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Original research paper

Long-term soil quality and C stock effects of tillage and cover cropping in a conservation agriculture system

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ABSTRACT

Conservation and regenerative agriculture are widely considered as approaches aiming at addressing challenges in relation to climate change and soil quality. In that respect, no tillage (NT) and use of cover crops (CCs) are seen as key elements in achieving the sustainability goals of these approaches, but the long-term individual and interactive effects of these elements remain unknown. We examined the impact of tillage (NT and mouldboard ploughing) combined with a fodder radish CC in continuous cereal cropping. Soil was sampled in the 0–10 and 10–20 cm layer after two decades of treatments in the CENTS experiment at AU Viborg (Denmark) situated on a sandy loam with 9% clay. We assessed soil structural quality, SOC stocks, clay dispersibility (CD), wet stability of aggregates (WSA) and soil pore characteristics. Neither tillage nor cover cropping affected the SOC stock in the 0–20 cm soil layer. No tillage improved CD, WSA and plant available water capacity in 0–10 cm depth as compared to ploughing. The marked improvement in CD and WSA of NT soils could not be explained by SOC per se, but rather positive effects ascribed to absence of disturbance. In contrast, soil porosity, especially in 10–20 cm depth, the fraction of soil volume represented by >30 μm pores and gas diffusivity decreased, and NT soils resulted in a less good soil structural quality. The inclusion of CCs improved soil structural quality and the functionality of the soil macropore system. Hence, CCs have the potential to alleviate negative effects of NT on pore characteristics at macroscale. Furthermore, we found that the positive effects of NT on CD and WSA and of CCs on pore characteristics at macroscale were much more pronounced after long- (20 yrs NT; 13 yrs CC) than after medium-term (10 yrs NT; 5 yrs CC) underlining the value of long-term conservation agriculture experiments.

1. Introduction

Globally, agriculture is faced by key challenges in relation to climate change, food security, biodiversity, and environmental impacts. Conservation agriculture (CA) and regenerative agriculture (RA) are widely considered as approaches aiming at addressing the current and future challenges (Kumawat et al., 2023; Rehberger et al., 2023). Both CA and RA are based on several integrated management practices. The key principles in CA are minimal soil disturbance, permanent soil cover, and species diversification (https://www.fao.org/3/cb8350en/cb8350en.pdf). These principles are also essential to RA – and in addition use minimal external inputs, mixed farming, and manure and compost (Rehberger et al., 2023). Thus, for both CA and RA the use of no tillage (NT) and cover cropping (CC) are key elements in achieving the sustainability objectives of these approaches (Kumawat et al., 2023; Rehberger et al., 2023).

No tillage affects a range of soil physical, chemical, and biological properties, and key soil functions and ecosystem services (Jayaraman and Dalal, 2022; Skaalsveen et al., 2019). The soil organic carbon content (SOC) is generally increased in the surface 0–10 cm layer of NT soils whereas the effect is minor in deeper layers, i.e. causing increased stratification of SOC in the soil profile (Mondal et al., 2023). For soil physical properties, NT has been shown to increase soil structural stability (SSS) as well as water infiltration and retention in the surface layer (0–10 cm) as compared to mouldboard ploughing (MP) (Blanco-Canqui and Ruis, 2018). This reduces the risk of erosion and the loss of particle bound nutrients and other chemicals to the water bodies, and increases drought resistance (Skaalsveen et al., 2019). The beneficial impact of NT

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on soil physical properties of the surface layer has partly been linked to an increase in SOC content in the surface layer (Blanco-Canqui and Ruis, 2018). Generally, SOC positively influences the soil physical properties such as SSS (Chebet et al., 2023; Jensen et al., 2019). The question is, however, whether other factors also play a part in the NT induced improvement of soil structure of the surface layer. For instance, Chebet et al. (2023) showed that the relationship between SOC and SSS markedly changed shortly after the conversion from annually tilled cereals to semi-natural grassland with absence of tillage.

The benefits of NT on the physical properties of the surface layer may to some extent be counteracted by increased topsoil compaction – especially in the layer below the seeding depth and down to the previous tillage depth (typically 5–25 cm layer) (Soane et al., 2012). This can result in poor aeration under wet conditions (Kadžienė et al., 2011; Martínez et al., 2016; Schjønning and Rasmussen, 2000) and high root penetration resistance under dry conditions (Kadžienė et al., 2011; Tormena et al., 1999). The formation of a system of continuous and stable macropores (cracks and biopores) under NT will – over time – improve the conditions for gas and water transport (Soane et al., 2012). Recent results have shown that cover crops may stimulate the formation of continuous biopores in the topsoil and the upper subsoil (Pulido-Moncada et al., 2020; Zhang et al., 2022). Further, cover crop induced biopores mitigated the negative effects of NT on topsoil compaction in some short- and medium-term studies (Abdollahi and Munkholm, 2014; Abdollahi et al., 2014; Villarreal et al., 2021), but not in others (Rücknagel et al., 2016). Cover crops have also been found to increase soil structural stability (Hao et al., 2023), soil C input and the SOC content (Joshi et al., 2023; Poeplau and Don, 2015). The SOC content is expected to increase for at least 10–20 years before a new equilibrium is established (Jensen et al., 2022). This increase in SOC content may, itself, improve the soil structural properties – especially of the surface layer under NT.

Overall, CC may alleviate NT induced soil structural problems. Little is, however, known about the long-term effects of such additional practices to NT, as pointed out by Blanco-Canqui and Ruis (2018).

This study explores long-term effects of no tillage and cover cropping practices on soil structural stability, pore characteristics and carbon stocks. The experiment was carried out on Danish sandy loam soil where CA management has been performed for 20 years (the CENTS experiment). We hypothesized that CC increased the anticipated positive impact of NT on soil carbon content and SSS in the soil surface layer, increasing soil C stock and limiting the risk of soil sealing, crusting and erosion. We also hypothesized that CC alleviated potential negative effects of NT on the pore system in lower part of the formerly ploughed layer limiting the need for mechanical soil loosening (i.e. tillage).

2. Materials and methods

2.1. The CENTS experiment

Soil was from CENTS experiment at AU Viborg, Foulumgaard Experimental Station, DK (56°30’N, 09°35’E), a sandy loam soil classified as an Mollic Luvisol (IUSS Working Group WRB, 2015) and a Typic Hapludalf (Soil Survey Staff, 2014). The study site is based on morainic ground deposits from the last glaciation (Munkholm et al., 2008). The topsoil (0–25 cm depth) contains 9% clay (<2 mm), 13% silt (2–20 mm), 75% sand (20–2000 mm), and 3.1% organic matter (Munkholm et al., 2008). The mean annual precipitation and temperature (2003–2020) were 733 mm and 8.0°C, respectively (Gómez-Muñoz et al., 2021).

The R5 crop rotation of the CENTS experiment was selected for the present investigation and was initiated in 2003. The experiment is arranged in a completely randomized split-plot design with two factors and three blocks. Tillage (T) is the main plot factor, whereas cover crop (CC) is the subplot factor. The tillage systems included in this study were no tillage (NT) and mouldboard ploughing to a depth of 20 cm (MP). A traditional Nordsten seed drill was used in the MP treatment and a chisel coulter was used in the NT treatment. Each tillage plot consisted of two 3-m-wide and 72.2-m-long tillage strips. Soil was sampled in the 0–25 cm layer in the tillage main plots in 2002 before the initiation of the experiment. The SOC concentration was 2.05 and 1.99 g 100 g^{-1} in the MP and NT treatments, respectively (Hansen et al., 2015). Paired subplots (13.7 by 3 m) with (+CC) or without (-CC) a fodder radish (*Raphanus sativus* L.) cover crop was selected within the main tillage plots. The CC treatment was introduced in 2007 (Table S1). Fodder radish seeds (13 kg ha^{-1}) were broadcast two weeks before the expected harvest of the main crop, and CCs were not present in 2016 and 2020. Spring barley (*Hordeum vulgare* L.) was grown every year during 2007–2021, except for four single years between 2016 and 2021, when spring oat (*Avena sativa* L.) or winter wheat (*Triticum aestivum* L.) was grown (Table S1). Spring barley and oat were sown in April and harvested in August, while winter wheat was sown in September and harvested in August. Tillage was performed prior to sowing of the main crop. Straw was chopped and left in the field after harvest. Fertilizer standards for the cereal main crops were given as an annual spring application of mineral fertilizer or manure (Hansen et al., 2015). Agrochemicals were used to control weeds and pests.

2.2. Soil sampling and field measurements

In February 2022, a sampling campaign was carried out. The average gravimetric soil water content (GWC) was 19.6 g 100 g^{-1} which was approximately at field capacity. Using a spade, minimally disturbed soil blocks were sampled at two sampling positions per plot and at two depths (0–10 and 10–20 cm). In total, 24 samples were collected (2 tillage systems × 2 cover crop treatments × 3 blocks × 2 depths = 24). The soil blocks were stored in sealed plastic boxes at 2°C until analyses could take place. Subsamples from the soil blocks were used for measuring wet stability of aggregates (WSA), clay dispersibility (CD), and soil organic carbon (SOC). Subsequently, undisturbed soil cores (6.1 cm in diameter, 3.4 cm in height, 100 cm^3) were sampled from the 4–8 cm and 12–16 cm soil layers. Six soil cores were sampled per depth in each plot. Three of the samples were taken from one location, while the other three were taken from a different location within the subplot. In total, 144 soil cores (2 tillage systems × 2 cover crop treatments × 3 blocks × 2 depths × 6 replicates = 144) were collected for analysis. The soil cores were stored at 2°C until analyses could be performed.

In May 2022, a visual evaluation of soil structure (VESS) analysis was conducted to evaluate the structural quality of the topsoil (0–22 cm) according to Guimarães et al. (2011). Using a spade, an undisturbed portion of soil (22 cm deep, 10 cm thick, and 20 cm wide) was dug out. It was gently broken down along the natural boundaries between the aggregates when possible. First, layers within the 0–22 cm soil were identified and then scored individually. The identified layers received a score in the range from 1 (the best) to 5 (poor soil structure) based on a visual evaluation of the size, shape, and strength of the aggregates, visible porosity, the appearance, color, and presence of aggregates or clods.

2.3. Soil organic carbon

Around 200 g of the soil blocks were left to air-dry, crushed, and passed through a 2-mm sieve. An aliquot was used for determination of SOC content by high-temperature dry combustion at 950 °C using a Vario Max Cube (Elementar Analysensysteme GmbH, Hanau, Germany). Droplets of 10% HCl were used to test for carbonates, but it was not detected. The SOC content was expressed as g 100 g^{-1} oven-dry soil (105 °C for 24 h).

2.4. Soil structural stability

Wet stability of aggregates (WSA) and clay dispersibility (CD) was measured at plot level for each depth on soil retrieved from the fieldmoist soil block using a small corer (22-mm diameter) and gently crumbled by hand to pass an 8-mm sieve. The methodology is described in detail in Kemper and Rosenau (1986) and Jensen et al. (2019). For WSA, 4 g soil was transferred to a sieve with 250-µm openings. The aggregates were rewetted using a vaporizer with artificial rainwater (0.012 mM $CaCl_2$, 0.150 mM $MgCl_2$ and 0.121 mM NaCl; pH 7.82; EC 2.24×10^{-3} S m^{-1}). The sieve was moved up and down for 3 min (34 cycles min^{-1}; stroke length 13 mm) in aluminum cans filled with 100 mL artificial rainwater. After 30 seconds, the >250-µm aggregates were transferred from the sieves to 100 mL beaker glasses using a spray bottle. After oven-drying (105°C, at least 24 h), the samples were transferred into 50 mL tubes. The sediment in the tube was corrected for particles >250-µm isolated by chemical dispersion to express WSA on basis of soil free of particles >250-µm (sand and stones).

For CD, 10 g of soil was transferred to 100 mL plastic tubes and artificial rainwater was added gently along the tube wall to obtain a soil: water ratio of 1:8 by weight. After end-over-end shaking for 2 min (40 rpm; 25-cm diameter rotation) using a Stuart Tube Rotator model SB3, the suspension was left to stand for 3 h 50 min (based on Stokes' Law) after which particles ≤ 2-µm was siphoned off. The weight of dispersed clay was determined after oven-drying (105°C, at least 24 h), and the sediment in the tube was corrected for particles >250-µm as described for WSA.

2.5. *Soil pore characteristics*

The minimal disturbed soil cores (100 cm^3) were weighed and then placed on top of a tension table and saturated with water from beneath (Dane and Hopmans, 2002). Soil water retention, air permeability (K_a) and gas diffusivity were determined at −10 kPa matric potential after which the soil cores were oven-dried (105° C, at least 24 h), and bulk density calculated. Soil porosity was estimated from bulk density and particle density. A particle density of 2.61 g cm^{-3} was used (Eden et al., 2011). Soil water retention at −1.5 MPa (permanent wilting point) was predicted based on clay and SOC content using Eq. (1) in Hansen (1976):

$$\mathrm{PWP} = 0.365 \times \mathrm{Clay} + 1.254 \times \mathrm{SOC\ content} + 0.630 \quad (1)$$

where PWP denotes the permanent wilting point (gravimetric water content, %), and clay and SOC contents are in units of g 100 g^{-1} oven-dry soil.

Volumetric water content at −10 kPa matric potential was calculated from the weight loss upon oven-drying. Pore-water suction was assumed to relate to an average pore size by the approximate relation:

$$d = \text{-}3000/\mathrm{h} \quad (2)$$

where d is the tube-equivalent pore diameter (µm), and h is he soil matric potential (hPa). The equation derives from the physics-based capillary rise equation of Young-Laplace. Plant available water capacity (PAWC) was determined as the difference in volumetric water content at −10 kPa and −1.5 MPa (PWP).

Soil air permeability (K_a) was measured using the Forchheimer approach at four pneumatic pressures as described in Schjønning and Koppelgaard (2017). The relative gas diffusivity (D_s/D_o) was calculated from measurements conducted using the one-chamber, one-gas method described by Schjønning et al. (2013). Pore organization (PO) was calculated from K_a and air-filled porosity (ε_a) as suggested by Groenevelt et al. (1984):

$$\mathrm{PO} = K_a / \varepsilon_a \quad (3)$$

Soil tortuosity (pore length to sample length) was calculated from D_s/D_o and ε_a as proposed by Ball (1981):

$$\mathrm{Tortuosity} = \mathrm{sqrt}(\varepsilon_a / D_s/D_o) \quad (4)$$

2.6. *Calculations and statistics*

For VESS, two replicates of each experimental plot were analysed. The average of its evaluation was used for statistical analysis. The final score was obtained by multiplying the score of each layer by its depth and dividing that result by the total depth (Guimarães et al., 2011):

$$\mathrm{VESS} = (\mathrm{Sq1}\times \mathrm{d1})/\mathrm{dtot} + (\mathrm{Sq2}\times \mathrm{d2})/\mathrm{dtot} \quad (5)$$

where Sq1 is the VESS evaluation of the top layer, Sq2 is the VESS evaluation of the bottom layer, d1 is the top layer thickness, d2 is the bottom layer thickness, and dtot is the total depth.

We calculated the changes in soil organic carbon stocks in the 0–20 cm soil layer from 2012 to 2022 based on the equivalent soil mass (ESM) approach outlined in Eq. (5-7) in Fowler et al. (2023). Soil organic carbon and bulk density in the two soil layers from 2012 were from Abdollahi and Munkholm (2014) and Abdollahi et al. (2014).

For statistical analysis, the K_a and PO data were logarithmically transformed to yield normality. The statistical analyses were performed using the R project software package Version 4.0.3 (R Foundation for Statistical Computing). The results were analysed by a linear mixed model using the *lmer* function of the *lme4* package. Tillage (T), cover crop (CC) and depth (D) were considered as fixed factors, while block was included as random factor. The significance of different factors was discriminated by analysis of variance (ANOVA) Type III. $P<0.05$ was the criterion used for the statistical significance of the different treatment effects. When T, CC, D, or the interaction between them were significant, further analyses were made to isolate differences between them (pairwise comparisons) using the estimated marginal means (*emmeans*) function implemented in the R *emmeans* package. Post hoc comparisons were performed by use of the Tukey's HSD test. To test if SOC was affecting WSA and CD differently depending on if the soil was under no tillage or mouldboard ploughing, linear mixed models including T and CC as categorical fixed factors, SOC as continuous fixed factor and block as random factor were used. When the ANOVA Type III revealed significant interactions between T and SOC, further analyses were made to test if the slopes were significantly different using the *emtrends* function implemented in the R *emmeans* package.

3. Results

No significant interaction between tillage (T), cover crop (CC) and depth (D) for any of the measured soil properties was observed (Table 1). Significant interactions between T and D were found for SOC, porosity and PAWC, while only one significant interaction between T and CC was observed (tortuosity).

3.1. *VESS evaluations*

Tillage (T) and cover crop (CC) both significantly affected VESS scores (Table 2). Ploughing and inclusion of CC resulted in a significantly lower VESS score as compared to no tillage and no CC, respectively (Figs. 1a and 3a).

3.2. *Soil organic carbon and stock*

The no tillage treatment in 10–20 cm depth had a significantly lower SOC content as compared to the upper soil layer and the ploughing treatment irrespective of depth (Fig. 1b). The SOC stock in the 0–20 cm soil layer was not significantly affected by tillage and use of cover crop (Table 2). The change in SOC stock from 2012 to 2022 was likewise unaffected by tillage and use of cover crop (Fig. 4). However, the decrease in SOC stock was 3.19 Mg C ha^{-1} without a cover crop and 1.07 Mg C ha^{-1} with a cover crop, although not significant (P=0.284).

Table 1
Analyses of variance (Type III) for soil organic carbon (SOC), wet stability of aggregates (WSA), clay dispersibility (CD), porosity, permanent wilting point (PWP), plant available water capacity (PAWC), pores >30 μm, air permeability at −10 kPa (K_a), relative gas diffusivity at −10 kPa, pore organization at −10 kPa and tortuosity in 2022 for the tillage treatment (T: no tillage or mouldboard ploughing), cover crop treatment (CC: with or without cover crop) and depth (D: 0–10 or 10–20 cm). Block was included as random effect.

	SOC	WSA	CD	Porosity	PWP	PAWC	Pores >30 μm	K_a	Gas diffusivity	Pore organization	Tortuosity
Tillage (T)	**<0.05**	**<0.001**	**<0.001**	**<0.001**	**<0.001**	**<0.01**	**<0.001**	0.135	**<0.001**	0.741	**<0.001**
Cover crop (CC)	0.443	0.217	0.232	**<0.05**	0.069	0.271	**<0.01**	**<0.05**	**<0.001**	**<0.05**	**<0.001**
Depth (D)	**<0.001**	**<0.05**	0.861	**<0.001**	0.501	**<0.001**	0.576	0.129	0.548	0.117	0.210
T × CC	0.603	0.656	0.118	0.966	0.708	0.804	0.959	0.190	0.998	0.171	**<0.05**
T × D	**<0.001**	0.759	0.365	**<0.01**	0.475	**<0.001**	0.977	0.254	0.472	0.218	0.835
CC × D	0.807	0.289	0.126	0.450	0.613	0.088	0.135	0.902	0.295	0.947	0.063
T × CC × D	0.807	0.378	0.449	0.367	0.525	0.332	0.220	0.382	0.141	0.435	0.051

Table 2
Analyses of variance (Type III) for VESS scores, SOC stock 2012, SOC stock 2022 and change in SOC stock from 2012 to 2022 in 0–20 cm depth for the tillage treatment (T: no tillage or mouldboard ploughing) and cover crop treatment (CC: with or without cover crop). Block was included as random effect.

	VESS-sq	SOC stock 2012	SOC stock 2022	Change in SOC stock
Tillage (T)	**<0.01**	0.770	0.396	0.904
Cover crop (CC)	**<0.01**	0.114	0.292	0.284
T × CC	0.455	0.159	0.062	0.615

3.3. Soil structural stability

Wet stability of aggregates (WSA) was significantly larger for NT than the MP plots with 69.3 and 50.0 g 100 g^{-1}, respectively (Fig. 1c), and WSA was 4.1 g 100 g^{-1} larger in the 10–20 as compared to the 0–10 cm soil layer. Clay dispersibility (CD) was significantly lower for NT than the MP plots with 2.81 and 3.69 g kg^{-1}, respectively (Fig. 1d).

3.4. Pore characteristics

The MP treatment had a larger porosity than the NT treatment irrespective of depth, and the porosity was lower in 12–16 than 4–8 cm depth for the NT treatment (Fig. 2a). Inclusion of cover crop increased porosity as compared to no cover crop irrespective of depth (Fig. 3b).

Plant available water capacity (PAWC; water retained in 0.2–30 μm pores) was significantly larger for NT in 4–8 as compared to 12–16 cm depth and the MP treatment irrespective of depth (Fig. 2b). The fraction of soil volume represented by pores >30 μm was 0.284 and 0.223 m^3 m^{-3} for MP and NT (Fig. 2c), respectively, and 0.246 and 0.261 m^3 m^{-3} for -CC and +CC (Fig. 3c), respectively. Tillage did not affect air permeability (K_a) and pore organization (PO), whereas K_a and PO were significantly larger with cover crop than without (Fig. 3d-e). The geometric mean for K_a was 53 and 83 μm^2, and PO was 124 and 172 μm^2 for -CC and +CC, respectively. Tillage and inclusion of cover crop both affected relative gas diffusivity (Figs. 2d and 3f). Relative gas diffusivity was 0.0319 and 0.0558 for NT and MP, respectively, and 0.0395 and 0.0483 for -CC and +CC, respectively. Pore tortuosity was in general larger for the NT than MP treatment irrespective of cover cropping, however, within the NT treatment -CC had larger tortuosity than +CC (Fig. 5).

3.5. Linking soil organic carbon to structural stability

A significant interaction between SOC and tillage was found for both WSA (P=0.041) and CD (P=0.039). This entails that SOC was affecting WSA and CD differently depending on if the soil was under no tillage or mouldboard ploughing, i.e., significant difference in the slopes of the linear regressions. A linear increase in WSA with increasing SOC was observed for MP (R^2=0.47, $P<0.05$), while no relation was found for NT (R^2=0.00, P=0.99) (Fig. 6a). A linear decrease in CD with increasing SOC was observed for both MP (R^2=0.69, $P<0.001$) and NT (R^2=0.62, $P<0.01$) (Fig. 6b). However, the slope of the linear regression was significantly lower for the NT treatment (-1.2) as compared to the MP treatment (-2.4).

4. Discussion

4.1. Tillage effect

A vertical stratification of SOC was found in the NT plots with a larger SOC content in the top 0–10 cm as compared to the 10–20 cm soil layer (Fig. 1b). This stratification was already observed in 2012 (Abdollahi and Munkholm, 2014) following ten years of NT treatment and is in correspondence with other studies, e.g., Mondal et al. (2023). However, taking the whole 0–20 cm soil layer as well as bulk density into account, the SOC stock did not differ between tillage treatments. Small or no changes in SOC stocks for NT as compared to MP have also been found in meta-analysis (Meurer et al., 2018; Mondal et al., 2023). Despite the similar SOC contents, a marked improvement in soil structural stability (SSS) was observed for NT as compared to MP with an increase in WSA by 39% and decrease in CD by 31% (Fig. 1 cd). In 2012, WSA was greater for NT in the 10–20 cm layer only and the difference was small, and CD was not affected by tillage (Abdollahi and Munkholm, 2014). Hence, 20 as compared to 10 years of tillage treatment displayed much more pronounced effects illustrating the importance of long-term experiments for assessing tillage effects on soil parameters as SSS continued to develop for NT plots. For WSA and CD we found individual correlations to SOC for each tillage treatment (Fig. 6). For WSA, the stabilizing effect of NT was independent of SOC although SOC ranged from 1.54% to 2.42%, while SOC significantly increased WSA for the MP treatment. For CD, the NT soils were less affected by SOC as compared to MP soils. Other studies have also found that SSS of arable soils with annual tillage were more affected by changes in SOC as compared to grassland soils with absence of tillage (Chebet et al., 2023; Jensen et al., 2019). Hence, the stabilizing effect of NT is related to other drivers rather than SOC per se. The absence of soil disturbance may improve the persistency and presence of aggregate-forming factors such as roots, fungal hyphae, and earthworms (De Notaris et al., 2021; Tisdall and Oades, 1982). In addition, crop residues left on the surface in NT plots may protect aggregates against disturbance by raindrops and reduce fluctuations of soil water content and temperature resulting in less abrupt cycles of wetting-drying and freezing-thawing and hence aggregate-disintegration (Blanco-Canqui and Ruis, 2018). Plant available water capacity was larger in the topsoil of NT (Fig. 2b). The difference in SOC compared to MP was not significant, which may indicate that the larger PAWC is due to the development of a more stable structure including increases in small pore size classes as indicated by the improvement in SSS. The tortuosity was larger for NT – especially for no CC. This indicates a more poorly connected macropore system in NT as compared to MP with higher macroporosity. Further, the results imply a larger proportion of marginal and remote pores in NT as

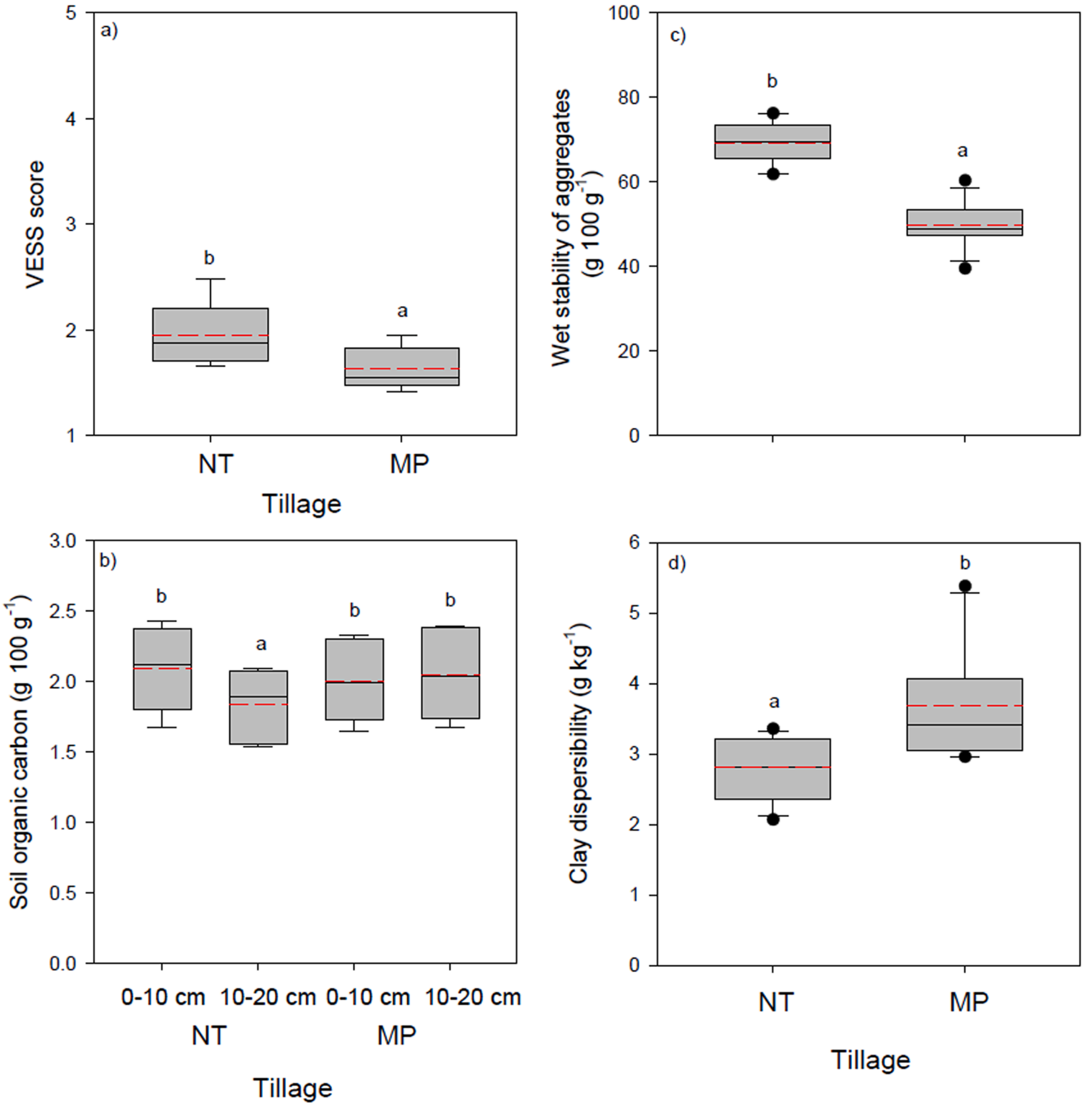

Fig. 1. Effects of tillage (no tillage [NT] and mouldboard ploughing [MP]) on (a) visual evaluation of soil structure (VESS), (b) soil organic carbon divided by depth (0–10 and 10–20 cm), (c) wet stability of aggregates and (d) clay dispersibility. Red dashed lines indicate mean values. Lines within the boxes represent median values, box boundaries include the 25th and 75th percentiles, and the whiskers extend from the box boundary to the 10th and 90th percentiles. Data points that lie outside the 10th and 90th percentiles are shown as symbols. Letters denote statistical significance at $P<0.05$.

compared to MP, which also could be the result of an increase in intra-aggregate pore space due to unrestricted aggregate development and hence stabilization. The VESS score was lower for MP than NT, but the NT plots also had a good soil structural stability with a score of 2, and the score for NT had decreased from 2.5 to 2 since the last sampling in 2012 (Abdollahi and Munkholm, 2014) indicating a further development in soil structural quality.

On the other hand, the NT treatment resulted in lower porosity, the fraction of soil volume represented by >30 μm pores and gas diffusivity as compared to the MP treatment. Hence, soil gas exchange may be reduced, and root growth may be negatively affected. This is not in agreement with an analysis of 14 studies showing that for studies running >20 years an increase in macroporosity for NT was found (Blanco-Canqui and Ruis, 2018).

4.2. Cover crop effect

Generally, cover cropping affected less parameters compared to tillage. Surprisingly, the change in SOC stock from 2012 to 2022 was not significantly affected by inclusion of a fodder radish cover crop, which probably was a result of the large variation between plots (Fig. 4). However, the difference between inclusion and no CC was 2.10 Mg C ha^{-1} (corresponding to a relative difference of 4%) resulting in a mean gain of 0.21 Mg C ha^{-1} year^{-1} from 2012 to 2022, which is at odds with Jensen et al. (2022) comparing inclusion of a ryegrass CC in continuous spring barley with no CC. A non-significant effect of cover cropping on

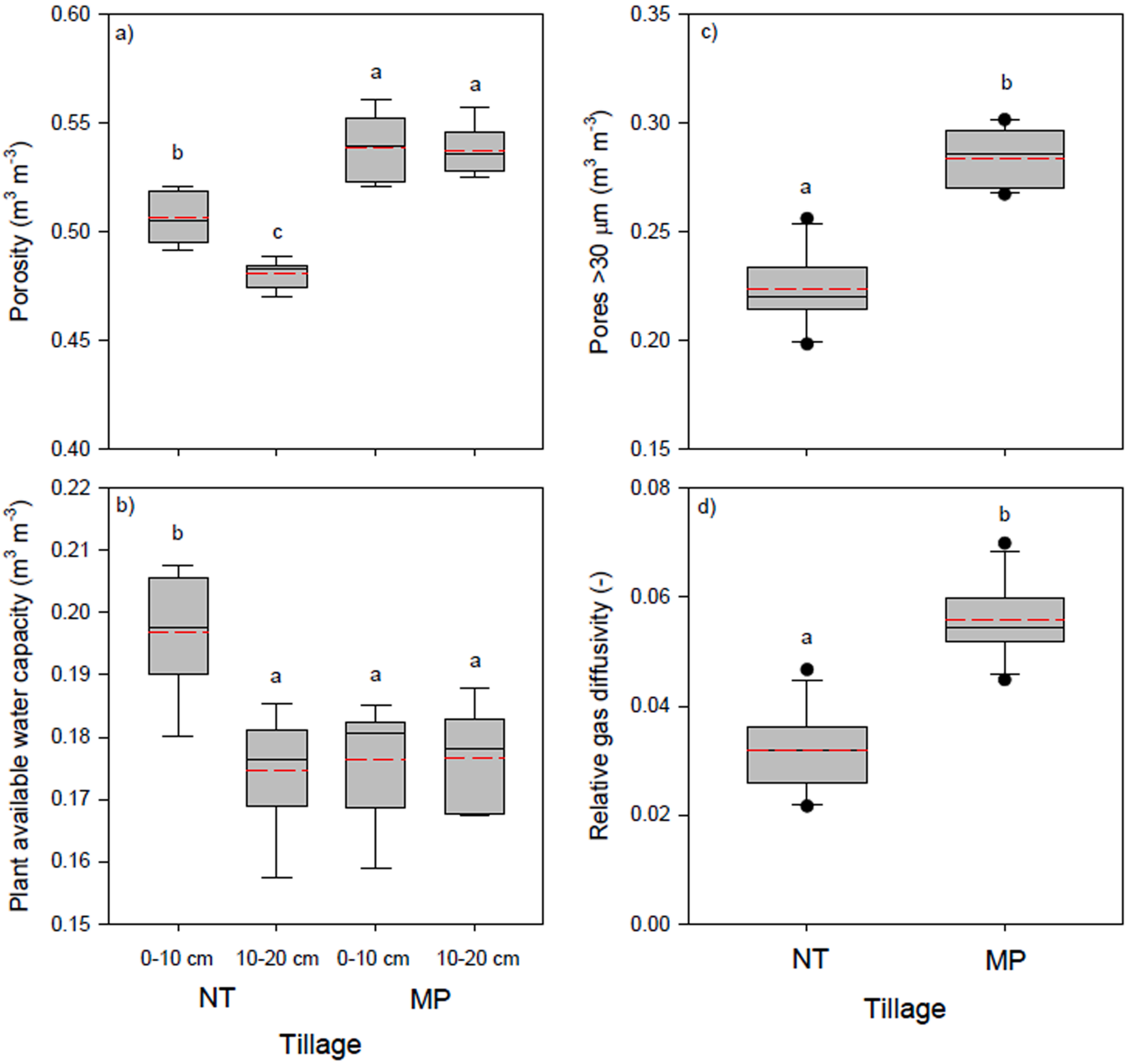

Fig. 2. Effects of tillage (no tillage [NT] and mouldboard ploughing [MP]) divided by depth (0–10 and 10–20 cm) on (a) total porosity and (b) plant available water capacity, and effects of tillage on (c) the fraction of soil volume represented by pores > 30 μm and (d) relative gas diffusivity at −10 kPa. Red dashed lines indicate mean values. Lines within the boxes represent median values, box boundaries include the 25th and 75th percentiles, and the whiskers extend from the box boundary to the 10th and 90th percentiles. Data points that lie outside the 10th and 90th percentiles are shown as symbols. Letters denote statistical significance at $P<0.05$.

SOC was also found by Jordon et al. (2022) in a meta-analysis study on regenerative agricultural practices in temperate soils from mainly Northwestern Europe, and in a recent opinion paper on effects of cover crops on SOC storage (Chaplot and Smith, 2023). Significant cover cropping induced gains in SOC of 7–9% has, however, been found in other recent global analysis (Hao et al., 2023; Joshi et al., 2023).

Soil structural quality was improved when a CC was grown, although the difference was small (0.3). Similarly, porosity, the fraction of soil volume represented by >30 μm pores, air permeability, pore organization and gas diffusivity were significantly larger when a CC was grown. All these parameters were either not significant or close to being significant in 2012 (Abdollahi and Munkholm, 2014; Abdollahi et al., 2014) following five years of CC inclusion. In 2022, CCs had been grown in 13 out of 15 years since 2007 (introduction of CC), indicating that soil pore characteristics continues to develop in the long-term when introducing CCs as a conservation agriculture element. A positive effect of cover cropping on soil structural properties agrees with reviews by Blanco-Canqui and Ruis (2020) and Hao et al. (2023) although they primarily found significant effects on soil strength and stability parameters and limited effects on soil pore characteristics. The reviews were primarily based on short-term studies and called for more long-term cover cropping studies. Our long-term study indicates that the duration of the experiment matters - especially with regards to soil pore characteristics.

4.3. Tillage and cover crop effects

We hypothesized a synergy effect of combining CC with NT by improving the positive effects of NT on SSS and SOC in the soil surface layer (0–10 cm) and by alleviating negative effects of NT on the soil pore system in the lower part of the arable layer (10–20 cm). No tillage improved, as anticipated, SSS of the surface layer, caused a stratification of SOC but not a significant increase in SOC as compared to MP. Cover cropping had, however, no significant effect on either SSS or SOC. Thus,

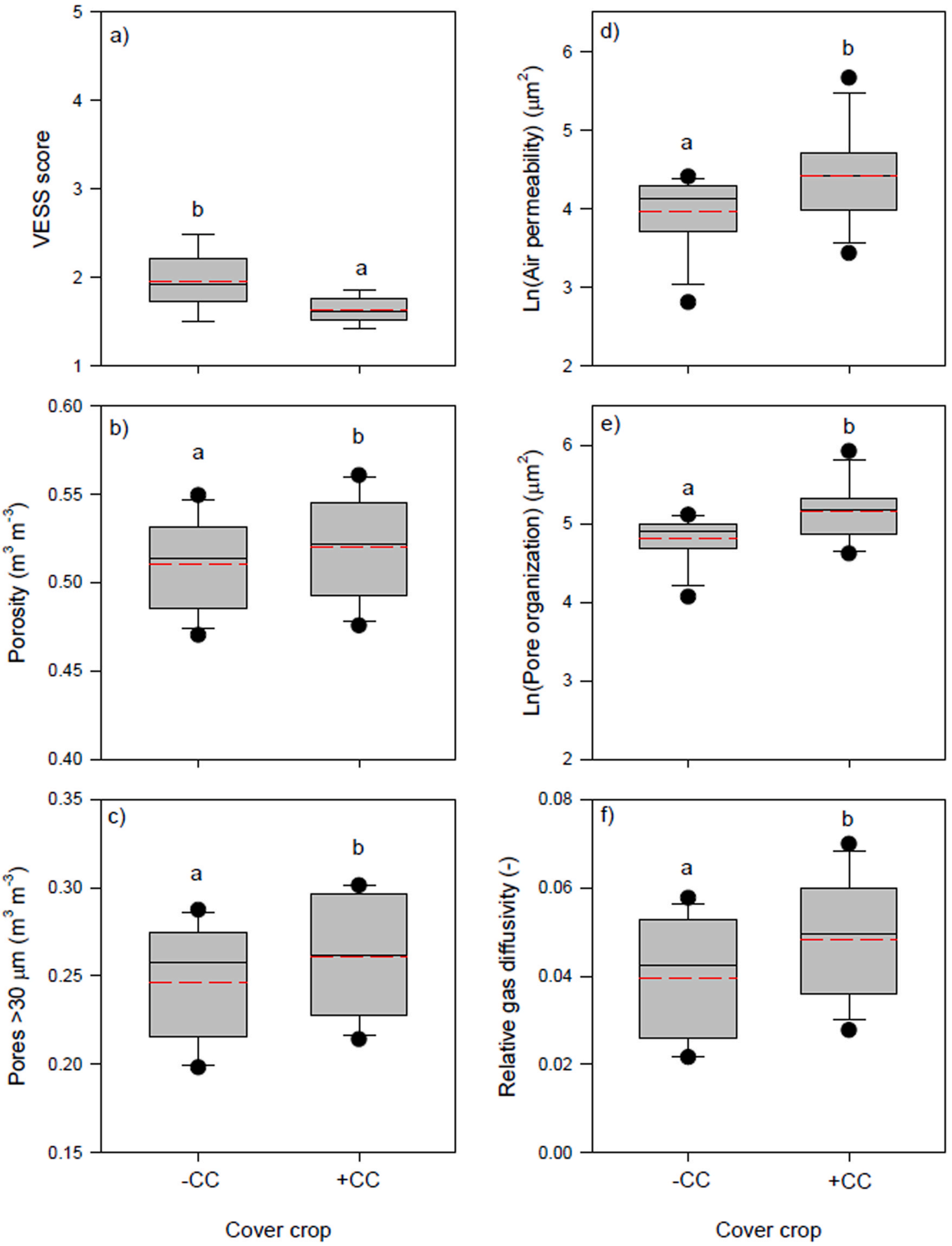

Fig. 3. Effects of cover crop (without [-CC] and with [+CC]) on (a) visual evaluation of soil structure (VESS), (b) total porosity, (c) the fraction of soil volume represented by pores > 30 µm, (d) the natural logarithm of air permeability at −10 kPa, (e) the natural logarithm of pore organization at −10 kPa and (f) relative gas diffusivity at −10 kPa. Red dashed lines indicate mean values. Lines within the boxes represent median values, box boundaries include the 25th and 75th percentiles, and the whiskers extend from the box boundary to the 10th and 90th percentiles. Data points that lie outside the 10th and 90th percentiles are shown as symbols. Letters denote statistical significance at $P<0.05$.

we were not able to confirm our hypothesized positive effect of combining NT and CC on SSS and SOC for the soil surface layer. On the other hand, CC improved visually evaluated soil structural quality (VESS) and the functionality of the soil macropore system for both the studied soil layers, while NT negatively affected these properties as compared to MP. Hence, we got confirmed that CCs have the potential to alleviate negative effects of NT on the soil pore system. However, it is worth noticing that the effect of CCs was less pronounced as compared to the effect of MP in many cases. Inclusion of CC could counteract the less good VESS score for NT as compared to MP, but CCs only increased the fraction of soil volume represented by >30 µm pores and gas diffusivity with 25 and 37%, respectively, while effects on porosity was

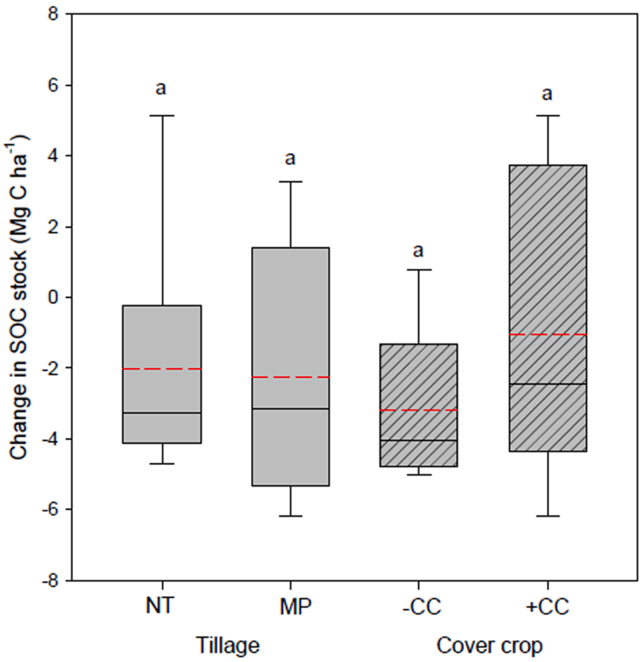

Fig. 4. Effects of tillage (no tillage [NT] and mouldboard ploughing [MP]) and cover crop (without [-CC] and with [+CC]) on the change in soil organic carbon (SOC) stock in 0–20 cm from 2012 to 2022. Red dashed lines indicate mean values. Lines within the boxes represent median values, box boundaries include the 25th and 75th percentiles, and the whiskers extend from the box boundary to the 10th and 90th percentiles. Data points that lie outside the 10th and 90th percentiles are shown as symbols. Letters denote statistical significance at $P<0.05$.

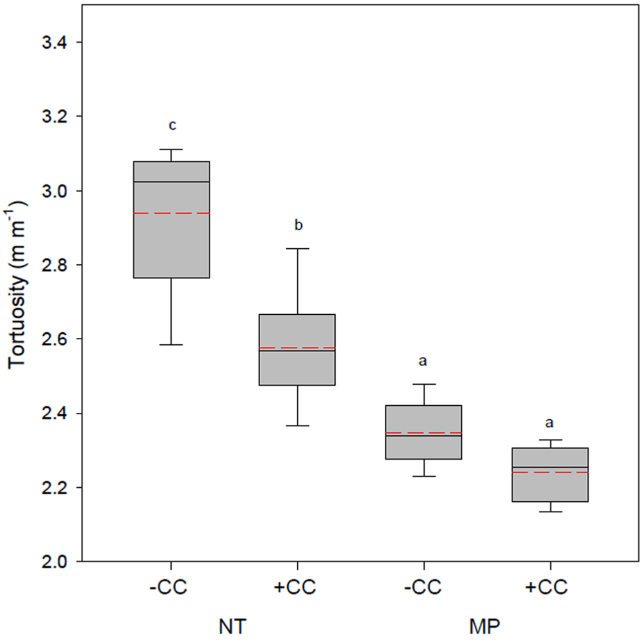

Fig. 5. Pairwise comparisons of the interaction between tillage (no tillage [NT] and mouldboard ploughing [MP]) and cover crop (without [-CC] and with [+CC]) on tortuosity. Red dashed lines indicate mean values. Lines within the boxes represent median values, box boundaries include the 25th and 75th percentiles, and the whiskers extend from the box boundary to the 10th and 90th percentiles. Data points that lie outside the 10th and 90th percentiles are shown as symbols. Letters denote statistical significance at $P<0.05$.

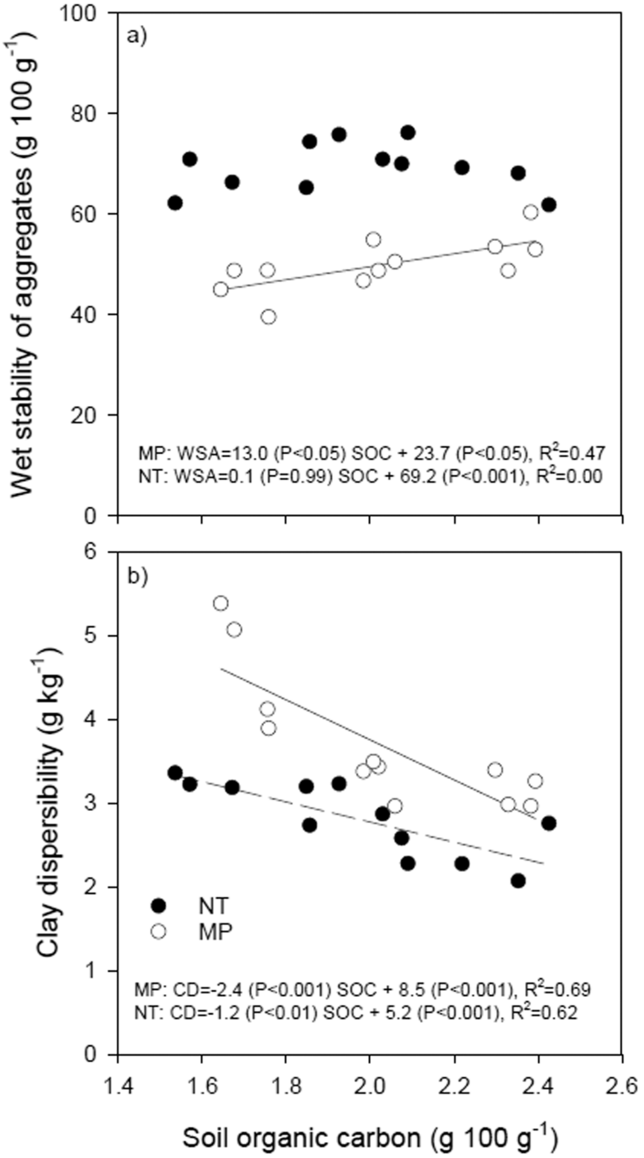

Fig. 6. (a) Wet stability of aggregates and (b) clay dispersibility as a function of soil organic carbon at individual sample level for the two tillage treatments. Black and white symbol fills highlight no tillage (NT) and mouldboard ploughing (MP), respectively. Each tillage treatment includes two cover crop treatments (with and without), two depths (0–10 and 10–20 cm) and three blocks providing a total of 12 samples. The linear regression models for individual tillage treatments are indicated and the regression lines are indicated if the slope of the regressions were significant.

counteracted by 34 and 19% in the 0–10 and 10–20 cm soil layers, respectively. However, air permeability and pore organization were affected by CC only entailing an improvement as compared to MP if NT is combined with CC and will result in a soil with pores >30 µm being more effective at conducting air by convection and having a higher degree of pore continuity.

Overall, our observed lack of synergy between NT and CC for SSS and SOC in the surface layer contrasts with observations summarized by Blanco-Canqui and Ruis (2018) and Blanco-Canqui and Ruis (2020), while the soil macropore results agree with their results.

5. Conclusions

We relied on a unique long-term field experiment with continuous cereal cropping to examine the impact of tillage (no till [NT] or mouldboard ploughing) and inclusion of a fodder radish cover crop (CC) on SOC, soil structural stability (SSS) and pore characteristics following two decades. Neither tillage nor cover cropping affected the SOC stock in the 0–20 cm soil layer. NT caused, however, a stratification of SOC in the top 0–20 cm layer as expected, i.e. higher SOC content at 0–10 cm than at 10–20 cm depth. Cover cropping did not, in contrast to what we hypothesized, increase the stratification effect of NT. No tillage improved SSS, PAWC in 0–10 cm depth and increased tortuosity as compared to ploughing. Notably, the marked improvement in SSS for NT soils was only to a very limited extent related to SOC, suggesting that the positive effects ascribed to absence of disturbance was the main driver. On the other hand, soil porosity, especially in 10–20 cm depth, the fraction of soil volume represented by >30 μm pores and gas diffusivity decreased and NT plots resulted in a less good VESS score (0–22 cm soil layer). The inclusion of CCs improved the VESS score, increased soil porosity, the fraction of soil volume represented by >30 μm pores, air permeability, pore organization and gas diffusivity. Hence, our results confirm, as hypothesized, that CCs can be used to alleviate the negative effects of NT on pore characteristics at the macroscale. Our study also emphasizes the value of long-term studies as we showed that the positive effects of NT on SSS and of CC on macropore characteristics were much more pronounced after long-term (20 yrs NT; 13 yrs CC) than after medium-term (10 yrs NT; 5 yrs CC) practices.

CRediT authorship contribution statement

Johannes L. Jensen: Writing – review & editing, Visualization, Supervision, Investigation, Funding acquisition, Formal analysis, Data curation, Conceptualization. **Sebastiano Rocco:** Writing – original draft, Formal analysis, Data curation, Conceptualization. **Lars J. Munkholm:** Writing – review & editing, Supervision, Resources, Project administration, Investigation, Conceptualization.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

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Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.still.2024.106129.

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