



## Short communication

**Soil macroaggregate turnover and microaggregate formation:  
a mechanism for C sequestration under no-tillage agriculture**J. Six\*, E.T. Elliott<sup>1</sup>, K. Paustian*Natural Resource Ecology Laboratory, Colorado State University, Fort Collins, CO 80523, USA*

Received 27 March 2000; received in revised form 4 July 2000; accepted 29 July 2000

**Abstract**

Soil disturbance from tillage is a major cause of organic matter depletion and reduction in the number and stability of soil aggregates when native ecosystems are converted to agriculture. No-till (NT) cropping systems usually exhibit increased aggregation and soil organic matter relative to conventional tillage (CT). However, the extent of soil organic matter changes in response to NT management varies between soils and the mechanisms of organic matter stabilization in NT systems are unclear. We evaluated a conceptual model which links the turnover of aggregates to soil organic matter dynamics in NT and CT systems; we argue that the rate of macroaggregate formation and degradation (i.e. aggregate turnover) is reduced under NT compared to CT and leads to a formation of stable microaggregates in which carbon is stabilized and sequestered in the long term. Therefore, the link between macroaggregate turnover, microaggregate formation, and C stabilization within microaggregates partly determines the observed soil organic matter increases under NT. © 2000 Elsevier Science Ltd. All rights reserved.

**Keywords:** Aggregate turnover; Carbon sequestration; No-tillage; Particulate organic matter

An accumulation of soil organic matter under no-tillage (NT) compared to conventional tillage (CT) confers important improvements in soil quality, soil fertility and sequestration of carbon. Mechanisms by which reduced tillage decreases soil organic matter decomposition (e.g. reduced soil disturbance, redistribution of residues) have been known for some time (Oades, 1984; Paustian et al., 1997) but the ultimate mechanisms have not been well elucidated. It has been established that the inclusion of organic materials within soil aggregates reduces their decomposition rate (Oades, 1984; Elliott and Coleman, 1988). Increases in aggregation concomitant with increases in organic C have been observed in NT systems (Paustian et al., 2000; Six et al., 2000). Tillage has been found to induce a loss of C-rich macroaggregates and a gain of C-depleted microaggregates (Six et al., 2000). However, this decrease in macroaggregates cannot explain the total C loss associated with tillage. Six et al. (1998, 1999a) suggested that increased macroaggregate turnover under CT is a primary mechanism causing decreases of soil C. Differential macroaggregate turnover induced by tillage and its influence on SOM dynamics are

represented in a conceptual model (Fig. 1) we developed to explain differences in C-sequestration between NT and CT. The model forms a basis for the hypotheses tested in this study.

At time 1 ( $t_1$ ), macroaggregates (250–2000  $\mu\text{m}$ ) are formed around fresh residue which then becomes coarse intra-aggregate particulate organic matter (iPOM) (Fig. 1). The fresh residue induces the formation of macroaggregates because it is a C source for microbial activity and the production of microbial-derived binding agents (Golchin et al., 1994; Jastrow, 1996; Six et al., 1999a). The model assumes similar rates of macroaggregate formation in NT versus CT (at  $t_1$ ) because the residue input is often similar in both management systems (Paul et al., 1997). Six et al. (1999a) found that the proportion of crop-derived C (i.e. newer C) relative to native grassland C (i.e. older C) was similar in NT and CT macroaggregates, confirming that the rate of macroaggregate formation was similar in both management systems.

Fine iPOM within a macroaggregate is derived from the decomposition and subsequent fragmentation of coarse iPOM ( $t_1$  to  $t_2$ ; Fig. 1). This process is supported by the observations that decomposition causes decreases in POM size (Guggenberger et al., 1994) and that fine iPOM is older than coarse iPOM (Six et al., 1998). Consequently, fine iPOM concentration is expected to increase with macroaggregate

\* Corresponding author. Tel.: +1-970-491-5599; fax: +1-970-491-1965.

E-mail address: johan@nrel.colostate.edu (J. Six).

<sup>1</sup> Present address: School of Natural Resource Sciences, University of Nebraska at Lincoln, Lincoln, NE 68583-0758, USA.

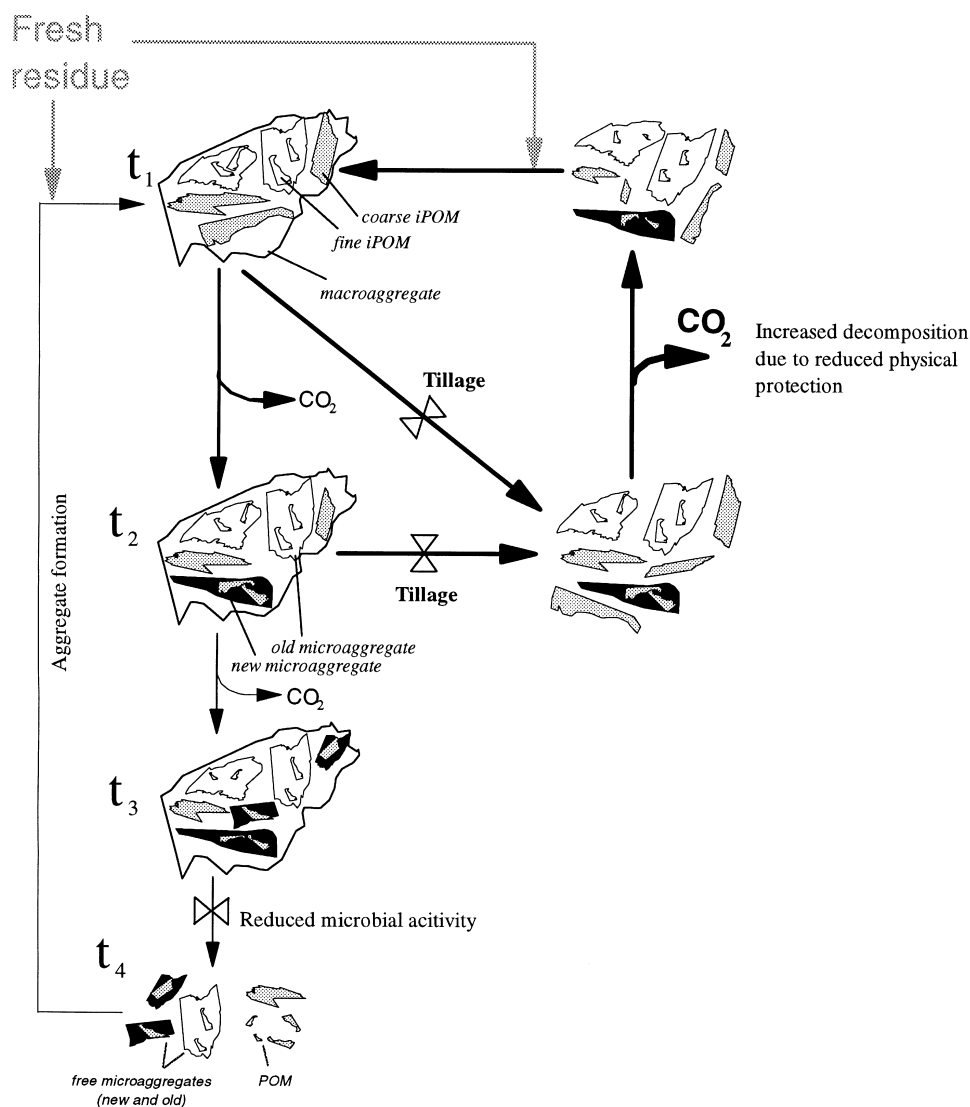


Fig. 1. This conceptual model shows the “life cycle” of a macroaggregate and the formation of microaggregates. These processes are the result of the interrelationship between the turnover of macroaggregates, the turnover of SOM and controlling factors such as disturbance (i.e. tillage). Turnover occurs through time as an aggregate is formed, becomes unstable and is eventually disrupted. Disturbances such as tillage shortcut this “life cycle” of macroaggregates and diminish the formation rate of new microaggregates and thus the sequestration of C within microaggregates and the soil as a whole. (iPOM = intra-aggregate particulate organic matter; POM = particulate organic matter;  $t$  = time;  $\text{---}\times\text{---}$  = process rate modifier).

age. Therefore, an abundance of older macroaggregates suggests a slower macroaggregate turnover and the ratio of fine iPOM to coarse iPOM within macroaggregates can be used as a relative measure of the turnover of macroaggregates. At four agricultural experiment sites this ratio was approximately twice ( $2.10 \pm 0.30$ ) as large in NT as in CT (Fig. 2), suggesting that macroaggregate turnover is approximately twice as slow in NT compared to CT.

As fine iPOM is formed it gradually becomes encrusted with clay particles and microbial products ( $t_2$  to  $t_3$ ) to form microaggregates within macroaggregates (Six et al., 1998, 1999a). In isotope tracer studies, labeled C was redistributed from macroaggregates to microaggregates with time (Angers et al., 1997; Gale et al., 2000) suggesting that microaggregates

are formed within macroaggregates (Oades, 1984; Elliott and Coleman, 1988). Eventually, the binding agents in macroaggregates degrade, resulting in loss of macroaggregate stability ( $t_4$ ) and the release of stable microaggregates, which become the building blocks for the next cycle of macroaggregate formation (Tisdall and Oades, 1982).

The model suggests that the increases in macroaggregate turnover induced by tillage yields fewer new free microaggregates in CT compared to NT at  $t_4$  (Fig. 1). Supporting this, less accumulation of crop-derived C in free microaggregates has been observed in CT compared to NT (Six et al., 1999a). This incorporation of new C into free microaggregates is an important factor contributing to C-sequestration (Skjemstad et al., 1990) since C contained in

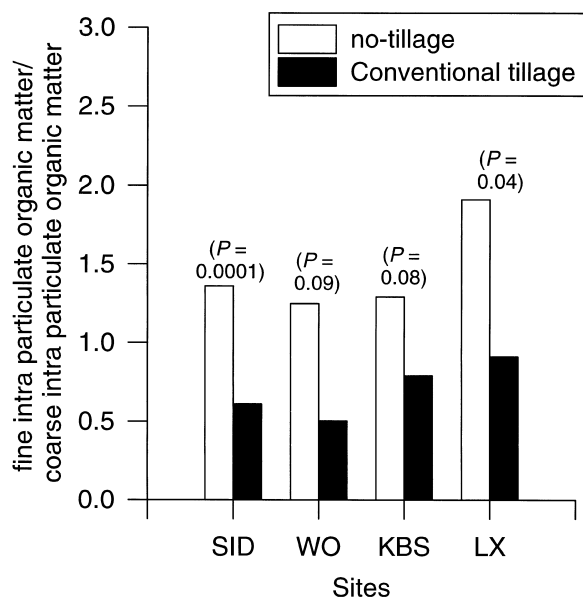


Fig. 2. Ratio of fine intra-particulate organic matter (iPOM) over coarse iPOM in the surface layer (0–5 cm) of NT and CT soils across four agricultural experiment sites (Sidney, NE; Wooster, OH; Kellogg Biological Station, MI; Lexington, KY). The ratio's are calculated based on data presented in Six et al. (1999a).

free microaggregates has a slower turnover than C in macroaggregates (Jastrow et al., 1996).

Based on the accumulated understanding embodied in our conceptual model, we advanced the following two hypotheses.

H1: in NT soils, there are more microaggregates contained within macroaggregates and there is more fine iPOM-C within these microaggregates than in CT soils.

H2: in NT soils, the SOM-C and iPOM-C of microaggregates that are contained within macroaggregates are younger than in CT soils.

The rationale for these two hypotheses is as follows. A slow macroaggregate turnover in NT allows time for the formation of fine iPOM from recent crop-derived coarse iPOM and the subsequent encapsulation of this fine iPOM by mineral particles and microbial byproducts to form stable microaggregates containing young crop-derived C. In contrast, the turnover of macroaggregates in CT is faster, providing less opportunity for the formation of crop-derived fine iPOM and stable microaggregates. Thus the organic matter contained in microaggregates of CT soils is dominated by older C derived from the pre-cultivation vegetation.

Direct testing of these hypotheses was accomplished by isolating microaggregates held within macroaggregates using a new method. Soil samples (0–5 cm) were taken at a long-term tillage experiment located at Sidney, NE. The experiment is on a Duroc loam, fine silty, mixed, mesic Pachic Haplustoll and includes NT and CT under wheat fallow, and a native grassland treatment. Prior to establish-

ment of the experiment (in 1969) the entire area was native grassland that had never been cultivated. To isolate microaggregates held within macroaggregates, we built a device to completely break up macroaggregates while minimizing the break down of the released microaggregates. We only analyzed macroaggregates from the 0–5 cm layer because we did not observe any significant differences between NT and CT in aggregate carbon and fine iPOM-C in the 5–20 cm layer at this site (Six et al., 1999a). Macroaggregates (10 g) were immersed in deionized water on top of a 250- $\mu$ m mesh screen and gently shaken with 50 glass beads (dia. = 4 mm). Continuous and steady water flow through the device ensured that microaggregates were immediately flushed onto a 53- $\mu$ m sieve and were not exposed to any further disruption by the beads. After all macroaggregates were broken up, the material on the 53- $\mu$ m sieve was sieved to ensure that the isolated microaggregates were water-stable. The inter-microaggregate POM retained together with the microaggregates on the sieve was isolated by density flotation in 1.85 g cm<sup>-3</sup> sodium polytungstate (Six et al., 1998). After density flotation, the microaggregates were dispersed in 5 g hexametaphosphate per liter and intra-microaggregate POM isolated by sieving (Six et al., 1998). To avoid cross contamination of C, and especially <sup>13</sup>C, among samples, sodium polytungstate was recycled according to Six et al. (1999b). The proportion of microaggregate (53–250  $\mu$ m) weight within macroaggregates (250–2000  $\mu$ m) was calculated as:

$$\frac{\text{microaggregate weight} - \text{weight of } 53\text{--}250 \mu\text{m sized sand}}{\text{macroaggregate weight} - \text{weight of } 250\text{--}2000 \mu\text{m sized sand}} \quad (1)$$

The weights of macro- and microaggregates were corrected for the sand content of the same size as the aggregates because sand of the same size as the aggregate is usually not a part of an aggregate and should consequently not be weighed as an aggregate. Carbon concentrations were measured with a LECO CHN-1000 analyzer and carbon-isotope ratios were determined using a Carlo Erba NA 1500 CN analyzer coupled to a Micromass VG isochrom-EA mass spectrometer.

Our hypotheses were corroborated with data from this new approach. We found that microaggregates within macroaggregates accounted for only 27% of the macroaggregate weight in CT, while 47% of the macroaggregate weight in NT consists of microaggregates (Table 1). Hence, the formation of new microaggregates within macroaggregates was reduced by a factor of about 2 (27 versus 47%) in CT compared to NT. These data are congruent with the previous observation that macroaggregates turn over twice as fast in CT compared to NT based on the ratio of fine iPOM to coarse iPOM (Fig. 2). Thus, macroaggregate turnover and microaggregate formation appear to be linked: a doubling of macroaggregate turnover reduces the microaggregate formation within macroaggregates by a factor of 2.

Within macroaggregates, the concentration of intra-

Table 1

Percent microaggregates in macroaggregates and concentrations of inter-microaggregate particulate organic matter (POM) and intra-microaggregate POM in no-tillage (NT) and conventional tillage (CT) surface layer (0–5 cm) at Sidney, NE

Treatment	Microaggregates in macroaggregates <sup>a</sup> (%)	Inter-microaggregate POM (g C kg <sup>-1</sup> sandfree macroaggregates)	Intra-microaggregate POM (g C kg <sup>-1</sup> sandfree macroaggregates)
NT	47.1 a <sup>b</sup>	1.3 b	11.1 a
CT	27.0 b	2.8 a	3.8 b

<sup>a</sup> The data presents the % of macroaggregate soil found in microaggregates (sand-corrected).

<sup>b</sup> Values followed by a different lowercase letter between management treatments are significantly different ( $P < 0.05$ ) according to Tukey's HSD mean separation test.

Table 2

$\delta^{13}\text{C}$  signature (per mille) of different aggregate size classes and particulate organic matter (POM) fractions in the surface layer (0–5 cm) of no-tillage (NT) and conventional tillage (CT) at Sidney, NE. Wheat residues had a  $\delta^{13}\text{C}$  of  $-27.57$  compared to a  $\delta^{13}\text{C}$  of  $-18.43$  for the native grassland soil present prior to initial cultivation in 1969. Thus more negative values indicate a greater proportion of crop-derived carbon and consequently a younger age

Fraction (per mille)	NT (per mille)	CT (per mille)
Macroaggregate (250–2000 $\mu\text{m}$ )	$-21.70$ a B <sup>a</sup>	$-21.96$ a B
Microaggregate within macroaggregate	$-21.39$ a B	$-20.32$ b C
Intra-microaggregate POM	$-23.56$ a A	$-22.73$ b A
Mineral fraction <sup>b</sup>	$-19.89$ a D	$-18.47$ b E
Free microaggregate (53–250 $\mu\text{m}$ )	$-20.55$ a C	$-19.19$ b D

<sup>a</sup> Values followed by a different lowercase letter between management treatments and within a fraction are significantly different ( $P < 0.05$ ) according to Tukey's HSD mean separation test. Values followed by a different uppercase letter between fraction and within a management treatment are significantly different ( $P < 0.05$ ) according to Tukey's HSD mean separation test.

<sup>b</sup> Macroaggregates contain  $<53 \mu\text{m}$  particles.

microaggregate POM-C was 3-fold greater in NT compared to CT while the concentration of inter-microaggregate POM-C (i.e. POM-C held within macroaggregates but not within microaggregates) was 2-fold greater in CT compared to NT (Table 1). In addition, the proportion of fine iPOM-C held within microaggregates that occurs within macroaggregates was much larger in NT compared to CT (90 versus 58%, respectively) leading us to conclude that the slower macroaggregate turnover in NT leads to a sequestration of C, primarily as fine iPOM held within stable microaggregates that are formed within macroaggregates. The greater accumulation of microaggregates rich in fine iPOM contributes to the greater total SOM-C under NT. Six et al. (1999a) found that the increase in total fine iPOM (i.e. inter- plus intra-microaggregate iPOM) alone accounted, on average, for 21% of the total C difference between NT and CT at four agricultural experiment sites.

Both the microaggregates within macroaggregates and the intra-microaggregate POM-C were younger in NT than in CT (Table 2). This is attributed to the slower turnover of macroaggregates in NT, resulting in more sequestration of crop-derived C within stable microaggregates formed within macroaggregates under NT. In addition, the  $\delta^{13}\text{C}$  signature of the mineral fraction ( $<53 \mu\text{m}$ ) contained within the macroaggregates suggests that C sequestration is also enhanced in this fraction (Table 2).

The halved rate of macroaggregate turnover, as suggested

by the data in Fig. 2, results in twice as many microaggregates formed within macroaggregates which, in turn, leads to twice as much fine iPOM accumulating in NT compared to CT. We have reported a doubling of both the amount of macroaggregates (Six et al., 2000) and the mean residence time of total soil C (Paustian et al., 2000; Six et al., 1999a) in NT compared to CT in several soils.

The linkage of macroaggregate turnover, microaggregate formation and SOM dynamics hypothesized in our model, was strongly supported by our data on the composition of macro- and microaggregates and their respective C dynamics. The linkage of these processes gives a mechanistic explanation as to why there is greater accumulation of SOM-C under NT agriculture. Our results underscore the importance of soil aggregation and more specifically the interactions of SOM and aggregate dynamics in controlling C-sequestration in soils.

## Acknowledgements

We appreciate the assistance of Dan Reuss, Karolien Deneff and Heleen Bossuyt during methods development. We thank Jason Kaye, Rich Conant, and Serita Frey for comments on the manuscript. Clay Combrink and Caroline Krumm are acknowledged for help with analyses. This work was funded by the US National Science Foundation and the US Department of Energy.

## References

- Angers, D.A., Recous, S., Aita, C., 1997. Fate of carbon and nitrogen in water-stable aggregates during decomposition of  $^{13}\text{C}^{15}\text{N}$ -labelled wheat straw in situ. *European Journal of Soil Science* 48, 295–300.
- Elliott, E.T., Coleman, D.C., 1988. Let the soil work for us. *Ecological Bulletins* 39, 23–32.
- Gale, J., Cambardella, C.A., Bailey, T.B., 2000. Root-derived carbon and the formation and stabilization of aggregates. *Soil Science Society of America Journal* 64, 201–207.
- Golchin, A., Oades, J.M., Skjemstad, J.O., Clarke, P., 1994. Study of free and occluded particulate organic matter in soils by solid state  $^{13}\text{C}$  CP/MAS NMR spectroscopy and scanning electron microscopy. *Australian Journal of Soil Research* 32, 285–309.
- Guggenberger, G., Christensen, B.T., Zech, W., 1994. Land-use effects on the composition of organic matter in particle-size separates of soil: I. Lignin and carbohydrate signature. *European Journal of Soil Science* 45, 449–458.
- Jastrow, J.D., 1996. Soil aggregate formation and the accrual of particulate and mineral-associated organic matter. *Soil Biology & Biochemistry* 28, 656–676.
- Jastrow, J.D., Boutton, T.W., Miller, R.M., 1996. Carbon dynamics of aggregate-associated organic matter estimated by carbon-13 natural abundance. *Soil Science Society of America Journal* 60, 801–807.
- Oades, J.M., 1984. Soil organic matter and structural stability: mechanisms and implications for management. *Plant and Soil* 76, 319–337.
- Paul, E.A., Paustian, K., Elliott, E.T., Cole, C.V. (Eds.), 1997. *Soil Organic Matter in Temperate Agroecosystems: Long-term Experiments in North America*. CRC Press, Boca Raton.
- Paustian, K., Collins, H.P., Paul, E.A., 1997. Management controls on soil carbon. In: Paul, E.A., Paustian, K., Elliott, E.A., Cole, C.V. (Eds.), *Soil Organic Matter in Temperate Agroecosystems: Long-term Experiments in North America*, CRC Press, Boca Raton, pp. 15–49.
- Paustian, K., Six, J., Elliott, E.T., Hunt, H.W., 2000. Management options for reducing  $\text{CO}_2$  emissions from agricultural soils. *Biogeochemistry* 48, 147–163.
- Six, J., Elliott, E.T., Paustian, K., Doran, J.W., 1998. Aggregation and soil organic matter accumulation in cultivated and native grassland soils. *Soil Science Society of America Journal* 62, 1367–1377.
- Six, J., Elliott, T., Paustian, K., 1999a. Aggregate and soil organic matter dynamics under conventional and no-tillage systems. *Soil Science Society of America Journal* 63, 1350–1358.
- Six, J., Schultz, P.A., Jastrow, J.D., Merckx, R., 1999b. Recycling of sodium polytungstate used in soil organic matter studies. *Soil Biology & Biochemistry* 31, 1193–1196.
- Six, J., Paustian, K., Elliott, E.T., Combrink, C., 2000. Soil structure and soil organic matter: I. Distribution of aggregate size classes and aggregate associated carbon. *Soil Science Society of America Journal* 64, 681–689.
- Skjemstad, J.O., Le Feuvre, R.P., Prebble, R.E., 1990. Turnover of soil organic matter under pasture as determined by  $^{13}\text{C}$  natural abundance. *Australian Journal of Soil Research* 28, 267–276.
- Tisdall, J.M., Oades, J.M., 1982. Organic matter and water-stable aggregates in soils. *Journal of Soil Science* 33, 141–163.