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Growing Season Nitrous Oxide Emissions from a Gray Luvisol as a Function of Long-term Fertilization History and Crop Rotation

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Abstract

A field study conducted over three growing seasons (2013-2015) assessed the effect of long-term fertilization history and crop rotation on growing season nitrous oxide (N₂O) and carbon dioxide (CO₂) emissions, wheat yield, wheat N uptake, N₂O emission intensity and soil properties on Gray Luvisolic soils. Long-term fertility treatments included check, manure, NPKS, NPK and PKS fertilizers in two contrasting crop rotations: a 2-year of wheat (*Triticum aestivum* L.)-fallow (WF), and a 5-year wheat (*Triticum aestivum* L.)-oat (*Avena sativa*)-barely (*Hordeum vulgare* L.) - alfalfa (*Medicago sativa*)/brome (*Bromus tectorum*) hay (WOBHH). Rotation significantly affected cumulative growing season N₂O emissions and, within each rotation, long-term fertilizer or manure N additions increased N₂O emissions over the check. Average, cumulative growing season N₂O emissions from the 5-year rotation were 1.29 kg N₂O-N ha⁻¹, significantly higher than the 0.58 kg N₂O-N ha⁻¹ in the WF rotation, but N₂O emission intensities were comparable between to the two rotations. Cumulative N₂O emissions were positively correlated to total soil N (0-15 cm) and wheat N uptake, but N₂O emission intensities were negatively correlated to total soil N. However, it is uncertain whether this result extends to annual emission intensities because of a lack of non-growing season measurements.

Key words: Crop Rotation. Long-term Fertilization. Nitrous Oxide. Crop N Uptake. Nitrous Oxide Emission Intensity

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Introduction

Approximately, 60% of the current increase in atmospheric N₂O emanates from agricultural soils (IPCC, 2007). Significant progress has been made with respect to identifying processes responsible for the production and emission of N₂O from soils, but quantifying the relative contribution of the different processes and how agricultural management influences these processes remains a challenge (Butterbach-Bahl et al. 2013). There is significant evidence that N₂O emissions increase in direct proportion to the sum of fertilizer and non-fertilizer N inputs in agricultural and non-agricultural ecosystems (Bouwman 1996; Bouwman and Boumans, 2002; Liu and Greaver, 2013), but other factors such as soil moisture and temperature, fertilizer N type/placement, P, K and S fertilizer applications, crop type/rotation, inclusion of legumes, lime applications, microbial community diversity and tillage contribute to the variability of N₂O emissions (Bouwman 1996; Bouwman and Boumans, 2002; Snyder et al. 2009; Siciliano 2014; Butterbach-Bahl et al. 2013; Barton et al. 2013; Drury et al. 2006; Koga, 2013; Li et al. 2002; Wagner-Riddle et al. 2007). Currently, there is a knowledge gap with respect to the links between management practices and 1) N₂O production processes/emissions; and 2) soil properties affecting N₂O emissions and production processes.

The influence of long-term management (e.g., crop rotation, tillage, fertilizer applications) on soil properties and processes such as soil C and N stocks and cycling is well recognized (Richter et al. 2007). There are relatively few examples, however, of long-term management effects or management legacy effect on current annual or growing season N₂O emissions. Drury et al. (2014) observed a significant long-term rotation effect on N₂O emissions, showing that growing season N₂O emissions in the corn phase of a cereal-forage rotation were lower than in a continuous corn rotation with comparable N inputs (fertilizer and biologically fixed). Venterea et

al. (2011) showed a significant long-term tillage effect, showing that growing season N_2O emission intensities were greater under no-till management compared to conventional tillage. LaHue et al. (2015) demonstrated that N_2O emissions from zero-N controls were a function of previous N applications. Pearce (2016) demonstrated that land with historical applications of manure had greater growing season N_2O emissions following applications of urea compared to land with no history of manure application despite comparable historical N inputs. Thus, the legacy of long-term cropping practices and nutrient management appears to be important for understanding the magnitude and variability of current and future soil N_2O emissions.

Implementing management practices to increase crop recovery of applied N may reduce N_2O emissions as demonstrated in the examples cited the review of Snyder et al. (2009). However, van Groenigen et al. (2010) observed that higher crop yields and N uptake can be associated with higher N_2O emissions. In their meta-analysis, they observed that increased crop recovery of applied N decreased N_2O emissions per unit of yield or per unit of crop N uptake (i.e., N_2O emission intensity), but not total N_2O emissions. N_2O emissions did not increase with added fertilizer N as long as crop uptake of N matched or exceeded N application rates. N_2O emission intensities had a non-linear relationship with N fertilizer rate, decreasing between rates of 0 – 90 kg N ha⁻¹, followed by stable intensities between 90 – 180 kg N ha⁻¹ and then increasing at rates greater than 180 kg N ha⁻¹. Therefore, fertilizer management practices that increase crop recovery of applied N, thus avoiding N applications in excess of crop demands, have the potential to reduce N_2O emissions from agricultural land.

Applying fertilizer to address all nutrient deficiencies is key to maximizing crop recovery of applied N (Snyder et al. 2009), which likely reduces the risk of increased N_2O emissions associated with fertilizer N applications. For example, addition of phosphorus (P) fertilizer with

fertilizer N reduced the soil profile NO_3^- and improved the N recovery in long-term corn nutrition research plots in Kansas and adequate K substantially improved crop N recovery in corn in Ohio (Gordon 2005). Adequate S is required for efficient N use and high yield in annual and forage crops. Similarly, long-term applications of N and S increased dry matter forage yield, N and S uptake, and reduced nitrate leaching (Malhi and Gill 2007; Malhi et al., 2010).

In light of the abovementioned examples of management effects on soil nutrient stocks, cycling, plant uptake – all of which appear to be linked to N_2O – and the few examples of direct N_2O emission measurements from agricultural land under consistent, long-term management, the main objective of this research was to assess the effects of long-term rotation and fertilization growing season N_2O emissions, N_2O emission intensities (N_2O -N per grain yield and N_2O -N per crop N uptake) and the relationships between these variables. The secondary objective was to explore the effects of long-term rotation and fertilization on soil C and N stocks and their relationship to crop N uptake and growing season N_2O emissions.

MATERIAL AND METHODS

Study Site Description and Field Management

The Breton Classical Plots (BCP) agroecological experiment was first established in 1930 near Breton, AB ($53^{\circ} 07' \text{ N}$, $114^{\circ} 28' \text{ W}$) on Gray Luvisolic soils with moderate to poor drainage, slightly acidic pH, and a loam texture (Dyck et al., 2012). Soil properties such as total organic carbon (TOC) and total N (TN) will be discussed in the Results and Discussion section because they vary due to long-term rotation and fertilization. The mean annual air temperature and precipitation at the site are 2.1°C and 547 mm, respectively (Grant et al., 2001). The potential evapo-transpiration is 732 mm (Izaurrealde et al. 1995).

The BCPs consist of 8 fertility treatments superimposed on 2 rotations. The two rotations of the BCP are: 1) a 2-year wheat (*Triticum aestivum* L.)-fallow (WF) rotation; and 2) a 5-year wheat (*Triticum aestivum* L.)-oats (*Avena sativa*)-barley (*Hordeum vulgare* L.)- alfalfa (*Medicago sativa*)/brome (*Bromus tectorum*) hay (WOBHH) rotation. In each rotation, each crop is grown in all fertility treatments on an annual basis, but isn't replicated in space. The replications for this study are in time (across growing seasons).

The 8 fertility treatments are: 1) check (control or no fertilizer); 2) manure; 3) NPKS; 4) NKS(-P); 5) lime; 6) NPK(-S); 7) PKS(-N); and 8) NPS(-K). For the research presented in this paper, N₂O emissions were only measured on the wheat phase of the check, manure, NPKS, PKS(-N) and NPK(-S) treatments. Past and current nutrient rates for the fertility treatments that are the focus of this research are described in Table 1. Manure and fertilizer nutrients are broadcast and incorporated with a disc or rotary tiller in the spring prior to seeding, except for the manure treatment in wheat phase of the WOBHH rotation when manure applied in the previous fall during hay plough-down.

Gas Flux (N₂O and CO₂) Measurements

Gas (N₂O and CO₂) fluxes were measured in selected fertility treatments (Table 1) of both wheat and fallow phases of the WF rotation and in the wheat phase of the WOBHH rotation during the growing seasons of 2013 (14 measurements, 8 June to 29 August), 2014 (14 measurements, 22 May to 21 August) and 2015 (15 measurements, 15 May to 20 August).

The nonsteady-state chamber method (Rochette and Bertrand, 2008) and a 1312 photoacoustic, multi-gas monitor (Innova Air Tech Instruments, Ballerup, Denmark) was used to measure the gas fluxes between seeding and harvest each growing season. Rectangular chambers (65 cm x 16

cm x 15 cm) consisted of a chamber body and a detachable, vented lid with a small fan. The chamber bodies were inserted 5 cm into the soil, perpendicular to the crop rows, such that 10 cm remained above the surface. In 2013, chambers bodies were installed at two locations in each plot (6 plots x 2 rotations x 2 chambers; N = 24). For 2014 and 2015, 4 chamber bodies were installed in each plot (N=48).

During gas flux measurements, the lids were fastened to the chamber bodies using rubber bands and the fan was run to mix the atmosphere in the head space between gas concentration measurements. Gas measurements, from the head space of the chamber, were done in sets of 3 with sampling at 0, 9, 18, 27 min, after placing the lid on the collar.

Instantaneous gas (N₂O) fluxes (mg N₂O m⁻² min⁻¹) were calculated by the following equation (Rochette and Hutchinson, 2005):

$$F = \frac{dc}{dt} \times \frac{V}{A} \quad [1]$$

Where, dc/dt is the rate of change of gases concentration in the chamber head space (mg m⁻³ min⁻¹), V= Volume of chambers (m³), A=area covered by chambers (m²). Daily fluxes were estimated by multiplying the instantaneous flux by a conversion factor (1440 min day⁻¹). Linear interpolation was used to estimate a continuous time series with the estimated daily fluxes and cumulative growing season emissions were estimated by integrating the area under the estimated time series.

Soil Sample Analysis

In fall 2013, after crop harvest, soil samples were taken from east and west halves of check, manure, NPKS, NPK and PKS treatments of all phases of both crop rotations (WF and

WOBHH), at a depth of 0-7.5 and 7.5-15 cm (four locations per plot, using a shovel). After removing the coarse roots, plant residues and stones, the soil samples were air dried, at room temperature, and stored until further use. In the fall of 2015, a portion of each sample was sent to the University of Alberta Natural Resources Analytical Laboratory (NARL) in Edmonton, AB for analysis of total TOC, TN, total sulfur (TS), and pH using standard methods.

Yield and N uptake

At crop maturity, four 1-m² subsamples of each wheat plot half were harvested for grain and straw yield. The samples were dried at 60⁰ C for 7 days and threshed and a CNS combustion analyzer was used to determine TN from representative samples of both straw and seed (Malhi et al., 2010). Furthermore, seed (or straw) yield in kg ha⁻¹ x TN concentration (g N kg⁻¹) in seed (or straw) x 0.001 kg g⁻¹) was used to calculate N uptake (kg N ha⁻¹) (Malhi et al., 2010). Intensity of N₂O emissions were calculated by dividing cumulative growing season N₂O-N emissions by grain yield (kg N₂O-N kg grain⁻¹) or wheat N uptake (kg N₂O-N kg N⁻¹)

Meteorological Measurements

Daily air temperature, precipitation, soil temperature, and soil moisture at 5 and 20 cm depths were collected from Alberta Agriculture and Forestry weather data website, which is linked to the meteorological station at the Breton Plots.

Statistical Analysis

General linear model (GLM) procedures in the Minitab statistical software package (v. 17) were used to identify factors having a statistically significant effect on cumulative growing season N₂O and CO₂ emission estimates, wheat yield, wheat N uptake at harvest (grain + straw), N₂O

emissions intensities and selected soil properties. Because of the unconventional experimental design, the following model with year as a random effect and fertility treatments nested within rotation as fixed effects was used:

$$\text{Response} = \text{Year} + \text{Fertility Treatment} + \text{Year} * \text{Fertility Treatment} + \text{Rotation (Fertility Treatment)}$$

[3]

Because the soil properties included in this analysis were measured in only one year, the statistical model for soil properties was:

$$\text{Soil Property} = \text{Fertility treatment} + \text{Rotation (Fertility Treatment)} \quad [4]$$

Pairwise comparison of least-square means for the nested, Rotation(Fertility Treatment) factor using the Tukey test was carried when possible for significant model factors.

RESULTS AND DISCUSSION

Effects of Long-term Rotation and Fertility Treatment on Soil Properties

The long-term rotation and fertility treatments have resulted in significantly different levels of soil TOC, TN, pH, and to a lesser extent, total S (Table 2). All fertility treatments in the 5-year rotation have accumulated more TOC and TN than their counterparts in the 2-year rotation. The greater levels of soil N in the 5-year rotation are likely a result of more frequent and greater N inputs in the form of fertilizers (in NPKS, NPK treatments), manure (in the manure treatment), and biological fixation and crop residues (in all treatments). There is much more variability in soil total S, resulting in no statistical differences, but the magnitude of the relative differences in

mean total S between the fertility treatments in the two rotations were similar to the relative differences between TOC and TN.

Within each rotation, the long-term fertility treatments have also resulted in some significant differences in TOC and TN and TS levels. The manure treatments had higher levels of TOC, total TN and TS and all other treatments within each rotation, but the remaining treatments were not statistically different from each other.

Growing Season Conditions and Wheat Yields

Table 3 summarizes the growing season conditions for the 2013, 2014 and 2015 growing seasons. Mean growing season temperature, growing degree days (GDDs) and reference ET were very consistent between years, but growing season precipitation was significantly lower in 2015. Long-term averages for growing season precipitation, air temperature, and GDDs are 340 mm, 13.6 °C and 1066, respectively. Thus, all three years were drier and warmer than the long-term average, but 2015 was especially dry, resulting in poor growing conditions and dry soil conditions for most of June and July, 2015. Because the variability in timing of and amount of growing season precipitation between the three growing seasons, year was a significant factor manifested as significantly lower 2015 growing season N₂O and CO₂ emissions, wheat grain yields and N₂O emission intensities (Table 4).

The drought in 2015 reduced wheat yields compared to the previous 2 growing seasons, but the relative differences in yields between the rotations and fertility treatments within and between rotations was consistent over the three growing seasons, and wheat N uptake did not appear to decrease because of the drier conditions (Table 4). Despite similar total growing season precipitation in 2013 and 2014, yields were higher in 2014 because of more favorable seeding conditions in the spring of 2014. Soil moisture was near saturation for the majority of June, 2013

(the month following seeding), and this likely inhibited root development in early growth stages which impacted yields. Overall, higher yields and N uptake were observed in the 5-year rotation compared to the 2-year rotation, which is likely a reflection of greater cumulative inputs of fertilizer, crop residue and biologically fixed N over the long-term. Reynolds et al. (2014) also observed higher yields and improved soil quality in a corn-oat-alfalfa-alfalfa rotation compared to a continuous corn rotation.

There are also rotation effects with respect to wheat yield and N uptake response to applied nutrients (Table 4). Yield and N uptake in response to fertilizer N was much greater in the WF rotation compared to the 5-year rotation as observed by comparing the NPKS and PKS (-N) treatments (Table 5). Despite no fertilizer N applications in the PKS (-N) treatment of the 5-year rotation, significant N is added to the soil through biological fixation and residue incorporation of the alfalfa-brome hay phases. Comparing the NPKS and NPK (-S) treatments, wheat yield and N uptake was much more sensitive to applied S in the 5-year rotation compared to the 2-year rotation, because of high S removals in the alfalfa-brome phases of the 5-year rotation causing S deficiency for the following wheat phase in the absence of long-term S fertilization. In fact, wheat yields were more sensitive to S than N in the 5-year rotation as is apparent when the PKS (-N) and NPK (-S) treatments are compared (Table 5). Wheat yields and N uptake in the manure treatment of the WF rotation were consistently lower than the NPKS treatment (but not statistically different), but were equal in the 5-year rotation. The reason for this difference is not clear.

Growing Season N₂O and CO₂ Emissions

Year was a significant factor affecting cumulative growing season N_2O and CO_2 emissions, but the 5-year rotation had consistently higher emissions and the ranking of fertility treatment mean N_2O and CO_2 emissions within each rotation was fairly consistent across years (Table 6).

The variation in cumulative emissions between years can be explained by the unique environmental conditions in each year as shown in Figs. 1, 2 and 3. N_2O and CO_2 emissions were influenced by precipitation and soil moisture in each growing season. For example, in 2013, spring soil moisture was high (near saturation) resulting in a steady increase in cumulative N_2O emissions in the early growing season, possibly from denitrification while CO_2 emissions (i.e., soil respiration) were low until soil aeration improved following decreased soil moisture in late June/ early July (Fig. 1). Cumulative N_2O emissions increased again following a significant precipitation event of 35 mm in the second week of July and emissions were relatively constant of the remainder of the growing season as indicated by the consistent increase in cumulative emissions (Fig. 1).

In 2014, spring moisture conditions were not as wet as 2013 so there was no lag in increased, cumulative CO_2 emissions (soil respiration; Fig. 2). Weekly N_2O emissions were fairly consistent and similar between the two rotations until a significant precipitation event of ~35 mm in the third week of July after which cumulative N_2O emissions from the 5-year rotation responded much more than the WF rotation. Weekly emissions in both rotations decreased after the soil moisture content decreased to about 20% at the end of the growing season, but the decrease was lower in the WOBHH rotation than the WF rotation (Fig. 2).

In the 2015 growing season, precipitation was below the 3-yr (2013, 2014 and 2015) average (222 mm). As a result of low rainfall, soil moisture decreased more or less continuously from mid- May until mid-July (Fig. 3). Because of this, early growing season cumulative N_2O and

CO₂ emissions from both rotations were low and of similar magnitude. Following a number of small precipitation events in the second and third weeks of June (cumulative amount ~20 mm), soil moisture increased slightly and was followed by a large increase in air and soil temperature. Thus, starting in the last week of June, cumulative N₂O and CO₂ emissions from the 5-year rotation increased to a much greater extent than the WF rotation, though both rotations' emissions increased and were sustained by more regular rainfall (Fig. 3).

Rainfall, soil moisture content and soil temperature appear to be the main drivers for the difference in pattern of cumulative N₂O and CO₂ emissions from both rotations. Soil N cycling processes that produce N₂O – nitrification and denitrification – are sensitive to soil moisture and temperature. In the spring of 2013, there was a long period of near-saturated soil conditions and the high N₂O emission and a low CO₂ emission during this period is consistent with anaerobic denitrification. For the rest of the measurement record, although soil moisture reached near-saturated conditions following significant precipitation events, it was very short-lived. For this soil, 50% and 70% water-filled pore space (WFPS) correspond to 33% and 24% volumetric moisture content, respectively, suggesting that both nitrification and denitrification were the processes contributing to the N₂O emissions in all growing seasons. The significant relationship between soil average cumulative N₂O and CO₂ emissions (Fig. 4) could be interpreted as: 1) dominant aerobic soil conditions resulted in conditions favorable for soil respiration and nitrification that resulted in N₂O production via the hydroxylamine pathway (Siciliano 2014); 2) dominant aerobic soil conditions resulted in conditions favorable for soil respiration that created oxygen-depleted conditions in the soil pore space that resulted in N₂O production via nitrifier denitrification and anaerobic denitrification; or 3) a combination of 1) and 2). Therefore, growing seasons with sustained soil conditions favorable for denitrification or nitrification had

higher cumulative N_2O emissions. The distribution of rainfall and air temperature in 2014 created a series of soil wetting and drying cycles favorable for both nitrification and denitrification resulting in higher cumulative N_2O emissions than 2013 and 2015. In 2013, conditions were favorable for denitrification in the spring, but this was interrupted by a dry period where conditions were less favorable for denitrification. In 2015, dry soil conditions in the first half of the growing season were not optimal for denitrification or nitrification and this corresponds to the lower cumulative emissions in 2015.

Despite annual variations in soil and growing conditions, average, cumulative N_2O and CO_2 emissions over the three growing seasons were consistently higher in the 5-year rotation compared to the 2-year rotation (Table 6: Figs. 1, 2 and 3) likely because of greater N inputs over the long-term in the 5-year rotation (higher frequency of cropping and N inputs) and higher net mineralization and nitrification of previous forage residues in the 5-year rotation. In line with this, Chen et al. (2008) cited results from researchers and reported that previous crops in the rotation can contribute to an increase in easily mineralizable soil C and N, and this ultimately induced N_2O emissions. Further, Farrell et al. (2015) observed that crop residues accounted for a greater fraction of N_2O emissions than chemical fertilizers.

Differences between N_2O and CO_2 emissions of the fertility treatments within each rotation were much smaller than between rotations. The cumulative N_2O emissions of the manure treatment in the 5-year rotation were significantly higher than the check, but not significantly higher than the remaining fertility treatments (Table 6). Similarly, in WF, the cumulative N_2O emissions in the manure treatment were significantly higher than the PKS and check treatments, but were not significantly different from the NPKS and NPK treatments. Beauchamp et al. (1989) and Wagner-Riddle (1997) observed that compared to inorganic N fertilizers addition, addition of

equivalent rates of N in the form of manure had a much higher effect on N₂O emissions. Koga (2013) and Li et al. (2002) also confirmed this and reported higher N₂O emissions from plots fertilized with cattle manure than plots fertilized with chemical fertilizers. In addition, Meng et al. (2005) reported that since long-term application of animal manure increase organic carbon in agricultural soil, it could support microbially-mediated N₂O production.

N₂O Emission Intensity

Table 7 illustrates significant effects of crop rotation and long-term soil fertility treatments on N₂O emission intensity, and demonstrates the relationship between soil fertility treatments, crop rotation, N₂O emission and yield. In the 5-year crop rotation (WOBHH), N₂O emissions intensities from the fertility treatments ranged from 5×10^{-4} to 2×10^{-3} kg N₂O per kg grain. In the 2-year crop rotation (WF), N₂O emissions intensities from the soil fertility treatments were in the range of 10×10^{-4} to 12×10^{-4} kg N₂O-N per kg grain. In other words, both crop rotations showed comparable N₂O emissions intensity, indicating similar N₂O emissions from the production of 1 kg grain of wheat when using either of the two rotations, although the cumulative N₂O-N emission per ha was higher in 5-year rotation. Therefore, even though the N₂O emission intensity of WOBHH was similar to that of WF, WOBHH provided higher yield, and had an agronomic and economic advantage compared to WF.

The 34% higher yield in WOBHH compared to WF with similar N₂O emission intensities in both crop rotation systems is remarkable. A significant decrease in N₂O emission intensity, relative to the growing season cumulative N₂O emissions, in WOBH, combined with a significant increase in overall yield suggests that the increase emissions associated with higher N inputs are at least partially offset by increased agricultural production and crop N uptake which is potentially more

beneficial to society than low emission, low productivity (yield), high emission intensity cropping systems (van Groenigen et al., 2010). In line with this, Cui et al. (2013) suggested that in order to achieve sustainable agricultural production, a substantial increase in grain yield with efficient management of N fertilizer is crucial. Zhao et al. (2015) also promote incorporating management practices that reduce N₂O emission intensity through increased yields is favorable over reducing yields in order to reduced total N₂O emissions.

It is uncertain if the lower growing season emission intensities of the 5-year rotation would hold if annual emissions were considered because up to 70% of annual N₂O emissions can occur during the spring thaw periods (Lemke et al. 1998). Therefore, further investigation is required in the spring snowmelt and late fall seasons. Furthermore, even though it is not statistically different from the other fertility treatments, the NPKS soil fertility treatment in the WOBHH crop rotation, had the lowest N₂O emission intensities. This suggests that long-term balanced fertilization addressing all nutrient deficiencies will likely reduce N₂O emissions intensities through increased crop yields. Therefore, the implication of our results for nutrient management, in terms of reduction the greenhouse gases emissions, is similar to those promoting balanced fertilization.

Relationship between TN, N Uptake and Cumulative N₂O Emissions

Figure 5 shows the relationship between TN and wheat N uptake. Despite some variability, all soil fertility treatments in both rotations showed a linear increase in wheat N uptake with increasing TN and the relationship between TN and cumulative growing season N₂O emissions is shown in Fig. 6.

All soil fertility treatments under both rotations showed a linear increase in cumulative growing season N_2O emissions and wheat N uptake with increasing TN. Despite the annual variability making it difficult to observed differences in N_2O emissions between different fertility treatments within rotations, the significant slope of the regression line in Fig. 6 strengthens the observed long-term effect of crop rotation and fertilizer or manure applications on TN, N_2O emissions and crop N uptake from the GLM analysis, and this is corroborated by a similar observation made by Gomes et al. (2009). Moreover, some mechanisms may contribute to this effect, for example, if there is a surplus TN in the soil, it might serve (following mineralization) as a substrate for both microbial N_2O production processes (nitrification and denitrification) (van Groenigen et al., 2010) and provide plant-available N. Consequently, treatments with larger N stocks because of greater long-term N inputs had higher N_2O emissions. Greater N stocks can increase N mineralization and there by contribute to increased N_2O emissions and, simultaneously, crop N uptake (Gomes et al., 2009).

Synchronicity between crop N demand and available N supplied by soil and fertilizer should contribute to higher yields, crop recovery of applied N and reduce the risk of increased N_2O emissions. It is reasonable to hypothesize that N_2O emissions will decrease as crop N uptake increases because there may be less mineral N available to be converted to N_2O , but, in the present study, growing season cumulative N_2O emission showed a significant positive linear relationship with wheat N uptake, which suggests a greater supply of N compared to crop N demand (Fig. 7). Although the wheat N uptake in soil treatments in WOBHH were greater than the wheat N uptake in WF, WOBHH exhibited greater cumulative growing season N_2O emissions compared to WF suggesting that management effect on the soil N balance also influences N_2O emissions.

The relationships presented in Figures 5, 6 and 7 show the influence of long-term rotation and fertilizer applications on soil N stocks and cycling, crop N uptake and N₂O emissions. Within both rotations, treatments showing an increase in TN and TOC relative to the check/PKS treatments over time indicate a positive N balance with net, long-term N inputs exceeding N removals in harvested grain or hay. These treatments have consistently higher yields and crop uptake of N. Thus the observed positive relationship between N₂O and crop N uptake in Fig.7 may be a reflection of excess N supply relative to crop demand. However, it appears that crop N uptake increases as a function of TN at a greater rate than N₂O increases as a function of TN as indicated by the negative relationship between TN and the N₂O per wheat N uptake emission intensity parameter (Fig. 8). In the non-manure treatments, there is a negative relationship between TN and N₂O per crop N uptake emission intensity. Recalling the discussion about annual variations in growing season N₂O emissions (Fig. 1-3), temperature and moisture conditions that are favourable for N₂O emissions are also favorable for crop growth and increased N uptake and result in lower emission intensities. Therefore, we hypothesize that a negative relationship between cumulative N₂O (or TN) and crop N uptake may be observed in a scenario where urease and nitrification inhibitors are applied with fertilizers because of reduced cumulative N₂O emissions and increased crop uptake of N in the form of NH₄⁺ rather than NO₃⁻ (e.g. Venterea et al. 2016).

CONCLUSIONS

In the present study, although the cumulative growing season N₂O emissions were affected by long-term crop rotation as well as fertilization history, long-term crop rotation explains most of the variability of N₂O emissions from soil treatments. Overall, soil treatments and crop rotations with a higher N input increased the soil TN which had a significant, positive linear relationship

to cumulative growing season N₂O emissions. Although the five-year rotation had higher wheat yield and N uptake, the cumulative growing season N₂O emissions were greater than the WF rotation, resulting in comparable N₂O emission intensities between the two rotations.

Aerobic soil conditions during the growing season favored N₂O production through nitrification and wheat N uptake was linearly correlated with cumulative growing season N₂O emissions, suggesting that conditions favorable for N₂O emissions are also favorable for crop growth and N uptake. Therefore, the use of nitrification inhibitors might be the good choice to reduce growing season N₂O emissions while maintaining crop yields and N uptake (Venterea et al. 2016).

The findings of this study provide insights into the effect of long-term fertilization history and crop rotation management practices on wheat yield and growing season N₂O emissions in Gray Luvisolic soils in western Canada. Our results suggest that long-term crop rotation and soil fertility treatments which increase yield and crop N uptake reduce the N₂O emission intensity should be considered best management practices. Since it is a win-win solution for N₂O emission reduction and yield increase, application of fertilizer nutrients to address all nutrient deficiencies should also be considered in N₂O reduction strategies. However, since the N₂O measurements in this study were done in summer, it would be prudent to compare the annual emissions from these and other common rotations in multi-year experiments.

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Table 1. Treatment descriptions in the Breton Classical Plots study, where the gas (N₂O and CO₂) samples were taken.

Treatments 1930-1979 inclusive						Treatments, 1980 onward				
kg ha ⁻¹						kg ha ⁻¹				
Plot ^a	Treatment	N	P	K	S	Treatment	N	P ^d	K ^d	S
2	Manure	76	42	91	20	Manure	^e			
3	NPKS	10	6	16	10	NPKS	^b	22	46	^c
5/11	Check	0	0	0	0	Check	0	0	0	0
7	NPKSL	11	6	16	9	NPK	^b	22	46	0
8	P	0	9	0	0	PKS	0	22	46	^c

^a “Plot” refers to the original design of the experiment (Dyck et al., 2012) and is associated with the physical location of the fertility treatments as the Breton Plots. It is included here for clarity and to be consistent with previous publications.

^bN (applied as urea) rate depends on the crop and its place on rotation: wheat after forage (50 kg N ha⁻¹), oat or barley after wheat (75 kg N ha⁻¹), barley under seeded to hay: 50 kg N ha⁻¹ and legume-grass forages: 0 kg N ha⁻¹.

^cS is applied as elemental S at a rate of 5.5 kg ha⁻¹ from 1980 – 2007 and 20 kg ha⁻¹ from 2007 – present.

^dRates represent rates of the nutrient element rather than P₂O₅ and K₂O convention. P is applied as triple super phosphate (TSP) (0-46-0) and Potassium (K) is applied as muriate of potash (0-0-62).

^eComposted cattle manure with bedding straw incorporated; N application via manure depends on crop rotation. i.e., wheat-fallow: 90 kg N ha⁻¹ during cropped years, and cereal crops in WOBHH rotation: 175 kg N ha⁻¹ every 5 years applied in two equal applications. Actual manure rate is calculated using % TN measured on composite samples.

Table 2. Soil properties of soil at surface soils (0-15 cm) of fertility treatments within each rotation. Values are averages of samples taken from fertility treatment plots in all phases of the rotation (see Dyck et al. 2012). Within each column, different lower case letters indicate that the means are significantly different ($P<0.05$)

Crop rotation	Soil fertility treatment	TOC (Mg C ha ⁻¹)	TN (Mg N ha ⁻¹)	C:N ratio	TS (Mg S ha ⁻¹)	pH
WF	Check	19.0 d	1.8 d	10.6	0.34 b	6.4 a
	Manure	38.0 ab	3.6 ab	10.6	0.61 ab	6.6 a
	NPKS	22.0 cd	2.2 bcd	10.0	0.44 ab	5.9 abc
	NPK(-S)	19.0 d	2.0 d	9.5	0.37 ab	6.3 abc
	PKS(-N)	18.0 d	1.7 cd	10.6	0.37 ab	6.4 ab
WOBHH	Check	31.0 bc	2.9 bc	10.7	0.48 ab	6.0 ab
	Manure	47.0 a	4.4 a	10.7	0.60 a	6.2 a
	NPKS	35.0 b	3.3 b	10.6	0.52 ab	5.5 c
	NPK(-S)	33.0 b	3.1 bc	10.6	0.47 ab	6.0 ab
	PKS(-N)	34.0 b	3.2 bc	10.6	0.53 ab	5.6 bc

Note: TOC, TN: combustion; TS: nitric acid digest; pH, 5:1 CaCl₂.

Table 3: Summary of precipitation, air temperature, growing degree days and reference evapotranspiration (ET) during the growing season (2013-2015) at Breton Classical Plots.

Year	Growing season ^a precipitation (mm)	Average growing season air temperature (°C)	Growing season* growing degree days	Growing season reference ET (mm)
2013	258	14.0	1123	442
2014	249	13.8	1085	437
2015	160	14.3	1137	491

^aMay 1 – August 31

1 Table 4: Summary of the P value of the statistical model comparing the effect of year, fertility treatments, year*fertility treatments and
2 fertility treatment nested in rotation on cumulative growing season N₂O emissions, CO₂ emissions, wheat grain yield, wheat N uptake
3 (grain + straw), N₂O emissions per grain yield, N₂O emissions per wheat N uptake, TN, total soil C and soil pH.

Model Term	Growing season N ₂ O-N	Growing season CO ₂ -C	Wheat grain gyield	Wheat N uptake (grain + straw)	N ₂ O-N per grain yield	N ₂ O-N per wheat N uptake	TN (0- 15 cm)	Total soil C (0-15 cm)	Total soil S (0-15 cm)	pH
Year	< 0.001	< 0.001	0.006	0.154	0.010	0.090				
Fertility Trt	< 0.001	0.033	0.001	< 0.001	0.014	0.101	< 0.001	< 0.001	0.015	0.002
Year * Fertility Trt	0.981	0.525	0.081	0.232	0.405	0.056				
Rotation (Fertility Trt)	< 0.001	< 0.001	< 0.001	< 0.001	0.010	0.005	< 0.001	< 0.001	0.112	0.008
Model R ²	0.71	0.81	0.87	0.84	0.62	0.68	0.71	0.78	0.27	0.47

Table 5: Effect of soil fertilization history x crop rotation interaction on growing season grain yield and wheat N uptake.

Rotation	Fertility treatment	Grain yield (kg / ha)			
		2013	2014	2015	3-year average ^a
WF	Check	522	698	823	681 d
	Manure	1572	1314	1672	1520 bcd
	NPKS	2574	3052	1975	2534 ab
	NPK(-S)	1824	2377	2038	2079 abc
	PKS(-N)	516	622	786	641 d
WOBHH	Check	1244	1499	533	1092 cd
	Manure	2245	4152	2689	3029 a
	NPKS	2475	4349	2378	3067 a
	NPK(-S)	1761	3021	802	1861 c
	PKS(-N)	2364	2531	2060	2318 ab
Wheat N uptake – grain + straw (kg N/ ha)					
WF	Check	27	19	29	25 d
	Manure	71	37	58	56 bcd
	NPKS	116	88	79	95 ab
	NPK(-S)	89	73	87	83 abc
	PKS(-N)	24	16	30	23 d
WOBHH	Check	58	55	34	50 cd
	Manure	94	147	104	115 a
	NPKS	97	153	96	115 a
	NPK(-S)	68	93	53	72 bc
	PKS(-N)	81	69	86	79 bc

^aValues in the same column by the same letters are not significantly different at ($P < 0.05$) probability level.

8 Table 6: Effect of soil fertilization history x crop rotation interaction on growing season N₂O and
9 CO₂ emissions.

Rotation	Fertility treatment	Cumulative growing season N ₂ O-N emissions (kg N / ha)			
		2013	2014	2015	3-year average ^a
WF	Check	0.38	0.33	0.28	0.33 d
	Manure	0.78	1.18	0.42	0.79 bcd
	NPKS	0.48	1.11	0.42	0.61 cd
	NPK(-S)	0.70	0.64	0.72	0.74 bcd
	PKS(-N)	0.48	0.50	0.31	0.43 d
WOBHH	Check	1.14	1.44	0.61	1.06 bc
	Manure	1.77	1.49	1.61	1.62 a
	NPKS	1.75	1.19	0.94	1.36 ab
	NPK(-S)	1.67	1.37	0.96	1.23 ab
	PKS(-N)	1.44	1.40	0.70	1.18 abc
Cumulative growing season CO ₂ -C (kg C / ha)					
WF	Check	1521	1072	714	1102 d
	Manure	3186	2194	1361	2247 abc
	NPKS	1801	1600	1036	1479 cd
	NPK(-S)	2195	2024	1144	1788 bcd
	PKS(-N)	1922	1306	781	1336 cd
WOBHH	Check	3221	3320	1710	2750 ab
	Manure	4365	3496	1697	3186 a
	NPKS	4065	2983	1800	2949 a
	NPK(-S)	4115	3127	1630	2957 a
	PKS(-N)	3830	3504	737	2690 ab

10 ^aValues in the same column by the same letters are not significantly different at (P < 0.05)
11 probability level.

Table 7: Effect of soil fertilization history x crop rotation interaction on growing season on N₂O emission intensities (N₂O-N emissions per grain yield and N₂O-N emissions per N uptake).

Rotation	Fertility treatment	N ₂ O-N per unit of grain yield (kg N ₂ O-N/ kg grain) x 10 ⁻³			
		2013	2014	2015	3-year average ^a
WF	Check	1.04	0.39	0.39	0.61 ab
	Manure	0.59	0.44	0.24	0.42 ab
	NPKS	0.28	0.25	0.21	0.25 b
	NPK(-S)	0.52	0.47	0.22	0.40 ab
	PKS(-N)	1.14	0.95	0.36	0.82 ab
WOBHH	Check	0.96	0.95	1.17	1.03 a
	Manure	0.86	0.35	0.59	0.60 ab
	NPKS	0.70	0.31	0.42	0.48 b
	NPK(-S)	0.95	0.39	1.34	0.90 ab
	PKS(-N)	0.61	0.55	0.33	0.50 b
		N ₂ O-N per N uptake (kg N ₂ O-N/ kg N wheat) X 10 ⁻²			
		2013	2014	2015	3-year average ^a
WF	Check	1.86	1.48	1.14	1.49 abc
	Manure	1.30	1.58	0.68	1.19 abc
	NPKS	0.61	0.88	0.53	0.67 c
	NPK(-S)	1.05	1.51	0.52	1.03 bc
	PKS(-N)	2.51	3.76	0.92	2.40 ab
WOBHH	Check	1.96	2.64	1.82	2.14 a
	Manure	2.46	1.00	1.54	1.50 abc
	NPKS	1.83	0.89	1.09	1.27 bc
	NPK(-S)	2.46	1.27	1.80	1.85 abc
	PKS(-N)	1.78	2.05	0.81	1.54 abc

^aValues in the same column by the same letters are not significantly different at (P < 0.05) probability level.

Figure 1: Daily precipitation, daily average air temperature, daily volumetric water content at 5 cm, daily soil temperature at 5 and 20 cm, and gas fluxes (N₂O and CO₂) over the 2013 growing season.

Figure 2: Daily precipitation, daily average air temperature, daily volumetric water content at 5 cm, daily soil temperature at 5 and 20 cm, and gas fluxes (N_2O and CO_2) over the 2014 growing season.

Figure 3: Daily precipitation, daily average air temperature, daily volumetric water content at 5 cm, daily soil temperature at 5 and 20 cm, and gas fluxes (N₂O and CO₂) over the 2015 growing season.

Figure 4. Relationship between cumulative growing season N_2O emissions and cumulative growing season CO_2 emissions averaged over the 2013, 2014 and 2015 growing seasons from soils with different fertilization history under wheat-fallow (WF) and wheat-oat-barley-hay-hay (WOBHH) crop rotations at Breton Plots, Western Canada. Symbols represent mean values and error bars represent 1 standard error. The line represents the orthogonal regression relationship: $y = 1858x + 507$. The slope and intercept are both highly significant ($P < 0.001$).

Figure 5: Relationship between wheat N uptake (grain +straw) and TN (0-15 cm) of soils with different fertilization history under wheat-fallow (WF) and wheat-oat-barley-hay-hay (WOBHH) crop rotations at Breton Plots, Western Canada. Symbols represent mean values and error bars represent 1 standard error. The line represents the orthogonal regression relationship: $y = 34.17x - 25.45$. The slope is significantly different from zero ($P < 0.05$), but intercept is not ($P = 0.59$).

Figure 6: Relationship between cumulative growing season N_2O emissions and TN (0-15 cm) of soils with different fertilization history under wheat-fallow (WF) and wheat-oat-barley-hay-hay (WOBHH) crop rotations at Breton Plots, Western Canada. Symbols represent mean values and error bars represent 1 standard error. The line represents the orthogonal regression relationship: $y = 0.45x - 0.33$. The slope is significantly different from zero ($P < 0.001$), but intercept is not ($P = 0.21$).

Figure 7: Relationship between cumulative growing season N_2O emissions and the wheat N uptake (grain + straw) from soils with different fertilization history under wheat-fallow (WF) and wheat-oat-barley-hay-hay (WOBHH) crop rotations at Breton Plots, Western Canada. Symbols represent mean values and error bars represent 1 standard error. The line represents the orthogonal regression relationship: $y = 81.9x - 5.7$. The slope is significantly different from zero ($P < 0.01$), but intercept is not ($P = 0.83$)

Figure 8: Relationship between TN (0-15 cm) and N_2O -emission intensity from soils with different fertilization history under wheat-fallow (WF) and wheat-oat-barley-hay-hay (WOBHH) crop rotations at Breton Plots, Western Canada. Symbols represent mean values and error bars represent 1 standard error.