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Incorporation and harvest management of hairy vetch-based green manure influence nitrous oxide emissions

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Abstract

In this study, we measured nitrous oxide (N_2O) emissions from plots of fall-planted hairy vetch (HV, *Vicia villosa*) grown as a green nitrogen (N) source for following summer forage crabgrass (*Digitaria sanguinalis*). Two treatments were compared: (i) HV grown solely as green manure where all biomass was incorporated by tillage, and (ii) harvesting of aboveground HV biomass prior to planting of crabgrass. Fluxes of N_2O were measured with closed chamber systems on 27 dates during a 2-month growth period of crabgrass after the termination of HV in early May. At termination, the average aboveground biomass yield of HV was 4.6 Mg ha^{-1} with 146 kg N ha^{-1} content. The N_2O emissions were as high as 66 g N_2O-N ha^{-1} day^{-1} on day 1 after HV incorporation, but reached close to zero within a week. Emissions of N_2O increased with subsequent rainfall and irrigation events from both treatments but emission peaks were not observed during the rapid growth of crabgrass. Two-month cumulative emission of N_2O (mean ± s.e., $n = 4$) from HV incorporated plots (921 ± 120 g N_2O-N ha^{-1}) was three times ($P < 0.05$) of HV harvested plots (326 ± 30 g N_2O-N ha^{-1}). However, crabgrass biomass yields, N concentrations and total biomass N uptake were decreased significantly by harvesting HV. In conclusion, the results suggested that whereas removal of HV biomass for use as forage may significantly reduce N_2O emissions, quantity and quality of the following recipient crops may be constrained.

Introduction

The cultivation of legume-based cover crops as green sources of nitrogen (N) is practiced in many parts of the world, including the US Southern Great Plains (SGP) (Tonitto *et al.*, 2006; Bergtold *et al.*, 2019; Kandel *et al.*, 2018). Summer crops such as corn (*Zea mays*), sorghum (*Sorghum bicolor*) and annual summer grasses are important components of agriculture in the region. These crops are generally cultivated from June to September, and have long fallow periods (October through May). Winter hardy annual legumes such as hairy vetch (HV, *Vicia villosa*) can be cultivated as green manures in these production systems (Kaufman *et al.*, 2013; Kandel *et al.*, 2018). High yielding winter legumes cultivated in the SGP may fix enough atmospheric N in biomass to meet the requirements of following summer crops (Tonitto *et al.*, 2006; Kandel *et al.*, 2018). The biomass N of winter legumes can be effectively transferred to the following summer crop after soil incorporation due to generally low C/N ratio of biomass. Such a low ratio is a conducive biomass trait for rapid decomposition and N mineralization after termination (Rosecrance *et al.*, 2000; Melkonian *et al.*, 2017). However, rapid decomposition and mineralization of biomass N can also contribute to elevated emissions of N_2O which is a highly potent greenhouse gas (Huang *et al.*, 2004; Basche *et al.*, 2014). An Intergovernmental Panel on Climate Change (IPCC) estimate suggests that on average 1% of N in biomass residue is converted into N_2O (IPCC 2007). However, the emission factor for legume-based green manures can be higher due to rapid decomposition and N mineralization rates, compared to non-legume residues (Sanz-Cobena *et al.*, 2014).

Denitrification is a major microbial process that contributes N_2O emissions from soil incorporated biomass residues (Li *et al.*, 2016). In denitrification, nitrate (NO_3^-) or nitrite (NO_2^-) are reduced to N_2 through intermediate products of nitric oxide (NO) and N_2O. When legume green manures are incorporated into the soil, rapid decomposition and mineralization may elevate concentrations of soil NO_3^- which can be eventually used by denitrifying bacteria in moist soil condition and emit large amounts of N_2O to the atmosphere. A recent field study in the US SGP demonstrated large emissions of N_2O during rainfall events following soil incorporation of HV resulting in cumulative emissions of 19.3 kg N_2O-N ha^{-1} within a month after soil incorporation (Kandel *et al.*, 2018). Although growth and yield of the

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following summer crop (crabgrass; *Digitaria sanguinalis*) were improved by HV green manure, its application did not have a positive atmospheric impact due to the high rates of N_2O emissions. The emissions were particularly high prior to the establishment of crabgrass, but remained close to zero during periods of rapid growth of crabgrass, even immediately after rainfall events (Kandel *et al.*, 2018). Thus, the application of management techniques that can decrease soil concentrations of mineral N during periods between the termination of legume green manures and establishment of the following recipient crops can be crucial to mitigating large emission of N_2O.

One potential management technique to limit N_2O emissions from legume green manure residues would be the removal of aboveground biomass for use as forage prior to soil incorporation (Shrestha *et al.*, 1999; Rao and Northup, 2008). Such a decision on biomass harvesting can be developed based on total biomass N, potential biomass N mineralization rates and the expected N requirements for following recipient crops (Kandel *et al.*, 2018). The harvested legume biomass could lower the risk of forage shortage for cattle in dry years, which are common in the SGP (Holman *et al.*, 2018). However, the harvest of biomass of legumes cultivated as green manures may significantly change the availability and turnover of N in the agroecosystems (Li *et al.*, 2015). Allocation of biomass by annual legumes to above and belowground components is not uniform, particularly for cool-season species. Biederbeck *et al.* (1993) reported that a collection of legumes allocated <20% of whole plant biomass to belowground components during a 6-yr study. Such an allocation pattern indicates that biomass removal would remove a major portion of N from the ecosystem. Therefore, such management may not provide enough N to support the growth of the following crop, unless the exported N is returned as manure or replenished with inorganic fertilizers (Singer and Meek, 2004). Nevertheless, the influence of harvesting biomass of winter legumes cultivated as green manure on N_2O emissions mitigation, and productivity and N concentration of the biomass of summer forages is not clear.

This study reports findings from a field experiment where fall-planted HV was either harvested or incorporated into the soil in early May prior to planting of crabgrass grown for hay production. The objective was to compare the effects of two management strategies of HV residues on N_2O emissions during the growing season of the following summer crop; (1) soil incorporation of whole HV biomass as green manure, and (2) harvesting of aboveground HV biomass for forage. We hypothesized that emissions of N_2O would be lower due to harvesting aboveground biomass of HV compared to incorporating it into the soil before planting of a summer crop.

Materials and methods

Study site and soil properties

This study was conducted at the USDA-ARS Grazinglands Research Laboratory (35°40′N, 98°00′W) near El Reno, OK, USA (Kandel *et al.*, 2018). The study site was situated on the side slope of a heavily degraded old field adjacent to a permanent stream. The site was historically (1950–2011) planted to different annual crops and heavily grazed by cattle during parts or all of many calendar years dating back to 1950. The predominant soil series was classified as Norge silt loams (fine, mixed, thermic, Udic Ustochrepts) with 3–5% slopes. The topsoil (0–0.15 m) sand, silt and clay contents were 37, 41 and 22%, respectively.

Experimental design and crop management

This study was conducted during 2017–2018 within a long-term field experiment that was initiated in 2014. The long-term (multi-decade) study was developed to examine the function of fall- and spring-planted legumes as green sources of N to support a forage grass during summer, within low-input management systems similar to those applied by landholders with limited resources (Ledgard, 2001; Bartholomew, 2015). A series of annual legumes were planted in sets of plots in fall or spring and the effects were compared to those of plots that were unfertilized or received a common rate of inorganic N. The current study utilized plots planted to HV in autumn, grown through the winter and spring, and incorporated into soil as a green manure in May to provide N for the following crabgrass. In 2017, HV was planted (seed rate: 20 kg ha^{-1}) in mid-September after the harvest of crabgrass in late-August. Prior to planting, HV seeds were inoculated with a *Bradyrhizobium* strain. HV received 45 kg P ha^{-1} as dry diammonium phosphate (DAP; 18% N and 46% phosphate) at planting. The lack of availability of P fertilizer without N for producers in the region necessitated the use of DAP. Crop management of HV during the period of this experiment was similar to activities applied in the previous years (2014–2016) of the long-term study.

Measurement of the influence of management strategies applied to residues of HV on emissions of N_2O was conducted in 2018. The study consisted of four replicate blocks (6 m × 6 m) within each of two landscape positions (shoulder and toe slopes) of the site. The blocks were planted to HV in mid-September 2017, and terminated on 2 May 2018. To impose the harvest treatment, HV biomass from 2 m × 3 m areas of each block was removed before termination. HV biomass on the remaining area of blocks was terminated on the same date by shredding with a flail mower. Both harvested and shredded areas of blocks were then tilled (disked once to ~10 cm depth and roto-tilled once). Crabgrass was then planted (seed rate: 5 kg ha^{-1}) on the same day with a row spacing of 0.03 m. Two replicate plots (2 m × 3 m) in each block from where HV was shredded were assigned for incorporation treatment. Thus, each block consisted of three experimental plots with two management treatments in a randomized block design. Large spot-specific heterogeneity of N_2O emissions was anticipated due to spot-specific differences in response to biomass termination and incorporation (Kandel *et al.*, 2018). Therefore, two replicate plots were assigned to each incorporation treatment. All measurements from the plots where HV biomass was incorporated in each block were subsequently averaged prior to data analysis and comparisons to the harvest treatment.

A long dry spell (>1 month) started 2 weeks after planting of crabgrass. This period restricted the growth of crabgrass, and plants showed severe symptoms of water shortage. Therefore, the plots were irrigated with 30-mm water on day 35, 43 and 47 after the termination of HV. The entire area of the plots, except that covered by collars placed to measure N_2O fluxes (described in the following section), was irrigated with a sprinkler system. Irrigation inside collars was subsequently applied using a rose can for uniform application.

Measurement and calculation of N_2O fluxes

A detailed description of methods used in flux measurements is presented in Kandel *et al.* (2018). In brief, fluxes of N_2O were measured with a closed chamber system from 3 May to 30 June

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2018. A set of PVC collars (0.65 × 0.65 m) were inserted to 0.10 m depth in each plot (total $n = 12$) immediately after field operations on 2 May. All three collars in a block were placed in a line with the same elevation. The collars had a 0.04 m wide outer flange that remained parallel to the soil surface to support the chamber used for flux measurements. Fluxes were measured by placing a white-colored chamber (0.70 × 0.70 × 0.21 m) on the preinstalled support collars. Flux measurements were taken more frequently (often daily) during and after rainfall and irrigation events to capture rainfall/irrigation induced emissions. Less frequent (longest interval 7 days) measurements were taken during dry spells when emissions remained close to zero. During measurements, headspace air was mixed by two small fans, which ran continuously while chambers were enclosed. The chamber was connected to a portable Fourier transform infrared-based analyzer (DX4040; Gasmet Technology Oy, Helsinki, Finland[1]). Air in the chamber headspace was circulated through 3 mm inlet and outlet tubing to the gas analyzer. Concentration of N_2O was measured at 20-s intervals during 6–8 min enclosures for each measurement. Flux measurements were taken between 10:00 and 12:00.

Fluxes were calculated by linear regression (Kandel *et al.*, 2016a) using the MATLAB® (MathWorks, Inc., Natick, MA, USA[1]) routine developed by Kutzbach *et al.* (2007). The first few records after chamber enclosure were discarded as dead-band based on visual inspection of the CO_2 flux curve which was also measured simultaneously with N_2O flux. Total cumulative emissions of N_2O during the measurement period were calculated using linear interpolation of measured fluxes between the measurement dates.

Measurement of CO_2 fluxes

Although the main interest of this study was N_2O emissions, fluxes of CO_2 were also measured simultaneously with N_2O fluxes, and reported to define the contribution of decomposing residues to N_2O emissions. The CO_2 fluxes measured with an opaque chamber represent ecosystem respiration (i.e., flux from ecosystem to atmosphere) with contributions by both living plants and soils. However, the study plots were free of any green plants for a week prior to germination of crabgrass. Therefore, the CO_2 fluxes during this initial period represent heterotrophic soil respiration. Collar-specific cumulative emissions of CO_2 obtained by the linear interpolation method were correlated to cumulative N_2O emissions.

Measurements of environmental variables

Volumetric water content (VWC) of the surface soils (0–0.15 m) was measured continuously at a plot using EC-5 soil moisture sensors (METER Environment, Pullman, WA, USA[1]). A sensor was inserted at each 0–0.05, 0.05–0.10 and 0.10–0.15 m soil depth and an average of three sensors is presented. The VWC were expressed in terms of relative VWC contents to the maximum VWC recorded at saturated field conditions, which represents water-filled pore space (WFPS) equal to 100% (Kandel *et al.*, 2018).

Soil temperature was recorded continuously at 1-h interval in the same plot used for VWC measurements. Three soil sensors (TMC-6; Onset Computer Corporation, Bourne, USA[1]) were placed at 0.05, 0.10 and 0.15 m soil depths, and the average is presented. Hourly air temperature during chamber enclosure and precipitation data for the study period were obtained from a weather station (Oklahoma Mesonet, Oklahoma Climatological Survey) located roughly 1 km from the study site.

Analyses of soil samples

Soil samples (0–0.15 m) were taken from all plots on 26 out of 27 dates of flux measurements to define concentrations of the two main fractions of mineral N (NO_3^- and NH_4^+), soil pH and electrical conductivity (EC). Two soil cores (diameter, 0.02 m) were taken at about 0.10 m distance from opposite sides of the collars at each soil sampling. The two cores were pooled to form a composite sample for analysis. Aliquots of samples were extracted in 1.0 M KCl and analyzed by flow injection (Timberline Instruments, Boulder, CO, USA[1]) to determine the concentrations of nitrate (NO_3^-) and ammonium (NH_4^+) N. Soil pH and EC were determined in 1:2 soil:water solution by a benchtop pH/conductivity meter (Orion Star A215; Thermo Scientific, Waltham, MA, USA[1]). Total organic C and total N concentrations in soil were determined from soil samples collected on 2, 36 and 59 days of HV termination. Samples were assessed by auto-analyzer at 900 °C for 10 min (Model VarioMacro, Elementar Americas, Inc., Mt. Laurel, NJ, USA[1]), and analyzing gases evolved from the samples.

In addition to the soil samples taken simultaneously with flux measurements, soil samples were also taken prior to HV termination on 2 May (day 0). Since the residue management treatments were not imposed at soil sampling on 2 May, one soil sample (pool of three cores) was taken from each block to represent the soil characteristics under both treatments. Therefore, the results from soil samples taken prior to the termination of HV on 2 May were not used for comparisons between the treatments.

Measurements of yield and quality of hairy vetch

Total aboveground biomass of HV was determined by drying all biomass from 6-m^2 plots ($n = 4$) of harvest treatment. The biomass was dried at 60 °C to constant weight in a forced-draft oven. A fraction of dried biomass was milled to pass through a 1-mm sieve to determine biochemical compositions. Total C and N concentrations were assessed by flash combustion (900 °C for 10 min) instrument (Model VarioMacro[1]). Amounts of N per hectare in HV biomass was obtained as a product of biomass yield and N concentrations in biomass. Neutral detergent fiber (NDF), acid detergent fiber (ADF) and acid detergent lignin (ADL) in samples were determined by procedures outlined for a batch fiber analyzer (Fairport, NY; https://www.ankom.com/analytical-methodssupport/fibre-analyzer-a2000). Concentration of cellulose was estimated as the difference between ADF and ADL, and hemicellulose as the difference between NDF and ADF. The ADL fraction was considered as lignin concentration. Biochemical composition was determined on three replicates and averages are presented.

Six soil cores (0–0.25 m) containing individual HV plant were excavated outside the study plots with a shovel to determine root/shoot ratios. The plants were partitioned to shoot and root biomass. Root biomass was cleaned manually, and root and shoot

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[1] Mention of trade names or commercial products in this publication is solely for the purpose of providing specific information and does not imply recommendation or endorsement by the US Department of Agriculture.

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were oven-dried at 60 °C to constant weight, and dry weights were recorded separately. Total root biomass per hectare was determined from the average root/shoot ratio and shoot biomass yield determined from 6-m^2 areas in harvest treatment. The pooled root biomass from the six plants was milled and chemical compositions were analyzed as similar to the shoot biomass.

Measurements of plant growth, yield and quality of crabgrass

Growth of crabgrass was monitored non-destructively by taking canopy reflectance measurements inside the collars using a full range PSR-3500 portable spectro-radiometer (Spectral Evolution, Lawrence, USA[1]) with a spectral range of 350–2500 nm. Canopy reflectance at red and near infrared (NIR) wavelengths (656 and 779 nm, respectively) was used to calculate the ratio vegetation index (RVI) as the ratio of reflectance at NIR and red wavelengths (Kandel *et al*., 2016b). Total aboveground biomass of crabgrass was measured inside the collars on 29 June, a day prior to the last flux measurement. All biomass inside the collars (0.42 m^2) was harvested manually and oven-dried at 60 °C to constant weight. Concentration of N and total N uptake per hectare in crabgrass biomass were determined with a similar method used for HV biomass.

Statistical analysis

Measurements are presented as averages of four plots per residue management treatment and standard errors denote spatial variations in responses unless stated otherwise. The two blocks at each slope position were at slightly different elevations (about 1 m), and therefore, treated as individual units rather than replications within a slope position. The difference of cumulative fluxes of the incorporation and harvesting treatments were determined using a mixed model in SAS (SAS Inc., Cary, NC, USA[1]) considering blocks as a random variable. The effect of sampling dates was included in the model and treated as repeated measurements for the measured dynamic variables. The compound symmetry covariance structure was applied to account for auto-correlation among dates of repeated measurements. Fisher's Least Significant Difference (LSD) method was used for pairwise comparisons at 5% level.

Relative importance of soil variables (soil moisture, concentrations of NO_3^- and NH_4^+, pH and EC) on dynamics of N_2O emissions was tested using PROC HPFOREST in SAS. Averages of individual soil variables and N_2O emissions at 26 measurement dates were used for the test. Additionally, statistical correlations of the soil variables to N_2O emissions were carried out using a Pearson correlation matrix. Pearson correlation (R) was also used to show the relationship between 7 days cumulated CO_2 and N_2O emissions from individual collars after HV termination.

Results

Yield and properties of hairy vetch biomass

At termination, HV produced 4.6 Mg ha^{-1} shoot biomass which contained 146 kg N ha^{-1} (Table 1). Root biomass comprised 5.5% of the total biomass present, indicating root biomass approximated 0.3 Mg ha^{-1} with 6.1 kg N ha^{-1}. Concentrations of N in HV shoots were 3.2%, while it was 2.3% in root biomass. Ratio of C/N in HV shoots was 14.3, and higher in roots (22.2). Concentrations of cellulose and hemicellulose in shoot and root biomass were similar but lignin concentration in roots was higher than in the shoot.

Table 1. Yield and properties of hairy vetch biomass at termination

Parameters	Shoot	Root
Yield (Mg ha^{-1})	4.6	0.3
N concentrations (% of DM)	3.2	2.3
C concentrations (% of DM)	46.0	51.2
C/N	14.3	22.2
Total N (kg ha^{-1})	146.4	6.1
Cellulose (% of DM)	29.4	27.6
Hemicellulose (% of DM)	17.0	15.5
Lignin (% of DM)	12.1	18.1

Climate and soil conditions during study period

Air temperature during flux measurements ranged between 9 and 25 °C and soil temperature ranged between 15 and 35 °C (Fig. 1a). The average air temperature at the study site in May was 23.2 °C, which was 3.0 °C higher than the long-term (1981–2010) average of 20.2 °C. Similarly, the average air temperature in June was 26.2 °C, being 1.5 °C higher than the long-term average of 24.8 °C.

Total precipitation in May (50 mm) was about 74 mm lower than the long-term average, while total precipitation in June (93 mm) was 40 mm lower than the long-term average. A total of 15 mm rainfall was recorded prior to the first flux measurement on 3 May, after the soil incorporation of HV on 2 May. Additional 10 and 15-mm rainfalls were recorded in the second and third weeks after termination of HV (Fig. 1b). A total of 86-mm rainfall was received between days 51 and 54 of HV termination.

Soil moisture (presented as WFPS) was high during the first measurement taken 1 day after termination of HV on 3 May but gradually decreased until subsequent rainfall events (Fig. 1b). Soil moisture was increased after irrigation events and reached up to saturation during heavy rainfall events in late June and then decreased.

Dynamics of CO_2 emissions

Emissions of CO_2 from HV incorporated plots were relatively high (0.25 Mg CO_2-C ha^{-1} d^{-1}) on day 1 after incorporation, then declined gradually with decreasing soil moisture (Fig. 1c). However, emission rates increased after rainfall on day 12 after biomass incorporation. Emissions of CO_2 from HV incorporated plots remained higher than the HV harvested plots during the early period of HV termination. Thus, cumulative emission of CO_2 by 7 days after the termination of HV (when no contribution of autotropic plant respiration occurred) from biomass-incorporated plots (1.04 Mg CO_2-C ha^{-1}) was 2.15 times higher than cumulative CO_2 emission from HV biomass-harvested plots (0.49 Mg CO_2-C ha^{-1}). The 7-day cumulative emission derived from decomposing HV shoot biomass calculated as the difference in cumulative emissions from HV incorporated and HV harvested plots (0.56 Mg CO_2-C ha^{-1}) was about 26% of total C in aboveground HV biomass (2.13 Mg C ha^{-1}).

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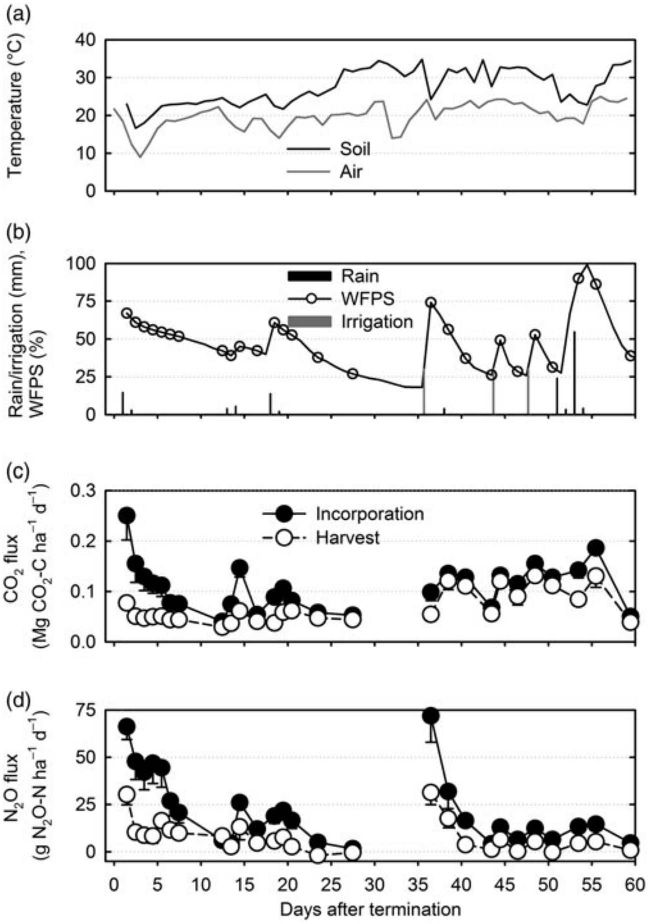

Fig. 1. (a) Average air and soil temperatures during flux measurement, (b) dynamics of daily precipitation and water-filled pore space (WFPS) measured at 0–0.15 m soil depth during flux measurement. Dynamics of (c) CO_2 and (d) N_2O emissions. Error bars (c–d) represent the spatial variations at the plot scale (s.e., $n = 4$). Unidirectional error bars are shown for clarity.

Dynamics of N_2O emissions

Similar to emissions of CO_2, emissions of N_2O from HV incorporated plots were high (66 g N_2O-N ha^{-1} d^{-1}) on the first day after soil incorporation, then declined gradually with decreasing WFPS (Fig. 1d). The emissions increased with increasing soil moisture after rainfall events and showed similar dynamics as CO_2 emissions before the rapid growth of crabgrass. Rainfall-enhanced N_2O emissions were also observed from HV harvested plots but magnitudes were smaller. The first irrigation event on day 35 after HV termination enhanced N_2O emissions to 72 and 36 g N_2O-N ha^{-1} d^{-1} from HV incorporated and harvested plots, respectively. The two subsequent irrigation events on days 43 and 47 after HV termination, however, did not enhance N_2O emissions to this level. Although soil moisture was close to saturation at flux measurements taken at day 55 and 57 after HV termination, the emissions of N_2O did not increase considerably from either treatment. Average rates of N_2O emissions from the HV incorporated plots across the measurement dates (21.8 g N_2O-N ha^{-1} d^{-1}) were significantly higher than that from the HV harvested plots (8.0 g N_2O-N ha^{-1} d^{-1}). Effects of soil sampling dates and interaction effects between treatments and the dates were also significant.

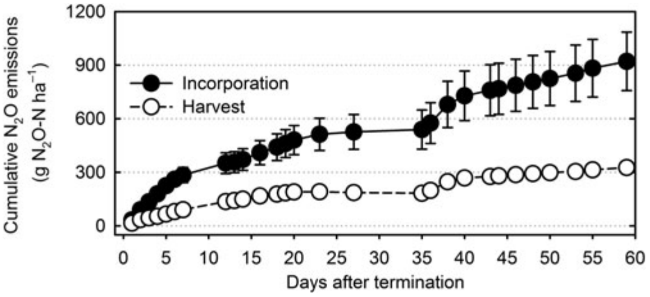

Fig. 2. Cumulative estimates of N_2O emissions during the 2-month study period. Error bars represent the spatial variations at the plot scale (s.e., $n = 4$). The cumulative emissions from hairy vetch incorporated treatment were significantly higher hairy vetch harvested treatment ($P < 0.05$).

Cumulative N_2O emissions

The cumulative N_2O emission during the 2-month study period from HV incorporated plots (921 ± 120 g N_2O-N ha^{-1}) was significantly higher than emissions from HV harvested plots (326 ± 31 g N_2O-N ha^{-1}) (Fig. 2). Since the rates of N_2O emissions were high during the early stage of termination of HV biomass (Fig. 1d), a total of 31% of cumulative emissions was observed within the first week in HV incorporated plots. The 7-day cumulative emissions from HV harvested plots also had similar (28%) contribution to total 2-month cumulative emissions. Although large amounts of rainfall were observed during the last week and WFPS remained high, cumulative emissions during this period contributed only 7% to total emissions in both treatments. Cumulative CO_2 emissions from individual collars were strongly correlated ($R = 0.95$) to cumulative N_2O emissions during the first week after termination of HV, when there was no contribution from plant respiration to CO_2 fluxes and no crop uptake of soil N (Fig. 3).

Dynamics of soil organic carbon and total nitrogen

Concentrations of soil organic carbon (SOC) in 0–0.15 m soil depth increased rapidly from 1.17% at pre-termination to 1.38% after 2 days of termination in the HV incorporated plots (Fig. 4a). However, the concentration increased only slightly (1.17–1.25%) in the HV harvested plots. Thereafter, concentrations decreased from both treatments and reached to levels slightly lower than pre-termination (1.13% in HV incorporated and 1.08% in HV harvested plots) within 2 months of HV termination. Average SOC concentration measured on three dates after HV termination in the incorporated plots (1.25%) was not significantly higher ($P = 0.056$) than that in the harvested plots (1.18%). Effect of soil sampling dates on SOC concentrations was significant since the concentrations decreased from both treatments over time. However, interaction effects between treatment and the dates were not significant.

Concentrations of total N in 0–0.15 m soil depth increased only slightly from 0.167% at pre-termination to 0.173% after 2 days of termination in the HV incorporated plots but remained unchanged in HV harvested plots (Fig. 4b). Thereafter, concentrations decreased from both treatments and reached to levels lower than the pre-termination (0.114% in HV incorporated and 0.105% in HV harvested plots) within 2 months of HV termination. Average N concentration measured on three dates after HV termination in the incorporated plots (0.138%) was not

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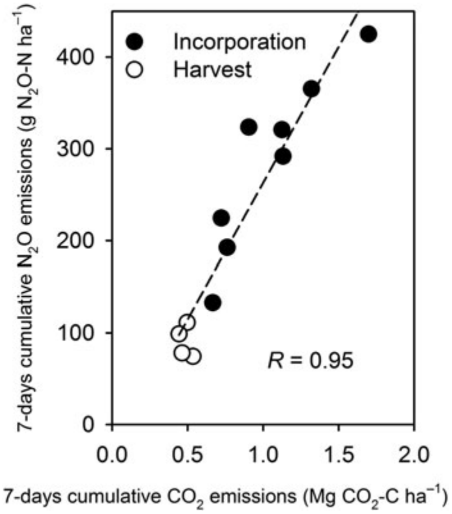

Fig. 3. Correlation of cumulative CO_2 and N_2O emissions during the first week of the termination of hairy vetch biomass. The fitted linear relations represent data for both residue management treatments. Pearson's correlation coefficient (*R*) between the cumulative emissions is shown.

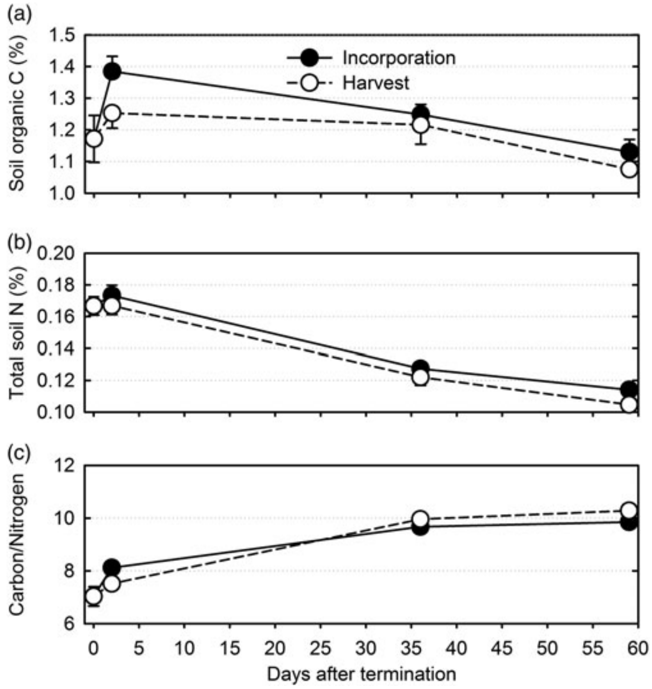

Fig. 4. Dynamics of (a) soil organic carbon (C), (b) total nitrogen (N) and (c) ratio of C and N in the 0–0.15 m soil depth. Error bars represent the spatial variations at the plot scale (S.E., $n = 4$). Unidirectional error bars are shown for clarity.

significantly ($P = 0.052$) higher than the harvested plots (0.131%). Effect of soil sampling dates on total N concentrations was significant since the concentrations decreased from both treatments over time. However, the interaction effect between treatments and the dates was not significant.

Ratios of soil C and N in 0–0.15 m soil depth increased slightly after HV termination in both treatments since soil C concentrations increased more rapidly than soil N concentrations (Fig. 4c). Thereafter, C/N ratios increased slightly in both treatments. Average soil C/N ratios measured on three different dates after HV termination in the HV incorporated plots (9.21) was similar to the harvested plots (9.25). However, the effect of sampling date was significant since the ratios measured on day 2 of HV termination were lower than ratios at later dates. Similarly, the interaction effect between treatments and dates of measurement was also significant.

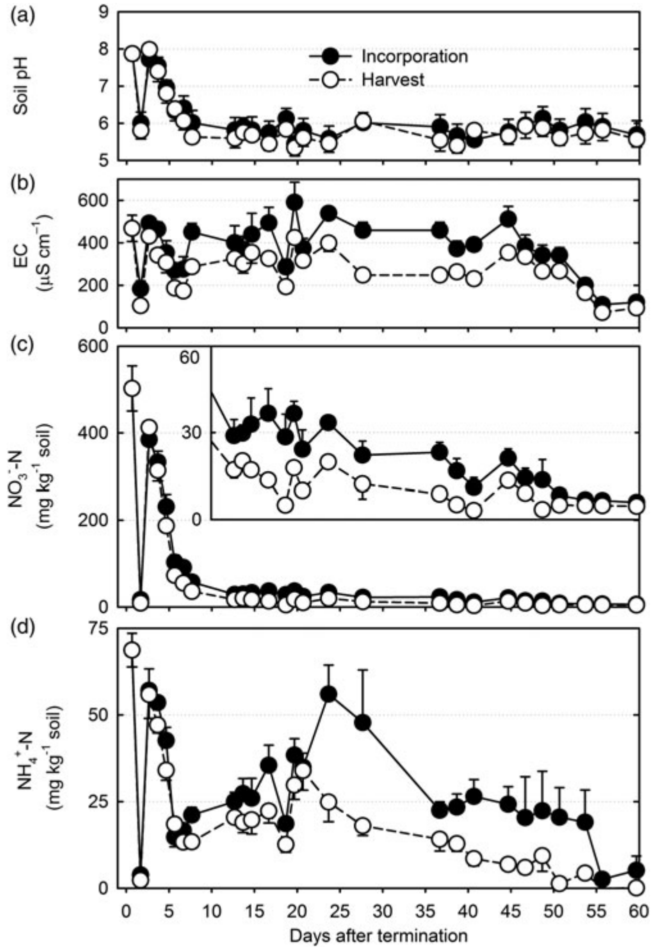

Fig. 5. Dynamics of (a) soil pH, and (b) electrical conductivity (EC), (c) nitrate (NO_3^-) and (d) ammonium (NH_4^+) N in the 0–0.15 m soil depth. The insert (panel c) is a zoom on the soil NO_3^- concentrations after 12 days of hairy vetch termination. Error bars represent the spatial variations at the plot scale (S.E., $n = 4$). Unidirectional error bars are shown for clarity.

Soil pH and electrical conductivity

Average soil pH in the HV incorporated plots across the sampling dates (6.13) was slightly higher than in the HV harvested plots (5.99) (Fig. 5a). Effects of soil sampling dates on soil pH were also significant. Interaction effects between HV residue management and soil sampling dates were not significant since the dynamics of soil pH was similar under both forms of residue management. Average soil pH prior to termination of HV was 7.85 which declined rapidly to 5.82 on day 1 of biomass termination but increased to pre-termination level on day 2. Thereafter, soil pH declined gradually but remained mostly stable 1 week after termination.

Average soil EC in the HV incorporated plots across the sampling dates (380 $\mu S\ cm^{-1}$) was significantly higher than in the HV

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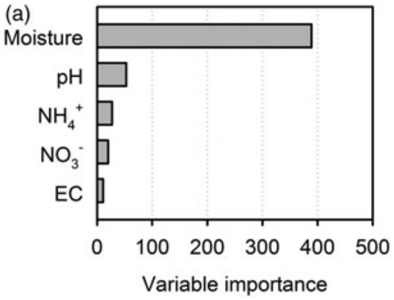

(b)

	pH	EC	NO_3^-	NH_4^+	Moisture
EC	0.20				
NO_3^-	0.95	0.38			
NH_4^+	0.67	0.68	0.80		
Moisture	0.14	-0.37	0.15	-0.01	
N_2O	0.06	-0.21	0.08	0.00	0.50

Fig. 6. (a) Variable importance (VIMP) for predicting the temporal trend of N_2O emissions. (b) Correlation matrices with Pearson's correlation coefficients (*R*) of N_2O emissions and soil variables measured on 26 measurement dates during the study period. EC, electrical conductivity.

harvested plots (276 μS cm^{-1}) (Fig. 5b). Similarly, effects of soil sampling dates had significant effects on soil EC. As with soil pH, soil EC declined on day 1 after biomass termination but increased to pre-termination level on the following day in both treatments. Soil EC declined rapidly from 462 μS cm^{-1} on day 2 after biomass termination to 224 μS cm^{-1} within 1 week but increased thereafter. Soil EC was fluctuating during the period of rainfall events during day 12–27 of termination. A rapid decline in EC was observed during the rapid growth of crabgrass biomass (day 44–55) when the plots were irrigated and received high amounts of rainfall. Interaction effects of HV biomass management and soil sampling dates were not significant since the dynamics of soil EC was mostly similar under both forms of residue management.

Dynamics of soil mineral N

Soil concentrations of NO_3^- was high prior to termination of HV, and declined on day 1 after HV termination, then increased to pre-termination levels on the following day in both treatments (Fig. 5c). Soil concentrations of NO_3^- declined rapidly from 398 mg kg^{-1} soil on day 2 post-termination to 47 mg kg^{-1} soil within 1 week, then declined at slower rates thereafter. Concentrations of soil NO_3^- on the last measurement were only 6 and 5 mg kg^{-1} soil from HV incorporated and harvested plots, respectively. Average soil concentrations of NO_3^- in the HV incorporated plots across the sampling dates (62 mg kg^{-1} soil) were significantly higher than in the HV harvested plots (49 mg kg^{-1} soil). Effects of date of soil sampling on NO_3^- concentrations were significant. Interactions of HV residue management and date of soil sampling were not significant, since the dynamics of soil concentrations of NO_3^- was similar under both forms of residue management.

Soil concentrations of NH_4^+ were high before termination of HV, declined on day 1 post-termination, then increased to pre-termination levels on the following day under both treatments (Fig. 5d). Average soil concentrations of NH_4^+ declined rapidly from 56 mg kg^{-1} soil on day 2 post-termination to 15 mg kg^{-1} soil within 1 week, but increased thereafter until a month after HV termination. The concentrations decreased to 3 mg kg^{-1} soil for HV incorporated plots, and to 0 mg kg^{-1} soil in HV harvested plots on the final week of measurements. Average soil concentrations of NH_4^+ in the HV incorporated plots across sampling dates (27 mg kg^{-1} soil) were significantly higher than in the HV biomass-harvested plots (17 mg kg^{-1} soil). Effects of date of soil sampling on concentrations of NH_4^+ were also significant. Interaction effects of HV residue management and dates of soil sampling were not significant, since the dynamics were similar under both forms of residue management.

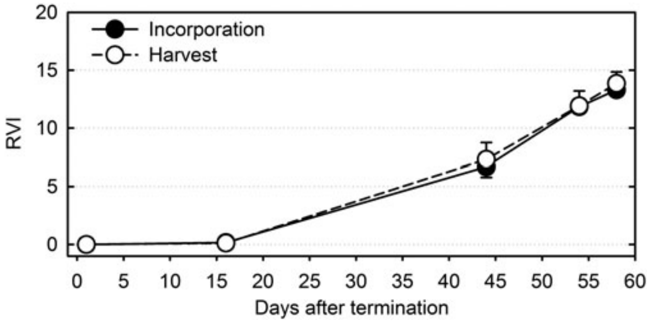

Fig. 7. Dynamics of ratio vegetation index (RVI) measured as a proxy for green biomass of crabgrass. Error bars represent the spatial variations at the plot scale (S.E., *n* = 4). Unidirectional error bars are shown for clarity.

Role of soil variables on temporal dynamics of N_2O emissions

The regression model with the random forest revealed the values of variable importance in the order as soil moisture > pH > NH_4^+ > NO_3^- > EC (Fig. 6a). In particular, soil moisture dominated the temporal dynamics of N_2O emissions. This was also supported by stronger correlations between soil moisture and N_2O emission (Fig. 6b). Correlations of N_2O emissions with other soil variables were poor and insignificant.

Crabgrass development, yield and biomass N concentrations and uptake

Growth of crabgrass, measured as RVI, was similar ($P > 0.05$) in both treatments (Fig. 7). Although germination of crabgrass was seen 1 week after planting, the germinated plants were severely affected by drought (small plant size, rolled leaves to limit transpiration, dark blue-green color), and thus, rapid increase in RVI from crabgrass was seen only after the first irrigation event.

Biomass yield of crabgrass in the HV incorporated plots (3.8 ± 0.5 Mg ha^{-1}) was 20% higher ($P < 0.05$) than the HV harvested plots (3.3 ± 0.4 Mg ha^{-1}) (Fig. 8a). Similarly, N concentration in crabgrass biomass from HV incorporated plots (2.9 ± 0.3%) was 35% higher ($P < 0.05$) than the HV harvested plots (2.1 ± 0.2%) (Fig. 8b). Thus, total N in harvested crabgrass biomass from HV incorporated plots (104 ± 9 kg N ha^{-1}) was 54% higher ($P < 0.05$) than the HV harvested plots (68 ± 8 kg N ha^{-1}) (Fig. 8c).

Discussion

Emissions of N_2O

Large emissions of N_2O following incorporation of HV into soils as seen in the current study are also reported in previous studies

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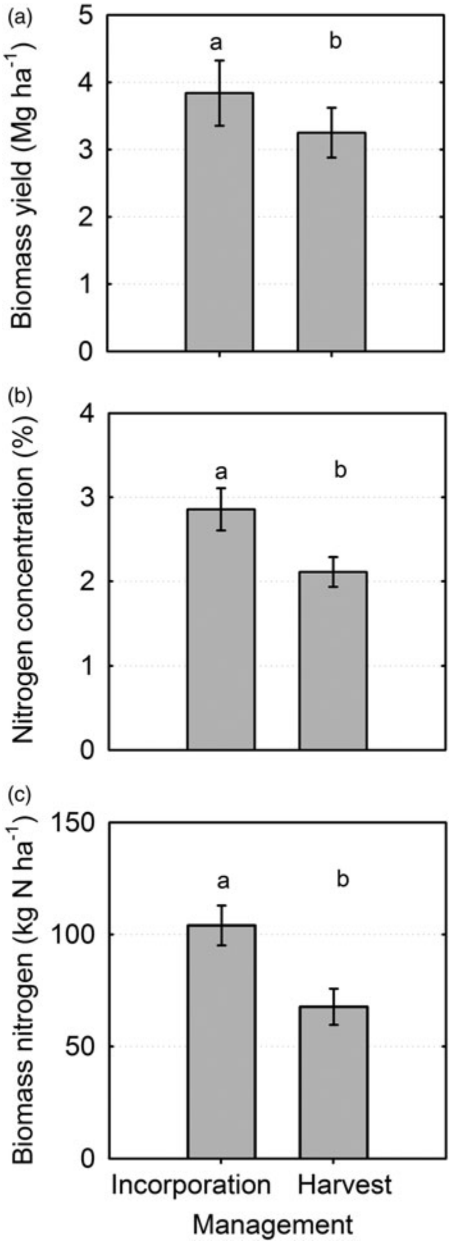

Fig. 8. (a) Mean aboveground biomass produced by crabgrass at first harvest in late-June. (b) Nitrogen concentrations of the harvested crabgrass biomass. (c) The total amount of nitrogen in the harvested crabgrass biomass. Standard error (S.E.) bars represent spatial variations at the plot scale ($n = 4$). The statistical differences ($P < 0.05$) in biomass yield are indicated by different letters on the top of bars.

conducted in other agro-climatic conditions (Sanz-Cobena *et al.*, 2014; Kim *et al.*, 2017). The enhanced emissions from such treatments are contributed by both increased concentrations of soil NO_3^- and mineralizable C (Mitchell *et al.*, 2013). We did not directly measure soil concentrations of mineralizable C in this study, but higher C mineralization in HV incorporated plots was evidenced by higher CO_2 emissions during the periods between the termination of HV and germination of crabgrass. Strong correlations between N_2O and CO_2 emissions obtained from individual collars also indicated the contribution of decomposing legume biomass to N-turnover and N_2O emissions (Li *et al.*, 2015). The correlations also support the findings from the previous studies that crop residues act as drivers for N_2O emissions from agricultural soils (Kravchenko *et al.*, 2017; Pugesgaard *et al.*, 2017). As seen in the current study, Li *et al.* (2015) found that the harvest of legume cover crops reduced N_2O emissions after soil incorporation. Similarly, the high rates of emissions after soil incorporation of HV residue in the study corroborate the findings from previous studies which reported large short-lived pulses of N_2O emissions immediately after incorporation of residues with low C/N ratio (Baggs *et al.*, 2000). Similarly, Aulakh *et al.* (1991) found large emissions of N_2O during the first 10 days of HV incorporation in an incubation study that included three different soil types. These results thus collectively indicate that large emissions of N_2O are possible after soil incorporation of legume-based green manures that have low C/N ratio. Therefore, residue removal can be an effective management option to limit N_2O emissions.

When the difference in N_2O emissions between the incorporation and harvest treatments is assumed as the emissions derived from incorporated aboveground biomass of HV, emissions from decomposing aboveground biomass were 0.6 kg N_2O-N out of 146 kg N in the aboveground biomass. The emission factor (0.4%) is lower than IPCC estimates of 1.0% (IPCC, 2007). Although additional emissions may be possible beyond the current study period, large emissions were not observed during active growth of crabgrass, even with favorable soil conditions (i.e., high soil moisture and warm temperatures) during summer.

In the previous year (2017), we measured the emissions of N_2O from the same plots as used in this study (Kandel *et al.*, 2018). Total N in incorporated aboveground biomass of HV in the previous year (185 kg N ha^{-1}; Kandel *et al.*, 2018) was slightly higher than in the current study year (146 kg N ha^{-1}). Although the difference in the amount of N was marginal, cumulative emissions of N_2O within 1 month after soil incorporation of HV (19,300 g N_2O-N ha^{-1}; Kandel *et al.*, 2018) in the previous year was 36 times higher than the emissions noted in this study year (530 g N_2O-N ha^{-1}). The large difference was likely due to the amount of rainfall received after soil incorporation of HV residues during the 2 years. The total amount of rainfall in 2017 within a month after termination (130 mm) was almost three times higher than that in 2018 (50 mm). In 2017, extremely large emissions (5200 g N_2O-N ha^{-1} day^{-1}) was observed after a large rainfall (80 mm day^{-1}) event (Kandel *et al.*, 2018). Such large rainfall events were not observed during the early period after termination of the current study. Consequently, no large peaks in N_2O emissions were recorded. Extremely high levels of soil NO_3^- concentrations and bare soil conditions within a week after termination in the current study were conducive conditions for N_2O emissions. However, denitrification during this period was constrained by lack of soil moisture generated by no rainfall events that resulted in low N_2O emissions compared to the previous year. A large rainfall event (81 mm within 48 h) was observed at 50 days following biomass incorporation. However, there was already a crabgrass stand by that period, which might have competed with denitrifying microbes in the soil for mineralized N derived from decomposing HV biomass. The rapid uptakes of mineralized N by rapidly growing crabgrass may have contributed for low concentrations of mineral N in the last week of measurements. The results indicate that intensity, frequency and timing of rainfall are key components for determining inter-annual variations in N_2O emissions from legume residues that are incorporated into soils (Skiba and Smith, 2000).

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Correlations of soil pH, EC to N_2O emissions

Previous studies have indicated that legume crops mostly acidify the soil during their growth (Yan *et al.*, 1996a; Goulding, 2016). However, high pH during the measurement period prior to termination of HV indicated that soil was not acidified during the growing season of HV. The large decrease of soil pH within the first day of soil incorporation of HV biomass was possibly due to formation of organic acids produced from sugars in the glycolytic pathway (Adeleke *et al.*, 2017; Yan *et al.*, 1996b). During the following days, rapid decomposition of these recently formed organic acids to H_2O and CO_2 may have contributed to the subsequent increase in pH (Yan *et al.*, 1996*a*, *b*). Additionally, decarboxylation and protonation of NH_3 formed by ammonification may be responsible for this pH increase because these two processes occur in parallel in soils during the decomposition of amino acids (Yan *et al.*, 1996*a*). This was also reflected in strong correlations between the dynamics of soil pH and soil NH_4^+ concentrations (Fig. 6b).

Higher EC of soil in the HV incorporated plots might be contributed by released components from the decomposition of crop residues. As seen in this current study, Moreno-Barriga *et al.* (2017) observed rapid declines in soil EC after incorporation of biomass-derived biochar in the early stage of incubation, but soil EC increased within few days and remained stable 2 weeks after incorporation. Soil concentrations of inorganic solutes containing calcium and magnesium were not determined in this study. They were expected to increase after decomposition of plant materials, which might have contributed to higher EC in the plots where HV biomass was incorporated compared to the plots where HV was harvested. The rapid decline of EC during the effective growth of crabgrass might be related to crop uptake of these solutes.

Previous studies have shown the strong predictive ability of soil pH and EC to emissions of N_2O (Adviento-Borbe *et al.*, 2006; Russenes *et al.*, 2016; Wang *et al.*, 2018). In our study, the average soil pH across all measurement dates among four blocks ranged from 5.7 to 6.8 (data not shown). However, there was no correlation between average seasonal pH from individual blocks and average N_2O emissions (data not shown). Similarly, average seasonal soil EC among the blocks ranged from 292 to 384 $\mu S\ cm^{-1}$ but there was no spatial correlation between soil EC and average N_2O emissions (data not shown). The poor spatial correlations of soil pH and EC to N_2O emissions might be related to superseding effects of mineralized soil C and N due to spatial heterogeneity in production and distribution of terminated HV biomass as evidenced by strong correlations between soil CO_2 and N_2O emissions (Fig. 3). Similarly, temporal dynamics of both soil pH and EC could not define the temporal dynamics of N_2O emissions. These poor temporal correlations might be related to the dominating effects of soil moisture on the relationship (Fig. 6a, b).

Nitrogen transfer from hairy vetch to crabgrass

To be effective as green sources of N, legumes must fix enough N in their plant tissues during growth, which in turn must be capable of effectively being transferred to the following recipient crop (Rao and Northup, 2011; Northup and Rao, 2016). Significantly higher biomass yield and N concentrations of crabgrass biomass from HV incorporated plots indicated that N from incorporated HV biomass was effectively transferred to the following recipient crop even during a drought-affected year. Since crabgrass biomass on HV incorporated plots contained 62 kg ha^{-1} more N than HV harvested plots, this showed 42% of N from HV shoot biomass, which contained 146 kg N ha^{-1} at incorporation, was transferred to the following crabgrass.

Our results indicate that removal of biomass of winter legumes used as cover crops can significantly reduce N_2O emissions from soil ecosystems during the cultivation of a following summer cash crop. However, the export of N in harvested biomass can have adverse effects on the quantity and quality of forage produced by the recipient crops. These results are in line with the results from previous studies which reported negative effects of removing legume green manures on yields of subsequent cash crops (Frøseth *et al.*, 2014). Other potential forms of management to reduce N_2O emissions from winter legume cover crops in the US SGP could be grazing of legume biomass prior to soil incorporation. This would allow the high-quality biomass provided by the green manure to be utilized without exporting significant amounts of N and other nutrients. Grazing is a more common management strategy than harvesting of forages in the SGP. Further research on N_2O emissions under this alternate form of management is needed.

Conclusions

The present study showed that the management strategies of HV cultivated as green manure to supply N for the following crabgrass hay can have significant effects on N_2O emissions. Two-month cumulative N_2O emissions from HV incorporated plots were three times higher than the HV harvested plots. Rainfall and irrigation events significantly enhanced N_2O emissions prior to the establishment of crabgrass stand but irrigation and high rainfall events during the rapid growth of crabgrass did not induce large peaks of N_2O. Although N_2O emissions were decreased by the harvest of HV biomass, crabgrass yield, concentrations and total uptake of N in the crabgrass biomass were also reduced. In conclusion, the results of this study indicate that N_2O emissions can be significantly reduced by removing legume biomass cultivated as green manure but such management can also affect quality and quantity of forage produced by the following recipient crop.

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