8-2022

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Quantification and Machine learning based N_2O-N and CO_2-C emissions predictions from a decomposing rye cover crop

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Core Ideas

- The rye cover crop decreased N_2O-N emission during early growth and increased emission during decomposition.
- The rye cover crop reduced the partial CO_{2e} from 496 to -1,061.
- In 2019 and 2020, decomposing cover crop emitted 27 and 69% of fixed C respectively.
- The random forest model outperformed by accounting 73% of the variation in the N_2O-N daily emissions.
- Daily CO_2-C emissions was also best predicted by random forest model with 85% of variation explained.

Abstract

Cover crops improve soil health and reduce the risk of soil erosion. However, their impact on the carbon dioxide equivalence (CO_{2e}) is unknown. Therefore, objective of this two-year study was to quantify the effect of cover crop-induced differences in soil moisture, temperature, organic C, and microorganisms on CO_{2e} and to develop machine learning algorithms that predict daily N_2O-N and CO_2-C emissions. The prediction models tested were multiple linear regression (MLR), partial least square regression (PLSR), support vector machine (SVM), random forest (RF), and artificial neural network (ANN). Models' performance was accessed using R^2, RMSE and MAE. Rye (*secale cereale*) was dormant seeded in mid-October and in the following spring it was terminated at corn's (*Zea mays*) V4 growth stage. Soil temperature, moisture, and N_2O-N

This article has been accepted for publication and undergone full peer review but has not been through the copyediting, typesetting, pagination and proofreading process, which may lead to differences between this version and the Version of Record. Please cite this article as doi: 10.1002/agj2.21185.

and CO_2-C emissions were measured near continuously from soil thaw to harvest in 2019 and 2020. Prior to termination, the cover crop decreased N_2O-N emissions by 34% (p=0.05) and over the entire season, N_2O-N emissions from cover crop and no cover crop treatments were similar (p=0.71). Based on N_2O-N and CO_2-C emissions over the entire season and the estimated fixed cover crop carbon remaining in the soil, the partial CO_{2e} were -1,061 and 496 kg CO_{2e} ha^{-1} in the cover crop and no cover crop treatments, respectively. The RF algorithm explained more of the daily N_2O-N (73%) and CO_2-C (85%) emissions variability during validation than the other models. Across models, the most important variables were temperature and the amount of cover crop-C added to the soil.

1.0 Introduction

Techniques to reduce agricultural greenhouse gas (GHG) emissions are needed to lower unknown future climate risks (Joshi et al., 2021; Shrestha et.al., 2019; Skinner et al., 2019). Of the numerous techniques proposed, planting a cover crop is a technique that can be rapidly adopted by many farmers (McClelland et al., 2021a). Despite many studies, there is not conclusive evidence that cover crops reduce the CO_{2e} (Basche et al., 2014; Behnke and Villamil, 2019; Thies et al., 2020; Reicks et al., 2021).

A growing cover crop can reduce soil moisture, inorganic N, and temperatures which in turn can reduce N_2O emissions (Cayuela et al., 2009; Thapa et al., 2018; Reicks et al., 2021). However, after cover crop termination the effect of the decomposing cover crops on GHG emissions is unclear (Antosh et al., 2020; Basche et al., 2016; Basche et al., 2014; Çerçioğlu et al., 2019). During cover crop decomposition, the release of inorganic N and organic substrates may increase and N_2O-N and CO_{2}-C emissions. To quantify the effect of cover crops on the carbon footprint, the CO_{2e} for the entire season must be determined. The CO_{2e} equivalence combines all GHG into a single value. However, due to the high cost of intensive trace gas measurements few studies measure emissions for the entire life cycle of both the cover and cash crops.

Aside from the difficulty of measuring N_2O-N and CO_2-C emissions, accurate and precise models are needed to provide guidance on how climate and management changes impact sustainability and GHG emissions. However, many process-based models are difficult to use, may not provide the desired accuracy (Sozanska et al., 2002; Roelandt et al., 2005; Zhang et al., 2016; Jiang et al., 2019), may require long-term field histories, and may not accurately predict management responses in real systems (Hamrani et al., 2020; Del Grosso et al., 2000, 2001; Jiang et al., 2019). In addition, following calibration process-based models often have a mixed ability to predict N_2O emissions. For example, McClelland et al. (2021b) used the DAYCENT model to predict the effect of cover crops on N_2O emissions. This work showed that the predicted and observed N_2O emissions were not correlated. In Colorado, Del Grosso et al. (2008) reported that DAYCENT overestimated N_2O emissions, whereas in Iowa, Jarecki et al. (2006) reported that DAYCENT over predicted emissions when the actual emissions were low and underestimated emissions when emissions were high. The mixed results of the model's ability to predict N_2O-N emissions may be attributed to many factors including field experiments that do not accurately measure N_2O-N emissions, process-based models that were not accurately parametrized, and/or mathematics that do not accurately describe the complexity of the system.

An alternative approach is to use the machine learning (ML) algorithms to predict GHG emissions. These models may be easier to use because they can be based on easy to measure values, may require fewer input variables than process-based models, and can be modified to account for different spatial and temporal resolutions. Therefore, the objectives were to quantify the effect of cover crop-induced differences in soil moisture, temperature, organic C, and microorganisms on CO_{2e} and to develop machine learning algorithms that can predict daily N_2O-N and CO_2-C emission.

2.0 Materials and Methods

2.1 Study Site, Experimental design, and treatments

The two-year study was conducted at the South Dakota State University Aurora Research Farm located at 44°18′20.57″N and 96°40′14.04″W in 2019 and 2020. The site was in the Dfb (humid continental climate) Köppen climatic subtype. The soil at the experimental site was a Brandt silty clay loam (fine-silty, mixed, superactive cold Calcic Hapludoll). The soil organic carbon content was 36 Mg ha^{-1} (1.8% SOC), and the surface 15 cm contained 28 g clay kg^{-1} and 650 g silt kg^{-1} (Reicks et. al., 2021). The production practices were a corn-corn rotation, no tillage, and N fertilizer was not applied.

The experimental design was completely randomized with two treatments: cover crop and no-cover crop. Each treatment was replicated 4 times. The dimensions for each experimental unit was 9.1 × 3.1 m. Winter cereal rye (*Secale cereale*) was drilled in two rows at a rate of 56 kg ha^{-1} at a depth of 2.5 cm in October in the fall of 2018 and 2019. The two cover crop rows were separated by 17.5 cm, and they were positioned in the center between 2 corn rows. The cover crop occupied about 25% of the area between the corn rows.

In the following spring, a 97-day relative maturity corn (*Zea mays*) cultivar was planted at the rate of 79,000 seeds ha^{-1} at a depth of 5 cm close to the rows of the previous corn crop. The row spacing was 76 cm. At V4 growth stage of corn and boot stage of rye, rye was terminated using glyphosate [N-(phosphonomethyl) glycine); Roundup Power Max] at the rate of 2.34-liter ha^{-1}. A non-ionic surfactant was added at 0.25% of the spray solution. Ammonium sulfate was also added to the spray solution at 10.2 g L^{-1}. Corn was harvested on 26 September 2019 and 8 October 2020. More details about field activities are provided in Table 1.

2.2 GHG emission measurements

Nitrous oxide-N and CO_2-C emissions were measured from cover crop termination to harvest using techniques described in Reicks et al. (2021). Glyphosate was used to kill the cover crops, but because the rye at termination was taller (approximately 45 cm) than the rings (6cm above soil surface), the plants were bent and twisted such that the cover crop fit inside the rings. At the corn V4 growth stage, PVC pipe rings 12-cm tall having a diameter of 20-cm and a surface area of 317 cm^2 were randomly placed in the production plots with and without cover crops. In plots with cover crops, the PVC rings were centered on the cover crop rows, whereas in plots without cover crops, the rings were centered between the corn rows. For GHG measurement, eight PVC rings (4 per treatment) were pushed 6 cm into the soil with 6 cm remaining above the soil surface. Directly before termination, similar rings were placed adjacent to the GHG microplots in the cover crop treatment. The cover crop within the ring was clipped near the soil surface, dried, weighed and analyzed for C and N in the laboratory.

To collect GHG from the microplots, the PVC rings were covered with LI-COR long-term opaque chambers (8100-104 LI-COR) six times daily for 15 minutes at four-hour intervals (between 0000 and 0230 h, 0400 and 0630 h, 0800 and 1030 h, 1200 and 1430 h, 1600 and 1830 h, and 2000 and 2230 h) (Reicks et al. 2021). Using a Picarro Cavity Ringdown Spectrometer (model G2508, Picarro Inc, Santa Clara, CA), gases extracted from the chambers were analyzed for N_2O-N and CO_2–C concentrations. Emissions were calculated using the LI-COR SoilFluxPro 4.01 software (v. 4.01; LI-COR). Standard N_2O, and CO_2 gases were used at the beginning and end of the experiment to ensure Picarro gas analyzer accuracy. Soil moisture and temperatures for the surface 0 to 5 cm were measured using LI-COR LI-8150-205 Soil Moisture Probes and LI-COR LI-8150-203 Soil Temperature Probes (LI-COR), respectively.

2.3 Soil sampling

Soil samples were collected from the 0 to 15 and 15 to 30 cm depth at cover crop termination in area adjacent to the PVC rings to avoid soil disturbance within the ring on June 24 (cover crop termination) and from inside the ring at the termination of the experiment on October 21 (each year) following corn harvest. Soil samples from the 0 to 15 cm depth was analyzed for bulk density, gravimetric soil moisture, inorganic N, soil organic carbon and the soil microbial community (Table 1). Samples from the 15 to 30 cm depth were analyzed for bulk density, gravimetric soil water, inorganic N, and soil organic carbon. Gravimetric soil moisture content and bulk densities were determined by drying the soil at 105 ^{o}C for 24 hours. Air dried subsamples were ground and analyzed for total C and N, $NH_4{}^+$–N and $NO_3{}^-$–N (Clay et al., 2015).

2.4 Soil microbial biomass and composition

Soil samples were collected from 0 to 15 cm soil depth at the same timings as above for microbial biomass and composition following procedures outlined in Veum et al. (2019). Microbial community composition was determined using PLFA (Phospholipid Fatty Acid) protocols described by Buyer and Sasser (2012), Thies et al. (2019), and Fiedler et al. (2021). In this analysis, 19:0 phosphatidylcholine was used as an internal standard for PLFA and a 19:0 trinonadecanoin glyceride was used as an internal standard for NLFA (neutral lipid fatty acids).

A Shimadzu GC-2010 Plus gas chromatograph (Shimadzu Corporation, Japan) with a flame ionization detector was used to analyze the extracts. The PLFAD2 method was used to calibrate the gas chromatograph using a standard provided by MIDI Sherlock (No. 1208, MIDI, Inc., Newark, DE). Using the MICSOILV2 approach from the MIDI Sherlock Software system (MIDI, Inc., Newark, DE) fatty acids were assigned to distinct functional groups associated with each community type to determine the number and types of microorganisms within the microbial population (Veum et al., 2019). Terminally branched chain fatty acids were used to identify gram-positive bacteria, while monounsaturated and hydroxy substituted fatty acids were used to identify gram-negative bacteria. Methyl branched chain fatty acids were used to identify actinomycetes (Zhang et al., 2016). Total microbial biomass was the summation of all fatty acids (Quideau et al., 2016).

2.5 Statistical Analysis

2.5.1 Carbon dioxide equivalence

The experiment used a completely randomized design where each treatment was replicated 4 times per treatment. Total N_2O-N and CO_2-C emissions were determined by integrating the emissions over the study period. The experiment was repeated in 2019 and 2020. The analysis of variance was conducted to compare the total N_2O-N and CO_2-C emissions, inorganic nitrogen, total carbon, and microbial population from each treatment using "agricolae" package in Rstudio (R core Team 2017). Tukey HSD test was conducted after ANOVA analysis to determine significant differences between treatment means at p-value 0.05.

Based on the cover crop occupying 25% of the area between the corn rows the N_2O-N and CO_2-C emission data were area weighted. For this correction, the emissions from the cover crop were multiplied by 0.25 which was added to product of 0.75 times the emissions from the no-cover crop. The CO_{2e} was determined by converting N_2O-N kg ha^{-1} values to N_2O kg ha^{-1} and CO_2-C kg ha^{-1} to CO_2 kg ha^{-1}. The N_2O was then converted to CO_{2e} determined by multiplying N_2O by 298. The partial CO_{2e} value was the summation of $CO_{2e\ N2O}$ and CO_2 which was then subtracted from the amount of CO_2 that was fixed by the cover crop during the growth phase. This analysis did not consider the effect of the cover crop on methane emissions or any factors other than those directly involved in the production of N_2O-N and CO_2-C during the cover and cash crop growing seasons.

2.5.2 Machine learning models

"Hmisc" package and "rcorr" function in Rstudio was used to determine the Pearson's correlation (r) between all the variables. Following correlation analysis of all the variables, CO_2-C and N_2O-N emissions were predicted using five models. Those five models tested were multiple linear regression (MLR), partial least square regression (PLSR), support vector machine (SVM), random forest (RF) and artificial neural network (ANN). MLR model was considered the traditional linear regression model whereas rest of the models were machine learning models. The PLSR method is well-known for its ease of use when dealing with highly correlated variables. It was selected because it generalizes and combines features from principal component analysis and multiple linear regression (Abdi, 2003). The SVM algorithm creates a line or a hyperplane which separates the data into different classes. The line or hyperplanes are considered as the decision boundary, and they are utilized to predict continuous outputs. It was selected due to its ability to solve non-linear regression prediction problem (Ahmad et al. 2014). The non-linear "svmRadial" algorithm from the R “caret” package was utilized to implement SVM in our analysis. The RF is a machine learning (ML) algorithm for classification and regression which is based on the recursive partitioning principle, and specific information about the relationships between the response and predictor variables is not required (Breiman, 2001; Hamrani et al., 2020; Sharma et al., 2022). It creates a forest with several decision trees. With the RF approach, the accuracy and robustness of model is directly correlated with the number of trees in the forest (Breiman, 2001). The ANN adapts to the computing environment by adjusting neuron weights and thresholds repeatedly. When the network's output error approaches the expected value, the network training is complete. This model is gaining in popularity because of its ability to develop predictive relationships even when there is not a coherent theoretical framework (Maind and Wankar, 2014). The model predicted daily emissions, that were calculated by integrating the hourly measurements (every 4 hours = 6 samples/ day). The whole dataset was randomly divided into training (75%) and validation (25%) datasets. On the training data set, k-fold cross-validation (CV) was carried out for resampling procedures using “caret” package. The CV technique splits the data into different folds, estimates the error rate based on machine learnings algorithms, and then generates the final model with the lowest error rate (Yang et al., 2011). In this work, 10 folds with three replications of the repeated k-fold CV were used. The model performance was assessed by comparing the coefficients of determination (R^2), root mean square errors (RMSE), and mean of absolute value of error (MAE) that were determined with the equations,

$$R^2 = 1 - \frac{\sum_{i=1}^{n}(y_i - y_p)^2}{\sum_{i=1}^{n}(y_i - \bar{y}_i)^2} \quad \text{[Eq 1]}$$

$$RMSE = \sqrt{\frac{1}{n}\sum_{i=1}^{n}(y_i - y_p)^2} \quad \text{[Eq 2]}$$

$$MAE = \frac{|(y_i - y_p)|}{n} \quad \text{[Eq 3]}$$

where y_i and y_p were measured and predicted values (N_2O-N or CO_2-C) respectively, and $\bar{y}_i$ was the mean of all measured values and n was the number of samples. All the models were built using "caret" package (Version 6.0-88) in Rstudio. In the model, N_2O-N and CO_2-C were used as dependent variables whereas soil temperature, air temperature, soil moisture, amount of cover crop-C remaining, and rainfall were used as predictor variables. The best performing models has high R^2 (closer to 1) and low RMSE and MAE values.

The total daily cover crop-C was calculated using equations 4 and 5 as shown below,

$$CO_2\text{-}C_{CC\ emitted} = [CO_2\text{-}C_{CC+soil\ emitted}] - [CO_2\text{-}C_{soil\ emitted}] \quad \text{[Eq 4]}$$

where CO_2-$C_{CC\ emitted}$ was the daily amount of CO_2-C that was mineralized from the cover crop over a 24-hour period, CO_2-$C_{CC\ +\ soil\ emitted}$ was the total amount of CO_2-C that was emitted over a 24-hour period in cover crop treatment, and CO_2-$C_{Soil\ emitted}$ was the total amount of CO_2-C that was emitted over a 24-hour period in the no-cover crop treatment. The amount of cover crop-C remaining in the soil was calculated with the equation,

$$\text{Cover crop-}C_{remaining} = [\text{Cover crop-}C_{initial}] - [CO_2\text{-}C_{CC\ emitted}] \quad \text{[Eq 5]}$$

where, Cover crop-$C_{remaining}$ was the amount of cover crop-C remaining in the chambers, [Cover crop-$C_{initial}$] was the amount of cover crop-C in the soil when the cover crops were termination, and CO_2-$C_{CC\ emitted}$ was defined in equation 4.

The importance of the variables was determined following validation. Variable importance was determined using the "varImp" function from the “caret” package. The function used scaled important score between 0 to 100. The higher the score the more important.

3.0 Results and Discussion

3.1 Weather and climatic conditions

At the study area, the 30-year (1989 to 2019) average annual rainfall was 640 mm, the average growing season rainfall (May to September) was 452 mm, the average growing degree days (10 ^{o}C base and 30 ^{o}C maximum temperature) from April to October was 1256 GDD’s, the average annual temperature was 6.3 ^{o}C, and the growing season average temperature was 17.9 ^{o}C (NOAA, 2022). At the study site, the average annual and growing season temperature in 2019 were 5.37 and 17.9 ^{o}C, whereas in 2020 it was 7.15 and 18.9 ^{o}C respectively (Figure 1). Total annual rainfall in 2019 was 825 mm of which 607 mm occurred during the growing season. In 2020, total rainfall was 441 mm of which 324 mm occurred during the growing season. In 2019 and 2020 the numbers of accumulated growing degree days based on corn were 1266 and 1436, respectively. Additionally, from 1 October 2018 to 31 March 2019 and from 1 October 2019 to 31 March 2020 the average snow depth was 8.7 cm and 12 cm, respectively. The temperature of the snow-covered soil at 0 to 5 cm depth, ranged from -5.12 to 13.17 ^{o}C in 2019 and from -0.93 to 13.99 ^{o}C in 2020. Between cover crop termination and harvest, the soil moisture content of the cover crop treatment in the 0 to 5-cm soil depth was greater (0.32 cm^3 cm^{-3}) than the no-cover crop treatment (0.26 cm^3 cm^{-3})

(Table 2). On average across years, the average soil temperature for the surface 0 to 5cm was 3.1 ^{o}C cooler in the cover crop (14.2 ^{O}C) than the no-cover crop (17.3 ^{o}C) treatment.

3.2 Cover crop biomass and corn grain yield

The amount of dried above-ground rye biomass contained within the microplot was 4156±576 and 3166±353 kg biomass ha^{-1} in 2019 and 2020, respectively. Based on previously reported value of 0.497 g root (g shoot)$^{-1}$ for the root to shoot ratio (Sawyer et al., 2017), the amount of rye roots was calculated. Rye roots were then multiplied by 2 to estimate the root exudates (Kuzyakov and Domanski, 2000; Kuzyakov and Larionova, 2006). Finally, to determine total rye biomass the shoot + root + root exudates were summed which was then multiplied by the amount of carbon in the above ground biomass samples [0.42 g carbon (g biomass)$^{-1}$]. The amount of cover crop-C added to each chamber was 4,349 and 3,312 kg C ha^{-1} in 2019 and 2020, respectively. The measured C to N ratio of the above ground cover crop biomass was 31:1 and 25:1 in 2019 and 2020, respectively. Based on these values, the amount of N contained in the above ground cover crop biomass was 56 and 43 kg N ha^{-1} in 2019 and 2020, respectively. This calculation does not consider N contained in root biomass.

The above cover crop C and N values represent the additions to area between the corn rows that were seeded with cover crops. The area seeded with cover crops represented about 25% of the area between corn rows. Based on this percentage, the amount of cover crop biomass in the production plot was 1120 and 702 kg biomass ha^{-1} in 2019 and 2020, respectively.

The effects of the cover crop on corn growth and yield have been reported by Miller et al. (2021). Across years, corn grain yields at 15.5% moisture ranged from 7.7 to 12.8 Mg ha^{-1}. The no cover crop treatment had 40% greater yield than treatment with cover crop that was terminated at corn's V4 growth stage.

3.3 N_2O and CO_2 emissions

N_2O-N and CO_2-C emissions in 2019 and 2020 were separated into two periods when the cover crops were growing and when they were decomposing. Reicks et al. (2021) reported on emissions between soil thaw and cover crop termination at V4. To summarize, this growth period N_2O-N emissions were 90 and 192 g ha^{-1} in the cover crop and no-cover crop treatments in 2019, respectively. In 2020, similar results were observed, and N_2O-N emissions were 168 and 209 g N_2O-N ha^{-1} in the cover crop and no-cover treatments, respectively. Lower N_2O-N emissions in the cover crop compared with the no-cover crop treatment was attributed to the cover crop reducing soil moisture and inorganic N (Reicks et al. 2021). Due to higher soil temperatures, N_2O-N was slightly higher in 2020 than 2019. Based on these values, the cover crop-induced decrease (cover crop - no-cover crop) in N_2O-N emissions was 0.11 in 2019 and 0.04 kg ha^{-1} in 2020. These decreases were equivalent to 0.42 and 0.78% of the N contained in the above ground cover crop biomass. Higher emissions in 2020 than 2019, were attributed to higher temperatures and nitrous oxide being produced during nitrification and denitrification.

Greater N_2O-N emissions were observed during cover crop decomposition than the growth phase. In 2019, N_2O-N emissions in the cover crop and no-cover crop treatments were 537 and 301 g N_2O-N ha^{-1} and in 2020 N_2O-N emissions in the cover crop and no-cover crop treatments were 953 and 537 g N_2O ha^{-1}, respectively (Figure 2, Table 2). Differences in N_2O-N emissions during the growth and decomposition cover crop phases were attributed to the decomposing cover crop biomass releasing NH_4^+ into the soil. The NH_4^+ was subsequently nitrified of which 0.03 to 1% of the N can be emitted as N_2O-N (Farquharson, 2016).

The amounts of CO_2-C that was emitted in 2019 prior to corn's V4 growth stage were 1379 and 882 kg CO_2-C ha^{-1} in the cover crop and no-cover crop treatment, respectively (Reicks et al. 2021). During decomposition, CO_2-C emissions within the chambers were 5093 and 3935 kg CO_2-C ha^{-1} in the cover crop and no-cover crop treatment, respectively. The cover-crop induced increase in CO_2-C emissions represented 27% of the estimated amount of carbon contained within the above and below ground cover crop biomass.

In 2020, CO_2-C emissions during the growth phase were similar in the cover crop and no cover crop treatments and averaged 1500 kg CO_2-C ha^{-1} (Reicks et al. 2021). However, during decomposition, CO_2-C emissions in the cover crop and no-cover crop treatments were 7970 and 5690 kg CO_2-C ha^{-1}. The difference between CO_2-C emitted in the cover crop and no-cover crop treatment was equivalent to 69% of the estimated amount of above and below ground cover crop biomass-C. The increased CO_2-C emissions were attributed to the cover crop providing organic C to the soil which was subsequently mineralized (Poeplau and Don, 2015; Rosecrance et al., 2000; Aulakh et al., 2001; Smith et al., 2011). Lower emissions in 2019 than 2020 were attributed to cooler temperatures.

In 2019, CO_2-C emissions tended to decrease as the season progressed, whereas in 2020 CO_2-C increased or remained relatively constant and then decreased after September 15 (Figure 2). In both years, the ratio between CO_2-C and N_2O-N varied across the seasons. Since the CO_2-C is a function of the aerobic respiration and N_2O-N emission is a function of both nitrification and anaerobic respiration, a higher CO_2-C/ N_2O-N ratio suggests that there was an increased importance of aerobic respiration or a change in the soil microbial community structure. For example, from June 24 to September 10, 2019, the ratio between CO_2-C and N_2O-N in the cover crop and no-cover crop treatments were 10,500 and 16,500 (kg CO_2-C h^{-1}) (kg N_2O-C ha^{-1})$^{-1}$, respectively (p=0.006). This apparent cover-crop induced decrease in the CO_2-C and N_2O-N ratio suggests that the biota in cover-crop treatment has a higher reliance on anerobic respiration than the no-cover crop treatment. This apparent increased reliance on anaerobic respiration was associated with increased CO_2 emissions, which most likely reduced soil O_2 concentrations.

Between September 11 and October 20, 2019, similar results were observed and the CO_2-C to N_2O-N ratios in the cover crop and no-cover crop were 3940 and 5905 (kg CO_2-C h^{-1}) (kg N_2O-C ha^{-1})$^{-1}$ (p=0.08), respectively. Again, these results suggest that the cover crop treatment had a higher reliance on anaerobic respiration than the no-cover crop treatment.

In 2020, between June 24 and September 10 the CO_2-C to N_2O-N ratio in the cover crop and no-cover crop treatment were 7,720 and 15,970 kg CO_2-C h^{-1}) (kg N_2O-C ha^{-1})$^{-1}$, respectively (p=0.004). Later in the season (September 11 to October 20) the CO_2-C to N_2O-N emissions ratios were similar in the cover crop and no-cover crop treatment and had a ratio of 14,980 (kg CO_2-C h^{-1}) (kg N_2O-C ha^{-1})$^{-1}$. Temporal changes in the CO_2-C to N_2O-N ratio for this same soil were also observed by Thies et al. (2020), where the impact of different fertilizer application dates on N_2O-N and CO_2-C emissions were investigated. It was observed that fertilizer applied on 20 September 2017 had a CO_2-C to N_2O-N ratio of 1360 whereas fertilizer applied on 1 October 2017 had a ratio of 24,000. These values suggest that the relative amount of N_2O-N that is emitted per unit or respired CO_2-C can vary widely.

3.4 Change in soil total inorganic nitrogen and carbon during decomposition

In the linked experiment, Reicks et al. (2021) reported that the cover crop reduced soil inorganic N and soil moisture during the cover crop growth phase compared to no cover crop treatment. However, when the chambers were moved to a new location slightly different results were observed. At the new location, the amount of NO_3 + NH_4-N contained in the surface 30 cm at cover crop termination was not affected by the cover crop. However, at harvest the cover crop increased the amount of inorganic N in the soil (Table 3). These results suggest that N mineralization of the cover crop biomass may provide N to corn. However, the timing of the mineralization is critical to assess if it will reduce the N requirement in the current or future crop. In this example, an increase of 14 kg N ha^{-1} was observed following harvest.

An increase in N at harvest would not reduce the N requirement in the harvested crop, however it might influence the N requirement in the upcoming crop if the N remains in the soil profile. In the past, fertilizer replacement values for cover crops in corn have been mixed. According to Mahama et al. (2016), the N fertilizer requirement in the cash crop can be reduced by introducing legume cover crops. However, different results have been reported for non-legume cover crops. Sawyer et al. (2017) reported that the rye cover crop reduced corn yield by 5% in Iowa and that the economic optimum N rate for corn were similar in the rye cover crop and no-cover crop treatments. Pantoja et al. (2016) extended this discussion and reported that the rye cover crop does not provide a meaningful amount of N to the growing corn plant in the year of termination. However, neither study considers what happens in following years.

The amount of soil organic C contained in the surface 30 cm at V4 growth stage of corn (cover crop termination) was not affected by cover crop in either 2019 or 2020. However, when the experiment was terminated in October the cover crop increased the amount of soil organic carbon 3,031 kg SOC-C ha^{-1}. This increase in SOC indicates that a relatively large portion of the cover crop biomass remained in the soil after 117 to 119 days of decomposition.

3.5 Change in the soil microbial biomass due to cover crop decomposition

Microbial biomass was higher when the cover crop was termination than following harvest and it was higher in the cover crop than the no-cover crop (Table 4). These temporal differences were consistent with Kaiser et al. (1995) where it was reported that microbial biomass was generally lowest during the winter and highest in the summer. Across years, the fungi concentration was lower than the bacteria concentration. In 2019, the fungi to bacteria ratio was higher in the cover crop than the no-cover crop treatment at both sampling dates. For example, at cover crop termination the ratio was 0.44 in the cover crop and 0.24 in no cover ($p=0.01$). Similarly, following harvest the fungi to bacteria ratio was 0.29 for the cover crop treatment and 0.18 for the no-cover crop treatments ($p=0.06$). Apparent relative cover crop induced increases in fungi may be associated with the composition of the cover crop biomass, cooler soil temperatures, and higher soil moisture contents. Our observations were consistent with Malik et al. (2016), where it was reported that following litter addition there was an increase in fungal phyla.

Associated with the higher fungus to bacteria ratio in the cover crop than the no-cover crop treatment was higher CO_2-C to N_2O-N emission ratios. Changes in the microbial community structure are important because there are fundamental differences between fungi and bacteria. These differences include that: 1) fungi decompose more complex organic molecules than bacteria, 2) fungi have slower growth rates than bacteria, and 3) fungi may store more carbon in the soil than bacteria (Helfrich et al., 2015).

In 2020 slightly different results were observed and the fungi to bacteria ratios were similar in cover crop and no cover crop treatment. In addition, the fungi to bacteria ratios were similar ($p=0.18$) at both sampling dates (Table 4). These finding suggest that cover crops in addition to reducing soil temperature and increasing soil moistures, have the potential to change the microbial community structure, which in turn can affect the relative amount of N_2O-N and CO_2-C that is emitted.

3.6 Partial Carbon Dioxide Equivalence (CO_{2e})

Rye cover crops have mixed results on N_2O-N and CO_2-C emission over the entire year. Our investigation found that during the cover crop growing phase, rye lowered soil moisture and inorganic nitrogen, and reduced N_2O-N emissions by 66% relative to no-cover crop. Different results were observed during the decomposition phase, when the cover crop increased N_2O-N and CO_2-C emissions. The increase in emissions during decomposition may be related to the cover crop providing organic carbon as well as lowering the soil temperature and increasing the soil moisture. When combing both phases, the rye cover crop did not influence (p=0.71) N_2O-N emissions and were 565 g N_2O-N ha^{-1} in the rye cover crop and 530 g N_2O-N ha^{-1} in the no-cover crop treatment. This finding suggests that reduced N_2O-N emission during cover crop growing phase offsets the increased emission during decomposition. However, the cover crop had greater (p-value= 0.001) CO_2-C emission (6750 kg CO_2-C ha^{-1}) than the no cover crop treatment (5951 kg CO_2-C ha^{-1}). This increase does not account for the large amount of CO_2 removed from the atmosphere by the cover crop. The partial CO_{2e} was determined by considering CO_2-C and N_2O-N emissions and the amount of CO_2-C that was removed from the atmosphere during photosynthesis. In the cover crop and no cover crop treatment the average CO_{2e} across years and the entire cover and cash crop growth cycles were -1,061 and 496 kg CO_{2e} ha^{-1}, respectively. These values suggest that cover crops have the potential to reduce the agricultural carbon footprint.

3.7 N_2O-N and CO_2-C emission prediction using a machine learning algorithm

Correlation analysis across years and treatments showed that the daily N_2O-N emissions were positively correlated to CO_2-C, air temperature, soil moisture, soil temperature, cover crop-C remaining in the soil, and rainfall (Figure 3). Similarity, analysis showed that daily CO_2-C emissions were positively correlated to N_2O, air temperature, soil moisture, soil temperature and cover crop-C remaining in the soil. However, CO_2-C emissions and rainfall were not correlated.

After determining which input parameters were statistically related to the N_2O-N and CO_2-C emissions, models based on soil temperature, air temperature, soil moisture, amount of cover crop-C remaining, and rainfall were developed. The RF model that predicted daily N_2O-N and CO_2-C emissions over two years outperformed all models and had with highest R^2, lowest RMSE and MAE during training and validation (Table 5). These findings were consistent with Philibert et al. (2013), Hamrani et al. (2020), and Saha et al. (2021).

During training, the RF model explained 95% of the N_2O-N emissions variability in the cover crop and no-cover crop treatments over two years. The RMSE and MAE for this model was 1.85 g N_2O-N ha^{-1} and 0.92 g N_2O-N ha^{-1}. For the validation data set, the R^2, RMSE and MAE values were 0.73, 3.7 g N_2O-N ha^{-1} and 2.1 g N_2O-N ha^{-1} respectively. The MLR, PLSR, SVM, and ANN models did not perform as well as the RF model (Figure 4).

The importance of the variables was determined for each model (Figure 5). In this analysis, variables were assigned scaled score between 0 to 100, with 100 being most important and 0 being least important. Variable importance differed among models and between the two emission gasses. For the N_2O-N RF model, cover crop carbon was most important variable followed by air temperature, soil temperature, soil moisture and lastly rainfall. For the CO_2-C RF model, soil temperature was the most important variable, and rainfall was the least important.

Models such as these can be used to improve our understanding of the factors affecting emissions and provide insights into how to minimize CO_2-C and N_2O-N emissions. For example, decreasing the soil temperature 1^{o} C reduced RF N_2O emissions predictions by 0.52%. Similar analysis can be conducted to predict how changes in soil moisture or cover crop biomass would affect emissions. This analysis suggests that additional research is needed to extend the use of the N_2O-N and CO_2-C machine learning algorithms to assess different climate and management scenarios (McLennon et al., 2021).

4.0 Conclusion

The decomposing rye cover crop stimulated microbial activity and changed the microbial community structure, which in turn increased N_2O-N and CO_2-C emissions. During cover crop decomposition, the amount of N_2O-N that that was emitted was equivalent to 0.24 and 0.42% of the N contained in the above ground cover crop biomass in 2019 and 2020 and an amount that was equivalent to 39% and 76 % of cover crop-C was released as CO_2-C in 2019 and 2020, respectively. Furthermore, the cover crop increased soil total carbon, total inorganic nitrogen, and moisture, all of which promote soil metabolic activity and respiration. During the rye cover crop growing phase, it reduced the N_2O-N emission which was attributed to nutrient and moisture uptake by the rye. This means that the cover crops had opposite effects on GHG emissions during growth and decomposition. For this reason, measuring cover crop emissions over the whole growing season is essential to fully understand their emission pattern.

Analysis suggests that only a relatively small portion of the N contained in the cover crop was contained in the soil at harvest or emitted into the atmosphere as N_2O-N. Although the cover crop increased N_2O-N and CO_2-C emissions, it also released inorganic nitrogen into the soil. This increased N contained in the soil at harvest has the potential to reduce the crop plants nutrient requirement in subsequent years. These results suggest that the mineralization of N from the rye biomass and N uptake by the growing corn plant were not synchronized. This question will be considered in subsequent papers. In the cover crop and no cover crop treatments the average CO_{2e} across years was -1,061 and 496 kg CO_{2e} ha^{-1}, respectively. These values suggest that cover crops have the potential to reduce the agricultural carbon footprint.

Additionally, our results demonstrate that ML based algorithm may can be useful for predicting N_2O-N and CO_2-C emission. Of the models tested, the Random Forest explained the most amount of variability over two seasons. Additionally, our results suggest that we may be able to improve GHG predictions by merging machine learning and process-based models into a common analysis. Models such as these, can be used to predict the effects of different management systems and climatic conditions on N_2O and CO_2 emissions.

5.0 Acknowledgments

Research supported in part by the National Science Foundation under Grant Number 2202706 and 2026431, South Dakota Corn Utilization Council, USDA-NRCS, and the South Dakota Agricultural Experiment Station. Any opinions, findings, and conclusions or recommendations expressed in this paper are those of the authors and do not necessarily reflect the views of the National Science Foundation.

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Table 1. Summary of activities and dates of operations performed during the two-year experiment.

Field activities and operations	2019	2020
Rye cover crop dormant seeded	16-Oct-18	23-Oct-19
GHG measured in growing cover crop	April 26 to June 24	April 8 to June 24
Corn planted	16-May	14-May
Rye cover crop termination at boot stage (corn V4). Soil samples and rye tissue samples collected.	24-Jun	24-Jun
GHG measurements started at rye cover crop termination.	24-Jun	24-Jun
Corn harvest	26-Sep	8-Oct
Termination of GHG measurements. Soil samples collected.	21-Oct	21-Oct

Table 2. Cumulative N_2O-N and CO_2-C emissions, soil temperature and moisture in 2019 and 2020 during cover crop decomposition.

Cover crop	Year	N_2O-N	CO_2-C	Soil Temp	Soil Moist
		g ha^{-1}	kg ha^{-1}	^oC	cm^3 cm^{-3}
No-cover crop	2019	301	3935	17.19	0.32
Cover crop	2019	537	5093	13.02	0.33
No-cover crop	2020	359	5691	17.93	0.22
Cover crop	2020	955	7969	15.89	0.3
p-value		0.1	0.41	0.41	0.07
2019		419	4518	12.4	0.34
2020		657	6829	16.95	0.27
p-value		0.003	0.001	0.03	<0.001
No-cover crop		330	4813	17.32	0.26
Cover crop		746	6531	14.21	0.32
p-value		<0.001	0.004	0.05	<0.001

Table 3. Cover crop impact on soil inorganic nitrogen (NO_3 + NH_4) and organic C contained in the surfaced 30 cm at cover crop termination and following harvest in 2019 and 2020. Difference in lowercase letters indicate significant different in mean at p = 0.05.

		Total inorganic N		Total organic C	
Treatment	Year	Cover Crop Termination	Following Harvest	Cover Crop Termination	Following Harvest
		-------------------kg ha^{-1}---------------			
No-cover crop	2019	48	36 a	79,910	81,290
Cover crop	2019	42	42 a	81,280	84,870
No-cover crop	2020	38	50 a	74,790	75,280
Cover crop	2020	48	73 b	71,340	77,760
p-value		0.1	0.05	0.24	0.58
2019		45	39	80,600	83,080
2020		40	46	73,060	76,520
p-value		0.9	<0.001	0.01	0.001
No-cover crop		43	43	77,350	78,290
Cover crop		45	58	77,810	81,320
p-value		0.7	0.004	0.8	0.05

Table 4. The impact of the cover crop on total biomass, bacteria, and fungi at cover crop termination and following harvest in 2019 and 2020. Difference in lowercase letters indicate significant different at p = 0.05.

		Total biomass		Total Bacteria		Total Fungi	
Treatment	Year	Cover Crop Termination	Following Harvest	Cover Crop Termination	Following Harvest	Cover Crop Termination	Following Harvest
		------------------------------mg C (kg soil)$^{-1}$ ----------------------------------					
No-cover crop	2019	4.7 a	1.4	2.3	0.8	0.5 a	0.1
Cover crop	2019	8.5 b	2.5	2.9	1.2	1.3 b	0.4
No-cover crop	2020	3.2 a	1.7	1.4	1.0	0.3 a	0.2
Cover crop	2020	4.6 a	2.7	2.1	1.3	0.4 a	0.4
p-value		0.03	0.9	0.98	0.56	0.01	0.8
2019		6.5	1.95	2.6	1.05	0.9	0.25
2020		3.9	2.2	1.75	1.15	0.35	0.3
p-value		<0.001	0.25	0.001	0.09	<0.001	0.4
No-cover crop		3.95	1.55	1.85	0.9	0.4	0.15
Cover crop		6.55	2.6	2.5	1.25	0.85	0.4
p-value		<0.001	0.004	0.005	0.001	0.001	0.030

Table 5. Performance comparisons during training and validation for a traditional regression-based model (MLR) and machine learning (PLSR, SVM, RF and ANN) models for predictingN_2O-N and CO_2-C emission.

N_2O-N	Training dataset			Validation dataset		
Models	R^2	RMSE	MAE	R^2	RMSE	MAE
MLR	0.26	6.41	3.61	0.30	5.94	3.72
PLSR	0.23	6.52	3.78	0.28	6.03	3.97
SVM	0.69	4.61	0.95	0.60	4.69	2.24
RF	**0.95**	**1.85**	**0.92**	**0.73**	**3.71**	**2.08**
ANN	0.56	5.56	2.87	0.61	4.67	2.27
CO_2-C	Training dataset			Validation dataset		
Models	R^2	RMSE	MAE	R^2	RMSE	MAE
MLR	0.60	17.86	13.96	0.57	19.28	14.67
PLSR	0.56	18.8	14.68	0.55	19.91	15.06
SVM	0.81	12.6	8.51	0.73	15.47	10.05
RF	**0.96**	**5.71**	**4.05**	**0.85**	**11.92**	**8.55**
ANN	0.69	16.07	12.44	0.68	16.18	10.65

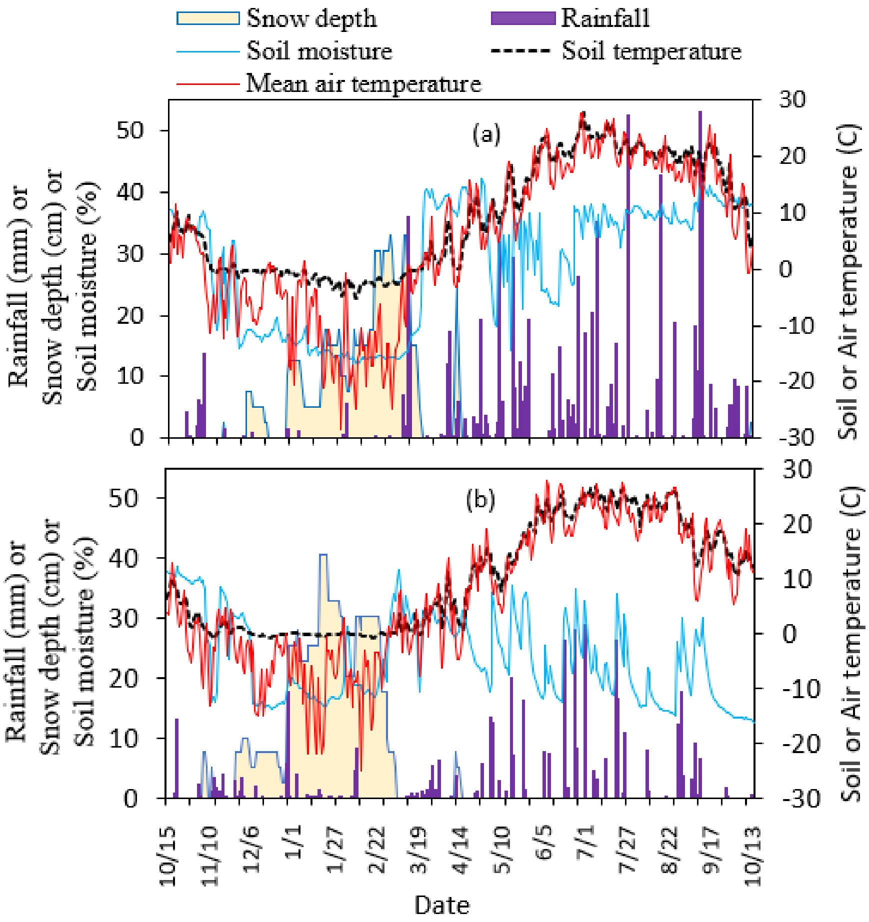


Figure 1. Daily distribution of snow depth, rainfall, air temperature, soil moisture, and soil temperature during first (Oct 2018- Oct 2019) (a) and second (Oct 2019- Oct 2020) (b) year of experiment. Data source: South Dakota Mesonet (2022).

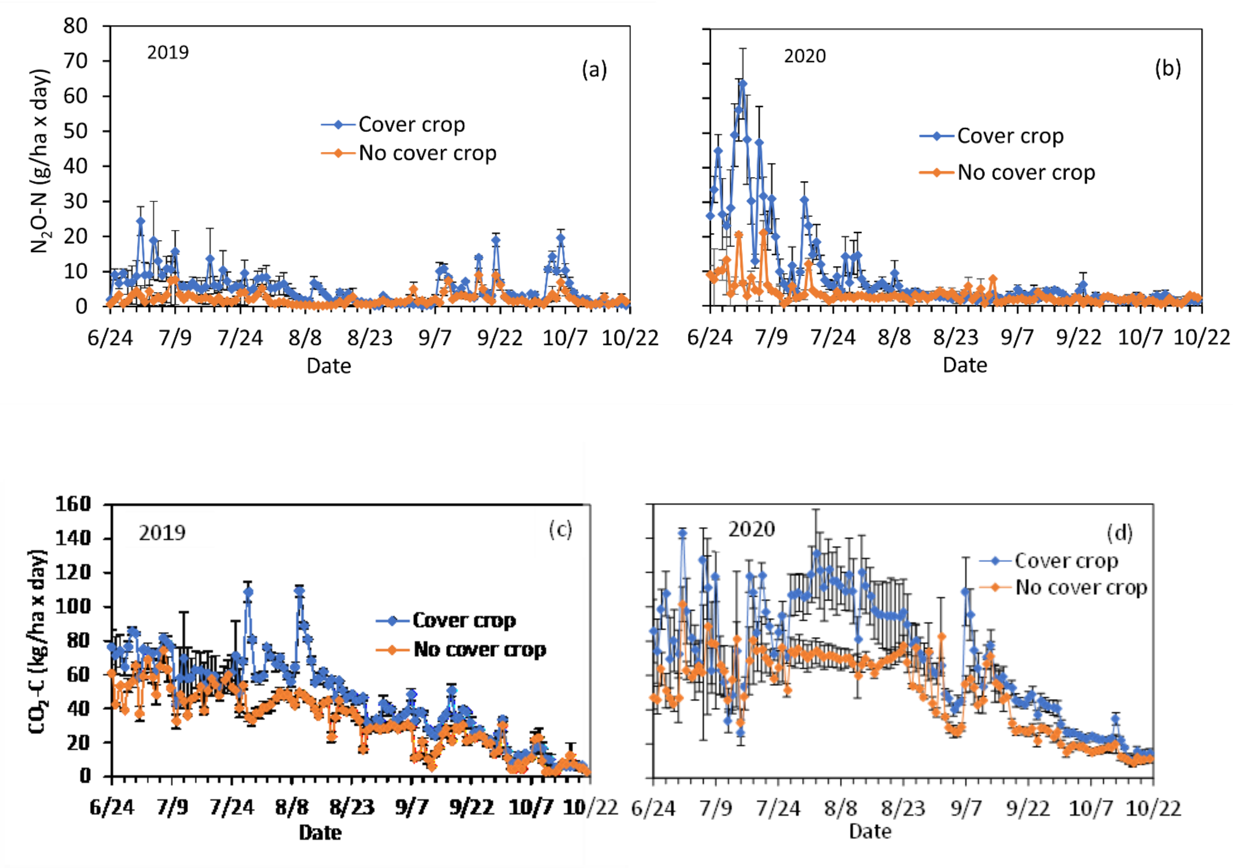


Figure 2. The impact of the rye cover crop on daily average N_2O-N (a and b) and CO_2-C (c and d) emissions in 2019 and 2020. Error bars represent standard error (SE) (n=4).

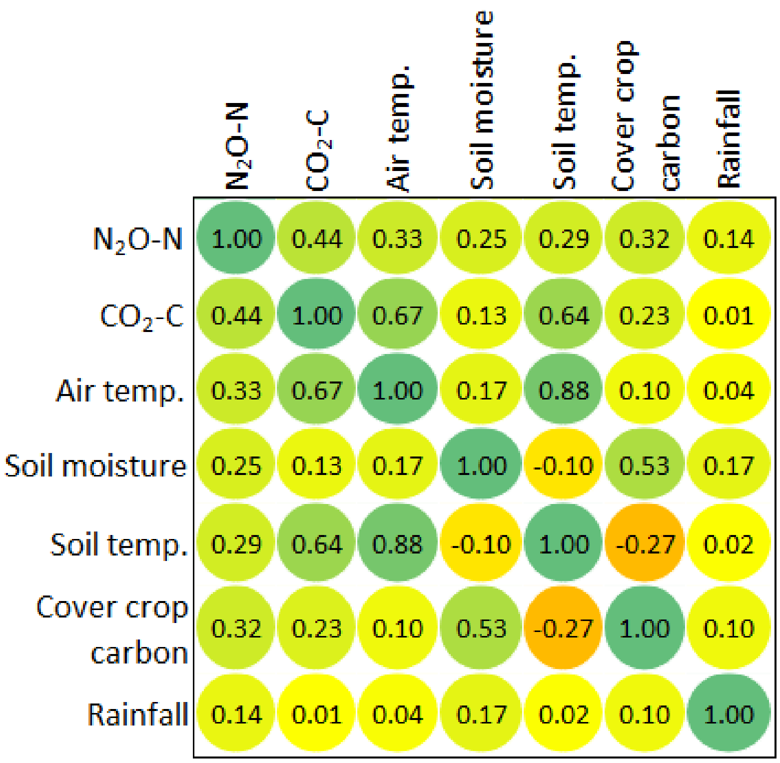
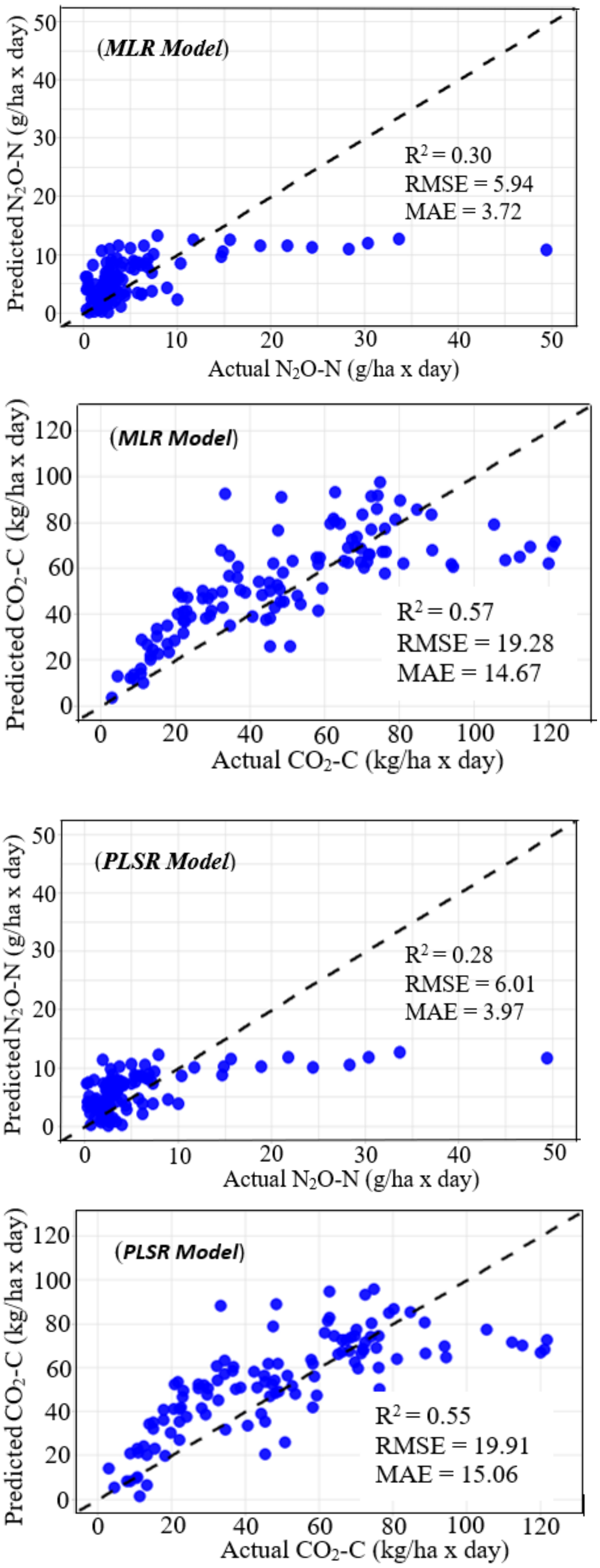
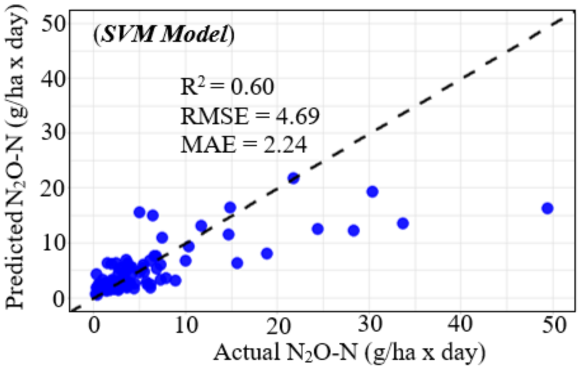
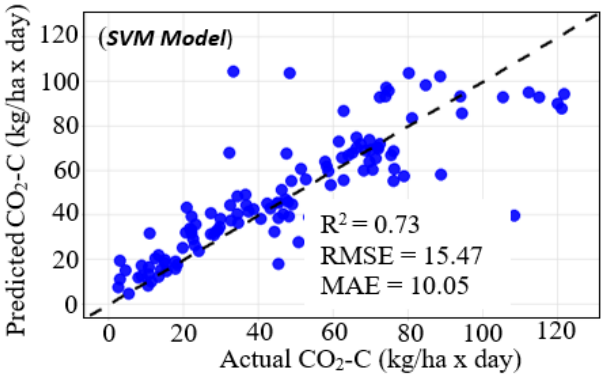


Figure 3. Correlation matrix between the different daily measurements in 2019 and 2020 (n=480). All correlation values (either negative or positive) equal or above 0.25 are statistically significant at p<0.001, between 0.13 to 0.17 are statistically significant at p=0.05 and values below 0.13 are not statistically significant. Positive values indicate positive relation whereas negative is just reverse.


(MLR Model)
R2 = 0.30
RMSE = 5.94
MAE = 3.72
Predicted N2O-N (g/ha x day)
Actual N2O-N (g/ha x day)
(MLR Model)
R2 = 0.57
RMSE = 19.28
MAE = 14.67
Predicted CO2-C (kg/ha x day)
Actual CO2-C (kg/ha x day)
(PLSR Model)
R2 = 0.28
RMSE = 6.01
MAE = 3.97
Predicted N2O-N (g/ha x day)
Actual N2O-N (g/ha x day)
(PLSR Model)
R2 = 0.55
RMSE = 19.91
MAE = 15.06
Predicted CO2-C (kg/ha x day)
Actual CO2-C (kg/ha x day)


(SVM Model)
R² = 0.60
RMSE = 4.69
MAE = 2.24
Predicted N₂O-N (g/ha x day)
Actual N₂O-N (g/ha x day)
0
10
20
30
40
50


(SVM Model)
R² = 0.73
RMSE = 15.47
MAE = 10.05
Predicted CO₂-C (kg/ha x day)
Actual CO₂-C (kg/ha x day)
0
20
40
60
80
100
120

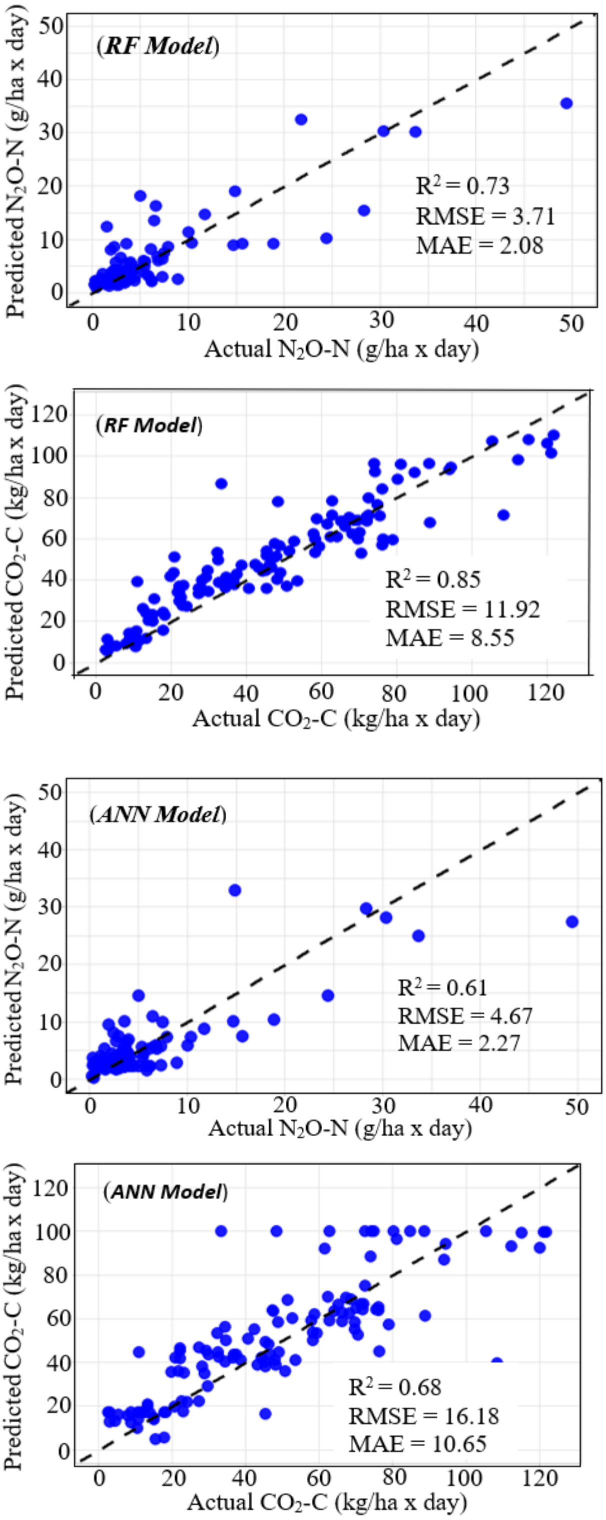
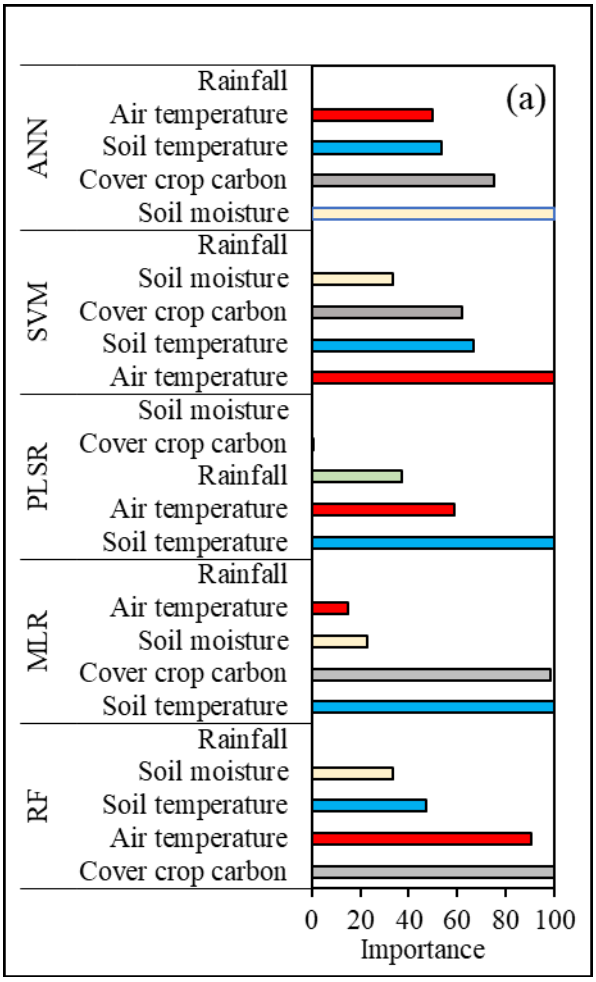


Figure 4. Validation of the actual vs. predicted N_2O-N and CO_2-C emissions using MLR, PLSR, SVM, RF and ANN models.


(a)
ANN
Rainfall
Air temperature
Soil temperature
Cover crop carbon
Soil moisture
SVM
Rainfall
Soil moisture
Cover crop carbon
Soil temperature
Air temperature
PLSR
Soil moisture
Cover crop carbon
Rainfall
Air temperature
Soil temperature
MLR
Rainfall
Air temperature
Soil moisture
Cover crop carbon
Soil temperature
RF
Rainfall
Soil moisture
Soil temperature
Air temperature
Cover crop carbon
0 20 40 60 80 100
Importance

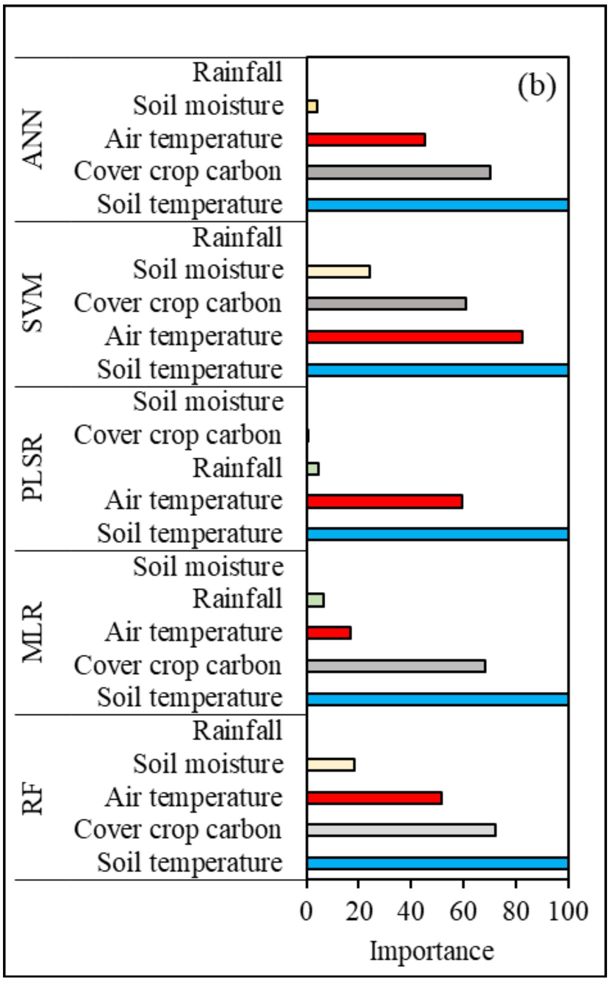


Figure 5. Relative importance of each variable used to model N_2O-N (a) and CO_2-C (b) emissions. Scaled importance score (0 to 100) was generated and higher scores indicate that the variable is of greater importance in the model.