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Aggregate Distribution and the Associated Carbon in Norfolk Soils under Long-term Conservation Tillage and Short-term Cover Cropping

Binaya Parajuli (b^a, Min Luo^b, Rongzhong Ye^a, Thomas F. Ducey^c, Dara Park^a, Matthew Smith^a, and Gilbert Sigua^c

^aPlant and Environmental Sciences, Pee Dee Research and Education Center, Clemson University, Florence, SC, USA; ^bSchool of Environment and Resource, Fuzhou University, Fuzhou, China; ^cUSDA - Agricultural Research Service, Coastal Plains Soil, Water, & Plant Research Center, Florence, SC, USA

ABSTRACT

Conservation agriculture practices have been widely implemented to improve soils and their sustainability. Here, we investigated the impacts of long-term conservation tillage (CS) on soil aggregates and their associated carbon (C) in a typical sandy Ultisols. The short-term effect of cover cropping was also investigated. Soils (0-5 and 5-15 cm) were collected from fields under 40years CS and conventional tillage (CV), in which cover crop and fallow treatments were embedded as a split-plot design for four years. Soils were tested for bulk density, pH, mean weight diameter, dry aggregate distribution, and C contents. The CS only resulted in higher total C (TC) concentrations $(13.56 \pm 1.14 \text{ g kg}^{-1})$ than CV $(10.06 \pm 0.53 \text{ g kg}^{-1})$ at 0–5 cm. Similar depth impacts were observed in various aggregate size fractions (2000-250 µm, macroaggregates; 250–53 µm, microaggregates; 0–53 µm, clay-silt fraction). Soils were dominated by macroaggregates (>50%) at both depths with associated C concentrations following the order of clay-silt fraction > microaggregates > macroaggregates. Nonetheless, after accounting for soil bulk density long-term CS did not result in higher TC stocks than CV at both depths. Cover crops demonstrated no effect on C stocks at both depths. Poor soil structure and low clay contents of the tested soils may partially explain these neutral impacts, while relatively lower residue returns of cover crops to cash crops may also confound the positive management outcomes. Novel management practices to increase soil organic C in the bulk soils by increasing organic inputs and the soils' capacity to preserve the inputs are needed.

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Soil organic carbon; soil aggregates; conservation tillage; cover crop; sandy coastal plain soils; ultisols

Introduction

Soil organic carbon (SOC) is one of the major determinants of soil fertility and a useful indicator of soil health (Lal 2016). The SOC storage in soils is a balance between inputs as primary production (e.g., root exudates, plant litter, and residues) and outputs, mainly as microbial decomposition (Jastrow, Amonette, and Bailey 2007). Current management strategies to improve SOC are therefore mainly focused on increasing organic inputs, including crop residue retention, applications of animal manure and compost materials, and cover-cropping, while reducing the outputs through minimized soil disturbance and permanent soil cover. Both strategies have been widely demonstrated to yield positive outcomes in many agricultural soils (Jastrow, Amonette, and Bailey 2007; Schmidt et al. 2011).

Conservation tillage (CS) is a common and effective practice in reducing SOC loss by maintaining natural soil structure and the stability of soil aggregates protecting SOC against microbial

decomposition (Bottinelli et al. 2017; Bronick and Lal 2005a; Curaqueo et al. 2011; Tian et al. 2015). Soil aggregation is a main mechanism of SOC stability making SOC inaccessible to microbes and their degradative enzymes. For instance, no-till was found to increase SOC by 325 ± 113 kg C ha⁻¹ year⁻¹ in both tropical and temperate systems (Six et al. 2002a; 2002b). Similarly, 14-year CS increased SOC, water-soluble C and microbial biomass C by 48%, 78% and 72%, respectively, as compared to conventional tillage (CV) (Madejón et al. 2007). However, the CS have been found to impact only the top soils (Álvaro-Fuentes et al. 2004; 2006; Novak, Bauer, and Hunt 2007) most likely due to the residues being remained at the surface. Angers and Eriksen-Hamel (2008) found higher C in surface soil for CS whereas CV had higher C at lower plow layer (15 cm). This discrepancy on C distribution due to tillage has showed the necessity of assessing soil C to lower depths. Lal (2009) suggested the extension of soil sampling depth to 1 m to precisely study the management induced changes of SOC stock.

Like CS, cover crops, i.e., the unharvested crops planted in rotation between cash crops (Farooq and Siddique 2016), have been demonstrated to have diverse benefits, including improved soil aggregate stability (Abdollahi and Munkholm 2014), and increasing SOC (Poeplau and Don 2015). Poeplau and Don (2015) estimated that cover crops increased C sequestration at a rate of 320 ± 80 kg ha⁻¹ yr⁻¹. Inclusion of cover crops with no tillage was estimated to accumulate 100 to 1000 kg C ha⁻¹ when compared with no-till without cover crops (Blanco-Canqui et al. 2013). However, some researchers suggest that higher amounts of organic residue inputs accelerated SOC decomposition (i.e. priming effect) reducing SOC storage (Fontaine et al. 2007; Manzoni et al. 2012) whereas, some studies demonstrate no effect on SOC with cover crops (Fronning, Thelen, and Min 2008; Mubiru and Coyne 2009). The discrepancy may be partially explained by selecting different cover crops with distinctive functional traits e.g., nitrogen fixation capacity and biomass production potentials, in addition to variable climatic and edaphic factors, highlighting the need for regional or site-specific research.

Coastal Plain soils in the southeastern US is often characterized by degraded soil health with low nutrient and water holding capacities, largely attributable to the hot and humid climate favorable for the decomposition of organic materials and historical intensive managements resulting in the losses of SOC. Improving SOC, and subsequently, soil health, is the management priority for sustainable production in the region, where nearly a quarter of US agricultural products are produced (Ruth, Coelho, and Karetnikov 2007). Recent research suggests that long-term applications of minimal tillage increases SOC in a typical Coastal Plain soil, in which, however the SOC is seemingly approaching the C saturation point (Nash et al. 2018; Novak, Bauer, and Hunt 2007). Similar results were also observed in a study on 48 farmer's fields in Virginia Coastal Plain soil (Stewart et al. 2012). Despite these findings, increasing organic inputs combined with reduced soil disturbance is still considered the best management option for Coastal Plain soils.

It is generally accepted that the soil clay plus silt fraction determines the soils' capacity to preserve SOC, while soil aggregates provide additional variable capacity through additive physical protection that can be modified by soil management (Hassink and Whitmore 1997; Carter 2002; Six et al. 2002a; Denef et al. 2004). A better understanding of whether and how the management practices improve soil structure and SOC distributions in different soil aggregates may provide insights to optimize or improve management outcomes of the sustainable practices (Bronick and Lal 2005b). In the present study, we investigated the impacts of long-term (~40 years) CS with short-term (4 years) cover crops on soil aggregates distribution, aggregate stability, the associated C contents, and stocks in bulk soils. It was hypothesized that: (1) CS improves soil aggregation and TC stocks in aggregates, especially the microaggregates and (2) integrations of cover crops with CS result in additive positive effects.

Materials and methods

Site description

The experimental site located at Pee Dee Research and Education Center of Clemson University at Florence, SC, USA (34°18' N, 79°44' W), and has been under continuous management by the

Agricultural Research Service, USDA for over 40 years. Annual mean temperature was 17.1°C with the maximum in July (23.8°C) and the minimum in December (11.1°C), while the mean annual precipitation was 1186 mm (US Climate Data; https://www.usclimatedata.com).

The research plots were established in 1979 to examine tillage impacts on soil properties and crop yield with all crop residues returned to the field. There were two types of tillage treatments: CV (i.e. disking soils at 15 cm depth for 2–3 times and subsoiling to 42 cm -depth once before spring crops) and CS (subsoiling to 42 cm -depth soils once before spring crops). Each was replicated five times in the field. Since the mid-1980s, the main crop has been corn, rotated with soybean or cotton. In 2015, cover crops mixture consisting of rye and crimson clover was introduced as a split plot design within the tillage. The plots therefore have four treatments imposed: CS with cover crops (CS- Cover), CS without cover crops (CS-Fallow), CV with cover crops (CV-Cover), and CV without cover crops (CV-Fallow).

The soils were primarily Norfolk loamy sand (Fine-loamy, siliceous, thermic Typic Kandiudults). Cash crops were planted between April and May and harvested between October and November, while the cover crop mixtures were planted after the harvests and terminated with herbicides prior to planting the cash crops. Fertilizers were applied when necessary according to recommended guidelines provided by the Agricultural Service Laboratory of Clemson University, based on soil testing results. Further details about site description, climate, crop rotation and, management practices can be found in Bauer, Hunt, and Camp (1997), (2006), Campbell, Karlen, and Sojka (1984), Hunt et al. (2004), Novak, Bauer, and Hunt (2007), Nash et al. (2018).

Soil sampling

In May 2018 (prior to the cover crops termination), eight soil cores (0–15 cm) were randomly collected with an AMS soil core sampler (5 cm in diameter) from each plots of the first four replications, totaling to 16 plots (4 replicates by 4 treatments). Each soil core was sectioned into 0–5 and 5–15 cm fractions and sieved through a 2 mm sieve. Samples from same plots at the same depth were composited and stored at 4°C until used.

Soil analyses

Bulk density was determined with the AMS soil core sampler (5 cm in diameter, 15 cm in depth). The collected soils were oven-dried at 60°C until constant weight was attained. Soil pH was analyzed with an Orion 8107 pH probe (Thermo scientific, Waltham, MA) with DI water (1:1 ratio) after equilibrating the mixture for 30 mins. Soil particle size distributions were estimated via micropipette method (Miller and Miller 1987). Moisture content was analyzed as loss of mass after oven drying at 60°C until constant weight was attained. Wet soil aggregate stability was measured via wet sieving (Six, Elliott, and Paustian 2000; Six et al. 2000; Márquez et al. 2004). Mean weight diameter (MWD) was calculated after correcting for sand content (Márquez et al. 2004). Distribution of soil aggregates (2000–250 μ m, 250–53 μ m and, 0–53 μ m) was also measured via sieving the airdried soil through stacked sieves. Different size samples were used for total C (TC) analysis with a Carlo- Erba NA 1500 CNS analyzer (Haak-Buchler Instruments, Saddlebrook, N.J.) to analyze the distribution of C in aggregates.

Statistical analyses

At each soil depth, a two-way analysis of variance (ANOVA) was used to test the main plot factor (i.e. tillage), the subplot factor (i.e. cover cropping), and their interaction for sand, silt and clay content, pH, soil bulk density, MWD and TC. A three-way ANOVA was used to test same factors, aggregate size fraction, and their interactions for aggregate-associated C. All data were tested for normality (Levene's test) and homogeneity (Shapiro-Wilks test). Non-normal data (MWD and aggregate-associated C) were log-transformed. The statistical analyses were performed using JMP Pro 14 (SAS

Institute, Cary, NC) with $\alpha = 0.05$. When effects were found significant, means were compared using the Student's t-test. Data presented are in its raw form.

Results

Soil physical properties

The tested soil had a loamy sand texture. Both long-term tillage and short-term cover cropping did not affect the soils' clay (7.0% to 8.1%) and silt (13.2% to 16.1%) contents at both soil depths (Table 1). Similarly, soil bulk density (1304 to 1516 kg m⁻³) and MWD (0.26 to 0.43 mm) showed no significant response to tillage treatments, which was not affected by integrating winter cover crops (Table 1). In addition, both tillage and cover crops had no effects on aggregate distribution (Figure 1). The aggregates were dominated (> 50%) by macroaggregates (250–2000 μ m), followed by microaggregates (53–250 μ m) and clay-silt fractions (0–53 μ m) (Figure 1).

Soil chemical properties

No significant treatment effects were found for soil pH (5.6 to 5.9) at both soil depths (Table 1). Cover cropping had no effects on TC concentrations, while the tillage effect was significant for TC only at 0–5 cm, in which it was higher in CS soils (13.2 to 13.9 g kg⁻¹) than in the CV soils (10.0 to 10.2 g kg⁻¹) (Table 1). However, after accounting for soil bulk density, the soil C stocks were not affected by the two treatments (Figure 3). At 0–5 cm depth, tillage has marginal effect (p = .08) with the C stocks being 9.11 and 6.97 in t ha⁻¹ in CS and CV soils, respectively.

Aggregates associated C

Cover cropping had no significant effects on the concentrations of aggregate-associated C (Figure 2). In contrasts, both tillage and aggregate size treatments affected the concentration in soils at 0-5 cm with significant tillage and aggregate size interactions (Figure 2). Regardless of cover crops effects, at 0-5 cm the CS had higher C concentrations than CV soils at all the three aggregate size fractions. Similarly, the concentrations associated with clay-silt fraction were the highest, followed by micro-aggregate and macroaggregate fractions, in both CS and CV soils (Figure 2). In soils at 5-15 cm, except fraction size, no significant treatment effects were found (Figure 2). Aggregates C concentrations at

Table 1. Selected soil properties of a Norfolk soil under different management practices. Values are means $(n = 4) \pm$ standard error. The results of *p* values are italicized.

	pН	BD	Clay %	Silt %	Sand %	MWD (mm)	TC	
0–5 cm								
CS-Fallow	5.7 ± 0.1	1304 ± 86	7.3 ± 0.3	13.3 ± 2.5	79.4 ± 2.85	0.30 ± 0.04	13.2 ± 1.8	
CS-Cover	5.7 ± 0.1	1370 ± 34	7.3 ± 0.3	13.2 ± 1.6	79.5 ± 1.80	0.27 ± 0.04	13.9 ± 1.6	
CV-Fallow	5.6 ± 0.1	1430 ± 14	7.8 ± 0.2	13.3 ± 1.0	78.9 ± 0.86	0.29 ± 0.03	10.0 ± 0.7	
CV-Cover	5.8 ± 0.1	1331 ± 48	8.0 ± 0.3	13.2 ± 1.5	78.8 ± 1.26	0.26 ± 0.01	10.2 ± 0.9	
Tillage (T)	0.82	0.49	0.11	0.99	0.80	0.92	0.04*	
Cover (C)	0.36	0.73	0.62	0.95	0.96	0.27	0.72	
Τ×C	0.51	0.12	0.74	0.98	0.93	0.96	0.84	
5–15 cm								
CS-Fallow	5.9 ± 0.1	1511 ± 37	7.0 ± 0.4	13.7 ± 3.2	79.3 ± 3.50	0.32 ± 0.04	7.10 ± 2.3	
CS-Cover	5.9 ± 0.2	1479 ± 86	7.2 ± 0.3	14.6 ± 3.6	78.2 ± 3.55	0.34 ± 0.06	6.05 ± 0.8	
CV-Fallow	5.8 ± 0.1	1409 ± 43	7.7 ± 0.5	16.1 ± 2.0	76.1 ± 1.78	0.42 ± 0.04	7.69 ± 1.0	
CV-Cover	5.8 ± 0.1	1516 ± 54	8.1 ± 0.5	15.2 ± 2.8	76.7 ± 2.37	0.43 ± 0.07	7.40 ± 1.0	
Tillage (T)	0.37	0.65	0.17	0.73	0.58	0.21	0.58	
Cover (C)	0.55	0.44	0.45	0.99	0.79	0.67	0.58	
Τ×C	0.61	0.17	0.73	0.31	0.40	0.93	0.75	

Note: * indicate significant difference at α = 0.05; BD, bulk density (kg m⁻³); MWD, mean weight diameter; TC, total C (g kg⁻¹ soil); CS, conservation tillage; CV, convention tillage; Fallow, without winter cover crops; Cover, with winter cover crop.



Figure 1. Soil aggregate distribution (%) at 0–5 and 5–15 cm depths. Bars indicate one standard error of means (n = 4). Main effects of Tillage (T), Cover crop (C) and their interactions were presented with significant level at α = 0.05. CS, conservation tillage; CV, conventional tillage; Cover, with cover crops; Fallow, without cover crops.

5–15 cm depth were higher in clay-silt fractions and microaggregates followed by macroaggregates, respectively (Figure 2).

Discussion

It is generally believed that beyond the clay-silt protective mechanism, additional C storage occurs primarily in aggregates (Dungait et al. 2012; Stewart et al. 2012), which can be attained with minimal soil disturbance and soil cover from residue retention and cover crops (Lal 2015; Six et al. 2004). However, in the present study, we demonstrated that 40-year of CS, when compared with CV, did not result in significant improvement of soil structure (described by aggregate distribution and stability) and increase of TC stocks in sandy soils at 0–15 cm (Table 1; Figure 3) (not supporting the Hypothesis 1), although it did increase TC concentrations in the



Figure 2. Total C concentration (g kg⁻¹) of different aggregate size fractions. Bars indicate one standard error of means (n = 4). Main effects of Tillage (T), Cover crop (C) and their interactions were presented with significant level at α = 0.05. CS, conservation tillage; CV, conventional tillage; Cover, with cover crops; Fallow, without cover crops; Ft, fraction size.

bulk soil and soil aggregates (Table 1; Figure 2) at 0-5 cm, which, unexpectedly, was not affected by inclusion of cover crops (not supporting Hypothesis 2).

Tillage effect on aggregate distribution and stability

Conventional tillage practices often result in the breakdown of aggregates, which degrades soil structure resulting in loss of SOC (Jat et al. 2019; Six et al. 2002b). The CS is therefore widely considered as an alternative management. While many studies have demonstrated improved soil structure and increased SOC (Six et al. 2002a; 2002b; West and Post 2002), some researchers also suggested no impact of CS on soil structure and C content (Chivenge et al. 2007). In present study, 40-year CS did not change soil bulk density, aggregate distribution, and MWD in soils at either 0–5 or 5–15 cm depths (Table 1; Figure 1). According to the aggregate hierarchy theory, soil aggregates are formed in ordered stages with different binding agents (Tisdall and Oades 1982). Microaggregates are formed from progressive binding of primary particles (i.e. clay and silt) with the persistent binding



Figure 3. Carbon stocks (Mg ha⁻¹) in bulk soil sat 0–5 and 5–15 cm depths. Bars indicate one standard error of means (n = 4). Main effects of Tillage (T), Cover crop (C) and their interactions were presented with significant level at α = 0.05. CS, conservation tillage; CV, conventional tillage; Cover, with cover crops; Fallow, without cover crops.

agents e.g. polyvalent metal cations, while macroaggregates formed by the bindings of microaggregates with temporary or transient binding agents, such as microbial and plant-derived organics (Six et al. 2000; Six, Elliott, and Paustian 2000; Six et al. 2002b). The tested soils were dominated by nearly 80% sand (Table 1). It is highly possible that the relatively low clay and silt contents of the surface soils impeded the realization of positive outcomes of CS. In agreement with this assumption, Ye, Parajuli, and Sigua (2019) demonstrated that amending top soil with clay soils improved soil structure by promoting soil aggregation and aggregate stability resulting in preservation of organic C in a degraded Coastal Plain Ultisol.

Tillage effect on C associated with aggregates

Despite its insignificant impacts on soil aggregates distribution (Figure 1), CS significantly increased TC concentrations in the topsoil (0–5 cm) by promoting C accumulations in aggregates (Figure 2), especially in the clay-silt fractions, highlighting the importance of clay and aggregation in C sequestration and stabilization (Barré et al. 2014; Singh et al. 2018; Tisdall and Oades 1982). However, like many similar studies (Novak, Bauer, and Hunt 2007; Novak et al. 2020; Somasundaram et al. 2017), such effects were not observed in soils at 5–15 cm, likely because the retained crop residues were not incorporated into deep soils. Soil C stocks are the balance between inputs (e.g., plant residues) and outputs (e.g., microbial decomposition), which can be regulated by various soil management practices (Mazzilli et al. 2014; West and Post 2002). Chivenge et al. (2007) proposed a conceptual model describing the interactive effects of management practices and soil texture on SOC, in which residue management can increases TC in sandy soils mainly by increased

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organic inputs, while tillage management increases TC by promoting aggregate formation in clay soils (i.e. reducing outputs). The concept underlined the importance of increasing plant biomass inputs to improve TC in sandy soil such as the Coastal Plain soils (Nash et al. 2018; Novak, Bauer, and Hunt 2007), where soil structure is poor and protection of SOC against microbes and their degradative enzyme is weak.

As expected, TC concentrations associated with aggregates followed the order of clay-silt fraction > microaggregate > macroaggregate (Figure 2) (Bronick and Lal 2005a; Jagadamma and Lal 2010). Clay and silt participate actively in the formation of organo-mineral complex with SOC due to their high charge and specific surface area (Chenu, Plante, and Puget 2017). Chivenge et al. (2007) suggested that sandy soils are often unable to store TC in a long term since the TC is often not protected by aggregation and can be easily decomposed without soil disturbance. Novel management practices to preserve the increased organic inputs against microbial decomposition are therefore needed to support the productivity and sustainability of the sandy coastal plain soils.

Tillage effect on TC stocks

Although, when compared to CV soils, significant C accumulations were observed in clay-silt fractions in CS soils at 0-5 cm (Figure 2), only marginal tillage affects (p = .08) were found on the TC stocks (accounting for soil bulk density) in soils at the same depth (Figure 3). The dominance of macroaggregates over clay-silt fractions likely weakened such tillage impacts (Figure 1). However, similar study conducted in 2002 on 0-5 cm soils from the same experimental plots demonstrated significant increase of TC stocks when CS was compared with CV (Novak, Bauer, and Hunt 2007). The different results between the present study and the one by Novak, Bauer, and Hunt (2007) may be explained by the observation that these soils are approaching C-saturation point (Nash et al. 2018; Novak et al. 2020; Stewart et al. 2012), reducing the efficiency and effectiveness of CS soils in preserving organic inputs from crop residues during the past 18 years when compared with CV soils, confounding the treatment effects (Chivenge et al. 2007). The distinctive changes in TC stocks can also be due to soil sampling at different time, resulting in different soil bulk density. For instance, Madejón et al. (2007) found variation in TC between soil samples measured at summer and fall season. Our soil samples were collected in May prior to spring crop planting whereas Novak, Bauer, and Hunt (2007) collected soil samples after the spring crop harvest. Farm machineries used during the harvest may compact soils increasing in bulk density (McNabb, Startsev, and Nguyen 2001).

Cover cropping effect on soil structure and TC

Integrating cover crops increased organic inputs to soils and was therefore expected to increase TC content. Nonetheless, documented research reported positive (Lal 2004; 2015; Poeplau and Don 2015), neutral (Fronning, Thelen, and Min 2008; Mubiru and Coyne 2009) and negative (Fontaine et al. 2007) impacts. Causarano et al. (2006) suggested that TC contents can be doubled in southeastern US fields when cover crops are combined with CS. However, in the present study, regardless of tillage method, cover crops impacts were insignificant on soil structure (Table 1; Figure 1), TC concentrations and stocks (Figures 2 and 3). There are several explanations for such findings. Firstly, the soil has intrinsically low clay content limiting its capacity to improve soil structure to protect the increased organic inputs (discussed above). Secondly, cover crops were introduced within 4 years and any changes of TC stocks are difficult to detect. Goidts, van Wesemael, and Crucifix (2009) suggested that cover crops resulted in annual TC sequestration of 320 ± 80 kg ha⁻¹ yr⁻¹, which however largely dependent on the time since introduction and site elevation (Poeplau and Don 2015).

It is believed that the capacity of cover crops to increase TC in agricultural soils are often dependent on the absolute carbon inputs (Poeplau and Don 2015), which may be more evident in the coastal plain

regions where the decomposition is very high (Hubbard, Strickland, and Phatak 2013). The relatively low C inputs by cover crops compared to that of cash crop residue in 16 years observed in the present study may also confound the cover-cropping impacts (Nash et al. 2018). Indeed, cover crop biomass production in these soils, e.g., 2300 kg ha⁻¹ for rye and 1300 kg ha⁻¹ for crimson clover (Bauer and Roof 2004), was often lower than crop residue retention, e.g., 7600 kg ha⁻¹ for soybean and 14,400 kg ha⁻¹ for corn (Nash et al. 2018). In this context, managing cover crops to improve their biomass production may be essential, e.g. cover crops selection, breeding, fertilization and planting and termination timing.

Summary

We investigated long-term (40 years) impacts of CS on the distribution and stability of soil aggregates and associated SOC stocks in a typical Ultisol in the Coastal Plain of the southeastern US. We also examined whether the integration of cover crops improve CS management outcomes. Regardless of the tillage and cover crops, the soils were dominated by macroaggregates, followed by microaggregates and clay-silt fraction. The results suggested that there was no difference between the CS and CV treatments in soil structure (described by MWD, bulk density, and aggregate distribution) and TC stocks in soils at 0–5 and 5–15 cm. The intrinsically low clay contents likely limit the extent to which the soil structure can be improved with CS, and hence the protection of SOC against decomposition. The introduction of winter cover crops for 4 years did not affect the measured variables. Conservation tillage promoted C accumulation in all soil aggregates at 0-5 cm, with the highest concentrations observed in the clay-silt fractions. However, after accounting for soil bulk density, CS did not result in higher TC stocks than CV at both soil depths with no cover crop impacts observed. The relatively low and short-term organic inputs of winter cover crops compared to long-term cash crop residues inputs likely confounded possible cover crop impacts. Novel management practices to increase organic inputs, while improving soil structure to preserve the increased inputs, are essentials to restore TC in the degraded Coastal Plains soils.

ORCID

Binaya Parajuli (D) http://orcid.org/0000-0002-1067-5947

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