



## Winter cover crops decreased soil mineral N contents and increased soil organic C stocks and N<sub>2</sub>O emission

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### ABSTRACT

Cover crops (CC) can contribute to climate protection as a result of their effects on soil nitrogen (N) cycling and nitrous oxide (N<sub>2</sub>O) emission and by increasing soil organic carbon (SOC) stocks. This study explored the influence of different winter CC (saia oat, winter rye and spring vetch) compared with bare fallow, followed by silage maize on N<sub>2</sub>O emissions and soil mineral N (SMN) dynamics, as well as on SOC stocks in year-round replicated field plot experiments in four fields located at two sites in northern Germany (Kiel, Uelzen) over two consecutive years (2018/19 and 2019/20). Non-legume CC decreased SMN contents in 0–30 cm during the CC period, but this did not result in decreased cumulative N<sub>2</sub>O emissions over that time. Decreased emissions during CC growth were offset by increased emissions during CC mineralisation after frost and incorporation. Higher cumulative N<sub>2</sub>O emissions during the maize period in all CC treatments compared with bare fallow (significant for non-legume CC) indicated that the incorporated CC biomass still boosted N<sub>2</sub>O emissions under the following crop. Overall, including CC in the cropping system increased annual and yield-related N<sub>2</sub>O emissions compared with bare fallow (significant only for non-legumes). The increase in annual N<sub>2</sub>O emissions of  $0.84 \pm 1.06$  kg N<sub>2</sub>O-N ha<sup>-1</sup> yr<sup>-1</sup> was only partly offset by the estimated mitigation potential for indirect N<sub>2</sub>O emissions of  $0.52 \pm 0.14$  kg N<sub>2</sub>O-N ha<sup>-1</sup> yr<sup>-1</sup>. The mean annual increase in SOC induced by growing CC every fourth year over a 50-year period was 40–60 kg C ha<sup>-1</sup> yr<sup>-1</sup>. In summary, CC had both positive and negative effects on greenhouse gas exchange. Site and crop rotation optimised CC systems, and a more precise and site-specific consideration of fertilising effects might help improve the net greenhouse gas budget of CC.

### 1. Introduction

Cover crops (CC) affect numerous ecosystem services provided by agroecosystems. They can decrease soil erosion by wind and water, moderate soil temperature, boost soil organic carbon (SOC) stocks and water-holding capacity, and enhance soil porosity, aggregate stability, nutrient cycling, and pest and weed control (Thorup-Kristensen et al., 2003; Blanco-Canqui et al., 2011; Kaspar and Singer, 2011; Blanco-Canqui et al., 2013; Schipanski et al., 2014; Poepflau and Don, 2015). Positive yield effects in crop rotations have been reported in particular for legume CC, and are explained by the additional N supply from symbiotic N<sub>2</sub> fixation (Vendig et al., 2023). Cover crops have the ability to take up considerable amounts of soil mineral N (SMN) in autumn if they are established sufficiently early (Kristensen and

Thorup-Kristensen, 2004; Schipanski et al., 2014). This can reduce nitrate (NO<sub>3</sub>) leaching and contribute to groundwater protection.

The effects of CC on greenhouse gas (GHG) exchange and climate change mitigation are less clear. Several processes contribute to these effects. Among the most important are the direct and indirect emission of nitrous oxide (N<sub>2</sub>O), SOC accumulation, and saving on fertiliser through the transfer of CC-derived N to the subsequent main crop. Mineral N uptake by CC can lead to a reduction in both direct and indirect N<sub>2</sub>O emissions, particularly when established after crops with low N-use efficiency (Ruser et al., 2001; Henke et al., 2008). However, enhanced N<sub>2</sub>O emission can occur after non-persistent CC are killed by frost and/or incorporated into the soil, making the net effect of CC on N<sub>2</sub>O emission variable and highly uncertain. Cover crop species and time of termination can affect soil mineral N dynamics and N<sub>2</sub>O emission

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(Rosecrance et al., 2000; Alonso-Ayuso et al., 2014; Basche et al., 2014). In particular, different frost hardness (persistent and non-persistent CC), the composition of CC residues (e.g., C/N ratio, lignification), differences in CC biomass and N yield, and the ability to fix atmospheric N<sub>2</sub> (legume versus non-legume CC) are key factors that can influence the dynamics of SMN, NO<sub>3</sub> leaching in winter, and the extent of N<sub>2</sub>O emission during the CC period and in the subsequent main crop (Thorup-Kristensen et al., 2003; Daryanto et al., 2018).

Cover crops affect N<sub>2</sub>O emission not only by uptake of SMN and remineralisation of CC-derived N, but also by providing readily mineralisable C, which encourages denitrification (Mitchell et al., 2013). In the literature, the net effects of CC on N<sub>2</sub>O emissions are not consistent, and there have been reports of increasing, decreasing or no effect on N<sub>2</sub>O emissions for both legumes and non-legumes compared with bare fallow (Blanco-Canqui et al., 2015; Abdalla et al., 2019). In their meta-analysis, Basche et al. (2014) conclude that more work covering the entire year is needed to be able to clarify whether the cultivation of CC leads to an overall reduction in annual N<sub>2</sub>O emissions due to reduced N availability in autumn or whether emissions are only deferred to later periods.

With their ability to reduce N leaching, improve the N supply of subsequent crops and save on fertiliser usage, CC can present an attractive option for farmers (Abdalla et al., 2019). An additional benefit for soil fertility and climate change mitigation is the accumulation of SOC induced by the incorporation of CC residues. In a meta-analysis of 37 studies worldwide, Poepflau and Don (2015) estimate that winter CC can sequester about 320 ± 80 kg C ha<sup>-1</sup> yr<sup>-1</sup> in the topsoil (0–22 cm) over a period of 54 years when established annually. However, this value seems to be more a SOC sequestration potential since annual winter cover cropping is unusual and requires the annual cultivation of summer crops. More typical for many cropping systems is a change of winter and summer crops. Cover crop effects on SOC stocks are expected to be highly site-dependent and influenced by crop rotation (e.g., proportion of summer crops), the type and management of CC, and soil properties (McClelland et al., 2021; Qin et al., 2023).

The present study investigated the effects of three different CC (persistent, non-persistent and a legume CC) compared with bare fallow as the control prior to the cropping of silage maize on year-round N<sub>2</sub>O dynamics and maize yield-related N<sub>2</sub>O emission in four fields (two established in 2018 and two established in 2019). Soil mineral N, water-filled pore space (WFPS), and the N uptake and dry matter (DM) yield of CC were measured as potential explanatory variables and to estimate the potential to mitigate indirect N<sub>2</sub>O emissions. In addition, the effects of introducing these CC on SOC stocks in a typical crop rotation with maize in Germany was investigated.

The objectives and hypotheses of this study were:

- (i) to determine the effects of cultivation of different CC after harvest of oilseed rape on winter dynamics of SMN and N<sub>2</sub>O emission. Hypothesis: CC reduce winter N<sub>2</sub>O emissions by reducing SMN content, but this reduction is compensated by a rise in winter N<sub>2</sub>O emissions following the mineralisation of CC residues after freezing of frost-sensitive CC and/or CC incorporation;
- (ii) to assess the effects of these different CC on soil N dynamics and N<sub>2</sub>O emission in the subsequent main crop (silage maize). Hypothesis: CC increase N<sub>2</sub>O emissions from the subsequent maize crop, which is influenced by the biomass and N yield of the incorporated CC;
- (iii) to determine the effects of different CC on annual and maize yield-related N<sub>2</sub>O emission. Hypothesis: total annual N<sub>2</sub>O emission and maize yield-related N<sub>2</sub>O emission are higher for maize cropping systems with CC than for those without CC because in total, the potential reduction of winter N<sub>2</sub>O emissions through a reduced wintery SMN content is lower than increases in N<sub>2</sub>O emissions from mineralisation of CC residues;
- (iv) to determine CC effects on SOC sequestration at the experimental sites in a typical rotation with maize in Germany. Hypothesis: the

spatially variable biomass development of CC and specific conditions controlling mineralisation have a pronounced effect on the modelled increase in SOC stocks induced by winter CC, and  
(v) to discuss the advantages and disadvantages of introducing CC for the GHG budget of maize cropping.

## 2. Materials and methods

### 2.1. Experimental sites and fields

The study was carried out in four experimental fields at two experimental sites in northern Germany (Uelzen (UE) and Kiel (KI)) between 2018 and 2020. The chosen sites represent sandy soils on glacial till that are prone to leaching and typical sites for silage maize cropping in Germany. Two sandy sites were chosen in order to account for the natural variability between sites. In the first year (2018), the experiment began in one field at each site, and then in the following year (2019) the experiment was replicated in a neighbouring field at each site in order to account for climatic differences. Hereafter, the field experiments are referred to as UE2018/UE2019 (Uelzen) and KI2018/KI2019 (Kiel) based on the site and year in which the experiment started. Uelzen (Westerweyhe, Lower Saxony: 52.990° N, 10.500° E, 65 m a.s.l.) has a long-term (1991–2021) mean air temperature of 10.0 °C and mean annual precipitation of 743 mm (Merkel, 2022). Kiel (Hohenschulen, Schleswig-Holstein: 54.315° N, 9.979° E, 29 m a.s.l.) has a long-term (1991–2021) mean air temperature of 8.8 °C and mean annual precipitation of 724 mm (Merkel, 2022). Weather data were obtained from the experimental station at Hohenschulen (KI) and the nearby station of the German Weather Service at UE (5146) and are shown in Figure A.1 in the Appendix. According to the World Reference Base for Soil Resources (WRB), the soils of the two fields in UE are classified as Cambisol (UE2018) and Planosol (UE2019), both with sandy loam textures in the topsoil (glacial till). The soils of the two fields in KI are classified as Luvisols with sandy loam texture in the topsoil (glacial till). The main soil properties of the fields are summarised in Table 1.

### 2.2. Experimental design and treatments

Prior to this study, the experimental fields at UE had been managed by local farmers. The fields at KI on the experimental farm had been managed as commercial field. At both sites, two field experiments were set up using a randomised split plot design with CC as main plot and N-fertilisation of subsequent maize as sub-plot organised in four blocks as replicates (this study only investigated one fertilisation treatment; for more details, see chapter 2.3). At both sites, the first field experiments were established in August 2018 and the second on neighbouring fields in August 2019. Plot sizes were 3 m x 7.5 m at UE and 3 m x 14 m at KI. In all the experimental fields, oilseed rape (*Brassica napus* L.) had been grown as a pre-crop before the start of the experiment. The CC treatments evaluated in this study were: (i) bare fallow as the control, (ii) winter rye (*Secale cereale* L.; a non-leguminous persistent CC), (iii) saia

**Table 1**

Soil type, soil organic C, total N, pH, texture and bulk density in soil from 0 to 30 cm depth at the experimental sites in Uelzen (UE) and Kiel (KI) (means and standard deviation, n=4). The year-round experiments commenced in August 2018 and were replicated in August 2019.

	UE2018	UE2019	KI2018	KI2019
Soil type	Cambisol	Planosol	Luvisol	Luvisol
Organic C (%)	1.0 (0.1)	1.5 (0.2)	1.9 (0.2)	1.5 (0.4)
Total N (%)	0.08 (0.00)	0.10 (0.01)	0.14 (0.01)	0.14 (0.01)
pH (CaCl <sub>2</sub> )	5.9 (0.2)	5.2 (0.3)	6.5 (0.2)	6.2 (0.1)
Clay (%)	5.5 (0.8)	4.4 (0.3)	14.4 (2.6)	11.9 (3.1)
Silt (%)	33.9 (3.1)	26.5 (4.9)	29.2 (3.3)	25.8 (4.0)
Sand (%)	60.6 (3.8)	69.2 (5.2)	56.4 (5.5)	62.3 (6.3)
Bulk density (g cm <sup>-3</sup> )	1.5 (0.1)	1.5 (0.1)	1.3 (0.1)	1.3 (0.1)

oat (*Avena stringosa* Schreb.; a non-leguminous non-persistent CC), and (iv) spring vetch (*Vicia sativa* L.; a leguminous non-persistent CC). Silage maize (*Zea mays* L.) was grown as the subsequent crop in all experimental fields.

### 2.3. Agricultural management

#### Uelzen

For a detailed description of the management activities carried out in the two experimental fields in UE, see Table A.1 in the Appendix. Briefly, at UE2018, CC were sown on 22 August 2018. No fertiliser was applied to the CC. The fallow, saia oat and winter rye plots were treated with herbicide on 9 October 2018 in order to remove volunteer oilseed rape (vOSR). Chemical CC termination and incorporation took place on 23 March 2019 and on 6 April 2019, respectively. On 28 April 2019, winter rye plots were ploughed (25 cm) and recompacted (10 cm). All other plots underwent conservative seedbed preparation (Grubber, 20 cm). Maize was sown on 3 May 2019. The N fertilisation was adjusted to the expected N demand of maize (about 160 kg ha<sup>-1</sup>) considering the mean SMN content before seeding (36 kg N ha<sup>-1</sup>). The first application of 80 kg N ha<sup>-1</sup> took place three days after seeding (6 May 2019) and the second application of 40 kg N ha<sup>-1</sup> was carried out on 5 June 2019. Silage maize was harvested on 25 September 2019. At UE2019, CC were sown on 22 August 2019. The fallow, saia oat and winter rye plots were treated with herbicide on 19 September 2019 in order to remove vOSR. On 28 March 2019, the CC were chemically terminated. Incorporation of CC and seedbed preparation were performed on 9 April 2020. On 14 April 2020, winter rye plots were ploughed (25 cm). All other plots underwent conservative seedbed preparation (Grubber, 20 cm). On 28 April 2020, maize was sown and fertilised with 150 kg N ha<sup>-1</sup>. Winter rye plots were recompacted before sowing. Silage maize was harvested on 2 October 2020.

#### Kiel

For a detailed description of the management activities carried out at the experimental sites in KI, see Table A.2. In brief, for the experimental site KI2018, seedbed preparation and seeding of CC took place on 20 August 2018. The fallow was kept free of vegetation using a broad-spectrum herbicide. No fertiliser was applied to the CC. Chemical CC termination took place on 25 March 2019 and CC were incorporated on 5 April 2019. Maize was sown on 29 April 2019 and fertilised one day later. The expected N demand of maize (about 160 kg N ha<sup>-1</sup>) was met by SMN before seeding (50 kg N ha<sup>-1</sup>) and by N application of 110 kg N ha<sup>-1</sup>. Silage maize was harvested on 23 October 2019. Key dates for KI2019 were the seeding of cover crops on 24 August 2019, chemical termination of CC on 23 March 2020, and incorporation of CC on 5 April 2020. Seeding of maize and N fertilisation (130 kg N ha<sup>-1</sup>) were carried out on 22 April 2020. Silage maize was harvested on 13 October 2020.

### 2.4. Soil and plant sampling

#### Soil characterisation

Soil from 0 to 30 cm depth was sampled at the beginning of the experiment from each plot. For the determination of soil texture and C and N contents, the samples were dried at 40 °C until constant weight and sieved to ≤2 mm. Soil texture was analysed according to ISO 11277 by sieving and sedimentation. For C and N analyses, subsamples were milled and total C and N were determined by a C/N analyser (LECO TruMac, LECO Instruments, Mönchengladbach, Germany). The soil pH was determined potentiometrically from dried soil samples suspended in calcium chloride (CaCl<sub>2</sub> 0.01 M) and measured with a pH electrode (Mettler Toledo™ FE20 FiveEasy™ Benchtop pH Meter, Fisher Scientific, Gießen, Germany). For the determination of bulk density, 100 cm<sup>3</sup> cores were taken from 5 to 10 cm, 15–20 cm and 25–30 cm depths before cultivation and about one month after cultivation in the plots with saia oat and bare fallow. Visible stones were removed and the holes filled with sand. The cores were dried at 105 °C for 24 hours and weighed. The

bulk density was calculated from the determined DM and core volume.

#### Soil mineral N

Soil samples for the determination of SMN (sum of NO<sub>3</sub>-N and NH<sub>4</sub>-N) dynamics were taken bi-weekly from 0 to 30 cm soil depth using a Goettinger gouge auger with a diameter of 18 mm and 14 mm slot (Nietfeld GmbH, Quakenbrück, Germany). Seven samples were taken from each plot and mixed thoroughly. Soil mineral N was extracted from 80 g moist soil with a 0.0125 M CaCl<sub>2</sub> solution and quantified colorimetrically with flow-injection analysers in the laboratories of the participating research groups. Soil moisture of a further aliquot was determined gravimetrically by drying at 105 °C for 24 hours. The water-filled pore space (WFPS) was calculated using the mean soil bulk densities measured at the experimental fields and assuming a soil particle density of 2.65 g cm<sup>-3</sup>.

#### Yields

Aboveground DM and N yields of CC and maize were determined on samples taken on four dates in the CC period and at the time of maize harvest. The sampling areas for CC and maize in UE was 1 m<sup>2</sup> and 15.75 m<sup>2</sup>, respectively. In KI, it was 1 m<sup>2</sup> for CC and 19.5–21 m<sup>2</sup> for maize. Plant samples were weighed before and after drying at 60 °C to determine fresh and dry mass. A homogenised subsample was taken and milled for biomass C and N analysis. Maize yields are reported in Kühling et al. (2023), and yield-related N<sub>2</sub>O emissions were calculated based on these yields.

### 2.5. Gas flux measurements

Fluxes of N<sub>2</sub>O at the soil surface were measured using closed chambers at weekly intervals. Measurements started in August 2018 (UE2018, KI2018) or August 2019 (UE2019, KI2019) at the date of seeding of the CC and continued until the harvest of maize the following year. In UE, the chambers consisted of PVC collars (height: 15 cm, Ø 30 cm), which were installed approximately 5 cm deep into the soil. For measurements, PVC chambers (height 30 cm), each fitted with a venting tube, a thermometer and two gas-sampling ports, were placed on the collars and sealed air tight with rubber bands. Headspace air samples were drawn with a handheld electric air pump into 20-mL glass vials closed with a rubber septum. In KI, the chambers were 71 cm long, 27 cm wide and 10 cm high, and the chamber material was white opaque PVC (Ps-plastic, Eching, Germany). They were equipped with rubber sealing, a pressure vent and a ventilator. For measurements, the chambers were anchored on their frames using elastic straps, and gas samples were taken using vacutainers or stopcock vials. The frame height was 13 cm and they were installed in soil to a depth of 5–10 cm. In all the investigated fields, the chamber remained closed for 60 minutes, and gas samples were collected 0, 20, 40 and 60 minutes after chamber closure. Chamber and soil temperatures were recorded for each gas sample. In all the fields, the chambers were only removed for tillage and harvest events. Cover crops were included in the chambers. The small plots for N<sub>2</sub>O measurements (base frames of the soil covers) in the bare fallow were kept free of vegetation manually. Small vOSR seedlings that were occasionally found within the base frames of the treatments with CC were also removed by hand. For maize, chambers were placed between seed rows in all the investigated fields.

Gas samples were analysed for N<sub>2</sub>O concentration using a gas chromatograph (GC-2014, Shimadzu, Duisburg, Germany) equipped with an electron capture detector (ECD) and connected to an autosampler (Greenhouse Workstation AS-210; SRI Instruments Europe GmbH, Bad Honnef, Germany). Four standard gases with concentrations from 300 to 3000 ppb N<sub>2</sub>O in synthetic air were used for calibration. The precision of the GC was regularly tested by repeated measurement of standards with gas concentrations close to ambient, and the coefficient of variance (CV; n = 10) was always <2%. Air samples were generally analysed within 2 weeks after sampling with a maximum storage time of <8 weeks.

## 2.6. Potential mitigation of indirect N<sub>2</sub>O emission

The potential for mitigating indirect N<sub>2</sub>O emissions induced by NO<sub>3</sub> leaching was calculated from the maximum N uptake in aboveground CC biomass before winter (N in aboveground plant biomass in mid-December) and the IPCC (2019) N<sub>2</sub>O emission factor for indirect emissions induced by N leaching and run-off (EF<sub>5</sub> = 0.011 kg N<sub>2</sub>O-N kg<sup>-1</sup> N). The CO<sub>2</sub>-equivalents (CO<sub>2eq</sub>) were calculated using the N<sub>2</sub>O global warming potential of 273 (IPCC, 2022). This approach should not be used for legume CC, because biomass N in legume CC can originate from N<sub>2</sub> fixation and does not necessarily reflect a related reduction in SMN. As a consequence, due to the high uncertainty of the amount of N scavenged from the SMN pool, spring vetch was excluded from the analysis of a mitigation potential for indirect N<sub>2</sub>O emissions.

Due to different contributions of vOSR to total N uptake, we separately determined N uptake into aboveground biomass of CC and vOSR. The amount of N in vOSR biomass was subtracted from the maximum N uptake of the respective CC treatment to calculate the maximum amount of N stored in shoots of the CC itself. At UE2018, vOSR largely contributed to total plant N uptake (44 ± 21%; Table A.6). We calculated potential reduction of indirect N<sub>2</sub>O emission with and without results from UE2018 to show how this site influenced the uncertainty of our results.

## 2.7. Calculations and statistical analyses

### Flux calculation and cumulative N<sub>2</sub>O emissions

Gas fluxes were calculated in R version 4.3.2 (RCoreTeam, 2023) using the Hutchinson-Mosier non-linear function (Pedersen et al., 2010) as implemented in the *gasfluxes* package (Fuss and Hueppi, 2020) or robust linear regression with a Huber-M estimator (Huber and Ronchetti, 1981) following the approach of Hüppi et al. (2018). If only three data points for a flux measurement were available, linear regression was used. The N<sub>2</sub>O fluxes were cumulated over the CC period ("CC", starting with the seeding of CC and ending with the seeding of maize) and the maize period ("M," starting at the time of maize seeding and ending with maize harvest). Cumulative fluxes were calculated based on linear interpolation between measurement dates. Since no measurements were taken on the date of maize seeding, fluxes from the last date before maize seeding and from the first date after maize seeding were horizontally extrapolated to the date of maize seeding in order to be able to calculate cumulative N<sub>2</sub>O fluxes for CC and M. Annual N<sub>2</sub>O emissions were calculated as (CC+M)/days from seeding of CC until harvest of M x 365. Furthermore, to take a closer look at periods that have particular potential to decrease (CC growth and major N uptake) and increase N<sub>2</sub>O emissions (CC mineralisation), the CC period was divided into three phases: (1) a pre-winter period from August to mid-December when the major CC growth and N uptake occurs, (2) a mid-winter period (from mid-December until CC incorporation) with lower to no growth and a potential for the dying off and increased mineralisation of non-persistent CC, and (3) the period after CC incorporation up to the seeding of maize, when SMN dynamics are heavily influenced by mineralisation of CC biomass.

### Statistical analyses

Statistical analyses were carried out using R version 4.3.2 (RCoreTeam, 2023). A small number of fluxes (four fluxes in total on different fields) showed exceptionally large associated uncertainties (standard errors >120 µg N<sub>2</sub>O-N m<sup>-2</sup> h<sup>-1</sup>). However, since calculations without these fluxes did not give statistically different results from calculations including these fluxes, they were not removed from the dataset. Standard errors of fluxes were not significantly correlated with flux magnitude. In order to ensure variance homogeneity of residuals in the statistical models, cumulative N<sub>2</sub>O fluxes were log<sub>10</sub> transformed (Stehfest and Bouwman, 2006). Variance homogeneity and approximate normality of residuals were assessed using diagnostic plots. In case of non-homogeneity of variance, a Welch-ANOVA followed by a

Games-Howell-Test as a post-hoc test (package *rstatix*; Kassambara, 2023) was conducted. The effects of CC on SMN, WFPS and N<sub>2</sub>O emissions were tested both field-wise and across all fields combined. Field-wise tests were carried out using analysis of variance (ANOVA) based on a linear mixed-effects model, with CC as the fixed effect and block as the random effect, using the package *tidyverse* (Wickham et al., 2019). In the event of a significant effect of CC treatments, a Tukey HSD post-hoc test (alpha = 0.05) was performed for pairwise comparisons. For synthesis of CC effects across all fields, a generalised least-squares regression model with CC, site and year of establishment and their interactions as fixed effects was fitted using the *nmle* package (Pinheiro et al., 2023). Variance heterogeneity was accounted for with the variance structure  $\sigma^2 \times |\hat{\gamma}|^{2\beta}$  (Zuur et al., 2009) if it was significant. In the event of a significant effect of CC treatment in the ANOVA, pairwise comparison of means with p-value adjustment (Tukey) was carried out on estimated marginal means using the *emmeans* package (Lenth, 2023). The relationships between different parameters such as N yield and DM yield of CC, SMN contents, WFPS, and N<sub>2</sub>O emission in different periods were estimated by fitting linear mixed-effects models using the *lmerTest* package (Kuznetsova et al., 2017) and the *lme4* package (Bates et al., 2015).

## 2.8. Modelling changes in soil organic carbon stocks

The effects of CC on changes in SOC stocks were simulated using a model ensemble consisting of the two process-based soil C models RothC (Coleman and Jenkinson, 1996) and C-TOOL (Taghizadeh-Toosi et al., 2014), as described by Seitz et al. (2022). Cover crop effects were analysed for the typical rotation: CC (or fallow) – silage maize – winter wheat – winter oilseed rape – winter wheat, resulting in a winter CC every fourth year. Organic C inputs via the residues of main crops were estimated from crop yields and allometric functions describing the relation to aboveground and belowground crop residues (Franko et al., 2011; Rösemann et al., 2017; Jacobs et al., 2020). The straw of wheat and oilseed rape remained on the fields. Silage maize was used for biogas production and the digestates (30 m<sup>3</sup>) were applied to fertilise the maize. Related C inputs via digestates were included. For CC, C inputs from aboveground crop residues were measured at the experimental sites in KI and UE. Crop residues from belowground inputs were estimated from the sampled aboveground crop residues and root/shoot ratios, which were derived from ratios of crop-specific aboveground and belowground biomasses (Grunwald et al., 2023). Local climate conditions (precipitation, temperature, radiation) and soil properties (Table 1) were additional model input parameters. Changes in SOC (0–30 cm) were simulated for the crop rotation and for a time horizon of 50 years, varying the CC species (winter rye, saia oat, spring vetch) and without a CC (fallow).

## 3. Results

### 3.1. Biomass and N uptake of cover crops

After the main CC growth period (i.e., from seeding in August until mid-December), their biomass was 1.2–1.8 t DM ha<sup>-1</sup> in UE and 1.1–3.5 t DM ha<sup>-1</sup> in KI (Table 2, left-hand section). Considerable amounts of vOSR had grown in the bare fallow plots at UE in both years (0.6–1.4 t DM ha<sup>-1</sup>; Table 2, left-hand section). It was also found in CC plots, with the highest proportions under spring vetch in all the investigated fields (up to 77% of DM biomass; Table 2, left-hand section). The maximum pre-winter N uptake of CC (including vOSR) ranged from 40 to 62 kg N ha<sup>-1</sup>, with no significant differences between CC species. For the non-legume CC, vOSR contributed 1.5±3.2% to the maximum N uptake except for UE2018, where contributions of vOSR to the total N uptake were much higher (44 ± 21%; Table A.6). In the spring vetch plots, the maximum N uptake in CC biomass excluding vOSR was 25 ±

**Table 2**

Aboveground DM and N yield of CC (including vOSR) and the proportion of vOSR at the time of maximum N uptake and CC incorporation in spring (means and standard deviation, n= 4).

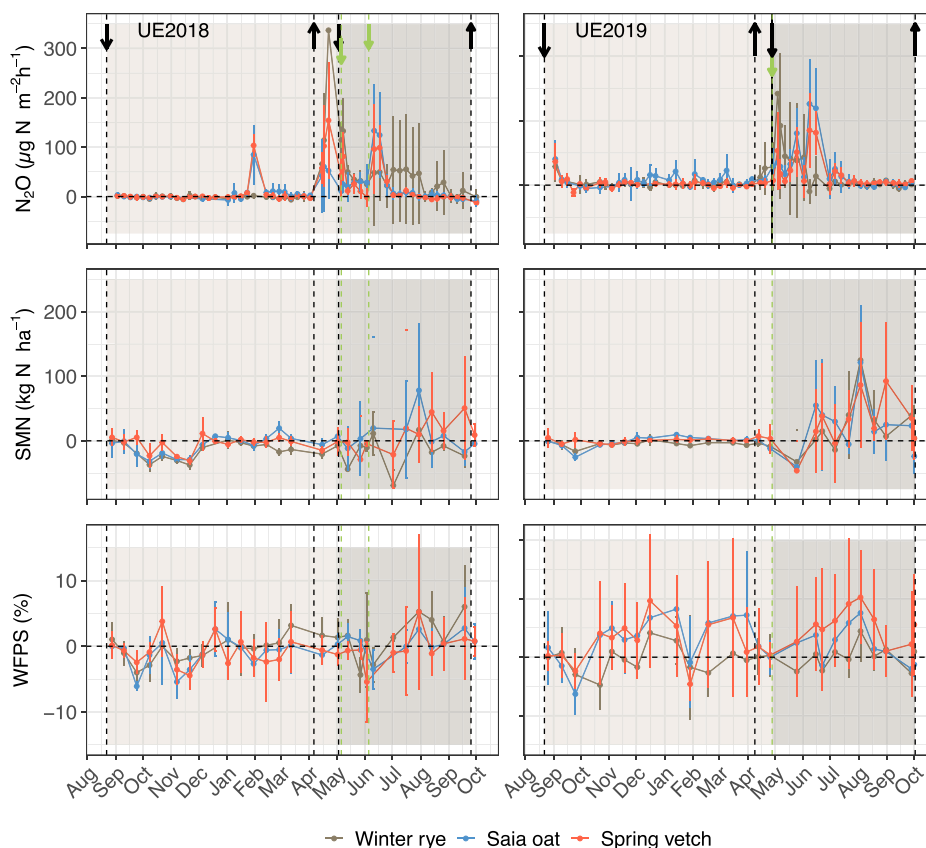
Field / Treatment	vOSR (%)	DM (t ha <sup>-1</sup> )	N yield (kg ha <sup>-1</sup> )	vOSR (%)	DM (t ha <sup>-1</sup> )	N yield (kg ha <sup>-1</sup> )
At the time of pre-winter maximum N uptake				At the time of CC incorporation in spring		
UE2018						
Fallow	100 (0)	1.4 (0.6)	a 47.4 (14.4)	a	0	
Winter rye	41 (31)	1.3 (0.2)	a 39.8 (4.7)	a	2 (3)	41.8 (11.3) ab
Saia oat	51 (9)	1.2 (0.1)	a 39.7 (2.9)	a	1 (3)	33.6 (7.6) a
Spring vetch	54 (6)	1.6 (0.2)	a 60.6 (17.5)	a	62 (21)	58.2 (10.1) b
UE2019						
Fallow	100 (0)	0.6 (0.3)	a 17.5 (9.8)	a	100 (0)	5.5 (2.1) a
Winter rye	5 (6)	1.5 (0.5)	b 40.6 (13.4)	ab	0	18.4 (2.6) b
Saia oat	0	1.8 (0.2)	b 48.4 (1.1)	b	0	n.a.§
Spring vetch	39 (14)	1.4 (0.4)	b 44.5 (9.7)	b	64 (5)	26.3 (8.4) b
KI2018						
Fallow	0				0	
Winter rye	0	2.3 (0.3)	a 62.4 (12.1)	a	0	54.4 (11.4) a
Saia oat	0	3.5 (1.1)	a 59.7 (20.2)	a	0	n.a.§
Spring vetch	77 (7)	2.0 (0.2)	a 61.0 (3.9)	a	100 (0)	51.0 (11.5) a
KI2019						
Fallow	0				0	
Winter rye	1 (1)	1.3 (0.3)	a 46.7 (9.2)	a	0	51.7 (3.6) b
Saia oat	5 (4)	1.7 (0.4)	a 43.8 (8.2)	a	0	51.3 (6.2) b
Spring vetch	52 (16)	1.1 (0.1)	a 45.2 (1.4)	a	61 (8)	40.0 (5.9) a

§n.a. – no biomass available for sampling due to CC dying off in winter

12 kg N ha<sup>-1</sup> (Table A.6). The plant biomass of saia oat (non-persistent) was strongly affected by site and year dependent frost periods. Saia oat was completely killed at KI2018 and UE2019 and biomass was largely decomposed before incorporation in April (Table 2). At the other sites, saia oat was only partly killed and the harvested biomass was a mixture

of dead and living plant parts.

Aboveground CC biomass before incorporation ranged from 0 to 3.2 t DM ha<sup>-1</sup>, with the highest amount in saia oat plots in KI2019, and values of zero where no plant biomass was available for sampling due to the CC being killed by frost and already largely decomposed (Table 2,



**Fig. 1.** Differences in N<sub>2</sub>O flux, SMN content and WFPS, both in 0–30 cm, of CC treatments compared with bare fallow (black dotted horizontal line at zero) for UE2018 (left) and UE2019 (right). Means and standard deviation (n=4). The CC phase is marked in light grey, the subsequent maize period in darker grey. Green arrows indicate fertilisation. Black downward and upward arrows indicate seeding and termination/harvest of CC (open arrow) and maize (closed arrow).

right-hand section). The N uptake in CC at this time ranged from 0 to 58 kg N ha<sup>-1</sup> with the highest amount the spring vetch plots, but again with high contributions of vOSR in the spring vetch plots (60–100% of DM biomass; Table 2, right-hand section). In the non-legume CC, vOSR made up < 2% of DM biomass.

### 3.2. Dynamics of soil mineral N, water-filled pore space and N<sub>2</sub>O emissions

Soil mineral N contents in 0–30 cm during the CC period (Figs. A.2–A.5) mostly consisted of NO<sub>3</sub><sup>-</sup> (data not shown). At the time of CC seeding, they amounted 38–75 kg N ha<sup>-1</sup> and decreased to 5–32 kg N ha<sup>-1</sup> up to mid-December (Figs. A.2–A.5). Soil mineral N contents did not increase immediately after incorporation of CC, but only after fertilisation of maize (Figs. 1–2 and Figs. A.2–A.5). During the main CC growth period (pre-winter), SMN contents were lower in CC treatments than bare fallow in all fields except for KI2019, but for this field, there were only a few measurement dates in winter (Fig. 2). The effects on WFPS during the CC period were inconsistent (Figs. 1–2), and no significant differences between treatments were found in the average WFPS throughout the CC period (Table A.4).

Emissions of N<sub>2</sub>O were generally at a low level during the CC period, with only a few differences between persistent and non-persistent CC and between CC treatments and bare fallow (Figs. 1–2 and A.2–A.5). Only in January 2019 was a frost-induced N<sub>2</sub>O peak visible in the non-persistent CC at UE2018 (Fig. 1 and A.2). Although this frost event also occurred in KI and led to the freezing of non-persistent CC, no such N<sub>2</sub>O peak was observed at this site (Fig. 2 and A.4). An increase in N<sub>2</sub>O fluxes under all CC species occurred after CC incorporation in April, with the greatest increase in winter rye plots at UE2018 (Figs. 1–2). Emissions

of N<sub>2</sub>O were generally at a higher level during maize cropping compared with the previous CC period until July or August, with distinct peaks after fertilisation (Figs. 1–2).

### 3.3. Cumulative N<sub>2</sub>O emissions and average SMN contents (0–30 cm)

#### Cover crop period

The average topsoil SMN contents during the CC period were at a lower level in UE2019 (11.1–17.6 kg N ha<sup>-1</sup>) than in UE2018 (30.4–43.6 kg N ha<sup>-1</sup>) or in KI (23.5–37.8 kg N ha<sup>-1</sup>, Table 3). The non-leguminous CC (winter rye and saia oat) tended to have lower average SMN contents compared with bare fallow, but this trend was only significant for different CC at different sites (winter rye in UE in both years, saia oat in KI2018; Table 3). Cumulative N<sub>2</sub>O emissions of the CC period ranged from 0.2 to 1.3 kg N ha<sup>-1</sup>, with no significant differences between the CC species at any site or in any year and no differences between CC treatments and bare fallow except for UE2018, where they were higher in all systems with CC compared with bare fallow (Table 3). Over the total CC period, N<sub>2</sub>O emissions were positively correlated with the DM of CC at the end of the CC period ( $p < 0.05$ ) and negatively correlated with the maximum DM yield ( $p < 0.01$ ; Table A.3). In mid-winter, N<sub>2</sub>O emissions were driven by the maximum N yield in CC ( $p < 0.05$ ). The WFPS affected N<sub>2</sub>O emissions during all phases of the CC period ( $p < 0.01$ – $p < 0.001$ ; Table A.3).

#### Maize period

Mean SMN contents during the maize period did not differ between CC treatments and bare fallow in any of the investigated fields (Table 3). Cumulative N<sub>2</sub>O emissions of the maize period were 0.5–2.1 kg N ha<sup>-1</sup> (Table 3), with no differences between CC treatments in any of the four fields. An effect of CC treatments compared to bare fallow was found

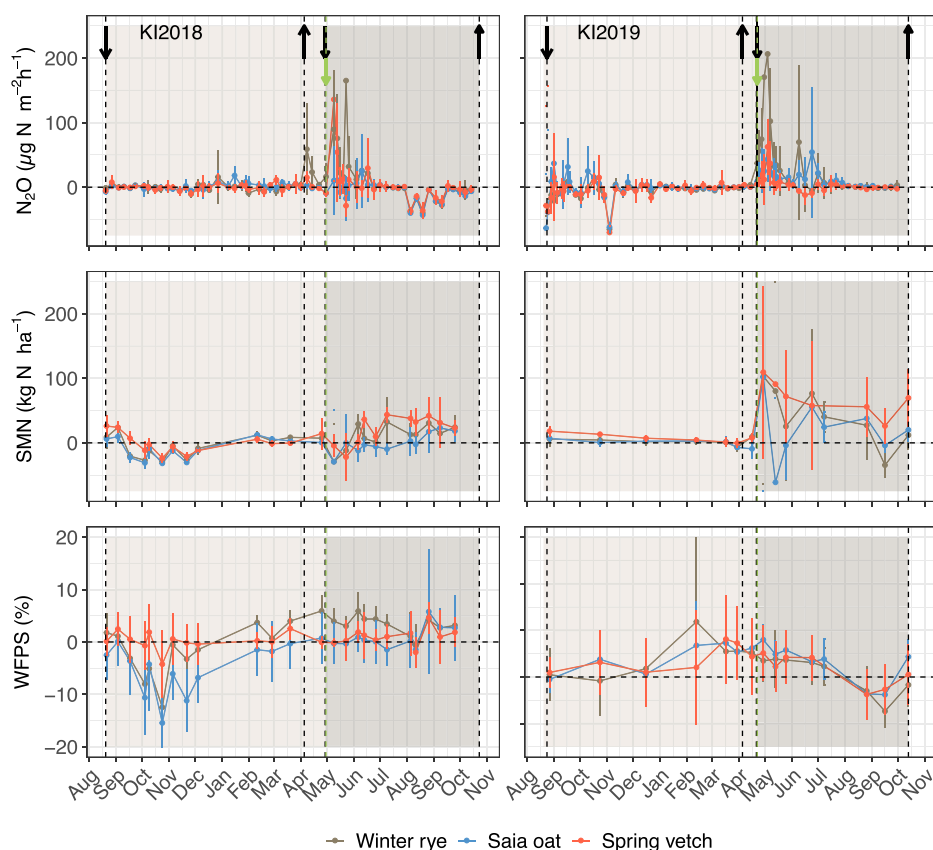


Fig. 2. Differences in N<sub>2</sub>O flux, SMN content and WFPS, both in 0–30 cm, of CC treatments compared with bare fallow (black dotted horizontal line at zero) for KI2018 (left) and KI2019 (right). Means and standard deviation ( $n=4$ ). The CC phase is marked in light grey, the subsequent maize period in darker grey. Green arrows indicate fertilisation. Black downward and upward arrows indicate seeding and termination/harvest of CC (open arrow) and maize (closed arrow).

**Table 3**

Cumulative N<sub>2</sub>O emissions and mean SMN content (0–30 cm) for the experimental fields during the CC period (seeding of CC to seeding of maize), the succeeding maize period (seeding to harvest), and the total annual experimental period (seeding of CC to harvest of maize) (means and standard deviation, n = 4). Different letters indicate significant differences between the treatments for each site (p < 0.05).

Field / treatment	Cum. N <sub>2</sub> O (kg N ha <sup>-1</sup> )	Mean SMN (kg N ha <sup>-1</sup> )	Cum. N <sub>2</sub> O (kg N ha <sup>-1</sup> )	Mean SMN (kg N ha <sup>-1</sup> )	Cum. N <sub>2</sub> O (kg N ha <sup>-1</sup> )	Mean SMN (kg N ha <sup>-1</sup> )
	Cover crop period		Maize period		Annual	
UE2018						
Fallow	0.2 (0.1)	a 43.6 (5.3)	b 0.8 (0.1)	a 76.8 (26.4)	a 0.9 (0.2)	a 59.3 (11.5)
Winter rye	1.3 (0.5)	b 30.4 (3.8)	a 2.1 (1.5)	a 61.0 (13.7)	a 3.2 (1.7)	ab 46.0 (9.0)
Saia oat	0.7 (0.3)	b 37.6 (4.5)	ab 1.5 (0.5)	a 84.5 (26.0)	a 2.1 (0.5)	b 61.9 (16.4)
Spring vetch	1.1 (0.4)	b 40.2 (0.7)	b 1.4 (0.2)	a 89.3 (16.0)	a 2.4 (0.5)	b 66.0 (9.1)
UE2019						
Fallow	0.2 (0.1)	a 16.8 (1.3)	b 0.6 (0.3)	a 115.2 (7.2)	a 0.7 (0.3)	a 57.2 (5.3)
Winter rye	0.4 (0.1)	a 11.1 (1.7)	a 1.2 (0.8)	ab 136.9 (9.9)	b 1.4 (0.8)	ab 62.5 (3.9)
Saia oat	0.5 (0.3)	a 15.1 (1.8)	b 1.6 (0.3)	b 139.0 (11.6)	ab 1.9 (0.6)	b 65.2 (4.0)
Spring vetch	0.3 (0.1)	a 17.6 (2.3)	b 1.3 (0.2)	b 142.5 (29.6)	ab 1.5 (0.3)	ab 68.5 (12.3)
KI2018						
Fallow	0.1 (0.0)	a 37.6 (3.1)	b 0.8 (0.7)	a 98.3 (19.0)	a 0.8 (0.6)	a 67.0 (9.9)
Winter rye	0.3 (0.3)	a 32.7 (2.1)	ab 1.0 (0.5)	a 112.3 (10.7)	a 1.1 (0.6)	a 70.9 (5.7)
Saia oat	0.2 (0.1)	a 29.2 (1.9)	a 0.5 (0.2)	a 100.5 (15.9)	a 0.6 (0.2)	a 63.4 (8.6)
Spring vetch	0.2 (0.2)	a 37.8 (3.2)	b 0.8 (0.5)	a 120.2 (8.2)	a 0.8 (0.5)	a 77.0 (2.6)
KI2019						
Fallow	0.7 (1.0)	a 23.6 (2.2)	a 0.5 (0.3)	a 116.2 (20.1)	a 1.0 (1.1)	a 75.6 (11.2)
Winter rye	0.4 (0.2)	a 26.1 (3.7)	ab 1.3 (0.8)	a 156.7 (44.1)	a 1.5 (0.6)	a 98.2 (23.8)
Saia oat	0.6 (0.3)	a 23.5 (4.6)	ab 0.9 (0.5)	a 139.8 (10.4)	a 1.4 (0.4)	a 87.2 (3.9)
Spring vetch	0.5 (0.4)	a 31.0 (1.1)	b 0.6 (0.2)	a 184.6 (60.8)	a 0.9 (0.5)	a 115.6 (34.9)

only for saia oat and spring vetch at UE2019.

#### Annual N<sub>2</sub>O emissions

On an annual basis, no differences in mean SMN contents were found between the CC treatments or between the CC treatments and bare fallow for any field (Table 3). Annual N<sub>2</sub>O emissions ranged from 0.7 to 3.2 kg N ha<sup>-1</sup> (Table 3). No significant differences in annual N<sub>2</sub>O emissions were observed between CC species in any of the four fields, but annual N<sub>2</sub>O emissions were higher in all treatments with CC compared with bare fallow for UE2018, and higher for the treatment with saia oat than for bare fallow for UE2019 (Table 3). No differences between CC and fallow were found in KI. The overall increase (calculated for the non-leguminous CC across all fields combined) in annual direct N<sub>2</sub>O emission after the implementation of CC into the crop sequence of oilseed rape followed by maize was 0.84 ± 1.06 kg N<sub>2</sub>O-N ha<sup>-1</sup> yr<sup>-1</sup> (equal to 361 ± 456 kg CO<sub>2-eq</sub> ha<sup>-1</sup> yr<sup>-1</sup>). With spring vetch included, the overall increase in annual direct N<sub>2</sub>O emission was 0.75 ± 0.98 kg N<sub>2</sub>O-N ha<sup>-1</sup> yr<sup>-1</sup> (equal to 320 ± 423 kg CO<sub>2-eq</sub> ha<sup>-1</sup> yr<sup>-1</sup>).

#### 3.4. Maize yields and yield-related N<sub>2</sub>O emissions

Nitrogen yields of maize were 114–226 kg ha<sup>-1</sup> with no differences between treatments in any field (Table 4). The generally lower N yields in UE2019 were due to the reduced growth of some plants where seed placement was too deep. Yield-related annual N<sub>2</sub>O emissions ranged from 0.04 to 0.19 kg N t<sup>-1</sup> maize DM yield ha<sup>-1</sup> and were generally lower in KI than in UE (Table 4). No significant differences were found between CC species in any field except for saia oat at UE2019, which had higher yield-related N<sub>2</sub>O emissions than the other CC treatments and bare fallow. Significant effects of CC on yield-related N<sub>2</sub>O emissions compared to bare fallow were only found for UE2018 and for saia oat in UE2019 (Table 4).

#### 3.5. N uptake of cover crops and mitigation potential for indirect N<sub>2</sub>O emission

The total aboveground N uptake into CC was used as an indicator of the potential of CC to reduce SMN and N leaching in winter. The N uptake into the non-legume CC excluding vO<sub>2</sub>R was 21–24 kg N ha<sup>-1</sup> at UE2018 and 38–62 kg N ha<sup>-1</sup> in the other three fields (Table 5). Multiplying the corrected maximum amount of N in CC biomass by the

**Table 4**

N yield of maize and DM yield-related N<sub>2</sub>O emission (means and standard deviation, n=4) calculated from the maize DM yield and annual N<sub>2</sub>O-N emission. Different letters indicate significant differences between treatments for each field.

Field	Treatment	N yield# (kg ha <sup>-1</sup> )	N <sub>2</sub> O-N per t DM* maize yield (kg t <sup>-1</sup> DM ha <sup>-1</sup> )
UE2018	Fallow	217.1 (15.0)	a 0.05 (0.01)
	Winter rye	175.9 (32.5)	a 0.19 (0.09)
	Saia oat	208.4 (5.5)	a 0.12 (0.02)
	Spring vetch	197.9 (20.2)	a 0.13 (0.02)
UE2019	Fallow	114.0 (38.8)	a 0.08 (0.04)
	Winter rye	136.9 (32.6)	a 0.15 (0.09)
	Saia oat	124.8 (17.5)	a 0.19 (0.06)
	Spring vetch	167.6 (20.4)	a 0.12 (0.03)
KI2018	Fallow	200.1 (8.9)	a 0.05 (0.04)
	Winter rye	226.0 (23.1)	a 0.06 (0.03)
	Saia oat	209.3 (15.5)	a 0.04 (0.01)
	Spring vetch	207.7 (16.5)	a 0.06 (0.03)
KI2019	Fallow	177.9 (43.9)	a 0.07 (0.05)
	Winter rye	193.6 (31.7)	a 0.10 (0.04)
	Saia oat	196.6 (31.3)	a 0.10 (0.04)
	Spring vetch	165.5 (15.3)	a 0.08 (0.05)

#low maize N yields for UE2019 were due to the placement of some maize seeds being too deep

\*DM yields of the investigated fields can be found in Kühling et al. (2023)

IPCC (2019) emissions factor for indirect N<sub>2</sub>O emission induced by leaching and runoff led to a mitigation potential for indirect N<sub>2</sub>O emissions of 0.42–0.69 kg N<sub>2</sub>O-N ha<sup>-1</sup> yr<sup>-1</sup> for non-legume CC, depending on CC and field (Table 5). On average over all fields combined, the mitigation potential for the non-legumes was 0.47 ± 0.19 kg N ha<sup>-1</sup> yr<sup>-1</sup> (equal to 201 ± 81 kg CO<sub>2-eq</sub> ha<sup>-1</sup> yr<sup>-1</sup>). Excluding UE2018 with its high contributions of vO<sub>2</sub>R, the potential was 0.52 ± 0.14 kg N ha<sup>-1</sup> yr<sup>-1</sup> or 224 ± 60 kg CO<sub>2-eq</sub> ha<sup>-1</sup> yr<sup>-1</sup> (Table 5).

#### 3.6. Synthesis of cover crop effects across all four investigated fields

Across all investigated fields combined, a significant reduction in the mean topsoil SMN content by CC was found for the non-legume CC (p < 0.001; Tables A.4/A.5; Fig. 3), which resulted to a major part from a significant reduction during the main CC growth period (pre-winter; p

**Table 5**

Mitigation potential for indirect N<sub>2</sub>O emissions induced by N leaching calculated using the pre-winter maximum N uptake of CC excluding vOSR, the IPCC (2019) default emission factor for N leaching (0.011 kg N<sub>2</sub>O-N kg<sup>-1</sup> N leached), and a GWP<sub>100</sub> value for N<sub>2</sub>O of 273 (means and standard deviation, n = 4). The overall mean was calculated across the two non-legume CC\* and three of four fields<sup>§</sup> (i.e., excl. UE2018; n = 32) as well as all four fields (i.e., incl. UE2018; n=48).

Field	CC#	N uptake in CC (kg N ha <sup>-1</sup> )	Mitigation potential (kg N <sub>2</sub> O-N ha <sup>-1</sup> yr <sup>-1</sup> )	Mitigation potential (kg CO <sub>2eq</sub> ha <sup>-1</sup> yr <sup>-1</sup> )
UE2018 <sup>§</sup>	Winter rye	23.5 (9.5) <sup>§</sup>	0.26 (0.1) <sup>§</sup>	111 (45) <sup>§</sup>
	Saia oat	20.5 (4.5) <sup>§</sup>	0.23 (0.1) <sup>§</sup>	97 (22) <sup>§</sup>
UE2019	Winter rye	38.2 (11.1)	a 0.42 (0.12)	a 180 (52)
	Saia oat	48.4 (1.1)	a 0.53 (0.01)	a 229 (5)
KI2018	Winter rye	62.4 (12.1)	a 0.69 (0.13)	a 294 (57)
	Saia oat	59.7 (20.2)	a 0.66 (0.22)	a 282 (95)
KI2019	Winter rye	46.2 (8.8)	a 0.51 (0.10)	a 218 (41)
	Saia oat	41.0 (8.5)	a 0.45 (0.09)	a 194 (40)
Overall mean (incl. UE2018)		42.5 (17.3)	0.47 (0.19)	201 (81)
Overall mean (excl. UE2018)		49.3 (13.8)	0.52 (0.14)	224 (60)

\*Spring vetch was excluded, because of an unknown contribution of biological N<sub>2</sub> fixation to total N uptake

<sup>§</sup>UE2018 was excluded because of high contributions of vOSR to total N uptake of 44 ± 21% (cp. Table A.6)

<0.001). In the mid-winter period, the non-persistent CC saia oat showed higher mean SMN contents than the persistent CC winter rye (p <0.001), but no significant differences were found between bare fallow and the CC (Table A.5; Fig. 3). The cumulated N<sub>2</sub>O emissions during mid-winter were also higher for non-persistent CC (saia oat and spring vetch) than winter rye (p < 0.05; Table A.5; Fig. 3). Further, emissions under saia oat were significantly higher than in the bare fallow during this period (p <0.01). In the period after CC incorporation, winter rye and spring vetch treatments showed increased emissions compared with bare fallow with the most pronounced increase in this period under winter rye (p <0.01; Fig. 3; Tables A.4/A.5). Over the total CC period, cumulative N<sub>2</sub>O emissions of the non-legume CC were increased compared with bare fallow (p<0.05; Fig. 3; Table A.5).

During the maize period, mean SMN contents in the topsoil were higher in the spring vetch treatment than for bare fallow (p <0.01; Fig. 3; Table A.5). Water-filled pore space did not differ between the bare fallow and CC during any of the investigated periods (Fig. 3; Table A.4). Emissions of N<sub>2</sub>O during the maize period (Fig. 3), as well as on the annual scale and for yield-related N<sub>2</sub>O emissions (Fig. 4) tended to be higher in all treatments with CC than for bare fallow, but this increase was only significant for non-legume CC (p <0.05, Tables A.4/A.5).

### 3.7. Changes in soil organic carbon

Crop yields and the mean annual organic C input with crop residues (shoots and roots) and digestates for the rotation without CC were higher for KI (C input of 3.97 t C ha<sup>-1</sup> yr<sup>-1</sup>) than for UE (3.07 t C ha<sup>-1</sup> yr<sup>-1</sup>). Growing CC every fourth year before maize increased this input by 330 kg C ha<sup>-1</sup> yr<sup>-1</sup> at KI and by 220 kg C ha<sup>-1</sup> yr<sup>-1</sup> at UE. Cultivation of CC increased the mean annual C input within the rotation by 10.7% and 7.2% at KI and UE respectively. Fig. 5 shows the related simulated

increase in SOC stocks over a 50-year period. The mean annual increase in SOC induced by growing CC in every fourth year was 60 kg C ha<sup>-1</sup> (spring vetch: 40 kg C ha<sup>-1</sup>, saia oat: 80 kg C ha<sup>-1</sup>, winter rye: 70 kg C ha<sup>-1</sup>) at KI and 40 kg C ha<sup>-1</sup> (spring vetch: 40 kg C ha<sup>-1</sup>, saia oat: 40 kg C ha<sup>-1</sup>, winter rye: 30 kg C ha<sup>-1</sup>) at UE. After 50 years, these rates sum up to a total increase in SOC stock of 3.1 t C ha<sup>-1</sup> at KI and of 1.9 t C ha<sup>-1</sup> at UE (Fig. 5), which equals a relative increase in SOC compared with the control without CC of 5.3% (KI) and 4.4% (UE). The highest soil C accumulation was found for saia oat at KI with a total increase of SOC of 4 t C ha<sup>-1</sup> after 50 years.

## 4. Discussion

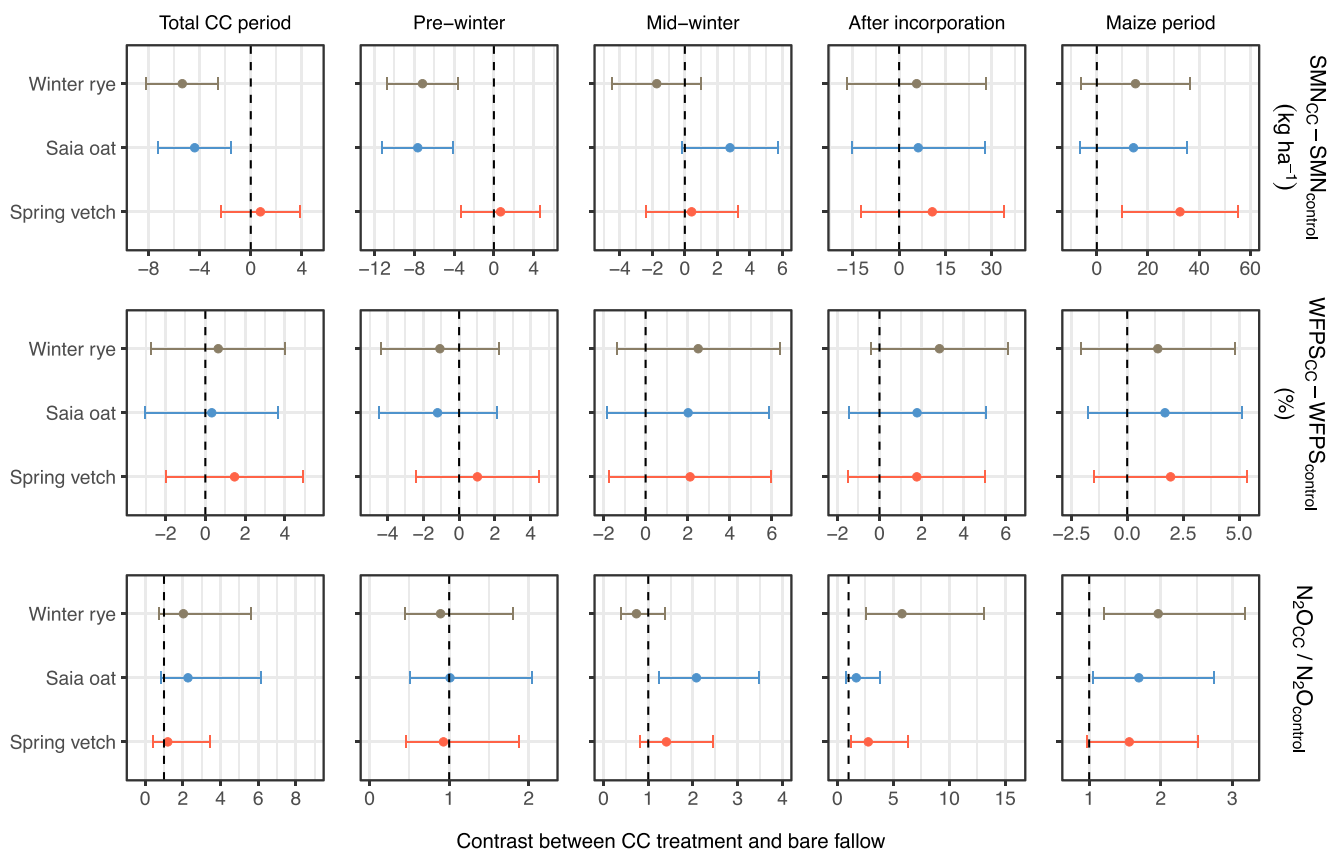
### 4.1. Effects of cover crops on water-filled pore space and soil mineral N contents in 0–30 cm and on direct N<sub>2</sub>O emissions in winter

The high water demand of CC, which can result in reduced soil water availability and a lack of water for the next main crop, can be a risk presented by CC (Smit et al., 2019). Cover crops slightly reduced soil moisture content during the main growing period in autumn (Figs. 1–2; exception KI2019), but there was no evidence that WFPS in the topsoil differed from the fallow treatment at the date of maize seeding. Precipitation during winter was evidently high enough to offset initial WFPS differences.

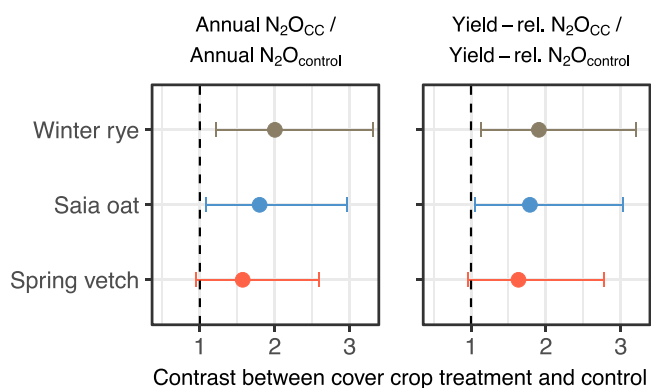
Among the postulated benefits of CC is the uptake of excess N in autumn and winter, which in turn may lead to a reduction in NO<sub>3</sub> leaching and in direct and indirect N<sub>2</sub>O emissions (Ruser et al., 2001; Kristensen and Thorup-Kristensen, 2004; Schipanski et al., 2014). A significant decrease in SMN in winter as found for non-legume CC in the investigated fields, but not under the legume spring vetch is in agreement with the findings of previous studies that legumes take up less SMN than non-legumes (e.g., Kaspar and Singer, 2011; Ramirez-Garcia et al., 2015; Valkama et al., 2015). This is generally attributed to the ability of legumes to fix N<sub>2</sub> from the atmosphere in addition to scavenging N from the SMN pool (Ramirez-Garcia et al., 2015; Daryanto et al., 2018), but may also have resulted from a shallower rooting system and a lower growth rate in autumn and winter compared with non-legume CC (Dabney et al., 2011; Grunwald et al., 2023; Kühling et al., 2023). The determined SMN contents were influenced by the N uptake of vOSR, which makes the assignment of the SMN effect to a specific CC type more uncertain. In the winter rye and saia oat treatments, proportions of N uptake by vOSR were generally low (1.5 ± 3.2% of the total plant N uptake at three of the four investigated fields) and changes in SMN were mainly due to CC N uptake. However, for UE2018, relative N uptake by vOSR was much higher (44 ± 21%) which makes the assignment of the measured SMN values to a specific CC more uncertain. Growth of the spring vetch was generally slow at the investigated fields, which was also reflected by rather low N yields of spring vetch biomass when correcting for the contributions of N in vOSR: while the N uptake in non-leguminous CC in this study was within the range found in other studies, the N uptake into spring vetch was lower than reported in the literature (between 50 and > 100 kg N ha<sup>-1</sup>; Mueller and Thorup-Kristensen, 2001; Thorup-Kristensen et al., 2003; Tonitto et al., 2006; Kaspar and Singer, 2011). Assuming a contribution of ~65% of N<sub>2</sub> fixation of spring vetch as found by Mueller and Thorup-Kristensen (2001), total N uptake in spring vetch biomass of 50–100 kg N ha<sup>-1</sup> correspond to a scavenge from the SMN pool between 33 and 65 kg N ha<sup>-1</sup>. This indicates that soil N uptake of spring vetch grown as a winter CC can be much higher than found in our study.

The decreased topsoil SMN contents under non-legume CC did not lead to a decrease in cumulative direct N<sub>2</sub>O emission for the CC period. Even during the time of the highest N uptake of CC (pre-winter), cumulative N<sub>2</sub>O emissions were no lower in CC treatments than for bare fallow. Several factors have probably contributed to this result, such as the differences in mean SMN contents in the topsoil between CC treatments and bare fallow being much smaller (<10 kg N ha<sup>-1</sup>) than





**Fig. 3.** Contrasts between CC and bare fallow (control) with 95% confidence intervals ( $n = 16$ ) for the average WFPS and SMN content, both in 0–30 cm, and cumulative N<sub>2</sub>O emissions for the various periods investigated (the total CC period, pre-winter (seeding of CC to mid-December), mid-winter (mid-December to incorporation), after incorporation (incorporation of CC until seeding of maize), and for the maize period). The contrasts of SMN and WFPS are calculated as the difference between estimated marginal means of CC and control while contrasts of N<sub>2</sub>O emissions are calculated as the ratio of estimated marginal means of CC and control.

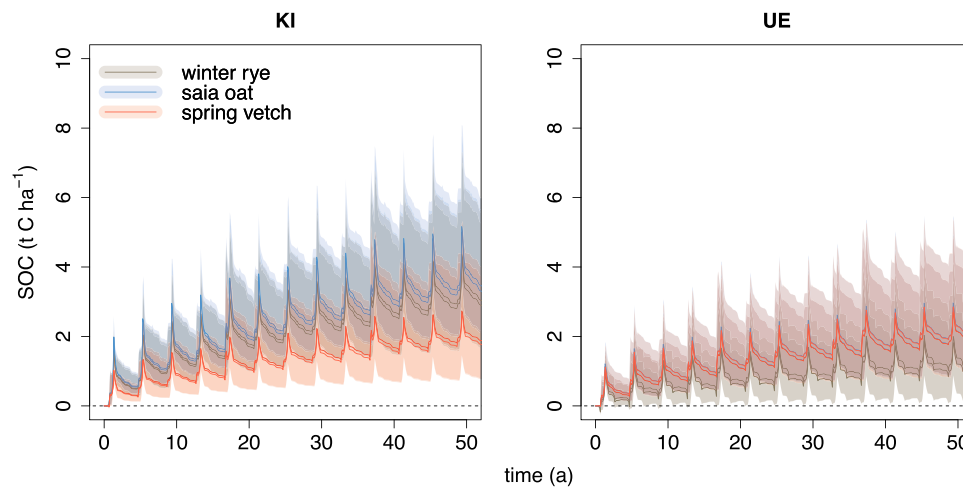


**Fig. 4.** Contrast between CC and bare fallow (control) with 95% confidence intervals ( $n = 16$ ) for the annual N<sub>2</sub>O emission (seeding of CC to harvest of maize) and yield-related N<sub>2</sub>O emission across all sites combined. Contrasts are calculated as the ratio of estimated marginal means of CC and control.

expected from the CC N uptake (50 kg N ha<sup>-1</sup> on average). One of the main reasons for these small SMN differences may have been NO<sub>3</sub> leaching to deeper soil layers in the fallow treatment (Fig. A.6). In addition, SMN did not appear to be a dominant factor limiting direct N<sub>2</sub>O emission during the CC period in the present study. Increased N<sub>2</sub>O emission rates after the killing of CC by frost (non-persistent species) or management (chemical termination and incorporation) were among the main reasons for CC not reducing overall winter N<sub>2</sub>O emission. This

increase can be explained by changes in microbial C and N availability and soil aeration due to the release of considerable amounts of easily available C and N during the decomposition of CC residues (Thorup-Kristensen, 1994; Clark et al., 1997; Davidson et al., 2000; Rosecrance et al., 2000; Alonso-Ayuso et al., 2014). This was corroborated by correlations of N<sub>2</sub>O emissions in the CC period with the DM yield of CC biomass and of emissions during the mid-winter period (i.e., the period when CC mineralisation after winter kill may occur) with the maximum N amount in CC biomass (Table A.3). The results of the present study are in agreement with the findings of Basche et al. (2014) who observed the highest response ratios of N<sub>2</sub>O emissions in systems with CC during periods of CC decomposition. Our results of higher SMN contents and increased N<sub>2</sub>O emissions under saia oat (non-persistent) in mid-winter compared to winter rye (persistent CC) indicate that winter hardness of CC affects dynamics of SMN and N<sub>2</sub>O emission because mineralisation starts earlier when CC are killed by frost. However, we found no evidence for a significant effect on cumulated N<sub>2</sub>O emission over the total CC period.

At field-scale, the effects were often less clear than when evaluated across all the fields combined, and they generally showed a high variability. This site-specific nature of the N<sub>2</sub>O response is probably due to the variety of factors driving N<sub>2</sub>O emissions (e.g., the presence of frost periods, management of CC, soil texture and WFPS, differences in SMN) that underlie complex interactions (Davidson et al., 2000; Rosecrance et al., 2000; Basche et al., 2014) and reinforce the importance of a larger dataset covering a wider range for each of these factors.



**Fig. 5.** Simulated effects of different CC on the increase in SOC stocks (0–30 cm) in the crop rotation “CC (or fallow) – maize – winter wheat – winter oilseed rape – winter wheat” at the KI and UE experimental sites (2 fields per site). The shaded areas indicate the uncertainty range of the model ensemble. Soil organic C dynamics of the treatments spring vetch and saia oat at UE were nearly identical; the saia oat line is not visible because it is masked by the spring vetch treatment.

#### 4.2. Effects of cover crops on soil mineral N dynamics and direct N<sub>2</sub>O emissions from the subsequent maize crop

This study’s hypothesis that emissions of N<sub>2</sub>O in the subsequent main crop maize are still boosted by the decomposition and mineralisation of incorporated CC residues was underlined by higher cumulative N<sub>2</sub>O emissions during the maize period in all CC treatments compared with bare fallow (significant only for non-legume CC). This is in agreement with the findings of [Basche et al. \(2014\)](#) who also report higher emissions after CC compared with bare fallow in the subsequent main crop due to ongoing mineralisation of CC biomass. However, an effect of winter hardiness of CC on N<sub>2</sub>O emission during the maize period or a correlation between CC biomass (DM amount) or CC N yield and N<sub>2</sub>O emissions in the main crop period was not found either in the present study or the study of [Basche et al. \(2014\)](#).

The present study’s result that N<sub>2</sub>O emissions in treatments with non-leguminous CC were higher than with bare fallow, but treatments with legume CC were not contradicts the more general finding in the literature that legume CC show a higher response ratio of N<sub>2</sub>O emissions than non-legume CC ([Rosecrance et al., 2000](#); [Basche et al., 2014](#); [Abdalla et al., 2019](#)). Increased emissions induced by legume CC are attributed to the additional N input from N<sub>2</sub> fixation and to an increased nitrification-denitrification activity that is favoured by a relatively narrow C/N ratio of legume residues (e.g., [Rosecrance et al., 2000](#); [Kaspar and Singer, 2011](#); [Basche et al., 2014](#)). Generally, legume CC are mineralised more quickly than non-legumes ([Waggoner, 1989](#); [Waggoner et al., 1998](#); [Rosecrance et al., 2000](#)). Based on the same experiment outlined in this study, [Kühling et al. \(2023\)](#) observed decreasing net N mineralisation in the order spring vetch > saia oat ≥ winter rye during the whole maize vegetation period. This finding is also in line with higher SMN contents in 0–30 cm under spring vetch compared with the other CC at the beginning of the maize vegetation period. It is well known that the time and extent of N mineralisation and immobilisation in the subsequent main crop are influenced by the C/N ratio and the lignin content of the CC biomass as well as by the time when it is killed by frost and incorporated into the soil ([Thorup-Kristensen, 1994](#); [Thorup-Kristensen and Dresbøll, 2010](#); [Kaspar and Singer, 2011](#)). Overall, the missing effect of spring vetch on N<sub>2</sub>O emission in the present study was probably due to several factors: i) N<sub>2</sub> fixation was restricted by poor CC development of vetch in autumn, which additionally led to high amounts of vOSR (a non-legume) in the spring vetch plots in all investigated fields, ii) a large part of the mineralisation of spring vetch C and N may have already occurred in the initial days after frost and CC incorporation (which was still counted within the CC period), and iii)

the mineralisation of spring vetch was in better synchronisation with the N demand of the succeeding crop maize.

#### 4.3. Effect of cover crops on annual and maize yield-related N<sub>2</sub>O emission

The hypothesis that winter CC before maize increase annual N<sub>2</sub>O emissions compared with bare fallow was confirmed although the increase was significant only for the two non-legume CC. Our results are in line with findings from [Blanco-Canqui et al. \(2015\)](#) and [Mitchell et al. \(2013\)](#) who explained the increased emissions in CC treatments by the stimulation of N<sub>2</sub>O production by increased C availability for denitrifiers. Especially in fertilised cropping systems, such as those in the present study, C availability through CC residues may even become a more important factor controlling N<sub>2</sub>O emissions than SMN supply ([Petersen et al., 2011](#); [Mitchell et al., 2013](#)). In this case, the amount of C and N incorporated might have been of greater importance for the N<sub>2</sub>O response than CC composition or C/N ratio.

As discussed in chapter 4.2, these results are not in agreement with the conclusions of several studies suggesting that, when considering annual N<sub>2</sub>O emissions, the response ratio of legume CC is generally higher than of non-legume CC ([Basche et al., 2014](#); [Lugato et al., 2018](#); [Abdalla et al., 2019](#)). Spring vetch appeared to be a risky choice of CC at these sites because growth and biomass yield were very variable. Mixing with other species might be advisable to reduce the risk of poor CC development. In general, giving consideration to the different effects of CC on the overall N balance and accounting for their effects on N immobilisation and mobilisation during the subsequent main crops are of vital importance for adequate fertilisation, crop yield and N<sub>2</sub>O mitigation.

The maize yields did not differ between CC treatments in any of the investigated fields ([Kühling et al., 2023](#)) and consequently yield-related N<sub>2</sub>O emissions reflected the trends observed for annual N<sub>2</sub>O emissions, i. e., higher yield-related emissions in non-legume CC treatments compared with bare fallow).

#### 4.4. Potential effects of cover crops on indirect N<sub>2</sub>O emissions

[Petersen et al. \(2011\)](#) have already stressed the importance of CC N uptake to indirect N<sub>2</sub>O emissions and concluded that increased N<sub>2</sub>O fluxes in systems with CC can potentially be offset by a reduction in NO<sub>3</sub> leaching and indirect losses of N<sub>2</sub>O in streams, lakes and drainage systems. Overall, leaching in agricultural systems is highly variable. [Kaye and Quemada \(2017\)](#) report leaching rates ranging from 0 to 150 kg N ha<sup>-1</sup> yr<sup>-1</sup>. The extent of N leaching depends on the amount of winter

rainfall, i.e., the amount of water percolating through the soil (Webb et al., 2000), as well as on the N concentration in the leachate (Sieling and Kage, 2010; Eysholdt et al., 2022). In particular the N concentration in the leachate can be greatly reduced by N uptake of winter CC (Kristensen and Thorup-Kristensen, 2004; Schipanski et al., 2014). The extent of this reduction varies greatly depending on CC species, CC growth, and the amounts of N and water in the soil. Reductions between 6% and 94% have been reported with legume CC generally being less effective compared to non-legume CC in depleting SMN (Thorup-Kristensen et al., 2003; Kaspar and Singer, 2011). Reduction of N leaching after implementation of non-legume CC by about 50–75% were reported in several studies (Tonitto et al., 2006; Dabney et al., 2011; Quemada et al., 2013). Considering the range of leaching of 0–150 kg N ha<sup>-1</sup> mentioned above (Kaye and Quemada, 2017), this results in a reduction of N leaching of 75–113 kg N ha<sup>-1</sup> yr<sup>-1</sup>. For agricultural systems with optimal N management, Kaye and Quemada (2017) suggest a value of 25 kg N leached ha<sup>-1</sup> yr<sup>-1</sup> and a reduction by CC of 12.5 kg N ha<sup>-1</sup> yr<sup>-1</sup>. The amount estimated from the maximum N uptake for the CC in this study lies in between the suggested values from these studies. A potential reduction of indirect N<sub>2</sub>O emission for the spring vetch treatment was not calculated, because such an estimation would be rather speculative due to a lack of knowledge about the contribution of biological N<sub>2</sub> fixation to total N uptake, which can be highly variable. In addition, we did not analyse if transformation of the additional N input by N<sub>2</sub> fixation resulted in increased indirect N<sub>2</sub>O emission.

This study's approach of using N uptake by winter CC as an indicator to estimate the potential amount of N protected from leaching is a simplistic method that does not take into account the water movement within the soil or N concentration in the leachate. Nevertheless, it is an estimate of the maximum amount of N that can potentially be protected from leaching by SMN uptake of a growing CC. Further, the maximum amount of N in the aboveground CC biomass was used. Additional N stored in roots or remineralisation of CC N in winter was not taken into account. Generally, N in crop roots corresponds to about 10–15% of plant N and up to 40% in legumes (Rochester et al., 1998; Kumar and Goh, 2000). For CC, Redin et al. (2018) report an average storage in roots of 9% of the total CC N.

#### 4.5. Soil organic C stocks

The model results in this study indicate that the different SOC accumulation rates induced by CC at KI and UE were mainly a result of different CC yields and related C inputs (much lower CC yields at UE). This is in line with the conclusion of McClelland et al. (2021) that the amount and management of CC residues are crucial for soil C accumulation. Early seeding and adequate water and nutrient availability are decisive factors for winter CC growth, and these factors can induce a high variability in CC performance between sites and years (Koch et al., 2017). In the present study, CC were sown relatively late (end of August) and they were not fertilised in order to promote uptake of residual SMN and reach low SMN levels during winter. Both factors probably contributed to relatively low CC biomasses at these sites, particularly at the UE site where the sand content was up to 69%. The relative performance of different CC for soil C accumulation was site-dependent. No effect of CC on the yield of maize was identified. However, CC effects on yields of the following main crops can be important for SOC accumulation, particularly in systems with low fertilisation rates and N-fixing legume CC (Fageria et al., 2005). According to the applied models, the lower clay content at UE than at KI favoured rapid mineralisation of CC biomass and contributed to the lower SOC accumulation at UE.

In a meta-analysis of 37 studies worldwide, Poeplau and Don (2015) estimate that winter CC can sequester about 320 ± 80 kg C ha<sup>-1</sup> yr<sup>-1</sup> in the topsoil (0–22 cm) over a period of 54 years when established annually. These rates are in line with those from a range of other studies on potential SOC accumulation due to the cultivation of CC (Dendoncker et al., 2004; Blanco-Canqui et al., 2013; Lugato et al., 2014; Sommer and

Bossio, 2014; Abdalla et al., 2019; Bolinder et al., 2020; McClelland et al., 2021). In cropping systems where winter crops dominate (e.g., at the sites in this study), this potential SOC sequestration is often reduced by 66–75%, depending on the frequency of summer crops such as maize, sugar beet, potato and summer grain (often every third to fourth year). The sequestration potential published by Poeplau and Don (2015) was divided by four to compare it with this study's model results (winter CC every fourth year rather than annually). The resulting SOC sequestration of 80 kg C ha<sup>-1</sup> yr<sup>-1</sup> matches the model results at KI for treatments with relatively high CC yields.

The simulated SOC accumulation rates are equal to a mean annual CO<sub>2</sub> sink of 220 kg yr<sup>-1</sup> at KI and 150 kg yr<sup>-1</sup> at UE calculated for a period of 50 years. However, soil C sequestration is a fairly unreliable measure of climate mitigation and its value to compensate for emission of long-life GHG is limited for several reasons: i) SOC accumulation is reversible and the permanence of the related climate mitigation effect is unsure, ii) the rate of SOC accumulation decreases over time until the SOC stocks approach a new equilibrium, thus increased SOC stocks require continuous maintenance with a decreasing, or even without an additional, climate mitigation effect, and iii) increased SOC stocks can induce increased emission rates of N<sub>2</sub>O that at least partly offset climate benefits from SOC sequestration (Lugato et al., 2018; Jian et al., 2020; Guenet et al., 2021).

#### 4.6. Climate mitigation potential of winter cover crops

The introduction of winter CC to crop rotations may contribute to climate mitigation mainly due to the following effects: it may i) change direct and indirect emissions of N<sub>2</sub>O, ii) result in binding CO<sub>2</sub>-C by increasing SOC stocks, and iii) reduce emissions related to the demand for N fertiliser by improving N cycling within rotations and N transfer to the main crops that follow. Another contribution to the GHG balance comes from GHG emissions related to agricultural management of CC, i.e., seeding of CC including seed production, termination and incorporation of CC, which we do not address in detail. Kaye and Quemada (2017) calculate a range of 10–100 kg CO<sub>2-eq</sub> ha<sup>-1</sup> yr<sup>-1</sup>, with 28 kg CO<sub>2-eq</sub> ha<sup>-1</sup> yr<sup>-1</sup> as a typical value for fuel required for farm operations due to extra field passes under CC, with the amount depending on the methods of planting and killing, which points to the minor importance of extra fuel use in farm operations.

The net change in N<sub>2</sub>O emissions (direct minus indirect emissions) across all fields was +0.31 ± 1.10 kg N ha<sup>-1</sup> yr<sup>-1</sup> or +136 ± 474 kg CO<sub>2-eq</sub> ha<sup>-1</sup> yr<sup>-1</sup>. Further, a potential sequestration of 50 ± 20 kg C yr<sup>-1</sup> or 183 ± 73 kg CO<sub>2-eq</sub> ha<sup>-1</sup> yr<sup>-1</sup> over a period of 50 years was modelled. However, the large standard deviations resulting from differences between CC species, sites and years show that the net effect of CC on the GHG balance of cropping systems is highly variable and depends on a range of factors, such as C and N availability, the water status of the soil and management. In addition, fertiliser savings should be examined for a more comprehensive view of climate mitigation effects, because CC may reduce GHG by decreasing the fertiliser N demand due to a CC-mediated N transfer to the subsequent main crop (Wittwer and van der Heijden, 2020; Kühling et al., 2023). The CC effect on the N fertiliser demand of maize could not be determined in this study, because fertilisation was similar in all treatments. However, Kühling et al. (2023) investigated the fertilised and unfertilised plots of the very same experimental fields under study here in terms of soil-plant N dynamics. They found a significant increase in effective N mineralisation during maize growth following CC compared with bare fallow by 43 kg N ha<sup>-1</sup> and by 51 kg N ha<sup>-1</sup> in the fertilised and unfertilised plots, respectively, which shows that the transfer of CC N to the following main crop depends on fertilization intensity. Using the value for the fertilised plots in combination with the reported mean GHG emission of synthetic N fertiliser production recently updated by Menegat et al. (2022); 3.4 kg CO<sub>2-eq</sub> kg<sup>-1</sup> N for the EU-28 and 4.1 kg CO<sub>2-eq</sub> kg<sup>-1</sup> N worldwide, respectively), the resulting climate mitigation potential is 0–146 (EU-28) and 0–176

(worldwide) kg CO<sub>2-eq</sub> ha<sup>-1</sup> yr<sup>-1</sup>. Reliable estimations of the N supply via CC and the net-N release after CC termination and/or incorporation in relation to management and site conditions are essential in order to adapt the fertilisation rate and achieve the related GHG savings.

## 5. Conclusions

Referring to our initial hypotheses, we found that (i) non-legume CC reduce winter SMN contents, but increase direct N<sub>2</sub>O emissions upon the mineralisation of CC residues after killing by frost and/or CC incorporation. Direct emissions of N<sub>2</sub>O (ii) were boosted by the mineralisation of CC residues in the subsequent crop leading to (iii) overall higher annual and yield-related N<sub>2</sub>O emissions for all CC treatments (although significant only for non-legumes). The increase of direct N<sub>2</sub>O emissions (v) was only partly compensated by the calculated potential for mitigation of indirect N<sub>2</sub>O emissions, but additional benefits were shown in the form of SOC accumulation over a certain period and potential N-fertiliser savings.

Overall, the results of this study indicate that growing winter CC had both positive and negative effects on GHG emission and climate mitigation. There was no generalisable positive or negative effect of CC on the net exchange of CO<sub>2-eq</sub> when direct and indirect N<sub>2</sub>O emission, SOC sequestration and emissions from fertiliser demand were considered together. Net effects can be highly variable depending on site conditions, soils, CC properties and management. Contributions to climate protection come from reduced NO<sub>3</sub> leaching, fertiliser savings and soil organic matter accumulation, while direct N<sub>2</sub>O emissions increased in most cases. Overall, the results suggest that cultivation of CC is not an effective measure *per se* for mitigating GHG emissions in fertilised arable cropping systems. However, it does offer many highly valuable benefits in the areas of soil and groundwater protection, soil organic matter accumulation, soil fertility and biodiversity. Thus, the relevant question is no longer whether the cultivation of CC contributes to climate mitigation, but rather how their mitigating effects can be optimised at different sites and in different cropping and fertilisation systems. The site and management-dependent risk of NO<sub>3</sub> leaching, the general N availability in the cropping system, and the matching of CC and fertiliser N inputs to the needs of the subsequent crops in terms of their quantity and timing are essential factors that must be taken into account if climate mitigation effects are to be optimised.

## CRedit authorship contribution statement

**Thomas Rübiger:** Writing – review & editing, Data curation. **Insa Kühling:** Writing – review & editing, Data curation. **Heinz Flessa:** Writing – review & editing, Supervision, Project administration, Methodology, Funding acquisition, Conceptualization. **Michaela Schlathöler:** Data curation. **Henning Kage:** Writing – review & editing, Conceptualization. **René Dechow:** Writing – review & editing, Writing – original draft, Visualization, Methodology. **Simone Merl:** Writing – review & editing, Methodology, Formal analysis, Data curation. **Mirjam Helfrich:** Writing – review & editing, Writing – original draft, Visualization, Methodology, Data curation. **Roland Fuß:** Writing – review & editing, Validation, Supervision, Funding acquisition, 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.agee.2024.108985](https://doi.org/10.1016/j.agee.2024.108985).

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