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Integrating Crop-Livestock System Practices in Forage and Grain-Based Rotations in Northern Germany: Potentials for Soil Carbon Sequestration

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Abstract: Integrating leys, cover crops, and animal manures constitute promising avenues to reach annual soil organic carbon changes (ΔSOC) >0.4% in forage and grain-based crop rotations, rates required to offset the increasing C emissions from fossil fuels ("4 per mille" initiative). How these practices and rotations perform in reaching this aim was object of analysis in this paper. Five cropping systems (CS), including three three-year forage and grain-based crop rotations containing annual grass-clover leys (FR and MR) or cover crops (GR), and two contrasting controls (continuous silage maize (CM), and permanent grassland (PG)) were compared for their impact on SOC stocks over eight years (2010–2018). The CS were unfertilized (N0) or fertilized using cattle slurry (N1) at a rate of 240 kg N ha^{-1} yr^{-1} applied in the non-leguminous crops. The ΔSOC of the top 30 cm soil layer and the annual carbon inputs (C_{in}) from slurry applications and plant residues were estimated, their relationship established, and the slurry-induced C retention coefficient was determined. The FR and MR SOC stocks remained stable at N1, while the GR and CM SOC decreased over time by tendency even at N1. Only the PG reached ΔSOC >0.4%. Differences in ΔSOC between CS and N rates were highly associated with the system-specific increase in belowground C_{in}, induced by slurry applications. Slurry-induced C retention coefficients differed strongly between CS: CM (3%) followed by GR (12%), and by FR and MR (20–15%), and lastly by PG (24%). Promoting belowground carbon inputs was identified as an efficient way to reach significant increases in ΔSOC. We conclude that a ley in only one out of three years is not sufficient to significantly increase SOC stocks in arable crop rotations of the study region.

Keywords: land-use intensification; leys; cover crops; animal manure; 4 per mille

Citation: De Los Rios, J.; Poyda, A.; Reinsch, T.; Kluß, C.; Taube, F.; Loges, R. Integrating Crop-Livestock System Practices in Forage and Grain-Based Rotations in Northern Germany: Potentials for Soil Carbon Sequestration. *Agronomy* **2022**, *12*, 338. https://doi.org/10.3390/agronomy12020338

Academic Editors: Zhongkui Luo and David W. Archer

Received: 22 December 2021
Accepted: 20 January 2022
Published: 28 January 2022

1. Introduction

Recent estimates suggest that obtaining annual soil organic carbon changes (ΔSOC) in agricultural soils >0.4% could offset the amount of C emitted annually from burning fossil fuels ("4 per mille initiative") [1,2]. This goal is confronted, however, with the increasing use of energy, protein, and fiber-rich feed crops [3]. Grasslands are turned into intensively-managed cropland, in either short crop rotation or continuous monocropping [4]. Likewise, crop-livestock farming units are being separated into either intensively-managed crop or livestock production units, leading to simplification and specialization of agricultural systems and regions [5,6]. With these changes, negative environmental impacts including higher greenhouse gas emissions, higher water pollution, and the depletion of soil organic matter associated with soil fertility and multiple ecosystems services have followed, as a result of nutrient imbalances and inadequate organic matter cycling [6–8].

To promote higher ΔSOC while reducing the negative impacts, cropping systems (CS) are required to foster ecological intensification practices like increasing crop diversification (in both crop rotations and seed mixtures), animal integration, and improving nutrient and organic matter cycling between plants and animals [4,6,9]. To do so, integrating crop-livestock system practices into crop rotations at farms (known as mixed farming systems or MFS) and regional scale (known as integrated crop-livestock systems or ICLS) are seen as avenues to implement these measures [10,11].

In crop-livestock systems, several practices linked to promoting SOC stocks are used to optimize nutrient cycling, increase agricultural output and the overall net primary productivity (NPP). The use of legumes, cover crops, and temporary grasslands in pure stands or in mixtures (also known as "leys") can increase soil aggregate stability, organic matter inputs, and reduce soil cultivation [12–15]. Other practices involved the incorporation of animal wastes, which provide nutrients and improve soil physical properties [16]. Of all these practices, the use of leys and cover crops in crop rotations have recently attracted great attention [12,17,18]. During the ley phase of a crop rotation, particularly in a legume-grass mixture, a significant amount of N can be added from atmospheric N_2 fixation, promoting the overall NPP of a rotation [18,19]. At the same time, SOC can be restored and increased by the high root biomass production of gramineous species, balancing the SOC degradation occurring during the arable phase [15]. Cover crops used as either green manure or catch crop in crop rotations can reduce nutrient leaching and supply these to the subsequent crops when ploughed. Furthermore, these provide additional carbon inputs (C_{in}), increasing SOC stocks compared to an annual or seasonal fallow period [14].

While these benefits are known, it remains yet unclear which integration or combination of practices are more effective to increase SOC stocks over time in intensive forage and grain-based crop rotations. From a system-specific view, increasing ΔSOC depends on the complex interaction of multiple factors affecting the C_{in} and the organic matter decay rates. Carbon inputs depend on the amount of residues derived from different sources, including shoots and roots of the crops grown in the system, as well as on the external inputs added, e.g., from animal manures [20,21]. Moreover, animal manures could induce indirect C_{in} via the increase in NPP and crop residues [22,23]. These different sources have shown, in addition, to have different resistances to soil decomposers. Higher humification or retention coefficients have been observed in roots compared to shoots in multiple studies [24,25], while the same has been observed for different manure types, favoring solid compared to liquid manures [16,26]. Conversely, tillage affects the organic matter decay rates of a system, by promoting the regular disruption of soil aggregates and increasing contact of organic matter to microbial degradation, which can result in small or negative ΔSOC despite increasing C_{in} as previously observed [23,27,28]. In this context, there is a need to elucidate the effects of combining the use of leys or cover crops with manure fertilization in forage and grain-based crop rotations, as well as their C_{in} on SOC stocks, as the integration of these different practices may produce ΔSOC that might be highly system-specific [15,23].

In this study, three three-year crop rotations containing annual leys or cover crops, and two contrasting control systems, namely continuous silage maize and permanent grassland, all fertilized with cattle slurry are assessed for their impact on the SOC stocks over time, over eight years. The annual soil C_{in} derived from crop residues will be calculated using annual crop yields and yield-based allocation coefficients obtained on-site, as well as from slurry applications over the study period in the different CS. The objectives were (i) to compare the ΔSOC of different forage and grain-based rotations containing leys or cover crops in combination with animal manures and observe whether annual ΔSOC > 0.4% is reached, and (ii) to analyse how annual C_{in} from crop residues and slurry influence the ΔSOC of the different CS. It was hypothesized that increasing C_{in} will result in higher ΔSOC in all CS, independently of the management system.

2. Materials and Methods

2.1. Study Site and Design

The study was conducted from 2010 to 2018 at the research station "Lindhof" of the Christian-Albrecht-University of Kiel, located in Kiel, northern Germany (N 54°27′55 E 9°57′55; 15 m a.s.l.). The area is characterized by a maritime climate zone, with a mean annual temperature of 8.9 °C and a mean total annual precipitation of 778 mm (1981–2010) (provided in Supplementary, Figure S1). The soil textural class is a sandy loam, formed by 61% sand, 26% silt, and 13% clay, containing 1.2% organic C (at a bulk density of 1.5 g cm^{-3}). The soil is classified as a Cambisol (Eutric Luvisol) group (FAO classification). Before the study period, the field was dominated by an organically managed four-year crop rotation, consisting of winter wheat—grass-clover—oats—potatoes, for over 15 years (1995–2010). The site was fertilized with solid manure three times over the whole period (2002, 2006, and 2010) in the potatoes only, with 24 Mg ha^{-1}, equal to 80 kg N ha^{-1} per application.

In 2010, a two-factorial randomized (split-plot) design was established. As main plot factor, five CS were introduced, including: (1) a continuous silage maize (CM); (2) a 3-year grain rotation (GR) with cover crops (spring oats—winter wheat/grass-clover—pulses/white mustard). The spring-oats, winter wheat and pulses (either blue lupin, field pea, or faba bean) were harvested as grain. The cover crops were a grass-clover undersown in the winter wheat managed with one cut in the sowing year, and a white mustard sown after the pulses, both ploughed as green manures in spring before the following main crop; (3) a 3-year forage rotation (FR) with a 1-year ley (silage maize—winter wheat/grass-clover—grass-clover). The winter wheat was harvested as whole-crop silage. The grass-clover was undersown in the winter wheat and managed with a cutting frequency of one cut in the sowing year and four cuts in the main year, then ploughed in spring; (4) a 3-year mixed rotation (MR) with a 1-year ley (spring oats—winter wheat/grass-clover—grass-clover). The spring oats and winter wheat were managed as in GR, and the grass-clover managed as in FR; and (5) a permanent grassland (PG) sown in 2010, managed with a cutting frequency of four cuts each year. A diagram of the rotational cycles and crops belonging to each CS is provided in Supplementary (Figure S2). The seed mixtures, cultivars, and seeding rates and durations used are presented in Table 1.

Table 1. Seed mixtures, cultivars, seeding rates, and crop type-dependent duration in months.

Crop		Cultivar	Seeding Rate	Crop Type	Length (Month)
Silage Maize	(*Zea mays*)	Ronaldinio	12 seeds m^{-2}	Forage	May–October (6 m)
Spring Oats	(*Avena sativa*)	Ivory Max	350 seeds ha^{-1}	Grain	March–August (6 m)
Winter Wheat	(*Triticum aestivum)*	Mulan Akteur Elixer	400 seeds ha^{-1}	Forage Grain	October–July (10 m) October–August (11 m)
Pulses	Peas (*Pisum sativum*) Lupin (*Lupinus angustifolius*) Faba bean (*Vicia faba*)	Santana Boruta Fanfare	80 seeds m^2 130 seeds m^2 50 seeds m^2	Grain	March–August (6 m)
Grass-clover Ley	Perennial ryegrass (*Lolium perenne*) Red clover (*Trifolium pratense*) White clover (*Trifolium repens*)	n.a. n.a. n.a.	20 kg ha^{-1} 8 kg ha^{-1} 2 kg ha^{-1}	Cover crop Forage	May–March (11 m) May–March (22 m)
Grassland	Perennial ryegrass (*Lolium perenne*) Timothy grass (*Phleum pratense*) Meadow grass (*Poa pratensis*) White clover (*Trifolium repens*)	n.a. n.a. n.a. n.a.	20 kg ha^{-1} 4 kg ha^{-1} 4 kg ha^{-1} 2 kg ha^{-1}	Forage	Permanent
White Mustard	(*Synapsis alba*)	n.a.	25 kg ha^{-1}	Cover crop	September–March (7 m)

n.a.: not available.

As subplot factors, two nitrogen (N) fertilization rates were used: unfertilized (N0), and fertilized using cattle slurry (N1) at a rate of 240 kg N ha^{-1}, applied annually using trailing hoses only to the non-leguminous crops. Leguminous crops (i.e., clover grass and pulses) were not fertilized, as these were considered as N sources via biological nitrogen fixation to the rotations. The mean dry matter (DM) content in the applied cattle slurry was 5.7%, and the C and N in DM were 40 and 3.66%, respectively (C:N 10.9). The application plan and the amount of N applied from cattle slurry for the N1 treatments are shown in Table 2. Other nutrients were applied equally to all plots (45 kg P ha^{-1}, 100 kg K ha^{-1}, 24 kg Mg ha^{-1}, and 68 kg S ha^{-1}, all as rock phosphate). Lime was applied bi-annually at the rate of 1 Mg ha^{-1} using $CaCO_3$ (23% Ca and 1.4% Mg).

Table 2. N application plan and the average amount of N applied from cattle slurry for the N1 treatment, to each main crop and cropping system, respectively. CM: continuous silage maize, GR: grain rotation, FR: forage rotation, MR: mixed rotation, PG: permanent grassland.

Cropping System	Crop	Fertilized (Yes/No)	Amount of N Supplied per Application (kg ha^{-1})	Mean N Input in System (kg ha^{-1} yr^{-1}) *
CM	Silage Maize	yes	80 + 80 + 80	240
	Spring Oats	yes	80 + 80 + 80	
GR	Winter Wheat	yes	80 + 80 + 80	160
	Pulses	no		
	Silage Maize	yes	80 + 80 + 80	
FR	Winter Wheat	yes	80 + 80 + 80	160
	Grass-clover Ley	no		
	Spring Oats	yes	80 + 80 + 80	
MR	Winter Wheat	yes	80 + 80 + 80	160
	Grass-clover Ley	no		
PG	Grassland	yes	80 + 60 + 60 + 40	240

*: Average of three years.

Each CS and N rate combination was initialized with 3 replicates by crop, i.e., 9 replicates for the crop rotation (3 crops with 3 replicates per crop) and three replicates for the CM and the PG (1 crop with 3 replicates) by N rate, summing 66 plots. The plot sizes were 12 × 3 m.

2.2. Measurements

2.2.1. Soil Sampling and SOC Analysis

Soil sampling started in autumn 2010 when the study began and was repeated annually over the study period until spring 2019. A composite of two samples per plot was taken from the 0–30 cm layer, using a soil auger (inner ⌀: 2 cm). Samples were oven-dried to constant weight at 30 °C, sieved to pass a 2 mm screen, and analyzed for C concentration by the dry combustion method using a CN analyser (Vario Max CN, Elementar Analysensysteme GmbH, Hanau, Germany) [29]. As no inorganic C was detected in the soil samples after treatment with 4 mol L^{-1} of Hydrochloric Acid (HCl) [30], the measured total C was considered as the SOC concentration. To calculate the annual SOC stocks on a hectare basis, a fixed depth of 30 cm and a soil bulk density of 1.5 g cm^{-3} was used. This density was used based on measurements conducted at the site at the beginning of the study. The SOC stocks were calculated following the equation:

$$\text{SOC stock [Mg C ha}^{-1}\text{] = OC concentration [\%]} \times \text{bulk density [1.5 g cm}^{-3}\text{ soil]} \times \text{soil depth [30 cm]} \quad (1)$$

2.2.2. Plant Sampling and Soil Carbon Input Estimations

The annual crop yields (Y_P) in the forage crops were obtained using a forage plot harvester (Haldrup, Løgstør, Denmark), and in the grain crops using a combine plot harvester (Wintersteiger, Ried, Austria) for the period 2011–2018. The silage maize was harvested with a target DM content of 30%, whereas the winter wheat for whole-crop silage was harvested at the soft dough stage. The leys were harvested once in the sowing year, whereas both permanent grassland and leys were cut in parallel 4 times in the main year (in May, June, August, and October). The grain crops were all harvested at the maturity stage. Subsamples of Y_P were oven-dried at 58 °C until constant weight to determine humidity content and calculate the DM yields. After harvest, the straw was removed in both cereal grain crops and was left on the field in the pulses. The mean annual Y_P of the different crops for the period 2011–2018 is provided in Supplementary (Table S1).

The yield-based allocation coefficients were obtained from measurements conducted in separate studies on the same trial. The total above (AGB) and belowground biomass (BGB) in the forage crops were sampled in the years 2012 and 2013 by [18], during and after the growing seasons (March–October and October–March, respectively). During the growing season, AGB samples were cut at the surface level and separated into harvestable biomass and stubbles, at the time of harvest, in 4 subsampling plots of 0.25, 0.5, and 1 m^2 in both grasslands (permanent and leys), winter wheat and silage maize, respectively. In the same plots, BGB was sampled using the ingrowth core method until 30 cm depth [31], using a sampling interval of four weeks. After the growing season, the same methods were applied to the grasslands, except that a single sampling of AGB and BGB was conducted at the end of March. Samples were dried at 58 °C. Harvest (HI) and stubble indices (SI) were calculated by the ratio of forage Y_P and stubbles to total AGB on a DM basis, respectively. The R:S was determined by the ratio between the annual sums of the ash-corrected BGB and AGB. The C content of the different plant fractions was analyzed using a CN analyser (Vario Max CN, Elementar Analysensysteme GmbH, Hanau, Germany).

The total AGB and BGB in the grain crops were sampled at the maturity stage, before harvest in 2019. In 6 subsampling plots of 0.25 m^2, AGB samples were cut at the surface level, dried at 58 °C, and separated into ear and straw. In the same plots, BGB was sampled within and between the crop rows using the soil core method until 30 cm depth [32]. The HI was determined using the grain Y_P to total AGB. The SI, R:S, and C content were determined as explained above. In the white mustard, AGB and BGB were sampled in March 2019 before ploughing, using the methods applied in the grain crops.

The annual soil C_{in} were calculated system-wise from cattle slurry applications (slurry C_{in}) and plant residues (plant C_{in}). The slurry C_{in} were calculated using the crop-specific N fertilization rates and the slurry CN ratio. The plant C_{in} from above (AG C_{in}) and belowground (BG C_{in}) were calculated annually, using the measured Y_P and yield-based allocation coefficients for the C_{in} produced during the growing season, and adding fixed inputs to account for the residues produced AG and BG (AW–AG_{in} and AW–BG_{in}, respectively) by the permanent grassland, leys and cover crops after the growing seasons, estimated above. For the residues occurring during the growing season, the stubbles were calculated using the SI for the forage and grain crops with straw removed (only the cereals), whereas the straw was calculated using the HI for the crops with the straw left on the field (only the pulses). In the grasslands, harvest residues and litter deposition were calculated using a SI of 15% as proposed by [20]. Ash-corrected dry matter (ACDM) values were used across calculations as these are preferred to using total dry matter values [20]. The BG residues occurring in the same season were calculated using the ash-corrected R:S ratios. To account for extra inputs derived from rhizodeposition, an additional 50% of the total BGB produced during and after the growing seasons was added to the BG C_{in} calculations, as suggested by [33]. For the C_{in} calculations, a 48% C content of the ash-corrected samples was adopted across plant fraction and species. The yield-based allocation coefficients and fixed inputs, as well as the equations used to calculate the plant and total C_{in} are provided in Supplementary (Tables S2 and S3).

2.3. Statistical Analysis

The interaction of the study factors CS and N rate with Time were tested for their influence on SOC stocks using an analysis of covariance (ANCOVA), within a linear mixed-effects model [34], in the statistical software R (version 3.6.3) [35]. When these factors interacted significantly, a t-test was performed to determine whether the differences between the mean annual ΔSOC were significant. In addition, the annual ΔSOC [Mg C ha^{-1} yr^{-1}] in the different CS and N rate combinations was evaluated whether these statistically differed from zero [36]. To test the relationship between the annual soil C_{in} and ΔSOC, a linear regression was performed using the mean BG C_{in}, plant $C_{in,}$ and total C_{in} (Plant C_{in} + Slurry C_{in}) of each CS and N rate combination. All statistical results were considered significant at a p-value < 0.05.

3. Results

3.1. Effect by Cropping Systems and N Rates on Annual SOC Change Rates

The SOC stocks were significantly affected only by the N rates over time across CS (N rate × Time, $p < 0.01$) (Table 3). Pooled over the CS, the differences in annual ΔSOC between the N rates were statistically significant ($p < 0.01$), with higher ΔSOC at N1 than at N0 (−0.1 and −0.4 Mg C ha^{-1} yr^{-1}, respectively) (Figure 1).

Table 3. Analysis of covariance (ANCOVA) of the effect of cropping system (CS), and N application rate (N rate) on soil organic carbon (SOC) stocks over time.

Actors	Degrees of Freedom †	F-Value	*p*-Value
Intercept	1, 300	11,045.4	<0.0001
CS	4, 247	8.0	<0.0001
N rate	1, 300	31.9	<0.0001
Time	1, 300	1.0	0.31
CS × N rate	4, 300	5.5	<0.0001
CS × Time	4, 300	1.3	0.27
N rate × Time	1, 300	10.1	<0.01
CS × N rate × Time	4, 300	0.6	0.63

†: Degrees of freedom (numerator, denominator); Model R^2: marginal = 0.15; conditional = 0.80.

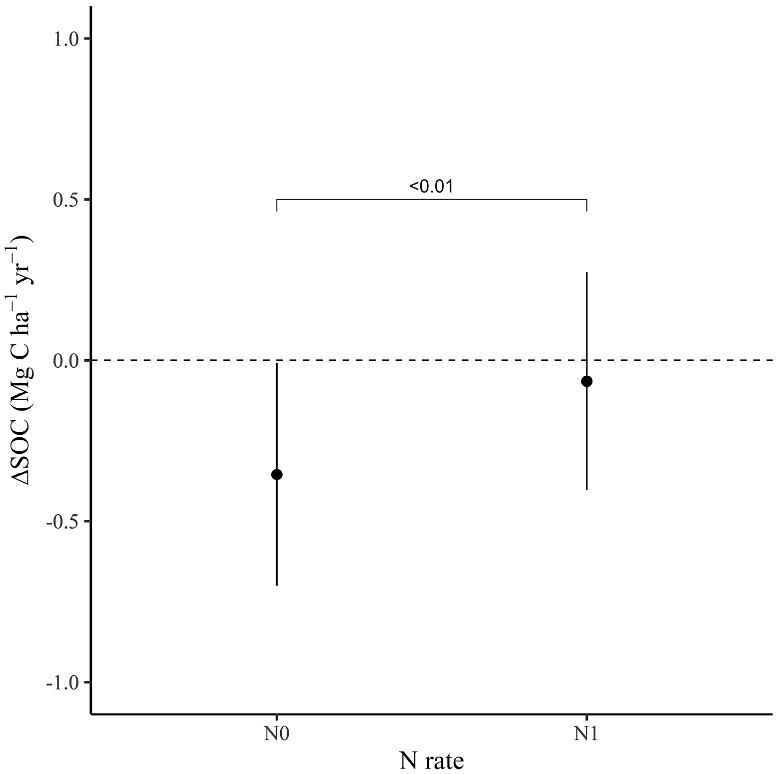

Figure 1. Mean annual SOC changes (ΔSOC) (±95% confidence intervals) for the different N rates. N0: unfertilized, N1: fertilized using cattle slurry at a rate of 240 kg N ha^{-1} in non-leguminous crops.

Differences in ΔSOC between the systems at the different N rates were observed (Figure 2a,b). The ΔSOC were negative at both N rates in CM (−0.7 and −0.6 Mg C ha^{-1} yr^{-1}) and in GR (−0.5 and −0.2 Mg C ha^{-1} yr^{-1}). In FR and MR, the ΔSOC were negative at N0 and positive at N1 (−0.3 and 0.1 Mg C ha^{-1} yr^{-1}), respectively. In PG, the ΔSOC were positive at both N rates (0.2 and 1.1 Mg C ha^{-1} yr^{-1}). Differences in ΔSOC between the N rates followed the order CM < GR < FR = MR < PG. While different tendencies were observed, only the ΔSOC of the PG at N1 statistically differed from zero ($p < 0.05$). In relative terms, only the PG reached ΔSOC slightly above 0.4% at N0 (0.5%) and considerably higher at N1 (2.3%), whereas for the crop rotations both FR and MR reached half of the target value at N1 (0.2%) during the measured period (Figure 2c).

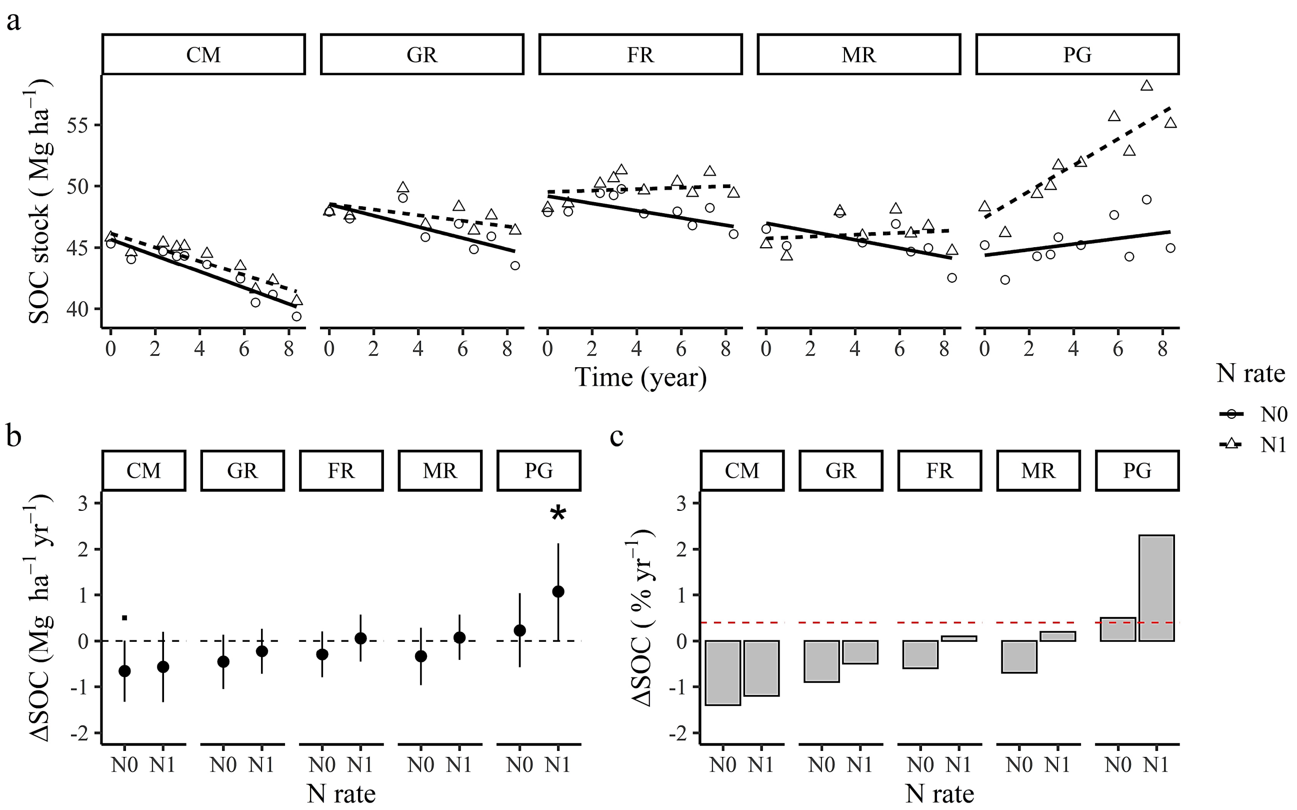

Figure 2. (**a**) Mean SOC stocks, (**b**) mean annual SOC change rates (ΔSOC) (±95% confidence intervals) (Mg ha^{-1} yr^{-1}), and (**c**) mean relative change (%) of the different cropping systems and N fertilization rates related to the initial SOC stock of 2010 (zero value) and to the target value of 0.4% (dotted red line), measured from autumn 2010 to spring 2019. Cropping Systems (CS) are ordered from smallest to largest annual SOC changes (ΔSOC). Significance at $p \leq 0.1$, $p \leq 0.05$ are indicated by ·, *, respectively. CM: continuous silage maize, GR: grain rotation, FR: forage rotation, MR: mixed rotation, PG: permanent grassland, N0: unfertilized, N1: fertilized using cattle slurry at a rate of 240 kg N ha^{-1} in non-leguminous crops.

3.2. Estimated Annual Soil C Inputs and Relationship to Soil C Inputs

The annual total C_{in} varied between the CS and N rates from 1.5 to 6.9 Mg C ha^{-1} yr^{-1} and followed the order CM < FR < PG < MR < GR at N0 and CM < FR < MR < GR < PG at N1 (Figure 3a). The smallest increment in plant C_{in} by N1 occurred in the GR (0.1 Mg C ha^{-1} yr^{-1}), followed by the CM (0.35 Mg C ha^{-1} yr^{-1}), then by the crop rotations containing leys (0.7 and 0.5 Mg C ha^{-1} yr^{-1} in FR and MR, respectively), and the greatest by the PG (1 Mg C ha^{-1} yr^{-1}), with 90% of the increment derived belowground. Across CS, N1 increased plant C_{in} from 2.9 to 3.5 Mg C ha^{-1} yr^{-1}, adding 20% more inputs via an increment in BG C_{in}. In contrast, the AG C_{in} were small, similar to N0. In relative terms, most of the plant C_{in} derived from BG (70 to 87%), and to a less extent from AG (13 to 30%).

Across CS, the total C_{in} derived mostly from plant C_{in} at both N rates, except for the CM where slurry C_{in} accounted for 55% of the total C_{in} at N1 (Figure 3b).

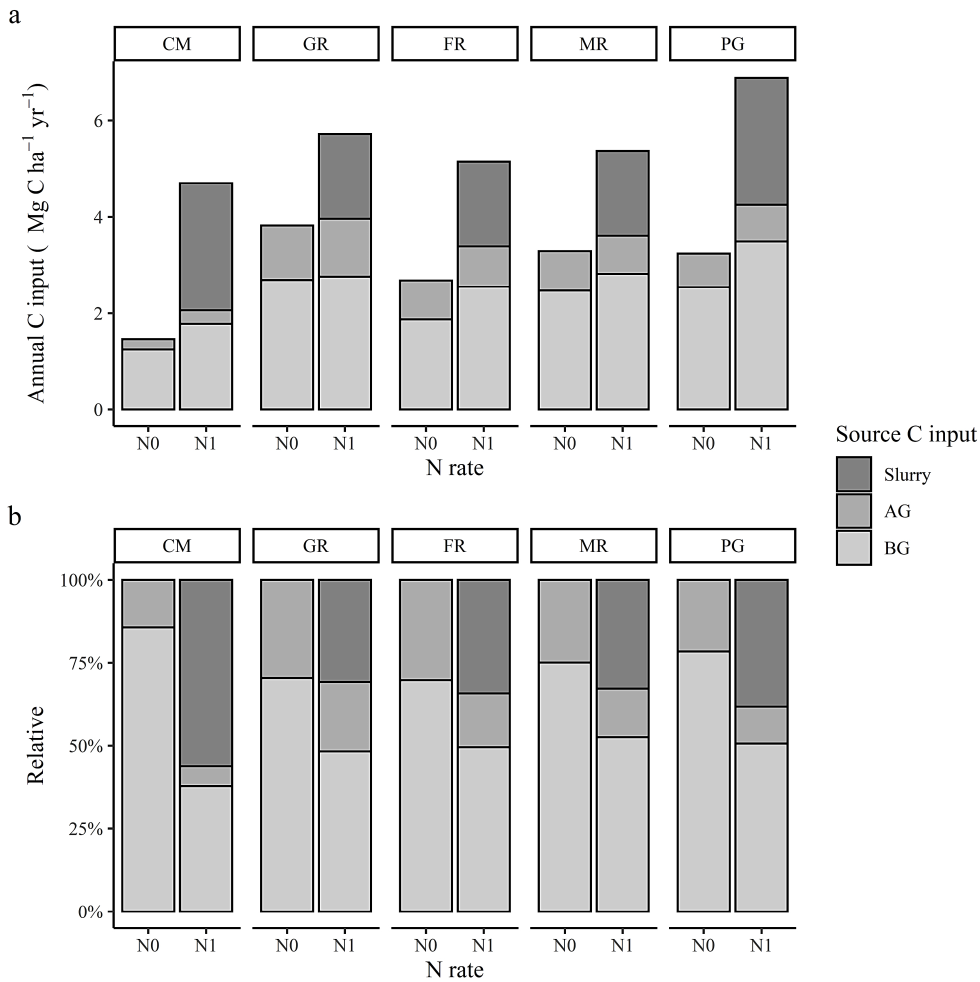

Figure 3. (**a**) Annual soil C inputs (Mg C ha^{-1} yr^{-1}) and (**b**) relative inputs derived from cattle slurry (Slurry C_{in}), from aboveground (AG C_{in}), and belowground residues (BG C_{in}) for the different cropping systems and N fertilization rates, estimated for the period 2011–2018. CM: continuous silage maize, GR: grain rotation, FR: forage rotation, MR: mixed rotation, PG: permanent grassland, N0: unfertilized, N1: fertilized using cattle slurry at a rate of 240 kg N ha^{-1} in non-leguminous crops.

The association between ΔSOC and annual C_{in} across the different treatments showed significant linear relationships for BG C_{in}, plant C_{in}, and total C_{in} (Plant C_{in} + Slurry C_{in}) (Figure 4), except for AG C_{in} alone (not shown). Consequently, changes in ΔSOC were more sensitive to altered BG C_{in} (slope = 0.68, $p < 0.01$, $R^2 = 0.64$) compared to altered plant C_{in} (slope = 0.41, $p < 0.05$, $R^2 = 0.46$) or total C_{in} (slope = 0.22, $p < 0.05$, $R^2 = 0.47$).

To further compare the relative increase in ΔSOC by the additional unit of total C_{in} induced by N1 between the CS, slurry-induced C retention coefficients were estimated system-wise following [23] approach. The lowest coefficient was obtained in the CM with 3%, followed by the crop rotation containing cover crops (GR) with 12%, then by the crop rotations containing leys (FR, MR) with 15–20%, and by the PG with 23%.

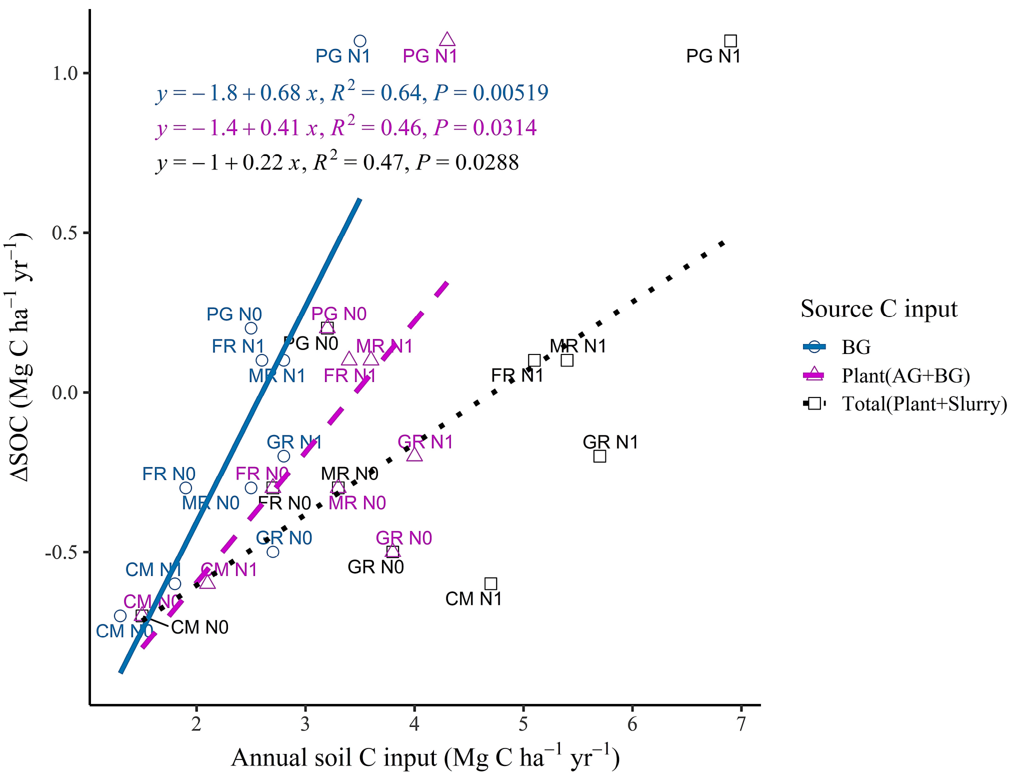

Figure 4. Relationship between annual soil C inputs from belowground (BG C_{in}), plant (AG C_{in} + BG C_{in}), and total (Plant C_{in} + Slurry C_{in}) with annual SOC changes (ΔSOC), for the different cropping systems (CS) and N rates in the topsoil layer (0–30 cm) in the period 2010–2018. CM: continuous silage maize, GR: grain rotation, FR: forage rotation, MR: mixed rotation, PG: permanent grassland, N0: unfertilized, N1: fertilized using cattle slurry at a rate of 240 kg N ha^{-1} in non-leguminous crops.

4. Discussion

4.1. Effect of Annual Leys and Cover Crops on SOC Stocks of Forage and Grain-Based Rotations

One of the objectives of this study was to compare the ΔSOC of different forage and grain-based rotations containing leys or cover crops in combination with animal manures and observe whether ΔSOC > 0.4% is reached in the different rotations. The rotations using an annual ley (FR and MR) showed no significant changes in SOC stocks over time but by the application of cattle slurry ΔSOC was increased from negative to stable (Figure 2). The ΔSOC in these rotations might have been already close to equilibrium, probably due to the similarities in management existing before and during the study period, containing an annual ley undersown in the winter wheat. Same results were also observed between the FR and MR, with similar ΔSOC at both N rates despite their differences in annual crops. These results are in agreement with those observed in a meta-analysis, which observed no effects in SOC stocks caused by exchanging one species of annual crop for another of different NPP when all other features of a rotation remain equal [12]. Regarding the relative ΔSOC, these rotations did not reach the target value of 0.4% obtained at N1, similarly to those reported in other studies varying between 0.03 and 0.08 Mg C ha^{-1} yr^{-1}, equivalent to 0.15% yr^{-1} in relation to the initial SOC stock [1,37]. These ΔSOC were, however, smaller than those reported for ley-arable systems with ley phases of 50 and 80%, estimated to increase SOC stocks in 0.2–0.3 Mg ha^{-1} yr^{-1}, equivalent to 0.4–0.7% yr^{-1} for 30 years [15,37,38], clearly above the 0.4%. These results indicate that using annual leys in forage and grain-based crop rotations might not be sufficient to increase SOC stocks according to the 4 per mille initiative at sandy loamy soils in northwest Europe.

In comparison to FR and MR, the grain rotation using cover crops (GR) (which replaced the use of annual leys in MR) was insufficient to maintain SOC stocks after the conversion and showed a tendency to decrease SOC stocks over time, despite applying slurry. Until now, limited studies are available that allow us to compare the effects of using either annual leys or cover crops in crop rotations in parallel at the same site. In several studies, however non-conclusive results regarding the potential of cover crops to increase SOC stocks can be observed. In a review, wide variations in ΔSOC from -0.55 Mg C ha^{-1} yr^{-1} or -1.8% yr^{-1} to 0.75 Mg C ha^{-1} yr^{-1} or 28.5% were reported for the use of cover crops in arable sandy soils with less than 1% of initial SOC, and in temperate regions like the present study [37]. In a global meta-analysis covering 37 different sites across climate regions and cover crop species, mean ΔSOC of 0.32 Mg C ha^{-1} yr^{-1} were estimated using cover crops compared to a winter fallowing or grain rotation without cover crops [14,39]. Although not measured, a higher soil cultivation frequency as a result of establishing pulses and cover crops in GR (tilled in two of the three-year rotational cycle), compared to establishing annual leys in FR and MR (tilled in one of the three-year rotational cycle), besides a low C retention of these crops might have promoted higher organic matter decomposition, as implied by previous studies [12,15,40].

As expected, the conversion to CM led by tendency to the greatest SOC losses of the CS evaluated. Very few studies are available for the conversion of cultivated land from a crop rotation containing annual ley to a continuous monocropping. Our results showed ΔSOC similar to those reported for the conversion from grassland to continuous monocropping, as a result of the drastic reduction in C_{in} and disruption of soil aggregate stability induced by soil cultivation [7,41]. In contrast to CM, the conversion to PG led to the greatest increase in SOC stocks by tendency without slurry, and with significant increases when fertilized with slurry. The ΔSOC obtained with slurry were similar to those found in a meta-analysis, where the mean ΔSOC after conversion from cropland to grassland was 1.01 Mg C ha^{-1} yr^{-1} or 3% yr^{-1} [42], showing that by the conversion of cropland to permanent grassland, ΔSOC > 0.4% yr^{-1} can be obtained at least during the first years after conversion. These results corroborate the need to make stronger modifications in cropping practices than using annual leys and cover crops in forage and grain-based crop rotations to produce significant increases in SOC stocks over time [15]. Otherwise, small non-positive SOC changes over time can be obtained, as currently observed. This might require the use of longer, perennial ley phases that reduces organic matter degradation, while adding a significant amount of C_{in}, as previously observed [15,37,38,43].

4.2. Influence of C Inputs from Slurry Application and Belowground C Input on Annual SOC Changes

Another objective of the study was to analyse how annual C_{in} from slurry and crop residues can influence the ΔSOC, and it was hypothesized that increasing C_{in} can result in higher ΔSOC in all CS. Overall, the application of cattle slurry increased SOC stocks considerably compared to the unfertilized systems across CS (Table 3). Differences in ΔSOC between N rates were, nevertheless, observed for the different CS (Figure 2), which were associated with the differences in C_{in} derived directly from slurry applications and indirectly from plant C_{in}. The presence of grasses for an extended period and their response to N supply from cattle slurry by producing higher BG C_{in}, improved SOC stocks in the CS, including grasses (PG) and grass-clover leys (FR and MR) more than those without leys (GR and CM) (Figure 3). The significant linear relationship between soil C_{in} and differences in SOC between CS confirmed the postulates of other studies, which associates higher ΔSOC with higher C_{in} [16,23,44,45]. In the present study, however, a stronger relationship with BG C_{in} than with plant C_{in} and total C_{in} (plant + slurry C_{in}) was identified (Figure 4), indicating the greater role played by BG C_{in} in achieving higher ΔSOC compared to the addition of slurry C_{in} and AG C_{in}, and confirming that roots are the most important contributor to SOC compared to other sources of C_{in} [22,24,25]. The dense rooting system of grasses adds greater amounts of recalcitrant C, contributing to the formation of stable aggregates and to the stabilization of SOC over time, compared to the less-dense rooting system observed in annual crops [13,39,46]. In agreement with these effects, the slurry-induced C retention coefficient was the highest in the CS with the highest increments in BG C_{in} (PG, FR, and MR) compared to those with the smaller increments in BG C_{in} (GR and CM) calculated in the current study. These results confirm those found by [23], who reported a higher increase in SOC concentration and higher C retention coefficients in ley-arable rotations compared to cereal monoculture after cattle slurry application, assuming higher BG C_{in} contributions in the ley-arable system compared to the arable system, and without separating the amount of C_{in} from the different sources as done in the current study.

In support of these observations, annual slurry applications in the CM did not reduce the SOC losses over time. In similar studies using annual slurry applications in continuous monocropping, SOC concentration was unaffected by regular slurry applications in continuous maize or continuous winter wheat [47,48]. In a five-year study conducted using continuous maize with several increments of slurry application of comparable DM content to the present study, a decrease in SOC content was reported except at an extremely high rate of 879 kg N ha^{-1} yr^{-1} [49]. This result strongly supports the low C retention coefficient of slurries as a direct source of C_{in} [16,26], and indicates that it is probable that the added slurry C_{in} and the low BG C_{in} could not compensate the negative C balance in the CM, as previously postulated [15,50,51]. As observed, increasing BG C_{in} is a more efficient source for increasing SOC stocks over time across CS than other sources of C_{in}. These results elucidate that the system-specific responses in ΔSOC produced by combining different practices are more dependent on the amount and increments in BG C_{in} than other C_{in} sources for a particular system.

5. Conclusions

In this study, the effects of integrating crop-livestock system practices like annual leys or cover crops in combination with animal manure on the SOC stocks of arable crop rotations were compared under northwest European conditions. Even with these management practices often assigned to increase SOC stocks, annual SOC changes were negative by tendency or stable and were not sufficient to reach the target ΔSOC of 0.4% yr^{-1} or higher, required to mitigate the increasing C emissions from fossil fuels in the short-term (4 per mille initiative). According to our results for sandy loamy soils, this rate can only be achieved by the establishment of permanent grassland. Further, belowground C inputs were identified as the main contributor to the differences observed in ΔSOC between CS. By promoting belowground inputs combined with reduced soil cultivation, moderate increases in SOC stocks over time could be reached. In arable crop rotations, this could be achieved most efficiently by introducing or increasing a ley phase for at least two consecutive production years.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/agronomy12020338/s1, Figure S1: Average monthly weather recorded during the study period (2010–2018) and average monthly long-term weather measured in the 30 years prior the study period (1981–2010). Figure S2: Diagram of the rotational cycles and crops belonging to each cropping system (CS) involved in the present study. Table S1: Mean annual crop yields (Y_P) (±standard deviation, (S.D)) of three replicates, measured for the different crops within each cropping system and N rate for the period 2011–2018. Table S2: Yield-based allocation coefficients and fixed inputs used to calculate the annual soil C inputs from plants (Plant C_{in}) during and after the growing season, respectively. Table S3: Steps and equations followed to calculate annual soil C inputs (C_{in}) in each crop and cropping system, using annual yields (Y_P, Table S1) and yield-based allocation coefficients (Table S2).

Author Contributions: Conceptualization, J.D.L.R., A.P., T.R., F.T. and R.L.; Methodology, J.D.L.R., A.P. and T.R.; Software, J.D.L.R.; Formal analysis, J.D.L.R.; Investigation, J.D.L.R.; Data curation, C.K., J.D.L.R.; Writing—original draft preparation, J.D.L.R.; Writing—review and editing, A.P., T.R. and F.T.; Visualization, J.D.L.R.; Supervision, A.P., T.R. and F.T.; Funding acquisition, T.R. and F.T. All authors have read and agreed to the published version of the manuscript.

Funding: J.D.L.R. is supported by the Evangelisches Studienwerk Villigst foundation, under the research program: "Third Ways of Feeding the World" who provided funding in the form of a doctoral scholarship. The European Commission through the 7th Framework Programme (Project ID: 289328, Funded under: FP7-KBBE, CANTOGETHER (Crops and ANimals TOGETHER) project) provided financial support in the first 3 years of the study. Land Schleswig-Holstein provided financial support within the funding program Open Access Publikationsfonds. Their support is gratefully acknowledged.

Acknowledgments: The authors would like to thank M. Hasler for his valuable statistical consultancy, and R. Kopp and T. Ehmsen for their technical support during the fieldwork.

Conflicts of Interest: The authors declare no conflict of interest.

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