Aggregation and Organic Matter Protection Following Tillage of a Previously Uncultivated Soil

A. Stuart Grandy* and G. Philip Robertson

ABSTRACT

Understanding the effects of tillage on soils following years or decades of no-till is critical for developing C conservation strategies. To date, short-term responses to tillage in previously uncultivated or other long-term no-till soils have primarily focused on total C changes, which are difficult to detect. Tillage effects on soil conservation and C permanence may be better predicted by changes in more readily detected factors known to affect C storage such as aggregation and physically protected C. We annually plowed replicated plots in a previously uncultivated midsuccessional field between 2002 and 2004 and investigated changes in the distribution of aggregates, physically protected C, and light fraction (LF) organic matter. Within 60 d of initial cultivation, soil aggregates in the 2000- to 8000-μm size class declined from 0.47 to 0.15 g g^{-1} at 0- to 7-cm soil depth and from 0.32 to 0.23 g g^{-1} at 7 to 20 cm. Lower levels of aggregation persisted through the winter and spring of the following year. Inter-aggregate, unprotected light fraction (LF) increased following cultivation, as did particulate C in soil fractions with densities < 1.9 g cm^{-3}. Changes in the mass of total soil C were not detectable after 3 yr but the vertical distribution of all soil C pools was altered by plowing. Our study demonstrates that plowing once immediately and substantially alters aggregation and LF and particulate C dynamics and that these conditions persist. Results suggest that no-till soils need to be continuously maintained to protect aggregation and physically stabilized C pools.

CHANGES in soil organic matter (SOM) and other soil properties following the conversion of native ecosystems to agriculture are well known. Over time, SOM declines, is redistributed between surface and subsurface horizons, and is released from protected microsites (e.g., Del Gado et al., 2003; DeGryze et al., 2004). Accompanying changes include increased soil temperature, modified trace gas fluxes (e.g., Smith et al., 2001; Smith and Conen, 2004), and altered microbial community structure and function (e.g., Cavigelli and Robertson, 2000; Buckley and Schmidt, 2001), among others. Typically, SOM declines over time to 30 to 60% of original values (e.g., Davidson and Ackerman, 1993; Buyanovsky et al., 1997; West and Post, 2002; Lal, 2003). In tropical ecosystems change occurs rapidly, and within months of initial cultivation measurable declines in soil C concentration can sometimes be detected (Houghton et al., 1985; Detwiler, 1986; Brown and Lugo, 1990). In temperate ecosystems it is more difficult to generalize because data for the first 10 yr of cultivation are not currently available (Davidson and Ackerman, 1993; West and Post, 2002; Miller et al., 2004).

Theory and a handful of relevant studies suggest that immediate C losses from temperate region soils may be less dramatic than from tropical ecosystems (Bowman et al., 1990). Tiessen and Stewart (1983) found no difference in total C 4 yr after initially cultivating a silt loam. Other authors (e.g., Pierce et al., 1994; VandenBygaart and Kay, 2004) have also failed to find an effect within the first few years of cultivating long-term no-till soils, perhaps because of detection difficulties. Short-term total soil C changes are notoriously difficult to detect because of high background C concentrations in most arable soils, spatial variability, and the redistribution of aboveground pools on cultivation (Robertson et al., 1988; Sollins et al., 1999). Consequently, soil C permanence may be better predicted by changes in more readily detected factors known to affect soil C storage such as aggregation, the vertical distribution of C, and the dynamics of specific SOM fractions.

Macroaggregates (>250-μm soil particles) may be especially good predictors of potential C responses to tillage because of their importance for protecting recently deposited, labile, LF organic matter (Angers and Giroux 1996; Jastrow et al., 1996). Isotope studies, including those using ^{13}C CP/MAS NMR spectroscopy, have demonstrated that newly incorporated crop residues, root derived C, and young SOM are found in macroaggregates (Golchin et al., 1994; Six et al., 1998; Gale et al., 2000a; 2000b). Physical protection of LF and other compounds within macroaggregates may limit its oxidation by 50% or more (Balesdent et al., 2000). In ecosystems with frequent soil disturbance, accelerated turnover rates of macroaggregates limit the physical stabilization of labile SOM compounds such as LF C within aggregates (Six et al., 1999). These and other studies documenting tillage effects on aggregation and SOM, however, were conducted in soils with a long history of cultivation. The immediate and short-term effects of tillage on soil-aggregate distribution and SOM protection still need to be resolved.

Our overall objective was to investigate short-term responses of aggregate-size distribution and SOM to tillage independent of other factors. We removed the potentially confounding effects of historical tillage and fertilization by cultivating a previously undisturbed soil profile, and minimized plant community changes that can also confound studies of agricultural conversion by leaving the site fallow after tillage. This approach provides a uniquely unobstructed view into the potential consequences of tillage on soil structure and the mechanisms controlling C persistence.

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Abbreviations: DI, deionized water; LF, light fraction organic matter; MWD, aggregate mean weight diameter; NAPT, sodium polytungstate; POM, particulate organic matter (>53 μm); SOM, soil organic matter.

MATERIALS AND METHODS

Site Description and Experimental Design

Our experimental site was a previously never-tilled field at the W.K. Kellogg Biological Station (KBS) Long-Term Ecological Research site in southwest Michigan, USA (42° 24′ N. lat., 85° 24′ W long.). The site was cleared of trees (roots were left in place to decompose) in 1956 and thereby converted from a northern hardwood forest to a midsuccessional grassland community that has since been mowed each fall, with mown biomass left in place. Soils at the site are Kalamazoo (fine-loamy) and Oshtemo (coarse-loamy) mixed, mesic, Typic Hapludalfs developed on glacial outwash (Crum and Collins, 1995). The two series co-occur at KBS with variation within a series often as great as variation between series. Before initial cultivation, soils contained 31.8 (±1.4) g C kg^{-1} and 2.59 (±0.11) g N kg^{-1}, and soil pH averaged 5.93 (±0.11). The same soil C levels had been measured 3 and 12 yr before this study and are statistically indistinguishable from mineral soil C levels in nearby late-successional forests on the same two soil types (Robertson et al., 2000). The six dominant species at the site are *Rubus occidentalis* (black raspberry), *Dactylis glomerata* (orchard grass), *Bromus inermis* (smooth brome), *Poa pratensis* (Kentucky bluegrass), *Arrhenatherum elatius* (tall oatgrass), and *Agropyron repens* (quackgrass).

In 2002, we established in this successional field eight 3 by 6 m plots to which we assigned four replicates of two tillage treatments (cultivated and uncultivated control) in a randomized complete block design. Cultivated sites were mowed with biomass left in place before cultivation to 19 cm. We used a moldboard plow and a disc for primary and secondary cultivation, respectively. Cultivation occurred on 25 June 2002 (DOY 176) and again in the same plots on 15 June 2003 (DOY 166) and 20 June 2004 (DOY 172).

Soil and Plant Sampling

Soil samples for aggregate and total C and N analysis were collected before initial cultivation on 18 June 2002. Additional soil samples were collected post-initial-cultivation on 24 Aug. 2002 to determine the short-term effects of initial cultivation (60 d), and on 21 Oct. 2003 and 24 Sept. 2004 to quantify the effects of subsequent cultivation. An additional sample was taken on 27 May 2003 for aggregate analysis to characterize the persistence of changes in aggregation over the winter and following spring. On each sample date, five 3.8-cm diam. soil cores were taken from each plot to a depth of 20 cm, placed in plastic bags, and refrigerated (<7 d) before sieving through an 8-mm sieve and then air-drying at 20°C. A subsample of each sieved soil core was dried for 72 h at 60°C and the gravimetric water content used to determine the core dry weight and bulk density.

Plant and litter C estimates before initial cultivation were estimated by drying and analyzing litter and plant samples collected on 21 June 2002 from two 625 cm^{-2} quadrats in each plot. Organic C and total N concentrations of plant and soil samples were determined by dry combustion and gas chromatography in a CHNS analyzer (Costech ECS 4010, Costech Analytical Technologies, Valencia, CA). Soil C and N concentrations were converted to an areal basis by correcting for bulk density and soil depth.

Particulate Soil Organic Matter Density Distribution

We used sequential fractionation in sodium polytungstate (NAPT) to determine cultivation effects on the distribution of particulate SOM in four density fractions. Whole soil samples (15 g) were dispersed in 0.5% (w/v) sodium hexametaphosphate and poured through a 53-μm sieve. Particulate materials > 53 μm were then sequentially fractionated in NAPT at four densities to separate POM into four fractions (>1.9, 1.6–1.9, 1.3–1.6, <1.3 g cm^{-3}). Clay plus silt associated SOM (<53 μm) was determined by the difference between whole soil C and total particulate C.

Water-Stable Aggregate Distribution

Aggregate distribution was determined on triplicate 40 g air-dried soil samples by wet-sieving in water through a series of 2000-, 250-, and 53-μm sieves (Elliott, 1986; Six et al., 1998). Soil was submerged for 5 min on the surface of the 2000-μm sieve, which was then moved up and down over 2 min with a stroke length of 3 cm for 50 strokes. Sieving was repeated on the 250-μm (50 strokes) and 53-μm (30 strokes) sieves using the soil plus water that passed through the next larger sieve. Aggregates remaining on each sieve were dried at 60°C. Sand content was determined on an aggregate subsample after dispersing soil in sodium hexametaphosphate (0.5%) for 48 h on a rotary shaker at 190 rpm. We determined mean weight diameters (MWD) of sand-free aggregates by calculating the sum of the products of the mean diameter of each size fraction and the proportion of the total sample weight in that fraction (Kemper and Rosenau, 1986).

Aggregate-Associated Light Fraction Organic Matter

Previously published protocols (Six et al., 1998, Gale et al., 2000a) were used to separate inter- and intra-aggregate LF (organic matter of relatively low density). Aggregate subsamples were prewetted before LF separation to minimize aggregate slaking. An 8-g subsample of aggregates was divided in half and placed on two membrane filters (47-mm diam.; Pall Supor-450) overlaying two paper filters (70-mm diam.; Whatman 42) in a 10-cm Petri dish. Four milliliters of deionized (DI) water were trickled onto the paper filters to slowly wet all of the aggregates by capillarity. Aggregates were transferred from the membrane filters to 100-mL beakers after 16 h with 5-mL aliquots of NAPT at a density of 1.62 g cm^{-3}. A total of 55 mL of NAPT was used for each sample. A preliminary test showed that the final density of NAPT was about 1.60 g cm^{-3} following equilibration with the water contained in aggregates.

After 24 h, LF was aspirated from the surface of the NAPT and then rinsed on a hardened, ashless filter paper with at least 600 mL of DI H_2O. We refer to this pool as inter-aggregate LF because it consists of SOM located between aggregates. After removal of this pool, we aspirated the remaining NAPT. Aggregates were then dispersed to release the intra-aggregate LF using sodium hexametaphosphate as described previously and resuspended in NAPT (d = 1.62 g cm^{-3}). The intra-aggregate LF was collected from the surface. Organic C and total N concentrations of organic matter and whole soil samples were determined by dry combustion methods in a CHNS analyzer (Costech ECS 4010, Costech Analytical Technologies, Valencia CA.).

Statistical Analysis

Soil aggregate-size distributions and aggregate-associated C pools were corrected for sand content >53 μm. Carbon content of SOM fractions was calculated on an areal basis by correcting for bulk density. Tillage effects on SOM pools were analyzed by Proc Mixed (Version 8.2, SAS Institute, 1999) using a randomized complete block design analysis of variance (ANOVA). Treatment, soil depth and, in the case of C pools, SOM fraction were considered fixed effects and block was considered a random effect.

Table 1. Tillage effects on soil C and N and bulk density at 0- to 7- and 7- to 20-cm depths. Tillage occurred for the first time on 25 June 2002 and again in the spring of 2003 and 2004 on the same plots. †‡

		24 Aug. 2002					21 Oct. 2003					24 Sept. 2004				
Depth		**C**	**N**	**Bulk density**	**C**	**N**	**C**	**N**	**Bulk density**	**C**	**N**	**C**	**N**	**Bulk density**	**C**	**N**
cm		%	%	g cm^{-3}	kg m^{-2}	kg m^{-2}	%	%	g cm^{-3}	kg m^{-2}	kg m^{-2}	%	%	g cm^{-3}	kg m^{-2}	kg m^{-2}
0–7	control	3.21a	0.27a	1.06d	2.38b	0.20b	3.17a	0.23	1.06c	2.33b	0.17b	3.13a	0.26a	0.92b	2.01b	0.17bc
	tilled	2.34b	0.21b	1.15c	1.89c	0.17c	2.42b	0.19	1.13c	1.90b	0.15b	1.84b	0.16b	1.33a	1.71b	0.15c
7–20	control	1.09d	0.10d	1.40a	1.98c	0.19bc	1.09c	0.09	1.45a	2.05b	0.18b	1.26c	0.12c	1.42a	2.30b	0.21b
	tilled	1.66c	0.15c	1.30b	2.80a	0.25a	1.71bc	0.14	1.31b	2.91a	0.24a	1.67b	0.15bc	1.41a	3.03a	0.27a
ANOVA F-tests																
Source									P values							
Tillage		0.103	0.199	0.989	0.114	0.182	0.788	0.978	0.242	0.247	0.254	0.063	0.015	0.002	0.323	0.202
Depth		0.001	0.001	0.001	0.026	0.005	0.002	0.001	0.001	0.064	0.011	0.001	0.001	0.001	0.003	0.001
Tillage × depth		0.001	0.001	0.001	0.001	0.004	0.017	0.050	0.005	0.005	0.022	0.001	0.002	0.001	0.032	0.046

† Means within a column followed by different letters are statistically different ($p < 0.05$).

‡ There were no differences in total soil C to 20 cm between control and cultivated plots in any year.

Single degree of freedom comparisons were made to compare differences among soil fractions and treatments at different depths. An LSD test was used to calculate a 95% confidence interval around the differences between means generated using the Diff option in Proc Mixed. The LSD was performed using the PDMIX800 algorithm (Saxton, 2003).

RESULTS

Total Soil Carbon and Nitrogen

On 24 Aug. 2002, 60 d after initial tillage, there were significant changes in the depth distribution of soil C and N (Table 1). Carbon and N concentrations in tilled plots were significantly lower than those in conventional systems at 0 to 7 cm but greater at the 7- to 20-cm depth. Expressed on an areal basis, soil C and N on 24 Aug. 2002 showed the same trend, despite tilled plots having a slightly higher bulk density at 0 to 7 cm and lower bulk density at the 7- to 20-cm depth. These changes in soil C and N generally persisted in 2003 and 2004 although differences between C concentrations at the 7- to 20-cm depth were not significantly different in 2003 (Table 1). Expressed on an areal basis, total soil C at the 0- to 20-cm depth was not different between treatments in any year.

Aggregation

Before tillage in June 2002 there were no differences in soil aggregation between treatments (Fig. 1 and 2). Within 60 d, tillage decreased 2000- to 8000-μm aggregates at both soil depths and increased the amount of soil in the 53- to 250-μm and <53-μm size classes at 0 to 7 cm. Two thousand– to 8000-μm aggregates declined from 0.47 to 0.15 g g^{-1}. These changes persisted over the winter and into the following spring when control plots had 0.40 g g^{-1} in the 2000- to 8000-μm size class and tilled plots had 0.24 g g^{-1}. Differences in 2000- to 8000-μm aggregates at both depths were also evident in October 2003 but not in September 2004. Tillage increased soil in size classes <250 μm at 0- to 7-cm depth throughout the experiment although there were generally no differences in these classes at 7 to 20 cm. Shifts in soil from 2000- to 8000-μm size classes into smaller sizes following cultivation are also evident from statistical comparisons among size classes within treatments. Control plots generally contained greater soil in macroaggregate than microaggregate classes, particularly at 0 to 7 cm, whereas in tilled plots the 53- to 250-μm class contained amounts of soil frequently equal to that in the 250- to 2000-μm class and higher than that in the 2000- to 8000-μm class.

Particulate and Aggregate-associated Carbon

Initial cultivation had significant effects on the distribution of SOM in different particulate organic matter (POM) density fractions and soil depths (Table 2). On 24 ‘Aug. 2002 cultivation reduced SOM in clay fractions and reduced POM > 1.9 g cm^{-3} at 0- to 7-cm depth while at 7- to 20-cm depth cultivation increased all POM

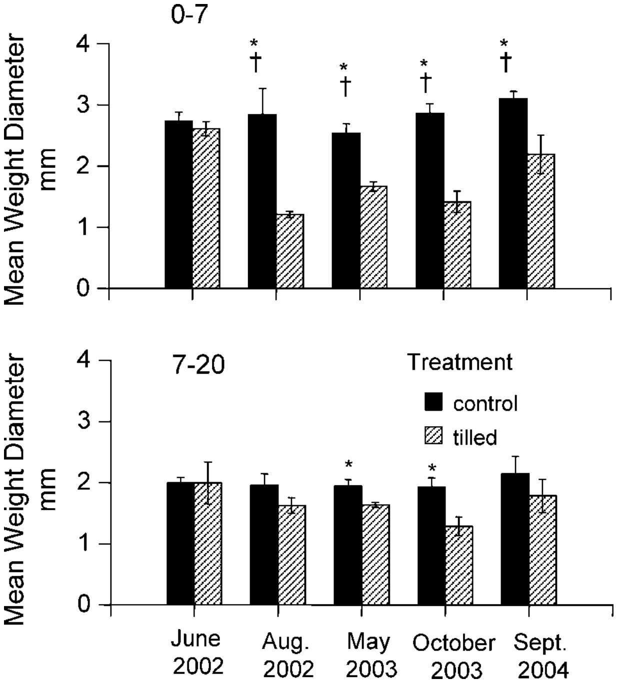

Fig. 1. Mean weight diameter (MWD) of soil aggregates at 0- to 7- and 7- to 20-cm soil depth. The June 2002 sampling was performed before cultivation. The May 2003 samples were taken before the second cultivation. Treatment means are shown ± standard error (n = 4). * Indicates statistically significant ($P < 0.05$) differences between control and tilled treatments within a single sampling date; † indicates significant differences between depths within a treatment and sampling date.

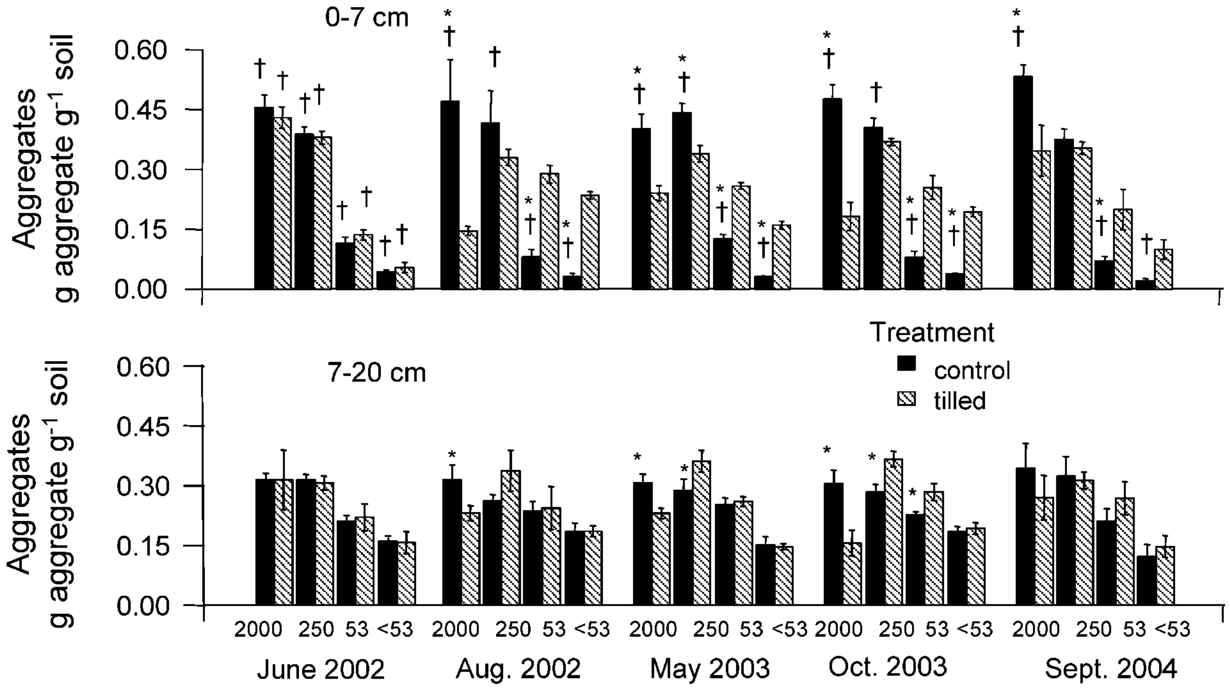

Fig. 2. Tillage effects on the distribution of soil in four aggregate size classes at 0- to 7- and 7- to 20-cm soil depth. Numbers along the *x* axis correspond with aggregate size: 2000 = 2000–8000 μm; 250 = 250–2000 μm; 53 = 53–250 μm; and <53 = <53 μm. The May 2003 samples were taken before the second cultivation. Treatment means are shown ± standard error ($n = 4$). * Indicates statistically significant ($P < 0.05$) differences between control and tilled treatments within a single sampling date; † indicates significant differences between depths within a treatment and sampling date. Size classes within a sampling date and treatment with different letters are significantly different.

classes and clay-associated SOM. Cultivation effects on POM distribution were similar in 2003 and 2004 although they were not as consistent across density fractions. Treatment differences were related to a homogenization of C availability at 0 to 20 cm due to cultivation. In control plots, there were significant differences in all POM fractions and clay associated C at all sampling dates after tillage (except the 1.3–1.6 g cm^{-3} in 2004); in contrast, the tilled plots had few differences in C concentrations between depths. At 0- to 20-cm POM increased in densities < 1.9 g cm^{-3} while there were no increases in the clay fractions or POM pools > 1.9 g cm^{-3} (Table 2).

Tillage changed the depth distribution of C associated with aggregate-size fractions (Fig. 3) and increased inter-aggregate light fraction C in the 2000- to 8000-μm class at 0 to 7 and 7 to 20 cm in 2002 and also in the 250- to 2000-μm class at 7 to 20 cm in 2002 (Fig. 4). In 2003, tillage increased inter-aggregate LF C in the 250- to 2000-μm class at 0 to 7 cm and LF C in the 2000- to 8000-μm class at 7 to 20 cm. Similar to whole soil C, there was a redistribution of inter-aggregate and intra-aggregate LF in the soil profile following cultivation. Intra-aggregate LF concentrations were generally indistinguishable among treatments; however, tillage increased intra-aggregate LF in the 250- to 2000-μm class in 2002 and the 2000- to 8000-μm class in 2003 at 7 to 20 cm (Fig. 5). The C/N ratios of several soil fractions at 0 to 7 cm were increased by tillage on 24 Aug. 2002 but on subsequent sample dates where

Table 2. Tillage and depth effects on the concentration of particulate organic matter (POM) in four density fractions and clay-associated C (<53 μm) on three sampling dates. Initial cultivation occurred on 25 June 2002 and again in the spring of 2003 and 2004 on the same plots.†

		24 Aug. 2002					21 Oct. 2003					24 Sept. 2004				
Depth		Clay	>1.9	1.6–1.9	1.3–1.6	<1.3	Clay	>1.9	1.6–1.9	1.3–1.6	<1.3	Clay	>1.9	1.6–1.9	1.3–1.6	<1.3
cm		mg C g^{-1} soil														
0–7	Control	21.43*†	8.06*†	0.91†	1.08†	0.64†	22.10†	5.66*†	1.47†	1.59†	0.85†	25.41*†	2.49†	1.33*†	1.32	0.75†
	Tilled	16.77†	4.43†	0.65	0.80	0.74	16.67†	3.21	0.87	1.61	0.84	13.77	1.83†	0.71	1.29	0.84
7–20	Control	8.67*	1.42*	0.23*	0.38*	0.20*	6.54*	3.31	0.16	0.56	0.35	11.12	0.56	0.25	0.35*	0.28
	Tilled	11.60	2.93	0.66	0.92	0.52	11.63	3.41	0.50	0.96	0.59	13.83	0.88	0.46	0.98	0.51
0–20	Control	12.35	3.34	0.42*	0.58*	0.33	11.02	3.98	0.53	0.85	0.49	14.83	1.05	0.53	0.61*	0.40*
	Tilled	13.01	3.41	0.65	0.88	0.59	13.38	3.34	0.62	1.16	0.68	13.37	1.19	0.54	1.08	0.62
		kg m^{-2}														
0–20	Control	3.16	0.86	0.11*	0.15*	0.84*	2.86	1.04	0.14	0.22	0.13	3.67	0.26	0.13	0.15*	0.10*
	Tilled	3.32	0.85	0.16	0.22	0.15	3.37	0.84	0.15	0.29	0.17	3.79	0.33	0.15	0.30	0.17

* Indicates significant differences between tillage treatments within a depth, POM fraction, and sampling date ($p < 0.05$).
† Indicates differences between depths within a treatment, POM fraction, and sampling date ($p < 0.05$).

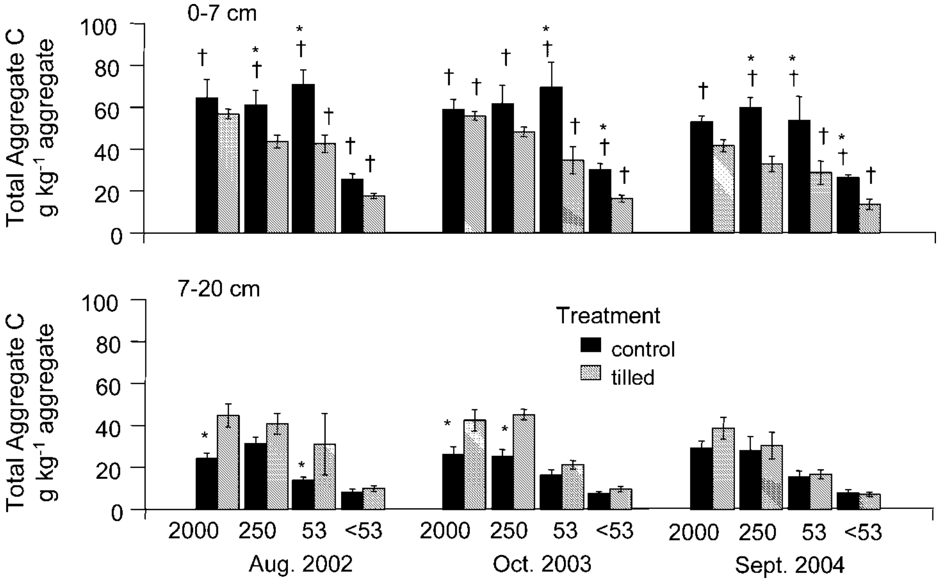

Fig. 3. Tillage effects on total sand-free aggregate C in 2002, 2003, and 2004. Initial cultivation was in June 2002. Treatment means are shown ± standard error ($n = 4$). * Indicates statistically significant ($P < 0.05$) differences between control and tilled treatments within a single sampling date; † indicates significant differences between depths within a treatment and sampling date. Size classes within a sampling date and treatment with different letters are significantly different.

there were differences between treatments, tilled plots had lower C/N ratios (Table 3).

DISCUSSION

Soil Aggregates

Changes in soil aggregate-size distribution were immediate and sustained following initial cultivation (Fig. 1). Within 60 d of cultivation, soil in the 2000- to 8000-μm size class at 0- to 7-cm depth (Fig. 2) declined to levels indistinguishable from those in an adjacent agricultural soil cultivated for more than 50 yr (19%; data not shown). Cultivation also reduced the amount of soil in this size class at 7- to 20-cm soil depth, demonstrating that declines at the soil surface were not solely due to a redistribution of aggregates to deeper in the profile.

Prior studies have demonstrated that long-term cultivation reduces soil aggregation (e.g., Gupta and Germida, 1988; Jastrow et al., 1996; Mikha and Rice, 2004). DeGryze et al. (2004) reported that at 0- to 7-cm soil depth, soil MWD in conventionally managed agricultural soils was approximately three-fold lower than in early succes-

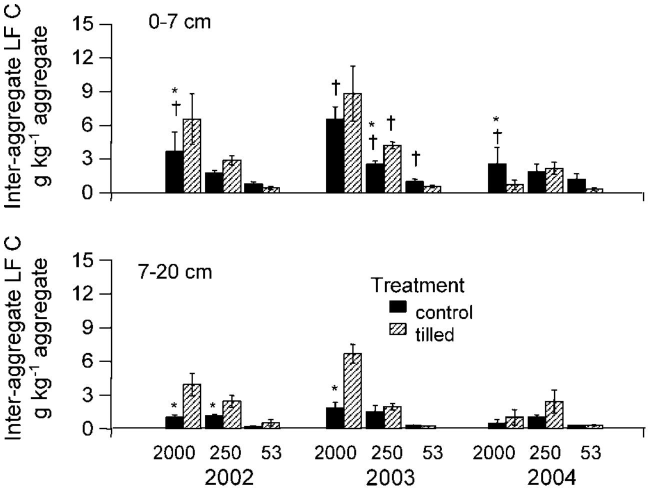

Fig. 4. Tillage effects on the distribution of sand-free inter-aggregate light fraction organic matter in 2002, 2003, and 2004. Initial cultivation was in June 2002. Treatment means are shown ± standard error ($n = 4$). * Indicates statistically significant ($P < 0.05$) differences between control and tilled treatments within a single sampling date; † indicates significant differences between depths within a treatment and sampling date. Size classes within a sampling date and treatment with different letters are significantly different.

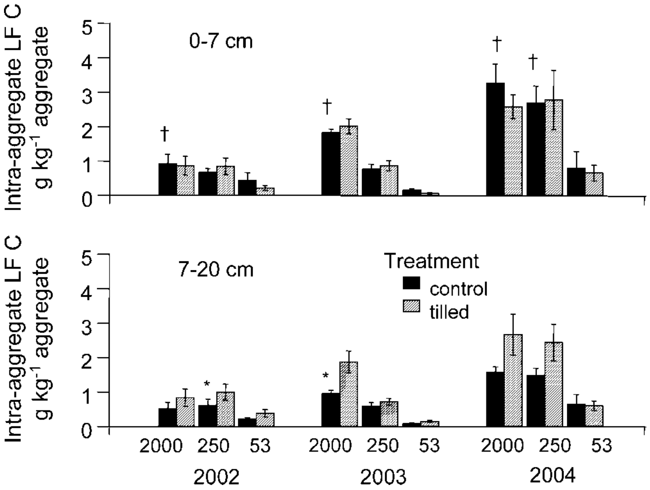

Fig. 5. Tillage effects on the distribution of sand-free intra-aggregate light fraction organic matter in 2002, 2003, and 2004. Initial cultivation was in June 2002. Treatment means are shown ± standard error ($n = 4$). * Indicates statistically significant ($P < 0.05$) differences between control and tilled treatments within a single sampling date; † indicates significant differences between depths within a treatment and sampling date. Size classes within a sampling date and treatment with different letters are significantly different.

Table 3. Tillage, depth and organic matter fraction effects on C/N ratios of different soil fractions in 2002, 2003, and 2004. Soil fractions include four aggregate size classes, interaggregate light fraction particulate organic matter (Inter-LF), intra-aggregate light fraction particulate organic matter (Intra-LF), whole soil particulate organic matter (WS POM) in four density fractions, and clay + silt associated C (clay). Initial cultivation occurred on 25 June 2002 and again in the spring of 2003 and 2004 on the same plots.†

		24 Aug. 2002				21 Oct. 2003				24 Sept. 2004			
		0–7		7–20		0–7		7–20		0–7		7–20	
Soil fraction		Cont	Till	Cont	Till	Cont	Till	Cont	Till	Cont	Till	Cont	Till
Whole soil		11.8	11.3	10.6	11.4	13.8	12.8	11.5	12.3	12.1	11.3	10.8	11.2
Aggregate size class	2000–8000	11.7	12.9	11.2	11.6	12.9	13.3	11.7	11.6	12.5	12.3	11.7	11.4
	250–2000	11.6	11.9	11.1	11.5	12.2	12.2	11.5	11.7	12.1	11.7	11.2	12.3
	53–250	11.5	11.8	10.0	10.9	12.2	11.7	10.1	10.9	11.2	10.6	9.7	9.9
	<53	8.7	9.8	7.6	7.9	10.3	10.1	7.9	9.0	10.3	9.6	8.1	8.3
Inter-LF	2000–8000	23.6*†	30.1	30.9	29.6	23.6†	25.3	29.5*	22.1	22.8†	23.6†	37.2*	29.2
	250–2000	21.8†	26.6	30.1	28.3	22.4†	21.2	31.9*	25.0	23.4	23.1	27.3	26.2
	53–250	24.0	22.4	29.0	25.6	20.6†	19.3	25.1	21.9	15.4†	18.2	20.6	18.3
Intra-LF	2000–8000	30.6	32.0	28.4	32.3	23.6†	24.0	29.7*	22.6	25.7	24.5	27.8	24.2
	250–2000	24.9*†	41.3	33.6	38.9	25.7†	27.1†	35.7	32.7	24.7	22.6	28.9*	23.7
	53–250	35.4	38.6	35.1	38.5	21.8†	25.5†	34.5	31.9	25.9†	30.5†	37.2	35.4
WS POM	Clay	10.7	10.5	9.5	10.2	11.5	11.3	7.8	9.9	11.2	9.9	10.2	10.3
	>1.9	13.2	11.5	15.5	12.4	21.6†	22.2	34.8*	22.8	15.6	16.1	14.7	15.9
	1.6–1.9	20.8†	22.0	27.4	26.1	16.1	15.8	17.8	16.4	18.4	19.6	20.2	18.1
	1.3–1.6	31.5*†	35.2	39.1*	31.0	23.1†	21.0	32.2*	25.7	23.2	22.7	27.3	22.9
	<1.3	25.7†	31.8	33.1	27.8	26.8†	26.4†	45.8*	39.6	26.2	23.5	29.4	25.6

*** Indicates significant differences between tillage treatments within a depth, soil fraction, and sampling date ($p < 0.05$).**
† Indicates differences between depths within a treatment, soil fraction, and sampling date ($p < 0.05$).

sional, afforested, and native forest ecosystems at our site. Six et al. (2000) showed that long-term cultivation reduced aggregation in four soils ranging in texture from sandy loam to silty clay loam. Our results show that tilling a never-previously cultivated soil immediately destroys macroaggregates and that this reduction persists throughout the growing season and following winter. Further, there is no measurable additional decline in macroaggregation following repeated tillage. Much of the decline in macroaggregation previously reported in long-term cultivated sites (e.g., Six et al., 2000; Grandy et al., 2002; Mikha and Rice, 2004) may have thus occurred almost immediately following initial cultivation, depending on soil type and cultivation intensity.

Carbon losses and changes in soil biological processes contribute to long-term declines in aggregation (Gupta and Germida, 1988; Grandy et al., 2002) following agricultural conversion, but short-term changes are likely driven by changes in plant communities and the direct effects of tillage (Angers and Caron, 1998; Six et al., 1999; Gale et al., 2000a). We minimized plant community change by leaving tilled sites fallow. As a result, control and tilled sites shared five of the top six dominant plant species (*Rubus accidentalis* was not a top biomass producer in tilled plots, data not shown). Changes in plant communities were thus relatively small compared with other studies where successional communities were converted to annual cropping systems (e.g., Tiessen and Stewart, 1983; Bowman et al., 1990). Fungal hyphae and fine-root networks contribute to macroaggregate stability (Bethlenfalvay and Barea, 1994). These networks are particularly susceptible to disruption following disturbance (Jansa et al., 2003) and their mechanical destruction during plowing and disking likely destabilized aggregates. Further, bare soil surfaces for 3 wk following tillage while plant communities recovered further exposed aggregates to the impacts of rainfall and made them more susceptible to slaking and dispersion following rapid wetting (Kooistra and van Noordwijk, 1996).

The persistence of cultivation effects on soil structure was evident before tillage in 2003 when aggregation in cultivated sites remained substantially lower (Fig. 1 and 2), despite the potential for root growth and freeze-thaw and wet-dry cycles to have affected aggregation in both tillage systems. Plant roots can increase aggregation by enmeshing small particles into stable macroaggregates; by supplying organic substrates such as root hairs, sloughed cells, and mucilage; and by influencing soil moisture content (Reid and Gross, 1981; Perfect et al., 1990). Freeze-thaw and wetting-drying cycles can directly break down aggregates but also influence soil structure by changing the proximity and alignment of mineral and organic particles to facilitate aggregation (Kemper and Rosenau, 1986; Denef et al., 2001). The direct and indirect destructive effects of tillage were greater than these other forces that form, stabilize, and destroy aggregates.

Soil Organic Matter

The vertical homogenization of soil C and N in the soil (Tables 1 and 2) following cultivation is consistent with results from other studies (Pierce et al., 1994; VandenBygaart and Kay, 2004). VandenBygaart and Kay (2004) studied the effects of moldboard plowing soils in three different textural classes (sandy clay loam, silty clay loam, and sandy loam) after 22 yr of no-till management. In each of these textural classes they found that soil C declined at 0 to 5 cm and increased at lower depths with the net result being no change in total soil C. Kettler et al. (2000) studied the effects of moldboard plowing a soil that had been in a no-till wheat–fallow system. Five years after plowing there remained differences in SOM stratification to 30 cm but no differences in total soil C. Pierce et al. (1994) found in Michigan that plowing fields in no-till for 6 to 7 yr redistributed C to 15-cm depth and that this effect persisted for 4 to 5 yr after plowing although there were no changes in total soil C. We similarly found changes in SOM stratification and that this homogenization occurred across SOM pools of different densities.

After 3 yr of annual tillage, we were still unable to detect differences in total soil C. Rates of SOM loss from temperate ecosystems immediately following cultivation have rarely been studied and the available data are inconclusive. Bowman et al. (1990), for example, found 40% organic C losses within 3 yr of cultivation (15.7 vs. 9.4 kg m^{-3}) and that between 3 and 20 yr after cultivation there was little additional C loss. These rapid rates of soil C change are unusual and may have been due to their sandy soil texture (Davidson and Ackerman, 1993; West and Post, 2002) as well as changes in vegetation that accompanied tillage. Substantial decreases in litter inputs associated with agricultural conversion likely contributed to the C losses as vegetation changed from short-grass steppe to an unfertilized wheat–fallow rotation. Tiessen and Stewart (1983) found no change in soil C concentration after 4 yr of cultivating a previously no-till silt loam soil. The high silt (49%) and clay (20%) contents of these soils may have increased their protective capacity and made them more resistant to the destructive effects of cultivation (Wander and Bidart, 2000).

Despite the lack of change in total soil C, we measured significant changes in POM pools and also in inter-aggregate SOM at 0- to 7- and 7- to 20-cm depths (Table 2; Fig. 2). Collectively, these changes, along with initial increases in C/N ratios of some organic fractions (Table 3), suggest increases in organic matter availability. In 2002, particulate organic matter quantity in the 0- to 20-cm soil layer increased in soil fractions < 1.9 g cm^{-3}, while in 2004 changes occurred in fractions < 1.6 g cm^{-3}. Light fraction pools have been shown to be correlated with soil surface respiration rates (Janzen et al., 1992; Alvarez and Alvarez, 2000) and its depletion represents a major portion of C loss in cultivated soils (Cambardella and Elliott, 1992, 1994). We measured no changes in clay fractions or in POM fractions with a density > 1.9 g cm^{-3}. Both these fractions are more chemically resistant to decomposition and more physically protected than LF (Golchin et al., 1994; Swanston et al., 2002).

Decreased physical protection of SOM is also suggested by the increases in inter-aggregate LF at both 0 to 7 at 7 to 20 cm. The biomass incorporated with tillage thus moved primarily into POM pools with low densities and inter-aggregate LF pools. In 2002, this aboveground C consisted of litter (142 ± 30 g C m^{-2}) and plant biomass (228 ± 11 g C m^{-2}). Transfer of aboveground and near-surface C to the 7- to 20-cm depth will additionally contribute to more rapid SOM turnover. For example, Lupwayi et al. (2004) found that incorporation of wheat litter into the soil with conventional tillage increased its decomposition rate by 48%. Similarly, Burgess et al. (2002) found that litter decomposition rates at 20 cm were 52 to 105% greater than those at the soil surface.

Implications for Soil Management

Marland et al. (2001) identified soil C permanence as the fundamental challenge to C sequestration. Although tillage is the major threat to stored soil C, most farmers for agronomic reasons periodically cultivate otherwise no-till fields. Rather than focusing on total soil C responses, which can be extremely hard to detect in short-term studies, we measured changes in the mechanisms underlying C and N cycling to infer potential long-term changes. We show immediate increases in unprotected C as well as homogenization of C in the soil profile. These changes drastically increased soil surface CO_2 emissions (1.0–1.9 g CO_2–C m^{-2} d^{-1} over three growing seasons) and microbial enzyme activity and over a period of time this will translate into detectable soil C losses (Grandy and Robertson, 2006). These results and recent research demonstrating that long-term no-till cropping can enhance environmental processes with no significant ecological or yield tradeoffs (Six et al., 2004; Grandy et al., 2006) highlight the need to maintain no-till management. Efforts should be made in specific regions to identify and overcome the agronomic limitations to no-till associated with different soil types.

CONCLUSIONS

We tilled a previously uncultivated field to investigate the immediate effects of tillage on aggregation and SOM dynamics. By choosing a previously undisturbed soil profile and leaving the site fallow following cultivation, we were able to isolate and measure the effects of cultivation without the confounding effects of changes in plant communities or prior cultivation. Our results include several changes in aggregation and organic matter availability that suggest immediate effects of tillage on destabilizing soil C and N pools.

1) Within 60 d, 2000- to 8000-μm aggregates declined from 0.47 to 0.15 g g^{-1}. These changes persisted over the winter and into the following spring when control plots had 0.40 g g^{-1} in the 2000- to 8000-μm size class and tilled plots had 0.24 g g^{-1};

2) In tilled plots, aggregates in the 2000- to 8000-μm size class declined to levels indistinguishable from those in adjacent agricultural fields tilled for >50 yr;

3) Total soil C changes were not detectable by the end of the third year but several lines of evidence suggest that tillage decreased SOM stability, including: (a) vertical homogenization of C; (b) increases in unprotected, inter-aggregate organic matter; (c) initial increases in C/N ratios of several SOM fractions; and (d) increases in POM with densities < 1.9 g cm^{-3}.

Our results demonstrate that aggregation, physical SOM protection, and the distribution soil C and N are immediately influenced by tillage, suggesting that no-till soils need to be continuously maintained to protect aggregation and physically stabilized C pools.

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