

**NITROUS OXIDE EMISSIONS FROM INCORPORATED AND
SURFACE-APPLIED DAIRY MANURE AFTER
SIMULATED IRRIGATION EVENTS**

by

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ABSTRACT

The more than 300,000 dairy cows in the Texas Panhandle generate a considerable amount of manure. This manure is a valuable fertilizer, but growing concerns with greenhouse gas (GHG) emissions has prompted research into methods for reducing GHG from land-applied manure. The objectives of this research were to 1) quantify nitrous oxide (N_2O) emissions from surface-applied and incorporated dairy cattle manure, 2) determine how irrigation affects N_2O emissions, and 3) determine the mechanism for N_2O emissions. A laboratory study was conducted to compare N_2O emissions from four treatments (TRT) consisting of commercial fertilizer (U, urea), surface-applied manure (MS), incorporated manure (MI), and soil alone (S, control). Soil and manure were placed into glass containers (4 reps per treatment) and monitored for a 14-day period, during which two simulated irrigation events were applied. Emissions were measured from each container once per hour using a multiplexer and real-time N_2O analyzer. Nitrous oxide emissions were ranked (high to low): U, MI, MS, and S. While MI is often used as a best management practice to reduce ammonia emissions following land application, it produced higher N_2O emissions than MS. Emissions of N_2O increased immediately after simulated irrigation in all TRT. Based on initial and final soil nutrient concentrations, the N_2O was most likely generated from the nitrification of ammonium to nitrate. Further research is warranted to quantify GHG emissions from land-applied dairy manure under field conditions.

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TABLE OF CONTENTS

Chapter		Page
I.	INTRODUCTION.....	1
II.	LITERATURE REVIEW.....	8
	Objectives	25
III.	MATERIALS AND METHDOS.....	26
	Experimental Design.....	26
	Nitrous Oxide Emissions Apparatus.....	26
	Soil and Manure Samples.....	27
	Calculation of Nitrous Oxide Emissions.....	32
	Final Soil Analysis.....	37
	Data and Statistical Analysis.....	38
IV.	RESULTS.....	39
V.	DISCUSSION.....	48
	Nitrous Oxide Emissions.....	48
	Mechanism for Nitrous Oxide Production.....	49
VI.	RECOMMENDATIONS FOR FUTURE RESEARCH.....	53
VII.	CONCLUSIONS.....	54
VIII.	REFERENCES.....	55
IX.	APPENDICES.....	63

LIST OF FIGURES AND TABLES

Figures	Page
1. Total US greenhouse gas emissions (2017)	3
2. US agricultural carbon dioxide emissions	4
3. 2008 GHG emissions sources from ag sectors	7
4. Global greenhouse gas emissions by gas 1990-2010	12
5. Global carbon dioxide emissions by region 1990-2012	13
6. Relative concentrations of $\text{NH}_4^+/\text{NH}_3$	20
7. Multiplexer system to jar connection	29
8. Schematic of sampling system	30
9. Manure collected from the pen surface of the dairy	31
10. Manure was weighed before being placed into the jars.....	33
11. Monitor displaying N_2O emissions over time	34
12. Photo of sample jars and treatments prior to water addition.....	35
13. Illustration of jars arranged in divider boxes	36
14. Cumulative nitrous oxide emissions	40
15. Mean cumulative nitrous oxide emissions	41
16. Final nitrate+nitrite-nitrogen concentrations	42
17. Final mean nitrate+nitrite-nitrogen concentrations	43
18. Final mean organic nitrogen concentrations	44
19. Final mean pH concentrations	45
20. Final mean total nitrogen concentrations	46
Tables	
1. Mean nutrient concentrations	47

CHAPTER I

INTRODUCTION TO GREENHOUSE GAS EMISSIONS

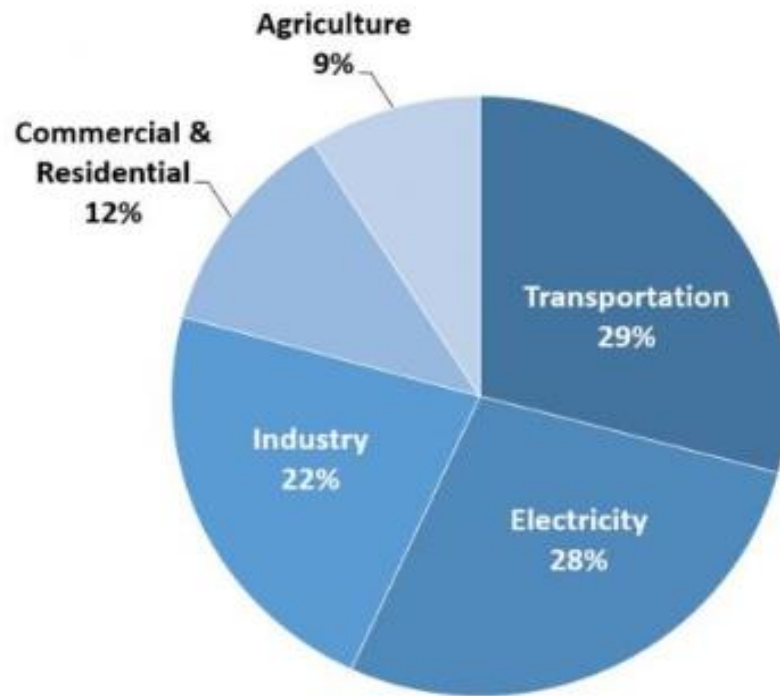
Gases that trap heat within the atmosphere are often referred to as greenhouse gases (GHG). They can be naturally occurring as well as anthropogenically produced (USEPA, 2015b). The main GHG of concern are carbon dioxide (CO₂), methane (CH₄), nitrous oxide (N₂O), and fluorinated gases including hydrofluorocarbons (HFC), perfluorocarbons (PFC) and sulfur hexafluoride (SF₆). Total GHG emissions are often expressed on the basis of metric tons of CO₂ and this convention will be used throughout this manuscript. These gases block heat from escaping through the atmosphere and amplify a condition commonly known as the “greenhouse effect” (IPCC, 2012; NASA, 2019). The greenhouse effect is a phenomenon that results when greenhouse gases absorb thermal infrared radiation that is emitted by the Earth's surface, the atmosphere, and clouds then reflect it back as heat. Many scientists and members of the public believe that the greenhouse effect is directly linked to global warming, although this correlation is often highly disputed (IPCC, 2012; NASA, 2019).

Global warming is the general increase of global temperatures, and there are natural drivers such as solar irradiance and volcanic activity (USGCRP, 2017). There are also anthropogenic drivers and it has been reported that GHG are a highly contributing factor behind this increase in global temperature (USEPA, 2015b). As a result of rising

global temperatures, scientists have drawn correlations that indicate climate change can be linked to global warming (IPCC, 2014). Climate change can occur due to natural internal processes, external forcing's, or continual anthropogenic changes in the structure of the atmosphere or in land use (IPCC, 2012). Greenhouse gas emissions can come from various sources as illustrated in Figure 1 (USEPA, 2017). Although there are varying percentages, each sector is involved with the other in one way or another. Thus, an intelligent research-driven approach in reducing greenhouse gas emissions is necessary.

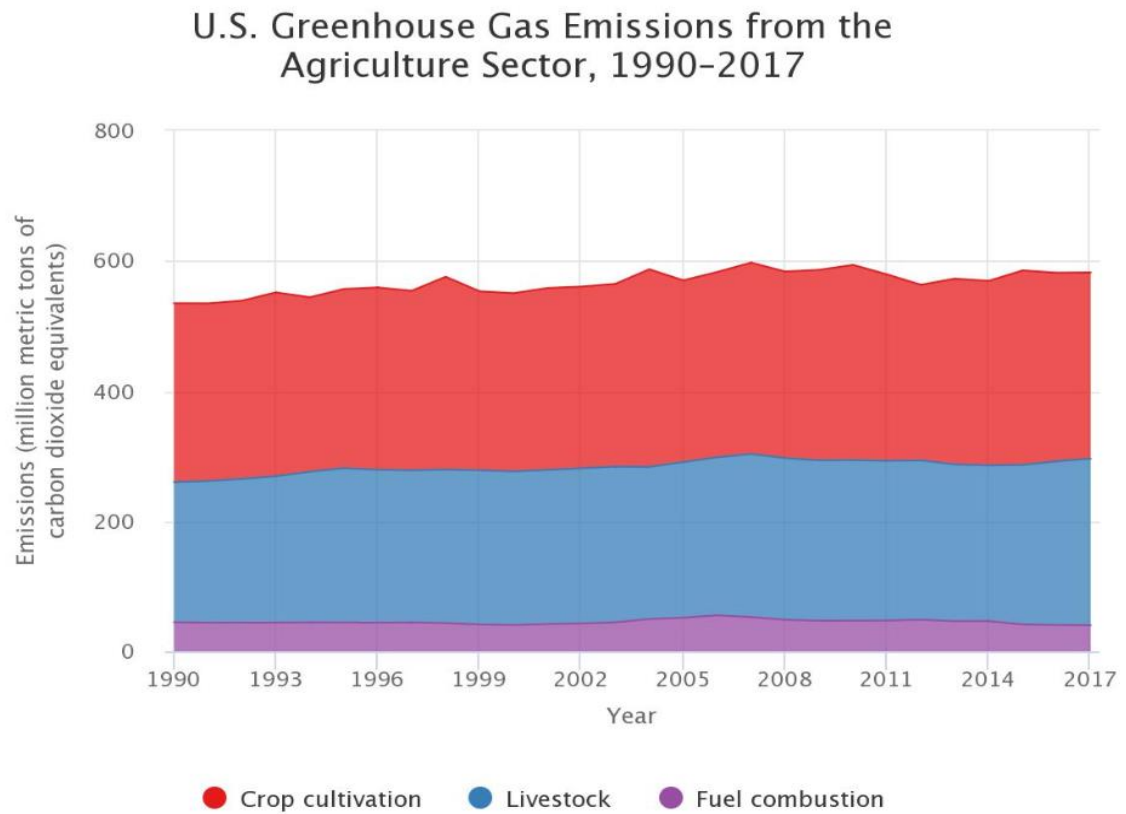
According to Figure 1, agriculture contributes a smaller amount of emissions relative to the other sectors. However, agriculture generates 58% of global non-anthropogenic non-carbon dioxide GHG emissions and 14% of all anthropogenic GHG emissions (Beach et al., 2008). A majority of the non-carbon dioxide emissions come from one of four sources within agriculture: 1) cropland soil management (primarily N_2O), 2) ruminant livestock enteric fermentation (primarily CH_4), 3) rice cultivation (primarily CH_4 from flooded rice paddies, though N_2O is also important under certain growing conditions), and 4) livestock manure management (both CH_4 and N_2O , with CH_4 from anaerobic manure management systems dominating) (Beach et al., 2008; USEPA, 2017). A general overview of US agricultural carbon dioxide emissions from 1990-2017 is presented in Figure 2 (USEPA, 2017b). Figure 2 also shows that a majority of agriculture-based emissions arise from crop cultivation and livestock.

Total U.S. Greenhouse Gas Emissions by Economic Sector in 2017



Total Emissions in 2017 = 6,457 [Million Metric Tons of CO₂ equivalent](#). Percentages may not add up to 100% due to independent rounding.

Figure 1. Total US greenhouse gas emissions for 2017 based upon economic sectors (USEPA, 2017).



Source: U.S. EPA's Inventory of U.S. Greenhouse Gas Emissions and Sinks: 1990-2017.
<https://www.epa.gov/ghgemissions/inventory-us-greenhouse-gas-emissions-and-sinks>

Figure 2. US agricultural carbon dioxide emissions from 1990-2017 (USEPA, 2017b).

A more detailed breakdown from 2008 of the emissions sources mentioned in Beach et al. (2008) and (USEPA, 2017) is shown in Figure 3 (Eve et al., 2014).

Agricultural emissions can come from various sources, however the application of manure and fertilizer as well as best management practices (BMP's) also play a role in emissions. Manure has historically been viewed and used as a valuable resource for soil nutrient restoration as well as a means for increasing crop production (VanderZaag et al., 2011). Manure contains several elements crucial to plant growth including nitrogen (N), phosphorous (P), and potassium (K). Manure also returns organic material to the soil along with other nutrients such as calcium (Ca), magnesium (Mg), as well as sulfur (S) and also helps contribute to soil fertility (Payne and Lawrence, 2019). To reduce financial costs and offset crop nutrient removal, integration of animal and crop production systems on the same farm are ideal (VanderZaag et al., 2011).

There are several manure land application methods. Selecting the best method depends on several factors including manure characteristics, cropping styles, soil types, and farm management constraints (VanderZaag et al., 2011). Manure is typically applied in one of two states: 1) a liquid state known as a slurry which is usually cattle, swine or poultry, or 2) a solid manure state. There are numerous application methods of these two states: application of slurry by trailing hose, application of slurry by trailing shoe, application of slurry by injection, rapid incorporation of slurry by cultivation of the soil, and incorporation of solid manures by cultivation (Webb et al., 2010).

Due to rising global population, animal production systems have intensified which has led to the creation of less costly synthetic N fertilizers (Liu et al., 2016; VanderZaag et al., 2011). Urea is one of these synthetic fertilizers, it is an inexpensive

form of nitrogen fertilizer that contains 46.65% N (UMN Extension, 2018). Urea is naturally occurring within the excreta of humans and land-based animals. Synthetic urea is the result of an industrial reaction of carbon dioxide with anhydrous ammonia and usually comes in the forms of prills or granules. Urea generally provides a high amount of N at a low cost compared to other available N sources. However, it is highly volatile so once it reaches the soil surface it can be easily converted into ammonia (NH_3) gas through volatilization (Killpack and Buchholz, 1993).

Volatilization is a chemical reaction that occurs at the soil surface when fertilizers such as urea are converted into ammonia gas and escape into the atmosphere (Schwenke, 2014; Killpack and Buchholz, 1993). With the various land application methods, there are several concerns that arise around the loss of nutrients and an increase in N_2O and NH_3 emissions following manure application procedures. One of the growing concerns is emission and nutrient loss when it comes to surface applied manure versus incorporating manure into the soil. Several studies have found that surface application of manure or urea-based fertilizers can result in ammonia losses ranging from 40-99% (Pfluke et al., 2011; Webb et al., 2010; Huijsmans et al., 2003; Meisinger and Jokela, 2000).

Ammonia volatilization is highly dependent upon environmental conditions and management practice such as weather conditions, soil moisture content, manure characteristics, and soil type (Meisinger and Jokela, 2000; Huijsmans et al., 2003). Application through injection and cultivation has shown to drastically reduce NH_3 emissions (Webb et al., 2010; Huijsmans et al., 2003). However, injection and cultivation have also demonstrated an increase in N_2O emissions, so there is a trade-off between NH_3 volatilization and N_2O emissions.

Emissions of N_2O are also increased in injection sites in moist soil due to the slurry concentrations at the injection sites causing anaerobic conditions which aid in N_2O production (VanderZaag et al., 2011).

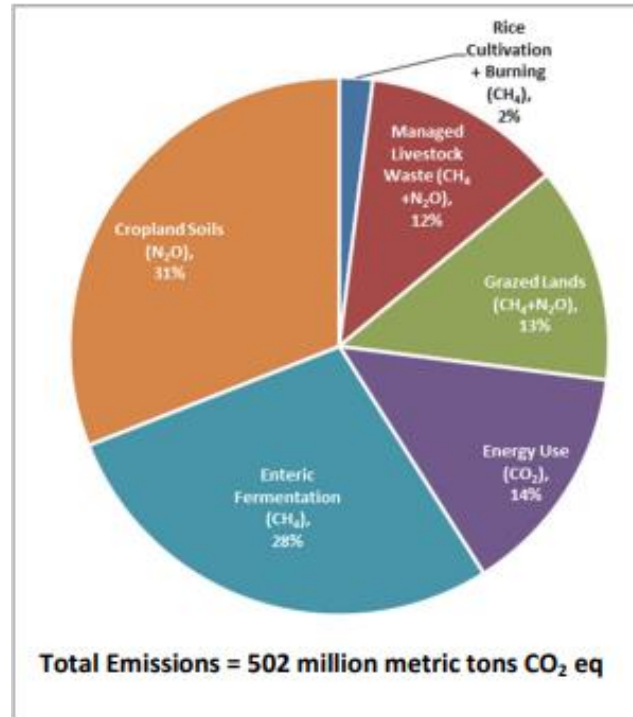


Figure 3. 2008 GHG emission sources within the agricultural sectors (Eve et al., 2014). Note that cropland soils emissions include emissions from major crops; non-major crops; histosol cultivation; and managed manure that accounts for the loss of manure nitrogen during transport, treatment, and storage, including volatilization and leaching/runoff.

CHAPTER II

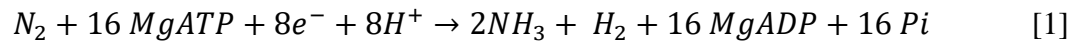
LITERATURE REVIEW

The nitrogen cycle is a complex cyclical system that has implications and effects well beyond its own internal system. Nitrogen is a primary nutrient that is fundamental for all living creatures because it is a biological component of proteins, DNA, and chlorophyll (Bernhard, 2010; Gruber and Galloway, 2008). Nitrogen is abundant within our atmosphere in the form of diatomic nitrogen gas (N_2), which is also referred to as molecular nitrogen. However, this form of nitrogen is largely unusable by plants via assimilation which makes it a limiting factor in primary plant tissue production and growth in both land and marine ecosystems (Bernhard, 2010; Gruber and Galloway, 2008). Sub optimal growth for plants can be attributed to lack of nitrogen while over supplying them can negatively impact water and nutrient acquisition of the plant as well as nitrogen uptake itself resulting in environmental and crop production issues (Topcu and Kirda, 2013).

For N_2 to become viable for plants or other primary producers it needs to be converted to ammonium (NH_4^+). To convert N_2 into a biologically available format a process known as nitrogen fixation must occur. Nitrogen gas (N_2) is very stable in the atmosphere due to the strength of its triple bonds. These bonds require a large amount of energy to break (Bernhard, 2010). Due to the high energy barrier that needs to be

overcome with directly breaking this bond, only a select group of prokaryotes can facilitate this fixation process. Nitrogen fixation can also be carried out abiotically by industrial processes due to high amounts of heat and pressure associated with these processes.

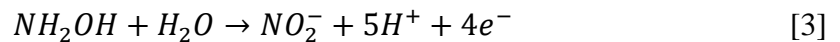
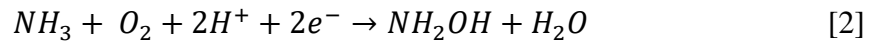
Nitrogen fixing organisms are either symbiotic or free-living, the symbiotic relationship that is required for nitrogen fixation among plants is facilitated through the nodules found on vascular plants in two specific groups of bacteria, *rhizobia* (*Alpha-proteobacteria*) and *Frankia* (Actinobacteria). Another group of nitrogen fixing bacteria are cyanobacteria which are found among fungi and algae (Frache et al., 2009). Legume plants work specifically with *rhizobia* by exuding signals from their roots that attract the bacteria. The bacteria are then absorbed via the root hairs of the plants where they then multiply and stimulate the formation of root nodules (Bernhard, 2010; Frache et al., 2009). Once the bacteria have integrated with the plant the enzymatic process of conversion from N_2 to NH_4^+ can occur. The conversion reaction is catalyzed by nitrogenase which is an oxygen-labile enzyme. The mechanism behind nitrogen reduction is extremely complex and has not been fully understood but the net reduction of N_2 to NH_4^+ have been generally described using the Eq.1.



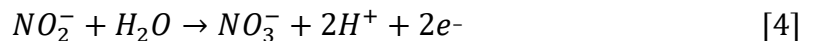
As previously stated, the triple bonds that are between N_2 require a large amount of energy to break that is why this reaction requires eight electrons and at least sixteen ATP molecules. A problematic characteristic of nitrogenase is that the enzyme is sensitive to oxygen and is deactivated if the presence of oxygen is high enough. As a result, nitrogen fixing bacteria have adapted via respiratory chains in oxygen conditions,

cyanobacteria have adapted by creating structures known as heterocyst's that provide a low oxygen environment and a site for nitrogen fixation (Bernhard, 2010; Franche et al., 2009).

Another pathway in the nitrification cycle is nitrification. Nitrification is the conversion process of ammonium to nitrite and then to nitrate. A majority of nitrification is aerobically occurring and is carried out exclusively by autotrophic prokaryotes that use ammonia as energy instead of light. This reaction is a two-step process, with the first step being the oxidation of ammonia to nitrite which is carried out by microbes known as ammonia-oxidizers bacteria (AOB). Common AOB's associated with this process are *Nitrosomonas* (Bernhard, 2010; USEPA, 2002). Aerobic ammonia oxidizers convert ammonia to nitrite via the intermediate hydroxylamine, a process that requires two different enzymes, ammonia monooxygenase and hydroxylamine oxidoreductase as shown in Eq. 2 and Eq. 3 (Prosser, 2005).

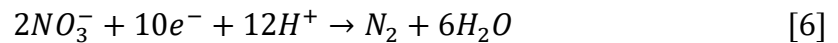
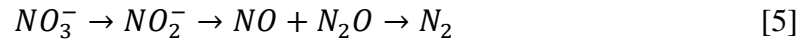


The second step of nitrification is the oxidation of nitrite (NO_2^-) to nitrate (NO_3^-) and this step is carried out by nitrite oxidizing bacteria. This process is commonly facilitated by *Nitrobacter* bacteria as shown in Eq. 4.



Still another pathway in the nitrogen cycle is denitrification which is the conversion of nitrate to N_2 . While N_2 gas is the end product of this reaction there are several intermediate gasses that are produced throughout the reaction. One of these intermediate gasses is N_2O which has been considered a greenhouse gas and a

contributor to climate change as well as global warming (Bernhard, 2010; Takaya et al., 2003). Denitrification unlike nitrification is an anaerobic process occurring mostly within the wet soils and lagoons with limited oxygen. One form of the denitrification reaction is shown in Eq. 5 and Eq. 6 shows the complete redox reaction of denitrification (Bernhard, 2010).



Denitrification is driven by several prokaryotes commonly found in the *Pseudomonas*, *Bacillus*, and *Paracoccus* genera (Bernhard, 2010; Takaya et al., 2003). Denitrification removes fixed nitrogen (nitrate) from various ecosystems and then returns it to the atmosphere in its unusable form of dinitrogen gas. With an increase in use of nitrogen-based fertilizers from agriculture over the past 40-50 years there has been a drastic increase in the amount of biologically available nitrogen which in turn has allowed for an increase in N₂O emissions of up to 0.3% per year (Takaya et al., 2003).

Nitrous oxide is one of three major GHG that result from agriculture. The other two are CH₄ and carbon dioxide CO₂. Greenhouse gases have been defined by the Intergovernmental Panel on Climate Change (IPCC) as “Those gaseous constituents of the atmosphere, both natural and anthropogenic, which absorb and emit radiation at specific wavelengths within the spectrum of thermal infrared radiation emitted by the Earth’s surface, by the atmosphere itself, and by clouds” (IPCC, 2012). These specific gasses cause an effect known as the greenhouse effect by absorbing solar radiation which has been reflected as thermal infrared radiation after being emitted by the Earth’s

surface, the Earth's atmosphere due to these very same gasses, and clouds. Greenhouse gasses essentially trap heat within the surface troposphere. This trapping of heat along with net incoming solar radiation causes the average temperature of the Earth's surface to be 14°C (IPCC, 2012; USEIA, 2019). This greenhouse effect is also naturally occurring. However global greenhouse gas emissions have been increasing over the past two decades. From 1990-2010 global emissions of all greenhouse gases significantly increased (Figure 4). Net emissions of CO₂ increased by 42%, N₂O increased by 9%, CH₄ had increased by 15%. Anthropogenic fluorinated gasses HFC's, PFC's, and SF₆ more than doubled (USEPA, 2016a).

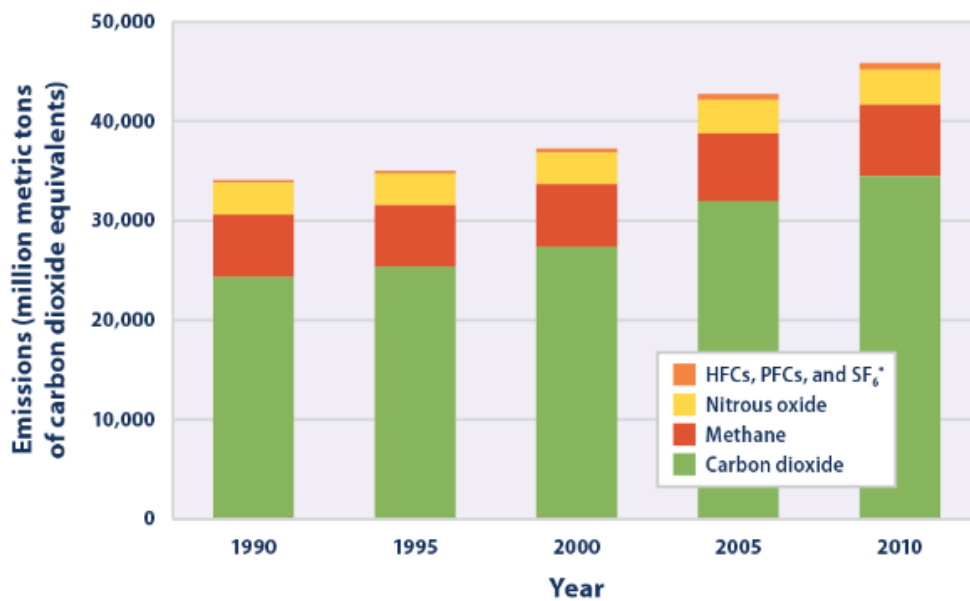


Figure 4. Global greenhouse gas emissions by gas for 1990-2010. For consistency emissions are expressed in million metric tons of carbon dioxide equivalents (from USEPA, 2016a).

In 2010 worldwide estimated emissions resulting from human activities had amounted to 46 billion metric tons of greenhouse gasses expressed as CO₂ equivalents.

This number represent an increase of 35% from 1990 (USEPA, 2015). These numbers are including forestry and land use which are typically viewed as emission sinks instead of sources. Land-use change, and forestry typically represent a net sink for emissions in the United States, absorbing carbon dioxide and offsetting emissions from other sources. However, due to global deforestation these sinks are now considered emission sources. Energy production represented 71% of global emissions in 2010 with agriculture representing 13% (USEPA, 2016a). Global emission percentages however can be skewed as they consider total emissions and tend to disregard or mask regional input. Some areas of the world are larger contributors to emissions than others. For example, in 2012 the three of regions; Asia, Europe and the United states accounted for 88% of total estimated global emissions (Figure 5; USEPA, 2016a).

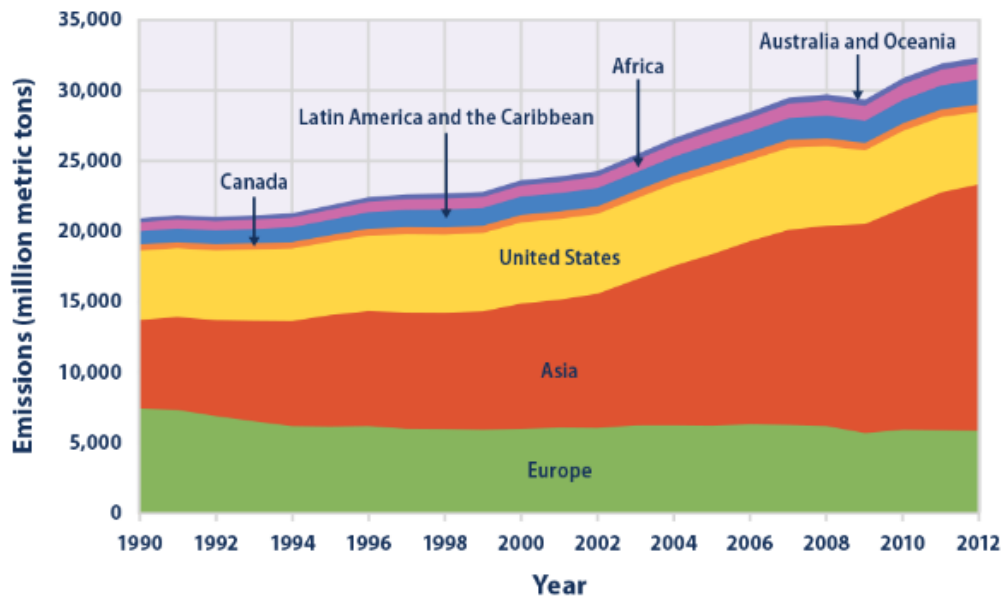


Figure 5. Global carbon dioxide emissions by region from 1990-2012 (from USEPA, 2016a). Totals do not include emissions or sinks related to land-use change or forestry.

For the United States during 1990-2014 carbon dioxide emissions increased by 9%. Methane emissions decreased 6% in this same time span and can largely be attributed to reduced emissions from coal mines, landfills and natural gas systems. Nitrous oxide emissions resulting from primarily agricultural practices decreased by only 1%. Fluorinated gasses emissions increased by 77%, largely due to increases in industrial, commercial and household uses (USEPA, 2016b).

Humans are often viewed as the primary drivers of global changes either through climate change, reduced water quality, loss of biological diversity or changes in ecosystems. The anthropogenic practices that drive these changes are increased population, energy use, and per capita consumption (Ussiri and Lal, 2013). Climate change is natural due to internal or external forcing's upon the Earth's system. However, excessive anthropogenic drivers or forcing's to Earth's natural energy balance can cause excessive climate change (Ussiri and Lal, 2013). A prime example of these forcing's is excess greenhouse gasses or changes to greenhouse gas emissions. These forcing's create positive or negative climate feedbacks. A positive climate feedback amplifies the response of climate forcing's. An example would be the increase of water vapor emissions caused by an increase in warming due to an increase in GHG emissions which in turn amplifies warming due to the properties of water vapor (Ussiri and Lal, 2013).

Climate forcing's can be radiative or nonradiative as well as direct or indirect. Radiative forcing's (RF) is the concept that was developed to quantify anthropogenic and natural influences upon the environmental climate system. (Ussiri and Lal, 2013;

Unger et al., 2010). Anthropogenic factors include: increases in GHG emissions, decreases in stratospheric ozone, and changes in land cover/use. Increased GHG emissions is the best understood scenario of the positive anthropogenic RF's. This is largely due to the absorption of IR radiation. Changes in land use and cover mainly for clearing of forested areas for cropland and farmland have changed the reflective properties of the land (Ussiri and Lal, 2013). The increase in anthropogenic drivers has increased the reflection of solar radiation from the Earth but due to the increase of GHG and the greenhouse effect this forcing effect is amplified and the larger the forcing the larger the disruption of global surface temperature (Ussiri and Lal, 2013).

Nitrous oxide emissions are major natural and anthropogenic drivers while CO₂ is cycled throughout the agriculture system in larger quantities than N₂O. However nitrous oxide has a global warming potential (GWP) 265-300 times that of CO₂ over a 100-year time scale making it a more potent GHG (Butterbach-Bahl et al., 2016; Snyder et al., 2009). GWP was developed to allow comparisons for global warming impacts of different GHG. Global warming potential is essentially a measure of how much energy the emissions of 1 ton of a gas will absorb over a period of time. This is relative to the emissions of 1 ton of CO₂ (USEPA, 2016c). Carbon dioxide has a GWP of 1 and the larger the GWP the more that specific gas warms the Earth relative to CO₂.

Nitrous oxide is also the dominant stratospheric ozone depleting emission with a substantial RF associated with agriculture (Griffis et al., 2017; Unger et al., 2010). The agricultural sector is the largest producer of anthropogenic N₂O with most emissions

resulting from soil application of nitrogenous fertilizers as well as manure management and application (Bracmort, 2010; O'Mara, 2011).

Nitrous oxide emissions are a result of the soil microbes that are active in nitrification and denitrification as they both release N_2O . However, the amount is dependent upon oxygen concentration within the soil as well as moisture content (Snyder et al., 2009). Nitrous oxide emissions can also occur indirectly when nitrogen is lost through ammonia volatilization or nitrate leaching which is converted to N_2O in another location (VanderZaag et al., 2011). Nitrous oxide emissions that are emitted from cropland soils are usually the largest source of greenhouse gasses from agricultural systems and manure application of nitrogen-based fertilizers are one of the major determinants of N_2O emissions (Beach et al., 2008; Huijsmans et al., 2003). Emissions are also heavily influenced by manure application method, weather conditions, soil moisture content, and manure characteristics (Huijsmans et al., 2003; Flessa and Beese, 2000). Manure application methods allow for high spatial variability in emission rates often resulting in either NH_3 loss via volatilization or NO_3^- and N_2O loss through leaching and emissions (Powell and Rotz, 2015). Excess N that is not utilized by plants is lost through emissions, leaching and runoff (Beach et al., 2008).

Manure application techniques are broad in the individual application but can fit within three general categories: 1) surface spreading, 2) surface incorporation, and 3) deep placement. There are large spatial differences and variability that accompany each application technique in terms of emission rates although it is generally agreed upon that ammonia volatilization greatly reduces the amount of nitrogen available

within manure applied to agricultural land (Huijsmans et al., 2003; Pfluke et al., 2011).

Ammonia volatilization occurs when NH_4^+ in manure or solution is converted to dissolved ammonia gas illustrated by Eq. 7 (Meisinger and Jokela, 2000).

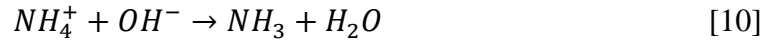
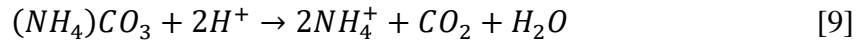
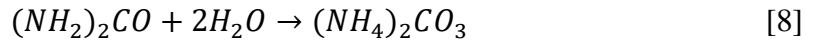


This reaction essentially produces more NH_3 as pH or temperature increases and as the NH_4^+ concentration increases therefore we see higher emission and volatilization rates in wetter and warmer soils (Meisinger and Jokela, 2000; Griffis et al., 2017).

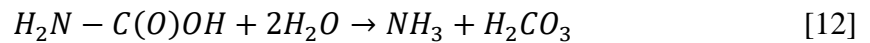
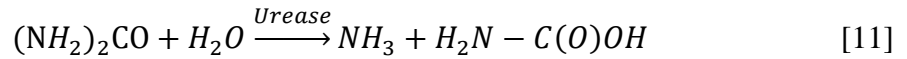
Ammonia volatilization can vary greatly with environmental conditions. However, with surface application ammonia volatilization rates can come close to 100% (Meisinger and Jokela, 2000). Meisinger and Jokela (2000) found that land application of manure contributed close to 46% of ammonia emissions from livestock in the UK, while also finding that liquid slurry surface application volatilization losses typically ranged from 40-70%. Huijsmans et al., (2003) found through records of 25 field experiments on various locations in the Netherlands between March and September using field applied liquid pig manure that the mean total volatilization which was expressed as a percentage of the total ammoniacal nitrogen (TAN) applied was 68% for surface spreading, 17% for surface incorporation and 2% for deep placement. However, McGinn and Sommer (2007) found that, in some instances, rainfall can increase nutrient filtration into soil from surface applied manure, thereby reducing volatilization. In one study conducted in Northern Germany NH_3 loss during windy and warm periods was 56% of applied $\text{NH}_4^+\text{-N}$, while during a cool rainy period this rate was reduced to 42%. There is also variability of NH_3 volatilization within the surface application of a

liquid cattle and poultry manure. Meisinger and Jokela (2000) confirmed that losses from dairy slurries in the spring in Ontario were 24% to 33%. However, losses from solid dairy manure application in several studies conducted in New York were 61% to 90%. Meisinger and Jokela (2000) also found that ammonia losses from surface-applied poultry litter in Europe are within the range of 15% to 45%, while ammonia losses from spring surface-applied poultry litter to pastures in Southeast United States ranged from 28% to 46%. This data shows that generally ammonia losses from poultry litter is between 20% to 45% TAN while cattle slurry losses are between 35% to 70% (Meisinger and Jokela, 2000). There is high spatial variability in the ammonia losses and volatilization between these studies. This is largely due to regional differences between soil composition, manure characteristics and moisture content. However, one thing is apparent, surface-applied manure tends to lead to a higher volatilization rate of NH_3 . Ammonia volatilization rates are usually highest immediately after slurry application Pfluke et al., (2011) found that on average 70% of total NH_3 losses occur within the first three hours after application while VanderZaag et al (2011) found in a study of cattle slurry surface application to grassland that more than half of total NH_3 loss occurred within the first six hours and up to 90% in the first 35 hours. Meisinger and Jokela (2000) reviewed 10 studies using cattle slurry surface application to grassland and found that 30% to 70% of total ammonia loss occurred within the first four to six hours and 50% to 90% within the first 24 hours. One explanation for rapid loss during slurry application can be attributed to the slurry matrix given that the well mixed liquid manure is well supplied with urease. The urease facilitates urea hydrolysis which increases the pH by degassing. Urea hydrolysis occurs once the slurry is applied,

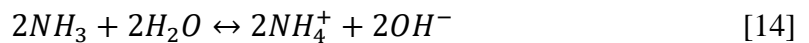
increasing pH which is a known contributing factor to increased ammonia volatilization (Meisinger and Jokela, 2000; Griffis et al., 2017). Urease is a metalloenzyme containing nickel, which catalyzes the hydrolysis of urea into ammonium carbonate. Ammonium carbonate spontaneously decomposes into ammonium, carbon dioxide and water. Ammonium is then converted to ammonia gas and carbon dioxide as shown in Eq. 8, 9, and 10.



Urea catalyzes the hydrolysis of urea to yield ammonia and carbamate. The carbamate spontaneously decomposes into ammonia and carbonic acid. A more detailed enzymatic process is shown in Eq. 11 and Eq. 12 (IPNI, 2019; Mazzei et al., 2016; Mobley, 2001).



In aqueous solutions carbonic acid and the two molecules of ammonia are in equilibrium with their deprotonated and protonated forms, respectively. The net effect of these reactions is an increase in pH (Eq. 13 and Eq. 14).



The conversion of ammonium to ammonia gas is controlled by pH. As hydrolysis occurs near the urea prills, the initial pH will rise. Once the pH exceeds 7.5 to 8.0 ammonia volatilization increases dramatically as shown in Figure 6 (Hunter et al., 2013; IPNI, 2019).

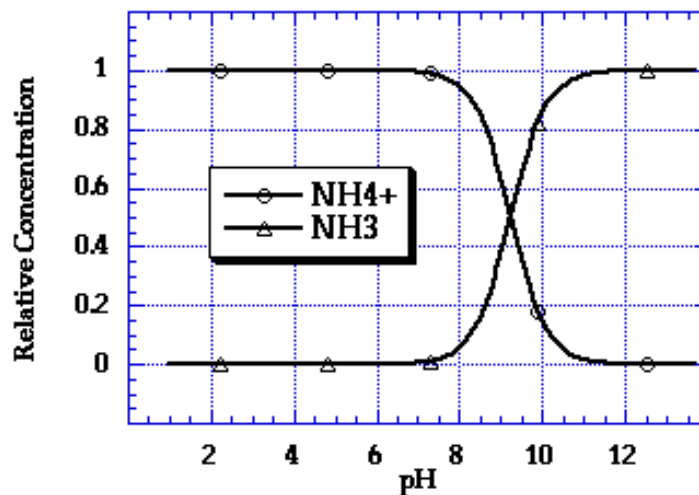


Figure 6. Relative concentrations of NH_4^+ and NH_3 as pH increases (from Cornell Waste Management Institute, 1996; Richard, 1996).

To reduce NH_3 volatilization and loss, researchers have discovered that slurry shallow injection greatly reduces ammonia losses by decreasing exposure time of the manure surface to air. Improved contact with the soil can yield reductions in NH_3 as high as 90% from shallow injection as compared to surface broadcasting (Pfluke et al., 2011; Webb et al., 2010). However, shallow injection is not as effective with higher soil moisture content or soil compaction, as these factors tend to lead to incomplete

injection coverage (Sommer and Hutchings, 2001). Surface incorporation of slurry is typically done through tillage immediately after application using rigid tines, spring tines, discs or harrows. Deep placement or shallow injection are done through spike injection, open slot injection (OSI), arable land injection or various other injection techniques.

Nyord et al., (2008) and Hansen et al., (2003) found that in order for shallow or deep injection to be effective the injection must be at least 50 mm deep, while deep injection is typically in depths greater than 100mm. In a review VanderZaag et al., (2011) reported that deep injection of slurry was the best application for reducing NH_3 losses within arable lands, with a 90% reduction compared to surface application on bare soils. However, for grasslands shallow injection showed lower reductions of 70%. These results were also confirmed by Misselbrook et al. (2002) who found that shallow injection of cattle or swine slurry reduced NH_3 losses by 73%.

In comparison Huijsmans et al. (2003) reviewed 25 field experiments conducted in the Netherlands and found that the weighted means expressed as total cumulative volatilization were 68% for surface spreading, 17% for surface incorporated and 2% for deep placement. Surface spreading was conducted using a tank filled with a splash plate with a net width of 8 m. Surface incorporation was conducted using the conventional methods previously mentioned and deep placement was done using an arable land injector at depths ranging from 150-200 mm. McGinn and Sommer (2007) found that tillage of the surface prior to manure application can enhance infiltration into the soil

reducing NH_3 volatilization between 27% and 50% for injection and surface applied slurry, respectively.

Thompson and Meisinger (2002) used mouldboard, chisel plow and disc harrow to immediately incorporate cattle slurry. They reported NH_3 emissions were reduced by 90%, 83% and 99% respectively. As with surface application techniques there will be variability within each situation and the optimal application methods depends largely on soil conditions. Shallow injection is not ideal in dry clay soil conditions because the injection slots cannot be cut, and in wet clay soils a trailing shoe can cause soil sealing which prevents infiltration and increases NH_3 volatilization (VanderZaag et al., 2011). While there is spatial variability in emissions with each manure application technique it has been widely acknowledged and reviewed that surface injection and incorporation of manure drastically reduces NH_3 volatilization and losses. However, application methods can also greatly affect N_2O emission rates as well.

Webb et al., (2010) found that techniques that tend to reduce NH_3 emissions are expected to increase N_2O emissions because more allowable N entering the soil via either incorporation or surface injection. VanderZaag et al., (2011) reviewed several studies and found that N_2O fluxes from injecting cattle slurry compared to surface application of the same slurry were higher. Injection did lower NH_3 emissions, further showing the tradeoff between application practices and reducing NH_3 emissions while increasing N_2O emissions. In a laboratory study conducted by Flessa and Beese (2000) cattle slurry was injected to 100 mm depth and resulted in higher N_2O emissions than

surface applied manure with total N₂O-N emissions at 0.2% for surface applied and 3.3% for slit injection.

VanderZaag et al. (2011) also reviewed a field study finding that N₂O emissions were highest when slurry was injected before planting spring barley and were about 50% lower when slurry was applied in surface bands and incorporated within the first hour. VanderZaag et al. (2011) also found that injection tends to decrease N₂O losses in dry soils, but it increases in moist soils due to the slurry being concentrated at the injection sites causing anaerobic conditions that promote N₂O production. Wulf et al. (2002) converted N₂O and NH₃ to CO₂ equivalents in a study comparing tilled cropland and grassland as well as various application techniques. They found that injection of slurry to 100 mm caused the lowest NH₃ emission but resulted in N₂O emission rates two to three times higher compared to other application methods. Nitrous oxide emission rates ranged from 10% to 200% on both sites with the mean variability of the arable land at 60% and the grassland site of 145%.

Regarding the presented information and literature, one of the conclusions that may be drawn is that one of the best approaches to reduce direct and indirect N₂O emissions is largely based upon soil conditions. From the literature it is evident that on well drained arable soils, injection is preferred to reduce NH₃ volatilization and has a minimal effect on N₂O. With soils that are considered moist surface spreading with immediate incorporation seems preferable as this minimizes NH₃ loss and it avoids slurry anaerobic conditions and N₂O emissions with injection. Regarding grasslands a trailing shoe is the preferred method as emissions from both N₂O and NH₃ are

minimized except if soil sealing takes place which typically occurs in wet clay soils preventing infiltration which increases NH_3 volatilization (VanderZaag et al., 2011).

It has become more apparent that there is a need for better management and application processes especially with the amount of manure produced from dairy cattle within the Texas Panhandle. In 2014 there was an estimated 285,000 dairy cattle within the Texas Panhandle according to (AgriLife Today, 2014). Assuming that the dairy cows are lactating and on average produce 35 kg (80 pounds) of manure per day per 448 kg (1,000-pound) animal unit, resulting in 4.92 Mg (5.422 million tons) of wet manure for that year (USDA, 1995). Parker et al. (2017a) reported that 6 million beef cattle produce around 14.5 Mg (16.0 million tons) of wet manure annually in the Texas High Plains in 2015. These large feedlots and dairies present themselves as large sources of potential N_2O emissions as well as NH_3 losses. While there has been substantial research conducted to validate and quantify emissions rates of N_2O from these facilities there is still a large amount of spatial variability in the biophysical and management conditions that affect production and emissions rates. There is also large variability within the presented literature, and this is largely due to different measurement methods and equipment. Accurate quantification and validation of emission rates is difficult since many factors need to be taken into consideration (time of year, soil moisture, type of slurry deposited, application technique, manure characteristics).

As a result of this literature review, a better understanding of the relationship between GHG emissions and agriculture is required to produce better BMP's with respect to manure land application.

Objectives

The objectives of this research were as follows:

1. Quantify and compare N₂O emissions from surface-applied and incorporated dairy cattle manure.
2. Determine the effect of simulated irrigation events on N₂O emissions, and
3. Determine the mechanism for N₂O emissions from land-applied dairy manure.

CHAPTER III

MATERIALS AND METHODS

Experimental Design

There were four treatments with four replications per treatment, for a total of 16 experimental units (EU). Nitrous oxide emissions were measured on each EU using a real-time N₂O analyzer. Each EU consisted of a glass container with a 750 ml volume, 75 mm diameter, and 4,418 mm² surface area. Treatments were applied randomly to each EU. Four containers of each treatment were labeled with container number and treatment to keep the treatments clearly defined:

Treatment 1 – Soil only (S), containers #5, 2, 1, 16

Treatment 2 – Soil + urea (U), containers #9, 4, 14, 3

Treatment 3 – Soil + manure on surface (MS), containers #6, 8, 11, 12

Treatment 4 – Soil + manure incorporated (MI), containers #15, 7, 10, 13

Nitrous Oxide Emissions Apparatus

The sampling apparatus consisted of 16 glass containers (Figure 7). Each container was connected to a multiplexer and a Los Gatos real-time N₂O analyzer (Figure 8). Each container was connected through two pieces of plastic tubing of 444.5 mm (17.5

in) and a diameter of 3.1 mm (0.125 in). When a single container was sampled, the sampling air was recirculated at a rate of 400 mL/min between the container and the analyzer. With this closed system, the concentration of N₂O in the sampling loop increased over time. Clean air was passed through each of the other 15 containers at a rate of 40 mL/min when not being sampled. A container was sampled every 3 min 45 sec, such that every container was sampled once per hour. Following a 165 sec flushing period the next chamber was sampled. During the 60 second sampling period the multiplexer valves were adjusted in a way, so the container became a small recirculating flow through – non steady state (RFT-NSS) chamber (i.e. a static chamber system) with N₂O concentrations being measured every 2 seconds.

Soil and Manure Samples

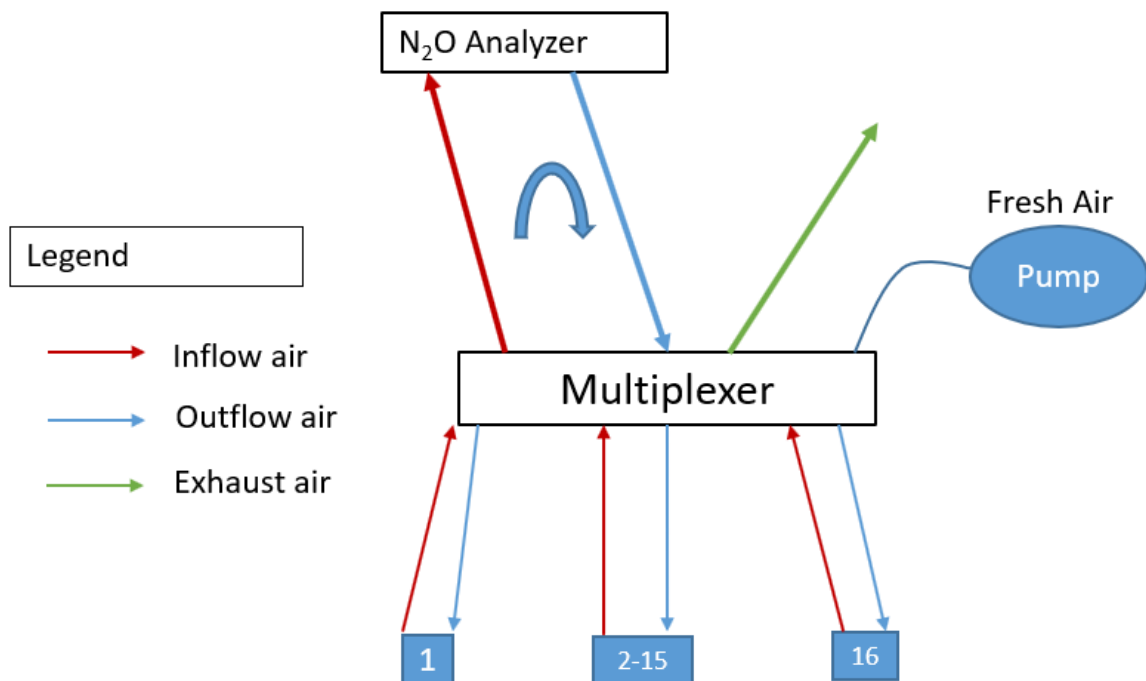
Manure samples were collected manually from a commercial dairy located near Hereford, TX on October 2019 (Figure 9). The loose manure was scraped from the top 50 mm of the pen surface. 50 mm is considered the dry, friable layer and anything below this not attainable by normal means due to compaction. Also, earlier research has shown that this loose surface layer is responsible for the majority of N₂O emissions (Parker, 2019).

Soil was collected from Terrace 47 at the USDA-ARS Conservation and Production Laboratory in Bushland, TX. Only the top 125 mm was disturbed and collected. The soil was Pullman clay loam which is a predominant soil type in the Texas Panhandle, with taxonomic class of *fine, mixed, superactive, thermic Torrertic Paleustolls* (Pringle, 1974).

Moisture content (MC) of manure and soil were determined using the gravimetric method with an isotemp oven. Soil samples with initial mass of 245 to 265 grams were analyzed for MC in triplicate. Manure samples with initial mass of 98 to 101 grams were also analyzed for MC in triplicate. All samples were weighed and oven-dried for 24 hours at 105°C.



Figure 7. Each of the 16 glass containers (top) were connected to the multiplexer system and real-time N₂O analyzer (bottom) with two lengths of tubing. During each measurement period, air was recirculated between the container and analyzer, resulting in a static chamber system.



While the multiplexer is sampling container it is a closed loop inflow/outflow system. While multiplexer is not sampling it is flushing with fresh atmospheric air via pump.

Figure 8. Schematic of the sampling system which consisted of 16 containers, a multiplexer and real-time N₂O analyzer. Sampling air was drawn by vacuum from each container, through the multiplexor, and into the analyzer.



Figure 9. Manure was collected from the pen surface of a local dairy.

A sample of manure was sent to ServiTech Laboratories in Amarillo, TX to determine total nitrogen content via the Kjeldahl Method. The treatments receiving manure or urea (treatments 2, 3, and 4) all received the same amount of total nitrogen by mass. Manure was applied at a rate of 22.42 Mg ha^{-1} (10 t ac^{-1}) to treatments 3 and 4. Each container received 100 mm (380.0 grams) of well mixed homogenous soil to create equivalent headspace within each EU. Treatments 3 and 4 received 9.91 grams of air-dried manure and treatment 2 received 0.277 grams of urea in the form of prills, equivalent to a nitrogen application rate of 288 kg ha^{-1} (257 lb ac^{-1}) (Figure 10).

Calculation of Nitrous Oxide Emissions

The monitors displayed concentrations of N_2O emissions within the headspace of each container as air was circulated continually through the inflow/outflow system. Temperature was also displayed in real time making the detection of inaccurate or false readings immediate (Figure 11).

The containers were set in numerical order in divider boxes, while the treatments among the units were randomly applied. Each divider box had spaces for nine containers. Temperature and relative humidity sensors were placed between the boxes to record measurements in the atmosphere outside of the containers. The thermostat in the room was set to 21.1° C (70° F) and was held constant throughout the trial. In addition, the containers were placed into an industrial heating blanket (model HB64-1440, Powerblanket, Salt Lake City, Utah) with internal and external temperature probes to keep the temperature constant. An example of how the samples were arranged is illustrated in Figure 12 and Figure 13, which shows the arrangement of jars that were

used in this experiment. The orange line in Figure 13 is a dividing line meant to show two separate divider boxes.



Figure 10. Manure was weighed (blue containers, foreground) prior to being placed into the glass containers. On the left are the two divider boxes into which the glass containers were placed prior to being placed into the temperature-controlled heating blanket (large black box on left).

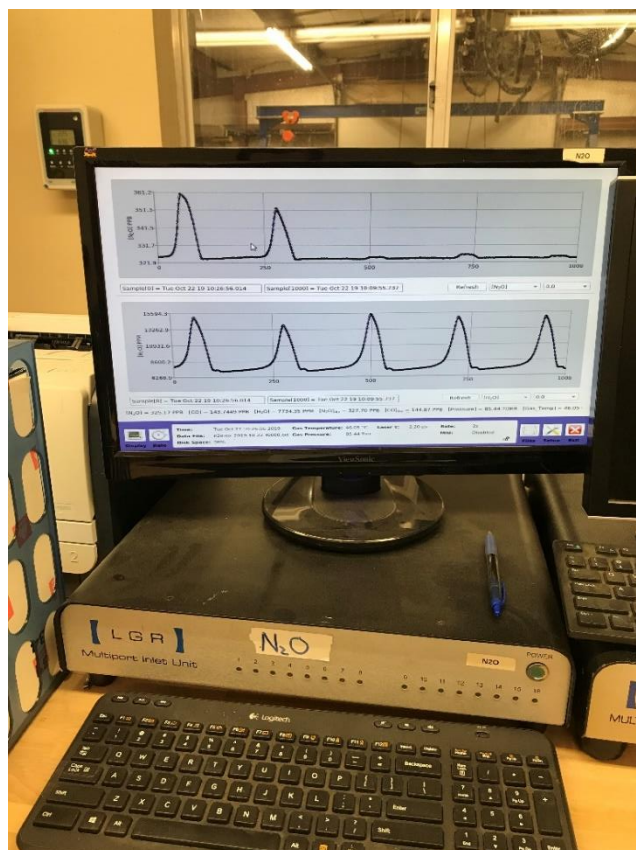


Figure 11. Monitor displaying N₂O emissions over time. Each rise and drop in concentrations corresponded with the measurement of a different container.



Figure 12. Sample illustration of jars arranged in divider boxes, prior to the addition of simulated irrigation. There were four replicates per treatments. Treatments were (top row to bottom row): Soil only (S), Urea (U), Manure on Surface (MS), and Manure Incorporated (MI).

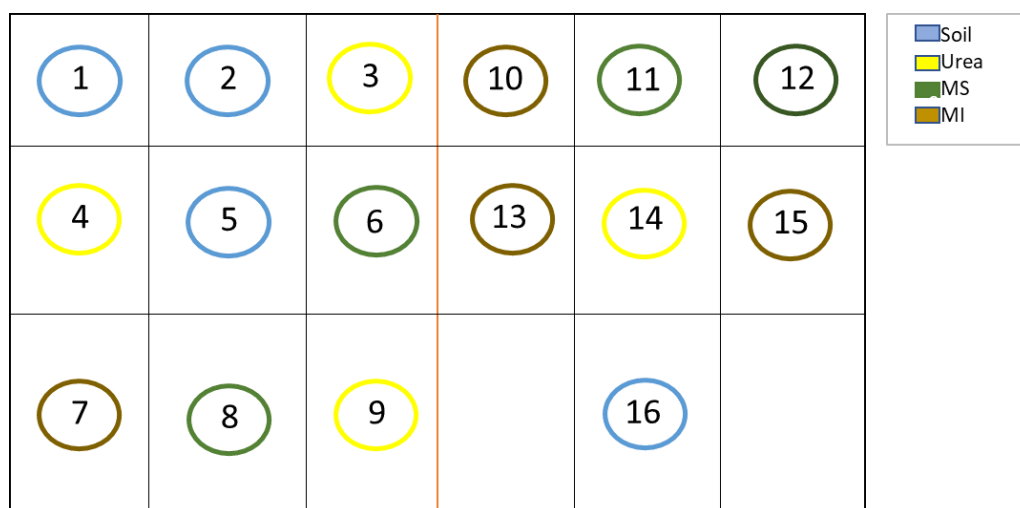


Figure 13. Illustration of how the sample jars were arranged in the divider boxes prior to placement within the temperature-controlled heating blanket.

Measurements were conducted using a real-time N₂O analyzer (model N₂O/CO-30-EP Enhanced Performance, Los Gatos Research, Inc., San Jose, CA). The N₂O analyzer uses laser absorption technology based on off-axis integrated cavity output spectroscopy. N₂O flux was calculated using the slope of the N₂O concentration versus time curve using linear regression (Parkin and Venterea, 2010). Nitrous oxide concentrations were measured from the container headspace every 2 seconds during each 60 second measurement time period (Parker et al., 2017a). After 24 hours of starting the

analyzers, 12.3 mm of water was added to container #1 via an automatic pipette, to simulate an irrigation event and monitor its effect on emission rates. This was repeated for each container 10 minutes before the analyzer performed a measured emission reading for each treatment. An additional 12.3mm of water was added to each container seven days after the initial irrigation event to simulate the second irrigation event. Emissions were continually measured from each container once per hour for 14 days, for a total of 5,376 individual emission measurements.

Final Soil Analysis

At the completion of this experiment, the soil and/or manure in each container was thoroughly mixed, and a sample was collected. Samples were analyzed for nitrate + nitrite nitrogen, pH, organic nitrogen, total nitrogen, and total Kjeldahl nitrogen concentrations by Servi-Tech Laboratories (Amarillo, Texas). Total and organic N were determined by the Kjeldahl method. Nitrate (NO_3^-) plus nitrite (NO_2^-) was analyzed by colorimetric flow injection analysis (FIA) according to EPA methods 353.2. Water content was determined using Standard Method 2540 by loss on drying. pH was determined with a probe meter 1:1 water to manure mixtures according to EPA method 9045D.

Data and Statistical Analysis

The cumulative nitrous oxide emission data was organized in a Microsoft Excel spreadsheet, where the raw data was converted from parts per million volume (ppmv) to mg m^{-3} using the ideal gas law. At the end of the 14-day trial the spreadsheet contained 632,168 lines of data amounting to 74 megabytes (MB).

Statistical analyses of the N_2O flux data was conducted using the SAS PROC ANOVA procedure (SAS Institute, 2013). Cumulative N_2O emissions were calculated using Microsoft Excel by numerically integrating the area under the flux versus time curves (Parker et al., 2017a). Post Hoc comparison of means was conducted using the Tukey's Test, which controls the experiment-wise error rate (Haynes et al., 2013). Statistical significance was determined by $\alpha = 0.05$ significance level.

CHAPTER IV

RESULTS

There was a malfunction in Jar 14 of the urea treatment. Nitrous oxide emissions in Jar 14 were essentially zero throughout the experiment, most likely due to a defective valve(s). The cumulative N₂O emissions from the other three jars of the urea treatment were 9.0, 9.80, and 10.58 mg Nm⁻²h⁻¹, with a mean of 9.79 mg Nm⁻²h⁻¹ and a standard deviation of 0.79 mg Nm⁻²h⁻¹. The cumulative N₂O emissions for Jar 14 were -0.32 mg Nm⁻²h⁻¹, which greatly is outside three standard deviations of the mean, (which encompasses 97% of datapoints). Thus, it was determined that Jar 14 was an outlier (Olewuezi, 2011). Therefore, the nitrous oxide data from Jar 14 was not used in the subsequent emissions analyses.

Cumulative nitrous oxide emissions varied greatly over time (Figure 14). All treatments had an immediate increase following water addition, only urea had a continued increase. During the first event incorporated manure displayed the highest peak, while at the second event surface applied manure was the highest. The urea treatments displayed a delayed slow rise emission rate as opposed to a spike.

The statistical distribution of groupings for N₂O emissions (A-D) shows distinct statistical differences between the treatments (Figure 15). Urea had the largest average emission rate and produced 3.45 times more N₂O than MI and 4.97 times more than MS.

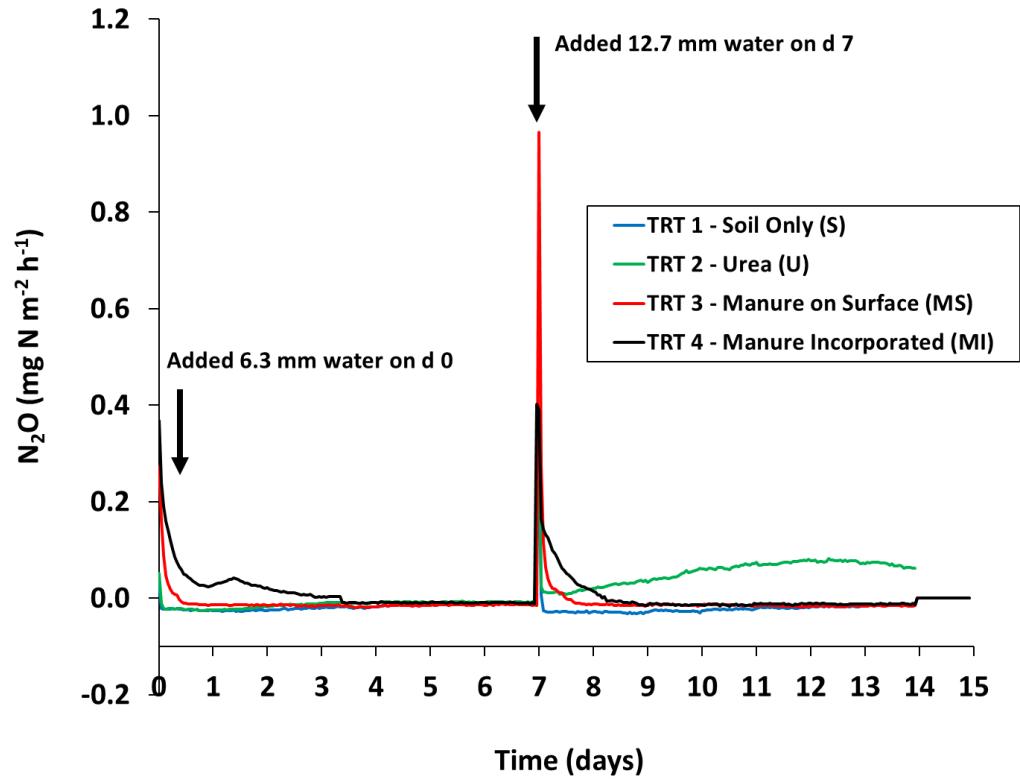


Figure 14. Cumulative nitrous oxide emissions varied greatly over time, with spikes in emissions immediately following simulated irrigation events.

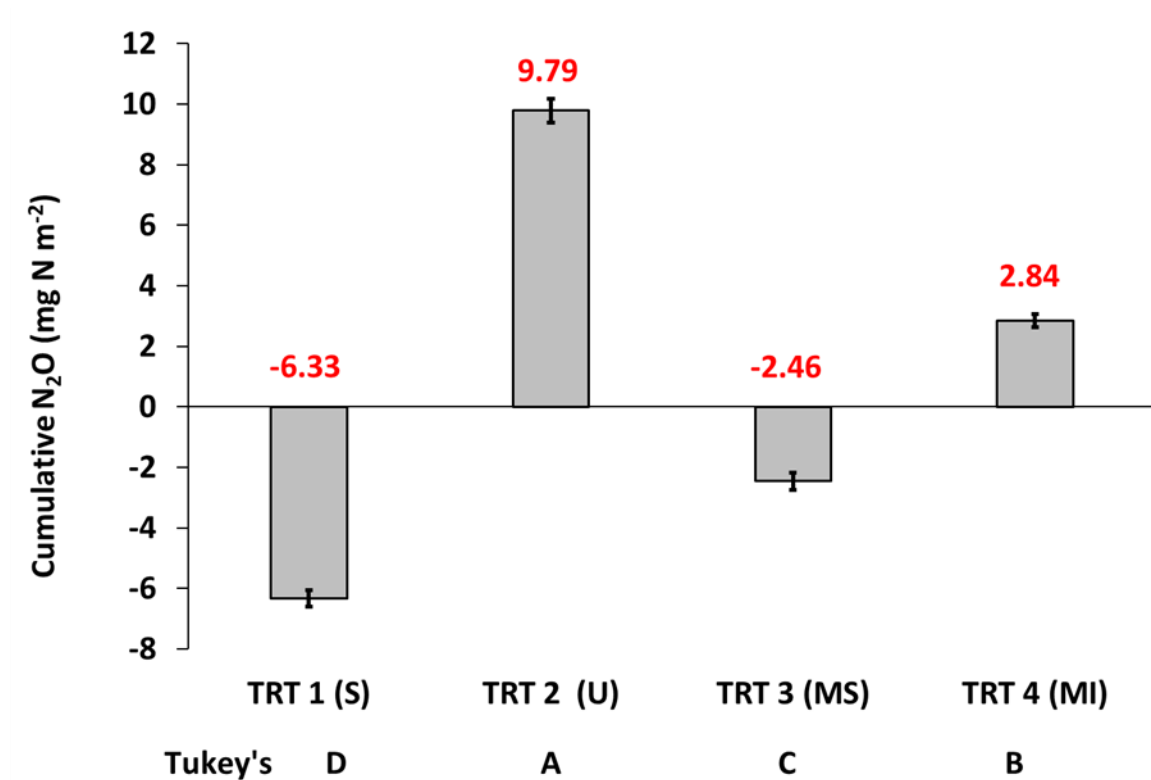


Figure 15. Mean cumulative nitrous oxide emissions were considerably different among the four treatments (S=soil; U=soil + urea; MS= soil + surface-applied manure; MI=incorporated manure). Values in red above the bars are the individual means. Error bars are the standard error of the mean for each treatment. Treatments with different letter (A-D) denote significant difference in means using Tukey's HSD test at $\alpha=0.05$.

Final total nitrate + nitrite concentrations show that the urea treatments had highest final concentrations (Figure 16). The manure incorporated treatments had the second highest followed by the surface-applied manure.

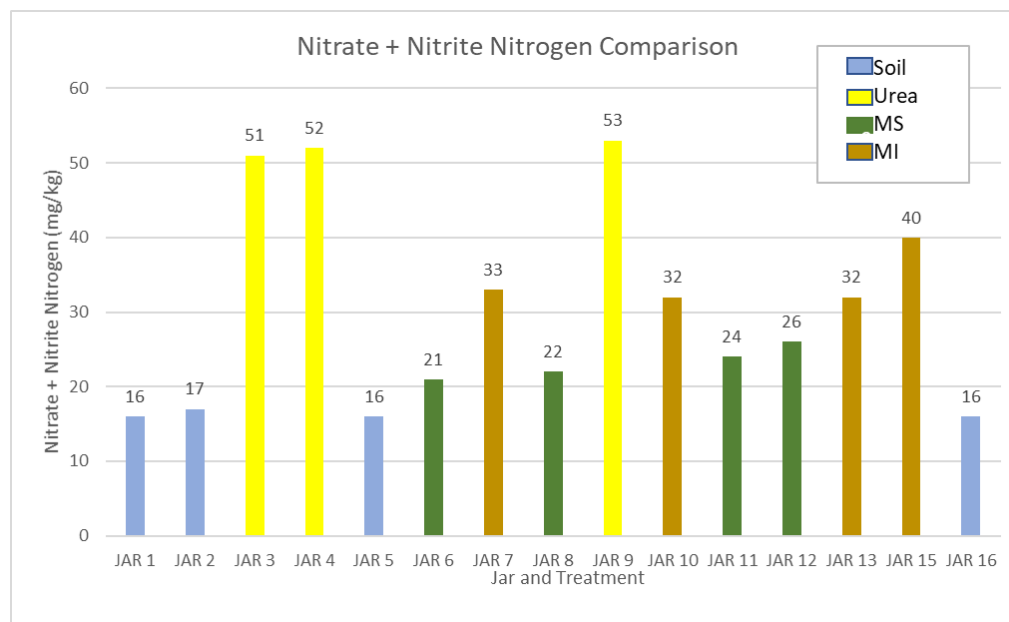


Figure 16. Final nitrate + nitrites-nitrogen concentrations for each treatment.

The distribution of groupings for ending soil nitrate concentration (A-D) shows distinct statistical differences between the treatments (Figure 17, Table 1). Urea had the largest final concentrations and contained 1.5 times more than the MI treatment and 2.2 more than the MS treatment.

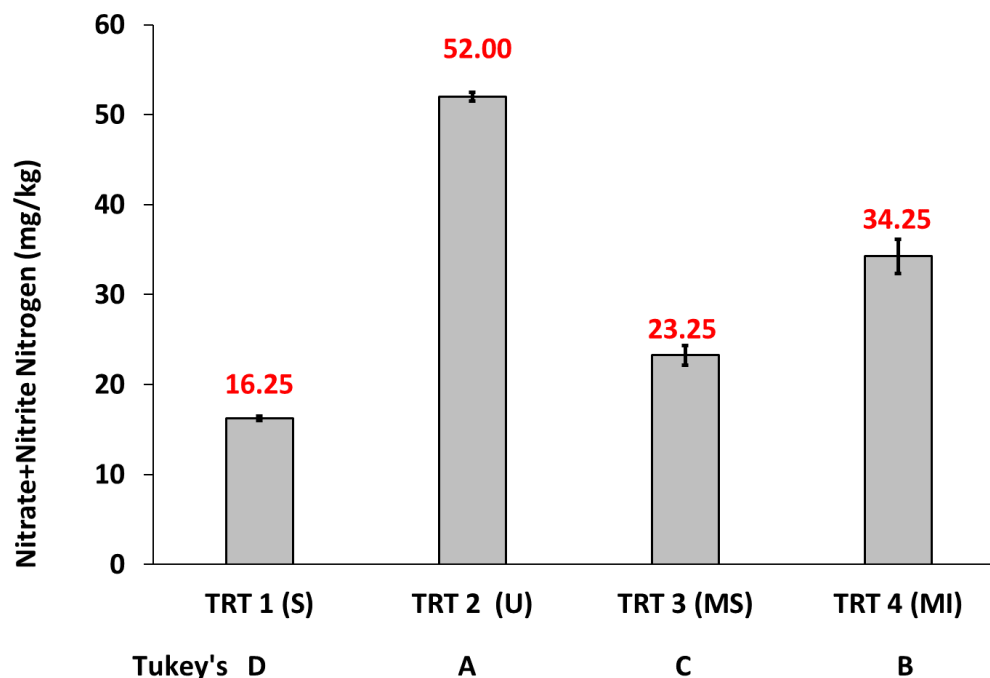


Figure 17. Final mean nitrate + nitrite nitrogen soil concentrations measured at the completion of the experiment. (S=soil; U=soil + urea; MS= soil + surface-applied manure; MI=incorporated manure). Values in red above the bars are the individual means. Error bars are the standard error of the mean for each treatment. Treatments with different letter (A-D) denote significant difference in means using Tukey's HSD test at $\alpha=0.05$.

The distribution of groupings (A-C) for organic nitrogen shows distinct statistical differences between the treatments (Figure 18, Table 1). Statistically MS and MI were similar as were U and MS treatments; S treatment was distinct from the other three treatments.

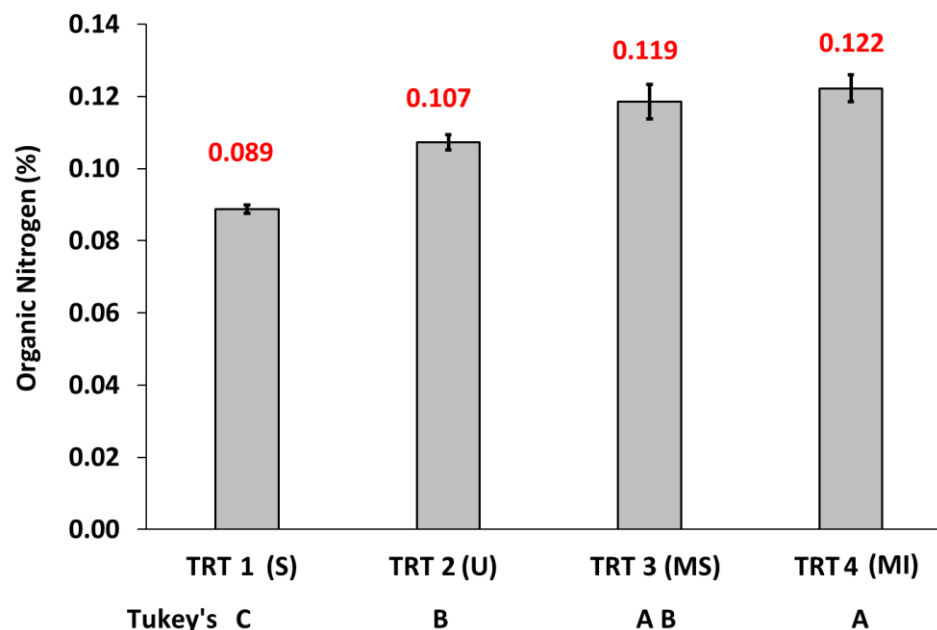


Figure 18. Final mean organic nitrogen soil concentrations measured at the completion of the experiment. (S=soil; U=soil + urea; MS= soil + surface-applied manure; MI=incorporated manure). Values in red above the bars are the individual means. Error bars are the standard error of the mean for each treatment. Treatments with different letter (A-D) denote significant difference in means using Tukey's HSD test at $\alpha=0.05$.

The distribution of groupings (A) for final soil pH shows no statistical differences between treatments at the end of the experiment (Figure 19, Table 1). Statistically treatments 1-4 were all similar.

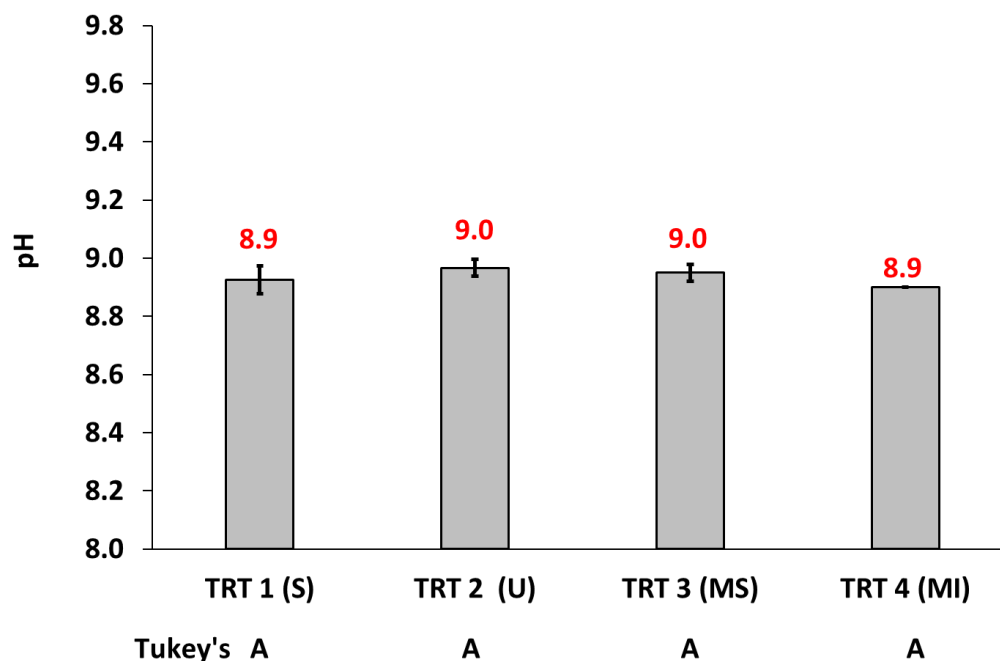


Figure 19. Final mean pH soil concentrations measured at the completion of the experiment. (S=soil; U=soil + urea; MS= soil + surface-applied manure; MI=incorporated manure). Values in red above the bars are the individual means. Error bars are the standard error of the mean for each treatment. Treatments with different letter (A-D) denote significant difference in means using Tukey's HSD test at $\alpha=0.05$.

The distribution of groupings (A-B) for final total nitrogen soil concentrations showed distinct statistical differences between the treatments at time of completion (Figure 20, Table 1). The S treatment was lower in total nitrogen compared to the other three treatments.

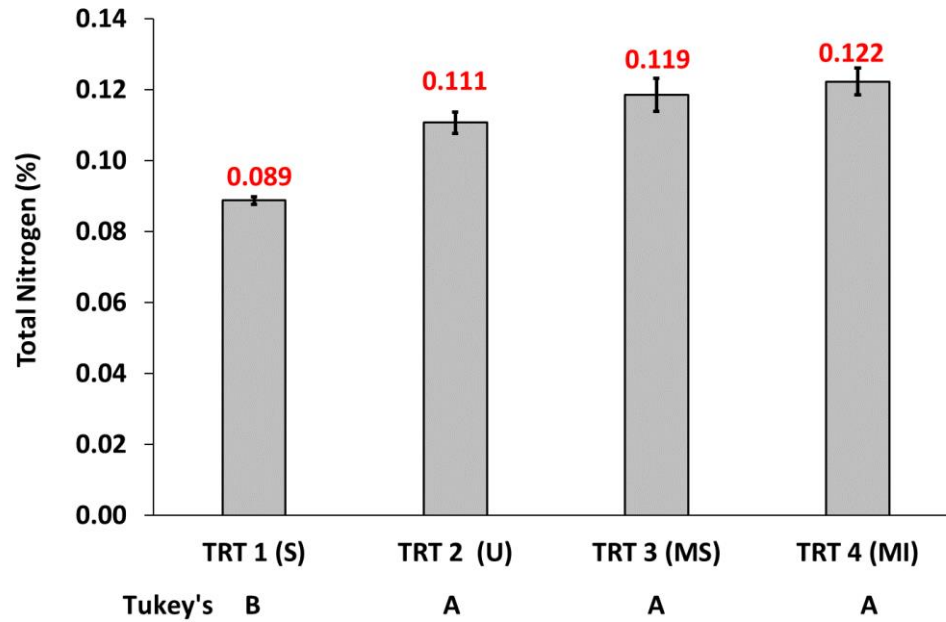


Figure 20. Final mean total nitrogen soil concentrations measured at the completion of the experiment. (S=soil; U=soil + urea; MS= soil + surface-applied manure; MI=incorporated manure). Values in red above the bars are the individual means. Error bars are the standard error of the mean for each treatment. Treatments with different letter (A-D) denote significant difference in means using Tukey's HSD test at $\alpha=0.05$.

Table 1. Mean N₂O emissions and nutrient concentrations for each treatment measured in the soil at the completion of the experiment.

	TRT 1	TRT 2	TRT 3	TRT 4
	(S)	(U)	(MS)	(MI)
Cumulative N ₂ O (mg N m ⁻²)	-6.33 _d	9.79 _a	-2.49 _c	2.48 _b
Nitrate + Nitrite -N (mg/kg)	16.25 _d	52.00 _a	23.25 _c	34.25 _b
Organic Nitrogen (%)	0.089 _c	0.107 _b	0.119 _{ab}	0.122 _a
pH	8.9 _a	9.0 _a	8.9 _a	9.0 _a
Total Nitrogen (%)	0.089 _b	0.111 _a	0.119 _a	0.122 _a

Means with a different letter (a-d) within a row are significantly different using the Tukey test at $\alpha < 0.05$.

Treatments: S=soil; U= Urea; MS= Surface-applied manure; MI= Incorporated Manure

CHAPTER V

DISCUSSION

Nitrous Oxide Emissions

Table 1 values show some indication as to the end products associated with each treatment. The nitrate and nitrite concentrations are indicative of the treatments that were driven toward nitrification (urea). Organic nitrogen concentrations show accessibility to nitrogen or nitrogen usage within the treatments. Final pH concentrations show a more alkaline or basic soil constitution across treatments which could be indicative of ammonia volatilization. In Figure 15, the distribution of groupings (A-D) shows distinct statistical differences in mean cumulative nitrous oxide emissions between the treatments.

Urea had the largest average emission rate and produced 3.5 times more N_2O than the incorporated treatment. Urea also had, 4.9 times more N_2O than the surface applied treatment, and 2.5 times more than the soil only treatment. The Tukey's HSD test showed that each treatment was statistically different than all other treatments (Figure 15). With a $P < 0.01$, it was statistically evident that the means of each respective treatment were different. This laboratory study has shown that there is a compelling statistical difference in nitrous oxide emissions between manure application practices (Figure 15).

Surface-applied manure seems to produce more nitrous oxide emissions initially while the urea treatment sustains more emissions over time. However, the surface applied treatment also becomes a sink relatively quickly due to what seems to be nitrogen sequestered within the soil via microbes or organic matter. This was not observed on the second irrigation event for urea which was also a surface application method.

The incorporated manure treatment behaved as expected. Due to incorporation of manure, the ammonia volatilization from the surface was minimized but the nitrous oxide emissions were increased. This can be seen in the cumulative means analysis (Figure 15).

Mechanism for Nitrous Oxide Production

It has been suggested that the manure characteristics in a dairy or beef feedyard pen which can include elevated nitrate, ammonium, and soluble carbon concentrations, together with a spatially-variable and temporally-variable water and oxygen concentrations, are ideal for production and release of N_2O through nitrification, denitrification, nitrifier denitrification, and/or coupled nitrification-denitrification (Firestone et al., 1980; Abbasi and Adams, 2000; Khalil et al., 2004).

While the determination of specific mechanisms is often difficult, some indication of the mechanisms can be determined based on soil nutrient concentrations and changes in concentrations over time. For example, the increase in soil nitrate concentrations, coupled with a decrease in ammonium concentrations, are indicative of the occurrence of nitrification (conversion of ammonium to nitrate). Likewise, a decrease in nitrate concentrations over time is indicative of denitrification (conversion of nitrate to N_2 gas). As further explained below, this logic has led to my understanding

that most (but not necessarily all) of the nitrous oxide produced during this experiment was from the nitrification process.

In well drained arable soils, injection is preferred to reduce NH_3 volatilization as it reduces NH_3 and has a minimal effect on N_2O . Webb et al., (2010) found that techniques that tend to reduce NH_3 emissions were expected to increase N_2O emissions on the grounds, that there is more allowable N entering the soil via either incorporation or surface injection. VanderZaag et al., (2011) reviewed several studies finding that N_2O fluxes were higher from injecting cattle slurry compared to surface application of the same slurry. Injection did lower NH_3 emissions further showing the tradeoff between application practices and reducing NH_3 emissions while increasing N_2O emissions. While results from this experiment agree with prior literature to a certain extent, the prolonged biological activity and emission rate of the urea treatment on the second irrigation event is an observed anomaly that deviates from the initial irrigation event.

When compared to the surface manure treatment, the urea treatment produced 3.5 times more nitrous oxide on average while both treatments had the same initial total nitrogen content. The urea hydrolysis reaction seems to be the main driver behind this difference in nitrous oxide emissions. This reaction happens almost instantly as water is added to the sample jars and, as described in Eq 8, 9, 10. The biological conversion of urea to ammonia (Eq, 11 and 12) is rather spontaneous, in the presence of the urease enzyme. Sigurdarson et al. (2018) describe the urease-catalyzed hydrolysis process with a half-life of only 20 milliseconds at 25°C . The net reaction in an aqueous solution results

in an increase in pH (Eq. 13 and 14). This increase in pH is a catalyst for a simultaneous increase in ammonia concentrations which is detailed in Figure 19. Now that there is rapid turnover of urea to readily available ammonia in an oxygen rich environment nitrification can take place thus driving the reaction to produce nitrate and nitrite (Eq. 2, 3, 4). This conclusion can be drawn from the final concentrations shown in Figure 17, which shows that the U treatment had the highest ending soil nitrate+nitrite-nitrogen concentrations. Under anaerobic conditions, nitrate and nitrite products sometimes proceed to denitrification, where additional nitrous oxide can be leaked as an intermediate gas through the chemical process (Eq. 5), which can contribute to higher N_2O emissions. However, since the urea treatment had the highest nitrate concentration at the end of the experiment, it is improbable that a lot of denitrification occurred in the urea treatment.

Another explanation for the higher emission rates in the urea treatments is that the urea prills used in this study were pure organic nitrogen, and this allowed microbes non-competitive accessibility to the nitrogen within the urea. Whereas, nitrogen access within the manure could have been hindered by the highly stable organic matter (humic and fulvic acids) and other microbes present in the manure. Figure 18 supports this via the representation of the final organic nitrogen concentrations. Although all of the initial N in the urea treatment was in the organic form, the final organic N concentration in the soil/manure was less in the urea treatment than in either the MS or MI treatments. Thus, it is evident that much of the organic N in the urea treatment was converted to ammonia (non-organic) form and eventually nitrate.

The urea treatment concentrations were lower than the incorporated or surface applied treatments. This supports the idea that microbes present within the EU's were able to access the nitrogen within the urea more readily than the nitrogen present within the other treatments. Figure 20 also supports this idea, the total nitrogen concentrations for the urea sample were lower as well which is an indication that the nitrogen within those treatments was more readily available. While being more readily available or useable the nitrogen within the urea samples is used through other channels within the nitrogen cycle. Therefore, a decrease in total nitrogen and organic nitrogen concentrations is observed and increased nitrous oxide emissions are observed.

The combination of the results presented in this study seem to agree with prior literature in that organic manure incorporation does reduce ammonia volatilization while increasing nitrous oxide emissions. However, this study also presents results that are contradictory to previous hypotheses regarding surface application practices, where typically surface application results in higher ammonia volatilization rates and lower nitrous oxide emissions.

CHAPTER VI

RECOMMENDATIONS FOR FUTURE RESEARCH

Field studies are needed to confirm the effects of irrigation events upon nitrous oxide emission and ammonia volatilization and to verify my results from the standardized controlled lab setting. With such large spatial variability present in field studies, conducting similar emission research to the standardized lab setting can act as a confirmation of prior results, if nothing else.

Another future experiment is recommended to further increase our knowledge concerning mechanisms of nitrous oxide production from urea and manure. This could be done by comparing a 50/50 blend of manure and urea with similar application practices to determine if emissions were inhibited by access to nitrogen stores. Another experiment could be conducted using heat to sterilize the manure of all microbes present, and determining emission rates to determine whether emission rates are tied to nitrogen accessibility, or if the microbes within the manure play a competitive role in nitrogen accessibility. In conjunction with the previously mentioned studies an incorporated urea study could also be helpful in determining the effect of nitrogen accessibility on emissions.

CHAPTER VII

CONCLUSIONS

The following conclusions were drawn from this research:

1. The urea treatment had the highest cumulative nitrous oxide emissions, followed by incorporated manure, surface applied manure, and soil only. While incorporation of manure is a best management practice for reducing ammonia emissions, it causes an increase in nitrous oxide emissions as compared to surface-applied manure.
2. There was an increase in nitrous oxide emission rate immediately follow a simulated irrigation event.
3. Based on changes in soil nitrate and organic nitrogen concentrations, it was concluded that nitrous oxide emissions were primarily due to nitrification (the conversion of ammonia to nitrate).

CHAPTER VIII

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CHAPTER IX

APPENDICES

Appendix A	Microsoft Excel Spreadsheet: 74MB (see attached electronic file)
Appendix B	Statistical Files: SAS Input and Output Files (see attached electronic files)
Appendix C	Servi-Tech Laboratories Soil and Manure Analysis Results