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Soil carbon stocks in different bioenergy cropping systems including subsoil

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ABSTRACT

Despite the growing importance of energy cropping systems, little is known about their soil organic carbon (SOC) stocks in topsoils and subsoils. Furthermore, information regarding the impact of N-fertilization on C-sequestration for perennial compared with annual energy cropping systems is scarce. In order to study SOC changes in the soils of different energy cropping systems, a long-term study was established in southwestern Germany with the following cropping systems: energy maize (*Zea mays*) with reduced tillage, Miscanthus (*M. x giganteus*), switchgrass (*Panicum virgatum*) and willow (*Salix schwerinii x viminalis*), as well as a crop rotation with conventional tillage (CT) and no-till (NT) consisting of oilseed rape (*Brassica napus* ssp. *oleifera*), winter triticale (*Triticale triticosecale*) and winter wheat (*Triticum aestivum*). The soil is a Haplic Luvisol (Siltic). For each cropping system three N-fertilization regimes adapted to the needs of each crop were applied. With the main hypothesis that perennial energy cropping systems increase SOC stocks compared with conventional annual cropping systems, the SOC stocks were analyzed to a depth of 90 cm after 11 years of continuous cropping (2002–2012). Compared with the control (CT) with 76 Mg SOC ha^{-1}, the perennial crops had significantly higher SOC stocks in all N-fertilization regimes, which amounted to 92–95 Mg SOC ha^{-1} in the N1-fertilization regime. The crop rotation with NT also had higher SOC stocks with 93 Mg SOC ha^{-1} in the N1-fertilization regime. N-fertilization generally led to higher SOC stocks in all cropping systems, although SOC stocks did not increase any further from reduced to highest crop-specific N-fertilization, with the exception of energy maize. The current findings also stress the importance of subsoil carbon analyses: SOC stocks at a depth from 30 to 90 cm made up 44–55% of the total stocks and differed significantly between cropping systems.

1. Introduction

Soil organic carbon (SOC) concentration and stock are considered significant soil parameters since their alterations affect not only the agricultural properties of the site itself, but also the global carbon cycle (Smith et al., 1993; Zeng et al., 2004). The potential capacity for all managed ecosystems as soil sinks is estimated to be 55–78 Gt CO_2 which equals the cumulative historic C loss from soils (Lal, 2004). Whether soils under human management serve as a source or sink for CO_2 depends strongly on the overall land management (forest, grassland, arable land and all intermediate management systems) as well as the implemented farming methods, such as reduced tillage or fertilization strategies using organic inputs (Smith, et al., 2008). Energy crops, designated to different pathways of conversion have widened the spectrum of cropping systems in the past decades (Wright, 2006). Whether such cropping systems have a negative carbon balance depends, among other aspects, on their ability to sequester carbon (Brandão et al., 2011).

Long-term studies on the changes in soil parameters of different bioenergy cropping systems below the topsoil (>30 cm depth) have rarely been conducted. There are indications that perennial crops increase SOC stocks as the extensive root system of perennial crops enmesh particles to aggregates and stimulate the formation of microbial polysaccharids that join particles together (Six et al., 2006). In addition, root exudates form organo-mineral complexes (Bolan et al., 2011) and promote microbial activity, which also promotes SOC sequestration (Miltner et al., 2012). Some authors

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(e.g. Shi et al., 2013; Syswerda et al., 2011) state that changes of SOC stocks due to management changes or the change from annual to perennial crops are more distinct in the topsoil (0–30 cm) than in deeper soil and tend to fade with increasing depth. However, the current debate on the role of subsoils for C-sequestration shows that the disregard of changes in subsoil SOC stocks can distort overall carbon sequestration rates (Liebig et al., 2005; Olson et al., 2005; Rasse et al., 2006). However, sampling of deeper soil is often avoided as it is more expensive and SOC changes are harder to detect as carbon concentrations are lower and more variable than in the topsoil due to pedogenic factors (Syswerda et al., 2011).

The availability of nitrogen (N) is an important factor affecting carbon dynamics in agricultural systems as N is often the limiting nutrient and determines biomass production, which therefore influences carbon flows. Moreover, the availability and form of N influences the composition and decomposition and eventually the stability of soil organic matter (Ludwig et al., 2011). Generally, an increased input of N leads to increased carbon accumulation until a new equilibrium is reached depending on pedogenic and climatic factors (Hellebrand et al., 2010; Jung and Lal, 2011; Mazzoncini et al., 2011).

To bring light to this knowledge gap, the aim of the current study is to evaluate the potential of perennial energy crops to sequester carbon in top- and subsoils, compared with annual energy crops, based on the rate of N-fertilization. In addition, for an annual energy crop rotation, different tillage systems (no-till vs conventional inversion tillage by moldboard plough) which affect C dynamics are analyzed.

Based on these research aims, the following hypotheses were set up:

a Perennial crops grown without tillage sequester more SOC than conventional tilled annual crops, which leads to significantly different overall SOC-stocks in the long term.
b No-till in annual cropping systems does not lead to an increase in SOC stocks compared to conventional tillage when the whole soil profile is considered.
c SOC stocks in subsoils do not differ significantly between energy cropping systems.
d Increased N-fertilization significantly increases the SOC stock in the topsoil in perennial and annual bioenergy cropping systems.

2. Material and methods

2.1. Site description and experimental design

This study was part of a long-term field trial located at the research station Ihinger Hof (48.75°N and 8.92°E, 480 m asl) of the University of Hohenheim, in southwestern Germany. Located in a fertile area where plane fields and gently sloping hills alternate, this site was agriculturally used for several centuries. The location is characterized by a mean annual temperature of 9.1 °C (ten-year average for 2002–2012) and a mean annual precipitation of 714 mm. The soil is a Haplic Luvisol (Siltic) (IUSS, 2007), (Table 1). Overall, the soil has an udic moisture regime and a mesic temperature regime. The slope on this site is below 1%. Before the trial was established in 2002, the soil was conventionally tilled with moldboard plough and showed a mean bulk density of 1.35 g cm^{-3} in the topsoil (0–30 cm) and 1.48 g cm^{-3} at 30–60 cm depth (Table 1).

The trial began in 2002 and the same management continued until 2012, when soil samples for the current study were taken. Six cropping systems for annual and perennial energy crops adapted to the environment in Central Europe were tested in four replications. Different rates of N-fertilization were included in a split plot design (Table 2). The selection of cropping systems reflects different plant-based energy production systems prevalent in Central Europe: (1) Combustion of the plant material, (2) fuel production and (3) biogas production. The 1st and 2nd cropping systems (Table 2) were comprised of one crop rotation including the annual crops winter oilseed rape, winter wheat and winter triticale with the application of two different tillage systems: conventional inversion tillage using a moldboard plough (CT, 20–30 cm) versus no-till (NT). The annual crop maize ('Mikado') in a maize monocropping system was selected as a 3rd system. Three perennial energy crops, Miscanthus, *switchgrass* and willow, (harvested every third year) were used to study both trees and grasses. Each cropping system was divided into three N-fertilization regimes (split-plot design). Each subplot had a size of 160 m^2 and was replicated four times. Spaces between blocks were 18 m, while split-plots had 4 m buffer between each other. Within each subplot, sampling was done with at least a 2 m buffer to the border. N-fertilizer regimes were adapted to the N-demand of the different crops and included a control with no N-fertilization (N0), a reduced fertilization regime (N1) and a fertilization regime based on the current agricultural practice (N2) (Table 2). The ammonium-stabilized N-fertilizer Entec 26 (K+S Nitrogen GmbH, Mannheim, Germany), containing 75 g kg^{-1} nitrate-N, 185 g kg^{-1} ammonium-N and 130 g kg^{-1} sulphur was used throughout the study. Phosphorus (P), magnesium (Mg), potassium (K) and sulphur (S) were applied to maintain a sufficient supply of plant available nutrients based on soil tests and in accordance with good agricultural practice.

2.2. Soil analyses

Soil samples were taken to a depth of 90 cm at the beginning of the study in March 2002 after the soil was ploughed 5 months before and again, 11 years later, in March 2013. Soil sampling was done with a soil probe three times in every subplot to a depth of 90 cm. The sample was divided into 0–30 cm depth, 30–60 cm depth and 60–90 cm depth. All three samples within each subplot were merged to one composite sample. In 2013, a second sampling was conducted for the topsoil (0–10 cm, 10–20 cm and 20–30 cm) in the described manner to gain a higher resolution of soil characteristics in the topsoil. All soil samples were air-dried and sieved to 2 mm. Total C (C_t) and total N were determined by combustion (Vario Max CNTS, Elementar). In 2013, the samples from 0 to 90 cm depth with 30 cm increments were treated with

Table 1
Soil characteristics of a Haplic Luvisol (Siltic) at the trial site (Particle-size classes according to FAO (2006)).

Horizon	Depth [cm]	Sand 2–0,063 mm [g kg^{-1}]	Silt <0,063–0002 mm [g kg^{-1}]	Clay <0,002 mm [g kg^{-1}]	Bulk density [g cm^{-3}]	Effective porosity [cm^3 cm^{-3}]	Field capacity [cm^3 cm^{-3}]	Effective field capacity [cm^3 cm^{-3}]	pH
Ap	−30	24	748	228	1.55	0.047	0.369	0.206	7.38
Bt1	−50	22	743	235	1.50	0.042	0.391	0.148	7.15
Bt2	−73	25	764	211	1.50	0.043	0.390	0.187	7.05
BCg	−112	15	846	139	1.54	0.048	0.368	0.206	7.34

Table 2
Cropping systems, N-fertilization regimes and tillage systems used for the 11 year long-term study on energy crops in Central Europe, University of Hohenheim, Germany (S 1 = conventional tillage as reference system).

System	Crops	Factors		
		N-fertilization N0–N1–N2 [$kg\,N\,ha^{-1}\,a^{-1}$]	Tillage	Annual/perennial
S1	–Winter oilseed rape (*Brassica napus* ssp. *oleifera*) –Winter wheat (Triticum aestivum) –Winter triticale (Triticale triticosecale)	0–120–240 0–80–160 0–80–160	Conventional till (CT): moldboard plough, 22 cm deep	Annual (crop rotation)
S2	See No. 1	See No. 1	No-till (NT)	See No. 1
S3	Maize *(Zea mays)*	0–120–240	Reduced till (RT): rotary harrow, 7 cm deep	Annual (monocropping)
S4	Miscanthus (*Miscanthus x giganteus)*	0–40–80	None	Perennial
S5	Switchgrass (*Panicum virgatum*)	0–40–80	None	Perennial
S6	Willow (*Salix schwerinii x viminalis*)	0–40–80	None	Perennial

10% HCl to eliminate potential carbonates before measuring SOC (VDLUFA, 2012). Carbonate concentration in the topsoil samples (0–30 cm) was measured by a volumetric determination of CO_2 from soil samples treated with 10% HCl (DIN ISO 10693, 1997).

Bulk densities were determined after drying the samples with 105 °C for 48 h by the core method (DIN ISO 11461, 2002). 100 cm^3 core samples were taken in the topsoil in the N1 subplots of each cropping system at a depth of 0–10, 10–20 and 20–30 cm. Core samples of the bulk densities from 30 to 60 cm and from 60 to 90 cm were taken at a representative position within the field trial.

Amounts of SOC were expressed as concentrations (g C kg^{-1}) and then recalculated to stocks (t C ha^{-1}) using the specific bulk densities. Since the soil was free of stones and gravel, no correction for particle size fractions >2 mm was necessary.

2.3. Calculation of carbon stocks

Since differing bulk densities were found in the topsoil, the calculation of SOC stocks was conducted with a soil mass correction to gain equivalent soil masses using the method published by Ellert and Bettany (1995) and modified by Lee et al. (2009). The difference in bulk densities in the topsoils, compared with standard conventional tillage management, resulted from different tillage methods, or their absence, respectively. The bulk densitiy of the CT treatment was set as standard (standard layer) and soil masses and SOC stocks of the other cropping systems were calculated accordingly (Eqs. (1)–(3)).

Since the CT treatment had the lowest bulk density, part of the soil mass (and SOC stock) in the other cropping systems had to be subtracted from the first sampling depth (in the following named as first layer) and added to the second layer. Subsequently, the difference in soil mass compared with the standard second layer, now expanded, was again subtracted and added to the third layer. In the deepest layer, the remaining bulge was cut off (since no deeper layer was measured), as if the sampling depth had been shallower, which led to slightly smaller sampling layers with the result of equal soil masses among all treatments.

The soil mass of the different trial treatments was calculated as:

Eq. (1):

$$M_i = BD_i \cdot T_i \cdot 0.01 \quad (1)$$

with M_i = soil mass of layer i [Mg ha^{-1}]
BD_i = bulk density of layer i [g cm^{-3}]
T_i = thickness of layer i [cm]
0.01 = unit conversion factor [from g cm^{-2} to Mg ha^{-1}]

Corrected SOC stocks in the first layers (0–10 cm) of the different treatments were calculated as:

Eq. (2):

$$C_1 = (SOC_1 \cdot M_1) - (SOC_1 \cdot \Delta M_1) \quad (2)$$

with C_1 = SOC stock of first layer [Mg ha^{-1} dm^{-1}]
SOC_1 = SOC concentration of first layer [%]
M_1 = soil mass of first layer [Mg ha^{-1}]
ΔM_1 = soil mass difference of first layer to the standard [Mg ha^{-1}]

Corrected SOC stocks in the sub layers (>10 cm) of the different treatments were calculated as:

Eq. (3):

$$C_i = (SOC_i \cdot M_i) + (SOC_{i+1} \cdot \Delta M_{i+1}) - (SOC_i \cdot ((M_i + \Delta M_{i+1}) - ESM_i)) \quad (3)$$

with SOC_i = SOC concentration of layer i [%]
SOC_{i+1} = SOC concentration of above layer [%]
M_i = soil mass of layer i [Mg ha^{-1}]
ΔM_{i+1} = soil mass formerly subtracted from the above layer [Mg ha^{-1}]
ESM_i = soil mass of the corresponding standard layer i (CT treatment) [Mg ha^{-1}]

The general concept can be found in (Lee et al., 2009).

2.4. Statistical analysis

Data analysis was performed with SAS 9.2 (SAS Institute Inc., Cary, NC, USA). A mixed model was used with block, crop and N-fertilization regimes, interactions of these as fixed effects, and the whole plot error (interaction of depth block and crop) as a repeated random effect. The degrees of freedom for testing fixed effects were estimated by the Kenward-Rogers approximation. The whole plot error was evaluated using unstructured covariance estimation.

All effects were tested at a *p* value of 0.05. Homogeneity of variance and normal distribution were verified; no data transformation was necessary. The equation of the mixed model reads as follows:

$$y_{ijrkt} = m + b_r + a_k + cna_{ijk} + cb(a)_{irk} + cnba_{ijrkt}$$

with
y_{ijrkt} = *t*-th measurement in *r*-th replication in *k*-th depth of *i*-th crop with *j*-th N regime
m = general effect
b_r = main effect of *r*-th replication
a_k = main effect of *k*-th depth
cna_{ijk} = interaction of *i*-th crop with the *j*-th N regime and the *k*-th depth
$cb(a)_{irk}$ = random deviation of *i*-th crop in *r*-th replication in each depth
$cnba_{ijrkt}$ = residual error term corresponding to y_{ijrkt}

3. Results

3.1. Bulk density

After 11 years of altered soil management, bulk density was highest in the top 10 cm of NT treatment and lowest in the CT treatment (Table 3). The comparatively high bulk density in the NT treatment may indicate a compaction of the first layer.

3.2. Gravimetric SOC concentrations in 2013

Mean gravimetric SOC concentrations in 2013 varied in the layer 0–10 cm between 10 and 17 g C kg^{-1} with the highest values measured in the N1-fertilization regime of Miscanthus and the N2-fertilization regime of willow (Table 4). Lowest mean SOC concentrations were found in the CT treatments. In the following layer from 10 to 20 cm, the highest mean SOC concentration was found in the N1-fertilization regime of the CT treatment, while the lowest mean was measured in the N0-fertilization regime of the NT crop rotation. In the deeper layers a similar pattern was observed, with the perennial crops and the NT crop rotation having mostly higher SOC concentrations than the CT rotation and maize.

3.3. SOC stocks in 2013

No general statistical relation of increasing SOC stock with increasing N-fertilization over all three N-fertilization regimes could be established as the interactions between N-fertilization, crop and depth were significant in the statistical model. However, the N0-fertilization regime led to the lowest total SOC stocks in all energy cropping systems except willow, even though this effect was not significant for each treatment (Table 5). Significantly lower SOC stocks for the N1-fertilization regime compared with the N2-fertilization regimes could not be detected for all cropping systems. For instance, in the crop rotation with NT, the N1-fertilization regime led to significantly higher SOC stocks than the N2-fertilization regime. These findings emphasize the necessity for the effect of N-fertilization to be analyzed separately for every energy-cropping system.

For all tested cropping systems, highest SOC stocks were measured in the first layer (0–10 cm), with the exception of the CT treatment, where a higher SOC stock was found in the 10–20 cm layer. In the soil depth 10–20 cm and 20–30 cm, the mean SOC stocks behaved in opposition to the top 10 cm: in these layers, the CT treatment showed partly higher SOC stocks. In the layers below 30 cm depth, NT again showed increased SOC stocks compared with the CT treatment; however, these were only significant in the N1-fertilization regime (Table 5).

The total mean SOC stock to a depth of 90 cm was highest in the N1-fertilization regime of Miscanthus, followed by the N2-fertilization regime of switchgrass and the N1-fertilization regime of willow. The Maize treatment in the N0- and N1-fertilization regimes and the CT treatment in all three fertilization regimes had significantly lower total SOC stocks than the perennial crops in the N0-, N1- and N2-fertilization regime (Table 5). The crop rotation with CT showed the lowest total SOC stock in all fertilization regimes. However, no statistical difference was found compared with the maize treatment in the N0 and N1-fertilization regimes. Contrary to these findings, under the N2-fertilization regime, the maize treatment showed a significantly higher SOC stock compared with the CT treatment.

In the N0- and N1-fertilization regimes, tillage had a significant effect on total SOC stocks: the CT and maize treatments had the lowest mean total SOC stocks, while the crop rotation with NT and the perennial cropping systems had significantly higher total SOC stocks. Furthermore, statistically significant differences between no-till annual cropping systems and perennial cropping systems could not be established. For the perennial cropping systems, no statistically significant differences were found.

3.4. Total carbon stock comparison between 2002 and 2013

As C_t was determined in 2002 without analyzing the carbonate concentration, soil deeper than 60 cm had to be excluded, since the higher and fluctuating carbonate concentrations in this depth leads to high inaccuracies. However, since carbonate concentrations were lower in the soil layers above 60 cm (<1 g kg^{-1}), the C_t stocks of these layers were compared between 2002 and 2013 to determine the general direction of carbon depletion or sequestration at this site.

The comparison of total gravimetric C stocks at the beginning of the study in 2002 and in 2013 revealed significant differences in some cropping systems. Although all fertilization regimes of the CT and NT rotations and also the N0- and N1-fertilized maize treatments showed no significant differences between 2002 and 2013 in the topsoil (0–30 cm), the N2-fertilized maize treatments and all treatments with perennial crops showed significantly increased total C stocks after 11 years of continuous management (Table 6).

Even in the subsoil (30–60 cm), tillage influenced SOC stocks: while SOC stocks with CT treatment did not differ when compared with 2002, the fertilized NT treatment had significantly higher C stocks in 2013. In addition, the perennial crops, with the exception of N0- and N2-fertilized willow treatments, had significantly increased total C stocks in the subsoil compared with 2002. In the maize treatment, the N2-fertilization regime only had a significant effect in the subsoil: the total C stock was higher in 2013 compared with 2002.

4. Discussion

4.1. The influence of cropping system, climate and soil on SOC stocks

SOC stocks illustrate carbon stratification according to soil depth, these stocks can be related to different site and management characteristics. Relatively low bulk densities suggest good

Table 3
Mean bulk density [g cm^{-3}] and standard deviation (±) in treatments with reduced N-fertilization in 2013 (0–30 cm: 2 replicate each with 3 samples; 30–90 cm: 1 replicate with 3 samples).

Depth [cm]	$CR_{ploughed}$ (CT)	$CR_{no\text{-}till}$ (NT)	$Maize_{reduced\text{-}till}$	Miscanthus [g cm^{-3}]	Switchgrass	Willow
0–10	1.21 ± 0.10	1.46 ± 0.07	1.26 ± 0.12	1.35 ± 0.16	1.30 ± 0.06	1.27 ± 0.04
10–20	1.32 ± 0.07	1.47 ± 0.06	1.41 ± 0.04	1.42 ± 0.06	1.45 ± 0.07	1.35 ± 0.03
20–30	1.49 ± 0.07	1.43 ± 0.03	1.45 ± 0.06	1.52 ± 0.06	1.41 ± 0.05	1.45 ± 0.08
30–60[a]	1.50	1.50	1.50	1.50	1.50	1.50
60–90[a]	1.54	1.54	1.54	1.54	1.54	1.54

[a] From one representative position in the field.

Table 4
Mean gravimetric SOC concentrations in the soil under the different energy cropping systems 11 years after trial establishment (± standard deviation).

Depth [cm]	Crop	No N-fertilizer input (N0)	Reduced N-fertilizer input (N1) [g C kg^{-1}]	High N-fertilizer input (N2)
0–10	$CR_{ploughed}$ (CT)	10.30 ± 0.6	13.30 ± 0.8	13.10 ± 1.0
	$CR_{no\text{-}till}$ (NT)	11.80 ± 0.7	10.90 ± 0.3	11.10 ± 0.6
	$Maize_{reduced\text{-}till}$	11.10 ± 0.6	12.10 ± 0.4	13.70 ± 0.7
	Miscanthus	13.20 ± 1.0	17.40 ± 2.1	16.60 ± 1.9
	Switchgrass	13.30 ± 0.4	13.50 ± 0.4	15.20 ± 1.4
	Willow	15.20 ± 2.1	16.60 ± 2.1	17.00 ± 1.6
10–20	$CR_{ploughed}$ (CT)	10.10 ± 0.5	9.50 ± 1.0	9.20 ± 0.5
	$CR_{no\text{-}till}$ (NT)	8.90 ± 0.5	11.10 ± 0.4	10.80 ± 0.3
	$Maize_{reduced\text{-}till}$	9.90 ± 0.8	10.30 ± 0.8	10.60 ± 0.3
	Miscanthus	9.80 ± 0.4	9.90 ± 0.3	10.00 ± 0.8
	Switchgrass	9.90 ± 0.2	9.60 ± 0.4v	10.30 ± 0.6
	Willow	9.60 ± 0.2	9.90 ± 0.7	9.60 ± 0.6
20–30	$CR_{ploughed}$ (CT)	7.80 ± 1.7	8.00 ± 0.6	7.60 ± 0.7
	$CR_{no\text{-}till}$ (NT)	7.70 ± 0.7	8.60 ± 1.3	8.20 ± 1.3
	$Maize_{reduced\text{-}till}$	9.00 ± 0.5	9.20 ± 0.9	9.50 ± 0.4
	Miscanthus	8.60 ± 0.7	8.80 ± 0.7	8.90 ± 0.6
	Switchgrass	8.50 ± 0.8	8.90 ± 0.4	9.10 ± 0.5
	Willow	8.60 ± 0.4	9.00 ± 0.5	8.30 ± 0.4
30–60	$CR_{ploughed}$ (CT)	4.20 ± 0.3	6.30 ± 1.2	5.00 ± 0.8
	$CR_{no\text{-}till}$ (NT)	4.80 ± 1.0	4.30 ± 1.0	4.10 ± 0.5
	$Maize_{reduced\text{-}till}$	3.80 ± 0.4	4.00 ± 0.3	4.80 ± 0.6
	Miscanthus	5.50 ± 0.3	5.70 ± 0.9	5.30 ± 0.6
	Switchgrass	7.00 ± 2.0	5.10 ± 0.9	5.90 ± 0.7
	Willow	5.70 ± 0.7	5.80 ± 0.4	5.50 ± 1.0
60–90	$CR_{ploughed}$ (CT)	3.40 ± 0.3	4.60 ± 0.4	3.50 ± 0.5
	$CR_{no\text{-}till}$ (NT)	4.70 ± 1.0	3.40 ± 0.2	3.50 ± 0.2
	$Maize_{reduced\text{-}till}$	3.10 ± 0.1	3.30 ± 0.4	3.90 ± 0.5
	Miscanthus	3.70 ± 0.4	4.20 ± 0.5	3.70 ± 0.1
	Switchgrass	4.20 ± 0.5	3.70 ± 0.6	4.60 ± 0.6
	Willow	4.30 ± 0.5	4.10 ± 0.1	3.90 ± 0.9

water infiltration and aeration, high temperatures during the vegetation period and many easily degradable compounds in the fresh organic matter ensure a high rate of decomposition (Syswerda et al., 2011; Fontaine et al., 2007). As expected, crops with a high amount of litterfall and larger C/N ratios, connected with higher lignin contents (Foereid et al., 2004) and lower turnover rates, such as Miscanthus and willow, had the highest SOC stocks in the topsoil. A recent study by Tiemann and Grandy (2015) found evidence for a higher microbial activity under perennial crops compared with annual cropping systems, which probably leads to a different aggregate formation and stabilization and thus contribute to higher SOC stocks. On the other hand, the CT treatments had the lowest SOC stocks in the topsoil, since plant residues were continuously incorporated throughout the whole ploughing depth, which may have a dilution effect on SOC stocks. A recent study by Wiesmeier et al. (2014) found that, in contrast to forest soils, only 50% of the potential SOC storage capacity of cropland soils in southern Germany was reached. This offers the opportunity to increase carbon storage of agricultural soils by altering the management systems and the incorporation of new crops.

Flessa et al. (2008) found SOC stocks in the same range as in this current study in similar climates in long-term maize cropping systems with 78 and 97 Mg SOC ha^{-1} on two sites in Germany with a sampling depth of 65 cm.

Liebig et al. (2005) measured higher SOC stocks of about 160 Mg SOC ha^{-1} to a depth of 120 cm in switchgrass and in crop rotations with annual crops in the northern Great Plains and in the northern cornbelt of the US; however, switchgrass had higher SOC stocks compared with different arable crops. The overall higher SOC stock in the study by Liebig is probably derived from the continental climate at the studied sites. The strength of the current study lies in the comparison of different systems at one site, since contrasting climates, cropping history and textures have a strong influence on SOC stocks and their dynamics (Saiz et al., 2012). Therefore the rate of carbon sequestration and the absolute stocks found in this study are only conferrable to sites with similar climate and soil conditions.

4.2. The interaction of cropping system with N-fertilization

After 11 years of continuous cropping with different cropping systems and continuous rates of N-fertilizer input, the SOC stocks in the top 10 cm generally increased. Jung and Lal (2011) observed an increase of SOC by about 2 Mg ha^{-1} $year^{-1}$ in highly fertilized switchgrass stands in the northern US. Mazzanconi (2011) also found that with increasing N-fertilization SOC accumulation is likely to be accelerated in the top 30 cm, especially under NT conditions. The generally lower SOC stocks in the N0-fertilization regime compared with the N1- or N2-fertilization regime emphasize the role of N in biomass production, carbon turnover and accumulation. Increased SOC stocks through N-fertilization could also be found by Hellebrand et al. (2010) in treatments with willow (annual SOC stock increase of 0.34 and 0.22 Mg ha^{-1} a^{-1} in willow fertilized with 150 kg N ha^{-1} a^{-1} and in unfertilized willow, respectively) and poplar (annual SOC stock increase of 0.53 and 0.23 Mg ha^{-1} a^{-1} in poplar fertilized with 150 kg N ha^{-1} a^{-1} and in unfertilized poplar, respectively).

However, the N2-fertilization regime did not lead to significantly higher SOC stocks than the N1-fertilization regime of the

Table 5
SOC stocks after 11 years of different cropping systems (Mg SOC ha-1 layer-1; ±standard error); different letters indicate significant differences of the mean SOC stocks within one layer ($\alpha = 0.05$).

Depth [cm]	Crop	No N-fertilizer input (N0)	Reduced N-fertilizer input (N1) [Mg SOC ha^{-1} $layer^{-1}$]	High N-fertilizer input (N2)
0–10	$CR_{ploughed}$ (CT)	$12.5^{a} \pm 0.7$	$13.2^{ab} \pm 0.7$	$13.5^{ab} \pm 0.7$
	$CR_{no\text{-}till}$ (NT)	$14.4^{abc} \pm 0.7$	$16.1^{cd} \pm 0.7$	$15.9^{cd} \pm 0.7$
	$Maize_{reduced\text{-}till}$	$13.5^{ab} \pm 0.7$	$14.7^{bcd} \pm 0.7$	$16.5^{def} \pm 0.7$
	Miscanthus	$16.0^{cd} \pm 0.7$	$21.1^{h} \pm 0.7$	$20.1^{gh} \pm 0.7$
	Switchgrass	$16.1^{cd} \pm 0.7$	$16.4^{de} \pm 0.7$	$18.4^{fgh} \pm 0.7$
	Willow	$18.4^{efg} \pm 0.7$	$20.1^{gh} \pm 0.7$	$20.6^{h} \pm 0.7$
10–20	$CR_{ploughed}$ (CT)	$13.4^{bcde} \pm 0.3$	$14.7^{g} \pm 0.3$	$14.2^{fg} \pm 0.3$
	$CR_{no\text{-}till}$ (NT)	$12.4^{a} \pm 0.3$	$13.5^{bcdef} \pm 0.3$	$13.1^{abc} \pm 0.3$
	$Maize_{reduced\text{-}till}$	$13.1^{ab} \pm 0.3$	$13.7^{bcdefg} \pm 0.3$	$14.1^{efg} \pm 0.3$
	Miscanthus	$13.4^{bcdef} \pm 0.3$	$14.0^{cdefg} \pm 0.3$	$14.0d^{efg} \pm 0.3$
	Switchgrass	$13.4^{bcdef} \pm 0.3$	$13.1^{ab} \pm 0.3$	$14.0^{defg} \pm 0.3$
	Willow	$12.9^{ab} \pm 0.3$	$13.4^{bcdef} \pm 0.3$	$13.1^{abcd} \pm 0.3$
20–30	$CR_{ploughed}$ (CT)	$11.6^{a} \pm 0.5$	$12.8^{abcdef} \pm 0.5$	$12.2^{abcd} \pm 0.5$
	$CR_{no\text{-}till}$ (NT)	$11.9^{ab} \pm 0.5$	$12.5^{abcde} \pm 0.5$	$12.0^{abc} \pm 0.5$
	$Maize_{reduced\text{-}till}$	$13.5^{defg} \pm 0.5$	$13.9^{efg} \pm 0.5$	$14.2^{fg} \pm 0.5$
	Miscanthus	$13.1^{bcdef} \pm 0.5$	$13.4^{cdefg} \pm 0.5$	$13.6^{defg} \pm 0.5$
	Switchgrass	$12.9^{abcdef} \pm 0.5$	$13.4^{cdefg} \pm 0.5$	$13.8^{efg} \pm 0.5$
	Willow	$14.1^{fg} \pm 0.5$	$14.6^{g} \pm 0.5$	$13.6^{defg} \pm 0.5$
30–60	$CR_{ploughed}$ (CT)	$19.1^{abc} \pm 1.7$	$19.3^{abc} \pm 1.7$	$18.7^{ab} \pm 1.7$
	$CR_{no\text{-}till}$ (NT)	$22.4^{abcde} \pm 1.7$	$29.0^{g} \pm 1.7$	$23.4^{bcdef} \pm 1.7$
	$Maize_{reduced\text{-}till}$	$17.7^{ab} \pm 1.7$	$18.6^{a} \pm 1.7$	$21.9^{abcd} \pm 1.7$
	Miscanthus	$25.5^{defg} \pm 1.7$	$26.4^{defg} \pm 1.7$	$24.7^{defg} \pm 1.7$
	Switchgrass	$27.4^{fg} \pm 1.7$	$23.4^{cdef} \pm 1.7$	$26.8^{efg} \pm 1.7$
	Willow	$25.6^{defg} \pm 1.7$	$26.4^{defg} \pm 1.7$	$25.0^{defg} \pm 1.7$
60–90	$CR_{ploughed}$ (CT)	$15.6^{ab} \pm 2.3$	$15.7^{ab} \pm 2.3$	$16.2^{ab} \pm 2.3$
	$CR_{no\text{-}till}$ (NT)	$21.7^{bc} \pm 2.3$	$21.9^{bc} \pm 2.3$	$16.7^{ab} \pm 2.3$
	$Maize_{reduced\text{-}till}$	$14.2^{a} \pm 2.3$	$15.5^{a} \pm 2.3$	$18.0^{ab} \pm 2.3$
	Miscanthus	$17.5^{ab} \pm 2.3$	$19.9^{abc} \pm 2.3$	$17.5^{ab} \pm 2.3$
	Switchgrass	$19.6^{abc} \pm 2.3$	$25.3^{c} \pm 2.3$	$21.2^{bc} \pm 2.3$
	Willow	$19.7^{abc} \pm 2.3$	$18.9^{abc} \pm 2.3$	$18.1^{ab} \pm 2.3$
0–90	$CR_{ploughed}$ (CT)	$72.2^{a} \pm 2.7$	$75.7^{ab} \pm 2.7$	$74.8^{ab} \pm 2.7$
	$CR_{no\text{-}till}$ (NT)	$82.8^{cde} \pm 2.7$	$93.1^{g} \pm 2.7$	$81.1^{bcd} \pm 2.7$
	$Maize_{reduced\text{-}till}$	$72.0^{a} \pm 2.7$	$76.4^{abc} \pm 2.7$	$84.8^{def} \pm 2.7$
	Miscanthus	$85.4^{def} \pm 2.7$	$94.8^{g} \pm 2.7$	$90.0^{efg} \pm 2.7$
	Switchgrass	$89.4^{defg} \pm 2.7$	$91.6^{fg} \pm 2.7$	$94.3^{g} \pm 2.7$
	Willow	$90.7^{defg} \pm 2.7$	$93.5^{fg} \pm 2.7$	$90.5^{defg} \pm 2.7$

respective cropping systems, with the exception of continuous maize cropping. Continuous maize significantly increased SOC stocks at the highest N-fertilization compared with the N1 fertilization regime. In comparison with other annual crops used in this study, energy maize produces the highest biomass above-ground (data not shown). This enables the plant to use high rates of N-fertilizer efficiently. In addition, compared with the perennial crops in the current study, breeding efforts for maize have been intensive in the last decades resulting in cultivars with high N efficiency.

Increased N-fertilization generally leads to increased soil emissions of N_2O. N_2O has a GHG equivalent of 298 (IPCC, 2006), therefore N_2O emissions can easily diminish positive effects of carbon sequestration (Hellebrand et al., 2010). If a life cycle approach is used, further drawbacks may be become evident, e.g. production of N-fertilizer by the Haber-Bosch technology is very energy-intensive, and CO_2 emissions caused by fertilizer production might also outweigh the effect of C-sequestration by increased biomass production. Measurements of trace gas fluxes in selected plots of the same trial in 2010 provide additional information about soil derived emissions: maize in the N2-fertilization regime emitted 3.6 kg N_2O ha^{-1} yr^{-1}, which was more than the double the emission from Miscanthus treatments. Willow, however, had N_2O emissions close to zero in the N2-fertilization regime, which illustrates the efficient uptake of N-fertilizer by trees and understory vegetation (Gauder et al., 2012). Even though yields in most cropping systems grew with increasing N-fertilization (N1 to N2) in this trial (results not shown), this increase in above ground biomass did not show significant effects on C storage in soils. Therefore, a reduced fertilization regime seems to be sufficient to induce high C accumulation rates, in particular in perennial crops.

4.3. SOC-sequestration in top- and subsoil

Even if the total SOC stocks were the same as, or at least similar between most of the studied systems, the distribution of SOC with soil depth differed. Maize treatment had about 55% of the SOC stored in the top 30 cm, which was significantly higher than in all other cropping systems. Specifically, the NT crop rotation and switchgrass had a high share of SOC stored below 60 cm. This could hint at higher root activity in the subsoils of these treatments compared with the other tested crops, since root exudates act as C sources and can have a negative priming effect on microbes (slowdown of SOC decomposition) (Fu and Cheng, 2002; Hamer and Marschner, 2005). De Graaf et al. (2014) found that the roots of switchgrass induced negative priming effects below 40 cm depth. Currently, data on the effect of crop management systems on

Table 6
Comparison of gravimetric C stocks (Mg SOC ha^{-1} layer^{-1}) before planting (2002) and after 11 years of different cropping systems (2013) for the depth between 0 and 30 cm and 30–60 cm (±standard error; stars indicate significant differences with $\alpha > 0.05$: *; >0.01:**; >0.001: ***).

Year	Depth [cm]	Crop	No N-fertilizer input (N0)	Reduced N-fertilizer input (N1) [Mg SOC ha^{-1} layer^{-1}]	High N-fertilizer input (N2)
2002	0–30	CR$_{\mathrm{ploughed}}$ (CT)	40.1 ± 1.5	40.1 ± 1.5	40.1 ± 1.5
		CR$_{\mathrm{no\text{-}till}}$ (NT)	41.6 ± 1.5	41.6 ± 1.5	41.6 ± 1.5
		Maize$_{\mathrm{reduced\text{-}till}}$	39.3 ± 1.5	39.3 ± 1.5	39.3 ± 1.5
		Miscanthus	39.8 ± 1.5	39.8 ± 1.5	39.8 ± 1.5
		Switchgrass	38.5 ± 1.5	38.5 ± 1.5	38.5 ± 1.5
		Willow	39.9 ± 1.5	39.9 ± 1.5	39.9 ± 1.5
	30–60	CR$_{\mathrm{ploughed}}$ (CT)	20.3 ± 2.5	20.3 ± 2.5	20.3 ± 2.5
		CR$_{\mathrm{no\text{-}till}}$ (NT)	19.3 ± 2.5	19.3 ± 2.5	19.3 ± 2.5
		Maize$_{\mathrm{reduced\text{-}till}}$	18.1 ± 2.5	18.1 ± 2.5	18.1 ± 2.5
		Miscanthus	19.4 ± 2.5	19.4 ± 2.5	19.4 ± 2.5
		Switchgrass	19.3 ± 2.5	19.3 ± 2.5	19.3 ± 2.5
		Willow	22.6 ± 2.5	22.6 ± 2.5	22.6 ± 2.5
2013	0–30	CR$_{\mathrm{ploughed}}$ (CT)	38.9 ± 1.5	39.3 ± 1.5	41.1 ± 1.5
		CR$_{\mathrm{no\text{-}till}}$ (NT)	42.4 ± 1.5	42.0 ± 1.5	40.4 ± 1.5
		Maize$_{\mathrm{reduced\text{-}till}}$	38.7 ± 1.5	39.4 ± 1.5	43.9 ± 1.5
		Miscanthus	47.1 ± 1.5	47.5 ± 1.5	47.4 ± 1.5
		Switchgrass	46.7 ± 1.5	45.3 ± 1.5	56.6 ± 1.5
		Willow	48.8 ± 1.5	46.9 ± 1.5	52.1 ± 1.7
	30–60	CR$_{\mathrm{ploughed}}$ (CT)	19.1 ± 2.5	19.3 ± 2.5	18.7 ± 2.5
		CR$_{\mathrm{no\text{-}till}}$ (NT)	22.4 ± 2.5	29.0 ± 2.5	23.4 ± 2.5
		Maize$_{\mathrm{reduced\text{-}till}}$	17.7 ± 2.5	18.6 ± 2.5	21.9 ± 2.5
		Miscanthus	25.5 ± 2.5	26.4 ± 2.5	24.7 ± 2.5
		Switchgrass	27.4 ± 2.7	23.4 ± 2.5	26.8 ± 2.5
		Willow	25.6 ± 2.5	26.4 ± 2.5	25.0 ± 3.0
Difference	0–30	CR$_{\mathrm{ploughed}}$ (CT)	−1.2	−0.8	1.0
		CR$_{\mathrm{no\text{-}till}}$ (NT)	0.8	0.4	−1.2
		Maize$_{\mathrm{reduced\text{-}till}}$	−0.7	0.0	4.6*
		Miscanthus	7.3***	7.7***	7.6***
		Switchgrass	8.3***	6.9**	18.1***
		Willow	8.9***	7.0***	12.2***
	30–60	CR$_{\mathrm{ploughed}}$ (CT)	−1.1	−1.0	−1.6
		CR$_{\mathrm{no\text{-}till}}$ (NT)	3.1	9.7***	4.0*
		Maize$_{\mathrm{reduced\text{-}till}}$	−0.4	0.5	3.8*
		Miscanthus	6.1**	7.0***	5.4**
		Switchgrass	8.1***	4.2***	7.5***
		Willow	3.0	3.8*	2.4

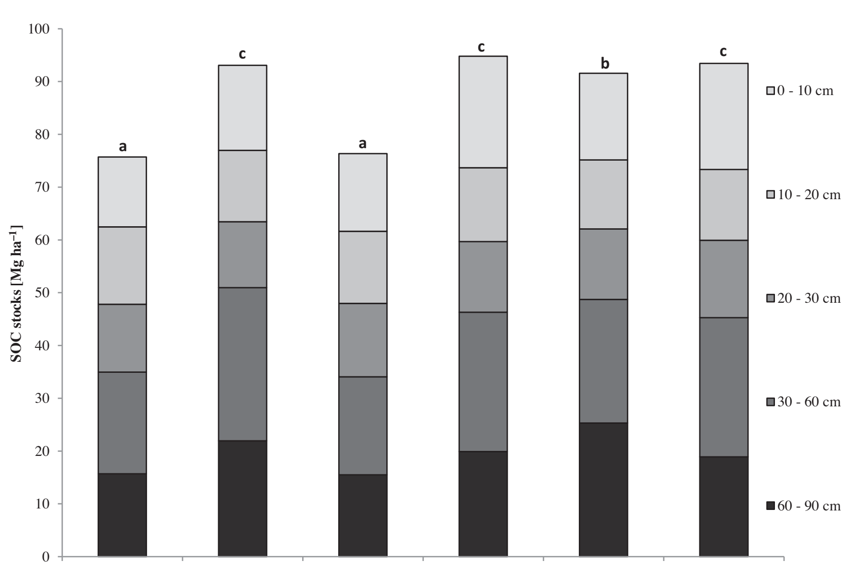

Fig. 1. Total SOC stocks [Mg ha^{-1}] in the N1-fertilization regime to a depth of 90 cm after 11 years of continuous cropping (bars with the same letters are not significantly different at $\alpha = .05$).

subsoil SOC sequestration are highly contradictory. Although depending on the soil conditions, no significant differences could be measured in deeper soil in some studies (e.g. Deen and Kataki, 2003; Syswerda et al., 2011), other studies verified the potential of carbon storage in deeper soil by different crop management systems (Olson et al., 2005).

Most studies assessing carbon sequestration in agricultural soils (e.g. when comparing NT and CT systems) limit sampling depth to the upper 30 cm or less (Baker et al., 2007; West and Post, 2002). Some studies document increased SOC stocks of NT systems compared with CT systems in the shallow topsoil, but in regard to deeper soil layers, results are scarce and often inconsistent; usually, either no significant differences can be detected (Spargo et al., 2008; Blanco-Canqui et al., 2011; Syswerda et al., 2011; Soane et al., 2012), or a stratification of SOC within the soil profile takes place, with the result of an accumulation of SOC in the topsoil and a reduction of SOC in the subsoil (Baker et al., 2007). In this study, when considering all layers to the depth of 90 cm, CT treatments had significantly higher SOC stocks in the N0- and N1-fertilization regimes, emphasizing the importance of measuring subsoil layers (Fig. 1). Regarding the bulk density, which were highest on the soil surface of NT treatment and lowest in the CT treatment, it could have been the case that the roots of the crops in the CT soil were more concentrated in the tilled top 20 cm compared with the NT treatment where the bulk density of the upper 30 cm indicate a compaction in the first 20 cm. However, below 20 cm the soil was less compacted in the NT treatment (Table 3). Roots of cereals and rape seed are known to reach depths of more than 100 cm (Smit and Groenwold, 2005) and could therefore be more oriented to deeper soil layers in the NT system compared with the CT treatment. Moreover, bulk density in the CT soil indicates a plough layer between 20 and 30 cm depth, which may act as a barrier in certain stages of root development. This could also be the case for maize, since the soil was tilled and the SOC stocks in deeper soil layers were in the same relatively low range as for the CT treatment. Maize can also root deeper than 1 m (Dardanelli et al., 1997); however, with the loose soil in the top 20 cm it is likely that root growth occurred predominantly in these layers. Nevertheless, rooting structures were not measured in this study.

Soane et al. (2012) reviewed NT studies in Europe and concluded that the increased vertical macro porosity in no-tilled soil leads to an increased air, water and root permeability through the soil profile. On the other hand, studies evaluating vertical root distribution between NT and plough systems came to inconsistent results, probably due to factors such as soil texture, climate and year effects, as discussed by Qin et al. (2006) and Dwyer et al. (1996). Our study detected higher SOC stocks in the NT treatments below 30 cm compared with the CT treatments. Therefore, our work does not support the hypothesis that NT lead to an enrichment of SOC in the topsoil, which is then offset by reduced SOC stocks in the subsoil (e.g. Baker et al., 2007). Hence, NT systems seem to be an appropriate tool to increase SOC stocks at the trial location, in particular as SOC in the subsoil is regarded as less susceptible to decomposition than SOC in the topsoil (Ajwa et al., 1998). Table 7 shows that for all treatments about 50% of the total SOC stocks are located in the subsoil below 30 cm. The current study underlines the importance of sampling to deeper soil layers to fully assess the potential of different cropping systems for C sequestration.

4.4. Annual SOC-sequestration

The results indicate that perennial crops increased total C stocks in the topsoil layer over the study's duration with and without N input (Table 8). N input has enhanced this trend in Miscanthus and maize, as well as in the NT treatment of the annual crop rotation. The role of plant breeding for high N efficiency became evident as only maize was able to increase C stocks in the N2-fertilizer regime. Up to now, breeding activities for perennial energy crops are limited; therefore, genotype selection for high N efficiency in Miscanthus, willow and switchgrass might even lead to higher yields and higher C-sequestration rates than shown in the current study.

The annual SOC sequestration rates of the topsoil layer are within or in the lower range of previously published sequestration rates. Regarding the no-till system, the annual SOC sequestration rates of 0.10–0.13 Mg C ha^{-1} yr^{-1} in this study correspond to sequestration rates found in other studies which ranged from 0.02–0.14 Mg C ha^{-1} yr^{-1} (Soane et al., 2012; VandenBygaart et al., 2010). However, more often higher rates of 0.30–0.57 Mg C ha^{-1} yr^{-1} were reported (Adam-Schumm et al., 2007; Freibauer et al., 2004; West and Post 2002). For the maize treatment with reduced soil tillage, the mean annual SOC sequestration rates of 0.15–0.45 Mg C ha^{-1} yr^{-1} are within the range of previously published results (0.27 and 0.40 Mg C ha^{-1} yr^{-1},Adam-Schumm et al., 2007; Freibauer et al., 2004). For Miscanthus, the sequestration rates were with 0.45–0.71 Mg C ha^{-1} yr^{-1} in the lower range of published data (0.60–1.00 Mg C ha^{-1} yr^{-1}, e.g. Don et al., 2012; Freibauer et al., 2004; Kahle et al., 2001; Poeplau and Don, 2014). The same is true for switchgrass with 0.20–0.57 Mg C ha^{-1} yr^{-1} in the topsoil and 1.45–1.77 Mg C ha^{-1} yr^{-1} in the whole soil profile, compared with the published data of 0.57–2.1 Mg C ha^{-1} yr^{-1} (Follet et al., 2012; Al-Kaisi et al., 2005; Lemus and Lal 2005 Al-Kaisi et al., 2005; Lemus and Lal 2005) and 2.00–4.00 Mg C ha^{-1} yr^{-1} respectively (Follet et al., 2012; Lee et al., 2007; Zan et al., 2001 Lee et al., 2007; Zan et al., 2001). For willow, however, the mean SOC sequestration rate was 0.67–0.72 Mg C ha^{-1} yr^{-1} in the topsoil and 1.43–1.68 Mg C ha^{-1} yr^{-1} in the whole soil profile, and therefor well in the range of comparable studies which found rates from 0.22–0.90 Mg C ha^{-1} yr^{-1} (Don et al., 2012; Hellebrand et al., 2010; Kahle et al., 2007; Lemus and Lal 2005) and −1.3–9.0 Mg C ha^{-1} yr^{-1} respectively (Walter et al., 2014; Zan et al., 2001).

Some authors came to the conclusion that on marginal or degenerated sites SOC sequestration rates outperform the rates which can be found on fertile sites (Soane et al., 2012; Lemus and Lal, 2005). This would explain the rather low accumulation rates in

Table 7
Distribution of C stocks with profile depth from 0 to 30 cm and 30–90 cm (mean values ± standard error).

	Average C-stocks 0–30 cm			Average C-stocks 30–90 cm			Percentage of average C-stocks in the subsoil%		
	Mg ha^{-1}								
Cropping system	N0	N1	N2	N0	N1	N2	N0	N1	N2
$CR_{plouhed}$ (CT)	37.5 ± 1.2	40 .7 ± 1.2	39.9 ± 1.2	34.7 ± 2.2	35 ± 2.2	34.9 ± 2.2	48 ± 1.4	46 ± 1.4	47 ± 1.4
$CR_{no-till}$ (NT)	38.7 ± 1.2	42.1 ± 1.2	41.0 ± 1.2	44.1 ± 2.2	50.9 ± 2.2	40.1 ± 2.2	53 ± 1.4	55 ± 1.4	49 ± 1.4
Maize $_{reducedtill}$	40.1 ± 1.2	42.3 ± 1.2	44.8 ± 1.2	31.9 ± 2.2	34.1 ± 2.2	39.9 ± 2.2	44 ± 1.4	45 ± 1.4	47 ± 1.4
Miscanthus	42.5 ± 1.2	48.5 ± 1.2	47.7 ± 1.2	43 ± 2.2	46.3 ± 2.2	42.2 ± 2.2	50 ± 1.4	49 ± 1.4	47 ± 1.4
Switchgrasss	42.4 ± 1.2	42.9 ± 1.2	46.2 ± 1.2	47 ± 2.2	48.7 ± 2.2	47.9 ± 2.2	53 ± 1.4	53 ± 1.4	51 ± 1.4
Willow	45.4 ± 1.2	48.1 ± 1.2	47.3 ± 1.2	45.3 ± 2.2	45.3 ± 2.2	43.1 ± 2.2	50 ± 1.4	49 ± 1.4	48 ± 1.4

Table 8
Mean annual SOC change [Mg C ha^{-1} yr^{-1}] compared with the reference, i.e. $CR_{ploughed}$ (± standard error).

Depth [cm]	Crop	No N-fertilizer input (N0)	Reduced N-fertilizer input (N1) [Mg C ha^{-1} yr^{-1}]	High N-fertilizer input (N2)
0–30	$CR_{ploughed}$ (CT)	0.00 ± 0.14	0.00 ± 0.14	0.00 ± 0.14
	$CR_{no\text{-}till}$ (NT)	0.11 ± 0.14	0.13 ± 0.14	0.10 ± 0.14
	$Maize_{reduced\text{-}till}$	0.24 ± 0.14	0.15 ± 0.14	0.45 ± 0.14
	Miscanthus	0.45 ± 0.14	0.71 ± 0.14	0.71 ± 0.14
	Switchgrass	0.45 ± 0.14	0.20 ± 0.14	0.57 ± 0.14
	Willow	0.72 ± 0.14	0.67 ± 0.14	0.67 ± 0.14
0–90	$CR_{ploughed}$ (CT)	0.00 ± 0.25	0.00 ± 0.25	0.00 ± 0.25
	$CR_{no\text{-}till}$ (NT)	0.96 ± 0.25	1.58 ± 0.25	0.57 ± 0.25
	$Maize_{reduced\text{-}till}$	−0.02 ± 0.25	0.06 ± 0.25	0.91 ± 0.25
	Miscanthus	1.20 ± 0.25	1.74 ± 0.25	1.38 ± 0.25
	Switchgrass	1.56 ± 0.25	1.45 ± 0.25	1.77 ± 0.25
	Willow	1.68 ± 0.25	1.62 ± 0.25	1.43 ± 0.25

the topsoil found in the current study. The soil type on the research sites was classified as Haplic Luvisol, which can be regarded as a fertile soil. Based on the available literature, the current SOC sequestration rates are supposed to decline as soon as the equilibrium stage for the cropping systems is reached. For the NT treatments, this equilibrium is assumed to be established after 15–25 years of continuous management, for the perennial crops after 40–100 years (Alvarez 2005; Potter et al., 1999; Sauerbeck 2001; West and Post 2002).

4.5. Stability and turnover rate of SOC

A comparison between the initial total carbon stocks and the stocks in 2013 showed that an equilibrium stage in SOC fluxes was reached in the control treatment with CT. If it is unknown whether the field site has reached an equilibrium stage or if SOC is still depleted, one can confuse a reduced SOC depletion rate in one variant with SOC sequestration (VandenBygaart and Angers, 2006). Since the field site was cropped in a similar way for several centuries, the arable soil in these plots is supposedly close to an equilibrium stage and the other cropping systems used for the study induce new equilibria. However, recent data indicate that increasing mean temperatures over the past decades triggered SOC losses in unchanged soil management systems (Senthilkumar et al., 2009). Furthermore, Lefèvre et al. (2014) found evidence that stabile C fractions in the soil react more sensitively to increased temperatures than labile fractions. Such a decline in SOC stocks could not be detected in the current study despite increasing mean temperatures in the last years (mean of the last 10 years 9.1 °C, mean of the last 40 years 8.3 °C).

Literature on the stability of SOC with respect to soil depth reveals inconsistent findings: while some data suggest that SOC in deeper soil layers is relatively protected from decomposition (Ajwa et al., 1998; Fontaine et al., 2007), others found a high rate of microbial decomposition in deeper soil layers, especially when organic fertilizers were applied (Erich et al., 2012; Liang and Balser, 2008). These findings suggest that chemical, physical and biological differences in the soils influence the decomposition rates of SOC in the subsoil (Sanaullah et al., 2011). Density or size fractionations of SOC as well as spectroscopic methods could contribute to a better understanding of the dynamics in subsoil SOC on the trial site.

5. Conclusions

The results of this study indicate that perennial energy crops can increase soil organic carbon (SOC) stocks (soil depth 0–90 cm) by 15–20 Mg ha^{-1} compared with annual energy crops in conventional arable systems (crop rotation, moldboard plough) over 11 years in southwest Germany on a Haplic Luvisol. The no-till (NT) treatment also led to significantly increased SOC stocks compared with conventional ploughing, especially in the soil layers below 30 cm. The data support hypothesis 1 only. Significantly higher SOC stocks of no-tilled annuals compared with conventional tilled annuals were also measurable in subsoil layers. Therefore, the data do not support hypothesis 2 or 3. Reduced N-fertilization adapted to each cropping system led to increased SOC accumulation in most cropping systems compared with the control without N input. Therefore hypothesis 4, N-fertilization leads to increased carbon sequestration, was verified. The SOC stock was not further increased by a higher N-fertilization, except for energy maize which could utilize high N inputs for biomass production and subsequently in higher SOC stocks. Further research is needed to determine the stability of the carbon pools in the differing cropping systems to evaluate long-term effects on soil fertility and the persistency of the systems as sinks for CO_2.

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