

COMPARING WATER-STABLE AGGREGATE DISTRIBUTIONS, ORGANIC MATTER
FRACTIONS AND CARBON TURNOVER USING ^{13}C NATURAL ABUNDANCE IN
CONVENTIONAL AND NO TILLAGE SOILS

by

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(Under the Direction of David C. Coleman, Ph.D.)

ABSTRACT

Water-stable aggregates (WSA) physically protect soil organic matter (SOM) from decomposition, resulting in SOM pools with longer turnover times. Intensive cultivation disrupts soil aggregates, whereas conservation management practices increase aggregate stability and carbon concentration in the soil. Soil samples were taken from No-tillage (NT) and Conventional-tillage (CT) plots at Horseshoe Bend field site in Athens, Georgia. Soil cores were divided in depths and wet sieved into aggregates >2000 , $250\text{--}2000$, $53\text{--}250$, and <53 μm . I determined aggregate distribution, total carbon (C) and nitrogen (N), mineral and particulate organic matter (POM) associated C and N, and used ^{13}C natural abundance to trace recent organic inputs and C turnover in the soil. These results support the conceptual model of aggregate turnover; the net input rate increased with aggregate size fractions, indicating a slower turnover for microaggregates than macroaggregates and suggesting physical protection of old C in the smaller aggregate fraction.

INDEX WORDS: Water-stable aggregates, No tillage, Conventional tillage, Long-term study, Soil Organic Matter dynamic, ^{13}C natural abundance, Wet sieving, C turnover

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CHAPTER 1

INTRODUCTION

Maintenance of Soil Organic Matter (SOM) is desirable in agroecosystems because of its beneficial effects on nutrient dynamics and soil structure (Bossuyt et al., 2002). Soil organic matter improves soil structural stability, soil fertility and crop production, and ensures long-term sustainability of agricultural ecosystems (Tisdale et al., 1993; Stevenson, 1994; Deneff et al., 2004). Furthermore, SOM has gained much attention due in part to its potential key role in the global C cycle by acting as a sink for atmospheric CO₂ when appropriate management practices are used (Bruce et al., 1998; Paustian et al., 1997b; Paustian et al., 2000). Soil is estimated to be the largest terrestrial pool of C, containing 1500 Pg or twice as much as the atmosphere (Sundquist, 1993; Schlesinger, 1997).

Farming systems that utilize appropriate management practices hold promise for sequestering soil C, which has the potential to enhance agricultural sustainability, reduce negative environmental impacts, and attenuate anthropogenic carbon dioxide emissions (Kong et al., 2005). This study was conducted in the Horseshoe Bend Agroecosystem site, Athens, Georgia, and was designed as part of a long term study to help explain the differences in organic matter dynamics after years of No tillage and Conventional tillage regimes in its plots. Our objectives were (i) to determine the influence of both tillage methods on water-stable aggregate distribution and SOM dynamics and (ii) to investigate the change of ¹³C in both NT and CT agricultural practices. We hypothesized that the contrast between tillage treatments was going to change in

time, having (i) greater macroaggregation and organic matter fractions in NT compared to CT (ii) and faster C turnover in both macro- and microaggregates for CT compared to NT.

Aggregates and Soil Organic Matter

The relationship between aggregates and associated SOM dynamics has been explained in several studies (Elliot, 1986; Jastrow, 1996; Six et al., 1998). Tisdall and Oades (1982) developed a conceptual model for aggregate hierarchy in which free primary particles and silt-sized aggregates (<20 μm) are bound together into microaggregates (20–250 μm) by persistent binding agents (i.e. humified organic matter), oxides and aluminosilicates. These stable microaggregates, are bound together into macroaggregates (>250 μm) by temporary (i.e. fungal hyphae and roots) and transient (i.e., microbial- and plant-derived polysaccharides) binding agents (Six et al., 2004) According to Six et al. (2004), the aggregate hierarchy concept proposed by Tisdall and Oades (1982) is probably the most significant theoretical advancement in the understanding of aggregate–SOM interactions. This conceptual model has been employed to explain the positive correlation between aggregation and SOM with cultivation. (Elliott, 1986; Cambardella and Elliott, 1993; Beare et al., 1994a) and C sequestration under No tillage and Conventional tillage practices (Beare et al., 1994a; 1994b; Six et al., 1998). A breakdown of macroaggregates results in release of labile SOM, making it therefore available to microbial decomposition. According to Jastrow (1996) and Six et al. (1998), higher microbial activity depletes SOM, which eventually leads to decreased microbial biomass and activity and consequently a lower production of microbial-derived binding agents and a loss of aggregation.

Oades (1984) formulated an important modification to the concept of the hierarchical aggregates, stating that the roots and hyphae holding together the macroaggregate form the

nucleus for microaggregate, being the latter formed within a macroaggregate. His new model came up to be really important for the understanding of SOM and aggregate dynamics. Elliott (1986) then suggested that macroaggregates have elevated C concentrations because of the organic matter binding microaggregates into macroaggregates and that this organic matter is labile and less highly processed than the organics stabilizing microaggregates. Further, since cultivation generally results in reduced stability and amount of macroaggregates but does not affect microaggregate stability (Tisdall and Oades, 1982), Elliott (1986) proposed that intermicroaggregate binding agents are the primary source of organic matter lost when native sod soils are cultivated. With this concept, it was identified for the first time the direct link between agricultural disturbance, decrease of aggregation and loss of labile SOM (Six et al., 2004). After this, Cambardella and Elliott (1992; 1993; 1994) suggested that most of the labile pool Elliott (1986) referred to within macroaggregates is either light-fraction particulate organic matter (POM) or relatively low-density, mineral-associated organic matter.

Elliott and Coleman (1988) hypothesized that different types of macroaggregate binding mechanisms will appear to be important depending on the process being observed. They pointed out that intermicroaggregate organic matter seems most important when native grassland soils are cultivated since it is more exposed, disintegrating macroaggregates into microaggregates at a faster rate. In contrast, when long-term cultivation soils are converted to reduced-tillage systems, roots and hyphae appear important because they probably serve as initial binding agents. They also suggest that with time, accumulation of POM and deposition of microbial by-products between microaggregates become important mechanisms in further stabilizing macroaggregates and result in the accrual of soil organic matter (Jastrow, 1996).

Golchin et al. (1994) proposed a model for the formation of microaggregates, saying that when fresh plant material enters the soil, it induces the formation of aggregates because it stimulates the production of microbial-derived binding agents. During decomposition, plant material fragments become encrusted with clay particles and microbial products to form the core of stable microaggregates (combining microbial metabolites with the mineral crust). In time, these cores are depleted of available SOM and with the decrease of microbial activity microaggregates become unstable. After their breakdown, the mineral crusts impregnated with microbial by-products are released to form stable silt-sized organo-mineral complexes. (Six et al., 2004)

Angers et al. (1997) corroborated the concept of microaggregate formation within macroaggregates. They traced C and N in macro- and micro-aggregates during decomposition of wheat straw and observed ^{13}C was accumulated in the macroaggregates during the initial decomposition period and decreased after that.. At the same time, a stabilization of ^{13}C was observed in the microaggregates at the same time as the decrease in ^{13}C enrichment of the macroaggregates was occurring,. This redistribution of C from macroaggregates to microaggregates with time showed that microaggregates are formed within macroaggregates.

According to Six et al. (2004), since the study of Angers et al. (1997), other studies have proposed variants of conceptual models of the role of SOM and POM play in the formation of microaggregates within macroaggregates (Jastrow and Miller, 1998; Six et al., 1998; Puget et al., 2000), however, these variants are small and do no change the main idea of this conceptual model.

Tillage practices and Soil Organic Matter

Tillage practices disrupt soil aggregates and this may lead to an enhanced aggregate turnover and increased decomposition of SOM (Six et al., 1998). Paustian et al. (1997a) found in most cases there was an increase in C content under (NT) compared to CT systems after several long-term field studies. They attributed this increase to a combination of reduced litter decomposition and less soil disturbance under NT.

Paustian et al. (1999) stated that reduced aggregation leads to subsequent lower levels of SOM in CT than in NT. This reduction is a result of indirect effects on aggregation. In CT, subsurface soil is brought to the surface and exposed to wet-dry, freeze-thaw cycles and raindrop impact (Beare et al., 1994; Paustian et al., 1997). Also, incorporating plant residues in soil can affect the soil microclimate and increase plant residue contact with soil. This will increase residue decomposition and organic matter transformation (Beare et al., 1992; Cambardella and Elliott, 1993). Tillage management changes the soil conditions (aeration, temperature, and moisture) and increases the decomposition rates of litter (Cambardella and Elliott, 1993). In NT, there is residue accumulation at the surface where the litter decomposition rate is slowed due to drier conditions and reduced contact between soil microorganisms and litter (Salinas-Garcia et al., 1997), and there is a greater proportion of microbial biomass composed of fungi, which contribute to macroaggregate formation and stabilization (Tisdall and Oades, 1982). Six et al. (1999) developed a Conceptual model of aggregate turnover, which shows organic C accumulation and mineralization under no-tillage (NT) and conventional tillage (CT) systems. Briefly, following the incorporation of fresh crop-derived residue (free LF), soil fungi and other microorganisms utilize the more easily available C and produce mucilages, forming macroaggregates around coarse intra-aggregate POM (coarse iPOM) (Six et al. 1999). Coarse

iPOM is further decomposed and fragmented into fine iPOM. The fine iPOM and associated mucilages become encrusted with minerals to form the stabilized organic core of a microaggregate within a macroaggregate. In time, C is depleted and microbial activity and production of binding agents decrease, leading to a destabilization and potential disaggregation of the macroaggregates. Upon disaggregation there is a release of microaggregates (old and new), the mineral fraction, and POM. In CT, some macroaggregates go through the same sequence (the decomposition loop). However, a greater proportion of macroaggregates in CT are disrupted by cultivation and/or slaking in the field and go through a shorter cycle (the cultivation loop) resulting in a faster macroaggregate turnover rate (Six et al., 1999). This turnover inhibits the formation of microaggregates within macroaggregates and the long-term sequestration of C within microaggregates. The faster macroaggregate turnover in CT than in NT results in (i) fewer macroaggregates being maintained but more free microaggregates being present in CT and (ii) less fine iPOM and new microaggregates formed in CT. In the end of the cycle, fewer microaggregates have crop-derived C incorporated in CT than in NT. (Six et al. 1999).

Carbon 13 ^{13}C and Soil Organic Matter

Plant residues incorporated into soils are subjected to contrasted stabilization and biodegradation processes and may contribute SOM pools with different turnover times (Dignac et al., 2005). During photosynthesis, plants discriminate against ^{13}C for the lighter isotope ^{12}C giving plants a lower $^{13}\text{C}/^{12}\text{C}$ ratio than the surrounding atmosphere. For this reason, when a vegetation cover is converted from C_4 plants to C_3 plants (or vice versa), it is possible to track the isotope ^{13}C to trace the turnover of recent organic matter inputs (Cerri et al. 1985; Balesdent et al. 1987). Different plants discriminate to different extents. C_3 plants discriminate more than

C₄ plants, and therefore the two plants have different natural enrichments of ¹³C. The ¹³C/¹²C ratios are usually reported as approximately –28.00 ‰ for C₃ and –12.00 ‰ for C₄ plants (Bender 1968). The negative values indicate a ¹³C deficit compared to the surrounding atmosphere. This difference is maintained during decomposition, so SOM originating from C₃ plants can be distinguished from C₄ plants (Balesdent et al. 1987).

CHAPTER 2

**COMPARING WATER-STABLE AGGREGATE DISTRIBUTIONS, ORGANIC
MATTER FRACTIONS AND CARBON TURNOVER USING ¹³C NATURAL
ABUNDANCE IN CONVENTIONAL AND NO TILLAGE SOILS¹**

¹ Arce Flores, S. and D.C. Coleman. To be submitted to *Soil Biology and Biochemistry*.

Abstract

Water-stable aggregates (WSA) physically protect soil organic matter (SOM) from decomposition, resulting in SOM pools with longer turnover times. Intensive cultivation (i.e. conventional tillage) is known to disrupt soil aggregates, whereas conservation management practices (i.e. no tillage) increase aggregate stability and carbon concentration in the soil. Soil samples were taken from No-tillage (NT) and Conventional-tillage (CT) plots at Horseshoe Bend field site in Athens, Georgia, United States. All soil samples were divided in two depths (0-5 and 5-15 cm) and divided through wet sieving in four aggregate size classes: large macroaggregates (>2000 μm), small macroaggregates (250–2000 μm), microaggregates (53–250 μm), and the silt + clay fraction (<53 μm). For size classes in both depths, we determined aggregate distribution, total carbon (C) and nitrogen (N), mineral and particulate organic matter (POM) associated C and N, and we used ^{13}C natural abundance to trace recent organic inputs and C turnover in the soil. Prior to 1998 the crops grown on these plots were a mix of C_3 and C_4 plants, since 1998 only C_3 crops have been planted. The distribution of WSA from surface (0-5 cm) soils was significantly ($P < 0.001$) influenced by tillage practice in all aggregate size classes. Macroaggregates (> 2000 μm) made up the largest percentage of the whole soil, and were nearly 1.6 times greater in NT than in CT. 250-2000, 53-250 and < 53 μm of WSA were 1.3, 2.9 and 2.2 times ($P < 0.05$) greater in CT than in NT, respectively. For all aggregate size classes in 0-5 cm soils, total sand free C and N were 1.7 times higher ($P < 0.01$) in NT than in CT. The reduction in C and N with depth was greater in NT than in CT. Mineral associated C and N were 1.5 and 1.7 times higher ($P < 0.05$) in NT than CT, respectively, for 0-5 cm soils. POM associated C and N were significantly higher in NT than CT for aggregates >250 μm in soil surface and lower in 5-15 cm. The turnover rate of aggregates in NT were slower than CT for

both depths, and within tillage net input rate was increasing with an increase in aggregates size fraction, which indicated a slower turnover for microaggregates than macroaggregates. This suggested physical protection of old C in the smaller aggregate fraction.

Keywords: Water-stable aggregates, No tillage, Conventional tillage, Long-term study, Soil Organic Matter dynamic, ^{13}C natural abundance, Wet sieving, C turnover.

Introduction

Maintenance of soil organic matter (SOM) is desirable in agroecosystems, because of its beneficial effects on nutrient dynamics and soil structure (Bossuyt et al., 2002). SOM improves soil structural stability, soil fertility and crop production, and it ensures long-term sustainability of agricultural ecosystems (Tisdale et al., 1993; Stevenson, 1994; Deneff et al., 2004). Furthermore, SOM has gained much attention due to its potentially key role in the global C cycle as a sink for atmospheric CO_2 when appropriate management practices are used (Bruce et al., 1998; Paustian et al., 1997b; Paustian et al., 2000). Soil is estimated to be the largest terrestrial pool of C, containing 1500 Pg or twice as much as the atmosphere (Sundquist, 1993; Schlesinger, 1997).

Farming systems that utilize appropriate management practices hold promise for sequestering soil C, which can enhance agricultural sustainability, reduce negative environmental impacts, and attenuate anthropogenic carbon dioxide emissions (Kong et al., 2005). Tisdall and Oades (1982) developed a conceptual model for aggregate hierarchy in which free primary particles and silt-sized aggregates ($< 20 \mu\text{m}$) are bound together into microaggregates ($20 - 250 \mu\text{m}$) by persistent binding agents (i.e. humified organic matter), oxides and aluminosilicates. These stable microaggregates are bound together into macroaggregates ($> 250 \mu\text{m}$) by temporary

(i.e. fungal hyphae and roots) and transient (i.e., microbial- and plant-derived polysaccharides) binding agents (Six et al., 2004). This conceptual model has been employed to explain the positive correlation between aggregation and SOM with different cultivation managements (Elliott, 1986; Cambardella and Elliott, 1993; Beare et al., 1994a) and C sequestration under No tillage (NT) and Conventional tillage (CT) practices (Beare et al., 1994a; 1994b; Six et al., 1998). Oades (1984) formulated an important modification to the concept of the hierarchical aggregates, stating that the roots and hyphae holding together the macroaggregate form the nucleus for microaggregate, with the latter being formed within a macroaggregate. This provided a significant advancement for understanding aggregate–SOM interactions. Since then, the relationship between aggregates and associated SOM dynamics has been explained in several studies (Elliott, 1986; Jastrow, 1996; Six et al., 1998).

Tillage practices disrupt soil aggregates and this may lead to an enhanced aggregate turnover and increased decomposition of SOM (Six et al., 1998). Paustian et al. (1999) noted that reduced aggregation leads to subsequent lower levels of SOM in CT than in NT. This reduction is a result of indirect effects on aggregation. In most cases there was an increase in C content under NT compared to CT systems in several long-term field studies (Paustian et al., 1997a). They attributed this increase to a combination of reduced litter decomposition and less soil disturbance under NT.

Six et al. (1999) developed a conceptual model of aggregate turnover, which shows organic C accumulation, mineral fraction, and POM. The faster macroaggregate turnover in CT than in NT results in (i) fewer macroaggregates being maintained but more free microaggregates being present in CT and (ii) less fine POM and new microaggregates formed in CT. In the end of

the cycle, fewer microaggregates have crop-derived C incorporated in CT than in NT (Six et al. 1999).

Furthermore, recent studies have demonstrated the usefulness of ^{13}C natural abundance for estimating long-term C turnover and dynamics in soils. This tracer allows the study of C produced *in situ*, and it can be used over time scales varying from 1 year to hundreds or thousands of years (Balesdent and Mariotti, 1996). Plant residues incorporated into soils are subjected to contrasted stabilization and biodegradation processes and may contribute SOM pools with different turnover times (Dignac et al., 2005). During photosynthesis, plants discriminate against ^{13}C for the lighter isotope ^{12}C , giving plants a lower $^{13}\text{C}/^{12}\text{C}$ ratio than the surrounding atmosphere. For this reason, when a vegetation cover is converted from C_4 plants to C_3 plants (or vice versa), it is possible to track the isotope ^{13}C to trace the turnover of recent organic matter inputs (Cerri et al. 1985; Balesdent et al. 1987). C_3 plants discriminate against ^{13}C more than C_4 plants, and therefore two plants have different natural enrichments of ^{13}C . The $^{13}\text{C}/^{12}\text{C}$ ratios are usually reported as approximately -28.00‰ for C_3 and -12.00‰ for C_4 plants (Bender 1968). The negative values indicate a ^{13}C deficit compared to the surrounding atmosphere. This difference is maintained during decomposition, so SOM originating from C_3 plants can be distinguished from C_4 plants (Balesdent et al. 1987). The introduction of a new vegetation with a different photosynthetic pathway allows the quantification of both the loss rate of the original organic matter and the net input rate from the new source (Jastrow, 1996).

This study was conducted in the Horseshoe Bend Agroecosystem site, Athens, Georgia, and was designed as part of a long term study to help explain the differences in organic matter dynamics after years of No tillage and Conventional tillage regimes in its plots. Our objectives were (i) to determine the influence of both tillage methods on water-stable aggregate distribution

and SOM dynamics and (ii) to investigate the change of ^{13}C in both NT and CT agricultural practices. We hypothesized that the contrast between tillage treatments was going to change in time, having (i) greater macroaggregation and organic matter fractions in NT compared to CT (ii) and faster C turnover in both macro- and microaggregates for CT compared to NT.

Materials and Methods

Study Area

The study was conducted in the long-term Conventional Tillage (CT) and No Tillage (NT) plots at the Horseshoe Bend experimental area near Athens, Georgia. This experimental site was established in 1978 on a bottomland terrace within a meandering loop of the North Oconee River (Hendrix, 1997). The site was divided into eight plots, 0.1 ha in size and assigned to either NT or CT management in a completely randomized design with four replicates per treatment. These tillage regimes have been maintained since 1978 to the present, with the cropping system consisting of a summer grain crop followed by a winter cover crop. The CT plots were moldboard plowed, disked, and rotary tilled (0-15cm) twice annually to incorporate crop residues and prepare the seed bed. Summer crops were direct drilled, and winter crops were broadcast seeded. The soil is a fine loamy siliceous thermic Rhodic Kanhapludult in the Hiwassee series with 66% sand, 13% silt, and 21% clay, and pH ~5.8. Mean annual precipitation and temperature are 1270 mm and 16.5 °C, respectively. Prior to 1998, the crops grown on these plots were a mix of C_3 and C_4 plants. In most years the plots were double cropped with a rye/clover split during winters and corn as summer crops. Since 1998, only C_3 crops have been planted. The winter crop has remained a rye/clover split, but Kenaf was the summer crop in 1998

and cotton was planted from 1999 onward. Further details about the Horseshoe Bend site can be found in Hendrix (1997).

Soil Samples

Soil samples were collected from the CT and NT plots on May, 1997 and on May, 2000 with a thin-walled steel cylindrical corer fitted with a removable plastic insert (5.8 cm diameter). Three core samples were taken randomly from each plot. The intact cores were sectioned into 0-5 and 5-13 cm increments and composite within depths (Kimber Collins 2001, unpublished data).

On May 2005, eight cores (5.8 cm diam.) were collected from each of the four replicate plots in each tillage treatment and composite by depth increment (0-5 and 5-15 cm). Soils were stored field moist (3 °C,) in crush-resistant air-tight containers prior to analysis. For all of the three different dates 1997, 2000 and 2005 soils were passed through a 10-mm sieve to remove crop litter and stones. This was done by gently breaking apart the larger soil peds along points with least resistance until the peds were able to fit through the 10-mm openings. The sieved soils were air-dried and stored at room temperature.

Aggregate Distribution

The distribution of aggregates was measured using a modified Yoder (1936) wet-sieving apparatus that was designed to handle larger masses of soil on stacked sieves (21.6-cm diameter) and allow for complete recovery of all particles from individual samples (Beare and Bruce, 1993). The initial moisture content of the soil prior to wet sieving can alter the aggregate size distribution and partitioning of the organic matter associated with the aggregate size fractions

(Kemper and Rosenau, 1986). Soil was air dried prior to sieving and suspended in distilled water at room temperature on the largest sieve 5 minutes before sieving. Each 50 g subsample was distributed on top of the sieve of the 2 sieve-stack (with 2000 μm on top of 250 μm). The soil was wet sieved oscillating 31 times/min for 5 minutes, through a series of three sieves to obtain four aggregate size fractions (i) 2000 μm (large macroaggregates), (ii) 250 to 2000 μm , (iii) 53 to 250 μm (microaggregates), and (iv) < 53 μm (silt- plus clay size particles). Following the sieving, the sieves were lowered to the bottom of the stroke and the fresh organic matter from crop residue was aspirated from the surface before draining and put into an aluminum cake pan. After wet sieving, the water columns were drained and the soil sieves were backwashed into aluminum cake pans and left to dry in a dehumidifying chamber (10 °C). Subsamples of soil were taken to convert sample weight to oven-dry weight. The 53 μm pan was removed from the bucket and a stirrer machine was used to mix the water in the bucket. Once it was well mixed, a subsample of 200 mL was taken and placed into its respective tin pan. The total volume of water and soil particles < 53 μm was measured as well. Particles < 53 μm in diameter were collected in a bucket and left there for 24 hrs, after that the supernatant was removed and the particles dried. Subsamples from each size fraction were ground and analyzed for total C and ^{13}C . Sub-samples were taken from the dry aggregate size fractions 53-250 μm , 250-2000 μm and > 2000 μm to separate the POM associated and Mineral associated C and N. The method for the separation is described in detail by Cambardella and Elliott (1992). Subsamples of intact aggregates were combined with sodium hexametaphosphate and shaken for 12 hours on a reciprocal shaker. The dispersed organic matter plus sand was collected on a 53 μm mesh sieve; and the water in the soil slurry was evaporated in a forced-air oven at 50 °C and the dried sample ground and analyzed by Dumas combustion on a Carlo/Erba analyzer. The difference between the C and N

values for the evaporated soil slurry and those obtained from a non-dispersed soil sample was considered to be equal to the C or N retained in the sieve. Residual hexametaphosphate does not influence the recoveries of C and N by Dumas combustion.

Isotope Analysis

Total carbon content and ^{13}C content of aggregate size fractions was measured on a Finnigan Delta C Mass Spectrometer coupled to a Carlo Erba, NA 1500, CHN Combustion Analyzer via Finnigan's Conflo II Interface. The $^{13}\text{C}/^{12}\text{C}$ ratios are usually reported as $\delta^{13}\text{C}$ values measured relative to a standard and are expressed in parts per thousand:

$$\delta^{13}\text{C} = \left[\left(\frac{^{13}\text{C}/^{12}\text{C} \text{ sample}}{^{13}\text{C}/^{12}\text{C} \text{ reference}} - 1 \right) \right] \times 1000$$

The reference is Peedee Belemnite (PDB).

The fraction of new C was calculated using the equation:

$$F = (\delta t - \delta A) / (\delta B - \delta A)$$

where $\delta t = \delta^{13}\text{C}$ at time t , $\delta A = \delta^{13}\text{C}$ of the soil when a mix of C_3 and C_4 plants were grown (at time 0, 1997), $\delta B =$ the average $\delta^{13}\text{C}$ of C_3 plants, and $F =$ fraction of new C in the soil. F is the direct expression of the turnover of soil C (Balesdent and Mariotti, 1996).

Turnover and Net Inputs of Organic Carbon

Skjemstad et al. (1990) used the assumption of exponential decay as a means for comparing relative decay rates and turnover times in micro and macro-aggregates. We determined first order rate constants (k) as

$$k = -\ln (C_{\text{mix cotton}} / C_{\text{mix time 0}}) / t$$

where C_{mix} is the concentration of C from the mixture of C3 and C4 plants remaining in each size fraction from the soil at the present time, $C_{mix\ 0}$ is the concentration of C from the mixture at time zero, before the switch to only C3 plants was made, and t is the length of time since that change took place (2005-1997= 8 yrs). The net input rate was calculated as the concentration of new C in each size fraction divided by the length of time since only C3 plants have been planted.

Sand-free Calculations

Because of the difference in amount of sand between aggregate-size classes, Elliott et al. (1991) suggested that comparisons of C content across aggregate-size classes should be based on sand-corrected C data. The sand content of the aggregate-size class was determined by dispersing 5 g of the fraction in 20 mL of sodium hexametaphosphate and sieving through a 53- μ m sieve. Sandfree C concentration was calculated as follows:

$$\text{Sand free C (g kg}^{-1}\text{)} = \text{C fraction} / [1 - (\text{sand proportion}) \text{ fraction}]$$

Statistical Analysis

The experiment was analyzed as a Nested split plot design, data subjected to the SAS statistical package for analysis of variance (ANOVA-PROC GLM, SAS Institute, 1990). Tukey's HSD ($p < 0.05$) was used for mean separation.

Results

The distribution of water stable aggregates was significantly influenced by tillage practice in both depths of soils (Fig. 1, a and b). Macroaggregates ($> 2000 \mu\text{m}$) made up the

largest percentage of the whole soil, and they were 1.6 times greater ($p < 0.001$) in NT than in CT. For aggregate sizes of 250-2000, 53-250 and $< 53 \mu\text{m}$, the aggregates in CT plots were nearly 1.3, 2.9 and 2.2 times greater than NT, respectively ($p < 0.05$). There was a significant difference on the distribution of macroaggregates in depth 5-15 cm (Fig. 1b). The distribution of macroaggregates from NT was 1.2 times greater than in CT. However, there was no significant difference in the smaller WSA size classes.

Total C as well as N was significantly different by tillage, aggregate size and their interaction ($p < 0.01$, Fig 2). For all aggregate size classes, total sand free C and N in 0-5 cm soils were significantly higher in NT than of CT. In surface soils (0-5 cm) of both NT and CT, sand-free C and N were highest in the 250-2000 μm aggregates, where total C and N were 1.6 and 1.5 times greater in NT than in CT, respectively. There were no significant differences on aggregate C and N at the 5-15 cm depth between tillage practices. Within size classes, the reduction in sand-free C and N content with depth was much greater in NT than in CT.

The mineral associated C and N were also significantly affected by tillage practices in all aggregate size classes, with higher values for NT than CT ($P < 0.05$, Fig 3). The differences between tillage treatments did not vary much within aggregate size classes.

Aggregates 53-250 μm had higher C and N concentrations in NT, being in both cases 1.5 times higher than in CT ($p < 0.01$). Deeper in the soil profile CT trended higher than NT for all aggregate size classes in both C and N concentrations. Furthermore, for the NT tillage practice, total C and N for 5-15 cm depth were much smaller than for surface soils, where as in CT the values varied little within the soil profile.

Carbon and nitrogen associated with particulate organic matter (Fig. 4) were calculated as the difference between total aggregate C or N and mineral C or N, respectively. The differences

in POM associated C or N were significantly affected by tillage and aggregate size class for all aggregate size classes in surface soils in the case of N, and for C it was significant in aggregates >2000 μm and 250-2000 μm . The POM associated C was greater in the 2000-250 μm class for both CT and NT, and POM C in NT was 1.5 times greater than CT. For POM associated N, the greatest difference was in the aggregates >2000 μm , with NT value 6 times greater than CT. The concentrations of C and N varied within depth for both NT and CT. For POM associated C, only the values for >2000 μm showed a significant difference, with CT 1.7 times greater than that for NT. POM associated N in aggregates >2000 μm in CT was 1.5 times higher than NT, nevertheless, the greatest difference between tillage practice was in the aggregate size class 250-53 μm , with NT being 8 times greater than CT. In general, the C/N ratios were much greater between size classes for the POM associated organic matter than for the Mineral Associated.

Total carbon is compared within the same tillage practice for the three different sampling dates 1997, 2000 and 2005 (Fig. 5). The total C in the surface soils in NT showed a significant loss of C for aggregate size classes >2000, 2000-250 and 250- 53 μm ($p < 0.01$) and a constant concentration of C in the aggregate <53 μm . The net loss of C from 1997 to 2005 accounted for an average 1.6 times reduction (40 g C kg^{-1}) of C concentration in the NT tillage. In surface soils of CT, there were no significant differences in time for the aggregate size classes >2000, 2000-250 and 250- 53 μm , however, there was a significantly different total C concentration for the aggregates <53 μm .

We observed significant differences in ^{13}C ratios in time within tillage method (Fig. 6). Ratios for all aggregate size classes were shifting towards the ^{13}C ratios of C_3 plants. The values for NT in the surface soils were generally lower (more negative) compared to CT. In both NT and CT, the aggregates >53 μm had the youngest age (most negative ^{13}C ratio) and the

aggregates $< 53 \mu\text{m}$ showed the oldest signature, being older in CT than NT. Both the 0-5 and 5-15 cm depths in CT were significantly more negative in the year 2000 than in 2005, and the 5-15 cm depth in NT was not significantly different in 1997 and 2000.

For surface soils there were similar values of new C in CT than in NT; in lower depths, the larger aggregates in CT had more new C incorporated than NT (Table 1). The proportion of new C did not show a significant variation for both depths within the NT practice, with a similar behavior in CT except for the deeper soil profile.

Table 2 shows the average rate constant (k) for loss and turnover time ($1/k$) of the mixture of C_4 and C_3 -C and average net input rate of C_3 -C in aggregate size fractions as determined by ^{13}C natural abundance. The k was lower for microaggregates than for macroaggregates in both tillage treatments and the turnover was faster in CT than in NT for both surface and deep soil in most all of aggregate size fractions. The turnover of aggregate associated old C in microaggregates was 27.08 years for NT and 26.12 for CT (0-5 cm) and for the deeper soil profile the difference becomes greater (34.5 and 12 yr, respectively). In the deeper profile the aggregates $< 53 \mu\text{m}$ for both NT and CT have a very little net input rate of new C. Furthermore, within tillage there is a difference of turnover within aggregate size fractions.

Discussion

Aggregate Distribution

Macroaggregates ($>2000 \mu\text{m}$) from surface soils of NT were more abundant than those of CT soils and the situation was inverted for smaller aggregates (2000-250, 250- 53 and $<53 \mu\text{m}$) for surface soils (Fig. 1). There were no significant differences in the amount of WSA between NT and CT for the lower depth samples (5-15 cm) except for the macroaggregate size fraction ($>$

2000 μm). Several researchers found more macroaggregates in NT soils compared with CT soils (Carter, 1992; Beare et al., 1994a, Paustian et al., 1997; Six et al., 2000a; Bossuyt et al., 2002). Our results are similar to the values obtained by Beare et al. (1994a) and Kimber Collins (unpublished data) in the same field site. Tisdall and Oades (1980) showed that long term tillage decreases the length of roots, hyphae, and soil organic matter and causes a reduction in macroaggregation. Thus tillage practices disrupt large macroaggregates and increase the proportion of smaller aggregates. A breakdown of macroaggregates results in the release of labile SOM, then making it available to microbial decomposition. Tillage management changes soil conditions (aeration, temperature, and moisture) and decomposition rates of litter (Cambardella and Elliott, 1993). In NT, there is residue accumulation at the surface where the litter decomposition rate is slowed due to drier conditions and reduced contact between soil microorganisms and litter (Salinas-Garcia et al., 1997), and a greater proportion of microbial biomass composed of fungi, which contribute to macroaggregate formation and stabilization (Tisdall and Oades, 1982). In CT, subsurface soil is brought to the surface and exposed to wet–dry, freeze–thaw cycles and raindrop impact (Beare et al., 1994; Paustian et al., 1997). Also, incorporating plant residues in soil can affect the soil microclimate and increase plant residue contact with soil. This will increase residue decomposition and organic matter transformation (Beare et al., 1992; Cambardella and Elliott, 1993).

Higher microbial activity depletes SOM, which eventually leads to decreased microbial biomass and activity and consequently a lower production of microbial-derived binding agents and a loss of aggregation (Jastrow, 1996; Six et al., 1998). Six et al. (1999) developed a conceptual model of aggregate turnover, which shows that the faster macroaggregate turnover in CT than in NT results in fewer macroaggregates being maintained but more free microaggregates

being present in CT than in NT. Our results support this model, since there are more macroaggregates in NT than in CT and more microaggregates in CT than in NT (Fig. 1, 0-5 cm).

Increasing aggregation with depth in CT indicates that tillage has a greater effect on the surface soils than on the deeper soils. According to Molope et al. (1987), aggregate stability might increase after disruption of aggregates by tillage due to greater microbial activity and the burial of crop residues. If the soil is not subject to the dispersive energy of raindrop impact (Sumner, 1992) and wet-dry and freeze thaw cycles (Rovira and Greacen, 1957; Adu and Oades, 1978; Hadas, 1990; Degens and Sparling, 1995), these factors could explain the increase in macroaggregates in 5-15 compared to 0-5 cm in CT.

Carbon and Nitrogen Concentrations

The total sand-free concentrations of aggregate C and N were up to 1.6 and 1.5 times higher in the surface samples of NT than of CT (Fig. 2). When aggregates are disrupted, more organic substrates become available for microbial attack, resulting in an increase in SOM decomposition, and therefore a decrease in C content. Since residues are placed on the surface in NT soils and aggregates are less disturbed, higher C concentrations are found in these surface layers. Beare et al. (1994a), Dick et al. (1997) and Six et al. (1999) also found higher C concentrations in surface samples of NT soils than of CT soils. These results suggest that where macroaggregates are relatively stable and resistant to frequent disruption, the additional inter-microaggregate organic matter should increase macroaggregate concentrations of total sand-free C and N compared with that of free microaggregates. Furthermore, disturbance-related SOM losses in CT and NT may be attributed to (i) reduced aggregation in CT in comparison with NT and (ii) increased decomposition due to aggregate disruption in CT (Beare et al., 1994b; Adu and

Oades, 1978; Rovira and Greacen, 1957). Paustian et al. (1997a) as well found in most cases there was an increase in C content under NT compared to CT systems after several long-term field studies and attributed this increase to a combination of reduced litter decomposition and less soil disturbance under NT.

Elliott (1986) and Gupta and Germida (1988) noted that concentrations of C and N were higher in aggregates of native sod than in those of long-term cultivation. Again, Beare et al. (1994a) and Kimber Collins (unpublished data) observed similar results in the same study area. In the present study, total sand-free concentrations of C and N were highest in the 2000-250 μm of NT and were significantly lower in larger and smaller aggregate size classes. Beare et al. (1994a) observed similar results but in a different aggregate size class that we did not study (106-250 μm) and they supported their results with the formation of microaggregates within macroaggregates viewed by Elliott and Coleman (1988) and since then studied by many others (Jastrow and Miller, 1998; Six et al., 1998; Puget et al., 2000).

There were no significant differences between tillage systems in deeper samples. Six et al. (1998) and Beare et al. (1994a) also reported no differences in aggregate C and N concentrations in the lower depth of no tillage and plowed treatments. It is important to notice that even when the WSA distribution (Fig. 1) shows a greater percentage of microaggregates in CT than in NT, there is more C and N in microaggregates of NT than in CT.

We found significant differences for all aggregate size classes (0-5 cm) for C and N mineral associated organic matter, with NT values being up to 1.3 times greater than CT values for C concentrations and 1.4 times greater for N concentrations (Fig. 3). Beare et al. (1994a) found similar results and they suggest that a significant part of the mineral associated organic matter in NT is being protected from microbial attack by its physical isolation within

macroaggregates. Studies in grassland soils have shown that cultivation results in a proportionally greater decline in POM than in Mineral Associated organic matter (Tiessen et al., 1983; Cambardella and Elliott, 1992). This could partly explain why we do not observe significant variation among aggregate size classes within the treatment. Six et al. (1998) as well found no differences in CT and NT for the lower soil profile. Jastrow (1996) noted that most of the accumulated C occurred in the mineral associated fraction of macroaggregates, suggesting that inputs of organic debris were rendered relatively rapidly into particles or colloids that are associated with mineral matter and thus are physically protected, slowing decomposition and promoting the development of stable microaggregates within macroaggregates.

POM associated C and N values were greater in NT than in CT in all aggregate size classes in surface soils, and they were greater for bigger than for smaller aggregates (Fig. 4). There is strong evidence that, in no-till, aggregate formation and stabilization processes are directly related to the decomposition of root-residue and the dynamics of POM C in the soil (Gale et al., 2000). Cambardella and Elliott (1992) suggested that the whole soil POM accounts for much of the SOM lost with cultivation of native sod and that NT practices can reduce these losses. Six et al. (1999) also showed less fine POM in smaller aggregates, which agreed with the results in this study. Conventional tillage disrupts macroaggregates and reduces the accumulation of fine POM within macroaggregates, in addition to the fact that the amount of macroaggregates is lower in CT than in NT. Beare et al. (1994a) presented similar results and noted that the differences in distributions of POM between depths in NT and CT may be attributed to biological activity near the soil surface, including fungi (Doran, 1980), roots (Cheng et al., 1990) and soil fauna (Parmelee et al., 1990), that help incorporate POM within macroaggregates and to increase their stability. Jastrow (1996) also found that organic C concentrations were greater in

macroaggregates than in microaggregates and Golchin et al. (1995) suggested that macroaggregates are stabilized mainly by carbohydrate-rich root or plant debris occluded within aggregates. This relates the percentage of water-stable aggregates (Fig 1) with total C content (Fig 5): the increase in C concentration in NT vs. CT helps explain the greater amount of WSA in NT compared to CT. In the case of CT for surface soils (Fig. 5) there were no significant differences in time for the aggregate size classes >2000, 250-2000 and 53-250 μm , however, there was a constant significantly different concentration of total C for the aggregates <53 μm , suggesting for CT and NT of this same size class that C in this fraction is stabilized by intimate associations with mineral particles. Gregorich et al. (1995) also found that the most stable organic matter is associated with the small size fraction. For the deeper soil profile a similar behavior was observed, with the <53 μm being significantly different than the other aggregate size classes. For both depths the old C associated with microaggregates may be physically protected. Christensen (1992) in a review of several studies observed that organic matter of the fine silt and coarse clay fractions tended on average to be more stable than organic matter of the other size fractions.

Carbon-13 Concentrations and Carbon Turnover

Carbon turnover for NT and CT was calculated in surface soils (0-5 cm) and deeper (5-15 cm) (Fig. 6). Data are shown in a timeline, since in 1997 as well as in 2000 the values of $\delta^{13}\text{C}$ were also measured. Once a chronology of land use is established, the decay rate of organic matter derived from previous vegetation can be calculated (Vitorello et al., 1989; Veldkamp, 1994; Gregorich et al., 1995, 1997). We observed significant differences in time within tillage method. Increasing tillage intensity enhanced the turnover of soil organic matter SOM and

decreases soil aggregation (Six et al., 1998). Since the average annual inputs of aboveground crop plus weed residues to NT and CT are very similar in HSB (Beare et al., 1994a), the differences in SOM content can be attributed to differences in the assimilation and decomposition of SOM under both tillage treatments. In this study however, we found a reduction of total C in NT plots for 2005 compared to previous years (Fig. 5), and this is not the case for CT. We do not have a satisfactory explanation for this result.

Gregorich et al. (1995) also found that $\delta^{13}\text{C}$ was more negative in the sand fraction than in smaller aggregate size classes. In both NT and CT, the aggregates $>53\ \mu\text{m}$ had the youngest age (most negative ^{13}C ratio) and the ones $<53\ \mu\text{m}$ the oldest signature, being older in CT than NT. Six et al. (1998) reported similar results and suggested that due to more intensive tillage in CT, macroaggregates turn over faster in CT than in NT, hence there is less formation stabilization of finer, more decomposed particles within CT fractions. In NT, aggregates were not disrupted by tillage, and fine POM was formed and stabilized through physical protection. This could explain the more negative values for $<53\ \mu\text{m}$ in NT compared to CT, leading to a slower rate of microaggregate formation within macroaggregates and less stabilization of new SOM in free microaggregates under CT.

Both the 0-5 and 5-15 cm depths in CT were significantly more negative in the year 2000 than in 2005, illustrating the tillage effect throughout both depths. In NT as well, both depths became more negative in year 2005. Since 5-15 cm depth in NT was not significantly different in 1997 and 2000, this suggests that new SOM was more impeded in moving towards lower depths and thus it is only a significant shift between 2000 and 2005. Furthermore, there was a significant difference between the aggregate size of 250- 53 μm for NT than CT in the soil surface, NT being more negative, which suggests that in NT more new C was being protected in

smaller aggregates, thus giving it a lower ^{13}C ratio. Higher CT values of new carbon for the 5-15 than NT 5-15 cm show how new carbon was incorporated more readily throughout the soil profile.

Table 1 shows the fraction of new C in the soil. This fraction is a direct expression of the turnover of soil Carbon. For surface soils there were similar values of new C in CT than in NT, suggesting the increase of new C is proportionate in both tillage practices in time. Similar results were reported by Six et al. (1998). In lower depths however, for the larger aggregates there was more new C incorporated in the soil of CT than in NT, once again suggesting more new organic matter movement through the soil profile. In the aggregate class 250-53 μm , in NT the C that made it through the soil profile stayed well occluded in this aggregate size class, whereas in CT it was still disrupted by cultivation. The proportion of new C did not show a significant variation for both depths within the NT practice, with a similar behavior in CT except for the deeper soil profile, suggesting that there was practically no new carbon occluded in this fraction ($p < 0.001$)

Shang and Tiessen (2000) isolated different density fractions within sand- and silt-size fractions and used the natural abundance of ^{13}C to determine C turnover in each sub-fraction. Highly variable turnover of C in the density fractions across and within size fractions indicated that mean residence time was not a singular property of soil C but rather a composite of a spectrum of mean residence times of different C fractions. Stabilization and destabilization of SOM can follow different pathways and involve different fractions; and there may be no unique pools having specific stability (Olk and Gregorich, 2006).

Experiments using ^{13}C natural abundance show up to a two-fold increase in mean residence time of SOM under no-till vs. intensive tillage (Paustian et al., 2000). The turnover in

years was faster in CT than in NT for both surface and deep soil in most all of aggregate size fractions (Table 2) , the turnover of aggregate associated old C in microaggregates was 27.08 years for NT and 26.12 for CT (0-5 cm) and for the deeper soil profile the difference becomes greater (34.5 vs. 12 yr). Studies of soil food webs associated with buried litter and surface litter have indicated that buried litter systems are more bacteria-dominated, while surface litter systems are more fungi dominated (Hendrix et al., 1986; Holland and Coleman, 1987). This results in faster turnover of inputs in CT with greater stabilization of inputs into organic matter in NT soil. In the deeper profile the aggregates < 53 μm for both NT and CT have a very little net input rate of new C. Furthermore, within tillage there is also a difference of turnover within aggregate size fractions. The measured turnover times for micro and macroaggregates support the concept of aggregate hierarchy (Oades and Waters, 1991) and the proposed differences in the persistence of organic materials involved in the binding of micro- vs. macroaggregates (Tisdall and Oades, 1982). Calculated turnover times for macroaggregates are actually a function of the turnover of microaggregate-associated C and of the organic materials binding micro aggregates into macroaggregates (Jastrow, 1996) and thus the binding agents themselves probably have shorter turnover times.

The net input rate of C_3 pathway organic matter to aggregates increased from microaggregates to macroaggregates. This finding supports the hypothesis that larger aggregates are bound together initially by the growth of roots and mycorrhizal hyphae and as they senesce and begin comminution will probably be incorporated into the intraaggregate POM of larger macroaggregates first (Jastrow, 1996). Furthermore, mycorrhizal fungi mycelium products would be expected to influence directly and strongly, aggregation at scales smaller than macroaggregate. However, little research has been devoted to the role of mycorrhizal fungi in the

formation of microaggregates (Rillig and Mummey, 2006). Bossuyt et al. (2000) noted in our same research area that more young C is accumulated in the subsurface soil of CT than NT, but this C is not stabilized in the long term, and they found that short- and long-term stabilization of C is higher in the soil surface layers under NT compared with CT. This C stabilization occurs mainly at the microaggregate level. The net input rate for microaggregates in our study was smaller for micro than for macroaggregates, which is consistent with the concept of stable microaggregates with slow turnover proposed by Tisdall and Oades (1980, 1982).

Conclusions and Future Directions

The results of this study support the conceptual model of aggregate turnover developed by Six et al. (1999): tillage practices disrupt large macroaggregates, having a faster macroaggregate turnover in CT than in NT, and in CT there is an increase in the proportion of free smaller aggregates. There is more total C and N in NT compared to CT, suggesting that where macroaggregates are relatively stable and not subject to frequent disruption, the additional inter-microaggregate organic matter should increase macroaggregate concentrations of total sand-free C and N compared with that of free microaggregates.

Even when the WSA distribution shows a greater percentage of microaggregates in CT than in NT, there is more C and N in microaggregates of NT than in CT. Disturbance-related SOM and thus C losses in CT vs. NT may be attributed to (i) reduced aggregation in CT in comparison with NT and (ii) increased decomposition due to aggregate disruption in CT. This suggests that a significant part of the mineral associated organic matter in NT is being protected from microbial attack by its physical isolation within macroaggregates.

POM associated C and N values were greater in NT than in CT all aggregate size classes in surface soils in the case of N, and for aggregates >53 μm in the case of C. Cambardella and Elliott (1992) suggest that the whole soil POM accounts for much of the SOM lost with cultivation of native sod and that NT practices can reduce these losses. Furthermore, conventional tillage disrupts macroaggregates and reduces the accumulation of fine POM within macroaggregates, in addition to the fact that the amount of macroaggregates is lower in CT than in NT.

The differences in distributions of POM between depths in NT and CT may be attributed to biological activity near the soil surface, including fungi (Doran, 1980), roots (Cheng et al., 1990) and soil fauna (Parmelee et al., 1990), that help incorporate POM within macroaggregates and to increase their stability.

The fact that total C concentration in the three different samples over time shows similar values within tillage practice suggests that old C associated with microaggregates may be physically protected. Because surface soils in CT are exposed to variable abiotic conditions, these factors contribute not only to the more frequent disruption of soil aggregates, releasing the aggregate protected SOM for mineralization (Beare et al., 1994b) but also to a lower production of aggregate stabilizing agents (Angers et al., 1993)

As expected, ^{13}C ratios for all aggregate size classes are becoming more negative, shifting towards the ^{13}C ratios of C_3 plants. Our results suggest that due to more intensive tillage in CT, macroaggregates turn over faster in CT than in NT so there is less formation and stabilization of finer, more decomposed particles within CT fractions. The proportions of crop-derived C in macroaggregates were similar in NT and CT, but were greater in microaggregates from NT than microaggregates from CT. This suggests that a faster turnover rate of

macroaggregates in CT compared with NT leads to a slower rate of microaggregate formation within macroaggregates and less stabilization of new SOM in free microaggregates under CT than NT. The net input rate increases from microaggregates to macroaggregates, consistent with the concept of stable microaggregates with slow turnover rates.

For future research it would be important to explore the dynamics of these soil fractions, not only within yearly time frames but also within seasonal time frames that would capture the dynamics of labile SOM in response to short-term soil processes. Furthermore it would be interesting to take soil cores deeper than the plow layer so as to compare mass and distribution of carbon derived from the C₃ and C₄ mixture deeper in the soil profile. On the other hand, another study could compare the values of ¹³C from the NT and CT plots to ones of the forest sites near it, to determine the loss rate of the original organic matter and the net input rate from the new source.

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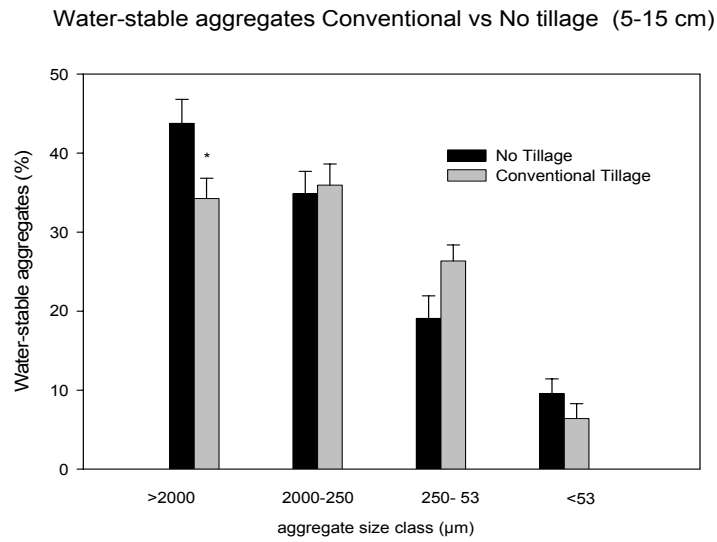
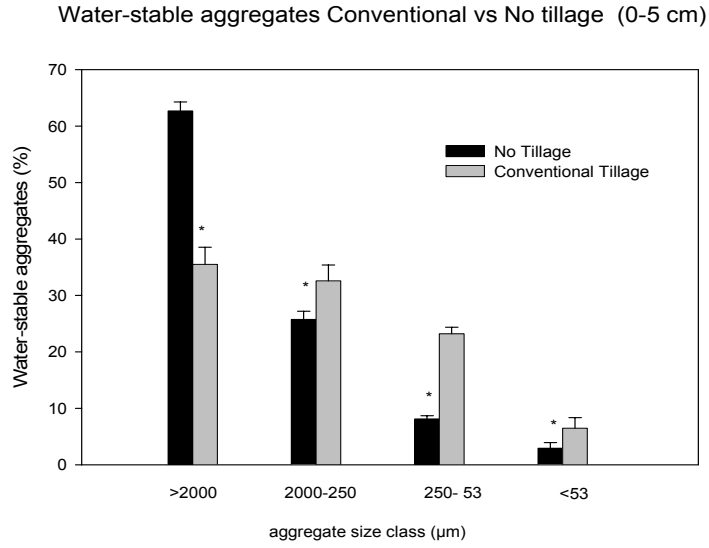


Figure 1. Distribution of water-stable aggregates from conventional and no tillage soils at 0-5 (a) and 5-15 (b) depth. Bars are means \pm S.E. Asterisks indicate significant differences ($P < 0.05$, Tukey's HSD) between tillage treatments within size class; $n=32$.

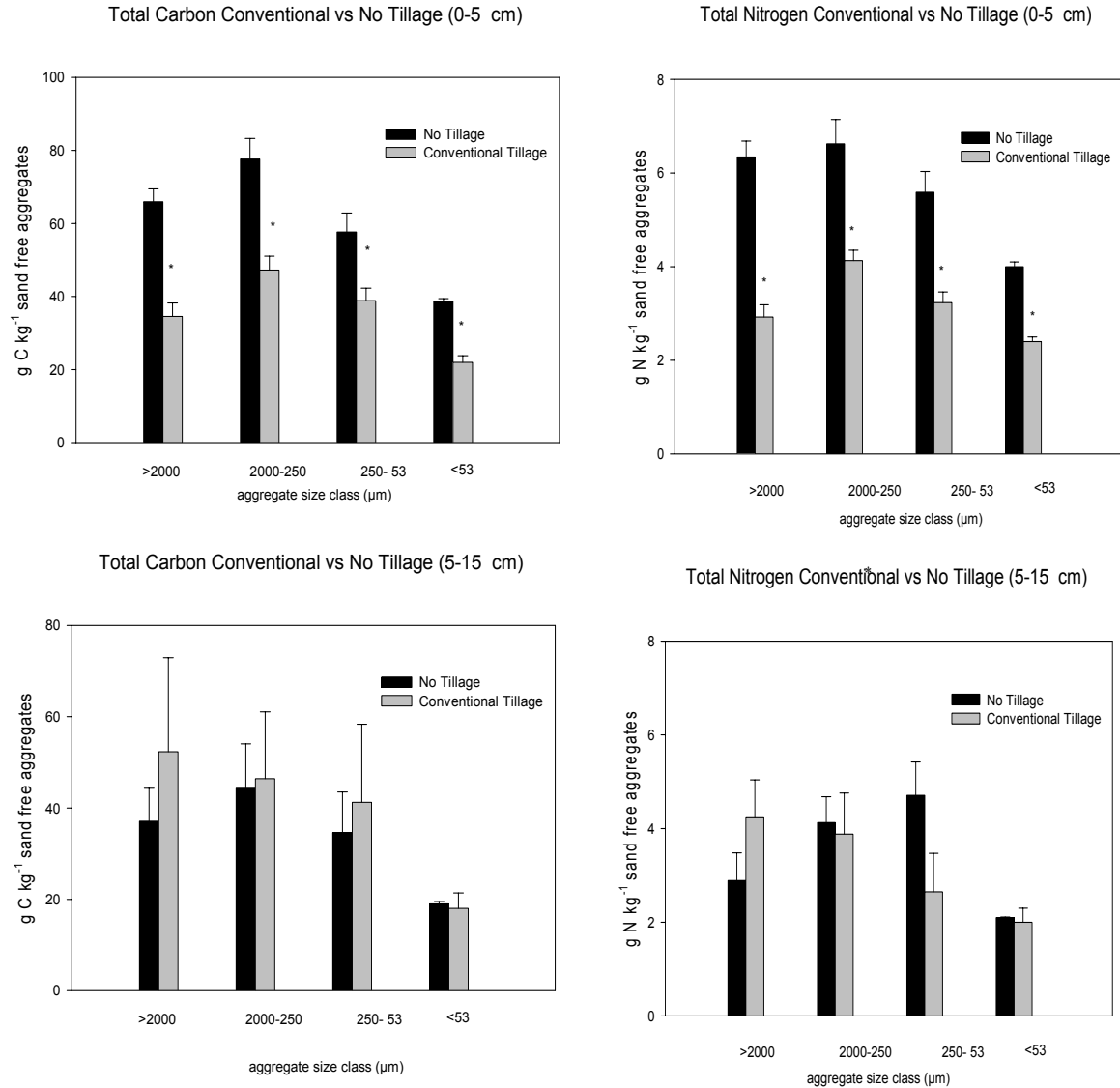


Figure 2. Total C and N (g kg⁻¹; normalized to a sand-free basis) in water-stable aggregates from conventional and no tillage soils at 0-5 and 5-15 depth. Bars are means \pm S.E. Asterisks indicate significant differences ($P < 0.05$, Tukey's HSD) between tillage treatments within size class; $n=32$.

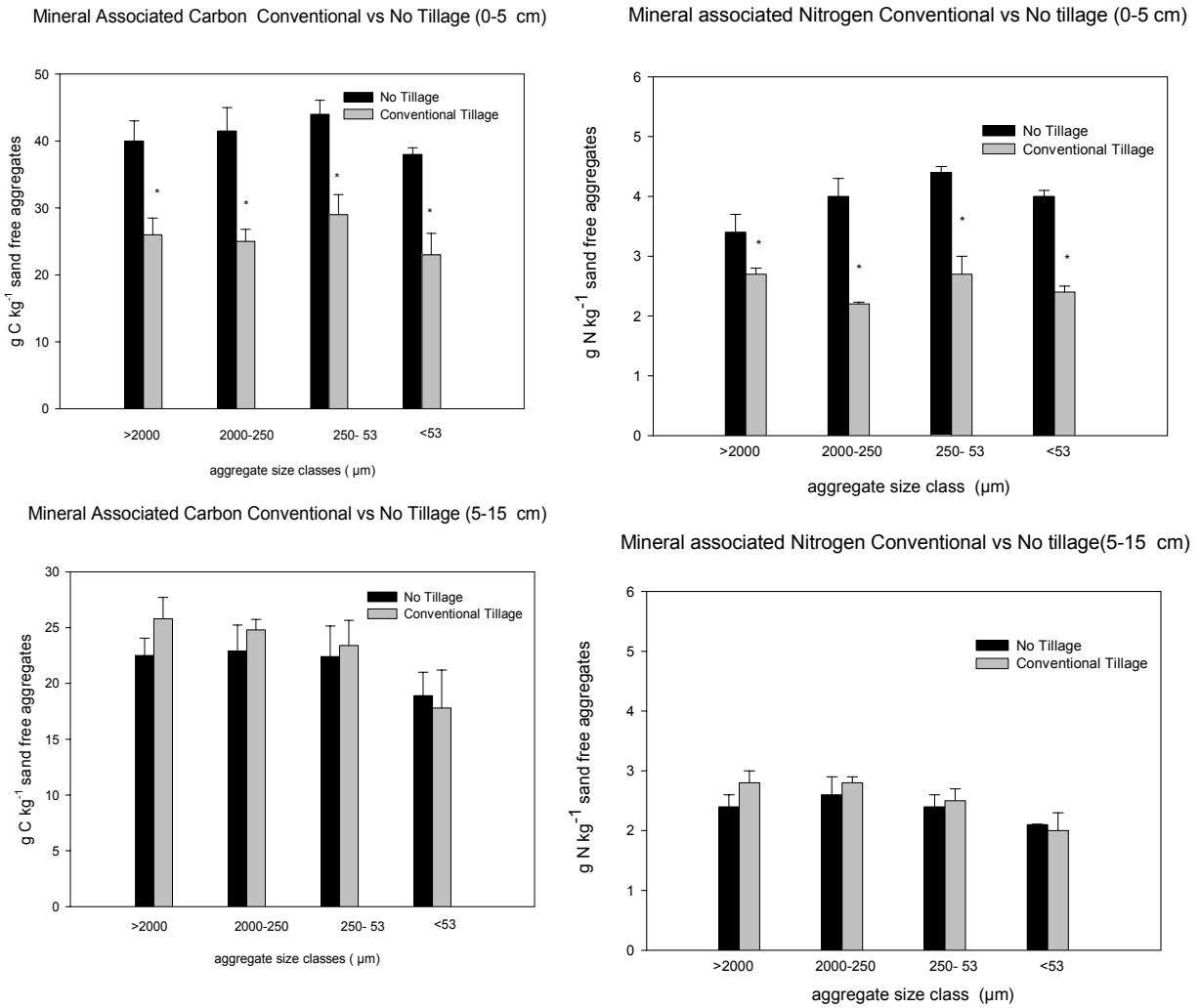


Figure 3. Mineral Associated Carbon and Nitrogen (g kg⁻¹; normalized to a sand-free basis) in water-stable aggregates from conventional and no tillage soils at 0-5 and 5-15 depth. Bars are means \pm S.E. Asterisks indicate significant differences ($P < 0.05$, Tukey's HSD) between tillage treatments within size class; $n=32$.

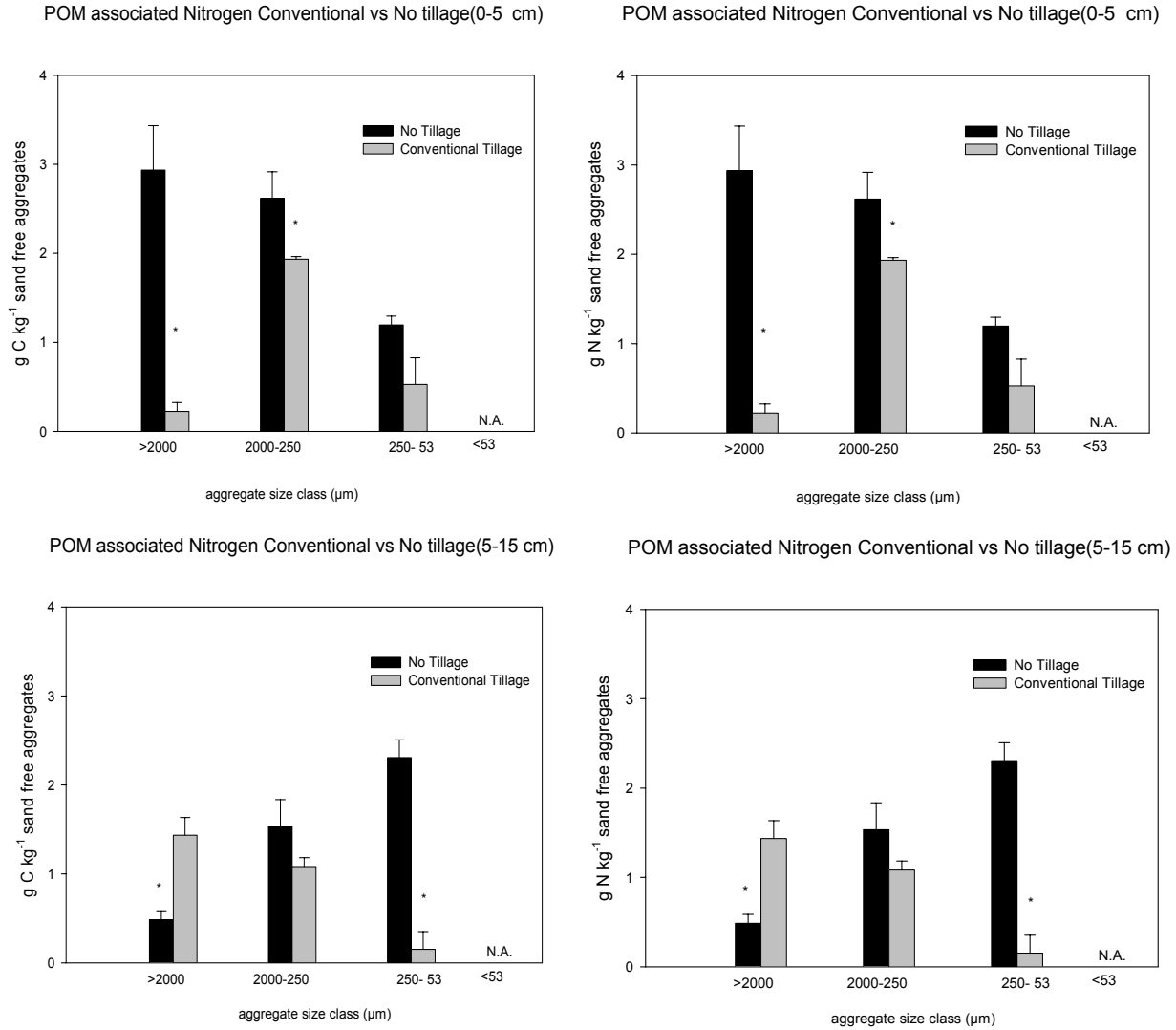


Figure 4. Particulate Organic Matter Associated Carbon and Nitrogen (g kg⁻¹; normalized to a sand-free basis) in water-stable aggregates from conventional and no tillage soils at 0-5 and 5-15 depth. Bars are means ± S.E. Asterisks indicate significant differences (P < 0.05, Tukey's HSD) between tillage treatments within size class; n=32.

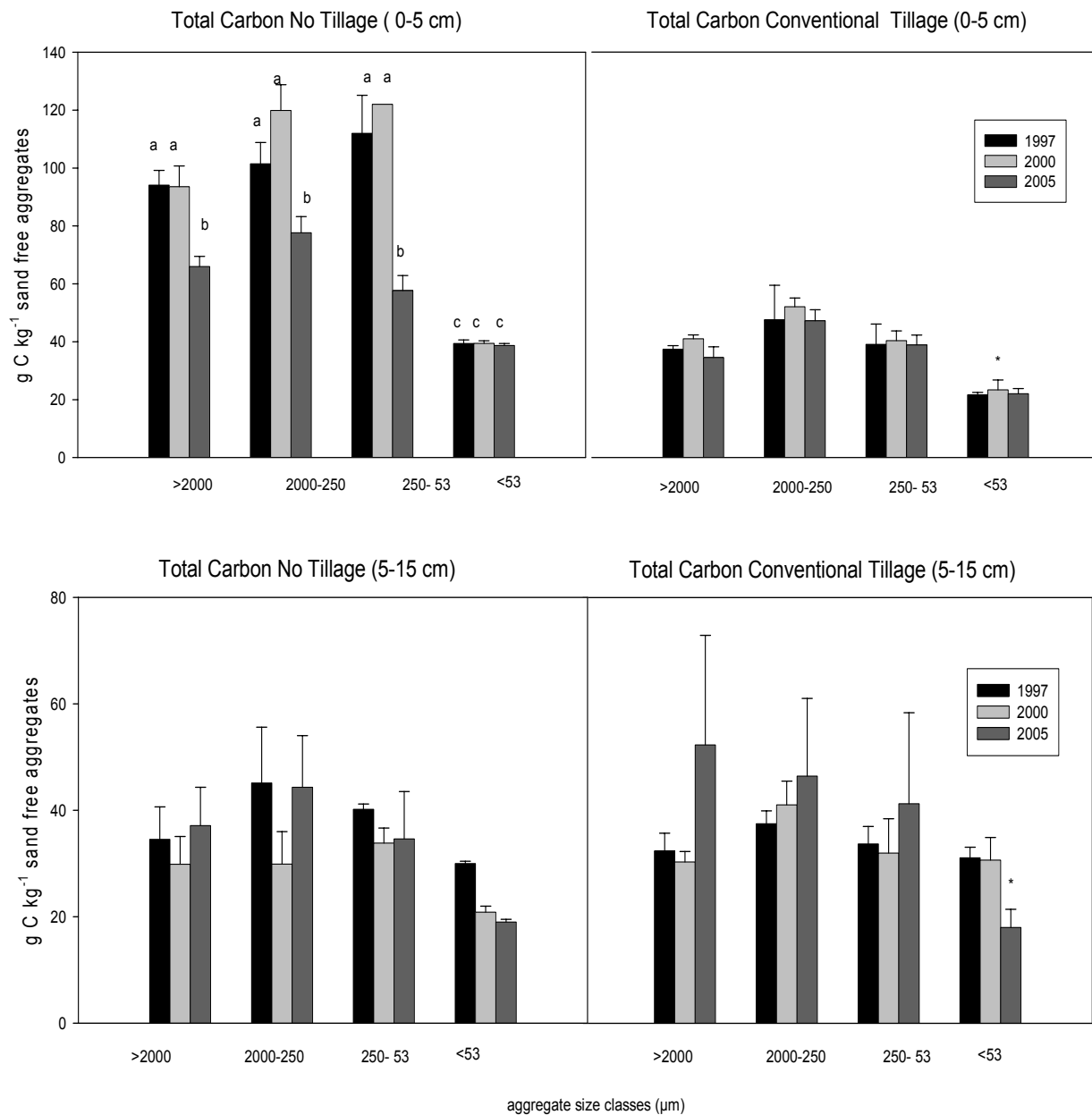


Figure 5. Total Carbon concentration in time for No Tillage and Conventional Tillage in different depths (0-5 and 5-15 cm) and aggregate size fractions.

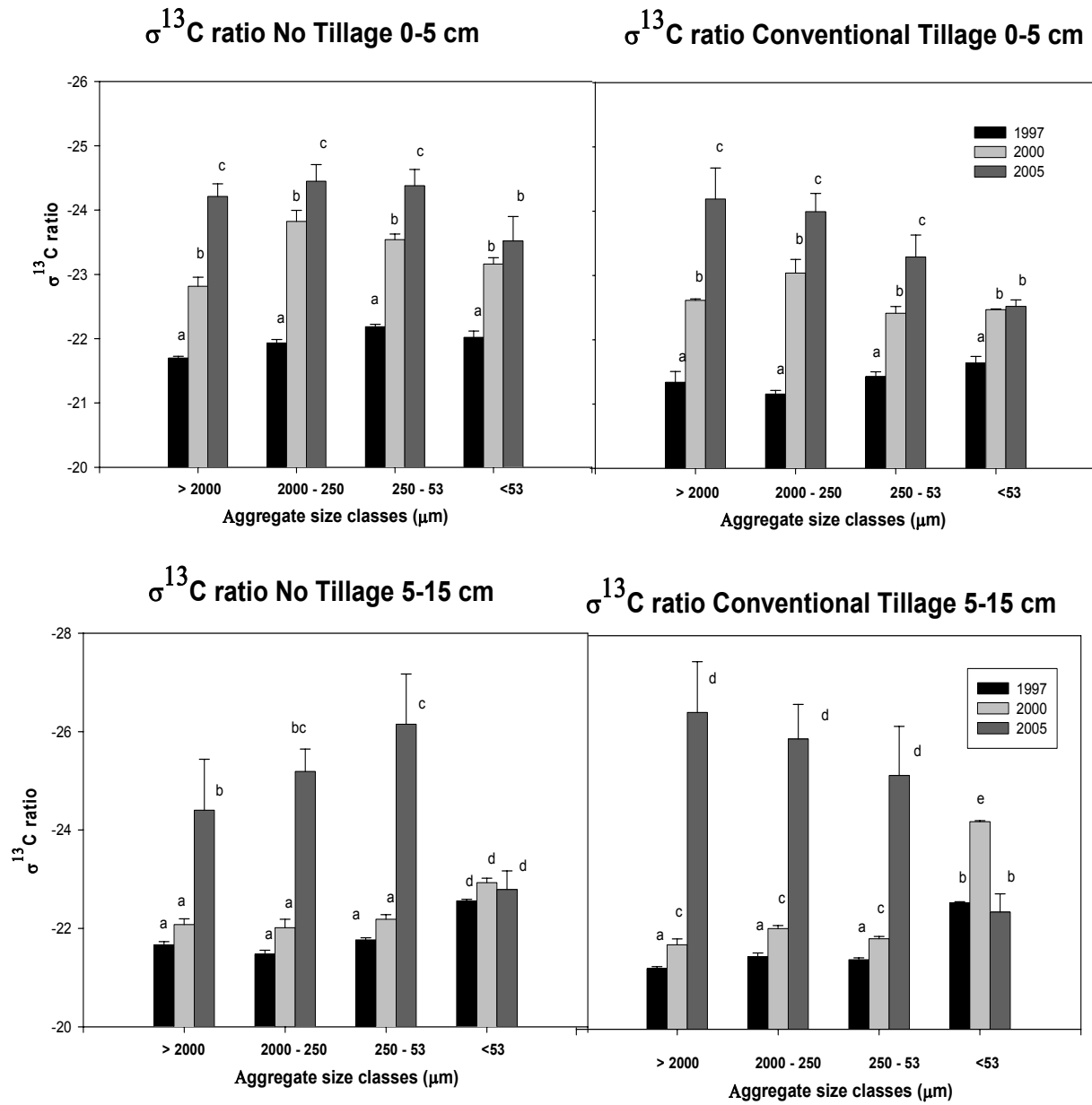


Figure 6. ¹³C ratios of No tillage and Conventional Tillage in time in different depths (0-5 and 5-15 cm) and aggregate size fractions.

CHAPTER 3

CONCLUSIONS

The results of this study support the conceptual model of aggregate turnover developed by Six et al. (1999): tillage practices disrupt large macroaggregates, having a faster macroaggregate turnover in CT than in NT, and in CT there is an increase in the proportion of free smaller aggregates.

There is more total C and N in NT compared to CT, suggesting that where macroaggregates are relatively stable and not subject to frequent disruption, the additional inter-microaggregate organic matter should increase macroaggregate concentrations of total sand-free C and N compared with that of free microaggregates.

Even when the WSA distribution shows a greater percentage of microaggregates in CT than in NT, there is more C and N in microaggregates of NT than in CT. Disturbance-related SOM and thus C losses in CT vs. NT may be attributed to (i) reduced aggregation in CT in comparison with NT and (ii) increased decomposition due to aggregate disruption in CT. This suggests that a significant part of the mineral associated organic matter in NT is being protected from microbial attack by its physical isolation within macroaggregates.

POM associated C and N values were greater in NT than in CT all aggregate size classes in surface soils in the case of N, and for aggregates $>53 \mu\text{m}$ in the case of C. Cambardella and Elliott (1992) suggest that the whole soil POM accounts for much of the SOM lost with cultivation of native sod and that NT practices can reduce these losses. Furthermore, conventional tillage disrupts macroaggregates and reduces the accumulation of fine POM within

macroaggregates, in addition to the fact that the amount of macroaggregates is lower in CT than in NT.

The differences in distributions of POM between depths in NT and CT may be attributed to biological activity near the soil surface, including fungi (Doran, 1980), roots (Cheng et al., 1990) and soil fauna (Parmelee et al., 1990), that help incorporate POM within macroaggregates and to increase their stability.

The fact that total C concentration in the three different samples over time shows similar values within tillage practice suggests that old C associated with microaggregates may be physically protected. Because surface soils in CT are exposed to variable abiotic conditions, these factors contribute not only to the more frequent disruption of soil aggregates, releasing the aggregate protected SOM for mineralization (Beare et al., 1994b) but also to a lower production of aggregate stabilizing agents (Angers et al., 1993)

As expected, ^{13}C ratios for all aggregate size classes are becoming more negative, shifting towards the ^{13}C ratios of C_3 plants. Our results suggest that due to more intensive tillage in CT, macroaggregates turn over faster in CT than in NT so there is less formation and stabilization of finer, more decomposed particles within CT fractions. The proportions of crop-derived C in macroaggregates were similar in NT and CT, but were greater in microaggregates from NT than microaggregates from CT. This suggests that a faster turnover rate of macroaggregates in CT compared with NT leads to a slower rate of microaggregate formation within macroaggregates and less stabilization of new SOM in free microaggregates under CT than NT. The net input rate increases from microaggregates to macroaggregates, consistent with the concept of stable microaggregates with slow turnover rates.

For future research it would be important to explore the dynamics of these soil fractions, not only within yearly time frames but also within seasonal time frames that would capture the dynamics of labile SOM in response to short-term soil processes. Furthermore it would be interesting to take soil cores deeper than the plow layer so as to compare mass and distribution of carbon derived from the C₃ and C₄ mixture deeper in the soil profile. On the other hand, another study could compare the values of ¹³C from the NT and CT plots to ones of the forest sites near it, to determine the loss rate of the original organic matter and the net input rate from the new source.

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