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- 3 receiving Liquid Manure Injection in the Fall versus Spring
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- 18 A key participant in this research project is Ms. Sisi Lin; the graduate student in the project with
- 19 my supervision. Her tremendous contributions made this project possible.
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1 Executive Summary

2 Nitrous oxide (N₂O) contributes to global warming and ozone depletion. Two-thirds of the
3 global N₂O emissions are derived from agricultural soils receiving manure or fertilizer
4 applications. The goal of this project was to identify and develop management practices that can
5 decrease N₂O emissions from cropland receiving liquid manure. We tested early fall versus late
6 spring application of liquid manure in combination with two nitrification inhibitors (NIs;
7 nitrapyrin vs. DMPP) admixed with the liquid manure. Two field experiments in central Alberta,
8 Canada. Barley for silage was planted, and productivity and N uptake were recorded. Soil
9 ammonium and nitrate concentrations and N₂O fluxes were repeatedly monitored. Compared to
10 fields without manure controls, field N₂O emissions were increased with by manure application
11 (3.15 vs. 0.45 kg N ha⁻¹ yr⁻¹), but emissions were sharply reduced with NIs. For instance, in the
12 Lacombe site, fall manure treated with DMPP reduced annual N₂O emissions by 81%, and
13 nitrapyrin reduced emissions by 58%. The emission reductions caused by NIs were also evident
14 in the spring manure field treatments and at our Edmonton site, but the reductions magnitudes
15 were typically smaller in associations with periods exhibiting drier conditions in particular in
16 Edmonton. Compared to the spring manure timing, fall manure without NIs resulted in an
17 approximate two-fold increase in N₂O emissions, due to major peak fluxes following the early
18 spring snow-melt, which accounted for at least 65% of the annual N₂O emissions. Fall manure
19 timing also reduced plant productivity and N uptake. In sum, spring manure with NIs can
20 mitigate N₂O emissions in Alberta's agriculture and in regions with comparable agro-ecological
21 conditions.

22 Keywords: nitrous oxide, liquid manure, nitrogen, nitrification inhibitors, nitrogen

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25 report are not necessarily the views of the Emissions Reduction Alberta (ERA)

26

1 Introduction and Project Overview

2 Agriculture and livestock act as important sources of greenhouse gases to the atmosphere.
3 Moreover, escalating climate change can further increase greenhouse gas emissions from
4 agricultural systems to the atmosphere. One of the predominant biogenic greenhouse gases
5 emitted from agricultural landscapes is nitrous oxide (Granli and Bøckman 1994; van Kessel et
6 al. 2013). Globally, 58 % of the agriculturally-derived greenhouse gas emissions is nitrous oxide,
7 and two-thirds of these nitrous oxide emissions occur in cropping systems where synthetic
8 nitrogen fertilizer or manure are recurrently added to the soil. In fact, increased nitrous oxide
9 emissions are an indication of inefficient nitrogen utilization in production systems (Cassman et
10 al. 2002). This is important as N is one of the most expensive agricultural inputs, and hence,
11 environmental N losses become directly connected to the efficiency and economic viability of
12 the production system.

13

14 The abundance and high concentration of manure in Alberta is an environmental issue faced by
15 various animal production systems such as swine, dairy, and beef. Adequate land disposal of
16 manure can be a feasible solution as manure can represent a source of nitrogen for enhanced
17 plant productivity (Hernandez-Ramirez 2009). However, manure also adds available organic
18 carbon to the soil which can accelerate some of the microbial processes that lead to nitrous oxide
19 emissions.

20

21 In soils, recently incorporated nitrogen can form ammonia and ammonium in dynamic
22 equilibrium within the aqueous phase. This equilibrium and transformation depends on
23 environmental factors including temperature, soil texture, water content, oxygen concentration in
24 soil air, pH, and available organic carbon. In non-flooded soils, the N transformation continues
25 with fast ammonium oxidation to nitrate via autotrophic nitrification. Nitrate can be subjected to
26 heterotrophic denitrification. Both nitrification and denitrification can separately produce nitrous
27 oxide which is stable and prone to gaseous loss (Baggs 2011; Bremner 1997). To prevent this
28 outcome, nitrification inhibitors can be used to slow nitrification by inhibiting the enzymatic
29 activity of ammonia monooxygenase, hence maintaining nitrogen as ammonium available for

1 plant uptake, and indirectly avoiding denitrification (Arp and Stein 2003). Three available
2 nitrification inhibitors with potential beneficial effects are nitrapyrin, DMPP (3,4-
3 dimethylpyrazole phosphate), and dicyandiamide (Subarrao 2006). Although this theoretical
4 knowledge has been developed in various countries, there has not been a complete experimental
5 confirmation of these various shifts in nitrogen transformation pathways as a response to
6 additions of nitrogen stabilizers in fields receiving fall vs. spring manure in Canada. We
7 anticipate that the use of these inhibitors as part of improved management systems will create
8 opportunities to effectively reduce nitrous oxide emissions, making manure disposal more
9 effective as a fertilizer (nutrient source) and less of an environmental issue.

1 Project Goals

2 The objective of this study is to identify and develop best management practices for manure
3 injection into soils with specific focus on efficiency of nitrification inhibitors, timing of manure
4 additions, associated quantities of nitrous oxide losses, and plant nutrient utilization.

5

6 Our continual research vision is reflected in our long-term objective:

7 - To reduce nitrous oxide emissions from agricultural landscapes where manure is applied, and in
8 this way to enhance both the reputation of the industry and farming profitability, and ultimately,
9 to mitigate greenhouse gas emissions and associated climate change. We will specifically
10 address the following short-term research objectives:

11 - To examine if nitrification inhibitors can consistently reduce nitrous oxide emissions from soils
12 receiving manure.

13 - To quantify what nitrous oxide reduction level is feasible.

14 - To estimate a reasonable/realistic emission reduction coefficient.

15 - To elucidate which manure management practices lead to increased effectiveness of
16 nitrification inhibitors (by contrasting manure-inhibitor injection during the cold fall season with
17 minimal plant growth vs. the wet warm spring season with increased biological activity).

18 - To quantify the contribution of spring thawing to the overall annual emissions under various
19 manuring practices.

20 - To assess whether nitrification inhibitors can enhance nutrient use efficiency by crops (barley
21 for silage) following manure injections.

22

1 Project Final Outcomes

2 Literature review

3 With an increasing global demand of livestock products (Herrero et al. 2009), the appropriate
4 disposal of the abundant manure has become a great concern. One feasible solution is to apply
5 the manure to the soil since the manure is conducive to biomass productivity increases as a
6 source of N (Hernandez-Ramirez et al. 2009a). However, this practice can lead to serious
7 environmental issues at the same time (Basso and Ritchie 2005; Webb et al. 2010). It has been
8 reported that manured or synthetic N fertilized soils account for 67% (1851.3 Tg CO₂e) of global
9 total nitrous oxide (N₂O) emissions from agriculture in 2005 (USEPA 2012). N₂O plays a
10 dominant role in global warming with a 100-year global warming potential (GWP) value of 298
11 (GWP for CO₂ is 1) (Myhre et al. 2013), as well as in stratospheric ozone depletion
12 (Ravishankara et al. 2009).

13 In soils, autotrophic nitrification (Bremner and Blackmer 1978; Butterbach-Bahl et al. 2013) and
14 denitrification (Braker and Conrad 2011) are two major pathways to generate N₂O. Soil moisture
15 is one of the crucial environmental drivers for N₂O fluxes (Zheng et al. 2000), since it is
16 interrelated with the oxygen consumption by the soil microbial community (Schindlbacher et al.
17 2004; Meixner and Yang 2006), and oxygen serves as an electron acceptor in nitrification
18 (Velusamy and Krishnani 2013). Autotrophic nitrification tends to be a predominant pathway in
19 producing N₂O under a lower water-filled pore space (WFPS) (i.e., 35-60%) (Davidson and
20 Schimel 1995; Bateman and Baggs 2005) . It has been found that the denitrification rate
21 increased with a higher soil water content (Luo et al. 2000), and the end product depends on the
22 level of anaerobic condition. It has been proposed that the maximum amount of dinitrogen (N₂)
23 and greatest decrease of N₂O were observed at an absolutely restricted aeration condition (i.e.,
24 ~2% O₂ v/v) (Morley and Baggs 2010). Additionally, soil temperature is another dominant
25 contributor for N₂O fluxes. As the temperature increased by 10 °C, the N₂O production was
26 observed to double (Phillips et al. 2015) .

27 The identification of best management practices for manure applications provides potential
28 opportunities to mitigate N₂O emissions, and address the global warming effect. Liquid manures

1 have been suggested to be injected into the soil profile instead of surface spreading since this
2 incorporation not only decreases ammonia volatilization (Laboski et al. 2013) but also increases
3 the proportion of N₂ derived from denitrification (Smith and Mukhtar 2015) . Moreover, with the
4 purpose of enhancing fertilizer-N usage efficiency, the addition of nitrification inhibitors (NIs)
5 with manure applications has been proposed (Zerulla et al. 2001). Effective NIs widely used in
6 agriculture include nitrapyrin, dicyandiamide (DCD) and DMPP (Subbarao et al. 2006). The
7 function of NIs is to delay the oxidation reaction by depressing the nitrifiers (Subbarao et al.
8 2006); hence the subsequent process, denitrification, would be restricted by a low concentration
9 of nitrate as substrate (Saggar et al. 2013), preventing gaseous N losses. The majority of both
10 dairy and hog manure is typically applied to the soil in the spring and fall seasons in temperate
11 regions (Beaulieu 2004), thus N losses from manure could be hypothetically driven by manure
12 application timing (Chadwick et al. 2011). A study estimated the nitrate losses by leaching and
13 found that a fall/winter slurry application resulted in higher nitrate losses than a spring slurry
14 application (Van Es et al. 2006). Another recent study found that the fall/winter cattle slurry
15 application increased direct N₂O emissions from free draining grassland soils in England
16 compared to a spring application (Thorman et al. 2007) .

17 Even though several earlier studies have examined the effect of manure application timing on
18 N₂O emissions (Allen et al. 1996; Weslien et al. 1998; Rochette et al. 2004), it is still unclear if
19 the addition of NIs would amplify, narrow, or even eliminate the difference in N₂O emissions
20 between the fall and spring manure applications.

21

22 Meetings with potential industry sponsors

23 Sisi Lin / Guillermo Hernandez and seven coauthors. Reducing Greenhouse Gases Emissions by
24 using Nitrification Inhibitors in Field Manure Applications Future Fare – ALMA,
25 Edmonton, AB 16 June 2014 500

26 Sisi Lin / Guillermo Hernandez. Preliminary Assessment of Nitrous Oxide Emissions, Soil
27 Temperature and Soil Oxygen Concentrations following Fall Manure Injections. Alberta
28 Agriculture - Field visit to University of Alberta South Campus. 8 June 2015

1 Sisi Lin / Guillermo Hernandez and seven coauthors Reducing Greenhouse Gases Emissions by
2 using Inhibitor Additives at varying rates in Field Manure Applications Future Fare –
3 ALMA, Edmonton, AB 13 October 2016 300

4

5 Experimental procedures/methodology

6 Two field sites were established in Lacombe (52°27'17''N, 113°44'20''W) and Edmonton
7 (53°29'30''N, 113°31'53''W), Alberta, arranged in a split-plot experimental design. Treatments
8 were replicated four and t in Lacombe and Edmonton, respectively. Soil classifications are
9 Orthic Black Chernozem for Lacombe and Black Chernozemic for Edmonton. Prior to treatment
10 establishment, the soil in Lacombe had a sandy clay loam, clay to clay loam texture, a 1.22 g cm⁻³
11 bulk density and a 7.0 pH, and the soil in Edmonton had a clay to heavy clay texture, a 1.15 g
12 cm⁻³ bulk density and a 6.1 pH. The climate of these two sites is semi-arid continental.

13 Two types of NIs were admixed and applied with the liquid manure: 2-chloro-6-
14 (trichloromethyl) pyridine (nitrapyrin) and 3, 4-dimethylpyrazole phosphate (DMPP). Six manure
15 treatments and two controls (without any manure added) were established at each experimental
16 site. The eight treatments were: control where the soil disturbed using the manure injector (CT),
17 control without any soil disturbance (CZ), fall-manured soil with no NIs added (FW), fall-
18 manured soil with DMPP (FD), fall-manured soil with nitrapyrin (FN), spring-manured with no
19 NIs added (SW), spring-manured soil with DMPP (SD), and spring-manured soil with nitrapyrin
20 (SN).

21 A coulter manure injector was used to establish CT and also to apply the liquid manure in the six
22 treatments receiving manure. All injections were conducted at a constant volume rate of 74.14
23 m³ manure ha⁻¹ and 0.5 kg active compound NIs ha⁻¹. The NIs were evenly added and
24 mechanically agitated with the liquid manure prior to manure injections. The injector created
25 ~2.5 cm width and ~12.7-15.2 cm deep injection bands, as well as ~28 cm spacing between
26 consecutive bands. Manure samples were taken at each time of manure applications for total N,
27 NH₄⁺-N and water content analyses. Barley for silage was planted at 300 seeds m⁻². To quantify
28 above ground biomass and plant N uptake, the crop was harvested in Lacombe and Edmonton

1 using a forage harvester, respectively. Crop phenology was recorded and photos were taken for
2 each treatment on the dates of N₂O flux measurements.

3 A manual static chamber method (Hernandez-Ramirez et al. 2009a) was used to measure the
4 field N₂O flux at both sites. The chamber bases (15 cm in height, 65.5 cm in length and 17.0 cm
5 in width) were installed in the middle of each plot inserted in the soil with a depth of 5 cm
6 perpendicular to the manure injection rows. Each chamber encompassed two manure injection
7 rows. The flux measurement frequency was two times weekly after manure injections, major
8 precipitations or during early spring-thawing period; otherwise, it was one flux measurement per
9 week. In order to improve the consistency of gaseous flux estimations, gas samples were
10 collected within the period between 11 am and 3 pm. Three gas samples were collected for each
11 chamber at 16, 32 and 48 minutes. To represent the time zero of chamber closure, three ambient
12 gas samples were randomly collected on each date of flux measurements at 10 cm above the
13 ground surface.

14 The field N₂O flux rate was determined by plotting a linear or a quadratic relationship between
15 measured N₂O concentrations versus time (as mentioned above, concentrations in ambient gas
16 samples were assumed as time 0). Zero N₂O flux rate was assumed if there was a non-significant
17 relationship (this statistical decision followed an alpha critical threshold of 0.20); otherwise the
18 flux rate was calculated by the modified ideal gas law as follows:

$$Flux = \frac{S * P * V * A^{-1}}{R * T} \quad [2-1]$$

19 where Flux is the N₂O flux rate ($\mu\text{mol min}^{-1} \text{m}^{-2}$); S is identified as the slope of the line from a
20 simple linear regression, or the first-order derivative at a certain time for a quadratic regression
21 curve (Yates et al. 2006; Pennock et al. 2010) ($\mu\text{L L}^{-1} \text{min}^{-1}$); P is the pressure of the gas (atm);
22 V is the volume of the gas chamber (L); A is the surface area of the gas chamber (m^2); R is the
23 gas constant ($\text{atm } \mu\text{L K}^{-1} \mu\text{mol}^{-1}$) and T is the temperature of the gas (K).

24 We recorded that 72, 18 and 10 % of the N₂O flux measurements in Lacombe were calculated by
25 linear, quadratic and zero regressions, respectively. Likewise, 79, 13 and 8% of the N₂O flux
26 measurements in Edmonton were calculated by linear, quadratic and zero regressions,
27 respectively. The cumulative emissions between two consecutive sampling dates were assumed

1 to equal the product of the average N₂O flux rate and the time interval between the two dates.
2 For the estimation of annual cumulative emissions (Fig. 0-1), flux quantities were assumed to be
3 negligible during the winter months (e.g., November to March) due to freezing ambient
4 temperatures leading to minimal soil biological activity and gaseous transport processes.

5 We conducted repeated soil samplings at the Edmonton site to determine any differences in the
6 temporal changes of ammonium (NH₄⁺-N) and nitrate (NO₃⁻-N) concentrations as a function of
7 experimental treatments and environmental conditions. Composite (≥ 3) 0-15 cm soil samples
8 were collected from each plot using a push probe (i.d. 2.5 cm). As the coulter manure injector
9 creates injection bands, soil samples were collected from both the injection row and non-
10 injection areas. The collected soil samples were stored at 5°C. Prior to the laboratorial analyses,
11 soil samples were air-dried and grinded through a 2 mm sieve.

12 We deployed 5TM sensors and Em50 data loggers to measure the average half-hour soil
13 temperature and moisture content data for each field treatment in the Edmonton site. The
14 installation of the sensors and data loggers was accomplished within 24 hours following manure
15 injections. Two 5TM sensors were installed in each plot horizontally at 10 cm and 20 cm below
16 ground surface. The middle prong of the sensor was established at these target soil depths. The
17 ECH2O utility software was used to collect the soil temperature and moisture content data.

18 The N₂O concentration in gas samples was measured by an electron capture detector in a Laurier
19 Varian 3800 gas chromatograph. The minimum analytical detectable N₂O flux was 2.84 g N ha⁻¹
20 day⁻¹. A 2M KCl solution was used to extract the NH₄⁺ and NO₃⁻ from the soil samples;
21 subsequently, the filtrate was colorimetrically evaluated using a SmartChem discrete wet
22 chemistry analyzer. Aboveground barley dry matter biomass was determined using oven dry
23 weight, and barley N concentration was quantified using near infrared spectroscopy (Hernandez-
24 Ramirez et al. 2011).

25 The Shapiro-Wilk test was conducted to examine the data normal distribution. The Bartlett or
26 Levene tests were conducted to examine homogeneous variances. The Box-Cox Power
27 transformation was used if the data did not fulfill these assumptions. One-way analysis of
28 variance (ANOVA) and a Fisher's least significant difference (LSD) test were conducted to
29 assess differences in (1) total N, ammonium and water content of the applied liquid manure

1 among the applications; (2) dry matter yield and plant N uptake among CZ, CT, FW and SW
2 treatments; (3) dry matter yield and plant N uptake among fall-manured soils or spring-manured
3 soils; (4) annual cumulative N₂O emissions among CZ, CT, FW and SW treatments and (5)
4 annual cumulative N₂O emissions among fall-manured soils or spring-manured soils at both
5 sites. The differences in total N, ammonium and water content in the applied liquid manure
6 between two sites were determined by a two sample t test. All statistical tests for treatment
7 effects were performed at a 90 or 95% confidence interval. All analyses were analyzed using the
8 version 3.1.3 R software. A split-plot linear model was applied to test the effect of manure
9 timing, nitrification additives and their interactions on annual cumulative N₂O, plant dry matter
10 yield and plant N uptake.

11

12 Results and analysis of experiments

13 Cumulative N₂O emissions were measured in both Lacombe and Edmonton sites to investigate
14 the effects of manure injection timing and nitrification inhibitors (NIs) (Fig. 2 1). Compared to
15 the control treatments (average 0.3 ± 0.1 and 0.6 ± 0.2 kg N ha⁻¹ yr⁻¹ in Lacombe and Edmonton,
16 respectively), adding the fall liquid manure without NIs (FW) resulted in a significant increase in
17 annual N₂O emissions (Fig. 2 1). This significant difference, nevertheless, was not found in the
18 spring manure without NIs (SW) at both sites (Fig. 2 1). Even though there was no statistically
19 significant effect of manure injection timing on the annual N₂O emissions (Table 2 1 and Fig. 2
20 1), the amount of annual N₂O emissions derived from fall manure without NIs was more than
21 double than that from spring manure without NIs at both sites (i.e., 6.2 ± 3.7 vs. 3.1 ± 1.0 in
22 Lacombe and 2.3 ± 0.7 vs. 1.0 ± 0.2 kg N ha⁻¹ yr⁻¹ in Edmonton, respectively).

23 Based on ANOVA results, there was a significant effect of NIs on the annual cumulative N₂O
24 emissions in our Lacombe site ($P < 0.05$), whereas there was no such significant effect in
25 Edmonton (Table 2 1 and Fig. 2 1). At the Lacombe site, the magnitude of our calculated
26 reduction coefficients (Eq. [2-2]) for the fall manure treatments with DMPP (FD) and nitrapyrin
27 (FN) were 81.0 and 57.8%, respectively (Fig. 2 1). Furthermore, the reduction coefficients were
28 less pronounced for the spring manure treatments (i.e., 64.3% for DMPP, and 32.7% for

1 nitrapyrin; Fig. 2 1). Overall, it should be highlighted that all soils receiving NIs resulted in
2 annual mean N₂O emissions consistently similar to or lower than a magnitude of 2.6 kg N ha⁻¹
3 yr⁻¹(Fig. 2 1). Moreover, when contrasting our field sites, the annual N₂O emissions derived
4 from both the fall and spring manure without NIs (FW and SW treatments) for Lacombe were
5 about three times larger than for Edmonton (Fig. 2 1). Additionally, N₂O emissions for all fall
6 manure treatments (FW, FN and FD) from 27 Mar to 10 Apr 2015 corresponded to 78% of the
7 annual emissions in the Lacombe site (Fig. 2 2b), and emissions from 27 Mar to 14 Apr 2015
8 represented 65% of the annual emission in the Edmonton site (Fig. 2 3b). Substantial N₂O
9 emissions from all spring manure treatments were also found at both sites following the spring
10 manure injections (Fig. 2 2b and Fig. 2 3b) when the average monthly temperature was around
11 11°C and the cumulative precipitation was about 21 mm (Fig. 2 4).

12 In order to investigate the amount and temporal changes of NH₄⁺ and NO₃⁻ concentrations in
13 the surface soil (i.e., 0-15 cm depth increment), repeated topsoil samples were collected in the
14 Edmonton site in the spring-summer. From 13 May to 20 Jul 2015, we found a general trend for
15 gradual decline in NH₄⁺ concentrations in particular for the treatments exhibiting a high initial
16 concentration, such as the fall manure with DMPP and certain spring manure treatments (Fig. 2
17 5b and e). Similarly, relatively quicker depletion patterns were observed for NO₃⁻ concentrations
18 for all fall and spring manure treatments (Fig. 2 5h and k). However, it should be emphasized
19 that these nitrate depletion progressions were delayed by approximately 15 days compared to the
20 ammonium depletion patterns (Fig. 2 5h and k). Unlike most of the treatments, NH₄⁺
21 concentration in the spring manure with DMPP moderately increased during 42 days following
22 the spring manure injection , and subsequently, decreased sharply on 7 Jul 2015 (Fig. 2 5e). In
23 particular, it took longer for the spring manure treatments than for the fall manure treatments to
24 reach asymptotic depletion plateaus for both NH₄⁺ and NO₃⁻ concentrations within the spring
25 2015 (i.e., about 42 vs. 31 days; Fig. 2 5). The amount of NO₃⁻ present in the soil treated with
26 fall manure injection with DMPP additive was in general three times larger than in the soil
27 receiving spring manure with DMPP (FD \approx 3 SD; Fig. 2 5h and k).

28 Our measured, temporal patterns of soil mineral N transformations – ammonium and nitrate
29 depletion progressions – were fitted and modelled by both first- and second-order kinetic models.
30 When focusing on the experimental treatments that had p-values lower than 0.1, the coefficients

1 of determination (R²) for nitrate depletions using second-order kinetic were relatively higher
2 than when employing first-order kinetic, with the only exception of SW treatment (Table 2 2).
3 Conversely, regarding ammonium depletion progressions, there was no clear differentiation in
4 the performance of first- vs. second-order kinetic when using R² and p-values as model
5 evaluation criteria (Table 2 2).

6 Compared to the two control treatments receiving neither manure nor additive, nearly all soils
7 receiving manure injections either in the fall or spring and with or without NIs required in
8 general longer periods to reach and settle into depletion plateaus for both NH₄⁺ and NO₃⁻
9 concentrations, with only few noticeable exceptions (FN and SW) (Table 2 2, Fig. 2 5). The
10 control zero (CZ) had comparatively faster soil mineral N depletion rates than the control treated
11 with manure injector disturbance (CT) (Table 2 2). Overall, the fall manure DMPP treatment had
12 the fastest rates of mineral N depletion among the fall manure treatments based on both first- and
13 second-order kinetic models , while the spring manure without NIs exhibited the fastest rates of
14 mineral N depletion among the three spring manure treatments (Table 2 2).

15 There was a significant effect of the manure injection timing on both aboveground barley dry
16 matter yield and plant N uptake in the Lacombe site (P_s<0.05), but there were no significant
17 effects of nitrification inhibitor additions at both sites (Table 2 1). In Lacombe, the soils
18 receiving spring manure amendment resulted in higher aboveground plant dry matter yield and
19 plant N uptake than the soils treated with fall manure (Table 2 3). In further details, when
20 comparing the fall manure treatments, the amount of N uptake under fall manure with DMPP
21 was significantly lower than for fall manure without NIs (FD was 82% of FW; Table 2 3).

22 Annual total precipitation in 2015 was 380 mm in Lacombe and 294 mm in Edmonton (Fig. 2 4c
23 and d). The monthly average temperature from Oct 2014 to Nov 2015 was higher than the
24 corresponding normal values in both Edmonton and Lacombe with the exceptions of Nov 2014,
25 as well as Feb and Sep 2015 (Fig. 2 4a and b). The monthly precipitation was generally lower
26 than the normal values during the growing season (Apr – Aug 2015) and higher over the cold
27 months with the exceptions of Oct and Dec 2014, and Oct and Nov 2015 at both sites (Fig. 2 4c
28 and d). A predominantly drier condition over the experimental period was even more obvious in
29 the Edmonton site during the spring 2015, when each monthly precipitation over the growing

1 season (i.e., Apr – Aug) was only about half of the corresponding normal average (Fig. 2 4d).
2 The majority of highest soil temperature occurred through Jun – Sep (Fig. 2 6a), while the
3 highest soil average volumetric water content occurred following the snow melt and soil thawing
4 in April with a range of 0.24-0.28 m³ m⁻³ (Fig. 2 6b).

5 Reflecting the variability in manure properties, there was a significant difference in both total N
6 and ammonium application rates across the three different times of manure injection in the
7 Lacombe site (P<0.05) in general (Table 2 4). More specifically, the highest total N and
8 ammonium loads were observed in the spring 2015, while the lowest manure-N rates were
9 quantified in the fall 2014 (Table 2 4).

10

11 Discussion

12 Our results showed that the fall manure injection led to a lower N input usage efficiency as
13 indicated by a higher annual cumulative N₂O losses from fall-manured soils compared to the
14 spring manure injection at both sites (Fig. 2 1), lower plant dry matter yield and also lower plant
15 N uptake and utilization particularly in the Lacombe site (Table 2 3). Our finding is consistent
16 with the reports by Weslien et al. (1998), Thorman et al. (2007), but it is opposite to the studies
17 by Rochette et al. (2004) and Hernandez-Ramirez et al. (2009a). The two latter studies
18 considered that the lower N₂O emission derived from the fall manure application was likely
19 attributed to the limited net nitrification under a wet and cool soil condition. Rochette et al.
20 (2004) measured N₂O emissions following fall and spring pig slurry injections in a Le Bras loam
21 soil near Québec City, Canada. As WFPS in their study ranged from 60-80% during the fall N₂O
22 measurements, it was likely that most N was lost via N₂ rather than N₂O (Morley and Baggs
23 2010). Likewise, the largest contribution to the total annual N₂O emissions in our three fall
24 manure treatments was the N₂O emissions that occurred during the early spring snow-melting
25 and soil thawing (March-April 2015; Fig. 2 2 and Fig. 2 3) in concurrence with an increasing
26 temperature and an abundance abundant soil moisture content (Fig. 2 6). This was also found by
27 Nyborg et al. (1997) when assessing the addition of synthetic N fertilizers. They reported an

1 increased N₂O flux during the spring thaw in a Black Chernozemic soil with high availability of
2 NO₃--N and moisture.

3 Adding the liquid manure caused extreme increases in annual N₂O emissions at both field sites,
4 and both inhibitors were effective in reducing N₂O emissions particularly in the Lacombe site
5 (Fig. 2 1). The proportion of N₂O losses to the manure applied total N for the fall and spring
6 manure with no inhibitors added at both sites was 0.19 – 1.93% (Table 2 5). This result is within
7 the range of <0.1 – 3% in an earlier report, which summarized the cumulative N₂O emissions
8 from cattle and pig slurry applications based on a compilation of eight studies (Chadwick et al.
9 2011) .

10 Contrasting the two NI additives, DMPP resulted was more effective in reducing cumulative
11 N₂O emission than nitrapyrin; this became evident in our wetter Lacombe site (Fig. 2 1). This is
12 because DMPP has a similar mobility as ammonium (Pasda et al. 2001), whereas nitrapyrin has
13 an even lower mobility (Subbarao et al. 2006). Hence, DMPP could more tightly and longer
14 remain in the soil solution in close contact with the soil ammonium ions than nitrapyrin, and
15 hence, decreasing decrease the likelihood for ammonium nitrification, and subsequently nitrate
16 denitrification. Our reduction coefficients for the spring-manured soil with NIs in Lacombe (i.e.,
17 DMPP: 57.8%; nitrapyrin: 32.7%; Fig. 2 1) are in line with an existing meta-analysis study, in
18 which the reduction coefficients for DMPP were about 55% [95% confidence interval (CI): ~21
19 to ~60%] and for and nitrapyrin 30% (95% CI: ~17 to ~40%) (Akiyama et al. 2010). However,
20 the reduction coefficients calculated for our fall-manured soil with NIs in Lacombe (i.e., DMPP:
21 81.0%; nitrapyrin: 64.3%; Fig. 2 1) were much higher than the range compiled by Akiyama et al.
22 (2010). This might be due to the variations and unique combinations of factors such as soil
23 texture, manure composition and rate, crop type and climate across different studies. All of these
24 factors have the scope to interact and influence the effectiveness of NIs.

25 It is noteworthy that both DMPP and nitrapyrin admixed and injected with liquid manure on
26 their fall 2014 were still active in the following early spring (Fig. 2 2b) after the soil had
27 undergone a six-month freezing period (Fig. 2 4a). In contrast, in a related incubation study using
28 the same soils collected from our experimental fields shortly after the spring manure injections
29 (See chapter 3), it was observed that DMPP activity decayed rather quickly within one week

1 after the incubation have had begun at a temperature about 3°C higher than that under field
2 conditions (i.e., 20.4 vs. 17.8 °C). This can be explained because the decay in activity of
3 inhibitors is highly dependent on the temperature, and inhibitors could persist over even longer
4 periods under colder temperatures (Guiraud and Marol 1992; Zerulla et al. 2001). Due to the
5 extended effectiveness of inhibitors under field conditions, our fall-manured soils with DMPP
6 (or nitrapyrin) did not show any obvious distinction in reducing (?) annual N₂O emissions
7 compared to the spring-manured soils with DMPP (or nitrapyrin) (Fig. 2 1a). However, most
8 notably, the fall-manured soil with NIs resulted in lower aboveground plant dry matter yield and
9 a plant N uptake than the spring-manured soil with NIs particularly in the Lacombe site (Table 2
10 3). This may be explained because a large amount of N₂ losses could have occurred during the
11 early spring snow melting and soil thawing via complete denitrification as soils underwent
12 predominant anaerobic conditions (Meixner and Yang 2006) . Overall, our data implies that
13 under certain circumstances shifts in plant productivity and variations in N₂O emissions not
14 necessarily trade off with each other, and hence, this can suggest that best management practices
15 need to be thoroughly identified to jointly address and attain simultaneously both an optimal
16 plant performance and effective mitigation of detrimental environmental effects.

17 Crop N uptake was the largest contributor for the total N output among treatments, followed by
18 the denitrified-N losses (i.e., the sum of N₂O and N₂), nitrate leaching and NH₃ volatilization
19 (Table 2 5). Similar N balance results were previously found under a continuous barley cropping
20 system where the dominant N input was also synthetic fertilizers and the main gaseous N output
21 was associated with denitrification (Ross et al. 2008). Based on these N balance results, we
22 recommend developing direct measurements of dinitrogen losses as well as N₂O to N₂ ratios
23 from manured soils to improve overall agroecosystem accounting of N fluxes and pools (Stevens
24 and Laughlin 1998).

25 As expected, our field results indicated a gradual pattern towards depletion of soil nitrate
26 concentrations following spring manure injection, whereas data shown in chapter 3 from a
27 microcosm incubation using the same soils showed nitrate accumulations. This apparent
28 divergence can be mainly explained as in the fields the barley crop assimilated and made use of
29 the available soil N for biomass growth leading to a net decrease in soil nitrate. Moreover, first-
30 and second-order ammonium depletion rates estimated in this field study (Table 2 2) were lower

1 than those found in the soil microcosms. We infer this numerical difference is attributable to
2 substantially lower temperature and soil moisture content in the field (during the period from 13
3 May to 20 Jul 2015) than in the incubation (18.2 vs. 20.4 °C, and 0.11 vs. 0.35-0.41 m³ m⁻³). As
4 shown in previous reports, lower temperature limits the soil microbial activities and lower
5 moisture is beneficial for ammonium nitrification (Davidson and Schimel 1995; Bateman and
6 Baggs 2005), thereby leading to reduced ammonium depletion rates in our field study.

7 The addition of nitrification inhibitors (NIs) clearly impacted the temporal dynamics of soil
8 mineral N. The spring-manured soils treated with NIs exhibited much slower first-order
9 depletion rates of ammonium concentration than the spring manure soils receiving no NIs (Table
10 2 2). This result is consistent with an earlier report (Omonode and Vyn 2013). Additionally, most
11 of the manure treatments receiving NIs took longer (about 13 extra days) to reach depletion
12 plateaus following their initial peaks in nutrient concentration, with the only exception of fall
13 manure with nitrapyrin and spring manure without NIs (FN and FW) (Table 2 2). In general, this
14 observation suggests that the assessed NIs remained still active even under the dry conditions
15 prevailing in the spring 2015 in the Edmonton site. However, these evident effects on nutrient
16 concentration patterns did not translate into strong reduction of N₂O emissions (Fig. 2 1b, Fig. 2
17 3). The effect of moisture on NIs activity is still not well documented, thus it is suggested to
18 further address this unknown using a wide range of soils under varying inhibitor rate, N addition
19 rate, moisture and temperature.

20 A key control on our N₂O emissions was the availability of substrate for denitrification (Havlin
21 et al. 2014). For instance, both the fall-manured soils with and without DMPP (FD and FW)
22 exhibited much higher N₂O fluxes than the fall-manured soil treated with nitrapyrin (FN) during
23 the week immediately after the manure injections conducted on 7 Oct 2015 (Fig. 2 3c). This
24 result can be attributed to the fact that both the fall-manured soils with and without DMPP still
25 kept comparatively higher concentration of residual nitrate in the 0-15 cm soil layer than the fall-
26 manured soil with nitrapyrin as quantified on 25 Sep 2015 after the barley growth cycle and
27 harvest had been completed (Fig. 2 5i).

28

1 Tables

2 Table 0-1. ANOVA results for annual cumulative N₂O emissions, barley dry matter yield and
3 nitrogen uptake in the Lacombe and Edmonton sites.

	Lacombe		Edmonton	
	F-value	p-value	F-value	p-value
Annual N ₂ O Emission				
Timing [†]	1.083	0.375	1.775	0.314
Additive [‡]	4.212	0.041	1.546	0.271
Timing:Additive	0.902	0.432	0.553	0.596
Plant Dry Matter Yield				
Timing	22.710	0.018	7.408	0.113
Additive	1.008	0.394	0.592	0.576
Timing:Additive	2.766	0.103	0.552	0.596
Plant Nitrogen Uptake				
Timing	87.300	0.003	5.770	0.138
Additive	0.838	0.456	0.823	0.473
Timing:Additive	2.605	0.115	1.348	0.313

4 [†] Timing factor includes fall 2014 and spring 2015 manure injection treatments.

5 [‡] Additive factor includes manure without nitrification inhibitors, manure with DMPP, and
6 manure with nitrapyrin treatments.

1 Table 0-2. Rates (k) of soil mineral nitrogen concentration changes with time in the Edmonton site based on first- and second-order
2 kinetic models. P-values (*P*) and coefficient of determination (*R*²) for each model and data subset are shown as criteria for model
3 evaluation.

Treatment	Time interval [†]	First-Order Kinetics [‡]						Second-Order Kinetics [§]					
		Ammonium Depletion			Nitrate Depletion			Ammonium Depletion			Nitrate Depletion		
		P	R ²	k	P	R ²	k	P	R ²	k	P	R ²	k
μg N kg ⁻¹ day ⁻¹			μg N kg ⁻¹ day ⁻¹			μg N kg ⁻¹ day ⁻¹			μg N kg ⁻¹ day ⁻¹				
CZ	1	0.001	0.980	-8.821**	0.011	0.978	16.68**	0.001	0.978	-2.161**	0.005	0.990	1.299**
FD	2	0.010	0.838	27.83**	0.008	0.930	42.06**	0.010	0.838	3.658**	0.006	0.940	1.064**
FN	1	0.284	0.361	-3.637	0.082	0.843	26.53*	0.242	0.413	-0.769	0.043	0.917	0.761**
FW	2	0.174	0.405	5.718	0.074	0.707	24.41*	0.174	0.405	1.307	0.044	0.790	0.743**
CT	1	0.647	0.079	0.001	0.038	0.925	13.32**	0.618	0.093	0.187	0.024	0.953	0.972**
SD	2	0.275	0.286	4.700	0.219	0.445	5.569	0.275	0.286	0.397	0.241	0.414	0.244
SN	2	0.022	0.767	13.93**	0.288	0.356	7.590	0.022	0.767	1.865**	0.401	0.241	0.264
SW	1	0.015	0.896	21.38**	0.055	0.893	32.19*	0.040	0.800	3.252**	0.095	0.819	1.109*
Mean		-	-	7.637	-	-	21.044	-	-	0.967	-	-	0.807
S.E.		-	-	4.600	-	-	4.793	-	-	0.710	-	-	0.150

4 S.E. = one standard error; * = significantly different at *P*<0.1; ** = significantly different at *P*<0.05.

5 † The time interval 1 corresponds to the period from 13 May throughout 4 Jul 2015 (time series of 5 sampling dates and data points)
6 for ammonium, and 24 May throughout 4 Jul 2015 (4 data points) for nitrate. The time interval 2 corresponds to the period from 13
7 May throughout 20 Jul 2015 (6 data points) for ammonium, and throughout 20 Jul 2015 (5 data points) for nitrate. The lapse between
8 the selected intervals for ammonium and nitrate can indicate the time necessary for an N transformation via nitrification in these soils.
9 Temporal patterns of ammonium and nitrate concentrations are shown in Fig. 0-4.

10 ‡ The first-order kinetic was calculated based on Eq. [2-3].

11 § The second-order kinetic was calculated based on Eq. [2-4].

1 Table 0-3. Aboveground barley dry matter yield and nitrogen uptake with one standard error in the Lacombe and Edmonton sites.

Treatment	Lacombe		Edmonton	
	kg ha ⁻¹			
Dry Matter Yield				
CZ	2946.4 ± 260.8	Bb [†]	4410.6 ± 440.4	Cb
FD	5865.5 ± 200.4	a	7996.4 ± 559.5	a
FN	6042.4 ± 393.2	a	7494.4 ± 891.2	a
FW	6691.2 ± 384.5	Aa	7896.4 ± 99.5	Aa
CT	2416.5 ± 365.4	Bb	3688.9 ± 189.5	Cb
SD	7414.4 ± 295.5	a	7042.5 ± 348.9	a
SN	7018.4 ± 401.1	a	6908.5 ± 893.0	a
SW	7048.9 ± 452.9	Aa	5942.5 ± 256.5	Ba
Aboveground Nitrogen Uptake				
CZ	35.3 ± 1.4	Cc	63.8 ± 8.8	Cb
FD	78.7 ± 3.2	b	143.2 ± 8.4	a
FN	88.8 ± 6.1	ab	134.0 ± 13.4	a
FW	95.8 ± 2.8	Ba	143.8 ± 2.8	Aa
CT	33.1 ± 3.6	Cb	56.3 ± 1.3	Cb
SD	138.6 ± 5.7	a	132.4 ± 4.4	a
SN	126.6 ± 8.3	a	126.8 ± 14.6	a
SW	133.1 ± 8.2	Aa	110.4 ± 6.8	Ba

2 † Values followed by different capital letters indicate significant differences among control zero (CZ), control disturbance (CT), fall
3 manure without nitrification inhibitor (FW) and spring manure without nitrification inhibitor (SW) treatments based on LSD test (*P*
4 <0.05); values followed by different lowercase letters indicate significant differences among three fall manure treatments (FD, FN and
5 FW) and control zero (CZ) or three spring manure treatments (SD, SN and SW) and control disturbance (CT) based on LSD test (*P*
6 <0.05).

7

1 Table 0-4. Average total N, ammonium and water contents with one standard error in the manure applied in fall 2014, spring 2015 and
 2 fall 2015. Different capital letters indicate significant differences among the manure applications based on LSD test ($P<0.05$), and
 3 different lowercase letters indicate significant differences between field locations (Lacombe vs. Edmonton) based on the two sample t
 4 test ($P<0.05$).

	Lacombe		Edmonton	
	Total N (kg ha ⁻¹)			
Fall 2014	323.7 ± 46.0	Ba	502.5 ± 8.9	Ab
Spring 2015	535.7 ± 7.2	Aa	526.8 ± 15.1	Aa
Fall 2015	416.7 ± 71.7	ABa	531.6 ± 28.3	Aa
	NH ₄ ⁺ -N (kg ha ⁻¹)			
Fall 2014	213.8 ± 0.1	Ca	286.1 ± 1.0	Ab
Spring 2015	304.4 ± 0.1	Aa	255.6 ± 2.1	Ab
Fall 2015	263.9 ± 3.7	Ba	281.0 ± 1.1	Bb
	H ₂ O m/m (%)			
Fall 2014	98.7 ± 0.1	Aa	91.8 ± 0.8	Bb
Spring 2015	97.8 ± 0.3	Aa	92.9 ± 0.1	Ab
Fall 2015	98.0 ± 0.7	Aa	93.7 ± 0.1	Ab

5

1 Table 0-5. Estimated annual N budget for Lacombe and Edmonton sites.

	Lacombe [†]								Edmonton [†]							
	CZ	FD	FN	FW	CT	SD	SN	SW	CZ	FD	FN	FW	CT	SD	SN	SW
	kg N ha ⁻¹ yr ⁻¹															
inputs:																
N deposition from atmosphere [‡]	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0
Non-symbiotic N fixation [§]	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0
Manure(Total N) [¶]	0.0	323.7	323.7	323.4	0.0	535.7	535.7	535.7	0.0	502.5	502.5	502.5	0.0	526.8	526.8	526.8
	0.0	(46.0) ^{¶¶}	(46.0)	(46.0)	0.0	(7.2)	(7.2)	(7.2)	0.0	(8.9)	(8.9)	(8.9)	0.0	(15.1)	(15.1)	(15.1)
Manure(Ammonium) [¶]	0.0	213.8	213.8	213.8	0.0	304.4	304.4	304.4	0.0	286.1	286.1	286.1	0.0	255.6	255.6	255.6
Total N inputs	10.0	333.7	333.7	333.7	10.0	545.7	545.7	545.7	10.0	512.5	512.5	512.5	10.0	536.8	536.8	536.8
outputs:																
N exported in crop harvest ^{¶¶}	35.4	78.7	88.9	95.9	33.1	138.6	126.7	133.2	63.8	143.3	134.1	143.8	56.3	132.8	126.9	110.0
	(1.5) ^{¶¶}	(3.2)	(6.2)	(2.9)	(3.6)	(5.8)	(8.4)	(8.3)	(8.8)	(8.8)	(13.5)	(2.9)	(1.4)	(4.8)	(17.6)	(6.2)
Gaseous N losses	1.0	61.2	61.2	61.2	1.0	94.1	94.1	94.1	1.0	88.4	88.4	88.4	1.0	86.9	86.9	86.9
Nitrous oxide from soil ^{¶¶}	0.3	1.2	2.6	6.2	0.3	1.1	2.1	3.1	0.6	1.8	2.2	2.3	0.6	1.0	1.4	1.0
	(0.3) ^{¶¶}	(0.3)	(1.4)	(6.4)	(0.0)	(0.4)	(0.8)	(1.5)	(0.8)	(1.0)	(1.8)	(1.3)	(0.0)	(0.1)	(0.3)	(0.3)
Dinitrogen from soil ^{¶¶}	0.7	32.2	30.7	27.1	0.7	53.5	52.5	51.5	0.4	49.5	49.0	49.0	0.4	52.7	52.2	52.2
NH ₃ volatilization ^{¶¶}	ngb	27.8	27.8	27.8	ngb	39.6	39.6	39.6	ngb	37.2	37.2	37.2	ngb	33.2	33.2	33.2
Nitrate Leaching ^{¶¶}	ngb	32.4	32.4	32.4	ngb	37.5	37.5	37.5	ngb	50.3	50.3	50.3	ngb	36.9	36.9	36.9
Surface N run-off losses ^{¶¶}	ngb	1.5	1.5	1.5	ngb	1.5	1.5	1.5	ngb	1.5	1.5	1.5	ngb	1.5	1.5	1.5
Total N outputs	36.4	173.7	183.9	190.9	34.1	271.8	259.8	266.3	64.8	283.4	274.2	284.0	57.3	258.0	252.1	235.0
System N balance	-26.4	160.0	149.8	142.8	-24.1	273.9	285.9	279.4	-54.8	229.1	238.3	228.5	-47.3	278.8	284.7	301.8
	(1.7) ^{¶¶¶}	(49.7)	(53.6)	(55.3)	(3.7)	(13.4)	(16.3)	(16.9)	(9.6)	(18.7)	(24.2)	(13.1)	(1.4)	(20.0)	(33.1)	(22.2)

2 ngb = negligible.

3 † The time period for the annual N budget in Lacombe corresponds to 7 Oct 2014 – 6 Oct 2015; the time period in Edmonton
4 corresponds to 1 Oct 2014 – 30 Sep 2015.

5 ‡ Deposition from atmosphere was assumed to be 5.0 kg N ha⁻¹ yr⁻¹ (Janzen et al. 2003).

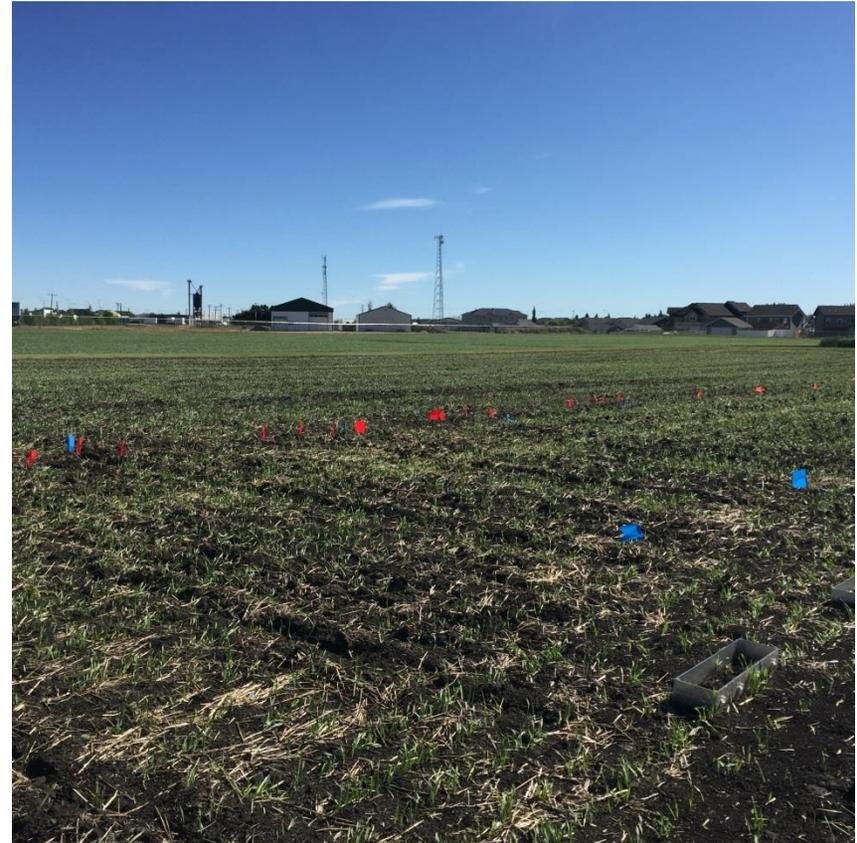
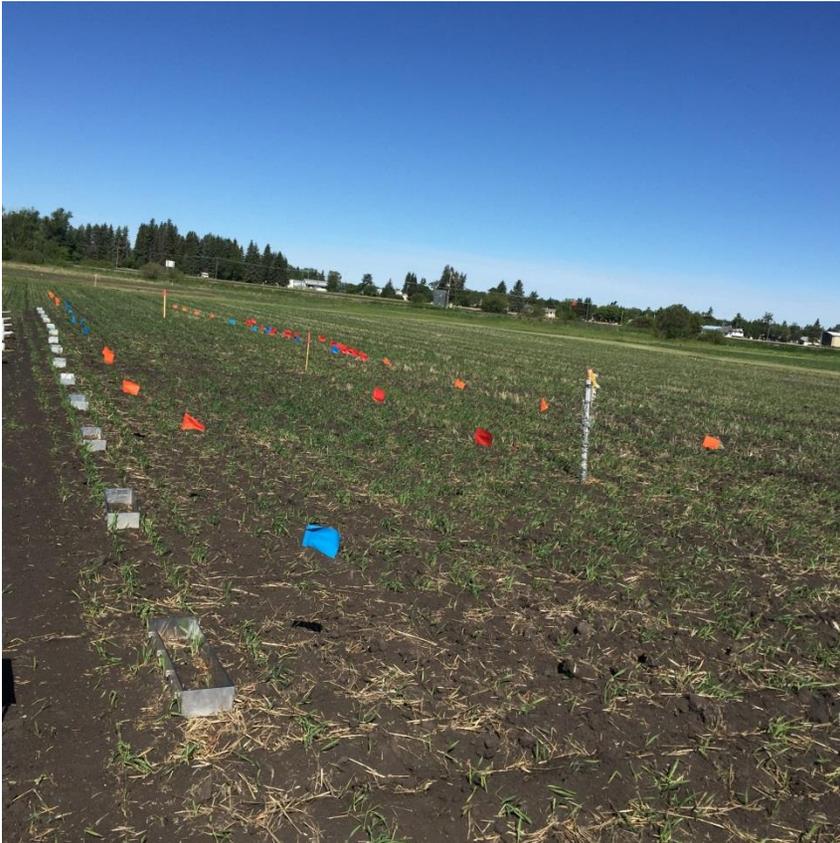
6 § Non-symbiotic N fixation and surface run-off losses were assumed to be 5.0 and 1.5 kg N ha⁻¹ yr⁻¹, respectively (Ross et al. 2008).

- 1 ¶ The values were directly measured in this study.
- 2 \\Dinitrogen from soil assume: 10% gaseous N losses from total N in manure (i.e., dinitrogen from soil = 10%*Total N in manure –
- 3 nitrous oxide from soil) (Janzen et al. 2003).
- 4 †† NH₃ volatilization assume: 13% of soluble ammonium in manure (Misselbrook et al. 2002).
- 5 §§ Nitrate leaching assume: 10 and 7% of total N in manure for the fall and spring treatments, respectively (Janzen et al. 2003; Van Es
- 6 et al. 2006).
- 7 ¶¶ Values in parenthesis correspond to one standard error.
- 8 \\These propagated errors for the system N balance closure were estimated by simple addition of the standard errors derived from the
- 9 direct measurements in this study: manure (total N) input, N exported in crop harvest, and nitrous oxide from soil.

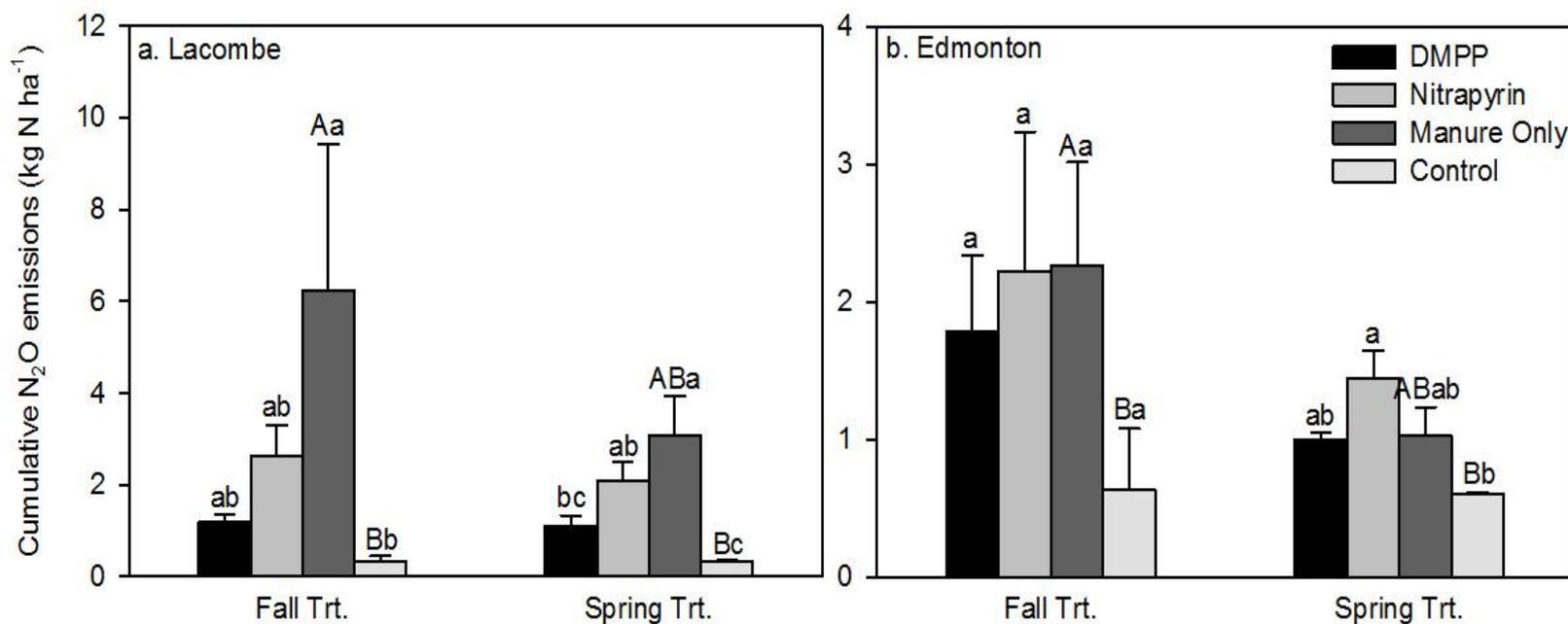
1 Figures

2 Lacombe site 15 June 2016

3

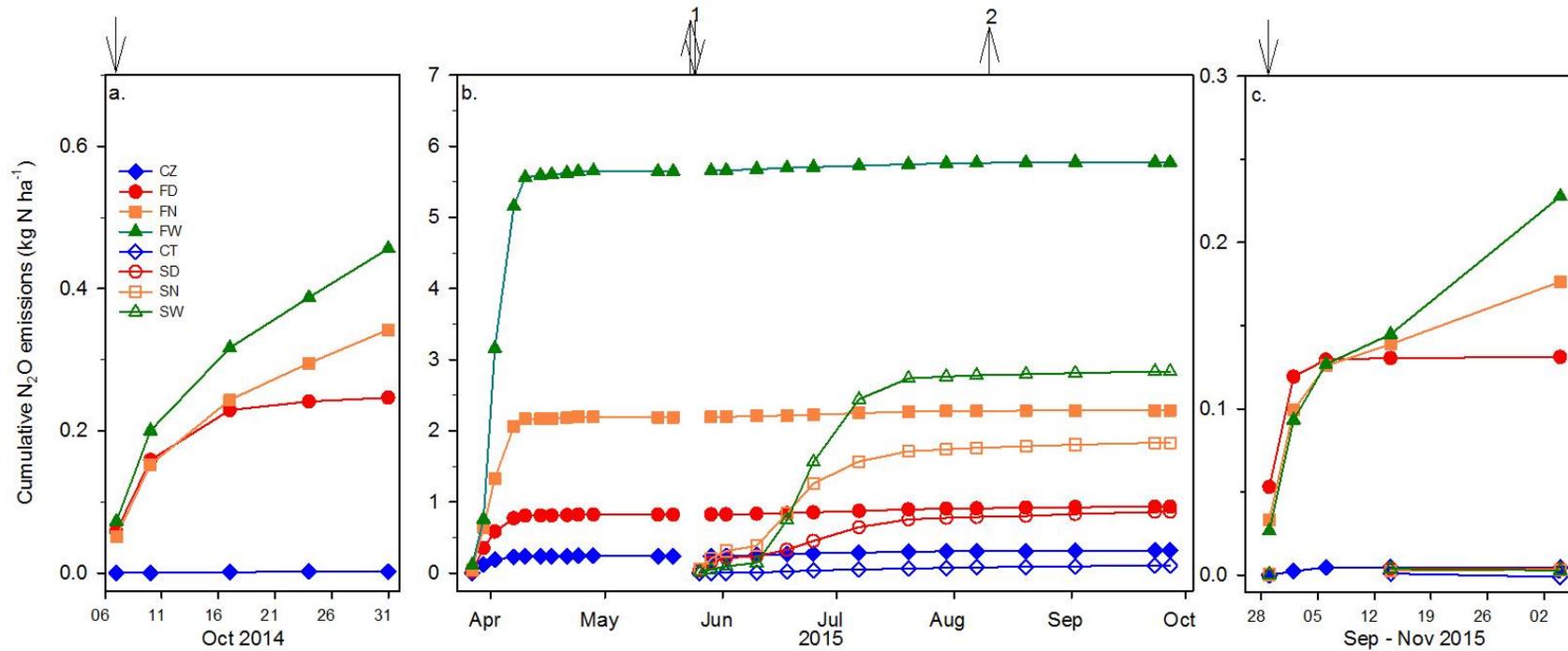


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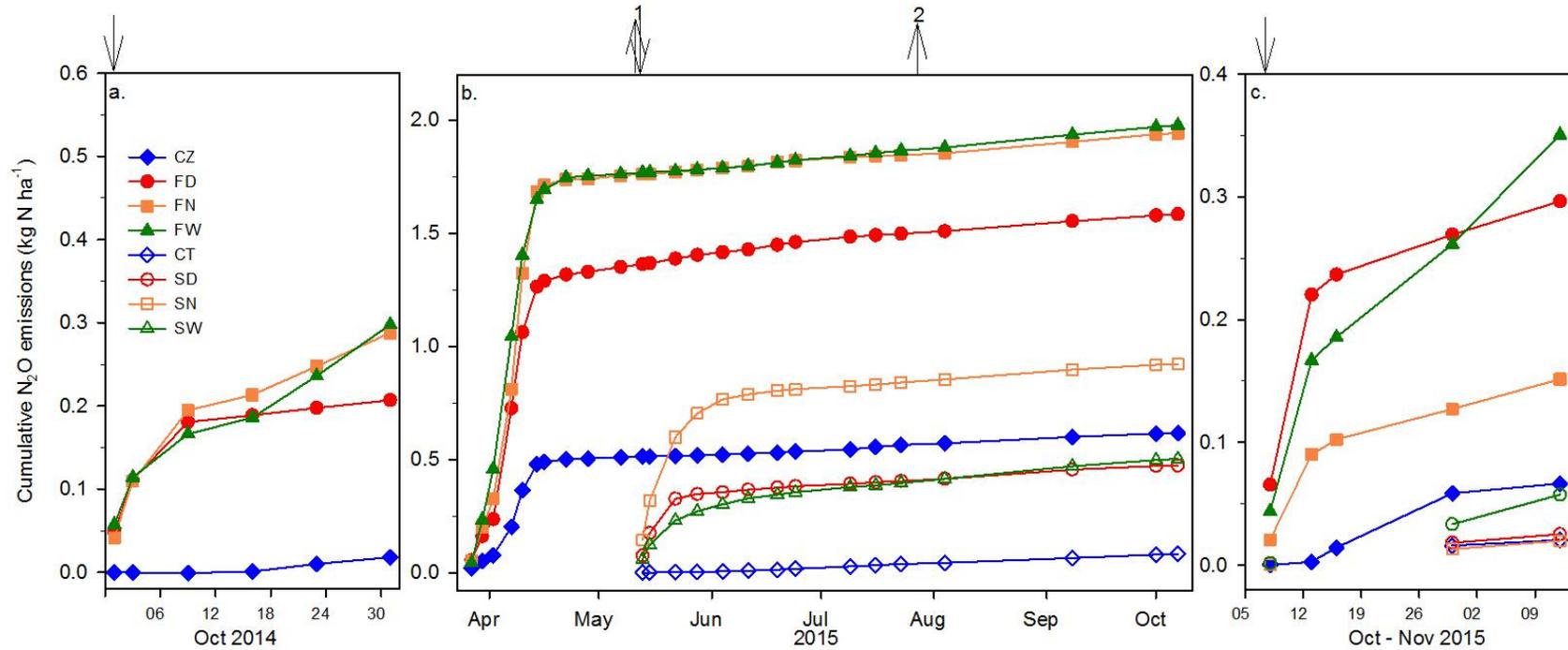
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3 Fig. 0-1. Annual cumulative N₂O emissions in the (a) Lacombe and (b) Edmonton sites. We assumed negligible N₂O emission from 1
 4 Nov 2014 to 26 Mar 2015 for both sites. Different capital letters indicate significant differences among control zero (CZ), control
 5 disturbance (CT), fall manure without inhibitors (FW) and spring manure without inhibitors (SW) treatments based on LSD test
 6 ($P < 0.05$); different lowercase letters indicate significant differences among three fall manure treatments (FD, FN and FW) and the
 7 control zero (CZ), or three spring manure treatments (SD, SN and SW) and the control disturbance (CT) based on LSD test ($P < 0.05$).
 8 Note the different y-axis scales across panels.



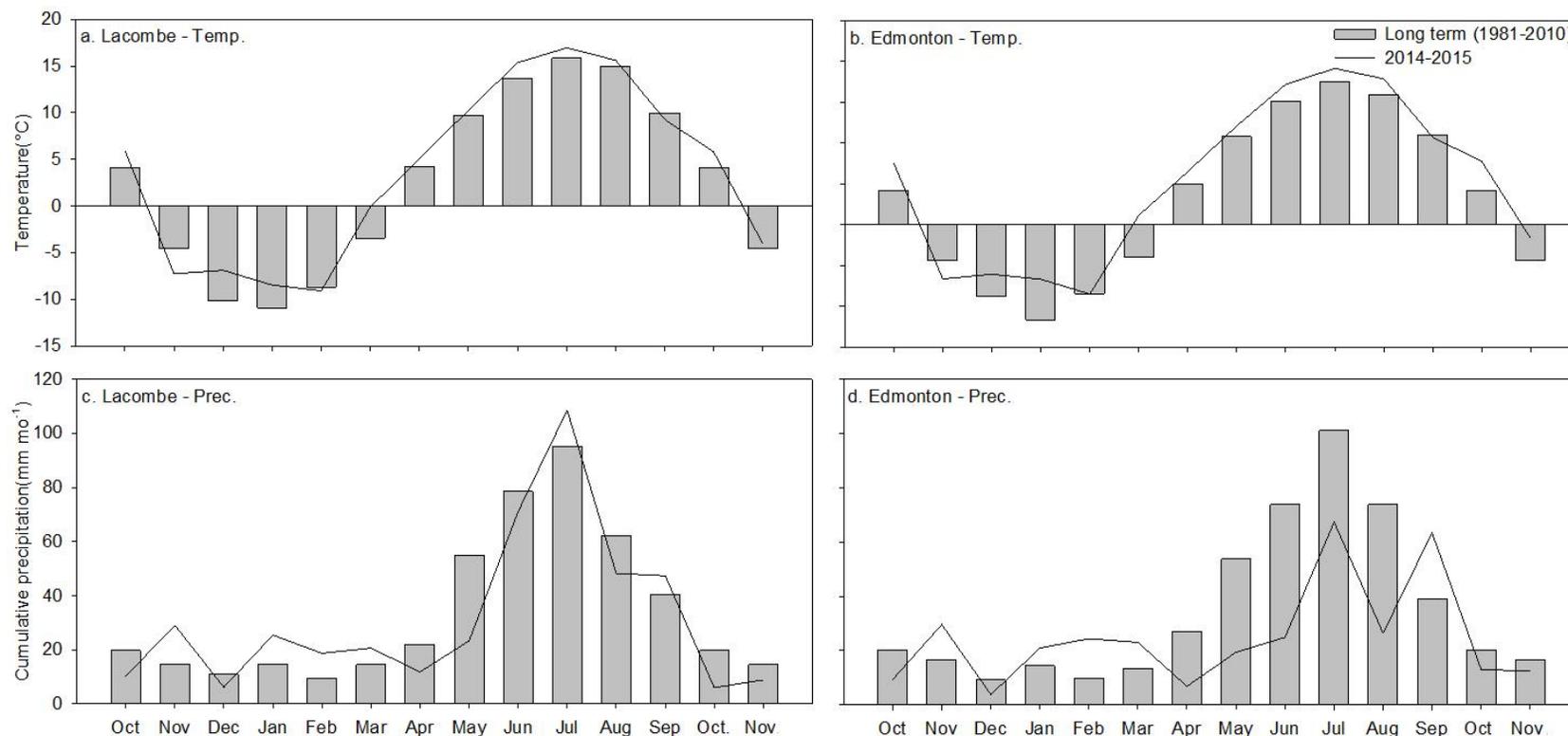
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2 Fig. 0-2. Cumulative N₂O emission patterns in the Lacombe site during the periods from (a) 4 Oct to 31 Oct 2014,
 3 27 Mar to 28 Sep 2015 and (c) 29 Sep to 4 Nov 2015. The upward arrows indicate the dates of seeding (↑¹) and harvest (↑²), and the downward (↓)
 4 arrows indicate the dates of manure injections. Standard errors were not included for clarity. Note the different y-axis scales across
 5 panels.



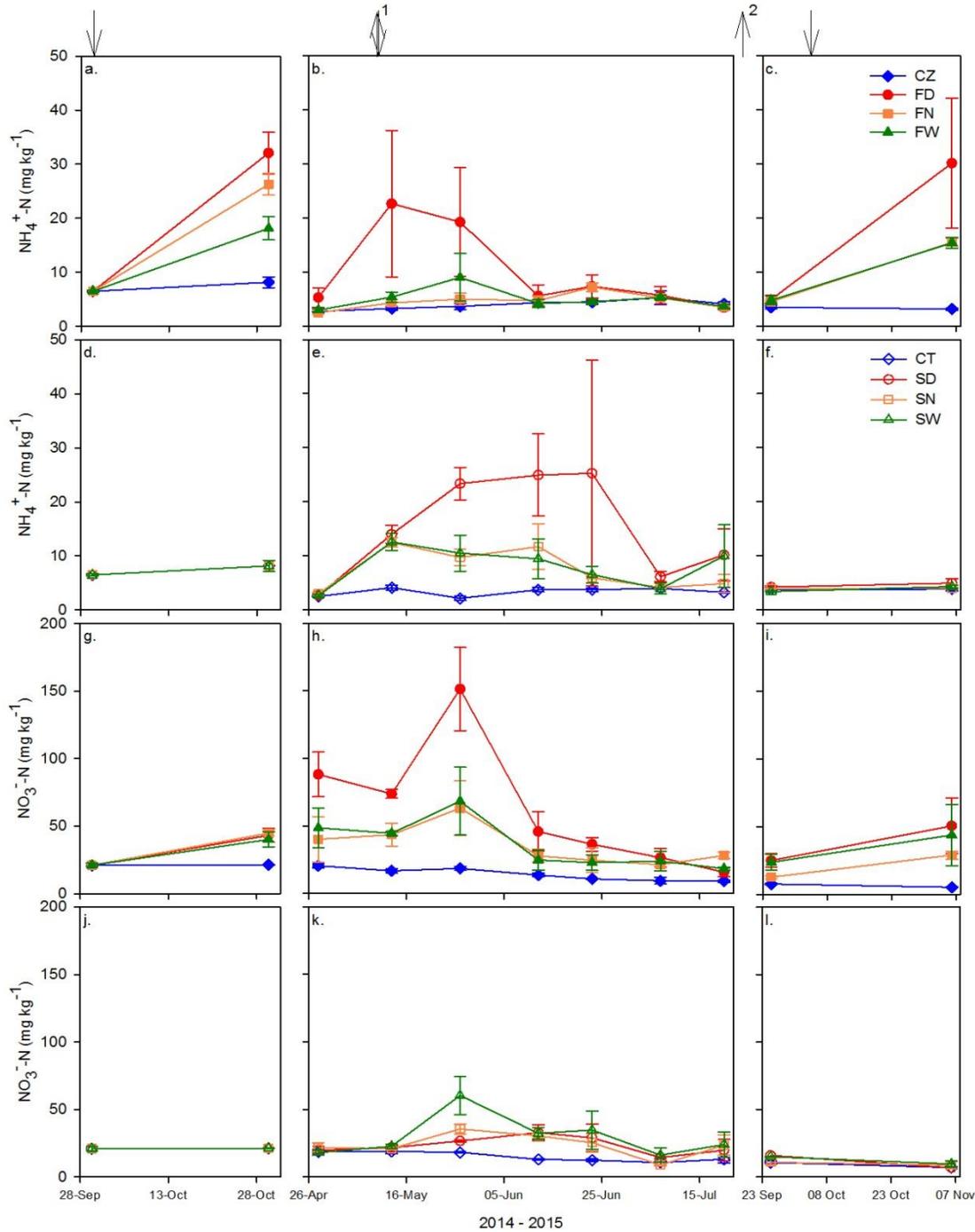
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2 Fig. 0-3. Cumulative N₂O emission patterns in the Edmonton site during the periods from (a) 1 Oct to 31 Oct 2014, (b) 27 Mar to 7
 3 Oct 2015 and (c) 8 Oct to 11 Dec 2015. The upward arrows indicate the dates of seeding (\uparrow^1) and harvest (\uparrow^2), and the downward (\downarrow)
 4 arrows indicate the dates of manure injections. Standard errors were not included for clarity. Note the different y-axis scales across
 5 panels.



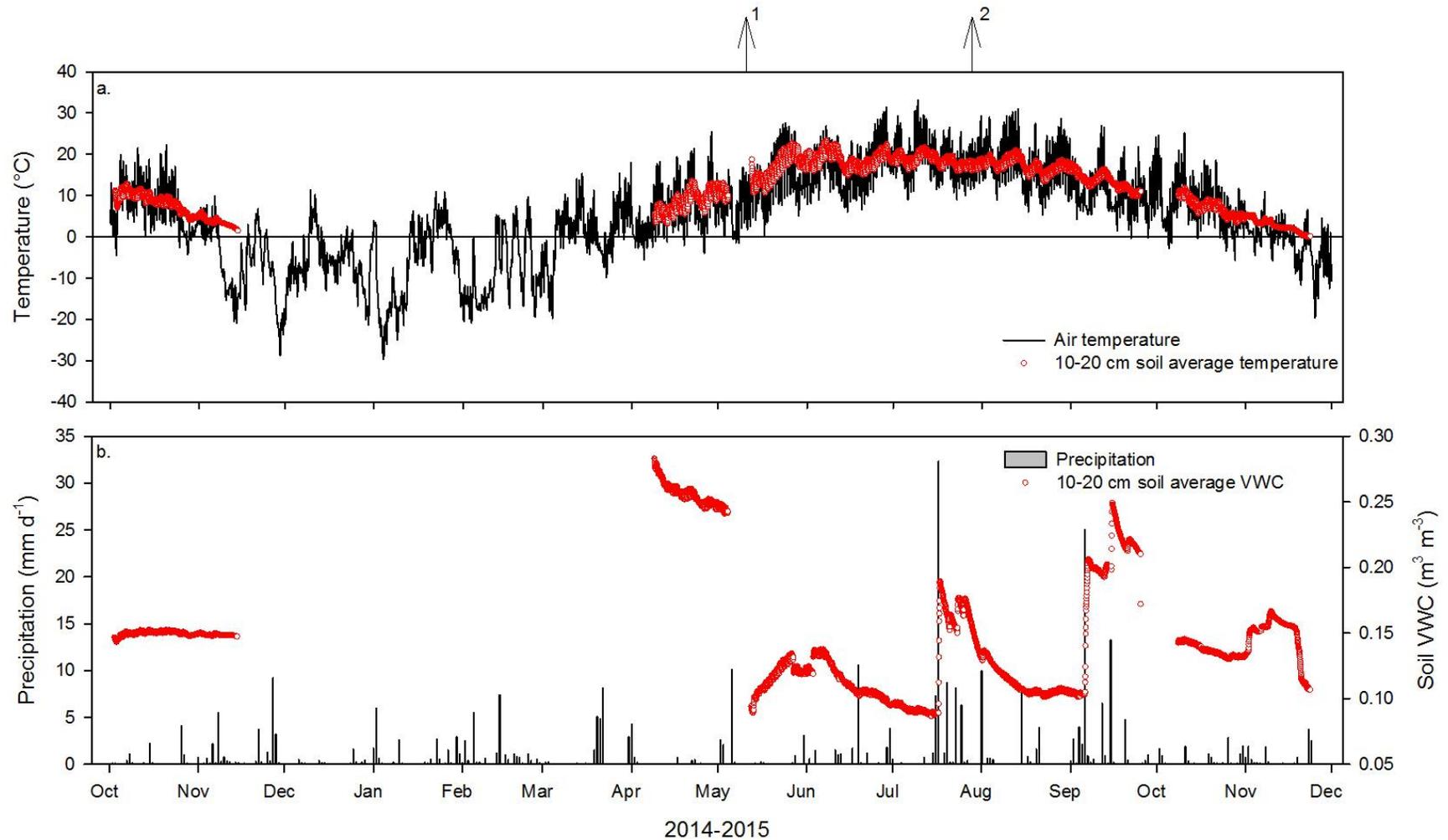
2014-2015

1
2 Fig. 0-4. Monthly average air temperature for (a) Lacombe and (b) Edmonton sites, and cumulative precipitation for (c) Lacombe and
3 (d) Edmonton sites during the experimental period. The 30-year normal monthly averages are also shown. Monthly average
4 temperature and cumulative precipitation data is derived from Alberta Agriculture and Forestry (2016). The 30-year normal monthly
5 temperature and cumulative precipitation data is derived from Government of Canada (2016).



1
2 Fig. 0-5. Soil (a, b, c, d, e, and f) ammonium and (g, h, I, j, k and l) nitrate concentrations at the
3 0-15 cm depth increment during the experimental period in the Edmonton site. Error bars
4 correspond to one standard error. The ammonium and nitrate values for all spring manure and
5 control treatments (SD, SN, SW and CT) from 13 May to 20 Jul 2015 were derived by using
6 weighted averages of the measurements taken from the band and interband zones in the field
7 plots. The same spatial zone sampling and weighted calculation were applied to derived the
8 ammonium and nitrate values for all fall manure treatments (FD, FN and FW) on 30 Oct 2014
9 and 6 Nov 2015. The ammonium and nitrate values for all fall manure treatments (FD, FN and

- 1 FW) on 30 Oct 2014 were the average for the 0-20 cm soil layer. The upward arrows indicate the
- 2 dates of seeding (\uparrow^1) and harvest (\uparrow^2), and the downward (\downarrow) arrows indicate the dates of manure
- 3 injections.



1

2 Fig. 0-6. (a) Hourly average air temperature and hourly average 10-20 cm soil temperature and (b) daily cumulative precipitation and
 3 hourly average 0-20 cm soil volumetric water content in the Edmonton site during the experimental period. The upward arrows (↑¹ or
 4 ↑²) indicate the dates of seeding (↑¹) and harvest (↑²), respectively. VWC = volumetric water content. Average hourly air temperature and
 5 cumulative daily precipitation data is derived from Alberta Agriculture and Forestry (2016).

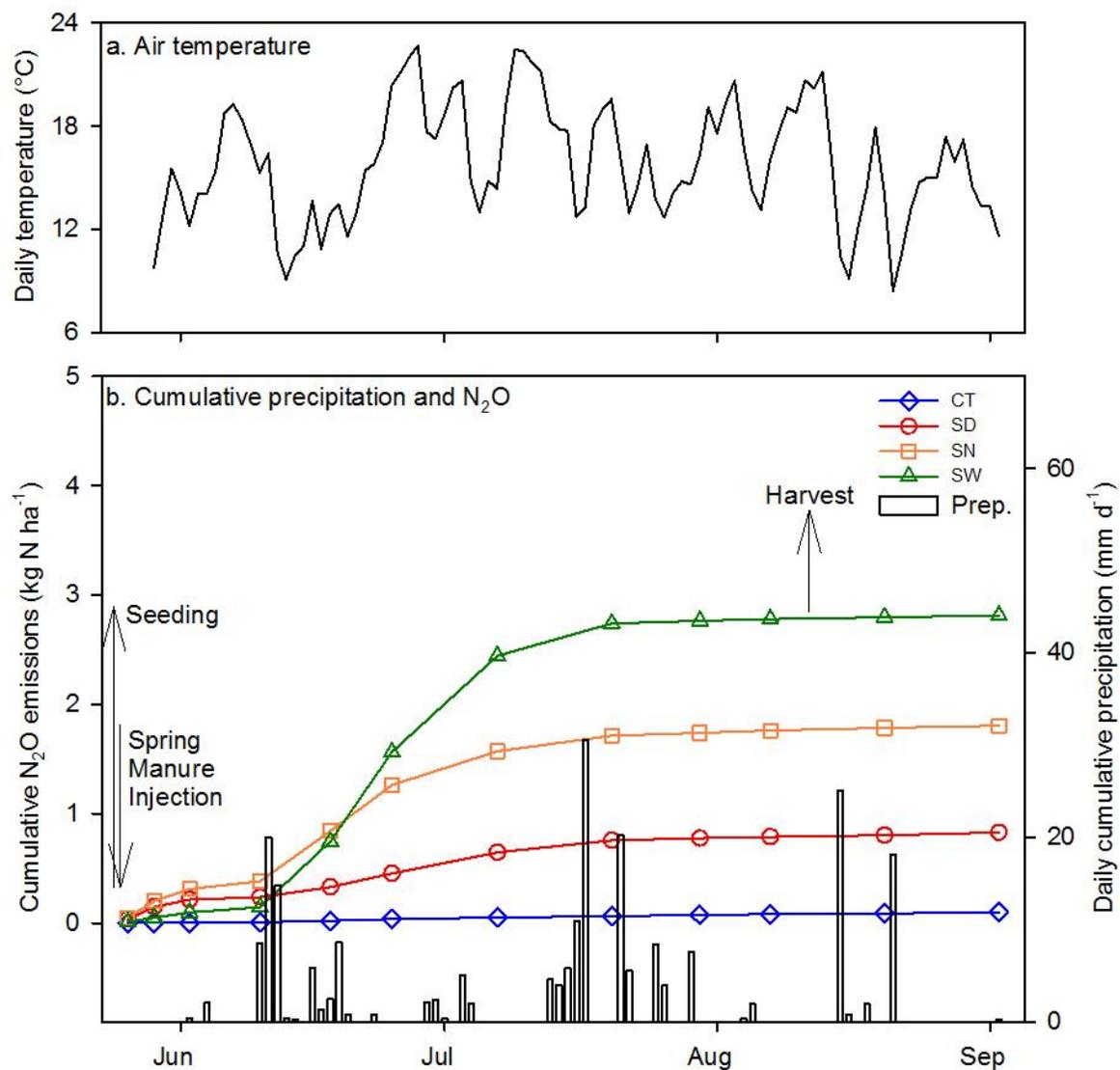


Fig. 0-7. (a) Average daily air temperature and (b) cumulative daily precipitation and N₂O emissions in the Lacombe site during the period from 26 May to 2 Sep 2015 after the spring manure injection. Error bars were not included for clarity. Average daily air temperature data is derived from Alberta Agriculture and Forestry (2016).

Scientific Achievements

(Note: We have acknowledged the funding agencies in every of these instances)

Sisi Lin; Guillermo Hernandez Ramirez; Len Kryzanowski; Trevor Wallace; Nils Berger; Rory Degenhardt; Craig Sprout; Huping Hou; Germar Lohstraeter; Leigh-Anne Powers. Use of Nitrification Inhibitors to reduce Nitrous Oxide Emissions from Crop Fields receiving Liquid Manure Injection in the Fall versus Spring. CCEMC AI Bio meeting. Edmonton AB. 1-2 Oct 2014

Sisi Lin*; Guillermo Hernandez Ramirez; Len Kryzanowski; Trevor Wallace ; Rory Degenhardt; Nils Berger; Craig Sprout; Germar Lohstraeter; Leigh-Anne Powers; Huping Hou. Preliminary Assessment of Nitrous Oxide Emissions, Soil Temperature and Soil Oxygen Concentrations following Fall Manure Injections. Alberta Soil Science Workshop, Edmonton. February 2015.

Sisi Lin*; Guillermo Hernandez Ramirez; Len Kryzanowski; Trevor Wallace ; Rory Degenhardt; Nils Berger; Craig Sprout; Germar Lohstraeter; Leigh-Anne Powers; Huping Hou. Assessment of Nitrous Oxide Emissions, Soil Ammonium and Nitrate under Controlled and Optimum Conditions. Soil Tillage & Research Organization, international meeting Nanjing. August 2015

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Journal papers submitted:

Assessment of Nitrous Oxide Emissions, Soil Ammonium and Nitrate under Controlled and Optimum Conditions. By S Lin* Hernandez Ramirez G, Kryzanowski L, Wallace T, Grant R, Degenhardt R, Berger N, Sprout C, Lohstraeter G, Powers L-A. Agriculture, Ecosystems and Environment AGEE16418

Nitrous Oxide Emission, Soil Temperature and Soil Oxygen Concentrations following Fall Manure Injections. By S Lin* Hernandez Ramirez G, Kryzanowski L, Wallace T, Grant R, Degenhardt R, Berger N, Sprout C, Lohstraeter G, Powers L-A.

Greenhouse Gas Impacts

We have redone calculations for nitrous oxide reduction in Alberta and carbon credit for manure and nitrification inhibitors using two scenarios for generation carbon credits: one conservative and one optimistic as follows:

- The conservative calculation follows that nitrous oxide emission can be typically reduced by inhibitors from 6.4 to 2.9 kg of N₂O per hectare per year generating 3.5 kg of N₂O per hectare per year as a net reduction. This net reduction corresponds to 1.04 Mg (or ton) CO₂ per hectare per year based on CO₂ equivalent (one N₂O molecule is equivalent to 298 CO₂ molecules). Based on manure production estimates about 55556 hectares of farmlands in Alberta receive manure application annually and a C credit price of \$30 per Mg CO₂, our 1.04 is multiplied by these factors resulting in 1.7 million dollars. Notice that the total existing farmlands in Alberta corresponds to 22 million hectares, and hence, our assumption of 55556 hectares of land is about 0.25% of the existing farmland in the province, so this is one of the conservative aspects of this estimate for GHG impacts.

- The optimistic calculation (based on our emission measurements in Lacombe in fields receiving manure injection using the best nitrification inhibitor) follows that nitrous oxide emission can be reduced from 9.1 to 2.4 kg of N₂O per hectare per year generating 6.7 kg of N₂O per hectare per year as a net reduction. This net reduction corresponds to 2.1 Mg CO₂ per hectare per year based on CO₂ equivalent. With an increase in livestock activities and assuming that 0.5% of the farmlands in Alberta receive manure application annually (110000 hectares) and the C credit price of \$30 per Mg CO₂, our 2.1 is multiplied by these two factors resulting in 6.6 million dollars. To promote the use of inhibitors with solid manure applications can further extend this market, so the potential is even larger for GHG impacts.

In addition, the implementation of inhibitors entails multiple benefits (not only the greenhouse gas reduction). The value proposition of this research includes also the gains in nitrogen use efficiency, plant productivity, and plant quality (nutritional composition). Nitrogen fertilization is one of the highest expenses in farming systems. The proposed research will generate savings to the industry by enhancing resources efficiencies and increasing the recycling of nutrients in

our production systems. Inhibitors have shown to increase plant nutrient availability and uptake; we will be quantifying and reporting these effects as a part of the proposed work.

Please notice that based on our new field measurements and the calculations done above the potential C credit can be stated for a conservative scenario as \$31 per hectare and for an optimistic as \$60 per hectare.

Furthermore, the retail price of nitrapyrin nitrification inhibitor (the commercial product is eNtrench) is 10.6 CAD per L (as consulted via phone with the manufacturer company). The active ingredient concentration is 200 g active ingredient (a.i.) per L, and hence, the cost of the inhibitor is CAD 53 per kg a.i.; since we are using a rate of 0.5 kg a.i. per Ha with liquid manure applications, this leads to a cost of CAD 26.5 per Ha in an area basis. As new inhibitor products are becoming available in the market, this cost will likely decrease. Recent experience indicates that every new inhibitor begins to be sold, the price of the new product is lower by 5 to 10 CAD compared to the pre-existing inhibitors. The cost of additives can also be expected to decrease as their use becomes more common and their market is further expanded in our region.

Overall Conclusions

Compared to the fall application of liquid manure, the spring timing enhanced the overall manure-N use efficiency and utilization by obtaining higher plant N uptake, higher plant dry matter yield, and lower risk of large annual N₂O emissions. Therefore, it is suggested that land injection of liquid manure should be conducted during mid-to-late spring in regions with comparable edaphic and climatic conditions as Alberta. Our study also indicates that the use of nitrification inhibitors (NIs) leads to more consistent N₂O emissions which otherwise are typically very variable, temporally erratic and unpredictable. Along these lines, the evaluated NIs were effective in reducing N₂O emissions although not statistically significant, and moreover, this effectiveness of the NIs was still functional in soils following a six-month freezing winter. Additionally, DMPP was even more effective in reducing N₂O emissions than nitrapyrin; however, this apparent advantage in retaining manure-applied N in the soils did not translate into differences in plant N uptake between these two NIs. Soil moisture content and nitrate concentration clearly arose as two key drivers of N₂O emissions.

Next Research Steps

We are already involved in research projects branching out from this project. For instance, we are conducting a follow up project to quantify the effects of varying nitrification inhibitor rates on N₂O emissions (where the liquid manure is treated with these nitrification inhibitors). This inhibitor rate project is a key follow up on this concluding manure project. A new project addressing the question of annual versus perennial grain on N₂O emissions. These perennial grains can putatively reduced the N₂O emissions during the early spring following soil thawing and snow melt.

Communications Plan

We will continue striving for publications of the key finding from this research project. The peer review process is a quality control. We will continue using popular media to spread the key findings from this research project. We have contacts with Grain News in Western Canada, and we will prepare public articles, research notes, and interviews in layman words to reach farmers and practitioners. We will keep using other local media for research translation purposes. We will continue sharing research insights in local conferences (e.g., FarmTech, Tri-Provincial Manure Management Conferences & Workshops) as well as regional meetings such as Agronomy Update and Alberta Soil Science Workshop.

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Final Financial Report

Please see enclosed financial reports.

The entire funding from ERA (CCEMC) Biological Management for a total CAD 75,000 for the duration of the project has been used for covering personnel expenses. This includes the graduate student stipend and other salary for technical personnel.

Please note that other funding sources (as a part of a larger partnership) have become available for covering other expenses such as travel, equipment, materials and supplies. On this respect, ALMA contributed with CAD 150,000 funding toward these various project expenses and Dow AgroSciences contributed with CAD 10,000 funding towards equipment for the project.

Non-Confidential Final Report

Final Outcomes Report for Project ID: B140392

Project title: Use of Nitrification Inhibitors to reduce Nitrous Oxide Emissions from Crop Fields receiving Liquid Manure Injection in the Fall versus Spring

Project outcomes

Sisi Lin; Guillermo Hernandez Ramirez; Len Kryzanowski; Trevor Wallace; Nils Berger; Rory Degenhardt; Craig Sprout; Huping Hou; Germar Lohstraeter; Leigh-Anne Powers. Use of Nitrification Inhibitors to reduce Nitrous Oxide Emissions from Crop Fields receiving Liquid Manure Injection in the Fall versus Spring. CCEMC AI Bio meeting. Edmonton AB. 1-2 Oct 2014

Sisi Lin*; Guillermo Hernandez Ramirez; Len Kryzanowski; Trevor Wallace ; Rory Degenhardt; Nils Berger; Craig Sprout; Germar Lohstraeter; Leigh-Anne Powers; Huping Hou. Preliminary Assessment of Nitrous Oxide Emissions, Soil Temperature and Soil Oxygen Concentrations following Fall Manure Injections. Alberta Soil Science Workshop, Edmonton. February 2015.

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Total project costs

CAD 738,250. This amount includes the in-kind contributions from the research collaborators as well as direct funding amounts for CAD 150,000 from ALMA Sustainability and CAD 10,000 from Dow AgroSciences.

ERA contributions

CAD 75,000

Expected greenhouse gas benefits

We have redone calculations for nitrous oxide reduction in Alberta for manure and nitrification inhibitors using two scenarios for generation carbon credits: one conservative and one optimistic as follows:

- The conservative calculation follows that nitrous oxide emission can be typically reduced by inhibitors from 6.4 to 2.9 kg of N₂O per hectare per year generating 3.5 kg of N₂O per hectare per year as a net reduction. This net reduction corresponds to 1.04 Mg (or ton) CO₂ per hectare per year based on CO₂ equivalent (one N₂O molecule is equivalent to 298 CO₂ molecules). Based on manure production estimates about 55556 hectares of farmlands in Alberta receive manure application annually. Notice that the total existing farmlands in Alberta corresponds to 22 million hectares, and hence, our assumption of 55556 hectares of land is about

0.25% of the existing farmland in the province, so this is one of the conservative aspects of this estimate for GHG impacts.

- The optimistic calculation (based on our emission measurements in Lacombe in fields receiving manure injection using the best nitrification inhibitor) follows that nitrous oxide emission can be reduced from 9.1 to 2.4 kg of N₂O per hectare per year generating 6.7 kg of N₂O per hectare per year as a net reduction. This net reduction corresponds to 2.1 Mg CO₂ per hectare per year based on CO₂ equivalent. With an increase in livestock activities and assuming that 0.5% of the farmlands in Alberta receive manure application annually (110000 hectares). To promote the use of inhibitors with solid manure applications can further extend adoption, so the potential is even larger for GHG impacts.

Final Outcomes Report for Project ID: B140392

Project title: Use of Nitrification Inhibitors to reduce Nitrous Oxide Emissions from Crop Fields receiving Liquid Manure Injection in the Fall versus Spring

Abstract and Keywords

Nitrous oxide (N₂O) contributes to global warming and ozone depletion. Two-thirds of the global N₂O emissions are derived from agricultural soils receiving manure or fertilizer applications. The goal of this project was to identify and develop management practices that can decrease N₂O emissions from cropland receiving liquid manure. We tested early fall versus late spring application of liquid manure in combination with two nitrification inhibitors (NIs; nitrapyrin vs. DMPP) admixed with the liquid manure. Two field experiments in central Alberta, Canada. Barley for silage was planted, and productivity and N uptake were recorded. Soil ammonium and nitrate concentrations and N₂O fluxes were repeatedly monitored. Compared to fields without manure controls, field N₂O emissions were increased with by manure application (3.15 vs. 0.45 kg N ha⁻¹ yr⁻¹), but emissions were sharply reduced with NIs. For instance, in the Lacombe site, fall manure treated with DMPP reduced annual N₂O emissions by 81%, and nitrapyrin reduced emissions by 58%. The emission reductions caused by NIs were also evident in the spring manure field treatments and at our Edmonton site, but the reductions magnitudes were typically smaller in associations with periods exhibiting drier conditions in particular in Edmonton. Compared to the spring manure timing, fall manure without NIs resulted in an approximate two-fold increase in N₂O emissions, due to major peak fluxes following the early spring snow-melt, which accounted for at least 65% of the annual N₂O emissions. Fall manure timing also reduced plant productivity and N uptake. In sum, spring manure with NIs can mitigate N₂O emissions in Alberta's agriculture and in regions with comparable agro-ecological conditions.

Keywords: nitrous oxide, liquid manure, nitrogen, nitrification inhibitors, nitrogen