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Distribution of organic carbon in humic and granulodensimetric fractions of soil as influenced by tillage and crop rotation

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Abstract. It is widely believed that soil disturbance by tillage is a primary cause of the loss of soil organic carbon (SOC) and that substantial SOC sequestration can be accomplished by conversion from conventional ploughing to reduced tillage. The objective of our study was to find alterations of the organic C content in soil humic and granulodensimetric fractions depending on soil tillage and crop rotation. The field experiment was carried out at the Joniskelis Experimental Station of the Lithuanian Institute of Agriculture on a drained clay loam *Endocalcari-Endohypogleyic Cambisol* (*CMg-n-w-can*). Two technologies – reduced tillage (RT) and conventional tillage (CT) – were compared in crop rotations with different proportions of overwintering and spring crops (0%, 25%, 50%, 75%, and 100% overwintering crops). The results of 2004–2006 are presented. Tillage had a greater influence than crop rotation on all soil C fractions. RT promoted the formation of all fractions of humic acids and FA1 and FA3 fractions of fulvic acids in the entire plough layer. Increasing the proportion of overwintering crops in the rotation to 100% tended to strengthen this effect. The C content in particulate organic matter (POM), light fraction (LF), and both clay-sized sub-fractions, expressed per unit mass of soil, significantly increased under RT in the top 15 cm of soil. The introduction of overwintering crops into the rotation and increasing their proportion had a significant positive influence on C content in POM and LF in the whole plough layer.

Key words: soil organic carbon, light fraction, particulate organic matter, clay fraction, humic acids, fulvic acids.

INTRODUCTION

However unique in form and function, soil is not an isolated body; it is, rather, a central link in the large chain of interconnected domains comprising the biosphere. Soil is a rich mixture of mineral particles, organic matter, gases, and nutrients, which, when infused with water, constitutes a fertile substrate for the initiation and maintenance of terrestrial life. The soil system is thus a self-regulating biological factory, utilizing its own matter, precipitated water, and solar energy (Hillel, 2009).

Soil organic matter (SOM) is considered to be a key attribute of soil quality and also environmental quality (Smith et al., 2010). SOM is one of the most complex components of terrestrial ecosystems (Cotrufo et al., 2011). It is involved in and related to many soil chemical, physical, and biological properties (Carter, 2002; Weil et al., 2003). Since SOM consists mainly of carbon, most analytical methods determine the soil organic carbon (SOC) content.

Organic C in the soil is located in discrete pools (Jones & Donnelly, 2004). SOC is divided into pools having different properties and rates of turnover depending on stabilization mechanisms: (1) easily decomposable – labile fraction with a short residence time of a few months to several years, (2) stabilized by physical–chemical interactions with soil mineral components – intermediary fraction with a residence time of decades, and (3) biochemically recalcitrant – stabile fraction with residence time of centuries to millennia (Six et al., 2002; Lal, 2009; Bruun et al., 2010).

The light fraction (LF) is comprised largely of organic residues in various stages of decomposition; it also contains appreciable amounts of microbial and microfaunal debris including fungal hyphae and spores (Janzen et al., 1992; Gregorich et al., 2006) and has a high concentration of organic C and N relative to that of the whole soil (Marriott & Wander, 2006). The LF content was found to be more sensitive to cropping practices than total organic C in the soil (Leifeld & Kögel-Knabner, 2005). Although the LF provides a sensitive and reasonably precise measure of organic matter changes, it probably only reflects short-term effects (Franzluebbers & Stuedemann, 2008).

Particulate organic matter (POM) is an uncomplexed fraction of SOM composed of particulate (>0.05 mm), partially decomposed plant and animal residues, fungal hyphae, spores, root fragments, and seeds with a recognizable anatomic structure (Baldock & Skjemstad, 2000; Causarano et al., 2008). It is a fraction that lies intermediate between litter and mineral-associated SOM (Franzluebbers, 2005). The largest concentrations of POM in the mineral soils of the temperate regions are situated in the topsoil horizons (Kaiser et al., 2002), and its C content often increases with a reduction in tillage intensity (Franzluebbers & Stuedemann, 2002).

A stabilization of SOM can be reached by its interaction with the mineral part of soil such as clay minerals, resulting in true chemical linkages or sorption complexes (Schulz, 2004; Kögel-Knabner et al., 2008; Schulz et al., 2011). According to Six et al. (2004), very stable clay-sized mineral–organic complexes contain the oldest and most processed organic particles.

The organic matter that does not degrade completely to carbon dioxide forms humic substances through secondary synthesis reactions (Lichtfouse et al., 1998). Humic substances are considered to be highly resistant to further biodegradation, thus providing a long-term sink for C in soils (Hayes & Clapp, 2001; Piccolo, 2002; West & Post, 2002). For many plants as much as 30–50% of the C fixed in photosynthesis is initially translocated belowground (Moran et al., 2005). There is also evidence that belowground plant C is a major source for subsequent conversion into more stable forms of SOC (Wilts et al., 2004; Baker et al., 2007;

Slepetys & Slepetiene, 2012). SOM transformations are affected by management including tillage systems (Lee et al., 2009; Svobodova et al., 2010) and its characteristics are mainly influenced by the tillage depth and intensity. Slepetiene et al. (2011) established that reducing tillage intensity in combination with crop rotations with perennial grasses enables to increase the quantity of SOC.

One of the most important tasks in land cultivation is to ensure a stable supplement of SOC (Velykis et al., 2005). Our hypothesis was that reduced soil tillage and increased proportion of overwintering crops in the rotation would promote quantitative changes in SOC. The aim of this research was to explore the influence of conventional and reduced soil tillage in the rotation with different proportions of overwintering crops on the total organic C content and C content in granulodensimetric SOM and chemical humus fractions in different plough layers.

MATERIALS AND METHODS

Site description and soil

The field experiment was conducted at the Joniskelis Experimental Station of the Lithuanian Institute of Agriculture, located in the northern part of Central Lithuanian lowland (56°21′N, 24°10′E) during the period 1998–2006. The average annual temperature over the last 40 years in this region has been 6.1 °C, and the average precipitation 547.4 mm. Detailed studies of SOC were conducted in the last three years of the experiment (2004–2006). During the investigation period, the annual precipitation varied considerably: 533 mm in 2004, 398 mm in 2005, and 479 mm in 2006.

The parent soil material in the region is glacial lacustrine clay lying on morainic loam. The predominant soil type in its overwhelming part according to FAO/UNESCO (1997) is *Endocalcari-Endohypogleyic Cambisol* (*CMg-n-w-can*) with the following average particle size distribution in the plough horizon (0–30 cm): 50.3% silt, 27% clay, and 22.7% sand. The pH_{KCl} in the 0–30 cm soil layer was neutral and ranged between 6.9 and 7.2. The arable soil layer was medium in humus (2.2%), medium in phosphorus (154 g P_2O_5 kg^{-1} soil), and high in potassium (304 g K_2O kg^{-1} soil).

The soil chemical analyses were performed in the Chemical Research Laboratory of the Lithuanian Institute of Agriculture.

Experimental design and parameters

The effects of conventional and reduced soil tillage in the rotations with different proportions of overwintering and spring crops on the distribution of soil carbon in chemical humus and granulodensimetric fractions were investigated observing the following experimental design: (1) conventional tillage (CT) – mouldboard ploughing and (2) reduced tillage (RT) – mouldboard ploughing after grasses for wheat, ploughless after all cereals. Research was conducted in the crop rotations with a different proportion of overwintering and spring crops: (1) without overwintering crops (spring vetch and spring oats → spring wheat → spring triticale → spring barley), (2) 25% overwintering crops (red clover and timothy → spring wheat → spring triticale → spring barley with undersown perennial grasses), (3) 50% overwintering crops (red clover and timothy → winter wheat → spring triticale → spring barley with undersown perennial grasses), (4) 75% overwintering crops (red clover and timothy → winter wheat → winter triticale → spring barley with undersown perennial grasses), (5) 100% overwintering crops (red clover and timothy → winter wheat → winter triticale → winter barley with undersown perennial grasses).

The field experiment was established using the fully expanded crop rotation method. All crops were grown every year in four replicates. The area of each plot was 90 m^2 and the area of the record sub-plot was 34.5 m^2 for cereals and 44 m^2 for grasses. The plots were arranged in blocks that had the same crops in the rotation treatments as numbered. According to tillage systems, the blocks were arranged in chess order.

General conditions of the field experiment

The main tillage in CT was ploughing with a mouldboard plough at a depth of 23–25 cm. In RT, the ploughing with a mouldboard plough was done at the same depth only for wheat after grasses, while after cereals the soil was loosened without turning at the same depth as ploughing. After harvesting, cereal straw was removed from the experimental plots, and the soil was tilled by a stubble breaker at a depth of 10–12 cm (except for the plots with perennial grasses under crop).

Soil sampling and preparation

For soil sampling, eight sub-samples per plot for all plots were taken randomly with a steel auger. Each soil sample core was separated into 0–15 and 15–25 cm depth, and combined across sub-samples by depth for each plot. All samples were air-dried; visible roots and plant residues were manually removed. Then the samples were crushed, sieved through a 2-mm sieve, and homogeneously mixed. For the analyses of humus content and humic acids' fractional composition the soil samples were passed through a 0.25-mm sieve. The sieved soil samples were dried in an oven at 60–65 °C for 24 h until constant weight. Since all samples were free of carbonates, the measured total C content was equivalent to the organic C content.

The fractionation according to particle size and density followed the protocol of Shaymuhametov (1985) modified by Schulz (2004). The original method separates clay fraction particles of <1 μm, which corresponds to the Russian classification of clays. To meet the international classification of clays (<2 μm) we applied the modified method that separates two clay fractions: clay fraction CF1 (<1 μm) and clay fraction CF2 (1–2 μm). The SOM associated with the clay fraction (particles <2 μm) was separated by applying ultrasonic energy for 1 min to a soil/water suspension (ratio 1 : 3.5; w : v) 12 times; after each sonication the clay fraction was isolated by sedimentation in a centrifuge at 1000 min^{-1}; later this fraction was subdivided into CF1 and CF2 fractions using a different centrifugation speed and time. The time of centrifugation was calculated according to the Shaymuhametov & Voronina method (1972).

The sediment that remained after the isolation of clay-sized SOM was dried to visual dryness, then 40 mL of NaI solution (1.8 g cm^{-3}) was added. The suspension was dispersed with ultrasonic energy and then centrifuged; floating material was siphoned onto filter paper. This procedure was repeated four times. The material collected on the filter paper, designated as the light fraction (LF), was rinsed out with distilled water for eliminating sodium iodide.

POM was isolated according to Franzluebbers & Stuedemann (2002) by shaking the soil samples in 0.01 M $Na_4P_2O_7$ solution, passing the mixture over a sieve with 0.053-mm openings, and collecting the contents remaining on the top of the sieve. The POM fraction was rinsed out with distilled water for eliminating sodium pyrophosphate.

All SOM fractions from each separation method were dried at 65 °C for 48 h and weighed to calculate each fraction's percentage of the total soil mass. After that SOM fractions were ground to fine powder, and C content was determined by the dry combustion (Dumas) method.

Humus content was determined by the Tyurin method modified by Nikitin (1999). SOM was fractionated into three humic acid (HA) and four fulvic acid (FA) fractions according to the Ponomareva & Plotnikova (1980) version of the classical Tyurin method following the scheme presented in Fig. 1. The organic C content of the isolated HA and FA was determined by the spectrophotometric procedure at a wavelength of 590 nm using glucose as a standard after wet combustion according to Nikitin (1999).

The following HA and FA fractions were identified: HA1, FA1 – the mobile fractions, free or weakly bound with clay minerals; FA1a – the so-called aggressive FA fraction; HA2, FA2 – fractions bound with calcium; HA3, FA3 – fractions strongly bound with soil clay minerals.

The experimental data were analysed by two-factor (A – proportion of over-wintering crops: 0%, 25%, 50%, 75%, and 100%; B – type of tillage: CT and RT) and correlation and regression analyses.

Significance of the differences between the means was determined according to the least significant difference (LSD) at 0.05 probability level. The data were processed using the software ANOVA and STAT ENG (Tarakanovas & Raudonius, 2003).

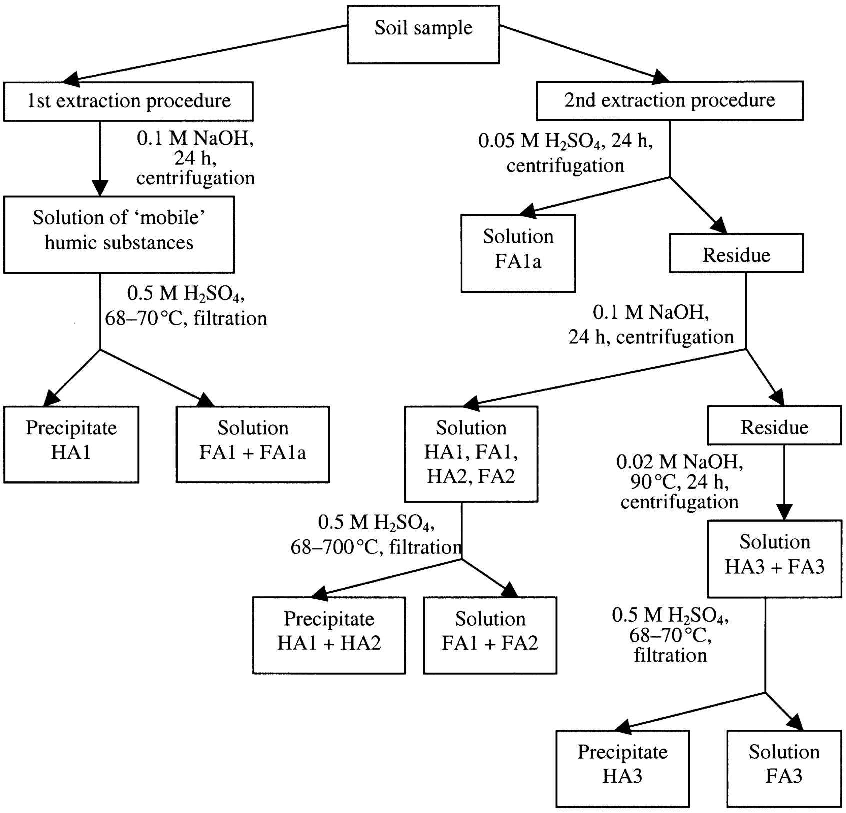

Fig. 1. Scheme of humus fractionation after Ponomareva & Plotnikova (1980). Carbon in HA1, HA3, and FA1a fractions was determined directly; FA1 C was determined by subtracting FA1a and HA1 C from the C in the extract of the 1st procedure; HA2 C was determined by subtracting HA1 C from the C obtained in 0.1 M NaOH extract after decalcification, and FA2 C was determined in a similar way. FA3 C was determined by subtracting HA3 C from the C obtained in 0.02 M NaOH extract after decalcification.

RESULTS AND DISCUSSION

It is widely believed that soil disturbance by tillage is a primary cause of the loss of SOC and that substantial SOC sequestration can be accomplished by changing from conventional ploughing to less intensive methods of tillage. In the present farming situation the largest part of phytomass produced by plants as marketable production is removed from field. In our experiment the cereal straw was also taken away from the experimental plots after harvesting, and only the below-ground plant residues were left in the soil. The data obtained in our experiment correspond to results of numerous studies (Franzluebbers & Stuedemann, 2008; Hermle et al., 2008; Slepetiene et al., 2011) that show an obvious influence of reduced tillage on the SOC content in topsoil. In RT a significantly higher SOC content was established in the 0–15 cm soil layer compared with CT: 13.8 and 12.8 g kg^{-1} soil, respectively (Fig. 2). SOC amounts in rotations with different proportions of overwintering crops under CT differed slightly; conversely, significant increases in the SOC content were established in RT after overwintering crops had been introduced into the crop rotation.

In the 15–25 cm soil layer the SOC content in RT was established as significantly lower compared with CT (Fig. 3). Also a significant influence of the proportion of overwintering crops in the rotation on the SOC content under RT was observed. Introduction of perennial grasses instead of annual grasses had a crucial influence on the SOC content in the plough layer, whereas the effect of the replacement of spring cereals with winter cereals was not significant.

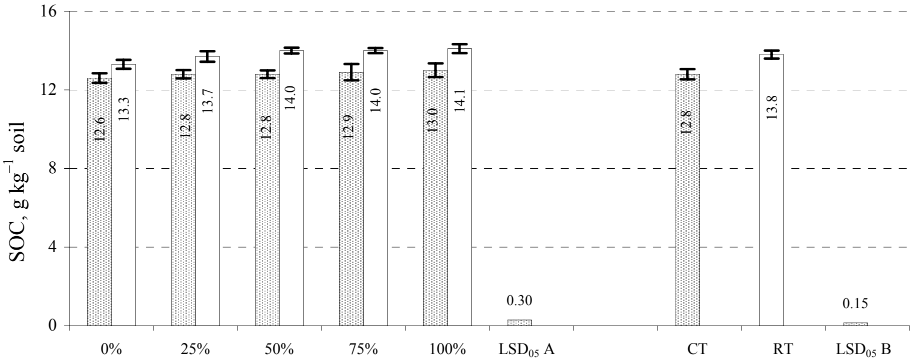

Fig. 2. The content of organic C in the 0–15 cm soil layer as influenced by tillage and the proportion of overwintering crops in the rotation (mean data 2004–2006). CT – conventional tillage, RT – reduced tillage; 0%, 25%, 50%, 75%, 100% – proportion of overwintering crops in the rotation. LSD$_{05}$ A – the least significant difference at 0.05 probability level of factor A (proportion of overwintering crops in the rotation); LSD$_{05}$ B – the least significant difference at 0.05 probability level of factor B (soil tillage system).

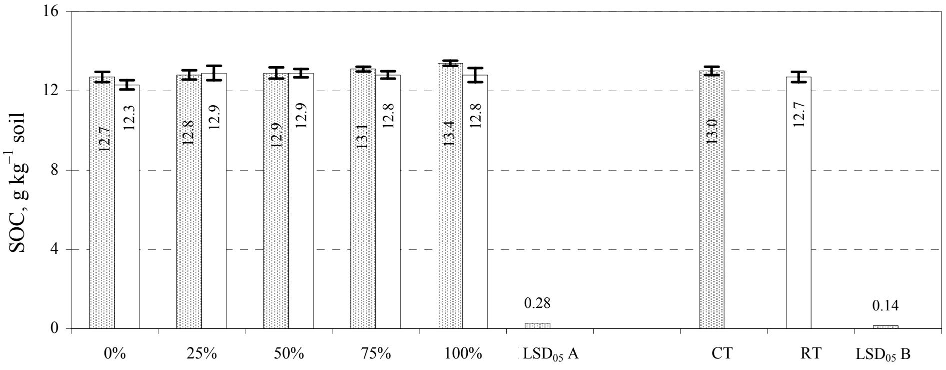

Fig. 3. The content of organic C in the 15–25 cm soil layer as influenced by tillage and proportion of overwintering crops in the rotation (mean data 2004–2006). For explanations see the caption of Fig. 2.

The influence of tillage on SOC seems to depend on the depth to which the tillage operation incorporated plant residues. RT had a higher SOC content in the 0–15 cm layer, but at 15–25 cm depth CT had a higher SOC content.

Easily mineralizable pools of SOM (LF and POM) are important indicators of soil quality dynamics because they are responsive to changes in soil management (Causarano et al., 2008). The inversion action of ploughing in the applied CT tends to homogenize the soil within depth, unlike in the case of RT where the residues accumulate in the topsoil. According to the results of our experiment both LF and POM were significantly higher under RT than CT in the 0–15 cm soil layer (Table 1). In the 15–25 cm plough layer the POM content was only by 0.67% higher at RT compared with CT, but the difference is significant, whereas the influence of tillage on the LF content was nonsignificant. Both the LF and the POM content increased significantly with increasing content of overwintering crops in the rotation (≥25%) compared with the rotation without overwintering crops. The accumulation of easily mineralizable pools of SOM should reflect the slower decomposition of residues under RT than CT.

The C content in the LF, expressed per unit mass of soil (this incorporates the influence of the amount of LF) in the 0–15 cm soil layer was significantly higher in RT compared with CT: 0.808 and 0.671 g kg^{-1}, respectively. The C content in POM in the 0–15 cm soil layer was 1.58 g kg^{-1} in RT compared with 1.33 g kg^{-1} in CT. In the 15–25 cm plough layer the C content in the LF was only 3% higher in RT than in CT, but the difference was significant, while in POM the C content was 0.98 g kg^{-1} in RT, which was significantly lower compared with CT.

Table 1. Dry matter (DM) and C content in the light fraction (LF) and particulate organic matter (POM) in the 0–15 cm and 15–25 cm soil layers as influenced by tillage and the proportion of overwintering crops in the rotation (mean data 2004–2006)

Treatment	0–15 cm				15–25 cm			
	DM, g kg^{-1} soil		C, g kg^{-1} soil		DM, g kg^{-1} soil		C, g kg^{-1} soil	
	LF	POM	LF	POM	LF	POM	LF	POM
Soil tillage systems (B)								
CT	3.33	5.90	0.671	1.33	2.64	4.47	0.454	1.03
RT	3.81*	6.85*	0.808*	1.58*	2.65	4.50*	0.468*	0.98*
LSD$_{05}$ B	0.022	0.052	0.0054	0.029	0.029	0.026	0.0057	0.015
Proportion of overwintering crops in rotation, % (A)								
0	3.39	5.24	0.671	1.29	2.45	3.66	0.340	0.82
25	3.47*	5.82*	0.700*	1.41*	2.47	4.10*	0.366*	0.94*
50	3.58*	6.42*	0.739*	1.42*	2.61*	4.64*	0.450*	1.04*
75	3.67*	6.94*	0.782*	1.50*	2.81*	4.94*	0.567*	1.09*
100	3.73*	7.48*	0.805*	1.67*	2.88*	5.10*	0.582*	1.13*
LSD$_{05}$ A	0.044	0.105	0.0110	0.058	0.059	0.052	0.0110	0.031

Asterisks indicate statistically significant differences between treatments at $p < 0.05$.

Minimizing mechanical soil loosening during tillage promotes the accumulation of unprotected SOM fractions in topsoil; moreover, these fractions have a higher C concentration.

Introduction of overwintering crops into the rotation and increasing their proportion (≥25%) augmented the C content in both the LF and POM in the whole plough layer significantly. The C content in the LF in the 0–15 cm soil layer increased from 0.671 g kg^{-1} in the rotation without overwintering crops to 0.805 g kg^{-1} in all rotations with overwintering crops (by 20%), and in the 15–25 cm soil layer the increment was 71%. In POM the C content in the 0–15 cm soil layer increased from 1.29 g kg^{-1} in the rotation without overwintering crops to 1.67 g kg^{-1} in all rotations with overwintering crops (+29%); in the 15–25 cm soil layer the increment was 37%.

The stable pool of SOC is mainly determined by site conditions, representing the primary products of mineral–organic interactions (Christensen, 2001). According to the data of our experiment, a tendency of the proportion of the smaller-sized clay fraction (CF1) to increase and of the larger-sized clay fraction (CF2) to decrease in soil was observed in RT compared with CT (Table 2). Irrespective of the tillage applied, the share of both clay fractions was higher in the 15–25 cm soil layer than in the 0–15 cm layer. An obvious increasing tendency of both clay fractions content could be observed after overwintering crops had been introduced into the crop rotation in the 0–15 cm soil layer.

Organic C content in clay-sized fractions mainly follows the soil type (Schulz, 2004). Compared with CT an about 5% higher C content, expressed per unit mass of soil, was detected in both clay-sized fractions in the 0–15 cm soil layer in RT,

Table 2. Dry matter (DM) and C content of clay-sized fractions CF1 (<1 μm) and CF2 (1–2 μm) in the 0–15 cm and 15–25 cm soil layers as influenced by tillage and the proportion of overwintering crops in the rotation (mean data 2004–2006)

Treatment	0–15 cm				15–25 cm			
	DM, g kg^{-1} soil		C, g kg^{-1} soil		DM, g kg^{-1} soil		C, g kg^{-1} soil	
	CF1	CF2	CF1	CF2	CF1	CF2	CF1	CF2
Soil tillage systems (B)								
CT	115.0	178.6	1.91	4.13	125.1	183.6	2.10	4.35
RT	115.3	176.7	2.03*	4.37*	127.5	181.2	2.07	4.33
LSD_{05} B	1.74	2.14	0.036	0.057	2.05	3.05	0.050	0.070
Proportion of overwintering crops in rotation, % (A)								
0	112.3	174.7	1.99	4.25	131.1	181.6	2.18	4.31
25	113.3	176.9	1.95	4.22	127.9	180.0	2.11	4.30
50	114.8	177.9	1.97	4.26	126.4*	182.8	2.09	4.34
75	116.8*	178.7	1.96	4.27	126.2*	184.5	2.06*	4.39
100	118.6*	179.7	1.98	4.26	120.1*	183.0	1.98*	4.35
LSD_{05} A	3.47	4.28	0.072	0.113	4.10	6.11	0.100	0.141

Asterisks indicate statistically significant differences between treatments at $p < 0.05$.

and this difference was significant. However, in the 15–25 cm layer the C content in RT was lower in both clay-sized fractions compared with CT. CT operations bury residues more deeply into the soil than RT does. Conditions at the bottom of the plough layer may restrict the mineralization of SOM. The preferential association of organic C with clay-sized particles with increasing soil depth may be explained by the fact that a major pathway for organic C to enter subsoil is via dissolved organic matter. Reactive clay minerals in the small-sized fraction are the most effective sorbent in soils for dissolved SOM (Kaiser et al., 2002). No dependence of the organic C content in the clay-sized fractions on the proportion of overwintering crops in the rotation could be identified.

In recent years a number of Lithuanian researchers published their findings about the effect of cover crops, green manure, and straw on soil agrochemical properties, as well as the content of humic substances in soil (Bučienė et al., 2003; Slepetiene & Slepetys, 2005; Velykis et al., 2005; Arlauskienė et al., 2008, 2011; Tripol′skaya et al., 2008; Slepetiene et al., 2010. During our investigation no additional organic residues were incorporated into the soil. The content of ‘mobile’ humic substances depended on the tillage and proportion of overwintering crops in the rotation (Table 3). The amount of the ‘aggressive’ FA1a fraction differed little between CT and RT in the 0–15 cm soil layer; however, in

Table 3. Contents (g kg^{-1} soil) of ‘mobile’ and bound with calcium humic and fulvic acids in the 0–15 cm and 15–25 cm soil layers as influenced by tillage and the proportion of overwintering crops in the rotation (mean data 2004 and 2006)

Treatment	0–15 cm					15–25 cm				
	HA1	FA1	FA1a	HA2	FA2	HA1	FA1	FA1a	HA2	FA2
Soil tillage systems (B)										
CT	0.67	0.67	0.68	1.58	1.06	0.65	0.71	0.67	1.47	0.92
RT	0.86*	0.79*	0.69	1.79*	1.03	0.76*	0.77*	0.71*	1.59*	0.84*
LSD_{05} B	0.047	0.042	0.012	0.036	0.032	0.048	0.053	0.015	0.078	0.060
Proportion of overwintering crops in rotation, % (A)										
0	0.70	0.66	0.69	1.67	1.15	0.64	0.63	0.68	1.41	0.87
25	0.74	0.70	0.69	1.67	1.06*	0.70	0.72	0.68	1.48	0.83
50	0.77	0.73	0.69	1.67	1.01*	0.70	0.76*	0.69	1.49	0.87
75	0.79	0.75*	0.69	1.69	1.00*	0.74*	0.78*	0.70	1.58*	0.89
100	0.82*	0.78*	0.68	1.75*	1.00*	0.73	0.80*	0.70	1.69*	0.93
LSD_{05} A	0.094	0.084	0.023	0.073	0.063	0.095	0.106	0.030	0.157	0.121

HA1 – humic acids extracted with 0.1 M NaOH and precipitated at pH 1.3–1.5; FA1 – fulvic acids extracted with 0.1 M NaOH and left in the solution after the separation of HA1, minus FA1a; FA1a – fulvic acids extracted with 0.05 M H_2SO_4, so-called aggressive fulvic acids; HA2 – humic acids bound with Ca, extracted with 0.1 M NaOH after removing Ca and precipitated at pH 1.3–1.5; FA2 – fulvic acids bound with Ca, extracted with 0.1 M NaOH after removing Ca and left in the solution after separation of HA, minus FA1.

Asterisks indicate statistically significant differences between treatments at $p < 0.05$.

the 15–25 cm layer it was significantly higher under RT than CT. The contents of both ‘mobile’ humic (HA1) and fulvic (FA1) acids in the whole plough layer were significantly higher under RT compared with CT. A significantly higher C content of HA bound with Ca (HA2) was established at RT compared with CT: respectively 1.79 and 1.58 g kg^{-1} in the 0–15 cm layer and 1.59 and 1.47 g kg^{-1} in the 15–25 cm layer. Conversely, the C content in FA bound with Ca (FA2) decreased substantially due to the application of RT at the bottom of the plough layer compared with CT. The strong association of humic substances with the inorganic soil components is regarded as a means by which carbon is protected against microbial degradation (Baldock & Skjemstad, 2000; Six et al., 2002; Kögel-Knabner et al., 2008).

The introduction of overwintering crops into the rotation and increasing their proportion influenced the accumulation of ‘mobile’ humic substances and HA2 in the whole plough layer. A significantly lower content of FA2 was established in the crop rotations with overwintering crops making up ≥25% compared with the rotation without overwintering crops in the upper soil layer; however, an increasing trend of FA2 content could be observed in the 15–25 cm soil layer.

The findings presented in Table 4 indicate that significantly higher HA3 and FA3 C contents in both soil layers were identified under RT treatment compared with CT. Since the HA3 and FA3 fractions are strongly bound with soil clay

Table 4. Contents (g kg^{-1} soil) of humic and fulvic acids strongly bound with soil clay minerals and total contents in the 0–15 cm and 15–25 cm soil layers as influenced by tillage and the proportion of overwintering crops in the rotation (mean data 2004 and 2006)

Treatment	0–15 cm				15–25 cm			
	HA3	FA3	ΣHA	ΣFA	HA3	FA3	ΣHA	ΣFA
Soil tillage systems (B)								
CT	2.01	1.98	4.26	4.39	2.11	1.99	4.23	4.29
RT	2.32*	2.11*	4.97*	4.62*	2.31*	2.15*	4.65*	4.46*
LSD$_{05}$ B	0.056	0.044	0.093	0.091	0.088	0.069	0.173	0.144
Proportion of overwintering crops in rotation, % (A)								
0	2.03	1.99	4.40	4.49	2.01	2.01	4.06	4.19
25	2.11	2.02	4.52	4.47	2.21*	2.05	4.39	4.28
50	2.19*	2.06	4.63*	4.49	2.19*	2.07	4.38	4.39
75	2.23*	2.06	4.71*	4.50	2.26*	2.05	4.58*	4.42
100	2.27*	2.10*	4.84*	4.56	2.38*	2.17*	4.79*	4.60*
LSD$_{05}$ A	0.112	0.087	0.186	0.182	0.176	0.138	0.347	0.288

HA3 – humic acids strongly bound with soil clay minerals, extracted with 0.02 M NaOH (hot extraction) and precipitated at pH 1.3–1.5; FA3 – fulvic acids strongly bound with soil clay minerals, extracted with 0.02 M NaOH (hot extraction) and left in the solution after the separation of HA3; ΣHA – total content of humic acids in all fractions; ΣFA – total content of fulvic acids in all fractions.
Asterisks indicate statistically significant differences between treatments at $p < 0.05$.

minerals, an increase in their contents improves the stability of soil humic substances. The introduction of overwintering crops into the rotation and increasing their proportion influenced the formation and accumulation of HA and FA strongly bound with soil clay minerals in the whole plough layer. In the HA3 fraction the C content increased from 2.03 g kg^{-1} in the rotation without overwintering crops to 2.27 g kg^{-1} (+11%) in all rotations with overwintering crops in the 0–15 cm soil layer, and from 2.01 to 2.38 g kg^{-1} (+18%) in the 15–25 cm layer; the C content of the FA3 fraction increased by 5.5% in the upper layer and by 8% in the lower layer.

Correlation–regression analysis was performed to find what affects the accumulation of organic C in soil. The correlation of SOC content with C contents in granulodensimetric and chemical humus fractions was positive from moderate to strong and significant at 95–99% probability level (Table 5). Across the tillage systems, the proportion of overwintering crops in the crop rotation and sampling depths showed a positive moderate relationship with the C contents in the chemical humus fractions and almost all clay-sized fractions. The C contents of the LF and POM fractions correlated positively with the C content in humic substances bound with calcium; this relationship was moderate and significant at 95% probability level.

The relationship between C content in clay-sized fraction CF2 (1–2 μm) and total SOC ($C_{CF2} = 1.453 + 0.215$ SOC; $R^2 = 0.355$, $n = 80$) suggests that C accumulation in temperate clay loam Cambisol was due to the increase in the CF2 fraction; i.e., for every unit of total SOC accumulated, 21.5% consisted of CF2 carbon.

The relationship between humified (ΣHA + ΣFA) C content and total SOC suggests that for every unit of total SOC accumulated, 63% consisted of humified carbon (Fig. 4).

Table 5. Correlation coefficients among SOC and C content in SOM granulodensimetric and chemical humus fractions (mean data 2004 and 2006; $n = 80$)

	SOC	CF1	CF2	LF	POM
SOC	–	0.298**	0.552**	0.483**	0.517**
HA1	0.548**	0.279*	0.387**	0.223*	0.280*
HA2	0.312**	–	–	0.451**	0.377**
HA3	0.517**	0.407**	0.313**	–	–
FA1	0.491**	0.341**	0.366**	–	–
FA1a	0.359**	0.274*	0.333**	–	–
FA2	0.296**	–	–	0.247*	0.247*
FA3	0.385**	0.358**	0.269**	–	–

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* Significant at $P \leq 0.05$; ** significant at $P \leq 0.01$.

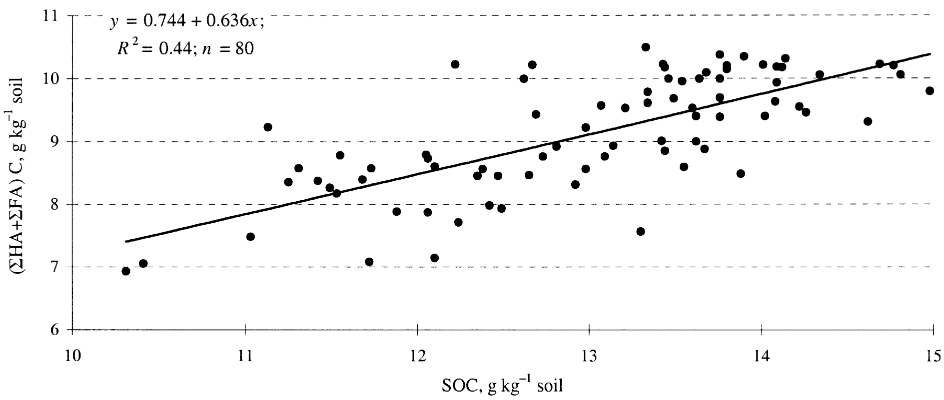

Fig. 4. The relationship between SOC and humified (ΣHA + ΣFA) C content, $n = 80$ (mean data 2004 and 2006).

CONCLUSIONS

The effect of conventional (CT) and reduced (RT) soil tillage in the rotations with different proportions of overwintering and spring crops on SOC and functional carbon pools was investigated in an 8-year field experiment. The results of the last three years of the second experimental period (2004–2006), when the complex of practices was applied once more in the field, suggest that RT influenced the vertical distribution of SOC. A considerable amount of SOC accumulation under the RT system in the upper soil layer may be attributed to the increase of organic C content in the light fraction and particulate organic matter pools (by 32% and 18%, respectively) compared with CT. Soil tillage mainly influenced the decomposable SOC pools, but it also affected more stabilized SOC fractions. Under RT the C content in both clay-sized fractions increased in the 0–15 cm soil layer and decreased in the 15–25 cm layer. Use of RT practices caused significantly increased contents of humic and fulvic acids strongly bound with the soil clay minerals and of humic acids bound with calcium in the whole plough layer.

The introduction of overwintering crops into the rotation and increasing their proportion boosted the SOC and C contents in the easily mineralizable pools in the whole plough layer. Introduction of perennial grasses instead of annual grasses was of crucial influence, whereas the influence of the replacement of spring cereals by winter cereals was not significant.

These findings give support to the conversion from CT to RT in Lithuania where the conventional mouldboard ploughing is still the predominant method of clay loam soil tillage. In our experiment such conversion introduced changes in the distribution of SOC within the soil profile, increased the SOC content in the topsoil layer in a relatively short time, and also improved soil quality.

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Orgaanilise süsiniku paiknemine huumuse ja granulodensimeetrilistes fraktsioonides sõltuvalt mullaharimisest ning külvikorrast

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Laialt on levinud arvamus, et mulla orgaanilise süsiniku vähenemise peamine põhjus on mulla segamine harimise käigus ja et olulise orgaanilise süsiniku talletumise suurenemise põllumulda tagab tavaharimise asendamine vähem intensiivse minimaalse mullaharimisega. Käesoleva uurimuse eesmärgiks on välja selgitada orgaanilise süsiniku sisalduse muutused erinevates huumusainete ja granulodensimeetrilistes (erineva osakese suuruse ning tihedusega humifitseerumata mulla orgaanilise aine) fraktsioonides sõltuvalt maakasutuse tehnoloogiast.

Põldkatse korraldati Leedu Põllumajandusinstituudi Joniskelise katsejaamas kuivendatud raske liivsavilõimisega *Endocalcari-Endohypogleyic Cambisol*'il (*CMg-n-w-can*). Erineva tali- (0, 25, 50, 75 ja 100%) ning suvekultuuride vahekorraga külvikorras, kus külvikorda järjest juurdevõetud talikultuurideks olid punase ristiku ja timuti segu, talinisu, talitritik ning talioder, uuriti võrdlevalt minimaalse ja tavamaaharimise tehnoloogiate ning külvikordade mõju mulla süsinikusisaldusele. Töös on esitatud aastate 2004–2006 tulemused, kui uuriti terviklikult agrotehnoloogiliste võtete kompleksi.

Külvikorraga võrreldes avaldas harimisviis suuremat mõju kõigile orgaanilise aine fraktsioonidele. Minimaalne harimine soodustas kõigi huumushappe fraktsioonide ja fulvohappe mobiilsete ning savimineraalidega seotud fraktsioonide moodustumist kogu künnikihi ulatuses. Talikultuuride osatähtsuse suurendamine külvikorras suurendas seda mõju veelgi. Süsinikusisaldus (kontsentratsioon) partikulaarses orgaanilises aines, kerges fraktsioonis ja mõlemas savi alamfraktsioonis suurenes minimaalse harimise korral usaldusväärselt pindmises 15 cm mullakihis. Taliviljade sisseviimine ja nende osakaalu suurendamine külvikorras avaldas usaldusväärselt positiivset mõju kogu künnikihi partikulaarse ja kerge fraktsiooni orgaanilisele ainele.