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Improvement of soil structure through organic crop management, conservation tillage and grass-clover ley

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Abstract

Conventional tillage is a widespread soil management practice that controls weeds and promotes nutrient mineralization at the expense of a degraded soil structure and soil carbon (C) loss. Although C dynamics and soil structure are widely recognized as pivotal to essential environmental and crop-related agroecosystem processes such as belowground C storage and crop root establishment, there is still a need to evaluate cropping practices most favorable for soil structure. For example, the effects on soil structure by continuous intensive tillage after a ley period remains unclear. To address these issues, we measured mean weight diameter, total C and total nitrogen (N) in whole soil and water-stable aggregate fractions after a 4-year arable crop rotation on a Cambisol where organic and conventional management was combined with intensive tillage and different types of conservation tillage. Measurements were repeated following a 2-year grass-clover ley period. Results showed that 4 years of organic management (including the application of cattle manure slurry) combined with reduced tillage significantly improved soil structure through increasing the proportion of large macroaggregates and hence the aggregate mean weight diameter (MWD) in the 0-6 cm soil layer. Although an increase in MWD after ley was observed in organic intensive tillage and a marginal increase in conventional intensive tillage, a significant increase in total C was observed only for the organic cropping systems, which also showed a high C stratification between 0-6 cm and 6-20 cm depth. Thus, a ley period enhances soil structure after continuous cropping under intensive tillage and when organic management is combined with reduced tillage. In conclusion, soil structure is best maintained when combining organic management with reduced tillage due to additive effects.

1 Introduction

The impacts of tillage and fertilization extend well beyond crop productivity; they influence soil microbial activity (Lori et al., 2017; Pérez-Piqueres et al., 2006; Zuber and Villamil, 2016), greenhouse gas emissions (Stavi and Lal, 2013), soil structure and C sequestration (Gattinger et al., 2012; Guo and Gifford, 2002). Widespread and long-term use of intensive tillage, prevalent in high yield cropping systems, has led to significant soil degradation with a concomitant loss in nutrients and organic matter; all of these effects limit current-day productivity and threatens future farming potential. This situation is particularly pressing in Europe, where there is a high degree of mechanization and limited land for agriculture. Although organic agriculture can benefit the environment in comparison with conventional crop management (Geiger et al., 2010; Gomiero et al., 2011), it is reliant on intensive tillage for weed control (Armengot et al., 2013).

Sustainable agricultural intensification has been touted as an alternative to reconcile productivity with environmental sustainability (Govers et al., 2017; Rockström et al., 2017). In arable systems, it includes organic management and conservation tillage practices (Hobbs et al., 2008). These improved variations of tillage (no till, chisel, disk, sweep till types of reduced tillage), alone or combined, have shown to be effective in adapting to environmental demands (Lal, 2009) by, for example reducing soil erosion (Lynch, 2012).

Soil structure controls the movement of water, solutes, microorganisms, gases and plant roots, which are necessary components to maintain soil functions (Bronick and Lal, 2005; Carter, 2002; Nicolodi and Gianello, 2014). Soil aggregation is often used as a measure for soil structure (Six et al., 2000a) despite ongoing difficulties in defining critical limits (Carter, 2002). The formation of aggregates, i.e., water-stable soil size classes (Elliott, 1986) with intrinsic varying physical and chemical characteristics, is essential to soil development and fosters C stabilization through physically protecting C from mineralization by microorganisms (Balesdent et al., 2000; Schimel and Schaeffer, 2012). During the formation of aggregates, intra-aggregate organic matter is incorporated and stabilized through physical protection (Six et al., 2004a). Considering that physical access to occluded substrates is the limiting process factor for organic matter breakdown in mineral soils (Jastrow, 1996; Schimel and Schaeffer, 2012; Tisdall and Oades, 1982), protected stabilized SOM can contribute to longer-term soil C sequestration (Kong et al., 2005). Aggregate stability is often lower under intensive tillage (Elliott, 1986; Kravchenko et al., 2011; Six et al., 2000b), encouraging a loss of C-rich macroaggregates while increasing C-poor microaggregates (Six et al., 2000a). Increasing SOM is sought, since it improves soil quality through altering nutrient availability, water holding capacity, soil porosity, cation exchange capacity and soil aggregation (Bronick and Lal, 2005; Carter, 2002; Kaiser et al., 2008; Rawls et al., 2003). Given the multiple advantages of good soil structure in supporting fundamental systems processes, management practices that foster aggregate formation should be encouraged.

In this study, we used the Swiss farming system and tillage experiment (FAST) to quantify the effect of four different cropping systems: conventional intensive tillage, conventional no tillage, organic intensive tillage and organic reduced tillage after a 4-year arable crop rotation on soil aggregate stability and aggregate-associated C and N storage. These same parameters were evaluated following a 2-year grass-clover ley period to test whether a temporary grass-clover pasture can compensate for the negative effects of intensive tillage on soil structure in comparison to systems with no- or reduced tillage. We hypothesized that reduced tillage and no-tillage would improve soil structure by reducing soil physical disturbance in comparison with intensive tillage, resulting in a higher total carbon (TC) content in comparison to intensive tillage. Similarly, we hypothesized that organic management would result in better soil structure than conventional management given that C-rich cattle slurry was used as fertilizer. Finally, that the use of a 2-year grass-clover ley would increase C content and improve soil structure overall due to the combination of no physical disruption and particle binding action of the plant roots and their exudates.

2 Methods

2.1 Field site and experimental design

The Swiss Farming System and Tillage experiment (FAST), described in detail by Wittwer et al. (2017), compares conventional and organic management strategies with different tillage intensities in 6-year rotations. The field experiment is located at the Swiss federal research station Agroscope, Reckenholz near Zurich, Switzerland (47°26’20”N, 8°31’40”E). The soil is a Cambisol on glacially deposited Pleistocene sediments with a loamy texture (23 % clay, 34 % silt, 43 % sand). Mean annual temperature is 9.4°C (Swissmeteo), while annual precipitation averages 1054 mm (1981-2010 data).

The crops rotating in the first 4 years are representative of local Swiss farming practices. The rotation started with winter wheat (*Triticum aestivum* L. cv. Titlis), followed by maize (*Zea mays* cv. Padrino), field bean (*Phaseolus vulgaris* cv. Fuego) and winter wheat (*Triticum aestivum* L. cv. Titlis). Finally, a grass-clover mixture (UFA 330) was sowed for a 2-year period. FAST consists of two identical field trials that are staggered one year. The first experiment (FAST I) started in August 2009 and the second (FAST II) started in August 2010. Both trials are set-up in a split-plot design with four cropping systems as main plots (6 m x 30 m) and four cover crop treatments as subplots (3 m x 15 m). The main plots are set-up as a randomized complete block design replicated four times. For this study, we only sampled FAST I at the cropping system (main plot) level. The factor cover crop was not considered and samples were taken in the cropping system without cover crop (“no cover crop”).

The farming practices tested are management type (i.e., conventional and organic) and tillage (i.e., intensive and conservation tillage. The combination of both factors resulted in four main investigated cropping systems: conventional intensive tillage (C-IT), conventional no tillage (C-NT), organic intensive tillage (O-IT), and organic reduced tillage (O-RT). Intensive tillage was applied to 0.2 m depth using a moldboard plow (Menzi B. Schnyder, Brutten, Switzerland) followed by seedbed preparation at 0.05 m with a rotary harrow (Amazone, H. Dreyer GmbH, Germany) for both conventional and organic management. Conservation tillage consisted of no tillage and direct seeding under conventional management whereas in organic plots were superficially tilled at 0.05 m depth with a disk harrow in the first year and a rotary harrow thereafter.

Both organic cropping systems were fertilized with cattle manure slurry whereof on the average 40 % was in the form of NH_4-N, and the rest organic N. This was distributed among the wheat crops (107 kg N ha^{-1} yr^{-1} in two applications), maize (137 kg N ha^{-1} yr^{-1} in two applications), and the grass-clover ley (205 kg N ha^{-1} $year^{-1}$ in four applications). Conventional fertilization consisted of ammonium-nitrate applications with an input of 110 kg N ha^{-1} yr^{-1} for the wheat crops, 90 kg N ha^{-1} yr^{-1} for maize, and for the grass-clover ley 130 kg N ha^{-1} the first year and 100 kg N ha^{-1} the second year. The grass-clover was harvested five times the first year and four times the second year. Fertilizer applications occurred after each cut.

2.2 Soil sampling

Soil was sampled at the end of the fourth growing season of FAST I (August 2013 after wheat harvest), as well as immediately following two years of grass-clover ley (August 2015). Four intact soil cores (5.5 cm x 20 cm) were taken at 3 m intervals from the center of each replicate plot using a Giddings hand sampler (Giddings Machinery Co, Windsor, Colorado, USA). Each 20 cm-length core was manually cut at 6 cm, separating the top 0-6 cm from the bottom 6-20 cm. All cores were kept cool during transport. Field-moist cores were sieved at 8 mm by manually crumbling along natural fracture lines in order to minimize aggregate disruption. The four cores from each plot were combined and each composite sample was air-dried and stored at room temperature.

2.3 Physical fractionation of soil aggregates

Air-dried whole soil was wet-sieved following Elliott (1986) to separate into four aggregate size classes: large macroaggregates (LM; > 2000 μm), small macroaggregates (SM; 2000-250 μm), microaggregates (mi; 250-53 μm) and silt and clay (S+C; < 53 μm). Eighty grams of air-dried soil was distributed evenly on a 2000 μm sieve and submerged in deionized water for 5 minutes for slaking. Then, the sieve was manually raised and lowered rhythmically 50 times over the course of two minutes. The water-stable aggregates remaining on the sieve (i.e. macroaggregates) were backwashed into a tin and oven dried at 60°C, while the remaining soil-water mix was poured over the next smaller 250 μm sieve and the procedure was repeated; also for the other two size classes. Mean weight diameter (MWD), used as a measure of soil structure, was calculated using the proportional abundance of each aggregate fraction and the mean diameter of each size class as defined in equation [1].

$$MWD = \sum_{i=1}^{n} \chi_i \omega_i \quad [1]$$

Where χ_i is the mean diameter of the particle range of each size class, ω_i is the weighted abundance of each aggregate fraction in whole soil and n is the number of aggregate size classes used.

Occluded microaggregates within macroaggregates were separated from the 0-6 cm layer according to Six et al. (2000a). This layer was chosen considering that its richer microbial composition is more responsive to change, and surface proximity increases the impact of management practices compared to the 6-20 cm layer. A total of 15 g of large and small macroaggregates were mixed in similar proportion as their occurrence in whole soil. After slaking in deionized water for 20 minutes, they were shaken atop a 250 μm metal mesh on a reciprocal shaker at 150 rpm together with fifty 4 mm diameter metal balls under a constant flow of deionized water. A clear outflow stream indicated the disruption of all soil macroaggregates. The outflow tube was placed atop a 53 μm sieve positioned over a basin. Subsequent sieving was performed as described previously for the separation of water-stable microaggregates from silt and clay. The mass of all soil fractions collected was recorded after oven drying at 60°C in pre-weighed aluminum tins.

2.4 Carbon and nitrogen quantification

For each bulk soil and fraction sample, approximately 2 g of dry soil was finely ground and subsampled for total carbon (TC) and total nitrogen (TN) determination by combustion on an elemental analyzer (LECO Corporation, United States). TC and TN of the entire 0-20 cm soil profile was calculated using weighted C and N amounts per soil layer. Annual C accumulation rates per year at 0-6 cm and 6-20 cm depth were calculated as the difference in C concentration between the end and start of the grass-clover period divided by 2 (duration of grass-clover period, in years).

2.5 Statistical analyses

All analyses and figures were performed in R 3.3.2 (R Development Core Team, 2016). Linear mixed effect models with restricted maximum likelihood were used to estimate effect size differences in MWD, TC, TN and fraction proportions using R package lmer4 (Bates et al. 2014). Block, cropping system, depth and year (i.e. before and after the 2-year grass-clover ley) were set as fixed effects, whereas a varying intercept was fit for the random effects of treatment within block and treatment within block and year. The data were log-transformed when visual inspection of residual plots revealed deviations from homoscedasticity or normality. To facilitate comparison, figure means and standard deviations presented are based on untransformed data. Type III ANOVA with Satterthwaite degrees of freedom approximation were calculated using the afex package (Lenth, 2016) and followed by Tukey familywise adjustment of least squares means post-hoc testing. Statistical significance was tested at $p<0.05$. Management type (organic vs. conventional) differentiated using orthogonal contrasts are noted as “M”. Similarly, differences within a cropping system between depths (identified throughout figures and tables with “D”) and between years (denoted in figures and tables with “Y”) were tested with custom contrasts using least squares means with Tukey multiple comparison correction.

3 Results

3.1 Soil aggregation

The distribution of aggregate fractions indicates a generally highly aggregated soil at the FAST trial, where macroagreggrates were the dominant fraction overall, accounting for between 68 % and 79 % of whole soil across both depths and years sampled (Table 1). The proportions of large macroaggregates (LM) and small macroaggregates (SM) showed significant interactions between treatment, depth and year. After the 4-year crop rotation, O-RT had significantly more LM compared to the other cropping systems while the opposite was true for SM abundance at 0-6 cm depth (Table 1). Mean weight diameter (MWD) followed the same trend in LM at both depths: O-RT had a significantly greater MWD than all other cropping systems after the 4-year rotation (Figure 1). In the top soil layer, the MWD decreased in the order: O-RT (1417 μm) > C-NT (1155 μm) > O-IT (1139 μm) > CIT (1053 μm). Orthogonal contrasts between management type (O-RT + O-IT vs. C-NT + C-IT) indicated that organic cropping systems contained significantly more LM ($p<0.001$) while at the same time significantly less SM ($p<0.05$) than the conventionally managed plots in the 6-20 cm depth. Among all cropping systems, macroaggregates at 0-6 cm depth were formed predominantly by occluded microaggregates (58 % – 62 %) and to a significantly lesser degree, occluded silt and clay (14 % – 18 %). The proportion of these fractions was, however, unaffected by cropping system (data not shown).

Following the 2-year grass-clover ley, the differences in LM and SM proportion abundance within cropping systems were no longer significant because of a general increase in LM; nevertheless, C-IT showed a significant increase in LM at both depths and O-IT only at 0-6 cm depth, when compared to corresponding values before the ley. This coupled with a corresponding significant decrease in SM. Likewise, significant decreases in SM proportion compared to its former state at the end of the crop rotation were observed for C-NT at both depths and for O-RT at 6-20 cm depth. Hence, the effect of the grass-clover ley was evident in the shift of dominating fraction at 0-6 cm depth in all but O-RT, which already had a higher proportion of LM after the crop rotation. The remaining mi and S+C aggregate fractions both followed a similar trend across cropping systems, depth and year: those under conventional management (C-IT and C-NT) had an increased proportion, significant at 6-20 cm depth, in comparison with the organic management (O-RT and O-IT, Table 1). These changes in aggregate proportion were reflected in the trend of MWD; O-IT increased significantly and C-IT increased marginally ($p<0.06$) the MWD at 0-6 cm depth compared to the start of the 2-year ley (Figure 1).

3.2 Total C and total N concentrations

With the exception of O-RT at 6-20 cm depth after the grass-clover period, the mi fraction consistently held significantly ($p<0.05$) less TC and TN per gram aggregate fraction compared to all other fractions within cropping systems at both depths and years. However, no other significant trends were found regarding within-fraction dynamics of C and N.

Significant interactions were found between cropping system x depth x fraction, cropping system x year x fraction and cropping system x year x depth x fraction for aggregate TC and TN contributions to the whole soil level (g aggregate-C kg^{-1} whole soil) (Table 2). After the 4-year crop rotation, the LM fraction contributed significantly more TC and TN to whole soil in the O-RT treatment (11.5 g C kg^{-1} and 1.3 g N kg^{-1}), whereas for the other cropping systems it was the SM fraction (between 7.1 - 8.5 g C kg^{-1} and 0.8 - 0.9 g N kg^{-1}) or the SM together with the LM fraction (between 5.3 - 6.2 g C kg^{-1} and 0.5 - 0.8 g N kg^{-1}) at 0-6 cm depth (Figures 2 & 3). After the 4-year crop rotation, all cropping systems had significantly higher TC and TN in the occluded mi than in the occluded S+C fraction at 0-6 cm depth (Table 3). Occluded S+C within macroaggregates contained 1.9 % and 13.4 % more TC than free S+C for C-IT and C-NT, respectively, but 14 % and 5 % less TC than free S+C in O-IT and O-RT, respectively.

The 2-year grass-clover ley increased TC associated with LM in C-IT and O-IT, while TN increased for C-NT and O-RT. SM contribution to whole soil TC and TN decreased in all but the O-RT at 0-6 cm and O-IT at 6-20 cm depth (Fig. 2). Significant increases of TC were observed for S+C after the ley period across all cropping systems and in both depths.

Despite no significant differences in TC in whole soil after the 4-year crop rotation between all cropping systems at 0-6 cm depth, C-IT and O-RT showed depth stratification, with higher TC at 0-6 cm than at 6-20 cm depth (Table 2). At the end of the grass-clover ley, both O-RT and O-IT increased TC significantly at 0-6 cm depth. The resulting TC after the ley period at 0-6 cm depth was significantly higher in O-RT (24.3 g C kg^{-1} dry soil) than C-NT (17.9 g C kg^{-1} dry soil, Table 2). These differences in TC translated into a C-accrual of between -0.69 and 2.2 g C kg^{-1} dry soil $year^{-1}$ across cropping systems and depths. Here, O-IT and O-RT gained significantly more C than both conventional cropping systems at 0-6 cm depth. Also, both organic cropping systems at 0-6 cm depth had accrued significantly more than at 6-20 cm depth, where there was even a net loss in C (Figure 4). The higher TN after the 4-year crop rotation between depths of O-RT was maintained after the ley period, where O-IT and C-IT also had significantly higher TN at 0-6 cm compared to 6-20 cm depth. Similar to TC between cropping systems at 0-6 cm after ley, TN was highest at O-RT compared to C-NT and C-IT.

Table 1. Distribution of soil aggregate size class fractions of whole soil after a 4-year crop rotation and following 2 years of grass-clover ley. LM: large macroaggregates, SM: small macroaggregates, mi: microaggregates, S+C: silt and clay.

Cropping system	Whole soil fraction (% dry soil)							
	LM (2000 µm)		**SM** (250-200 µm)		**mi** (53-250 µm)		**S+C** (53 µm)	
	0-6 cm	6-20 cm	0-6 cm	6-20 cm	0-6 cm	6-20 cm	0-6 cm	6-20 cm
After a 4-year crop rotation								
Conventional intensive tillage	26 b	27 b	44 a	43 a	23 a	26 a	5 a	4 a
Conventional no tillage	34 b	34 ab	39 a	41 ab	21 a	20 ab	5 a	4 a
Organic intensive tillage	32 b D	47 a	42 a D	32 bc	21 a D	17 b	5 a	3 a
Organic reduced tillage	57 a D	46 a M	22 b D	31 c M	17 a	18 ab M	3 a	4 a
Following 2 years ley								
Conventional intensive tillage	43 a Y	43 a Y	27 a Y	25 a Y	23 a	24 a	7 a Y	7 a Y
Conventional no tillage	42 a	43 a	30 a Y	26 a Y	20 a	21 a	6 a Y	6 a Y
Organic intensive tillage	47 a Y	42 a	30 a Y	27 a	18 a	21 a	5 a	6 a Y
Organic reduced tillage	50 a	50 a	28 a	22 a Y	17 a	17 a	5 a Y	5 a Y
Source of variation								
Block	n.s.		n.s.		n.s.		*	
Cropping system	***		*		*		*	
Year	**		***		n		***	
Depth	n.s.		n.s.		n.s.		n.s.	
Cropping system*year	*		**		n.s.		n.s.	
Cropping system*depth	n.s.		n.s.		n.s.		n.s.	
Year*depth	n.s.		n.s.		n.s.		*	
Cropping system*year*depth	*		*		*		n.s.	

Statistical significance was tested at *** $p<0.001$, **$p<0.01$ and * $p<0.05$ for all parameters evaluated. Different letters represent significant $p<0.05$ differences between least-squares means of cropping systems within a depth and year."D" represents the significant difference ($p<0.05$) between depths of a cropping system, within year, while "Y" denotes a significant difference ($p<0.05$) between years of a cropping system, within depth. Significant effects ($p<0.05$) across management types (organic vs. conventional), within depth and year tested with orthogonal contrasts are marked with "M".

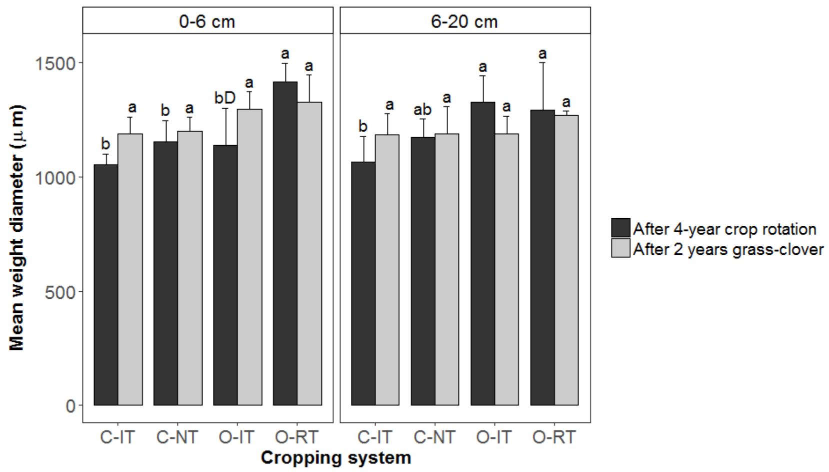

Figure 1 – Mean weight diameter (MWD) at 0-6 cm and 6-20 cm depth after a 4-year crop rotation and after a 2-year ley period. Error bars represent standard deviation of the arithmetic mean. Different letters represent least-square means tested significant differences ($p<0.05$) between cropping systems, within year and depth. Differences ($p<0.05$) between years, within depth are identified with "Y", while "D" identifies differences between depths within cropping system and year.

Table 2. Total carbon (TC), total nitrogen (TN) and C:N of whole soil after the crop rotation, and following the grass-clover ley period at two soil depths and for the entire 0-20 cm profile.

Cropping system	Total C (g kg $^{-1}$ dry soil)			Total N (g kg $^{-1}$ dry soil)			C:N		
	0-6 cm	6-20 cm	0-20 cm	0-6 cm	6-20 cm	0-20 cm	0-6 cm	6-20 cm	0-20 cm
After a 4-year crop rotation									
Conventional intensive tillage	18.9 a D	15.6 a	16.6 a	2.0 a	1.9 a	1.9 a	9.5 a D	8.2 a	8.6 a
Conventional no tillage	16.7 a	15.9 a	16.1 a	2.0 a	1.9 a	1.9 a	8.5 a	8.2 a	8.3 a
Organic intensive tillage	17.8 a	16.6 a	16.9 a	2.1 a	2.1 a	2.1 a	8.6 a	7.9 a	8.1 a
Organic reduced tillage	19.8 a D	17.0 a M	18.0 a	2.2 a D	2.0 a	2.1 a	8.8 a	8.5 a	8.6 a
Following 2 years ley									
Conventional intensive tillage	20.6 ab D	15.3 a	16.2 a	2.0 bc D	1.9 a	1.9 a	10.2 a	9.7 a	8.6 a
Conventional no tillage	17.9 b	14.4 a	16.6 a	1.9 c	1.8 a	1.9 a	9.1 a	8.7 a	8.8 a
Organic intensive tillage	22.3 ab D Y	14.3 a	17.8 a	2.4 ab D Y	1.9 a Y	2.0 a	9.4 a D	8.3 a	8.7 a
Organic reduced tillage	24.3 a D Y M	15.5 a	18.2 a	2.5 a D Y M	1.8 a Y	2.0 a	9.9 a D	8.5 a	9.1 a
Source of variation									
Block	n.s.		n.s.	n.s.		n.s.	n.s.		n.s.
Cropping system	n.s.		n.s.	n.s.		n.s.	n.s.		n.s.
Year	*		n.s.	n.s.		n.s.	**		n.s.
Depth	***		n.s.	***		n.s.	***		n.s.
Cropping system*year	n.s.		n.s.	n.s.		n.s.	n.s.		n.s.
Cropping system*depth	*		n.s.	**		n.s.	n.s.		n.s.
Depth*year	**		n.s.	***		n.s.	n.s.		n.s.
Cropping system*year*depth	n.s.		n.s.	*		n.s.	n.s.		n.s.

Statistical significance was tested at *** $p<0.001$, ** $p<0.01$ and * $p<0.05$. Lowercase letters represent significant ($p<0.05$) differences between least-squares means of cropping systems within depth and years "D" represents differences ($p<0.05$) between depths within year, while "Y" denotes a significant difference ($p<0.05$) between years, within depth. Significant differences between management types (organic vs. conventional), within depth and year tested with orthogonal contrasts are marked with "M".

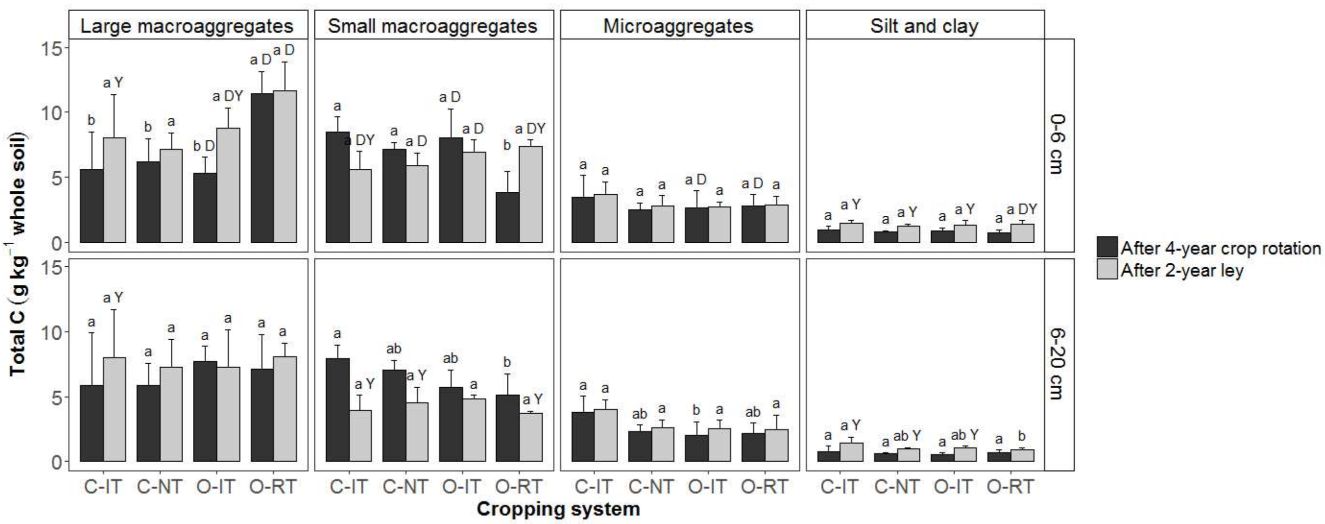

Figure 2 – Total C contribution to whole soil per aggregate fraction. Error bars represent standard deviation of the arithmetic mean. Different letters represent significant ($p<0.05$) differences between cropping systems within year, depth and fraction. Significant ($p<0.05$) differences between depths within cropping system and year are noted with "D", while "Y" marks significant ($p<0.05$) differences between years, within cropping system, depth and fraction.

Table 3. Amount of TC, TN and C:N in macroaggregate-occluded microaggregates (M-mi) and macroaggregate-occluded silt and clay (M-S+C) after a 4-year crop rotation at 0-6 cm, weighted per total proportion in macroaggregates.

Cropping system	M-mi[1]			M-S+C[2]		
	Total C (g kg $^{-1}$ macros)	Total N (g kg $^{-1}$ macros)	C:N	Total C (g kg $^{-1}$ macros)	Total N (g kg $^{-1}$ macros)	C:N
C-IT	12.1 a	1.2 a	10.1 a	18.8 b	2.5 b	7.3b
C-NT	11.1 a	1.2 a	9.1 ab	18.8 b	2.2 b	8.5 ab
O-IT	9.1 a	1.1 a	8.6 ab	16.5 b	2.3 b	7.2 ab
O-RT	11.5 a	1.2 a	9.8 ab	19.4 b	2.3 b	8.4 ab

Different letters represent significant differences between least-squares means ($p<0.05$) of cropping systems within fraction.
[1] Microaggregates within macroaggregates (pooled amount of large and small macroaggregates in similar proportions as present in whole soil).
[2] Silt and clay within macroaggregates (pooled amount of large and small macroaggregates in similar proportions as present in whole soil).

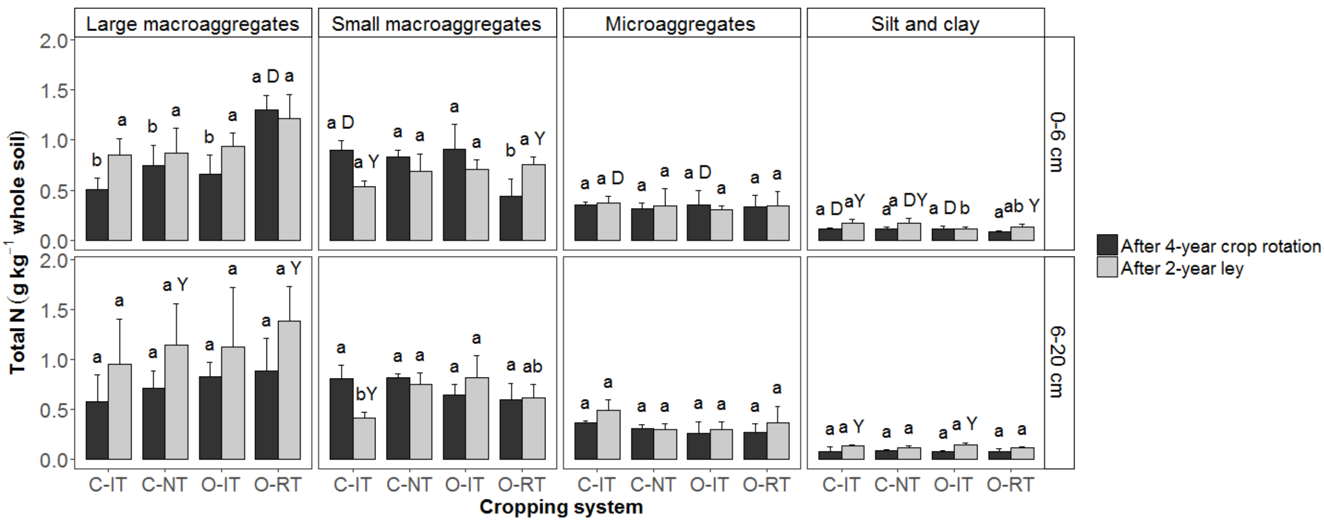

Figure 3 – Total N contribution to whole soil per aggregate fraction. Error bars represent standard deviation of the arithmetic mean. Different letters represent significant ($p<0.05$) differences between least-squares means of cropping systems tested within year, depth and fraction. Significant ($p<0.05$) differences of a cropping system between depth, within year and fraction are noted with “D”, while “Y” marks significant ($p<0.05$) differences between years, within depth and fraction.

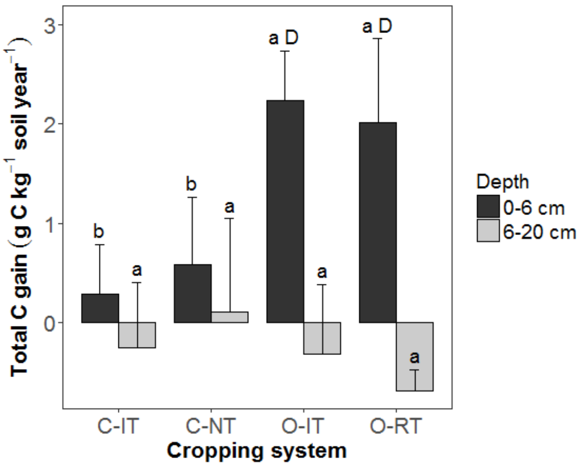

Figure 4 – Total C gain in each cropping systems and depth after ley period. Different letters indicate significant ($p<0.05$) differences between cropping systems within depth, while differences between depths of a cropping systems are noted with "D".

4 Discussion

Our results show that organic management in combination with reduced tillage increased aggregate stability (MWD) after 4-years arable cropping, indicating a synergistic effect between these two management practices (Fig. 1). This is in agreement with results by Bottinelli et al. (2017), who after seven years of a 4-year crop rotation under three tillage types (conventional plowing, surface tillage and no tillage) and two fertilization practices (organic and conventional) in a Humic Cambisol, found the highest aggregate stability under no tillage with organic fertilizer. Similarly, Bissonnette et al. (2001) and Whalen et al. (2003) report greater aggregate stability when combining organic fertilization and conservation tillage (cattle slurry with chisel plowing, and composted cow manure with no tillage, respectively) in crop rotations. These results can be attributed to the combined effect of additional C input from the organic fertilizers and lower tillage intensity.

Organic farming can lead to higher microbial abundance and activity (Francioli et al., 2016; Lori et al., 2017; Peacock et al., 2001). As a by-product of microbial activity, microbial exudates including polysaccharides increase aggregation by binding primary soil particles (Degens, 1997; Golchin et al., 1994; Oades, 1984; Tisdall and Oades, 1982). Increases in soil structure under organic management compared with conventional management have been documented previously (Kong et al., 2005). Abiven et al. (2009) reported in a literature review aggregate stability improvement with organic matter input, along with temporal variation depending on the nature of the organic inputs. However, they found no trend relating to the rate of organic inputs and soil C and clay contents. It is possible that the type of C input significantly affect C turnover and consequently microbial response (Berti et al., 2016). Hence, the need for trials under diverse soil types and conditions to capture site-specific responses and interactions.

The types of conservation tillage used (i.e., no tillage and reduced tillage) were expected to increase aggregate stability. Previous reports of conservation tillage increasing aggregate stability under different conditions include, for example, at the soil surface in a long-term Typic Kanhapludalf (Devine et al., 2014), in boreal soils under spring barley and spring wheat (Sheehy et al., 2015), in a Kenyan Ferrasol under a soy maize rotation (Paul et al., 2013), in a long-term study in Typic Hapludalfs in Michigan, U.S. (Kravchenko et al., 2011) and in Oxisols under different three year crop rotations in Brazil (Zotarelli et al., 2005). Bronick and Lal (2005) analyzed the relationship between soil structure and management in a literature review, making a case for encouraging practices that decrease soil disruption and thereby increase aggregate stability. Both direct and indirect effects are most likely responsible for this increase in MWD. For example, a reduction in physical disruption directly preserves macroaggregates and reduces their turnover, thereby increasing the MWD (Fiedler et al., 2016; Six et al., 1999). An increased microbial activity in these less disrupted systems may also raise the MWD indirectly through an increased production of microbial-derived binding agents (Nivelle et al., 2016; Six et al., 2006; Watts et al., 2001;

Zuber and Villamil, 2016). Improvements in soil structure benefits water fluxes (Horn et al., 1994), reduces susceptibility to erosion (Barthès and Roose, 2002) and enhances biodiversity (Vos et al., 2013). Added value in adopting conservation agriculture practices can reach beyond soil-related aspects and include reduced management costs. At this same experimental site, Wittwer et al. (2017) calculated an average management intensity score (considering energy use, N supply and pesticide use) per cropping system to account for different associated management costs. They found O-RT had the lowest intensity score; 36 %, 109 % and 118 % lower than O-IT, C-NT and C-IT, respectively.

We found that the 2-year grass-clover ley period improved soil aggregate stability under both intensive tillage cropping systems at 0-6 cm depth, as well as at 6-20 cm depth for C-IT. This was evidenced by the significant increase in LM abundance, while the accompanying decrease of SM proportion indicates that the SM were incorporated into the newly formed LM (Table 1). The increase in LM abundance led to a marginal increase in MWD in C-IT at 0-6 cm depth ($p<0.06$), which shows that changes in aggregate distribution do not always result in a significant change in MWD. Of the few studies on the use of temporary pastures together with arable cropping, several reports have shown mixed results in relation to soil structure (Haynes et al., 1991; Panettieri et al., 2017; Studdert et al., 1997). Variation between studies can be attributed to differences in experiment duration, plant composition and soil texture, e.g. higher clay contents can allow for higher C stabilization (Lützow et al., 2006). Different plant compositions can differ in quality (a grass-clover ley may have a lower C:N compared to prairie grasses), allowing more microbial plant residue decomposition and higher diversity of root exudates during the ley period which provide additional C and result in a soil binding effect (Kuzyakov, 2002). Extended season rhizodeposition during the ley period (Jones and Donnelly, 2004) and resulting increased biological activity as opposed to under bare fallow may support aggregate-forming mycorrhizae and also increase soil C input (Oades, 1993; Rillig and Mummey, 2006). An example is Zhang et al. (2016), who reported an increase in macroaggregates in Anthrosols under natural vegetation succession but not under bare fallow. The combination of this effect with the lack of physical disruption most probably allowed for uninterrupted aggregate-forming processes, thus favoring the formation of the higher-aggregated LM fraction measured in our study.

After the 4-year crop rotation, the LM fraction contributed the most C to whole soil correcting for relative abundance of the fraction in whole soil under O-RT at 0-6 cm depth, whereas for other cropping systems it was the SM fraction. Increased physical protection of soil organic carbon (SOC) in the highly-abundant LM fraction may reflect on longer-term SOC stabilization (Lützow et al., 2006; Six et al., 2000a) for O-RT. Beyond C and N, other dynamics such as organic phosphorus storage in LM may have been enhanced (Garland et al., 2017; Nesper et al., 2015). The microaggregate fraction held less TC per gram fraction than LM, SM and S+C across all cropping systems. Differently than Mikha and Rice (2004) who found an increase in aggregate TC and TN due to no tillage and manure application at 0-5 cm depth after 10 years of continuous corn in a Kennebec silt loam, in our study macroaggregates did not increase TC and TN under no tillage and organic management; the latter indicates no management or tillage effect in fraction TC enrichment in our study. At the end of the ley period, C-IT and O-IT had increased the amount of TC contributed by LM in whole soil. This change was caused by an increase in LM abundance rather than changes in LM-associated TC; again indicating that there is no macroaggregate enrichment of TC. Reallocation of TC within fractions can happen without significant changes in bulk soil TC, such as those demonstrated by Sheehy et al. (2015) in a long-term (9-11 years) study of no tillage, reduced tillage and conventional tillage in four boreal soils.

Balesdent et al. (2000), who summarized TC and TN for no-till and conventionally tilled soil in a number of studies under different management histories, reported generally higher C levels under no-till compared to conventional tillage. Similarly, in a meta-analysis Luo et al. (2010) found greater C stocks under no tillage compared to conventional tillage. These and other similar reports of C accumulation in conservation tillage compared to conventional tillage, e.g. in the U.S. (Johnson et al., 2005; West and Marland, 2008), continental and maritime Mediterranean climates (Aguilera et al., 2013; González-Sánchez et al., 2012), tropical and temperate climates (Ogle et al., 2005; Six et al., 2004b; VandenBygaart et al., 2003), have led to the general belief that transitioning from conventional tillage to no tillage can lead to increased sequestered C. Consequently, the additional C sequestered in soil has been thought to contribute to mitigating climate change (Lal, 2010, 2004; Paustian et al., 1997). In contrast, we found no significant cropping system differences in whole soil TC values after 4-years crop rotation although O-RT, and surprisingly C-IT, showed higher TC in 0-6 cm depth than in 6-20 cm depth. C sequestration rates can peak in 5 to 10 years (West and Post, 2002), therefore it is possible that at 4 years experiment start, the full potential of C sequestration had not been realized. However, any gains should be viewed cautiously, considering Powlson et al. (2014), who make a case that the apparent increases of organic carbon in soil under no tillage largely result from an altered depth distribution compared to conventional tillage, such that the quantity of additional carbon is relatively small.

After the ley period, there were no differences in TC at 0-20 cm depth. Nonetheless, in addition to O-RT and C-IT continuing to have greater TC in 0-6 cm depth than in 6-20 cm depth, O-IT also showed TC stratification between depths. Increased TC in the top 10 cm depth after conversion from conventional tillage to no tillage, coupled with decreased TC at 20-40 cm depth was reported by Luo et al. (2010) in an analysis of 69 paired conventional and no tillage experiments where sampling extended deeper than 40 cm. Overall, total SOC to 40 cm depth did not increase except in double cropping systems (Luo et al., 2010). Reduced soil disturbance together with organic matter inputs can lead to the stratification of soil C, which has been suggested as an indicator of soil quality (Sá and Lal, 2009; Franzluebbers, 2002; Gadermaier et al., 2011; Kay and VandenBygaart, 2002).

A key component of organic farming is the use of organic fertilizers, commonly in the form of composted vegetable residues or animal manures. Compared to conventional mineral fertilizers, organic fertilizers, such as the cattle slurry used in this study, include an array of C sources which, can enhance microbe abundance and activity in organic farming (Lori et al., 2017). Consequently, organic fertilization can have a direct impact on soil C input, stabilization and turnover. In a meta-analysis assessing 74 studies from pairwise comparisons of organic and conventional farming systems of temperate zones, Gattinger et al. (2012) reported higher SOC concentrations in organic compared to conventional farming. Despite other reports of organic agriculture increasing C contents (Gomiero et al., 2011; Munro et al., 2002; Tuomisto et al., 2012), Leifeld and Fuhrer (2010) hold that that methodological inconsistencies such as greater fertilizer rates in organic than conventional, lack of a true control and differing crop use between organic and conventional, caused the perceived increase of SOC in organic systems. Observations from other studies have shown C gain in organically fertilized systems after conservation tillage. For example, Crittenden et al. (2015) reported a stronger tillage effect: after four years of crop rotations SOC was higher in reduced tillage compared to conventional tillage across both organic and conventional fertilization in a calcareous marine clay loam. Bottinelli et al. (2017) and Whalen et al. (2003) found aggregate stability to be related to TC. This increase may be explained by higher C stabilization in larger aggregates by physical protection and occlusion in microaggregates (Six et al., 2004a; Lutzow et al., 2006). However, we found higher TC under organic management compared to conventional management at 0-6 cm depth only after the ley period. Therefore, for our conditions, the combined effect of additional plant root deposition and no disturbance during the ley period, with added C input from the organic fertilizer lead to increased TC.

A review on the short-term impacts of tillage found a 1-11 % of soil C loss after even one tillage event (Conant et al., 2007). However, these short-term losses may be ameliorated in the long-term. As a strategy to avoid C losses, a one-time tillage (every 10 or more years) has been suggested for homogenizing C stratification after no tillage without increased loss of labile SOC (Quincke et al., 2007). In loamy sandy soils of northern Germany, Linsler et al. (2013) demonstrated that occasional tillage of grassland had only short-term effects on C stocks. Although a single tillage event decreased C stocks and water-stable aggregates especially at 0-10 cm soil depth, five years later, the differences were no longer significant (Linser et al., 2013). Likewise, Kettler et al. (2000) reported that five years after a single tillage event for controlling a grass weed in winter wheat-fallow system of a silt loam soil under no tillage for over 20 years resulted in a decline in soil organic C at 0 - 7.5 cm depth, but increase at 7.5 - 15 cm depth. Higher C and N contents were found by Omonode et al. (2006) after 17 years of conventional tillage followed by 6 - 7 year intermittent chisel or no tillage, in dark prairie soils under continuous corn and soybean-corn rotations. Future studies on the impact of re-introducing intensive tillage after a ley period will be needed to determine whether LM abundance and C content is maintained long-term.

5 Conclusions

Organic management and reduced tillage significantly increased soil structural stability in 0-6 cm soil layer within a 4-year period of arable cropping only when applied together, pointing towards the need of reducing tillage intensity under organic management. Considering that tillage and organic management are common practices used simultaneously, further studies should combine both organic management and reduced tillage practices to capture non-additive effects..

Integrating a grass-clover ley into the crop rotation as a type of restoration crop significantly increased soil structural stability for both intensive tillage cropping systems, suggesting short-term ley as a minimal input strategy to improve soil structure for intensive plow cropping systems. The grass-clover ley period also significantly increased TC accumulation for both organic fertilization cropping systems in 0-6 cm depth, showing that the use of organic fertilization (i.e., slurry) had a larger effect on short-term C-accrual than conservation tillage strategies. This highlights the potential of organic management to change soil dynamics in favor of C accumulation.

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