an oxidizer unit (Packard Model 507) under superfluous oxygen while $^{14}\text{CO}_2$ was trapped into 15 mL NaOH. Thereafter, the ^{14}C activity of NaOH solution was

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