

**TILLAGE AND ROTATION EFFECTS ON LABILE ORGANIC  
CARBON AND AGGREGATION IN A COTTON CROPPING SYSTEM**  
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**Abstract**

Evaluating the effects of management practices on soil organic matter storage has been a popular topic of study for some time. It is well documented that increases in soil organic matter improve soil physical properties, and increase the overall fertility and sustainability of the soil. Research in this area has recently been stimulated following the proposal that agricultural soils may provide a significant C sink that may aid in the mitigation of increasing atmospheric CO<sub>2</sub>. Observed differences in lint yield and N response from a cotton performance study at the Texas A&M University experimental farm near College Station, TX prompted us to examine the effects of tillage and rotation on soil organic C (SOC) storage, soil microbial biomass C (SMBC), mineralizable C, and water-stable aggregation in these cotton cropping systems. The three tillage/rotation treatments examined included conventionally tilled continuous cotton (CT), reduced tilled continuous cotton (RT), and conventionally tilled cotton after corn rotation (CC) treatments. SOC, SMBC, and C mineralization were significantly greater under RT and CC managed plots, when compared to CT plots. SOC, SMBC, and mineralizable C were 33, 58, and 79 % greater in the top 5 cm of RT managed plots and 29, 32, and 36 % greater in the top 5 cm of CC managed plots when compared to CT plots. Tillage and rotation had little effect on water-stable aggregation and aggregation did not correlate with SOC, SMBC, or C mineralization. C mineralization provided the most sensitive indicator of management-induced change, followed by SMBC, and SOC. Increases in SOC storage were relatively modest, suggesting that potential SOC storage under cotton in this region may be limited by warm climatic conditions. However, increases in labile organic matter fractions resulting from reduced tillage and rotation could have the potential to significantly impact soil fertility in cotton cropping systems.

**Introduction**

Managing soils to provide an environment conducive to organic matter storage has gained new impetus following the realization that agricultural soils have the potential to provide a significant C sink (Paustian et al., 1998). In addition to potentially mitigating atmospheric CO<sub>2</sub>, it has been well documented that increases in soil organic matter (SOM) also result in increased soil fertility, structural stability, water-holding capacity, and help maintain the long-term productivity of the soil (Lal et al., 1997).

Labile or active SOM fractions, for example, the soil microbial biomass (SMB) and mineralizable organic matter, play fundamental roles in regulating soil fertility (Smith and Paul, 1990; Collins et al., 1997). The SMB can act as a source or sink of nutrients, and also plays a critical role in nutrient transformations in the soil (Schnürer et al., 1985). The SMB pool constitutes approximately 2 to 5 % of SOC and 1 to 5 % of total soil N. These estimated pool sizes suggest that the SMB is large enough to significantly impact plant available soil N (Smith and Paul 1990). Bonde et al. (1988) found that SMBN was a significant component of potentially mineralizable N, and Follett and Schimel (1989) found that SMBN constituted 57 % and 71 % of the active soil nitrogen pools in native sod and tilled soil, respectively.

Soil organic matter is considered to be susceptible to oxidation by soil microorganisms if it is not protected from decomposition through (1) physical protection by aggregation, (2) association with silt and clay particles, or (3) through biochemical stabilization in the form of recalcitrant humic compounds (Six et al., 2002). Physical disruption by tillage generally reduces aggregation in soil (Beare et al., 1994a; Bruce et al., 1990; Carter, 1992), which renders SOM more susceptible to microbial oxidation. Physical protection of organic matter is thought to be enhanced in no-till (NT) or reduced-till (RT) vs. conventional-till (CT) systems due to increased protection by both macroaggregates (Beare et al., 1994b) and microaggregates (Six et al., 1999).

Numerous studies have documented increases in SOC storage in RT and NT vs. CT systems under various cropping schemes (Doran, 1980; Lamb et al., 1985; Havlin et al, 1990; Carter, 1992; Beare et al., 1994a). However, potential SOC storage as a result of RT in cotton-cropped soils is less documented. Potter et al. (1998) observed slight increases in SOC under NT and RT managed cotton vs. CT managed cotton. Salinas-Garcia et al. (1997) and Zibilske et al. (2002) observed 64 % and 58 % increases in SOC, respectively, in the surface soils (0-5 cm) of NT managed cotton/corn rotations as opposed to CT rotations. Zibilske et al. (2002) also observed low amounts of readily oxidizable carbon in both NT and CT treatments, suggesting that much of the SOC in the system was of a recalcitrant nature.

Data concerning the effects of tillage and rotation on labile organic carbon pools in cotton cropping systems are lacking. Entry et al. (1996) reported SMBC values of 20 mg kg<sup>-1</sup> under long-term continuous cotton with no N application in Alabama's

“Old Rotation” experiment. SMBC in a 2-year cotton/corn rotation with a winter legume in the same study was more than twice that of the continuous system ( $55 \text{ mg kg}^{-1}$ ). Balota et al. (2003) reported SMBC values of  $372 \text{ mg kg}^{-1}$  and  $145 \text{ mg kg}^{-1}$  under NT and CT cotton/wheat rotations, respectively, in southern Brazil. They concluded that NT systems resulted in higher SMB and suggested that nutrient cycling would be enhanced in NT systems due to the increased SMB pool. A better understanding of SMB and its activity under cotton systems could provide insight into both SOC dynamics and nutrient turnover in these systems.

An ongoing study of the effects of tillage and rotation on cotton performance at the Texas A&M University experimental farm near College Station has exhibited significant increases in lint yield under CT cotton after corn and RT continuous cotton treatments vs. CT continuous cotton. The CT cotton after corn and RT continuous cotton treatments have also shown decreased response to added N when compared to the CT continuous cotton treatment (Hons et al., 2004). These differences suggest that organic matter storage and dynamics have likely been affected by tillage and rotation in these systems. Such systems could provide valuable insight into SOC dynamics and microbial activity under varying cotton management. The objectives of this study were to (1) address the potential for SOC storage in cotton systems in south-central Texas, (2) examine the effects of tillage and rotation on soil structure in cotton-cropped soils in this region, and (3) assess tillage and rotation effects on two labile fractions of SOC, SMBC and mineralizable C, under cotton cropping.

## **Materials and Methods**

### **Site Characteristics, Treatments, and Sampling**

The experimental plots are located in the Brazos River floodplain in south-central Texas ( $30^{\circ}32'N$ ,  $96^{\circ}26'W$ ). The mean annual temperature and precipitation in the area are  $20^{\circ}C$  and  $978 \text{ mm}$ , respectively. The soil is a Weswood silty clay loam (fine, mixed, superactive, thermic, Fluventic Ustochrept) with an average of 35 % clay and 52 % silt. The soil is calcareous and has an average surface pH of 8.2.

The plots are arranged in an incomplete factorial within a randomized complete block design. The three management systems include conventionally tilled continuous cotton (CT), reduced tilled continuous cotton (RT), and a conventionally tilled cotton after corn rotation (CC). The tillage treatments have been in place for 10 years, and rotation with corn has been in place for 6 years. The type of reduced tillage being practiced is ridge-till, in which tillage is used to re-establish seed beds prior to planting. Ridge-till plots are also minimally tilled to help maintain furrows during the growing season. Various N rates are superimposed upon these management systems and each distinctive treatment (including N rate) contains four field replicates. Sets of replicate plots with N rates of 0 and  $90 \text{ kg N ha}^{-1}$  were sampled for this study.

Thirty-five soil cores (2.54 cm dia.) were taken from each replicate plot on March 24, 2003, prior to the planting of cotton, and separated into 0-5 cm and 5-15 cm depth sections. Additional 0-5 cm cores were taken to obtain equal amounts of soil for both depths. All soil samples were air-dried and gently crushed to pass through a 5-mm sieve in order to obtain initial homogeneity and to prepare samples for analysis of water-stable aggregation. Soils not examined for water-stable aggregates were then ground to pass a 2-mm sieve. All samples were stored at room temperature. Field replicates ( $n=4$ ) were used as laboratory replicates in the analysis of all parameters.

### **Water-Stable Aggregation**

Structural stability was measured as degree of water-stable aggregation. Water-stable aggregates were determined through a wet sieving procedure modified from Elliott (1986). 100-g subsamples from the 0-5 cm depth section were capillary wetted on Whatman #1 filter paper for 3 hrs and then washed through a succession of sieves. Material remaining on top of each sieve was then backwashed into pre-weighed containers, which were dried at  $55^{\circ}C$  to determine the weight of soil in each aggregate size class. Size classes collected included large macroaggregates ( $>2000\text{-}\mu\text{m}$  fraction), small macroaggregates ( $2000\text{-}\mu\text{m}$  to  $250\text{-}\mu\text{m}$ ), microaggregates ( $250\text{-}\mu\text{m}$  to  $53\text{-}\mu\text{m}$ ), and the silt-clay fraction ( $< 53\text{-}\mu\text{m}$ ). The amount of soil in the silt-clay fraction was determined by difference. Water-stable aggregates were not determined for soils from the 5-15 cm depth due to physical complications experienced when attempting to pass the soils through the 5-mm sieve, which would have biased the analysis.

### **Soil Organic Carbon**

Soil samples (40 g) were prepared for total SOC analysis by removing visible organic residues and grinding the soils to pass a 200-mesh sieve. SOC was determined by dry combustion on 200 mg subsamples using an Elementar multi-element analyzer. Combustion was performed at  $650^{\circ}C$  to allow SOC determination while preventing the combustion of inorganic carbonates.

### **Carbon Mineralization**

Carbon mineralization ( $\text{CO}_2$  evolution) was determined using a closed, static-chamber incubation method. Soil samples (100 g) were placed in 550-mL gas-tight jars and wetted to 25 % moisture. Alkali traps (10 mLs of 1 N NaOH) were placed inside the jars, which were sealed and incubated at  $25^{\circ}C$ . Alkali traps were removed and titrated with 1 N HCL to determine  $\text{CO}_2\text{-C}$  evolved (Anderson, 1982) after 3, 10, 24, and 38 days of incubation.

### **Soil Microbial Biomass Carbon**

Soil microbial biomass carbon (SMBC) was determined using the chloroform fumigation-incubation method without subtraction of a control (Voroney and Paul, 1984). Soil samples (35 g) were wetted to 25 % moisture, placed in gas-tight jars, and pre-incubated for 10 days at 25°C to allow for equilibration of the SMB (Franzleubbers et al., 1996). SMBC was quantified from the amount of CO<sub>2</sub>-C evolved over a ten-day period using an efficiency factor of 0.41.

### **Statistical Analysis**

Significant differences among treatments were evaluated using a one-way ANOVA and multiple comparisons analyses were performed with Fishers LSD. Kruskal-Wallis rank-based ANOVA and the Student-Newman-Keuls multiple comparison procedure were utilized for parameters with non-normal data. Linear regression was used to evaluate the correlation of parameters. All statistical analyses were performed using Sigma Stat, Version 3.0 (SPSS Inc., 2003).

## **Results and Discussion**

### **Water-Stable Aggregation**

Water-stable small macroaggregates dominated over other aggregate size fractions across all treatments, constituting roughly 50 % of the samples used in the analysis. The only significant difference in aggregation was observed in the CC plots receiving 90 kg N ha<sup>-1</sup>, which contained significantly more large macroaggregates and significantly fewer microaggregates than all other treatments (Fig. 1). We did not observe increased aggregation as a result of RT in our study. Researchers have documented that tillage reduces aggregation (Beare et al., 1994a; Bruce et al., 1990; Carter, 1992); however, we failed to see any significant differences in aggregation between CT and RT systems. The type of RT system used in our study was a ridge-till system that did incorporate tillage in the re-establishment of seed beds prior to planting, and in the maintenance of furrows throughout the growing season. These tillage steps likely played a role in the physical disruption of aggregates in the RT system, and it is possible that enhanced aggregation may have been observed within a no-till system.

The increase in large macroaggregates observed in the CC plots receiving N application was not observed in the CC plots not receiving N, and the CC plots receiving N did not exhibit significantly greater concentrations of SOC, SMBC, or mineralizable C than any of the other treatments. Therefore, we could not conclude that additional organic matter inputs due to corn rotation, or enhanced microbial activity under corn were likely responsible for enhancing macroaggregation in our study. Researchers have proposed that aggregation is largely a function of biological processes and soil organic matter content. Aggregation has been positively correlated with organic matter content (Chaney and Swift, 1984), microbial biomass (Haynes and Beare, 1997), root length (Oades and Waters, 1991; Jastrow et al., 1997), and length of fungal hyphae (Oades and Waters, 1991; Jastrow et al., 1997). In our study, no correlations were observed between aggregation within any size fraction and SOC, SMBC, or C mineralization. This was largely due to the lack of differences in aggregation that were observed.

Our data do not explain why the CC plots receiving N contained a significantly greater amount of large macroaggregates than the CC plots not receiving N. The addition of N to the CC treatment did not have any notable effects on any of the other parameters studied.

### **Soil Organic Carbon**

Reduced tillage and rotation with corn did result in increased SOC storage when compared to conventional tillage. SOC in the 0-5 cm depth of RT and CC plots was 33 % and 29 % higher than SOC in the 0-5 cm depth of CT plots (Table 1). In the 5-15 cm depth, these differences were 17 % and 23 %, respectively. SOC was significantly greater in RT plots when compared to CT plots, except in the 5-15 cm depth when 90 kg N ha<sup>-1</sup> were applied. SOC in CC plots was also not significantly greater than SOC in CT plots at the 5-15 cm depth when N was applied, or at the 0-5 cm depth when N was applied. These results suggest that N application may have increased SOC in CT relative to CC and RT systems. Previous data from this tillage/rotation experiment have shown that CT plots have exhibited a greater N response relative to RT and CC plots (Hons et al., 2004). However, N application had no significant effect on SOC storage when examined within any of the tillage/rotation treatments.

Depth also had no significant effect on SOC storage within any of the tillage/rotation treatments. The RT plots did show the greatest accumulation of SOC in the surface, containing 16 % more SOC in the 0-5 vs. the 5-15 cm depth, but this difference was not statistically significant. Many studies reporting increased SOC under NT or RT systems have observed stratification of SOC in the soil surface as a result of the surface accumulation of crop residues in these systems (Havlin et al., 1990; Salinas-Garcia et al., 1997; Zibilske et al., 2002) We failed to observe significant stratification of SOC under RT, suggesting that ridge-tillage practices may have redistributed crop residues within the top 0-15 cm of the soil and reduced organic matter stratification.

Increases in SOC storage under RT and CC vs. CT systems were relatively modest. However, these moderate increases were consistent with what has been seen in warm climates. Organic matter storage is generally lower in warmer climates (Jenny, 1941), which provide an environment suitable for enhanced microbial activity and organic matter decomposition. Potter et

al. (1998) assessed the potential for increasing SOC in Texas soils by utilizing data from long-term management studies, and came to the conclusion that C sequestration was possible, but limited by warm climatic conditions. Zibilske et al. (2002) reported that warm climate likely affected increases in SOC storage under NT and RT cotton/corn rotations. The authors observed 44 % greater SOC in the top 4 cm of ridge-till vs. CT rotations. Our study yielded comparable results, with RT (ridge-till) containing 33 % more SOC than CT in the 0-5 cm depth under continuous cotton.

### **Soil Microbial Biomass Carbon**

SMBC as a percentage of SOC ranged from approximately 3 to 5 % as examined across all treatments, including N application and depth. SMBC in the 0-5 cm depth was 58 % and 32 % greater under RT and CC management when compared to CT management (Table 2). RT and CC managed plots contained significantly more SMBC than CT plots at both 0-5 cm and 5-15 cm depths. This result suggests that altered residue distribution by RT management and additional organic matter inputs from corn in CC management provided the soil microflora with more substrate as compared to CT management. SMBC was positively correlated with SOC and 38-day cumulative C mineralization (Figure 2), also supporting the observation that increased available organic substrate under RT and CC managed systems supported greater SMB when compared to CT managed systems.

Depth or N application had no significant impacts on SMBC within any of the tillage/rotation treatments with the exception of RT when N was applied, which contained significantly more SMBC than RT with no N application. It is possible that the addition of fertilizer N may have lowered C:N ratios in the surface of RT soil, which may have led to increased decomposition of organic residues and subsequent increases in the SMB in these plots. However, the magnitude of this observed difference is partly a result of the data distribution of the RT plots receiving N, which were skewed by one of the field replicates. Therefore, we could not make any conclusions regarding the effect of N application in this situation.

Stratification of the SMB was observed in RT managed plots, which contained 31 % more SMBC in the 0-5 cm depth vs. the 5-15 cm depth. Researchers have observed increased microbial abundance in the surface of RT and NT managed soils (Doran, 1980; Carter and Rennie, 1982; Carter, 1992). The observed stratification of SMB in our RT plots was relatively modest, but it was greater than the stratification of SOC in these plots. Similar results were observed by Carter and Rennie (1982). Management induced changes in SOC can be hard to measure against background SOC, and SMB has proven to be a more sensitive indicator of management-related change (Powlson and Brookes, 1987). SMBC was a better indicator of change when compared to SOC concerning stratification under RT management in our study. SMBC was also a more sensitive indicator of change than SOC with respect to the increases in these parameters in both RT and CC vs. CT managed plots.

Microbial biomass as a percentage of SOC ranged from 3 to 5 % in our study (data not shown). Smith and Paul (1990) reported that SMBC constitutes approximately 2 to 5 % of SOC. Though organic matter stocks are generally lower in areas experiencing warmer climate (Jenny, 1941), it has been reported that active organic matter fractions, such as the microbial biomass and mineralizable C, can constitute relatively larger proportions of SOM in warmer regions (Franzluebbers et al., 2001). SOC storage in these cotton cropping systems may be limited by warm climatic conditions, but increases in SMBC are apparently not limited. The SMBN pool has the capacity to significantly impact plant available N (Smith and Paul, 1990; Follett and Schimel, 1989); thus, increases in the SMB resulting from reduced tillage and rotation could lead to increases in available N and potentially improve cotton yield.

### **Carbon Mineralization**

Thirty-eight day cumulative C mineralization (38-day CMIN) provided the most sensitive indicator of change resulting from tillage and rotation when compared to SOC and SMBC. In the 0-5 cm depth, 38-day CMIN in RT and CC managed soils was 79 % and 36 % greater than 38-day CMIN in CT managed soils (Table 3). In the 5-15 cm depth these differences were 20 % and 29 % respectively. 38-day CMIN in RT and CC managed plots was significantly greater than 38-day CMIN in CT managed plots at both depths suggesting that RT and CC management increased the amount of mineralizable organic matter in top 15 cm of soil.

The effects of depth in our study were also best represented by 38-day CMIN. In RT managed soils, 38-day CMIN was significantly greater (69 %) in the 0-5 cm depth vs. the 5-15 cm depth. This suggests that there is some stratification of organic matter occurring in RT managed plots. This stratification was insignificant as represented by SOC measurements, thus 38-day CMIN was more sensitive in assessing the effects of stratification by RT. The stratification of mineralizable organic matter in these soils is likely due to the distribution of more crop residues near the surface of RT soils in these systems.

With the exception of soils in the 5-15 cm depth under CT management, the application of N had no significant effect on 38-day CMIN as examined within any of the tillage/rotation treatments. Nitrogen application resulted in significantly greater 38-day CMIN at this depth in CT managed soils. This may possibly be a result of the higher N response exhibited by CT managed cotton. If a greater yield response to N application resulted in increased crop residues, one would expect that the incorporation of these residues by tillage would lead to greater C mineralization in these soils. This N effect was not as pronounced in the 0-5 cm depth of CT managed soils.

The enhanced organic matter mineralization observed under RT and CC managed plots suggests that the mineralization of organic N into plant available forms may also be enhanced in these systems. Nitrogen transformations in the soil are largely dependent upon C transformations (McGill et al., 1975), thus, enhanced C mineralization may be linked to enhanced N mineralization. This is dependent upon mineralization and immobilization processes which vary with changes in the quality and quantity of the SOM and SMBC (Jansson and Pearson, 1982). Franzleubbers et al. (1996) observed strong relationships between C mineralization and N mineralization in soils from several geographic regions, and short-term C mineralization has been reported as a good predictor of 24-day potential N mineralization (Haney et al., 2001). The increases in mineralizable C and SMBC that were observed in RT and CC managed plots vs. CT managed plots suggest that plant available N may be positively affected by these types of management. These differences in labile organic matter pools coincide with the yield differences observed on these plots by (Hons et al., 2004). Future work will involve assessment of the effect of labile organic matter pools on potentially mineralizable N in these systems.

### Conclusions

Reduced tillage and rotation with corn significantly affected SOC storage, SMBC, and C mineralization in the cotton systems studied. RT and CC managed plots contained greater SOC, SMBC, and mineralizable C than CT managed plots, suggesting that distribution of crop residues by RT management, and additional organic matter inputs from corn in CC management affected these systems. Differences between tillage/rotation treatments were the greatest for mineralizable C, and were the least for SOC. In general, C mineralization and SMBC were better indicators of management-induced change than SOC in these cotton cropping systems. Water-stable aggregation did not correlate well with SOC, SMBC, or C mineralization, and there were few differences in water-stable aggregation between tillage/rotation treatments. Improved SOC storage in cotton cropping systems in Texas may be limited by a warm climate, but significant increases in labile organic matter pools are attainable through utilization of reduced tillage and rotation. These labile organic matter pools have the ability to supply plant available N; thus, they have the potential to improve fertility, and possibly reduce fertilizer N requirements in cotton systems.

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Table 1. Organic carbon (%) in soils from cotton-cropping systems at the Texas A&M University experimental farm\*.

Treatment	%SOC		
	CT	RT	CC
<b>0-5 cm depth</b>			
0 kg N ha <sup>-1</sup>	0.82 b,A	1.09 a,A	1.14 a,A
90 kg N ha <sup>-1</sup>	0.85 b,A	1.14 a,A	1.01 ab,A
<b>5-15 cm depth</b>			
0 kg N ha <sup>-1</sup>	0.80 b,A	0.95 a,A	1.05 a,A
90 kg N ha <sup>-1</sup>	0.85 a,A	0.98 a,A	0.99 a,A

\* Values followed by the same lowercase letter w/ in rows are not significantly different; Fishers LSD (P<0.05). Values followed by the same uppercase letter within in columns are not significantly different; Fishers LSD (P<0.05).

Table 2. Soil microbial biomass carbon in soils from cotton-cropping systems at the Texas A&M University experimental farm\*.

Treatment	SMBC mg C kg <sup>-1</sup> soil		
	CT	RT	CC <sup>†</sup>
<b>0-5 cm depth</b>			
0 kg N ha <sup>-1</sup>	310 b,A	454 a,B	412 a,A
90 kg N ha <sup>-1</sup>	325 c,A	553 a,A	428 b,A
<b>5-15 cm depth</b>			
0 kg N ha <sup>-1</sup>	272 b,A	359 a,B	360 a,A
90 kg N ha <sup>-1</sup>	300 b,A	408 a,B	367 a,A

\* Values followed by the same lowercase letter w/ in rows are not significantly different; Fishers LSD (P<0.05). Values followed by the same uppercase letter w/ in columns are not significantly different; Fishers LSD (P<0.05).

<sup>†</sup> values within columns followed by the same uppercase letter are not significantly different; Kruskal-Wallis rank-based ANOVA and Student-Newman-Keuls multiple comparison procedure.

Table 3. Cumulative carbon mineralization (38-day) in soils from cotton-cropping systems at the Texas A&M University experimental farm\*.

Treatment	C Mineralization mg C kg <sup>-1</sup> soil		
	CT	RT <sup>†</sup>	CC <sup>†</sup>
<b>0-5 cm depth</b>			
0 kg N ha <sup>-1</sup>	189 c,AB	337 a,A	264 b,A
90 kg N ha <sup>-1</sup> †	204 b,A	366 a,A	272 a,A
<b>5-15 cm depth</b>			
0 kg N ha <sup>-1</sup>	162 b,C	203 a,B	227 a,A
90 kg N ha <sup>-1</sup>	184 b,B	213 a,B	219 a,A

\* Values followed by the same lowercase letter within rows are not significantly different; Fishers LSD (P<0.05). Values followed by the same uppercase letter within columns are not significantly different; Fishers LSD (P<0.05).

†- values within columns followed by the same uppercase letters or values in rows followed by the same lowercase letters are not significantly different; Kruskal-Wallis rank-based ANOVA and Student-Newman-Keuls multiple comparison procedure.

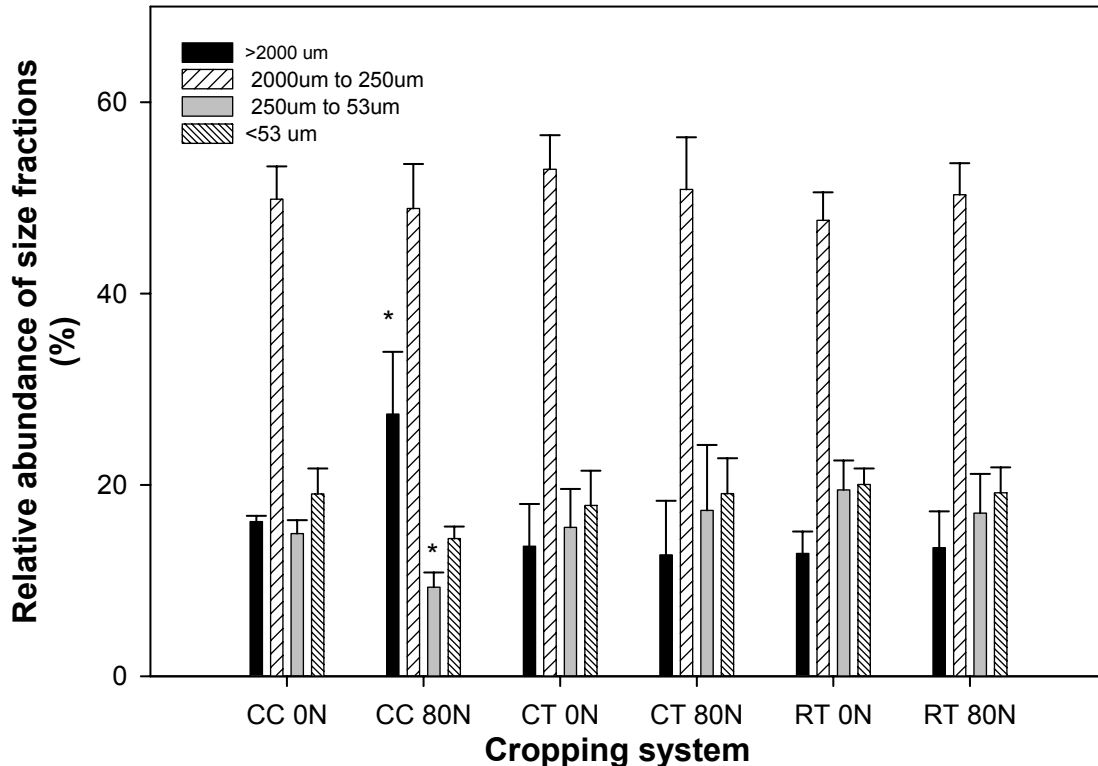


Figure 1. Water-stable aggregation in soils from cotton-cropping systems at the Texas A&M University experimental farm. Asterisks indicate significant differences (P<0.05) within aggregate size class across treatments; Kruskal-Wallis rank-based ANOVA and Student-Newman-Keuls multiple comparison procedure.



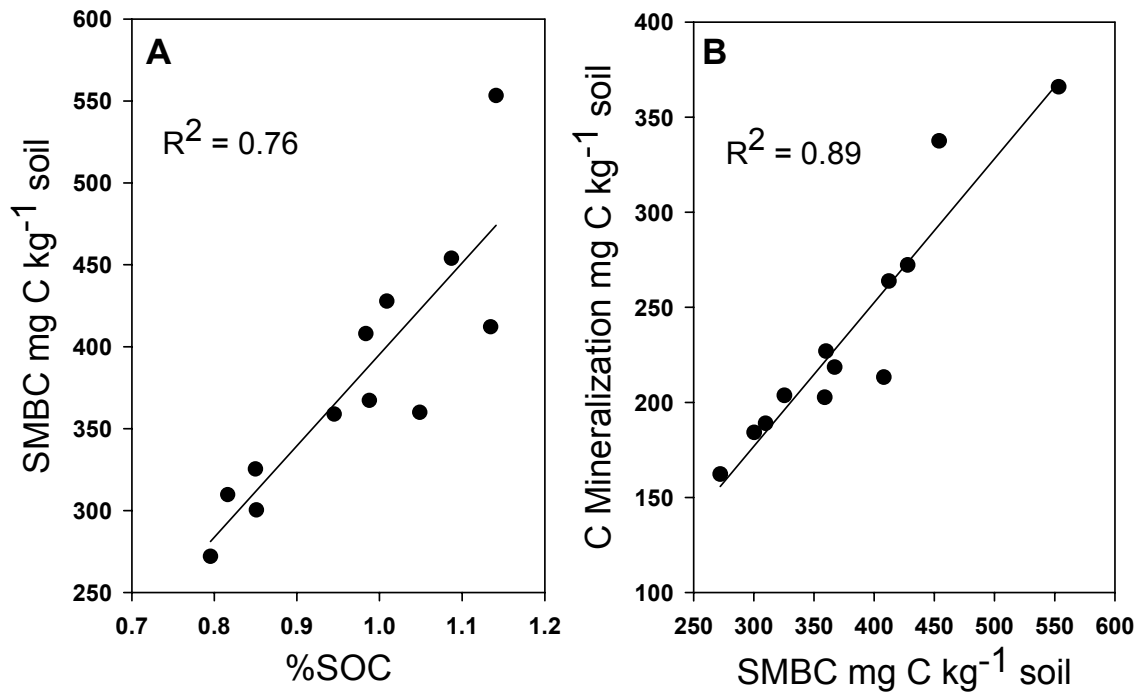


Figure 2. Relationship between soil microbial biomass C and soil organic carbon (A) and between carbon mineralization and soil microbial biomass (B).