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REGULAR ARTICLE

# Soil aggregate and particulate C and N under corn rotations: responses to management and correlations with yield

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**Abstract**
*Background* Soil aggregate and particulate organic matter (POM) C and N provide valuable insight into C cycling and storage, and are sensitive to management, but effect of these pools on corn yield is unknown.
*Methods* Corn yield, N uptake, and aggregate and POM C and N at 0–5, 5–25 and 25–50 cm were measured and correlated in continuous corn (*Zea mays* L.) (CC), strip-till corn/soybean [*Glycine max* (L.) Merr.] (CS), and organically managed corn/soybean/wheat (*Triticum aestivum* L.) with green manure (CSW).
*Results* The POM differed only at 0–5 cm, where greater POM mass was found in CS than CC and CSW. Lower POM-C and POM-N was found in CSW than CC and CS. Overall, CSW had fewer macroaggregates (>250 μm) and associated C and N than CC and CS, but free silt and clay (<53 μm) and microaggregates (53–250 μm) were enriched in C and N in CSW. Yield and macroaggregate-occluded C and N were negatively correlated. Yield and 5–25 cm free silt and clay C were positively correlated.
*Conclusions* While organic matter in aggregate-occluded fractions is beneficial for soil C storage, it was correlated with lower grain yields, highlighting a potential tradeoff between yield and long-term C sequestration.

**Keywords** Carbon storage · Cropping systems · Soil organic matter dynamics

**Abbreviations**

| | |
|---|---|
| SOC | Soil organic carbon |
| SOM | Soil organic matter |
| POM | Particulate organic matter |
| cPOM | Coarse particulate organic matter |
| AONR | Agronomic optimum nitrogen rate |

Annual grain production, critical for global food production, lags in provision of ecosystem services (Foley et al. 2005). Seasonally bare soil creates opportunities for erosion and nutrient runoff, and combining lower biomass inputs with regular tillage reduces soil organic carbon (SOC) compared to perennial systems (Gentile et al. 2005; Sanford et al. 2012; Zhou et al. 2014; Cates et al. 2016). Farmers are increasingly called upon to introduce practices which increase SOC in order to maintain productivity and sequester atmospheric $CO_2$ (Post and Kwon 2000; Lal 2004). Reducing tillage, for example, may increase SOC-storing soil aggregates (Denef et al. 2004; Plaza et al. 2013) while diverse crop

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rotations provide more extensive ground cover throughout the year and may stimulate soil microbial life (McDaniel et al. 2014; Kallenbach et al. 2015; Raphael et al. 2016). However, farmers are more willing to adopt conservation practices which increase SOC while enhancing or maintaining productivity (Snapp et al. 2005; Arbuckle and Roesch-McNally 2015; Swinton et al. 2016), so it's necessary to establish that SOC accumulation will have a positive effect on yield.

Research in a variety of ecosystems suggests that soil aggregates are critical to long-term SOC protection, because aggregates render SOC physically inaccessible to decomposer microbes (Jastrow 1996; Six et al. 2004; Gunina and Kuzyakov 2014). Based on analysis of SOC in aggregate fractions, theory suggests that macroaggregates (>250 μm diameter) are held together by more recent plant inputs, roots and fungal hyphae, while microaggregates (53–250 μm) are held together by more persistent plant and microbial byproducts (Tisdall and Oades 1982; Plante et al. 2002; Kong and Six 2010). Occluded microaggregates are those formed within macroaggregates, and SOC in these occluded microaggregates is stable due to protection from microbial access and subsequent mineralization (Plaza et al. 2013). The silt and clay fraction of soil (<53 μm) also protects SOC from decomposition through interactions between soil organic matter (SOM) and mineral surfaces. These organo-mineral associations are observed to be extremely long-lasting and to occur primarily with microbially-derived SOM, but the exact nature of the binding and the rates and mechanisms for turnover of mineral-bound SOC are not well understood (Kleber et al. 2007; Grandy and Neff 2008; Rumpel et al. 2015).

In addition to accumulation in aggregates, SOC may build up as free, labile or particulate organic matter (POM) which usually reflects the quality and quantity of recently incorporated, large-size organic material into the soil (Marriott and Wander 2006). Higher levels of POM have been found in systems with high biomass inputs (Cates et al. 2016), under monoculture corn compared to rotations (Coulter et al. 2009), with manure (Mirsky et al. 2008) or compost (Yu et al. 2012) fertilization, so POM is expected to be sensitive to a wide variety of management practices. There is also evidence that the chemically labile C, measured as either permanganate-oxidizable C or mineralizable C, is directly related to corn growth (Culman et al. 2013). Labile or particulate C is likely to be fresh plant inputs preferentially and rapidly decomposed by microbes, releasing associated N and other nutrients for plant growth.

Although the supply of N mineralized from the various pools and fractions of SOM is an important part of growing crops in Corn Belt Mollisols (Reddy and Reddy 1993; Stevens et al. 2005), it is difficult to predict from these pools the precise quantity of mineralized N supplied to crops. The pre-sidedress nitrate test is a widely used assay that recommends sidedress N rates to growers based on post-emergence soil nitrate levels (Magdoff 1991). But measuring nitrate only evaluates the currently available mineral N, rather than the identifying and quantifying the pool of organic soil N which is likely to be mineralized in a given growing season, allowing the grower to apply a sufficient and economical rate of N fertilizer. Amino sugar N, anaerobically mineralized N, permanganate-oxidizable C and potentially mineralizable C and N have been proposed (Mulvaney et al. 2001; Sainz Rozas et al. 2008; Culman et al. 2013), but none has been widely proven and adopted for predicting soil N available to maize. Precipitation and temperature variation influence how much N is mineralized from SOM over the course of a growing season, enhancing the difficulty of predicting theorganic N contribution (Andraski and Bundy 2002; Schomberg et al. 2009). In addition, inaccessibility of aggregate SOM may slow the release of organic N to plant roots, reducing yields in highly aggregated soils, but this has not been studied and none of the common tests for organic soil N take aggregation into account.

We investigated whether physically fractionated SOM pools were correlated with corn production. Although soil fractionation was developed to illuminate soil structure and C dynamics, they may also inform our understanding of annual grain productivity. In addition, aggregation has been shown to vary with soil type and mineralogy (Denef et al. 2004; Han et al. 2016), and in order to make recommendations to farmers working with Mollisols, it is important to evaluate C and N fractions on this soil type. Our objectives in this work were to: 1) describe the aggregate and particulate soil C and N in a productive Mollisol under three annual grain systems and 2) investigate the correlations across rotations between physical OM fractions and productivity, including yield and N uptake with no N and yield with optimum N. We hypothesized that less frequent tillage would mean more aggregates and associated C and N, that rotational diversity would be associated with greater yield, POM and aggregate SOM, and thus, that yield

would be correlated with particulate and unaggregated C and N. In delving into these questions, we hope to shed light on the processes by which SOM contributes to crop yield.

## Materials and methods

### Site description

The study took place within the Wisconsin Integrated Cropping Systems Trial (WICST), which was established at the University of Wisconsin-Madison's Agricultural Research Station in Arlington, WI in 1990, on Plano silt loam (fine-silty, mixed, superactive, Mesic Typic Argiuodoll). A complete description of the trial and site can be found in Posner et al. (1995). The mean annual temperature at Arlington is 6.9 °C and mean annual precipitation is 898 mm (1981–2010, National Climate Data Center). During the 2013 growing season, when N rate trials were conducted, the mean annual temperature was 7.6 °C and the mean annual precipitation was 978 mm.

The WICST is a randomized complete block trial, with six cropping systems represented in all phases and four replications. A staggered start was performed so that replication occurred in time as well as space. Plots are 155.5 × 18.3 m, or 0.3 ha, and commercial farm-scale equipment is used for all field work. The trial was designed to compare systems in productivity and sustainability in the most realistic conditions over time, and so management (hybrids, fertilizer rates, equipment, etc.) has changed slightly to reflect evolution of best management practices. The three annual grain systems are: (1) continuous corn (CC), (2) two year corn-soybean rotation which is no-till during the soybean phase and strip-till during the corn phase (CS), and (3) organically managed three year rotation of corn-soybean-winter wheat followed by oat (*Avena sativa* L.) cover crop interseeded with berseem clover (*Trifolium alexandrinum* L.) (CSW). Beginning in 2008, composted pelletized chicken manure has been applied in the spring prior to corn in CSW. Corn in CC and CS receives commercial fertilizer in the form of urea at planting at UW-Extension recommended rates based on the pre-plant nitrate testing (Laboski and Peters 2012). The CC and CSW systems are chisel plowed in the fall prior to corn planting and the CS system is strip tilled in the spring immediately prior to corn planting. A chisel plow is also used prior to soybean planting in CSW, and soybeans in CS are planted using a no-till drill. Wheat in CSW is drilled in following soybean harvest and harvested, including straw, the following summer. Oats and berseem clover in CSW are drilled in following wheat harvest and chisel plowed under before the ground freezes that winter. Additional cultivation for weed control is used as needed in all phases of CSW while in CC and CS weeds are chemically controlled.

### Nitrogen rate trials

In order to establish indices of productivity including yield and N uptake at 0 kg N $ha^{-1}$ and at agronomic optimum N rate (AONR) for each system, we conducted N rate trials during the 2013 growing season. Management of the N rate trial was unique from the standard whole plot management as described above, in order to evaluate how each system, after 23 years of consistent agronomics, would respond under a common set of circumstances. The N rate trials were designed as a randomized complete block split plot conducted in the corn phase of each of the three grain systems, with the WICST systems as whole plot treatments, and N rate as a subplot treatment at levels of 0, 67, 106, 146, 185 or 224 kg N $ha^{-1}$. Although cover crop plowdown in CSW amounted to 76 kg N $ha^{-1}$, no pelletized chicken manure was applied in the N rate trial subplots, and soil nitrate in the spring of 2013 was not different among systems at 0–30 cm. Therefor we feel confident that the inorganic N represented the primary N input for each system. The hybrid FS53VT4 was planted 5 May 2013 in six N-rate plots in the SW corner of each corn plot of CC, CS, and CSW. In order to avoid previous research plots, 2013 N-rate plots in CC were 8.53 m by 11.6 m (0.016 ha), but in CS and CSW, plots were 4.57 m by 11.6 m (0.021 ha). Fertilizer N was applied at planting in the form of urea with a stabilizing coating of N-methyl-2-pyrrolidone and N-(n-butyl) thiophosphorictriamide (Agrotain®, Koch Agronomic Services, LLC, Wichita, KS). The center four rows of each six-row plot were harvested using a two-row combine on 23 October 2013 and yields are reported as 100% dry matter. Nitrogen content in corn biomass was measured using a Flash EA 1112 CN Automatic Elemental Analyzer (Thermo Finnigan, Milan, Italy).

### Particulate organic matter (POM)

Soil analysis for POM were described in Cates et al. (2016). Briefly, eight cores (2.4 cm diameter) per WICST plot were removed prior to corn planting or tillage spring 2013. Samples were taken only from the corn phase of the rotations in order to compare soil with different long-term management history which would be planted to the same crop. Cores were split into 0–5, 5–25 and 25–50 cm depths, and composited into a single sample per plot. The POM analysis was conducted to reflect updates to the method outlined in Marriott and Wander (2006). After soaking overnight in sodium hexametaphosphate, oven-dried and ground soil subsamples (10.0 g) were dispersed, shaken, and rinsed. Material >53 μm, POM, was retained. The POM concentration was determined by dividing the mass of POM by the mass of soil analyzed. Note that the 25–50 cm data were previously reported in Cates et al. (2016).

### Aggregate fractionation

Aggregate fractionation sampling and analysis were described in Cates et al. (2016). Samples were taken Oct 29 2012 after corn harvest and prior to fall tillage, avoiding wheel tracks. Six cores (5.2 cm diameter) per WICST plot were kept refrigerated until analysis, when they were separated into 0–5, 5–25, and 25–50 cm depth segments. Composited samples for each plot was sieved to 4 mm; clumps were gently broken up and large organic material was removed by hand.

The fractionation procedure followed that outlined by Six et al. (2002). Sequential wet-sieving as described by Elliott (1986) separated soil into macroaggregates (>250 μm), microaggregates (53–250 μm) and silt and clay particles (<53 μm). A subsample of 15.0 g oven-dried macroaggregates was used for further fractionation in the microaggregate isolator (built by David Sloan, UW-Madison Department of Soil Science, based on specifications from Six et al. (2002)). Occluded microaggregates (53–250 μm), occluded silt and clay (<53 μm) and coarse particulate organic matter (cPOM; >250 μm) were isolated from within the macroaggregates. All material was dried at 50–60 °C for a minimum of 24 h and then weighed and stored in sealed plastic bags. Recovery from the wet sieving process averaged 100%, and recovery from the macroaggregate isolation process averaged 99.6%.

### Carbon and nitrogen analysis

A subsample of 8–10 mg of each whole soil, aggregate fraction, and POM sample was analyzed for total C and N using a Flash EA 1112 CN Automatic Elemental Analyzer (Thermo Finnigan, Milan, Italy). Since inorganic C is negligible in these soils (Paul et al. 2001), organic C was assumed to be total C. The POM-N was determined by multiplying the concentration of N in POM by the g POM $kg^{-1}$ bulk soil. The POM-N:TN was determined by dividing the POM-N, g $kg^{-1}$ soil, by the total N, g $kg^{-1}$ soil. The POM-C and POM-C:SOC were calculated in the same manner. Aggregate C and N is reported as g C or N $kg^{-1}$ bulk soil. Since we did not measure bulk density at these depths, all C and N are reported on a concentration basis. However, previous work suggests that bulk density does not differ in these three systems at 0–10 cm or 0–25 cm (Sanford et al. 2012; Cates et al. 2016), so we believe that C and N concentrations are representative of the soil stocks.

### Statistical analysis

The AONR was determined for each system separately ($n = 4$) by fitting a linear plateau model to N rate trial data using PROC NLIN in SAS 9.2. Soil data was analyzed using PROC MIXED in SAS 9.2 (SAS Institute Inc.) to develop ANOVA statistics. Depth and system were treated as fixed effects while block and block by system interactions were treated as random effects. Covariance structures that resulted in the lowest Akaike and Bayesian information criterion values were selected. Depth was analyzed as a repeated measure (REPEATED) with a compound symmetry (TYPE = cs) covariance structure for POM, SOC, and TN measures and with a ante-dependence symmetry (TYPE = ante(1)) covariance structure for aggregate measures. Since significant differences in each soil measure were only of interest within depth, if there was a significant effect of treatment at a depth interval (using SLICE = depth, $P < 0.10$) then LSMEANS were used as a means separation test ($P < 0.10$). Correlations between soil data and yield data were explored using the pairs function in R (R Core Team, 2014), and significance of these correlations determined using the cor.test function. These correlations are tools to explore the relative strength of the relationships between SOM fractions and corn productivity, and so no adjustment for multiple comparisons was made.

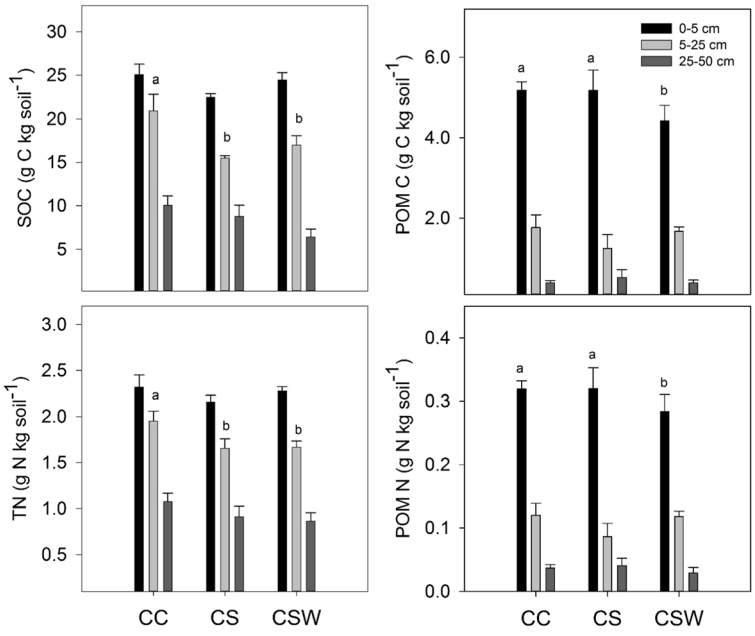



**Fig. 1** Whole soil C (SOC), total nitrogen (TN) and particulate organic matter (POM) C and N in three grain rotations at the Wisconsin Integrated Cropping Systems Trial: continuous corn, CC; strip-till corn/soybean, CS; organic corn/soybean/winter wheat, CSW. Error bars represent standard error of the mean and lowercase letters above bars indicates significant differences among systems at a given depth (LSD, $P < 0.10$). Data from 25 to 50 cm were previously reported in Cates et al. (2016)

## Results

### Soil organic matter measurements

The SOC and TN decreased with depth across systems (Fig. 1). At 5–25 cm, CC had greater SOC and TN than either CS or CSW. Systems differed in POM only at 0–5 cm, where greater total POM concentrations were found in CS (Fig. 2), while CSW had the least POM-C and POM-N (Fig. 1). The proportion of SOC and TN found in POM, and C:N ratios of both POM and whole soil all declined strongly with depth, but there was no significant difference among systems (Table 1).

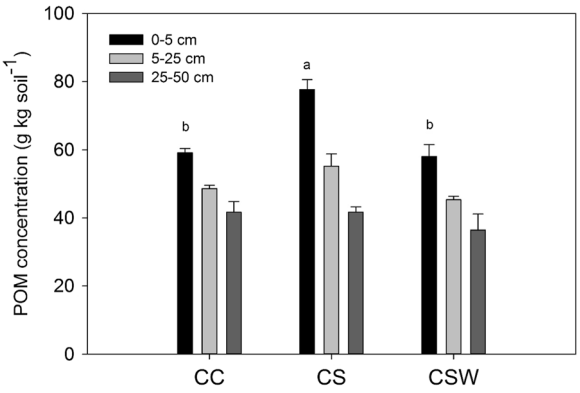



**Fig. 2** Concentration of particulate organic matter (POM) by depth in three grain cropping systems at the Wisconsin Integrated Cropping Systems Trial. *Error bars* represent standard error and *lowercase letters* denote significant differences among systems at a given depth: continuous corn, CC; strip-till corn/soybean CS; organic corn/soybean/winter wheat, CSW

Depth was a significant factor in all models of aggregate fraction C and N concentration. Broadly, soil at 0–5 and 5–25 cm had a greater proportion of macroaggregates, microaggregates, and occluded aggregate fractions, as well as higher concentrations of C and N in aggregates (Table 2). The proportion of soil found in free silt and clay was greater at lower depths, but the concentration of C and N associated with free silt and clay was greater nearer the surface. In many cases, the interaction between system and depth was a significant factor in the model, as C and N storage with depth clearly differed among the three systems (Table 2, Fig. 4). In CSW at 0–5 cm, less soil was found in macroaggregates and occluded components, but C and N within macroaggregates differed only in occluded microaggregate-C, which was depleted in CSW compared to CC (Fig. 3). At 5–25 cm, CS had less macroaggregate C and N. Microaggregate C and N differed among systems at all depths, with greater concentrations found in CSW except at 0–5 cm, where microaggregate-C was equivalent in CC and CSW. Silt and clay C varied among systems at all depths, with CSW having greater concentrations above 25 cm and lower concentrations below 25 cm. The CSW system also had greater concentrations of silt and clay N than CC above 25 cm (Fig. 4).

**Table 1** Treatment means and ANOVA *p*-values for particulate organic matter (POM) and whole soil C and N in three grain cropping systems at the Wisconsin Integrated Cropping Systems Trial (WICST). Lowercase letters indicate a significant difference across systems among depths, $P < 0.10$

| Treatment | POM g kg $^{-1}$ | POM-N | POM-C | POM C:N | POM-C: SOC | POM-N: TN | SOC g kg $^{-1}$ | TN | Whole C:N |
|---|---|---|---|---|---|---|---|---|---|
| System† | | | | | | | | | |
| CC | 49.8 | 0.159 | 2.445 | 16.0 | 0.11 | 0.078 | 18.7 | 1.78 | 12.0 |
| CS | 58.2 | 0.149 | 2.317 | 16.5 | 0.121 | 0.081 | 15.6 | 1.58 | 11.4 |
| CSW | 46.5 | 0.136 | 2.044 | 17.2 | 0.113 | 0.077 | 16.1 | 1.07 | 11.2 |
| Depth | | | | | | | | | |
| 0–5 cm | 64.9 a | 0.305 a | 4.889 a | 18.7 a | 0.208 a | 0.138 a | 24.1 a | 2.25 a | 12.5 a |
| 5–25 cm | 49.7 b | 0.106 b | 1.523 b | 16.7 b | 0.087 b | 0.060 b | 17.9 b | 1.76 b | 11.8 a |
| 25–50 cm | 39.9 c | 0.0335 c | 0.394 c | 14.4 c | 0.050 c | 0.037 c | 8.46 c | 0.951 c | 10.4 b |
| ANOVA | *P*-value | | | | | | | | |
| System (S) | 0.0003 | 0.3043 | 0.2620 | 0.2680 | 0.5169 | 0.5332 | 0.0486 | 0.0825 | 0.3412 |
| Depth (D) | <0.0001 | <0.0001 | <0.0001 | 0.0002 | <0.0001 | <0.0001 | <0.0001 | <0.0001 | 0.0005 |
| S x D | 0.0589 | 0.1373 | 0.2346 | 0.1377 | 0.2801 | 0.1747 | 0.0923 | 0.7435 | 0.2714 |

† CC, continuous corn; CS, corn/soybean; CSW, organic corn/soybean/winter wheat and oats/berseem clover

**Table 2** *P*-values of cropping system and soil depth effects from analysis of variance for aggregate weight, C and N distribution. See Figs 2 and 3 for evaluation of system effects at each depth

| | | System† | | Depth†† | | S x D |
|---|---|---|---|---|---|---|
| Fraction | Measurement | *P*-value | Treatment effects | *P*-value | Treatment effects | *P*-value |
| Macroaggregates | g g $^{-1}$ soil | 0.1527 | | 0.0005 | b = a > c | 0.0157 |
| | g kg $^{-1}$ C | 0.2174 | | 0.0001 | a = b > c | 0.0487 |
| | g kg $^{-1}$ N | 0.3084 | | 0.0001 | a = b > c | 0.0571 |
| Microaggregates | g g $^{-1}$ soil | 0.0001 | CSW > CC = CS | 0.0001 | a > b > c | 0.0001 |
| | g kg $^{-1}$ C | 0.0004 | CSW > CC > CS | 0.0016 | a > b > c | 0.2497 |
| | g kg $^{-1}$ N | 0.0002 | CSW > CC > CS | 0.0010 | a > b = c | 0.2124 |
| Silt & Clay | g g $^{-1}$ soil | 0.4293 | | 0.0140 | c > b = a | 0.0328 |
| | g kg $^{-1}$ C | 0.0613 | CSW > CC = CS | 0.0015 | b = a > c | 0.0089 |
| | g kg $^{-1}$ N | 0.039 | CSW > CC = CS | 0.0009 | b = a > c | 0.0092 |
| Occluded Microaggregates | g g $^{-1}$ soil | 0.0442 | CC > CS = CSW | 0.0001 | b = a > c | 0.0396 |
| | g kg $^{-1}$ C | 0.0270 | CC > CS | 0.0014 | b = a > c | 0.1809 |
| | g kg $^{-1}$ N | 0.0262 | CC > CSW = CS | 0.0010 | a = b > c | 0.1332 |
| Occluded Silt & Clay | g g $^{-1}$ soil | 0.0742 | CS > CSW | 0.0437 | b ≥ a > c | 0.0190 |
| | g kg $^{-1}$ C | 0.5505 | | 0.0019 | a = b > c | 0.1321 |
| | g kg $^{-1}$ N | 0.6680 | | 0.0029 | a = b > c | 0.6188 |
| Occluded Coarse POM | g g $^{-1}$ soil | 0.7378 | | 0.0074 | a > c = b | 0.0158 |
| | g kg $^{-1}$ C | 0.4681 | | 0.0032 | a > b = c | 0.2690 |
| | g kg $^{-1}$ N | 0.5524 | | 0.0045 | a > b = c | 0.1037 |

† CSW, organic corn/soy/wheat rotation; CC, continuous corn; CS, strip till corn/soy rotation

†† a, 0–5 cm; b, 5–25 cm; c, 25–50 cm

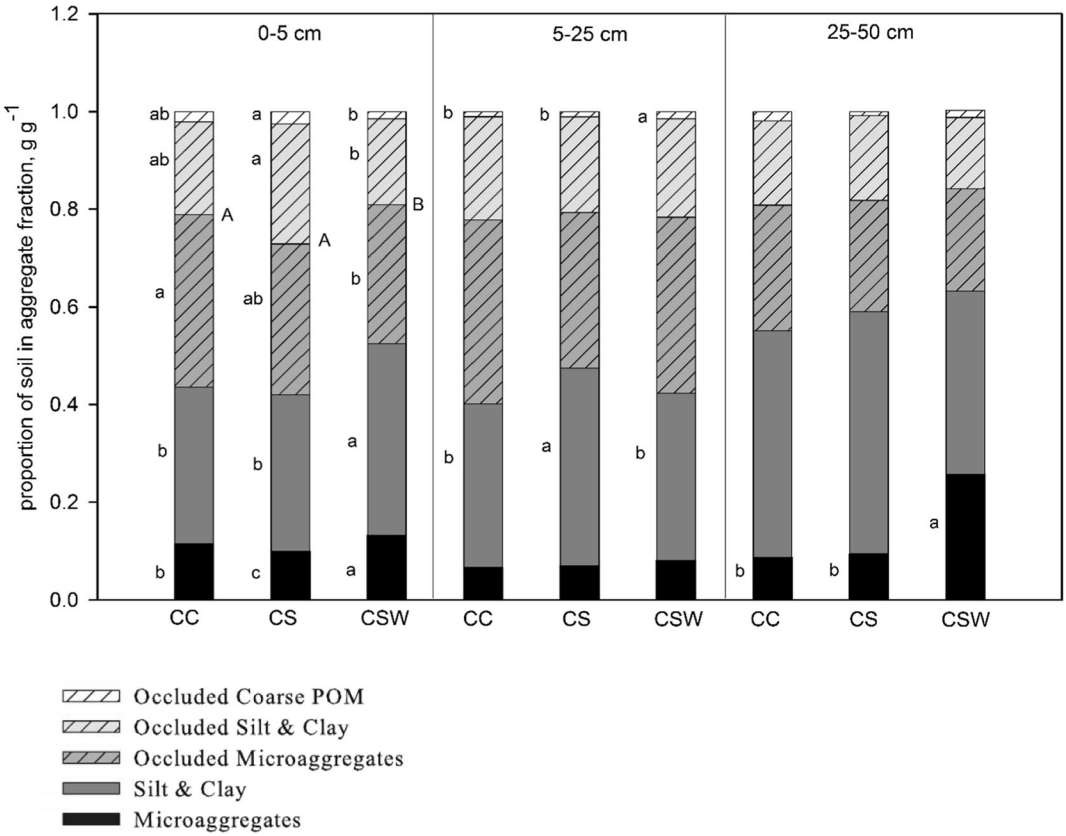



**Fig. 3** Soil aggregate distribution in three grain rotations at the Wisconsin Integrated Cropping Systems Trial: continuous corn, CC; strip-till corn/soybean, CS; organic corn/soybean/winter wheat, CSW. Bars representing fractions isolated from within macroaggregates include diagonal lines. Uppercase letters right of fractions represent a significant difference in macroaggregates among systems at a given depth. Lowercase letters left of fractions represent a significant difference among systems at a given depth for all other fractions

## Yield and correlations

When no fertilizer N was applied (0 N), there was a significant effect of system on yield according to the ANOVA ($P < 0.05$), but due to high variability, LS means were not different among systems (Table 3). At 0 N, plant N uptake was greater in CS and CSW than CC ($P < 0.06$). The AONR was greater in CSW and CS than CC ($P < 0.03$), and yield at the N rate closest to AONR was greater in CS and CSW than CC ($P < 0.03$, Fig. 5, Table 3). Plant N uptake at the N rate closest to AONR was not different among systems (Table 3) and so this was not included in correlation analysis.

We correlated SOM fractions to three metrics: N uptake at 0 N, yield at 0 N, and yield at the N rate closest to AONR. Broadly, more negative correlations were found with occluded SOM fractions and more positive correlations found with free silt and clay (Table 4). (For scatterplots of significant correlations, see Figure S1). Stronger correlations were found below the 5 cm depth. Patterns were driven by relationships across systems. Occluded microaggregate C and N at 5–25 cm were negatively correlated with yield at AONR, and occluded microaggregate weight was negatively correlated with N uptake at 0 N. Occluded silt and clay C and N at 5–25 cm were negatively correlated with N uptake and yield at 0 N, and occluded silt and clay C was negatively correlated with yield at AONR. Silt and clay C and N at 5–25 cm were positively correlated with yield at AONR and 0 N, but negatively correlated with yield at AONR at 25–50 cm. No significant correlations between yield or N uptake at 0 N and POM were found.

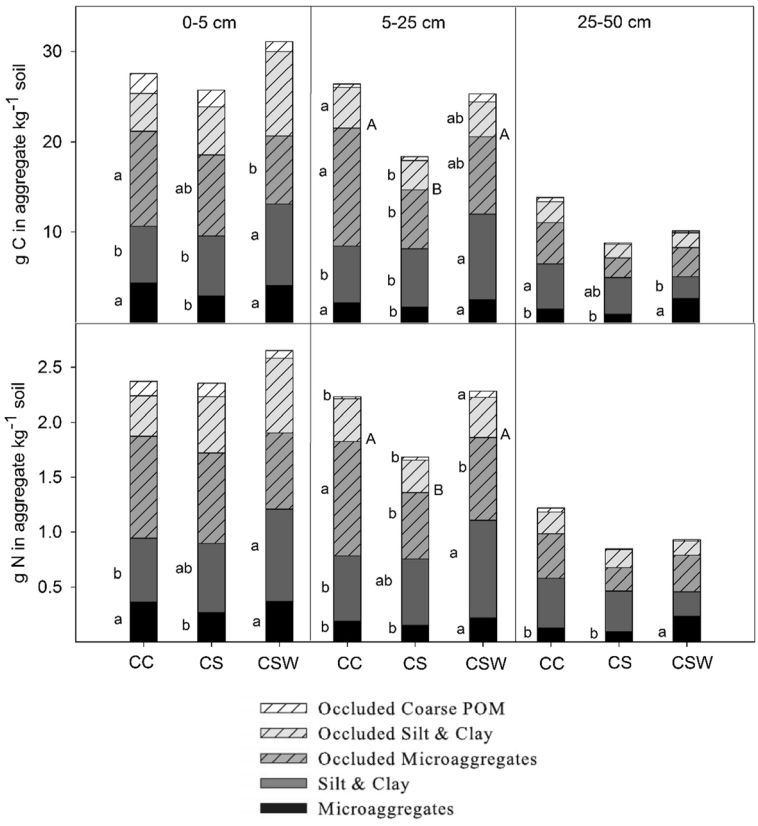



**Fig. 4** Concentration of soil C and N found in aggregate fractions of grain systems at the Wisconsin Integrated Cropping Systems Trial. Bars representing fractions isolated from within macroaggregates include diagonal lines. Uppercase letters right of fractions represent a significant difference in macroaggregates among systems at a given depth. Lowercase letters left of fractions represent a significant difference among systems at a given depth for all other fractions

## Discussion

Contrary to our hypothesis, tillage reduction was not associated with SOC accumulation in aggregates in the case of these loess-derived, deep, well-drained soils that comprise a large part of the U.S. Corn Belt. Aggregate theory predicts that a tillage gradient as studied here (in order of decreasing tillage intensity, CSW > CC > CS) should result in a gradient of aggregate distribution (Tisdall and Oades 1982; Six et al. 1999). For example, reducing tillage was found to increase aggregate mean weight diameter (Sheehy et al. 2015) and to increase the proportion of soil in macroaggregates (Singh et al. 2015). However, Grandy and Robertson (2006) found that a single tillage event on previously uncultivated soil decreased aggregate mean weight diameter for over a year. The CS system is strip-tilled every two years, so macroaggregates may not form in the intervening time. This is a good example of "reduced tillage" in a practical context, which may include occasional use of a chisel plow or field cultivator while maintaining high residue cover (Derpsch et al. 2014), leaving no opportunity to

**Table 3** Agronomic data (standard error in parentheses) from 2013 N rate trials in the Wisconsin Integrated Cropping System Trial. Agronomic optimum N rate (AONR) was calculated as the inflection point of linear plateau models (Fig. 5)

| System[†] | AONR<br>kg N ha$^{-1}$ | N rate closest to ANOR | Yield at N rate closest to AONR<br>Mg ha$^{-1}$ | Yield at 0 N | N uptake at N rate closest to AONR<br>kg N ha$^{-1}$ | N uptake at 0 N |
|---|---|---|---|---|---|---|
| CC | 153 (29.1) | 146 | 9.86 (0.35) | 5.21 (0.61) | 154 (17.4) | 71.9 (7.8) |
| CS | 99.2 (29.8) | 106 | 11.29 (0.24) | 7.50 (0.83) | 211 (54.5) | 134.2 (14.2) |
| CSW | 113 (19.9) | 106 | 11.96 (0.15) | 8.60 (1.34) | 176 (16.4) | 141.5 (22.8) |

[†] CC, continuous corn; CS, corn/soybean; CSW, organic corn/soybean/winter wheat and oats/berseem clover

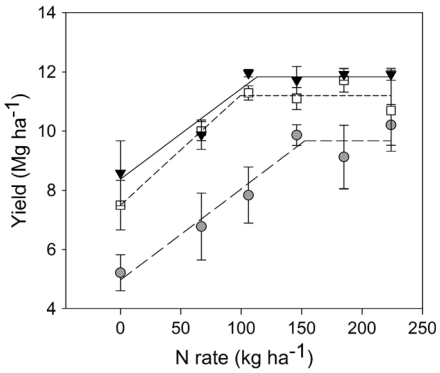



**Fig. 5** Yield of 2013 N rate trial plots at the Wisconsin Integrated Cropping Systems Trial. Gray circles, continuous corn; open squares, strip-till corn/soy; black triangles, organic corn/soy/wheat

build soil aggregates and subsequently SOC in annual grain rotations. However, our ability to isolate tillage effects was limited because rotating corn with soybean in the CS system decreases biomass C inputs compared to CC, which has a negative effect on SOC and aggregate and particulate C (Sanford et al. 2012; Brown et al. 2014; Cates et al. 2016). While previous work found slightly larger stocks of C and N in macroaggregates in CS, the difference between CS, CC and CSW was not statistically significant (Cates et al. 2016), and our more robust sampling in the present study (i.e. different depths and in a different year) confirmed that differences among the three grain systems were not based on a tillage gradient.

**Table 4** Correlations ($n$ = 11) between soil aggregate and particulate organic matter (POM) and 2013 yield and N uptake at 0 kg N ha$^{-1}$ (0 N) and N rate closest to the agronomic optimum N rate (AONR) determined by linear plateau model in the Wisconsin Integrated Cropping Systems Trial

| Fraction | Depth | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | 0–5 cm | | | 5–25 cm | | | 25–50 cm | | |
| | 0 N N uptake | 0 N yield | AONR yield | 0 N N uptake | 0 N yield | AONR yield | 0 N N uptake | 0 N yield | AONR yield |
| g kg$^{-1}$ soil | Correlation coefficient, r | | | | | | | | |
| POM | 0.27 | 0.08 | 0.22 | 0.23 | 0.10 | -0.12 | -0.05 | -0.04 | -0.19 |
| POM-C | -0.12 | -0.15 | -0.20 | -0.19 | -0.12 | -0.10 | 0.11 | 0.07 | 0.20 |
| POM-N | -0.12 | -0.13 | -0.22 | -0.19 | -0.11 | -0.09 | 0.00 | -0.02 | 0.03 |
| Macroaggregates | -0.26 | -0.36 | -0.33 | -0.26 | -0.19 | -0.37 | -0.03 | -0.04 | -0.18 |
| Macroaggregate C | -0.11 | -0.06 | -0.14 | -0.18 | -0.05 | -0.26 | -0.12 | -0.07 | -0.17 |
| Macroaggregate N | -0.15 | -0.13 | -0.17 | -0.15 | -0.05 | -0.15 | -0.12 | -0.07 | -0.18 |
| Microaggregates | -0.02 | 0.15 | 0.06 | 0.15 | 0.17 | 0.33 | 0.43 | 0.46 | 0.56† |
| Microaggregate C | -0.04 | 0.17 | -0.27 | -0.17 | -0.05 | -0.01 | 0.21 | 0.29 | 0.29 |
| Microaggregate N | 0.01 | 0.21 | -0.16 | -0.22 | -0.13 | -0.01 | 0.16 | 0.19 | 0.37 |
| Free Silt and Clay | 0.29 | 0.36 | 0.34 | 0.25 | 0.16 | 0.32 | -0.26 | -0.27 | -0.20 |
| Free Silt and Clay C | 0.02 | -0.17 | -0.02 | 0.46 | 0.52† | 0.58† | -0.31 | -0.23 | -0.58† |
| Free Silt and Clay N | 0.05 | -0.06 | 0.20 | 0.35 | 0.41 | 0.51 | -0.39 | -0.33 | -0.52 |
| Occluded Microaggregates | -0.53† | -0.54 | -0.52† | -0.14 | -0.04 | -0.27 | -0.02 | -0.28 | 0.01 |
| Occluded Microaggregate C | -0.20 | -0.22 | -0.43 | -0.51 | -0.39 | -0.56† | -0.13 | -0.08 | -0.18 |
| Occluded Microaggregate N | -0.23 | -0.21 | -0.37 | -0.49 | -0.38 | -0.55† | -0.16 | -0.12 | -0.18 |
| Occluded Silt and Clay | 0.20 | -0.01 | 0.11 | -0.41 | -0.43 | -0.49 | 0.07 | 0.02 | 0.01 |
| Occluded Silt and Clay C | 0.00 | -0.08 | 0.19 | -0.66* | -0.62* | -0.65* | -0.05 | -0.02 | -0.09 |
| Occluded Silt and Clay N | 0.12 | -0.06 | 0.14 | -0.57† | -0.53† | -0.52 | -0.08 | -0.11 | -0.03 |
| Occluded Coarse POM | -0.01 | -0.10 | -0.18 | 0.05 | 0.03 | 0.31 | -0.44 | -0.44 | -0.29 |
| Occluded Coarse POM C | 0.00 | 0.07 | -0.39 | -0.02 | -0.08 | 0.37 | -0.44 | -0.44 | -0.27 |
| Occluded Coarse POM N | 0.02 | 0.07 | -0.34 | 0.05 | -0.03 | 0.44 | -0.44 | -0.45 | -0.24 |

† Significant at the 0.10 probability level

*Significant at the 0.05 probability level

The CSW system yielded comparably to CS, so adding wheat and cover crops to CSW did not intensify the rotation effect on corn yield compared to a two-year rotation with soy (CS). In addition, CSW was enriched compared to CC and CS only in unaggregated C and N, suggesting that particulate and aggregate C and N were not accumulating in this system. Organic management, as practiced in the CSW system, requires the producer to "implement tillage and cultivation practices that maintain or improve the physical, chemical and biological condition of soil and minimize soil erosion" and refrain from using prohibited herbicides (U.S. Government Publishing Office 2016). While crop rotation commonly increases yield, and reduces disease and pest pressure (e.g. Bullock 1992; Munkholm et al. 2013; Farmaha et al. 2016), increased cultivation for weed control in place of herbicides is likely to increase mineralization of SOM and reduce aggregation, contrary to the above-stated goals of organic production. A complex rotation with cover crops may increase microbial activity and build SOC as microbial necromass accumulates (Kallenbach et al. 2015). However, in 2008, total microbial biomass to 20 cm was not significantly different among the three systems (Jokela et al. 2011). It is possible microbial biomass in the CSW system increased in the intervening time, as composted poultry manure has been applied in the corn and wheat phases of a three-year rotation since 2008 (Sanford et al. 2012; Cates et al. 2016). Although research on the decomposition of composted poultry manure is not extensive, Fauci and Dick (1994) found greater soil microbial biomass and enzyme activity with poultry manure application in a greenhouse experiment. A similar stimulation of soil biota by long-term poultry manure addition at WICST may partially explain the high yields in the CSW system.

Unaggregated C was positively related to yield at AONR, strengthening the argument that even when N is not limiting, SOM mineralization plays a part in supplying nutrients to corn growth. One potential mechanism for the relationship between yield and C on free silt and clay is that the silt and clay C and is indicative of microbial by-products and activity (Grandy and Neff 2008; Plaza et al. 2013; Cotrufo et al. 2015). It follows that we found negative correlations between yield and occluded microaggregate C and N, which has been shown to be largely plant-derived (Plaza et al. 2013). Perhaps slightly less soil aggregation in the intensively tilled CSW, where the highest free silt and clay C and N were found, allows for more access for microbes to mineralize SOM and provide N to the crop. Although Wood et al. (2016) found a negative relationships between yield and mineral-associated C on an African Oxisol, this could be due to differences in mineralogy. Soil C has been shown to interact differently and turn over at different rates in illite, kaolinite, and smectite clays, but the specific mechanisms for these phenomena have not been established (Wattel-Koekkoek et al., 2003; Denef and Six 2006; Han et al. 2016).

Lastly, although free, labile POM is the base of the supply for more processed and stabilized SOM (Gunina and Kuzyakov 2014), it was uncorrelated to yields for the year, suggesting it was of limited utility in describing soil fertility relevant to the current growing season. Decomposition of POM results in transformation of POM-C into $CO_2$, microbial biomass and other SOM fractions prior to release of plant-available nutrients (Lehmann and Kleber 2015). The greater C:N ratio of POM compared to whole soil indicates that decomposition of POM is in the early stages, similar to POM from an Iowa Mollisol measured by Brown et al. (2014). The POM analysis measures a pool of SOM, not the rate at which it is transformed to plant-available nutrients and perhaps the delay between pre-plant sampling for POM and crop demand was too great to evaluate a relationship. Previous work on an Alfisol in Michigan found that soil $NO_3$-N and N mineralization were significantly correlated with corn yield at V10 but not pre-plant sampling, and that permanganate-oxidizable C was never significantly correlated with yield (Culman et al. 2013). In contrast, Wood et al. (2016) found that POM-C from mid-season soil samples was positively related to corn yield on African Oxisols, which could be due to later sampling time or a greater agronomic impact of POM-C where decomposition is more rapid.

More macroaggregates and greater occluded microaggregate C in CC and CS are likely beneficial for long-term soil C accumulation as well as protection from erosion and increased water infiltration (Six and Paustian 2014; Tiemann and Grandy 2014; Castellano et al. 2015). However, macroaggregates were not correlated with yield, and occluded microaggregates and associated C and N were negatively correlated with yield, which might make farmers and land managers dubious of practices that increase formation of macroaggregates. Crop rotation in this study resulted in higher yields at all N rates, but did not result in soil with greater

total SOC or aggregate protected C. The common ground for researchers and land managers is that decreased SOC causes structural decline and increased erosivity (Oades 1984; Rhoton et al. 2002), so all parties benefit from maintaining SOC in agricultural soils. It is unclear whether there is a critical level of SOC below which productivity will be economically affected and soils cannot recover (Loveland 2003), and whether some metrics of SOC, particularly unprotected or active SOC, may be of greater importance to productivity (Culman et al. 2013). Researchers must consider the practical implications of their work and continue to work to develop metrics and predictors of short-term productivity as well as long-term SOC stabilization.

## Conclusion

We found that strip-tilled soil in CS did not have greater aggregate C and N, but CSW, an organic rotation relying heavily on tillage, had less POM-C and POM-N and greater free silt and clay and microaggregate C and N. Corn yields and associated AONR were significantly greater in CSW and CS than CC. In addition, positive correlations were observed between yield and free silt and clay C, and negative correlations were observed between yield and occluded microaggregate C and N, suggesting that occlusion of SOM in microaggregates is not beneficial to crops. The lack of correlation between POM and yield suggests that POM, recently incorporated SOM, may not be a relevant pool of SOM for nutrient release on an agronomic timescale. In order to make pertinent recommendations about SOC accumulation in agroecosystems, soil scientists and ecologists should work together with agronomists to develop more applicable metrics of SOM which assist growers in predicting their yields. In addition, potential declines in productivity must be considered when recommending farming practices to build SOC, because benefits to society of farming practices that promote SOC accumulation are weighed against the economic sustainability of farm businesses.

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