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Interactive effects of straw management, tillage, and a cover crop on nitrous oxide emissions and nitrate leaching from a sandy loam soil

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Abstract

Minimum tillage, residue recycling and the use of cover crops are key elements of conservation agriculture that play important roles in soil carbon (C) and nitrogen (N) dynamics. This study determined the long-term effects of tillage practice (conventional ploughing *vs.* direct seeding), straw management (retained *vs.* removed), and the presence of a cover crop (CC; fodder radish in this study) on nitrous oxide (N_2O) emissions, nitrate (NO_3^-) leaching, and soil mineral N dynamics between October 2019 and June 2020. In the factorial experiment with eight treatment combinations, cumulative N_2O emissions ranged from 0.04 to 0.8 kg N ha^{-1}, whereas NO_3^- leaching varied between 4 and 28 kg N ha^{-1}. The study did not find effects of straw retention on NO_3^- leaching or N_2O emissions. No-till reduced N_2O emissions by on average 46% compared to ploughing. Fodder radish reduced NO_3^- leaching by 80-84%, and there was little N_2O emission in the presence of the cover crop; however, after termination in spring there was a flush of N_2O,cumulative N_2O-N averaged 0.1 and 0.5 kg N ha^{-1} without and with a cover crop. With information about long-term soil C retention from straw and fodder radish, an overall greenhouse (GHG) balance was calculated for each system. Without straw retention after harvest there was always a positive net GHG emission, and the indirect N_2O emission from NO_3^- leaching was similar to, or greater than direct N_2O emissions. However, in the presence of fodder radish, the direct N_2O emissions after termination were much more important than indirect emissions, and negated the C input from fodder radish. Direct seeding, straw retention and the use of a cover crop showed positive effects on N retention and/or GHG balance and could substantially improve the carbon footprint of agroecosystems on sandy soil in a wet temperate climate.

Keywords: Conservation agriculture, nitrous oxide emissions, nitrate leaching, straw retention, cover crops.

Introduction

Within agriculture, sustainability will depend on land conservation and improved input management as identified in the United Nations Programme of Action (UN, 2020). Specifically, *conservation agriculture* is based on principles that include minimum tillage, residue recycling, use of cover crops, and diversification of crop rotations (FAO, 2021). The agronomic performance relative to conventional practices, however, is site-specific (Giller *et al.,* 2015), which indicates that environmental impacts are also variable (Sun *et al*., 2020). It is therefore necessary to determine at national or regional level to what extent the practices of conservation agriculture ensure nutrient recycling, avoid environmental losses, and reduce greenhouse gas emissions.

In agricultural systems, the nitrogen (N) cycle is "leaky", and efficient use of N has proven challenging; for instance, less than one-half of fertilizer N is recovered in the aboveground portion of cereal crops (Ladha *et al.*, 2005), suggesting that the fraction of applied N remaining in the soil is at risk of being lost to non-target ecosystems (Davidson, 2012). There are several undesirable mechanisms by which N may be lost, including nitrate (NO_3^-) leaching and gaseous emissions resulting from processes such as nitrification and denitrification (Butterbach-Bahl *et al.*, 2013). These emissions include nitrous oxide (N_2O), a powerful greenhouse gas with a contribution of 66% from the agricultural sector, mostly as direct emissions from soils (Davidson and Kanter, 2014). The profound effects of N_2O in the atmosphere (i.e. its high global warming potential and contribution to stratospheric ozone depletion), and of NO_3^- leaching on surface- or groundwater quality, are well documented, but mitigating N losses remains difficult (IPCC, 2019; Bakken and Frostegard, 2017).

Tillage can affect soil N availability through its influence on soil physical, chemical, and biological properties. Conventional inversion tillage favors the decomposition of crop residues and soil organic matter by enhancing aeration and soil-residue contact, and promoting soil microbial activity (Singh *et al.*, 2021), whereas conservation tillage practices (i.e., direct seeding) result in less soil disturbance, which slows down decomposition of crop residues but also reduces fuel costs and disturbance of soil organisms, and increases the volume of medium pores and soil water holding capacity (FAO, 2014; Munkholm *et al.*, 2020). More NO_3^--N has been reported to accumulate in soil under conventional tillage than in reduced or no-tillage soils, although reasons for the greater accumulation are not well understood (Carefoot and Janzen, 1997). Conservation tillage practices seem to reduce the activity of ammonia oxidizing bacteria (Wang *et al*. 2019), and the lower soil NO_3^--N content reported in some no-tillage systems has also been linked to leaching below the root zone, or greater soil N immobilization (Singh *et al.*, 2021). However, long-term studies in Denmark have not been able to confirm greater NO_3^- leaching from conventional compared with conservation tillage (Hansen *et al*., 2015).

Mutegi *et al*. (2010) found that straw recycling resulted in higher N_2O emissions under conventional tillage compared to reduced tillage six years after transition. Retention of crop residues at the soil surface will affect soil temperature, and the thermal insulation provided by surface residues tends to delay and dampen temperature variations in soil under conservation tillage when compared to soil under conventional tillage (Grevers *et al.*, 1986). Changing temperature regimes can, together with effects of conservation agriculture on organic matter and oxygen (O_2) availability and soil gas diffusivity, affect the potential for denitrification (Skiba and Ball, 2002).

In agroecosystems, the recycling of residues, including straw as a by-product of crop production, can improve soil structure, provide plant nutrients, and act as a primary energy source for soil microorganisms (FAO, 2014). Retention of straw along with appropriate fertilization may increase soil fertility and increase crop yields, and reduce N losses outside the growing season. Powlson *et al.* (1985) incorporated straw with 0.5% N into a soil that was subsequently sown with winter wheat, and they found that 78% of the N added in straw remained in the soil a year after incorporation, thereby increasing the quantity of mineralizable N. Nitrogen mineralization in synchrony with crop uptake (in spring and summer) can reduce crop fertilizer N requirements, whereas any N mineralization during autumn or winter increases the risk of NO_3^- leaching.

Establishing a cover crop, or catch crop, outside the main growing season, has frequently been shown to reduce NO_3^- leaching. Thapa *et al*. (2018) found in a global meta-analysis an average reduction of 56% by non-leguminous cover crops, and a higher efficiency for coarse-textured soil. By reducing soil mineral N availability outside the main growing season, cover crops may also reduce the potential for N_2O emissions. A meta-analysis by Basche *et al.* (2014) found mixed results for the effect of cover crops on N_2O emissions during the growth period, but a stimulation of N_2O emissions after incorporation of the cover crop in spring. Further, cover crops provide an input of carbon to the soil, which may help maintain soil C stocks (McClelland *et al.*, 2021).

A global meta-analysis found a higher risk for N_2O emissions under conservation tillage practices (Mei *et al.*, 2018), although there are results indicating a reversal of this effect after long-term continuous conservation tillage, possibly due to increasing total porosity created by roots and soil fauna (Six *et al.*, 2004; Finney *et al.*, 2015). Soil under conservation tillage has also been found to have a higher relative abundance of nosZ encoding the enzyme N_2O reductase, and more complete denitrification to N_2 would reduce N_2O emissions (Wang and Zou, 2020; Wang *et al.*, 2019). The effect of reduced tillage, however, may also depend on soil type and climate (Rochette, 2008; Mei *et al.*, 2018). These complex interactions between the different elements of conservation agriculture, i.e., residue recycling, use of cover crops, and minimum tillage, are key to understanding the environmental impacts of conservation agriculture practices.

The objectives of this study were to explore the interactions between tillage practice, a cover crop, and straw management with respect to: (1) dynamics of soil mineral N, (2) N_2O emissions, and (3) NO_3^- leaching. The study included the two main phases where a cover crop may influence nitrogen losses,i.e., the growth period (autumn-early spring) and the period where cover crop residues decompose in the soil (spring-early summer). The hypotheses were that: (i) minimum tillage would reduce N_2O emissions compared to ploughing; (ii) retention of straw in the field would reduce NO_3^- leaching compared to straw removal, but would increase N_2O emissions during autumn; and (iii) the presence of cover crop in the field would reduce NO_3^- leaching, but increase N_2O emissions after spring cultivation.

Materials and methods

Study site

Measurements were conducted between October 2019 and July 2020 within a long-term field experiment at Foulum (56°30´N, 9°35´E, elevation 56 m a.s.l.), Aarhus University, Denmark. The selected experimental plots were part of a larger field experiment, which was established in 2002 under temperate coastal climatic conditions to evaluate the effects of tillage, straw retention and cropping sequence, including cover crops, on crop yield and C and N cycling, and retention. The soil is a loamy sand based on ground morainic deposits from the last glaciation classified as a Mollic Luvisol according to the WRB (FAO) system (Krogh and Greve, 1999). The soil contains 8.1-8.8% clay, 3.5-3.8% organic matter, with pH 6.1 (1:2.5 suspension of soil: 0.01 M $CaCl_2$). Before the experiment was established, the field had been cropped and cultivated according to normal Danish agricultural practices (Hansen *et al*., 2015).

The field experiment had a split-plot design with different crop rotations in main plots and tillage treatments in subplots (Munkholm *et al*., 2013). This study included two cereal-based crop rotations which differed only with respect to straw management (removed or retained). Two tillage treatments were represented, i.e. conventional ploughing (PL) and direct seeding (DS), and tillage was applied to 72 m long and 6 m wide subplots, each of which had ten 3 x 10 m^2 sub-subplots. Fodder radish (*Raphanus sativus* L.) was grown as a cover crop from September 2019 to April 2020. Fodder radish followed oat (*Avena sativa* L.) as the main crop in 2019, and was followed in spring 2020 by faba beans (*Vicia faba* L.). One 3 × 10 m^2 plot with the cover crop was selected, as well as one plot kept free of vegetation with a herbicide cf. Table 1 (bare soil); bare-soil plots had been grown with cover crops before 2017. Thus, measurements in this study were restricted to net plot areas of 10 × 3 m^2. The PL treatments were mouldboard ploughed to 20 cm depth followed by levelling to make a good seedbed before sowing. Sowing in the CT treatment was performed with a traditional seed drill (Nordsten Lift-o-matic CLH300), whereas a chisel coulter drill (Horsch Airseeder CO3) was used in the DS treatments. In all treatments, crops were sown at the same row distance of 17.5 cm. In the crop rotation with straw retention, straw was chopped and retained after harvest, whereas straw was removed in the crop rotation without straw retention. Cover crop seeds were scattered from a seeding machine after harvest of the main crop (Table 1).

Table 2 shows the treatments and variables that were monitored in the experiment. With two tillage systems (PL and DS), each with or without straw retention (+S and –S), and with or without cover crop (+CC and –CC) in three replicate blocks, there were a total of 24 sampling positions. Precipitation and average temperatures for the experimental period were obtained from the meteorological station at Foulum and are shown in Fig. 1. Soil temperature was also monitored at 8 cm below the soil surface using Temperature-Moisture-Sensor (TMS) data loggers (Wild *et al.*, 2019).

Nitrate leaching

Nitrate leaching was determined from analyses of soil water samples collected *via* porous ceramic cups (two in each plot) installed in autumn 2002 at a depth of 1 m. The sampling system consists of suction cups (655x01-B1M1, 1 bar, standard, Soil moisture Equipment Corporation, Goleta, CA) mounted on PVC pipes (Hansen *et al.*, 2015). Suction and collection tubes are protected by rigid plastic tubes, which extend outside the experimental field to cupboards with three-way valves for sample collection. A suction of approximately 70–80 kPa was imposed 2–3 days before sampling of pore water. The soil water samples from each plot were bulked and frozen within a few hours, and later analyzed for NO_3^--N (Best, 1975). Sampling took place biweekly except in periods with frost or drought.

The daily drainage in each plot was calculated with the EVACROP model (Olesen and Heidmann, 1990; Hansen *et al.*, 2010), which applies a cascading model for calculating percolation at a depth of 1 m based on a simple conceptual water-balance model with a number of simple coupled linear reservoirs for snow, interception, evaporation, and transpiration. Soil-specific input parameters include the permanent available water content for each soil layer, and drainage constants of the root zone reservoir and the subsoil reservoir, which describe the fraction of the water that drains to the next deeper layer each day if the soil moisture in the layer is above field capacity. Data on daily precipitation, air temperature, and reference evapotranspiration from a meteorological station located close to the experimental field, were obtained to run the model. Precipitation was measured at 1.5 m height, and corrected to the soil surface (Allerup *et al.*, 2000).

Nitrate concentrations in the period between two observations were estimated by percolation-weighted interpolation, or 'drainage-linear interpolation' (Vogeler *et al.*, 2019), which provides the relative change in concentration based on daily percolation assuming that NO_3^- concentrations in the extracted soil water represented average flux concentrations (Askegaard *et al.*, 2005). Daily NO_3^- leaching (kg N ha^{-1}) was calculated for each treatment plot by multiplying daily percolation (mm) with flow-weighted interpolated daily NO_3^--N concentration (mg L^{-1}) and dividing by 100. Afterwards the cumulative NO_3^--N leaching was calculated for the whole period from 30 August 2019 to 2 April 2020.

N_2O and CO_2 measurements

Monitoring of N_2O and CO_2 took place between 15 October 2019 and 25 June 2020 with a total of 21 sampling days over the 36-week period. Fluxes were determined using 75 cm × 75 cm × 20 cm static chambers as previously described (Baral *et al.*, 2019); collars were inserted one day before the start of flux measurements. Chamber headspace air was continuously mixed by a battery-powered fan during chamber deployment. Gas samples were taken with a 10-mL syringe and hypodermic needle via a septum after first pumping air in and out of the syringe three times to flush the syringe. Five 10 mL gas samples were collected for flux measurements, the first sample immediately after chamber deployment and four other samples at 15–20 min intervals. Sampling was always initiated between 10:00 and 13:00. Gas samples were stored in pre-evacuated 6 mL exetainers (Labco, High Wycombe, UK) until analysis for N_2O and CO_2 using an Agilent 7890 (Agilent, Nærum, Denmark) gas chromatograph configured as previously described (Petersen *et al.*, 2012). The concentration-time series of CO_2 were mainly used for quality control purposes. Nitrous oxide fluxes were calculated by the free software package HMR avaiable as a package in R (Pedersen *et al.*, 2010).

Soil measurements

Soil samples were collected in the same plots where N_2O and CO_2 were monitored, and on the same days as gas sampling. Six individual samples were randomly collected from each treatment and block using an auger (2 cm dia., 0–20 cm depth); the subsamples were pooled to make a representative composite sample per plot, transferred to zip-lock plastic bags and kept at 2 °C until analyzed. Soil samples were sieved (<2 mm) and subsamples extracted for mineral N (ammonium (NH_4^+-N) and nitrate (NO_3^--N) analysis within two days of soil sampling. Approximately 10 g of soil was extracted in 40 mL of 1 M KCl. Additionally, two KCl blanks were prepared per batch to correct for any bias from KCl impurities. For extraction, both the soil-KCl solutions and blank samples were shaken for 30 min on an orbital shaker set at 150 rpm and then allowed to settle. The supernatant was then filtered through 1.6 µm glass microfiber filters (VWR, Sweden) and frozen at -20 °C for later analysis. Inorganic-N analysis was done with standard colorimetric methods on a Foss FIAstar 5000 flow injection analyzer (FOSS Denmark). Gravimetric soil water content was determined by drying approximately 10 g of soil for 24 h at 105°C. Using information about soil bulk density in the tillage experiment determined in November 2019 (Goméz-Mũnoz *et al*., 2021), gravimetric water content was used to calculate water-filled pore space (WFPS). For 0-20 cm depth, bulk density of PL-S, DS-S, PL+S and DS+S were 1.39, 1.39, 1.29 and 1.31 Mg m^{-3}, respectively.

Data analyses

Statistical analyses of the data were performed using R programming language version 4.0.0 (Noguchi *et al.*, 2012; R, 2018). All data, except cumulative N_2O emissions and NO_3^--N leaching, were split into pre-tillage (15 October 2019 – 2 April 2020) and post-tillage (8 April – 25 June 2020) seasons for analysis. Main and interacting effects of tillage, straw retention and cover crop on the daily and cumulative N_2O emissions and mineral N were analyzed with the linear mixed effect (*lme)* function of the *nlme* package using the restricted maximum likelihood (REML) method. Blocks were treated as a random factor in the analyses. Model assumptions, i.e., normality and homogeneity of variance, were assessed using diagnostic plots of residuals. In order to satisfy model assumptions, the daily and cumulative N_2O emissions and mineral N data were log-transformed. For time series of N_2O emissions and mineral N data, auto-correlation between sampling positions were accounted with the *corAR1* function. Pairwise comparisons between treatments were performed using the estimated marginal mean *(emmeans)* function. The *p*-values were adjusted by Tukey's HSD method, and the hypothesis rejection threshold was 0.05.

Effects of soil and environmental variables, such as mineral N (NH_4^+-N and NO_3^--N), soil temperature and moisture, to pre-tillage and post-tillage season N_2O emissions were analyzed using multiple regression analysis. Prior to the analysis, N_2O emission data were log-transferred and tested for collinearity between variables using a variance inflation factor (VIF) of 5 as threshold. During analysis, only two-way interactions of variables with respect to N_2O emissions were analyzed to make the results interpretable. The variable interactions that were not significant ($p > 0.05$) were removed in the final model.

Since daily NO_3^- leaching data violated model assumptions, even after transformation, the data were analysed using the non-parametric rank-based model *nparLD*, which is a robust method to analyse repeatedly measured data with non-normal distribution (Noguchi *et al*., 2012). We used F2-LD-F1 design during analysis, where F2 represents the two levels of independent factors, and F1 stands for repeatedly measured leaching data. Since this model can only handle interactions between two independent factors at a time, we made three different interaction models during the analysis, i.e., tillage × residue, residue × cover crop, and tillage × cover crop. Treatment effects within factors were evaluated using ANOVA-type statistics. As in parametric tests, the hypothesis rejection threshold was set at 0.05.

The overall carbon footprint of the eight treatment combinations was calculated from the direct emissions of N_2O and indirect N_2O emissions from NO_3^- leached, and C from straw and fodder radish stabilized in the soil. The indirect N_2O emission was estimated using an emission factor of 0.0075, as in the national inventory for Denmark (Nielsen *et al*., 2020), and a global warming potential (GWP) of 265 for direct and indirect emissions (IPCC, 2014). The annual net change of straw C was calculated from the change in soil C concentrations of PL and DS soil with and without straw retention between 2002 and 2019 (17 years), and bulk density determined in November 2019 (Gómez-Muñoz *et al*., 2021). The retention of cover crop C was based on dry matter in plant cuts collected in January 2020; the total above- and below-ground C from the cover crop returned to the soil was calculated using allometric functions embedded in C-TOOL (Taghizadeh-Toosi *et al.*, 2014), and assuming that 10% of this annual input would persist in the soil after 15-20 years (Mutegi *et al.*, 2013).

Results

Environmental conditions

During the monitoring period (15 October 2019 to 25 June 2020), daily mean air temperature ranged from -1 to 21°C, and total precipitation was 582 mm (Fig. 1). The winter period (December to February) was 2-4°C warmer than the average for the period 1991-2020), whereas precipitation was above average every month between October and February. Rainfall in February was particularly extreme at 134 mm against an average of 41 mm for the period 1991-2020.

The soil temperature at 8 cm depth was recorded continuously in all eight treatment combinations in one of the three experimental blocks. Main effects of tillage, straw residue recycling and cover crop, respectively, are shown as box plots representing the measurements during the 24-hour periods of individual sampling days (Fig. 2). For the autumn-winter period, paired-sample t-tests of mean soil temperature for these selected days showed small, but significantly higher soil temperature at 8 cm depth with no-till ($p < 0.001$), straw retention ($p < 0.05$) and the presence of a cover crop ($p < 0.01$). After termination of the cover crop, the cover crop treatment still had a positive effect on soil temperature ($p < 0.05$), while there was no main effect of tillage or straw retention.

Soil WFPS and mineral N dynamics

Soil WFPS (Fig. S1) remained close to 60% during autumn, winter and early spring independent of treatment, except for a transient increase for the sampling in February after a period with high rainfall (Fig. 1). During spring and early summer, there was a gradual decline in WFPS in all treatments to a final level of around 30%, and with a tendency for higher WFPS in cover crop treatments (+CC) that may reflect moisture retention by the plant material.

Across all treatments, NH_4^+-N content was always below 10 µg g^{-1} dry wt. soil. There was a minor increase in NH_4^+-N content in late autumn irrespective of management practice, and a more significant accumulation following termination of the cover crop and planting of faba bean (Fig. S2). During autumn and winter, there were significant effects of tillage practice and cover crop, but not straw retention, on soil NH_4^+-N content (~~Table 3~~Table S1), but the levels were low and often variable. After termination of the cover crop, with tillage of the PL treatments and then planting of faba bean in all plots, significant interactions between straw retention and both cover crop residues and tillage treatment were observed. In general, NH_4^+-N accumulation in spring was greater with direct seeding and presence of a cover crop, and with straw retention (up to 6 µg NH_4^+-N g^{-1} dry soil).

The soil NO_3^--N content was in the range 0-5 µg g^{-1} dry wt. soil during autumn and winter, and reached 10-15 µg g^{-1} during spring (Fig. S3). Like NH_4^+-N, NO_3^--N showed a minor increase in late autumn independent of treatment, but NO_3^--N was subsequently depleted during the wet winter and early spring before tillage and planting. There was in this period a significant effect of straw retention, and an interaction between cover crop and tillage treatment (~~Table 3~~Table S1). During spring, accumulation of soil NO_3^--N was observed independent of treatment; the content peaked in late May (close to 15 µg NO_3^--N g^{-1} dry soil in all treatments), and NO_3^--N was then depleted during June. There were in this period significant positive effects of cover crop and straw retention on NO_3^--N content, and a significant interaction between tillage and cover crop that suggested greater NO_3^--N accumulation with direct seeding (~~Table 4~~Table 3).

N_2O emissions

Despite a WFPS around 60% and some accumulation of NO_3^- in the soil, N_2O emissions remained low in all treatments during autumn, winter and early spring (Fig. 3). However, there was a small positive effect of cover crop on N_2O emissions during that period, and an also interaction between cover crop and tillage (~~Table 4~~Table 3) that may have been due to occasional higher emissions without a cover crop in PL (Fig. 3). After termination of the cover crop in +CC treatments, and planting of faba bean, there was still little N_2O emission from –CC treatments, but not significantly higher emissions from +CC treatments (Fig. 3, ~~Table 4~~Table 3) with a maximum flux of around 100 µg N_2O-N m^{-2} h^{-1} in treatment PL+S+CC. In all +CC treatments, the highest rates were observed between mid-April and mid-May, but N_2O emissions during June were also higher in +CC compared to -CC treatments. The statistical analysis indicated a significant effects of straw retention (+S), and a significant interaction between tillage treatment and cover crop (~~Table 4~~Table 3).

The regression analysis showed that soil temperature was a driving variable for N_2O emissions (~~Table 5~~Table S3) in autumn to spring (before planting faba bean). In contrast, during spring and early summer, NO_3^--N was a strong predictor of N_2O emission (~~Table 5~~Table S3), with a significant interaction of NO_3^--N with soil moisture.

Cumulative N_2O-N emissions ranged from 0.04 to 0.78 kg N_2O-N ha^1 (Fig. S4), with the highest amount in treatment PL+S+CC with conventional ploughing, straw retention, and cover crop.

Cover crop treatments consistently resulted in higher emissions than treatments without cover crop.

Nitrate leaching

Fig. S5 shows the NO_3^--N leaching during the monitoring period. Cumulated N loss ranged from 21 to 28 kg ha^{-1} in treatments without CC, and from 4 to 5 kg ha^{-1} in the presence of fodder radish, corresponding to a reduction of 80-84% (~~Table 7~~Table 4). There was no significant difference between tillage treatments, although leaching tended to be higher in PL treatments, and in treatments without straw retention. Cover crop was the only variable that affected NO_3^--N leaching significantly (~~Table 6~~Table S2, ~~Fig. S4~~Fig. S6).

Nitrogen uptake by faba bean

The faba bean N uptake at harvest ranged from 179 to 211 kg N ha^{-1} (Table ~~S1~~S4). There were no significant effects of experimental treatments on faba bean seed dry matter yield or N uptake, but a significant (p < 0.01) effect of straw retention on grain N content (Table ~~S2~~S5). Yield-scaled N_2O emissions were significantly higher in the presence of a cover crop during the previous winter

Overall carbon footprint

For each of the eight treatments, an overall GHG balance was calculated using the observed emissions of N_2O and leaching of NO_3^- during the monitoring period, the annual C stock change in each tillage × straw combination between 2002 and 2019, and a model-estimated long-term retention of C in fodder radish residues (Fig. ~~6~~4). In all treatment combinations, the overall carbon footprint was dominated by the long-term trends in soil C stock changes. With straw removal the carbon footprint was always positive, corresponding to C loss from the soil, and this was independent of the other components of the GHG balance. The emissions of N_2O resulting from degradation of cover crop residues, mainly after cultivation in spring, were comparable to the expected retention of C from cover crop residues. The indirect N_2O emissions from NO_3^- leaching were comparable to or greater than direct N_2O emissions from the soil in the absence of a cover crop.

Discussion

Transition from conventional to conservation agriculture will affect the degree of soil disturbance and distribution of crop residues returned. A meta-analysis by Pittelkow *et al.* (2014) indicated a small yield penalty in most cases that suggests a reduction in nutrient use efficiency with conservation agriculture, which in turn could increase environmental losses. In our experiment, fodder radish reduced NO_3^- leaching irrespective of other management factors, but this did not affect the N uptake of the following crop; any difference in N availability may have been compensated by N fixation of the faba bean crop (Rodriguez *et al.*, 2020). There was a positive effect of straw retention in general on grain N, which is consistent with immobilization during the initial phase of straw decomposition of N that is later re-mineralized (Powlson *et al.*, 1985). As an intermediate of denitrification, N_2O emissions may serve as an indicator of atmospheric losses that should be minimized to improve N use efficiency and reduce the carbon footprint of conservation agriculture. The following sections discuss possible controls of N_2O emissions as basis for the identification of mitigation strategies.

Tillage

The adoption of direct seeding in the long-term experiment in 2002, 17 years prior to this study, reduced N_2O emissions compared with conventional ploughing, as hypothesized (~~Table 6~~Table S2). Across straw retention and cover crop treatments, the reduction in N_2O emissions by direct seeding averaged 46%. Within the same field experiment, the corresponding reduction of N_2O emissions from direct seeding during autumn, winter and spring from winter barley in 2007-2008 (Mutegi *et al.*, 2010), and from fodder radish in 2008-2009 (Petersen *et al.*, 2011), were 27 and 31%, respectively. These observations are consistent with the meta-analysis of van Kessel *et al.*, (2013) concluding that long-term (>10 years) reduced- or no-till management enhances the N_2O mitigation potential.

Galdos *et al.* (2019) found, using X-ray computed tomography, a larger total pore volume and higher connectivity at 0-12 cm depth in a clay soil under zero-tillage compared to conventional tillage after 30 years. VandenBygaart *et al.* (1999) investigated pore structure of a silt loam soil in a no-till chronosequence and found that the number of biopores (>500 μm) was lower under no-till management four years after the transition to no-till, but higher after six and eleven years. In the field experiment used in the present study, Abdollahi and Munkholm (2017) found a lower total porosity in DS compared to PL at 11 years after transition, in accordance with the bulk density measurements given above. However, it should be noted that biopores may be under-represented in the 100-cm^3 samples used here for determination of bulk density when comparing with larger sample volumes (da Silva *et al.*, 2021), and aeration through biopores created by roots and earthworm activity could be a constraint on N_2O emissions in no-till soil.

Straw retention

Straw retained under conservation tillage is prone to slower decomposition compared to conventional systems because of the limited mixing with other soil constituents and organisms (Mitchell *et al.*, 2013). Also, the potential for long-term stabilization is less for residues decomposing at the soil surface (Goméz-Mũnoz *et al.*, 2021). Although significant effects of straw retention on mineral N were identified, straw was not related to N_2O emissions in neither autumn-winter nor spring. Aulakh *et al.* (1991) studied N_2O emissions and denitrification from residues of wheat and other crops when mixed with soil or left at the soil surface; in all treatments, the stimulation of N_2O emissions and denitrification occurred mainly during the initial 8-10 days of incubation, which implies that any direct effect would be associated with the initial stage of straw decomposition. In the present field study, oat straw was returned to the soil surface after harvest in both tillage systems in August 2019, two months before monitoring began, and therefore direct effects of straw retention on N_2O emissions could not be expected until spring ploughing in PL treatments.

Cover crop effects

Although the growth period is shorter and characterized by lower temperatures compared to the main growing season, fodder radish may contribute a substantial amount of C to the soil during growth and *via* crop residues (Mutegi *et al.*, 2011). Using $^{14}CO_2$ pulse labelling, these authors showed that more than 70% of the C fixed by fodder radish was incorporated at 0-10 cm depth. Yet, this C input from fodder radish did not lead to substantial N_2O emissions during the growth period, whereas termination of the cover crop triggered N_2O emissions.

The fact that N_2O emissions after termination were lower in DS treatments indicates that depletion of soil O_2 during the decomposition of CC residues, incorporated by ploughing in PL treatments, was a driver of N_2O emissions. Rochette *et al.* (2008), analyzing N_2O response ratios of no-till *vs.* ploughing as a function of soil wetness, found that tillage treatments had lower or similar N_2O emissions from soil with good and moderate drainage, but no-till showed higher emissions than ploughing in poorly drained soil, which corroborates the conclusion that soil aeration at the depth of residue decomposition is a key factor regulating emissions. Song *et al.* (2019) found a strong relationship between soil O_2 and N_2O in soil profiles, but N_2O emissions will also depend on the potential for N_2O reduction during transport to the surface.

The accumulation of NO_3^- observed in the topsoil during autumn and winter (Fig. S3) was mainly derived from organic matter input in previous years, which would have a very different distribution in the soil profile of no-till and ploughed soil. As a consequence, the contact between decomposing residues and soil NO_3^- was probably better in ploughed soil where residues and fertilizers are incorporated each year, and this in turn may have enhanced N_2O emissions as shown by Taghizadeh-Toosi *et al.* (2021) in a manipulation experiment with mixed or layered distribution of crop residues, and two levels of, respectively, soil moisture and NO_3^- content.

Previous meta-analyses have documented that cover crops could reduce N leaching by 50-70% compared with bare fallow (Basche *et al.*, 2014; Valkama *et al.*, 2015). The present study indicated a reduction of 80-84%, and hence the effect was largely independent of tillage and residue management on this soil type, in accordance with previous results (Hansen *et al.*, 2015).

Soil conditions

Soil WFPS followed the same pattern in all treatments. During autumn and winter with above-average precipitation, the WFPS level at sampling was always close to 60%, which has been proposed to be a threshold for N_2O emissions based on controlled laboratory experiments (Aulakh *et al.*, 1991; Shelton *et al.*, 2000). The N_2O emissions remained low in this period despite some mineral N availability (Figs S2 and S3), although higher rates associated with rain events could have been missed. In contrast, significant N_2O emissions occurred during April and May 2020 where soil WFPS was below 60% (Fig. S3), but mainly where fodder radish residues were returned. Together this indicates that, for the site investigated here, intense decomposer activity was more important as driver for N_2O emissions than soil drainage, in accordance with previous observations at this experimental site (Brozyna *et al.*, 2013; Pugesgaard *et al.*, 2017).

As reported by others, soil temperature during autumn-winter was higher with no-till (Yang *et al.*, 2018), straw retention (Chen *et al.*, 2007) and a cover crop (Kahimba, 2008). The cover crop maintained a positive effect on soil temperature after termination, possibly because cover crop residues promoted soil warming during spring due to a reduction in soil bulk density (Li *et al.*, 2015; Chalise *et al.*, 2019). Considering the importance of temperature for microbial transformations of C and N, such effects could influence N_2O emissions. Thus, Petersen *et al.* (2011) concluded that a large proportion of emitted N_2O was produced above a soil depth of 5 cm in both tilled and non-tilled soil after termination of a fodder radish cover crop. While there was a significant effect of temperature during autumn-winter (~~Table 5~~Table S3), N_2O emissions remained low independent of treatment in this period (Fig. 3); during spring, any effect of temperature was confounded by the main effect of cover crop.

Carbon footprints of conservation agriculture practices

The carbon footprints showed a GHG mitigation potential of direct seeding (no-till) and straw retention, and a neutral effect of cover crop, indicating that conservation tillage practices will have net benefits for the GHG balance of sandy loam soil in wet temperate climates. It should, however, be emphasized that N_2O emissions from main crops, i.e., fertilization of the previous oat crop, and decomposition of residues after harvest of oat and faba bean crops, were not included in the carbon footprints in Fig. ~~6~~4, which represent effects of management outside the main crop growing period. For example, Petersen *et al*. (2011) quantified N_2O emissions in PL+S and DS+S of the same long-term tillage experiment with and without fodder radish as winter cover crop, and here pig slurry was applied after termination of the cover crop. The combined N_2O emissions from residue and slurry decomposition reported by Petersen *et al.* (2011) corresponded to 0.7-1.6 Mg CO_2eq and thus were considerably higher than those from residues alone in the present study. Therefore, N_2O emissions after fertilization could outweigh the net benefits of the scenarios with straw retention reported in Fig. ~~6~~4.

The largest component of the carbon footprint was the change in soil C associated with straw retention. It should be noted that cover crops had been part of all treatments in previous years (Goméz-Mũnoz *et al.*, 2021), and therefore the soil C change in Fig. ~~6~~ 4 also included a contribution from cover crops. The soil C retention observed for fodder radish in 2019-2020 may indicate the relative importance of cover crops and straw retention, respectively, for the soil C stock changes. In a global meta-analysis, McClelland *et al.* (2021) estimated the annual C input from cover crops to be on average 0.21 Mg ha^{-1}. These authors also found that the response to cover crops was smaller in wet temperate compared to warmer climates, and that the apparent effect of cover crops on soil C stocks did not change with time since adoption. This has recently been corroborated by a long-term study in Denmark showing that the soil C effect of cover crops saturate after a period of approximately 15 years (Jensen *et al*., 2021). It indicates that the long-term retention of residue C in wet temperate climates is uncertain and, accordingly, that the retention of fodder radish C estimated from plant cuts in the present study, 0.23-0.43 Mg ha^{-1}, may be too high. If true, this would increase the relative importance of N_2O emissions for the GHG balance.

The use of a cover crop was highly effective in preventing NO_3^- leaching, which is a pollutant of recipient water bodies and an indirect source of N_2O, but the direct emissions of N_2O from cover crop residues were far greater than indirect emissions prevented in this study. This is in contrast to the results of a global meta-analysis of cover crop effects (Abdalla *et al.*, 2019) which concluded there was no significant effect of cover crops on N_2O emissions. The short duration (9 months) of the present study may have resulted in a higher relative importance of N_2O emissions associated with cover crop residues, but crop residues have also been identified as a main driver of N_2O emissions across entire four-year crop rotations on the light-textured soil investigated here (Brozyna *et al.*, 2013; Pugesgaard *et al.*, 2017). Thus, confounding effects of analysing field data across several soil types, climate zones and management practices may prevent the observation of effects that are seen with a greater disaggregation of the data (Chen *et al.*, 2013). As GHG mitigation strategy, the fodder radish crop in the present study depended on the concurrent C storage which, however, will be less permanent than N_2O in the atmosphere. Direct seeding resulted in less N_2O emission than spring tillage, and the combination of GHG mitigation methods under conservation agriculture should be further explored.

Conclusion

This study investigated effects of tillage practice, straw retention, and a winter cover crop on N_2O emissions and NO_3^- leaching, during the cover crop growth period and subsequent decomposition in spring. The study did not find effects of straw retention on nitrate leaching or N_2O emissions, but straw retention was critical for the overall GHG balance through soil carbon storage, and without straw retention after harvest there was always a positive GHG balance, i.e., net emissions. As expected, fodder radish was highly effective in reducing NO_3^- leaching during winter, but N_2O emissions after spring cultivation were enhanced; the increase in direct N_2O

emissions could in part be compensated by storage of residue C and by the adoption of direct seeding. Even though N losses associated with a main crop were not covered in this study, the results indicated that direct seeding, straw retention and the use of cover crops could substantially improve the carbon footprint of agroecosystems on sandy soil in a wet temperate climate.

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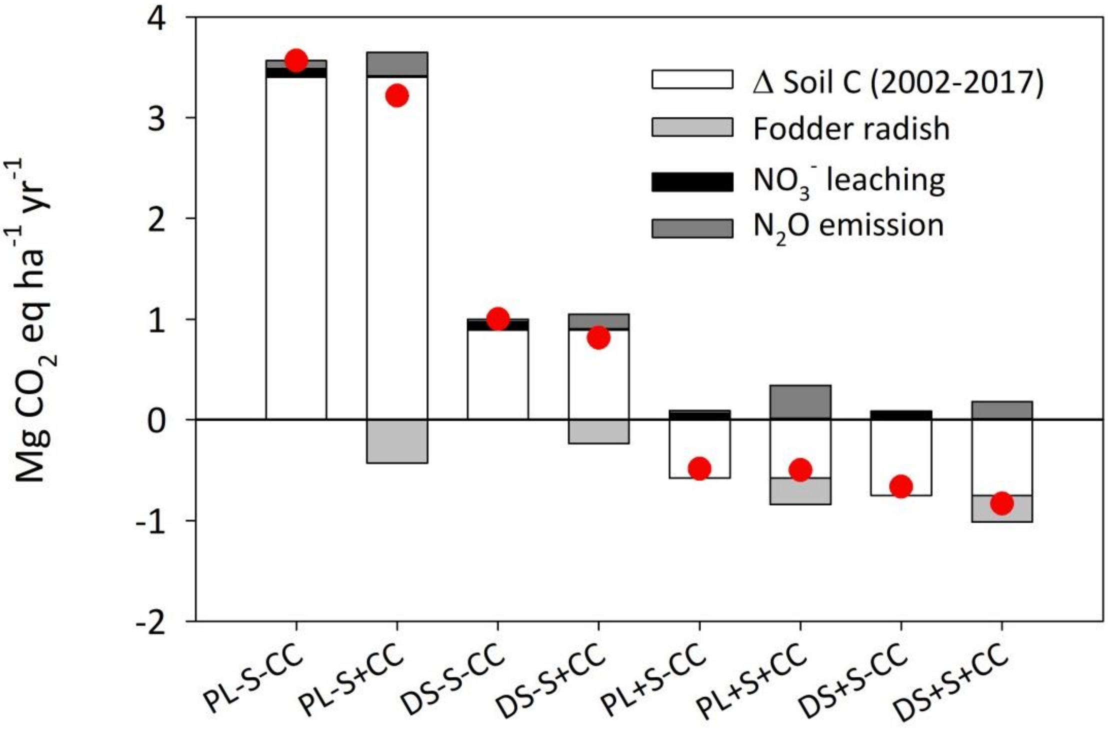
Graphical Abstract



Total greenhouse gas balance in red filled circles (October 2019 to June 2020) of the eight combinations of ploughing (PL) vs. direct seeding (DS), straw retention (+S) vs. straw removal (-S), and growth of fodder radish as a winter cover crop (+CC) vs. no cover crop (-CC). Positive effects indicate net emission of greenhouse gases from the soil.

Highlights

- Fodder radish, grown as a cover crop, reduced NO_3^- leaching by 80-84%.
- Termination of the cover crop in spring was followed by a flush of N_2O.
- No-till reduced N_2O emissions by on average 46% compared to ploughing.
- Straw retention in the rotation was more important than cover crop for C storage.
- Without straw retention there were net GHG emissions irrespective of tillage.

Manuscript (double-spaced and continuously LINE and PAGE numbered)-for final publication
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Interactive effects of straw management, tillage, and a cover crop on nitrous oxide emissions and nitrate leaching from a sandy loam soil

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Abstract

Minimum tillage, residue recycling and the use of cover crops are key elements of conservation agriculture that play important roles in soil carbon (C) and nitrogen (N) dynamics. This study determined the long-term effects of tillage practice (conventional ploughing *vs.* direct seeding), straw management (retained *vs.* removed), and the presence of a cover crop (CC; fodder radish in this study) on nitrous oxide (N_2O) emissions, nitrate (NO_3^-) leaching, and soil mineral N dynamics between October 2019 and June 2020. In the factorial experiment with eight treatment combinations, cumulative N_2O emissions ranged from 0.04 to 0.8 kg N ha^{-1}, whereas NO_3^- leaching varied between 4 and 28 kg N ha^{-1}. The study did not find effects of straw retention on NO_3^- leaching or N_2O emissions. No-till reduced N_2O emissions by on average 46% compared to ploughing. Fodder radish reduced NO_3^- leaching by 80-84%, and there was little N_2O emission in the presence of the cover crop; however, after termination in spring there was a flush of N_2O,cumulative N_2O-N averaged 0.1 and 0.5 kg N ha^{-1} without and with a cover crop. With information about long-term soil C retention from straw and fodder radish, an overall greenhouse (GHG) balance was calculated for each system. Without straw retention after harvest there was always a positive net GHG emission, and the indirect N_2O emission from NO_3^- leaching was similar to, or greater than direct N_2O emissions. However, in the presence of fodder radish, the direct N_2O emissions after termination were much more important than indirect emissions, and negated the C input from fodder radish. Direct seeding, straw retention and the use of a cover crop showed positive effects on N retention and/or GHG balance and could substantially improve the carbon footprint of agroecosystems on sandy soil in a wet temperate climate.

Keywords: Conservation agriculture, nitrous oxide emissions, nitrate leaching, straw retention, cover crops.

Introduction

Within agriculture, sustainability will depend on land conservation and improved input management as identified in the United Nations Programme of Action (UN, 2020). Specifically, *conservation agriculture* is based on principles that include minimum tillage, residue recycling, use of cover crops, and diversification of crop rotations (FAO, 2021). The agronomic performance relative to conventional practices, however, is site-specific (Giller *et al.,* 2015), which indicates that environmental impacts are also variable (Sun *et al*., 2020). It is therefore necessary to determine at national or regional level to what extent the practices of conservation agriculture ensure nutrient recycling, avoid environmental losses, and reduce greenhouse gas emissions.

In agricultural systems, the nitrogen (N) cycle is "leaky", and efficient use of N has proven challenging; for instance, less than one-half of fertilizer N is recovered in the aboveground portion of cereal crops (Ladha *et al.*, 2005), suggesting that the fraction of applied N remaining in the soil is at risk of being lost to non-target ecosystems (Davidson, 2012). There are several undesirable mechanisms by which N may be lost, including nitrate (NO_3^-) leaching and gaseous emissions resulting from processes such as nitrification and denitrification (Butterbach-Bahl *et al.*, 2013). These emissions include nitrous oxide (N_2O), a powerful greenhouse gas with a contribution of 66% from the agricultural sector, mostly as direct emissions from soils (Davidson and Kanter, 2014). The profound effects of N_2O in the atmosphere (i.e. its high global warming potential and contribution to stratospheric ozone depletion), and of NO_3^- leaching on surface- or groundwater quality, are well documented, but mitigating N losses remains difficult (IPCC, 2019; Bakken and Frostegard, 2017).

Tillage can affect soil N availability through its influence on soil physical, chemical, and biological properties. Conventional inversion tillage favors the decomposition of crop residues and soil organic matter by enhancing aeration and soil-residue contact, and promoting soil microbial activity (Singh *et al.*, 2021), whereas conservation tillage practices (i.e., direct seeding) result in less soil disturbance, which slows down decomposition of crop residues but also reduces fuel costs and disturbance of soil organisms, and increases the volume of medium pores and soil water holding capacity (FAO, 2014; Munkholm *et al.*, 2020). More NO_3^--N has been reported to accumulate in soil under conventional tillage than in reduced or no-tillage soils, although reasons for the greater accumulation are not well understood (Carefoot and Janzen, 1997). Conservation tillage practices seem to reduce the activity of ammonia oxidizing bacteria (Wang *et al*. 2019), and the lower soil NO_3^--N content reported in some no-tillage systems has also been linked to leaching below the root zone, or greater soil N immobilization (Singh *et al.*, 2021). However, long-term studies in Denmark have not been able to confirm greater NO_3^- leaching from conventional compared with conservation tillage (Hansen *et al*., 2015).

Mutegi *et al*. (2010) found that straw recycling resulted in higher N_2O emissions under conventional tillage compared to reduced tillage six years after transition. Retention of crop residues at the soil surface will affect soil temperature, and the thermal insulation provided by surface residues tends to delay and dampen temperature variations in soil under conservation tillage when compared to soil under conventional tillage (Grevers *et al.*, 1986). Changing temperature regimes can, together with effects of conservation agriculture on organic matter and oxygen (O_2) availability and soil gas diffusivity, affect the potential for denitrification (Skiba and Ball, 2002).

In agroecosystems, the recycling of residues, including straw as a by-product of crop production, can improve soil structure, provide plant nutrients, and act as a primary energy source for soil microorganisms (FAO, 2014). Retention of straw along with appropriate fertilization may increase soil fertility and increase crop yields, and reduce N losses outside the growing season. Powlson *et al.* (1985) incorporated straw with 0.5% N into a soil that was subsequently sown with winter wheat, and they found that 78% of the N added in straw remained in the soil a year after incorporation, thereby increasing the quantity of mineralizable N. Nitrogen mineralization in synchrony with crop uptake (in spring and summer) can reduce crop fertilizer N requirements, whereas any N mineralization during autumn or winter increases the risk of NO_3^- leaching.

Establishing a cover crop, or catch crop, outside the main growing season, has frequently been shown to reduce NO_3^- leaching. Thapa *et al*. (2018) found in a global meta-analysis an average reduction of 56% by non-leguminous cover crops, and a higher efficiency for coarse-textured soil. By reducing soil mineral N availability outside the main growing season, cover crops may also reduce the potential for N_2O emissions. A meta-analysis by Basche *et al.* (2014) found mixed results for the effect of cover crops on N_2O emissions during the growth period, but a stimulation of N_2O emissions after incorporation of the cover crop in spring. Further, cover crops provide an input of carbon to the soil, which may help maintain soil C stocks (McClelland *et al.*, 2021).

A global meta-analysis found a higher risk for N_2O emissions under conservation tillage practices (Mei *et al.*, 2018), although there are results indicating a reversal of this effect after long-term continuous conservation tillage, possibly due to increasing total porosity created by roots and soil fauna (Six *et al.*, 2004; Finney *et al.*, 2015). Soil under conservation tillage has also been found to have a higher relative abundance of nosZ encoding the enzyme N_2O reductase, and more complete denitrification to N_2 would reduce N_2O emissions (Wang and Zou, 2020; Wang *et al.*, 2019). The effect of reduced tillage, however, may also depend on soil type and climate (Rochette, 2008; Mei *et al.*, 2018). These complex interactions between the different elements of conservation agriculture, i.e., residue recycling, use of cover crops, and minimum tillage, are key to understanding the environmental impacts of conservation agriculture practices.

The objectives of this study were to explore the interactions between tillage practice, a cover crop, and straw management with respect to: (1) dynamics of soil mineral N, (2) N_2O emissions, and (3) NO_3^- leaching. The study included the two main phases where a cover crop may influence nitrogen losses,i.e., the growth period (autumn-early spring) and the period where cover crop residues decompose in the soil (spring-early summer). The hypotheses were that: (i) minimum tillage would reduce N_2O emissions compared to ploughing; (ii) retention of straw in the field would reduce NO_3^- leaching compared to straw removal, but would increase N_2O emissions during autumn; and (iii) the presence of cover crop in the field would reduce NO_3^- leaching, but increase N_2O emissions after spring cultivation.

Materials and methods

Study site

Measurements were conducted between October 2019 and July 2020 within a long-term field experiment at Foulum (56°30´N, 9°35´E, elevation 56 m a.s.l.), Aarhus University, Denmark. The selected experimental plots were part of a larger field experiment, which was established in 2002 under temperate coastal climatic conditions to evaluate the effects of tillage, straw retention and cropping sequence, including cover crops, on crop yield and C and N cycling, and retention. The soil is a loamy sand based on ground morainic deposits from the last glaciation classified as a Mollic Luvisol according to the WRB (FAO) system (Krogh and Greve, 1999). The soil contains 8.1-8.8% clay, 3.5-3.8% organic matter, with pH 6.1 (1:2.5 suspension of soil: 0.01 M $CaCl_2$). Before the experiment was established, the field had been cropped and cultivated according to normal Danish agricultural practices (Hansen *et al*., 2015).

The field experiment had a split-plot design with different crop rotations in main plots and tillage treatments in subplots (Munkholm *et al*., 2013). This study included two cereal-based crop rotations which differed only with respect to straw management (removed or retained). Two tillage treatments were represented, i.e. conventional ploughing (PL) and direct seeding (DS), and tillage was applied to 72 m long and 6 m wide subplots, each of which had ten 3 x 10 m^2 sub-subplots. Fodder radish (*Raphanus sativus* L.) was grown as a cover crop from September 2019 to April 2020. Fodder radish followed oat (*Avena sativa* L.) as the main crop in 2019, and was followed in spring 2020 by faba beans (*Vicia faba* L.). One 3×10 m^2 plot with the cover crop was selected, as well as one plot kept free of vegetation with a herbicide cf. Table 1 (bare soil); bare-soil plots had been grown with cover crops before 2017. Thus, measurements in this study were restricted to net plot areas of 10×3 m^2. The PL treatments were mouldboard ploughed to 20 cm depth followed by levelling to make a good seedbed before sowing. Sowing in the CT treatment was performed with a traditional seed drill (Nordsten Lift-o-matic CLH300), whereas a chisel coulter drill (Horsch Airseeder CO3) was used in the DS treatments. In all treatments, crops were sown at the same row distance of 17.5 cm. In the crop rotation with straw retention, straw was chopped and retained after harvest, whereas straw was removed in the crop rotation without straw retention. Cover crop seeds were scattered from a seeding machine after harvest of the main crop (Table 1).

Table 2 shows the treatments and variables that were monitored in the experiment. With two tillage systems (PL and DS), each with or without straw retention (+S and –S), and with or without cover crop (+CC and –CC) in three replicate blocks, there were a total of 24 sampling positions. Precipitation and average temperatures for the experimental period were obtained from the meteorological station at Foulum and are shown in Fig. 1. Soil temperature was also monitored at 8 cm below the soil surface using Temperature-Moisture-Sensor (TMS) data loggers (Wild *et al.*, 2019).

Nitrate leaching

Nitrate leaching was determined from analyses of soil water samples collected *via* porous ceramic cups (two in each plot) installed in autumn 2002 at a depth of 1 m. The sampling system consists of suction cups (655x01-B1M1, 1 bar, standard, Soil moisture Equipment Corporation, Goleta, CA) mounted on PVC pipes (Hansen *et al.*, 2015). Suction and collection tubes are protected by rigid plastic tubes, which extend outside the experimental field to cupboards with three-way valves for sample collection. A suction of approximately 70–80 kPa was imposed 2–3 days before sampling of pore water. The soil water samples from each plot were bulked and frozen within a few hours, and later analyzed for NO_3^--N (Best, 1975). Sampling took place biweekly except in periods with frost or drought.

The daily drainage in each plot was calculated with the EVACROP model (Olesen and Heidmann, 1990; Hansen *et al.*, 2010), which applies a cascading model for calculating percolation at a depth of 1 m based on a simple conceptual water-balance model with a number of simple coupled linear reservoirs for snow, interception, evaporation, and transpiration. Soil-specific input parameters include the permanent available water content for each soil layer, and drainage constants of the root zone reservoir and the subsoil reservoir, which describe the fraction of the water that drains to the next deeper layer each day if the soil moisture in the layer is above field capacity. Data on daily precipitation, air temperature, and reference evapotranspiration from a meteorological station located close to the experimental field, were obtained to run the model. Precipitation was measured at 1.5 m height, and corrected to the soil surface (Allerup *et al.*, 2000).

Nitrate concentrations in the period between two observations were estimated by percolation-weighted interpolation, or ‘drainage-linear interpolation’ (Vogeler *et al.*, 2019), which provides the relative change in concentration based on daily percolation assuming that NO_3^- concentrations in the extracted soil water represented average flux concentrations (Askegaard *et al.*, 2005). Daily NO_3^- leaching (kg N ha^{-1}) was calculated for each treatment plot by multiplying daily percolation (mm) with flow-weighted interpolated daily NO_3^--N concentration (mg L^{-1}) and dividing by 100. Afterwards the cumulative NO_3^--N leaching was calculated for the whole period from 30 August 2019 to 2 April 2020.

N_2O and CO_2 measurements

Monitoring of N_2O and CO_2 took place between 15 October 2019 and 25 June 2020 with a total of 21 sampling days over the 36-week period. Fluxes were determined using 75 cm × 75 cm × 20 cm static chambers as previously described (Baral *et al.*, 2019); collars were inserted one day before the start of flux measurements. Chamber headspace air was continuously mixed by a battery-powered fan during chamber deployment. Gas samples were taken with a 10-mL syringe and hypodermic needle via a septum after first pumping air in and out of the syringe three times to flush the syringe. Five 10 mL gas samples were collected for flux measurements, the first sample immediately after chamber deployment and four other samples at 15–20 min intervals. Sampling was always initiated between 10:00 and 13:00. Gas samples were stored in pre-evacuated 6 mL exetainers (Labco, High Wycombe, UK) until analysis for N_2O and CO_2 using an Agilent 7890 (Agilent, Nærum, Denmark) gas chromatograph configured as previously described (Petersen *et al.*, 2012). The concentration-time series of CO_2 were mainly used for quality control purposes. Nitrous oxide fluxes were calculated by the free software package HMR avaiable as a package in R (Pedersen *et al.*, 2010).

Soil measurements

Soil samples were collected in the same plots where N_2O and CO_2 were monitored, and on the same days as gas sampling. Six individual samples were randomly collected from each treatment and block using an auger (2 cm dia., 0–20 cm depth); the subsamples were pooled to make a representative composite sample per plot, transferred to zip-lock plastic bags and kept at 2 °C until analyzed. Soil samples were sieved (<2 mm) and subsamples extracted for mineral N (ammonium (NH_4^+-N) and nitrate (NO_3^--N) analysis within two days of soil sampling. Approximately 10 g of soil was extracted in 40 mL of 1 M KCl. Additionally, two KCl blanks were prepared per batch to correct for any bias from KCl impurities. For extraction, both the soil-KCl solutions and blank samples were shaken for 30 min on an orbital shaker set at 150 rpm and then allowed to settle. The supernatant was then filtered through 1.6 µm glass microfiber filters (VWR, Sweden) and frozen at -20 °C for later analysis. Inorganic-N analysis was done with standard colorimetric methods on a Foss FIAstar 5000 flow injection analyzer (FOSS Denmark). Gravimetric soil water content was determined by drying approximately 10 g of soil for 24 h at 105°C. Using information about soil bulk density in the tillage experiment determined in November 2019 (Goméz-Mũnoz *et al*., 2021), gravimetric water content was used to calculate water-filled pore space (WFPS). For 0-20 cm depth, bulk density of PL-S, DS-S, PL+S and DS+S were 1.39, 1.39, 1.29 and 1.31 Mg m^{-3}, respectively.

Data analyses

Statistical analyses of the data were performed using R programming language version 4.0.0 (Noguchi *et al.*, 2012; R, 2018). All data, except cumulative N_2O emissions and NO_3^--N leaching, were split into pre-tillage (15 October 2019 – 2 April 2020) and post-tillage (8 April – 25 June 2020) seasons for analysis. Main and interacting effects of tillage, straw retention and cover crop on the daily and cumulative N_2O emissions and mineral N were analyzed with the linear mixed effect (*lme)* function of the *nlme* package using the restricted maximum likelihood (REML) method. Blocks were treated as a random factor in the analyses. Model assumptions, i.e., normality and homogeneity of variance, were assessed using diagnostic plots of residuals. In order to satisfy model assumptions, the daily and cumulative N_2O emissions and mineral N data were log-transformed. For time series of N_2O emissions and mineral N data, auto-correlation between sampling positions were accounted with the *corAR1* function. Pairwise comparisons between treatments were performed using the estimated marginal mean *(emmeans)* function. The *p*-values were adjusted by Tukey's HSD method, and the hypothesis rejection threshold was 0.05.

Effects of soil and environmental variables, such as mineral N (NH_4^+-N and NO_3^--N), soil temperature and moisture, to pre-tillage and post-tillage season N_2O emissions were analyzed using multiple regression analysis. Prior to the analysis, N_2O emission data were log-transferred and tested for collinearity between variables using a variance inflation factor (VIF) of 5 as threshold. During analysis, only two-way interactions of variables with respect to N_2O emissions were analyzed to make the results interpretable. The variable interactions that were not significant ($p > 0.05$) were removed in the final model.

Since daily NO_3^- leaching data violated model assumptions, even after transformation, the data were analysed using the non-parametric rank-based model *nparLD*, which is a robust method to analyse repeatedly measured data with non-normal distribution (Noguchi *et al*., 2012). We used F2-LD-F1 design during analysis, where F2 represents the two levels of independent factors, and F1 stands for repeatedly measured leaching data. Since this model can only handle interactions between two independent factors at a time, we made three different interaction models during the analysis, i.e., tillage × residue, residue × cover crop, and tillage × cover crop. Treatment effects within factors were evaluated using ANOVA-type statistics. As in parametric tests, the hypothesis rejection threshold was set at 0.05.

The overall carbon footprint of the eight treatment combinations was calculated from the direct emissions of N_2O and indirect N_2O emissions from NO_3^- leached, and C from straw and fodder radish stabilized in the soil. The indirect N_2O emission was estimated using an emission factor of 0.0075, as in the national inventory for Denmark (Nielsen *et al*., 2020), and a global warming potential (GWP) of 265 for direct and indirect emissions (IPCC, 2014). The annual net change of straw C was calculated from the change in soil C concentrations of PL and DS soil with and without straw retention between 2002 and 2019 (17 years), and bulk density determined in November 2019 (Gómez-Muñoz *et al*., 2021). The retention of cover crop C was based on dry matter in plant cuts collected in January 2020; the total above- and below-ground C from the cover crop returned to the soil was calculated using allometric functions embedded in C-TOOL (Taghizadeh-Toosi *et al.*, 2014), and assuming that 10% of this annual input would persist in the soil after 15-20 years (Mutegi *et al.*, 2013).

Results

Environmental conditions

During the monitoring period (15 October 2019 to 25 June 2020), daily mean air temperature ranged from -1 to 21°C, and total precipitation was 582 mm (Fig. 1). The winter period (December to February) was 2-4°C warmer than the average for the period 1991-2020), whereas precipitation was above average every month between October and February. Rainfall in February was particularly extreme at 134 mm against an average of 41 mm for the period 1991-2020.

The soil temperature at 8 cm depth was recorded continuously in all eight treatment combinations in one of the three experimental blocks. Main effects of tillage, straw residue recycling and cover crop, respectively, are shown as box plots representing the measurements during the 24-hour periods of individual sampling days (Fig. 2). For the autumn-winter period, paired-sample t-tests of mean soil temperature for these selected days showed small, but significantly higher soil temperature at 8 cm depth with no-till ($p < 0.001$), straw retention ($p < 0.05$) and the presence of a cover crop ($p < 0.01$). After termination of the cover crop, the cover crop treatment still had a positive effect on soil temperature ($p < 0.05$), while there was no main effect of tillage or straw retention.

Soil WFPS and mineral N dynamics

Soil WFPS (Fig. S1) remained close to 60% during autumn, winter and early spring independent of treatment, except for a transient increase for the sampling in February after a period with high rainfall (Fig. 1). During spring and early summer, there was a gradual decline in WFPS in all treatments to a final level of around 30%, and with a tendency for higher WFPS in cover crop treatments (+CC) that may reflect moisture retention by the plant material.

Across all treatments, NH_4^+-N content was always below 10 μg g^{-1} dry wt. soil. There was a minor increase in NH_4^+-N content in late autumn irrespective of management practice, and a more significant accumulation following termination of the cover crop and planting of faba bean (Fig. S2). During autumn and winter, there were significant effects of tillage practice and cover crop, but not straw retention, on soil NH_4^+-N content (Table S1), but the levels were low and often variable. After termination of the cover crop, with tillage of the PL treatments and then planting of faba bean in all plots, significant interactions between straw retention and both cover crop residues and tillage treatment were observed. In general, NH_4^+-N accumulation in spring was greater with direct seeding and presence of a cover crop, and with straw retention (up to 6 μg NH_4^+-N g^{-1} dry soil).

The soil NO_3^--N content was in the range 0-5 μg g^{-1} dry wt. soil during autumn and winter, and reached 10-15 μg g^{-1} during spring (Fig. S3). Like NH_4^+-N, NO_3^--N showed a minor increase in late autumn independent of treatment, but NO_3^--N was subsequently depleted during the wet winter and early spring before tillage and planting. There was in this period a significant effect of straw retention, and an interaction between cover crop and tillage treatment (Table S1). During spring, accumulation of soil NO_3^--N was observed independent of treatment; the content peaked in late May (close to 15 μg NO_3^--N g^{-1} dry soil in all treatments), and NO_3^--N was then depleted during June. There were in this period significant positive effects of cover crop and straw retention on NO_3^--N content, and a significant interaction between tillage and cover crop that suggested greater NO_3^--N accumulation with direct seeding (Table 3).

N_2O emissions

Despite a WFPS around 60% and some accumulation of NO_3^- in the soil, N_2O emissions remained low in all treatments during autumn, winter and early spring (Fig. 3). However, there was a small positive effect of cover crop on N_2O emissions during that period, and an also interaction between cover crop and tillage (Table 3) that may have been due to occasional higher emissions without a cover crop in PL (Fig. 3). After termination of the cover crop in +CC treatments, and planting of faba bean, there was still little N_2O emission from –CC treatments, but not significantly higher emissions from +CC treatments (Fig. 3, Table 3) with a maximum flux of around 100 µg N_2O-N m^{-2} h^{-1} in treatment PL+S+CC. In all +CC treatments, the highest rates were observed between mid-April and mid-May, but N_2O emissions during June were also higher in +CC compared to -CC treatments. The statistical analysis indicated a significant effects of straw retention (+S), and a significant interaction between tillage treatment and cover crop (Table 3).

The regression analysis showed that soil temperature was a driving variable for N_2O emissions (Table S3) in autumn to spring (before planting faba bean). In contrast, during spring and early summer, NO_3^--N was a strong predictor of N_2O emission (Table S3), with a significant interaction of NO_3^--N with soil moisture.

Cumulative N_2O-N emissions ranged from 0.04 to 0.78 kg N_2O-N ha^1 (Fig. S4), with the highest amount in treatment PL+S+CC with conventional ploughing, straw retention, and cover crop.

Cover crop treatments consistently resulted in higher emissions than treatments without cover crop.

Nitrate leaching

Fig. S5 shows the NO_3^--N leaching during the monitoring period. Cumulated N loss ranged from 21 to 28 kg ha^{-1} in treatments without CC, and from 4 to 5 kg ha^{-1} in the presence of fodder radish, corresponding to a reduction of 80-84% (Table 4). There was no significant difference between tillage treatments, although leaching tended to be higher in PL treatments, and in treatments without straw retention. Cover crop was the only variable that affected NO_3^--N leaching significantly (Table S2, Fig. S6).

Nitrogen uptake by faba bean

The faba bean N uptake at harvest ranged from 179 to 211 kg N ha^{-1} (Table S4). There were no significant effects of experimental treatments on faba bean seed dry matter yield or N uptake, but a significant ($p < 0.01$) effect of straw retention on grain N content (Table S5). Yield-scaled N_2O emissions were significantly higher in the presence of a cover crop during the previous winter

Overall carbon footprint

For each of the eight treatments, an overall GHG balance was calculated using the observed emissions of N_2O and leaching of NO_3^- during the monitoring period, the annual C stock change in each tillage × straw combination between 2002 and 2019, and a model-estimated long-term retention of C in fodder radish residues (Fig. 4). In all treatment combinations, the overall carbon footprint was dominated by the long-term trends in soil C stock changes. With straw removal the carbon footprint was always positive, corresponding to C loss from the soil, and this was independent of the other components of the GHG balance. The emissions of N_2O resulting from degradation of cover crop residues, mainly after cultivation in spring, were comparable to the expected retention of C from cover crop residues. The indirect N_2O emissions from NO_3^- leaching were comparable to or greater than direct N_2O emissions from the soil in the absence of a cover crop.

Discussion

Transition from conventional to conservation agriculture will affect the degree of soil disturbance and distribution of crop residues returned. A meta-analysis by Pittelkow *et al.* (2014) indicated a small yield penalty in most cases that suggests a reduction in nutrient use efficiency with conservation agriculture, which in turn could increase environmental losses. In our experiment, fodder radish reduced NO_3^- leaching irrespective of other management factors, but this did not affect the N uptake of the following crop; any difference in N availability may have been compensated by N fixation of the faba bean crop (Rodriguez *et al.*, 2020). There was a positive effect of straw retention in general on grain N, which is consistent with immobilization during the initial phase of straw decomposition of N that is later re-mineralized (Powlson *et al.*, 1985). As an intermediate of denitrification, N_2O emissions may serve as an indicator of atmospheric losses that should be minimized to improve N use efficiency and reduce the carbon footprint of conservation agriculture. The following sections discuss possible controls of N_2O emissions as basis for the identification of mitigation strategies.

Tillage

The adoption of direct seeding in the long-term experiment in 2002, 17 years prior to this study, reduced N_2O emissions compared with conventional ploughing, as hypothesized (Table S2). Across straw retention and cover crop treatments, the reduction in N_2O emissions by direct seeding averaged 46%. Within the same field experiment, the corresponding reduction of N_2O emissions from direct seeding during autumn, winter and spring from winter barley in 2007-2008 (Mutegi *et al.*, 2010), and from fodder radish in 2008-2009 (Petersen *et al.*, 2011), were 27 and 31%, respectively. These observations are consistent with the meta-analysis of van Kessel *et al.*, (2013) concluding that long-term (>10 years) reduced- or no-till management enhances the N_2O mitigation potential.

Galdos *et al.* (2019) found, using X-ray computed tomography, a larger total pore volume and higher connectivity at 0-12 cm depth in a clay soil under zero-tillage compared to conventional tillage after 30 years. VandenBygaart *et al.* (1999) investigated pore structure of a silt loam soil in a no-till chronosequence and found that the number of biopores (>500 μm) was lower under no-till management four years after the transition to no-till, but higher after six and eleven years. In the field experiment used in the present study, Abdollahi and Munkholm (2017) found a lower total porosity in DS compared to PL at 11 years after transition, in accordance with the bulk density measurements given above. However, it should be noted that biopores may be under-represented in the 100-cm^3 samples used here for determination of bulk density when comparing with larger sample volumes (da Silva *et al.*, 2021), and aeration through biopores created by roots and earthworm activity could be a constraint on N_2O emissions in no-till soil.

Straw retention

Straw retained under conservation tillage is prone to slower decomposition compared to conventional systems because of the limited mixing with other soil constituents and organisms

(Mitchell *et al.*, 2013). Also, the potential for long-term stabilization is less for residues decomposing at the soil surface (Goméz-Mũnoz *et al.*, 2021). Although significant effects of straw retention on mineral N were identified, straw was not related to N_2O emissions in neither autumn-winter nor spring. Aulakh *et al.* (1991) studied N_2O emissions and denitrification from residues of wheat and other crops when mixed with soil or left at the soil surface; in all treatments, the stimulation of N_2O emissions and denitrification occurred mainly during the initial 8-10 days of incubation, which implies that any direct effect would be associated with the initial stage of straw decomposition. In the present field study, oat straw was returned to the soil surface after harvest in both tillage systems in August 2019, two months before monitoring began, and therefore direct effects of straw retention on N_2O emissions could not be expected until spring ploughing in PL treatments.

Cover crop effects

Although the growth period is shorter and characterized by lower temperatures compared to the main growing season, fodder radish may contribute a substantial amount of C to the soil during growth and *via* crop residues (Mutegi *et al.*, 2011). Using $^{14}CO_2$ pulse labelling, these authors showed that more than 70% of the C fixed by fodder radish was incorporated at 0-10 cm depth. Yet, this C input from fodder radish did not lead to substantial N_2O emissions during the growth period, whereas termination of the cover crop triggered N_2O emissions.

The fact that N_2O emissions after termination were lower in DS treatments indicates that depletion of soil O_2 during the decomposition of CC residues, incorporated by ploughing in PL treatments, was a driver of N_2O emissions. Rochette *et al.* (2008), analyzing N_2O response ratios of no-till *vs.* ploughing as a function of soil wetness, found that tillage treatments had lower or similar N_2O emissions from soil with good and moderate drainage, but no-till showed higher emissions than ploughing in poorly drained soil, which corroborates the conclusion that soil aeration at the depth of residue decomposition is a key factor regulating emissions. Song *et al.* (2019) found a strong relationship between soil O_2 and N_2O in soil profiles, but N_2O emissions will also depend on the potential for N_2O reduction during transport to the surface.

The accumulation of NO_3^- observed in the topsoil during autumn and winter (Fig. S3) was mainly derived from organic matter input in previous years, which would have a very different distribution in the soil profile of no-till and ploughed soil. As a consequence, the contact between decomposing residues and soil NO_3^- was probably better in ploughed soil where residues and fertilizers are incorporated each year, and this in turn may have enhanced N_2O emissions as shown by Taghizadeh-Toosi *et al.* (2021) in a manipulation experiment with mixed or layered distribution of crop residues, and two levels of, respectively, soil moisture and NO_3^- content.

Previous meta-analyses have documented that cover crops could reduce N leaching by 50-70% compared with bare fallow (Basche *et al.*, 2014; Valkama *et al.*, 2015). The present study indicated a reduction of 80-84%, and hence the effect was largely independent of tillage and residue management on this soil type, in accordance with previous results (Hansen *et al.*, 2015).

Soil conditions

Soil WFPS followed the same pattern in all treatments. During autumn and winter with above-average precipitation, the WFPS level at sampling was always close to 60%, which has been proposed to be a threshold for N_2O emissions based on controlled laboratory experiments (Aulakh *et al.*, 1991; Shelton *et al.*, 2000). The N_2O emissions remained low in this period despite some mineral N availability (Figs S2 and S3), although higher rates associated with rain events could have been missed. In contrast, significant N_2O emissions occurred during April and

May 2020 where soil WFPS was below 60% (Fig. S3), but mainly where fodder radish residues were returned. Together this indicates that, for the site investigated here, intense decomposer activity was more important as driver for N_2O emissions than soil drainage, in accordance with previous observations at this experimental site (Brozyna *et al.*, 2013; Pugesgaard *et al.*, 2017).

As reported by others, soil temperature during autumn-winter was higher with no-till (Yang *et al.*, 2018), straw retention (Chen *et al.*, 2007) and a cover crop (Kahimba, 2008). The cover crop maintained a positive effect on soil temperature after termination, possibly because cover crop residues promoted soil warming during spring due to a reduction in soil bulk density (Li *et al.*, 2015; Chalise *et al.*, 2019). Considering the importance of temperature for microbial transformations of C and N, such effects could influence N_2O emissions. Thus, Petersen *et al.* (2011) concluded that a large proportion of emitted N_2O was produced above a soil depth of 5 cm in both tilled and non-tilled soil after termination of a fodder radish cover crop. While there was a significant effect of temperature during autumn-winter (Table S3), N_2O emissions remained low independent of treatment in this period (Fig. 3); during spring, any effect of temperature was confounded by the main effect of cover crop.

Carbon footprints of conservation agriculture practices

The carbon footprints showed a GHG mitigation potential of direct seeding (no-till) and straw retention, and a neutral effect of cover crop, indicating that conservation tillage practices will have net benefits for the GHG balance of sandy loam soil in wet temperate climates. It should, however, be emphasized that N_2O emissions from main crops, i.e., fertilization of the previous oat crop, and decomposition of residues after harvest of oat and faba bean crops, were not included in the carbon footprints in Fig. 4, which represent effects of management outside the main crop growing period. For example, Petersen *et al*. (2011) quantified N_2O emissions in

PL+S and DS+S of the same long-term tillage experiment with and without fodder radish as winter cover crop, and here pig slurry was applied after termination of the cover crop. The combined N_2O emissions from residue and slurry decomposition reported by Petersen *et al.* (2011) corresponded to 0.7-1.6 Mg CO_2eq and thus were considerably higher than those from residues alone in the present study. Therefore, N_2O emissions after fertilization could outweigh the net benefits of the scenarios with straw retention reported in Fig. 4.

The largest component of the carbon footprint was the change in soil C associated with straw retention. It should be noted that cover crops had been part of all treatments in previous years (Goméz-Mũnoz *et al.*, 2021), and therefore the soil C change in Fig. 4 also included a contribution from cover crops. The soil C retention observed for fodder radish in 2019-2020 may indicate the relative importance of cover crops and straw retention, respectively, for the soil C stock changes. In a global meta-analysis, McClelland *et al.* (2021) estimated the annual C input from cover crops to be on average 0.21 Mg ha^{-1}. These authors also found that the response to cover crops was smaller in wet temperate compared to warmer climates, and that the apparent effect of cover crops on soil C stocks did not change with time since adoption. This has recently been corroborated by a long-term study in Denmark showing that the soil C effect of cover crops saturate after a period of approximately 15 years (Jensen *et al*., 2021). It indicates that the long-term retention of residue C in wet temperate climates is uncertain and, accordingly, that the retention of fodder radish C estimated from plant cuts in the present study, 0.23-0.43 Mg ha^{-1}, may be too high. If true, this would increase the relative importance of N_2O emissions for the GHG balance.

The use of a cover crop was highly effective in preventing NO_3^- leaching, which is a pollutant of recipient water bodies and an indirect source of N_2O, but the direct emissions of N_2O from cover crop residues were far greater than indirect emissions prevented in this study. This is in contrast to the results of a global meta-analysis of cover crop effects (Abdalla *et al.*, 2019) which concluded there was no significant effect of cover crops on N_2O emissions. The short duration (9 months) of the present study may have resulted in a higher relative importance of N_2O emissions associated with cover crop residues, but crop residues have also been identified as a main driver of N_2O emissions across entire four-year crop rotations on the light-textured soil investigated here (Brozyna *et al.*, 2013; Pugesgaard *et al.*, 2017). Thus, confounding effects of analysing field data across several soil types, climate zones and management practices may prevent the observation of effects that are seen with a greater disaggregation of the data (Chen *et al.*, 2013). As GHG mitigation strategy, the fodder radish crop in the present study depended on the concurrent C storage which, however, will be less permanent than N_2O in the atmosphere. Direct seeding resulted in less N_2O emission than spring tillage, and the combination of GHG mitigation methods under conservation agriculture should be further explored.

Conclusion

This study investigated effects of tillage practice, straw retention, and a winter cover crop on N_2O emissions and NO_3^- leaching, during the cover crop growth period and subsequent decomposition in spring. The study did not find effects of straw retention on nitrate leaching or N_2O emissions, but straw retention was critical for the overall GHG balance through soil carbon storage, and without straw retention after harvest there was always a positive GHG balance, i.e., net emissions. As expected, fodder radish was highly effective in reducing NO_3^- leaching during winter, but N_2O emissions after spring cultivation were enhanced; the increase in direct N_2O emissions could in part be compensated by storage of residue C and by the adoption of direct seeding. Even though N losses associated with a main crop were not covered in this study, the results indicated that direct seeding, straw retention and the use of cover crops could substantially improve the carbon footprint of agroecosystems on sandy soil in a wet temperate climate.

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Table 1. Field management during April 2019 to May 2020.

Date	Field operation
12 August 2019	Harvest of oat
19 August 2019	Spreading of fodder radish seeds (14 kg ha^{-1}) in +CC plots
19 November 2019	Spraying with glyphosate in Glyphomax HL, 1.5 l ha^{-1} in –CC plots (bare soil) [1]
14 March 2020	Spraying with glyphosate in Glyphomax HL, 2 l ha^{-1} (herbicide)
30 March 2020	Ploughing, PL plots
31 March 2020	Sowing of faba bean (228 kg ha^{-1})
6 May 2020	Fertilisation with PK 0-4-10 (800 kg ha^{-1})
20 May 2020	Spraying with Fighter 480, 1 l ha^{-1} (herbicide), Profi Mn nitrate 235, 1.5 l ha^{-1} and Contact, 0.2 l ha^{-1} (additive).

[1] All four tillage and straw treatment combinations

Table 2. An overview of treatments and variables that were measured in the experiment.

Main plot	*Subplots*	*Sub-subplots*	*Key*
Straw removal (-S)	Ploughing (PL)	Cover crop (+CC)	PL-S+CC
		No cover crop (-CC)	PL-S-CC
	Direct seeding (DS)	Cover crop (+CC)	DS-S+CC
		No cover crop (-CC)	DS-S-CC
Straw retention (+S)	Ploughing (PL)	Cover crop (+CC)	PL+S+CC
		No cover crop (-CC)	PL+S-CC
	Direct seeding (DS)	Cover crop (+CC)	DS+S+CC
		No cover crop (-CC)	DS+S-CC

Measurement variables

N_2O and CO_2 emissions
Soil mineral N dynamics
NO_3^- leaching
Soil moisture and temperature

Table 3. Analysis of variance (ANOVA) of temporal N_2O emissions. Significance of *p* values: *** < 0.001, ** < 0.01, * < 0.05, non-significance (ns) > 0.05.

	Before spring tillage				After spring tillage			
Treatments	numDF	denDF	*F*	*p*	numDF	denDF	*F*	*p*
Intercept	1	244	26224	***	1	178	3898	***
Tillage	1	244	0.022	ns	1	178	7.94	**
Straw	1	244	0.91	ns	1	178	0.062	ns
Cover Crop	1	244	12.67	***	1	178	72.78	***
Date	11	244	3.32	***	8	178	8.84	***
Tillage*Straw	1	244	0.29	ns	1	178	0.14	ns
Straw*Cover Crop	1	244	1.09	ns	1	178	1.2	ns
Tillage*Cover Crop	1	244	4.23	*	1	178	0.41	ns

Table 4. Cumulated emissions of N_2O, and of NO_3^- leaching, between October 2019 and June 2020. The experimental treatments were: Ploughing (PL); direct seeding (DS); straw retention (+S); straw removal (-S); growth of fodder radish as a winter cover crop (+CC); no cover crop (-CC).

Treatments	Cumulative loss (kg N ha^{-1})	
	N_2O-N	NO_3-N
PL-S-CC	0.19 ± 0.18	27.7 ± 3.3
PL-S+CC	0.56 ± 0.07	5.0 ± 0.8
DS-S-CC	0.07 ± 0.01	25.3 ± 3.9
DS-S+CC	0.35 ± 0.06	4.5 ± 1.9
PL+S-CC	0.06 ± 0.03	20.9 ± 1.2
PL+S+CC	0.78 ± 0.18	4.3 ± 0.3
DS+S-CC	0.04 ± 0.02	22.2 ± 0.7
DS+S+CC	0.41 ± 0.14	3.6 ± 1.1

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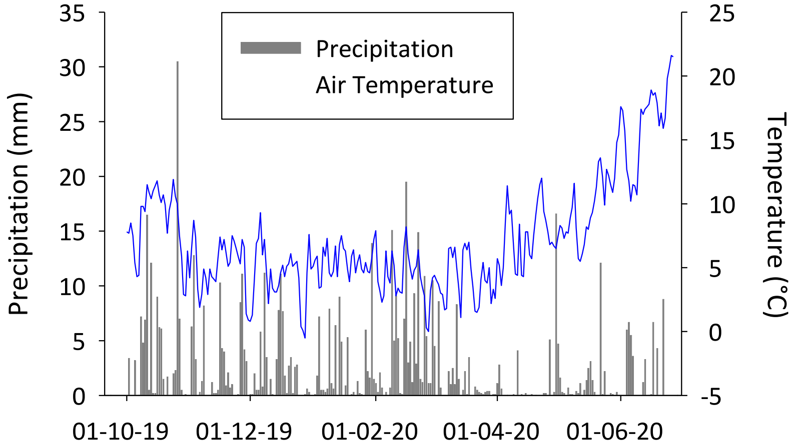

Figure 1. Daily average air temperature and precipitation during experimental period. Note: Blue line shows air temperature, and grey bars show Precipitation.

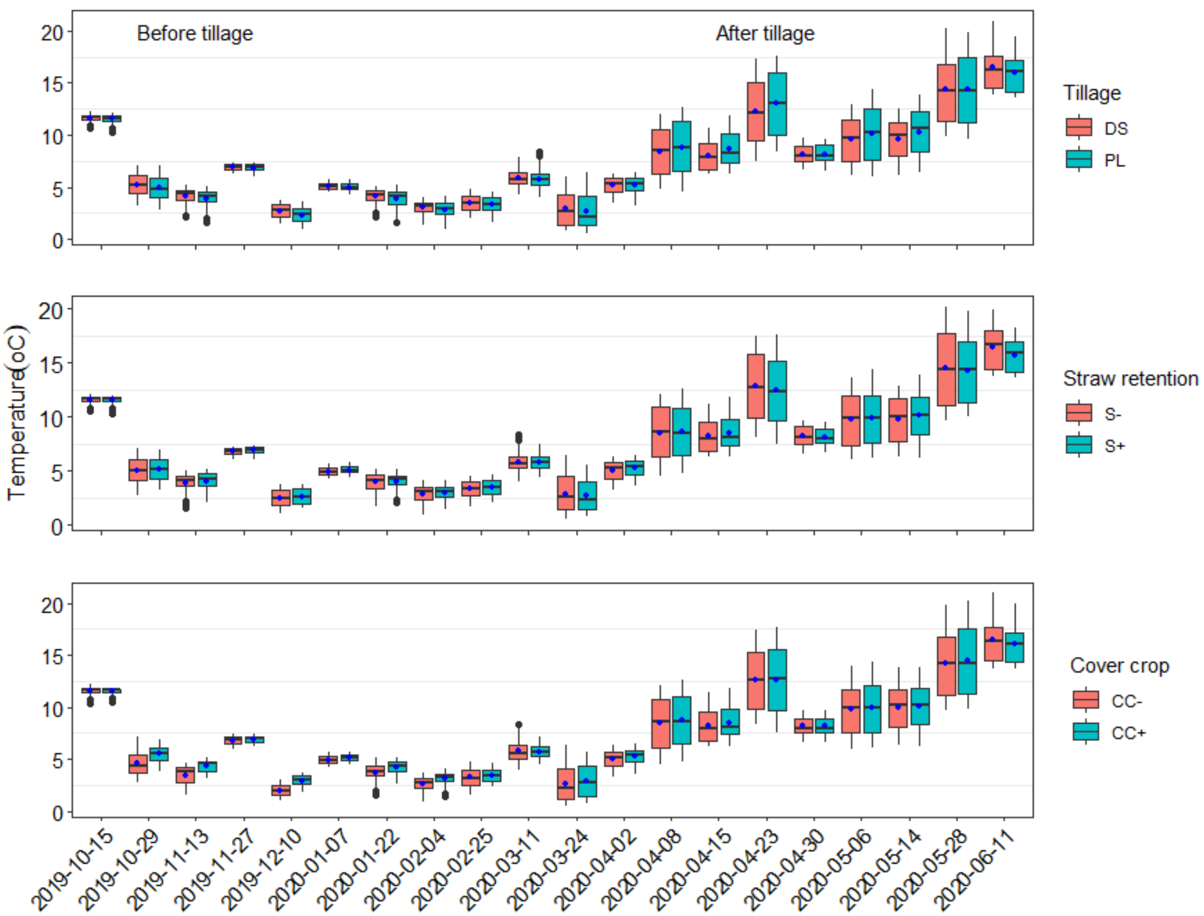

Figure 2. Average soil temperature at 8 cm below the soil surface in different treatments, which include tillage (PL: ploughing, DS: direct seeding), residue management (S+: straw retained, S-: straw removed), cover crops (CC+: with cover crop, CC-: without cover crop). Tillage was performed on 03 April 2020. Middle blue dots and solid lines of the boxplot represent mean and median values respectively. Whiskers represent minimum and maximum temperature at the particular dates of the measurements. Black dots represent extreme values which were outside the range. Error bars are standard errors of mean (s.e.m.).

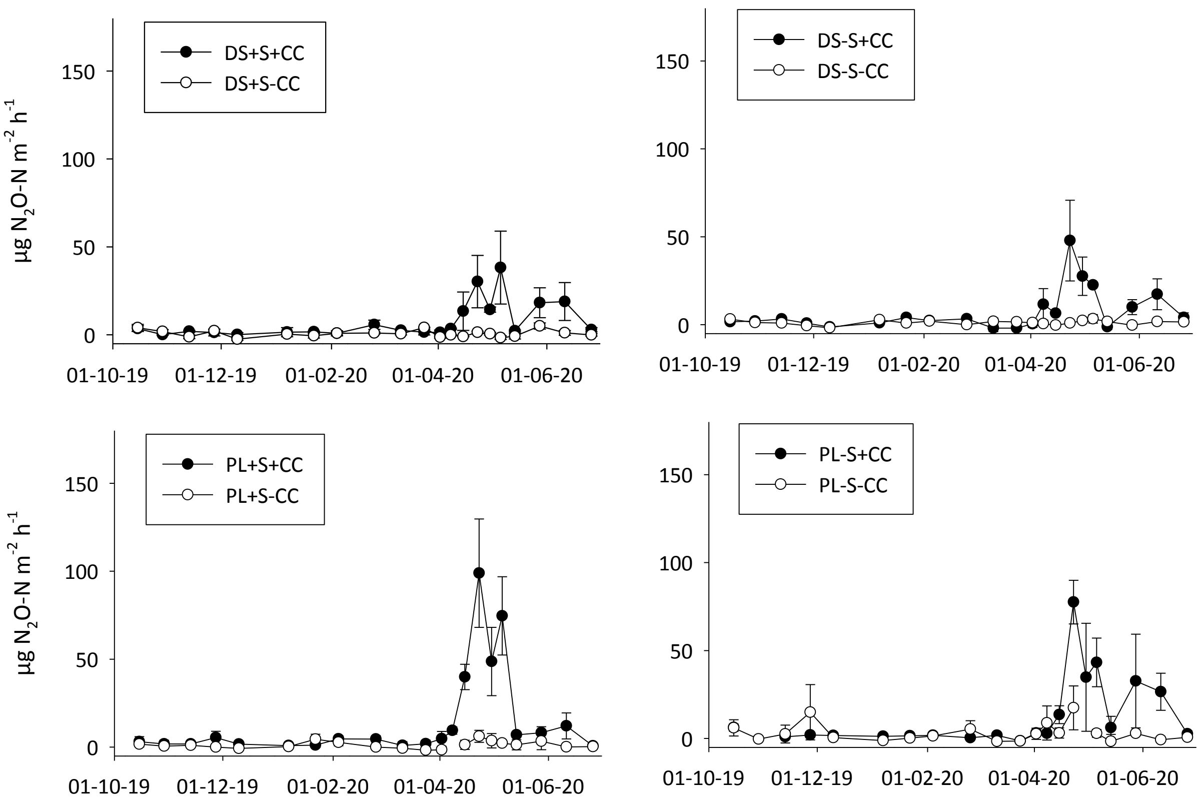

Figure 3. Nitrous oxide (N_2O) emissions from different treatments determined in the all treatments during October 2019 to July 2020 (error bars represent s.e.m.; n = 3) different treatments, which include tillage (PL: ploughing, DS: direct seeding), residue management (S+: straw retained, S-: straw removed), cover crops (CC+: with cover crop, CC-: without cover crop). On 14th March, the cover crop were sprayed with glyphosate, and on 30th March the cover crop residues were ploughed in PL treatments. Error bars are standard errors of mean (s.e.m.).

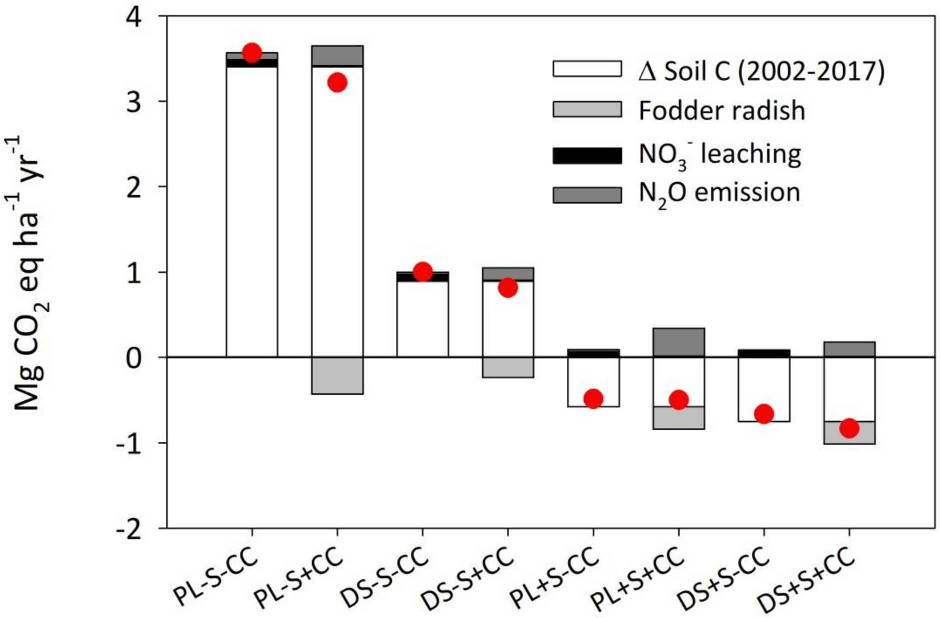

Figure 4. Net greenhouse gas balance (red filled circles) of the eight combinations of ploughing (PL) *vs.* direct seeding (DS), straw retention (+S) *vs.* straw removal (-S), and growth of fodder radish as a winter cover crop (+CC) *vs.* no cover crop (-CC). Positive effects indicate net emission of greenhouse gases from the soil. Nitrous oxide emissions and NO_3^- leaching was measured between October 2019 and June 2020. The changes in carbon stock represent the long-term retention after around 20 years.

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Supplementary Material
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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.

Authorship contribution statement

Arezoo Taghizadeh-Toosi: Writing of original draft, investigation, formal analysis, visualization, revieing, editing, final submission.

Khagendra R. Baral: Statistical analysis, formal analysis, editing.

Søren o. Petersen: Writing, Reviewing, editing, Investigation.

Elly M. Hansen: Nitrate leaching calculations, reviewing, and editing.

Jørgen E. Olesen: Reviewing, editing, and funding acquisition.