IMPROVING THE UTILITY OF PRECISION AGRICULTURE THROUGH MACHINE LEARNING AND CLIMATE-SMART PRACTICES

By

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THESIS ACCEPTANCE PAGE

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This thesis is approved as a creditable and independent investigation by a candidate for the master's degree and is acceptable for meeting the thesis requirements for this degree. Acceptance of this does not imply that the conclusions reached by the candidate are necessarily the conclusions of the major department.

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CONTENTS

ABBREVIATIONS

AgMIP	Agriculture Model Intercomparison and Improvement Project
ANOVA	Analysis of variance
AI	Artificial Intelligence
°C	Degrees in Centigrade
CO_2	Carbon Dioxide
CO_2 -C	Carbon Dioxide-Carbon
DAWN	Dashboard for Agricultural Water use and Nutrient Management
DNN	Deep Neural Networks
DSS	Decision Support System
FAO	Food and Agriculture Organization
FRST	Fertilizer Recommendation Support Tool
FICO	Fair Isaac and Company
GHG	Greenhouse Gas
GMO	Genetically Modified Organism
IPCC	Intergovernmental Panel on Climate Change
ML	Machine Learning
N_2O	Nitrous Oxide
N_2O-N	Nitrous Oxide-Nitrogen
NH_3	Ammonia
NH_3^--N	Ammonia-Nitrogen
NH_4^+	Ammonium
NH_4^+-N	Ammonium-Nitrogen
NO_3^-	Nitrate
NO_3^--N	Nitrate-Nitrogen
Picarro	Picarro Cavity Ringdown Spectrometer
PLFA	Phospholipid Fatty Acid
ppm	Parts Per Million
PA	Precision Agriculture
PVC	Polyvinyl Chloride
P value	Probability of Significance

UAV	Unmanned Aerial Vehicle

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ABSTRACT

IMPROVING THE UTILITY OF PRECISION AGRICULTURE
THROUGH MACHINE LEARNING AND CLIMATE SMART PRACTICES

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Climate Smart Practices are management strategies that focus on increasing soil and crop productivity, utilize site-specific strategies to increase resiliency against the effects of climate change, and mitigate these negative effects by reducing greenhouse gas (GHG) emissions. Decision Support Systems (DSSs) using machine learning (ML) can adjust models based on new information and help farmers make climate smart decisions within their operation. The 4R nutrient management model of right source, rate, location, and time also demonstrates a framework that may be considered climate smart by improving soil and crop productivity. However, when initially conceptualized, the 4R model did not consider GHG emissions. Additionally, the long-term adoption of DSSs has been low in agriculture, reducing the ability of farmers to collect and analyze farm data to the fullest. Therefore, the objective of the first chapter is to examine applications of, and barriers to, DSSs in precision agriculture (PA). The objective of the second chapter evaluates the 4R model to determine the impact of GHG emissions when utilizing near continuous chambers over a two-year period. The GHG emissions were measured by analyzing nitrous oxide and carbon dioxide emissions from a 50/50 split application of 157 kg N/ha that was applied to corn (*Zea mays*) at pre-emergence and V6 compared to a single application at pre-emergence 157 kg N/ha in a two-year replicated study. Results from the first chapter identify the barriers preventing farmers from using DSSs as well as suggesting solutions to these challenges. Results from the second chapter indicate that the

split application can reduce carbon dioxide and carbon equivalent emissions and therefore, may be a useful framework for DSSs to follow in achieving Climate Smart Practices.

STATEMENT OF THE PROBLEM

Climate Smart Practices are management strategies within the framework of Climate Smart Agriculture. These strategies focus on three pillars: 1) increase soil and crop productivity, 2) site-specific techniques that increase ecosystem resiliency, and 3) reduce greenhouse gas (GHG) emissions. Agriculture is a large producer of GHG emissions that include nitrous oxide (N_2O) and carbon dioxide (CO_2). For example, while on average less than 1% of the Nitrogen fertilizer that has been applied to corn is lost as N_2O, 60% to 75% of total anthropogenic loss of this gas comes from agriculture (Cavigelli et al., 2012; Syakila & Kroeze, 2011). Nitrous oxide is commonly emitted through denitrification, and nitrification, and because this gas absorbs infrared radiation at a rate approximately 298 times greater than CO_2, N_2O is a serious threat to the productivity of global agriculture (Weil & Brady, 2017; DeKlein et al., 2006; EPA, 2023). To balance environmental and economic concerns the 4R management model was developed in the early 2000s (Fixen, 2020). The goals of 4R management were to create locally based strategies that maximized efficiency while minimizing inputs and to produce healthier more productive soils (Fixen, 2020). The four R's: right time, right rate, right place, and right source, set a framework that allows for site-specific flexibility in reducing environmental impact and promoting more productive soils.

Climate smart goals can be implemented most effectively when frameworks such as the 4R framework are combined with Precision Agriculture (PA) technologies such as Decision Support Systems (DSSs). This has the potential to consistently, and at a large scale, increase yields while simultaneously reducing resource use and the impact of agriculture on the environment (Pretty, 1997; Petersen et al., 2015; Joshi et al., 2022; Hamrani et al., 2020). However, barriers to the widespread use of this technology exist including financial constraints of small farms, mistrust of DSSs, concerns about the privacy of farm data, and a lack of a specialized/ trained work force. Through acknowledging and addressing these challenges agriculture can work towards achieving climate smart goals.

Additionally, through new data management and measurement technologies, researchers are provided with the unprecedented opportunity to collect and evaluate a huge amount of environmental data. This opportunity allows researchers to re-test previously held assumptions. For example, much of what is known about GHG emissions from agriculture was produced through point measurements of data that were collected through static chambers. These points were averaged over the growing season to estimate total emissions. Studies using these techniques provided a baseline for policy by shaping how government agencies conceptualized agriculture's role in climate change (Venterea et al., 2015; Maharjan et al., 2013; Collier et al., 2014; DeKlein et al., 2006; EPA, 2023). However, new technology such as near continuous gas measurement systems, like the LI-COR LI-8100-104 long-term opaque chambers (8100-104 LI-COR, Lincoln, NE), can measure emissions nearly continuously and therefore significantly increase the accuracy of the data by measuring the area under the curves produced. This increased accuracy and new data collection system has revealed new information about GHG emissions and can also be used to re-test previously held assumptions about management strategies (Joshi et al., 2022; Thies et al., 2020; Reicks et al., 2021; Fiedler et al., 2021).

Through re-testing with more accurate PA technologies and DSS tools, researchers and extension specialists may begin to reconsider some management strategies. For example, in the 4R management model the right timing of nitrogen fertilizer is often associated with a split nitrogen application. This management strategy has been shown to reduce leaching and denitrification loss by applying fertilizer when it is needed (Graham et al., 2016; DeBruin & Butzen, 2014). In fact, this strategy is so ingrained as an environmentally beneficial strategy it is encouraged through crop insurance programs such as Post-Application Coverage Endorsement program in the U.S. Department of Agriculture's Risk Management Agency (USDA, 2021). In addition, it is an effective strategy to reduce nitrate leaching and denitrification losses in soils. However, weather conditions and soil parameters have been observed as significant variables impacting the success of split nitrogen application (Clark et al., 2020; Butzen, 2011). Additionally, split applications of nitrogen have not demonstrated significant reduction in N_2O emissions. In fact, research suggests that it can increase these emissions (Venterea and Coulter, 2015).

As researchers and farmers begin to collect data more accurately and use DSS tools for analysis and retesting we believe that accurate site-specific algorithms will be created (Gardezi et al., 2022). However, these algorithms will not be implemented without first reducing the adoption barriers. These barriers are systematic and social challenges that prevent farmers from collecting and utilizing data to make actionable decisions (Lindblom, 2017). By examining the adoption barriers that farmers face in using new technologies such as DSSs, researchers and stakeholders can work towards an ethical and equitable future to use PA and achieve the three pillars of Climate Smart

Practices that include increasing soil and crop productivity, increasing agriculture ecosystem resiliency, and reducing GHG emissions where possible.

Therefore, the objective of this thesis is to: 1) examine adoption barriers to long-term adoption of DSS tools that utilize ML and AI to move PA responsibly towards climate smart goals; and 2) demonstrate through example how this technology can advance PA towards a climate smart future by re-examining the 4R management model for its ability to reduce GHG emissions. The objective of the field experiment conducted in the second chapter was to reassess previous conclusions about nitrogen fertilizer timing concerning N_2O-N, CO_2-C and CO_{2e} emissions using a 50/50 split urea 157 kg N/ha application compared to a single urea application 157 kg N/ha at pre-emergence.

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CHAPTER I

IMPROVING DECISION SUPPORT SYSTEMS WITH MACHINE LEARNING: IDENTIFYING BARRIERS TO ADOPTION

ABSTRACT

Precision agriculture (PA) has been defined as a "*management strategy that gathers, processes and analyzes temporal, spatial and individual data and combines it with other information to support management decisions according to estimated variability for improved resource use efficiency, productivity, quality, profitability and sustainability of agricultural production.*" This definition suggests that because PA should simultaneously increase food production and reduce the environmental footprint, the barriers to adoption of PA should be explored. These barriers include: 1) the financial constraints associated with adopting DSS, 2) the hesitancy of farmers to change from their trusted advisor to a computer program often behaves as a black box, 3) questions about data ownership and privacy, and 4) the lack of a trained workforce to provide the necessary training to implement DSSs on individual farms. This paper also discusses the lessons learned from successful and unsuccessful efforts to implement DSSs, the importance of communication with end-users during DSS development, and potential career opportunities that DSSs are creating in PA.

Abbreviations: ML, machine learning; AI artificial intelligence; DSS decision support system; ICT information communication technology; UAV unmanned aerial vehicles; PA Precision agriculture; GMO, genetically modified organism.

1 INTRODUCTION

1.0 What is a Decision Support System (DSS)?

Decision Support Systems (DSSs) are models that use information and communication technologies (ICTs) for complex decision-making (Manos et al., 2004). The models embedded in DSSs take data stored in the database to produce a user-specific result (Alenljung, 2008; Manos et al., 2004). These models can be created using many techniques including process based, empirical models, and/or machine learning (ML)/artificial intelligence (AI) techniques. In agriculture, process-based models are generally mathematical representations of biogeochemical and physical systems; empirical models are statistical models based on observations among variables; and ML models make predictions based on patterns learned from the data set. A difference between process, empirical models, and ML/AI models is that ML/AI models learn as new input data are added, which makes them uniquely suited for PA.

However, as with all models, errors exist when the underlying understanding of the processes or model assumptions are incorrect. For example, a very common assumption is that weeds reduce yields by competing with the crop for water, nutrients, light, space, and carbon dioxide. However, current research, made possible by the ability to decipher the crop genomes, has shown that weeds can reduce yields regardless of resource availability. This finding is based on the ability to quantify transcriptomic changes (e.g. down-regulation of photosynthesis, root growth reductions) that are induced by weed presence, long before direct competition occurs (Clay et al., 2009; Horvath et al., 2023). Models, based on incorrect assumptions, may provide acceptable recommendations under some conditions but flawed recommendations in others.

2.0 Why are DSSs needed?

Agriculture is facing immense challenges, such as reducing greenhouse gas emissions and topsoil loss due to erosion, while increasing yields in an unstable climate (United Nations, 2022). The failure to manage these challenges can result in societal and environmental upheaval. As examples, the French revolution (1789-1799) has been linked to the Little Ice Age, when crops failed in cold climates, which led to starvation and societal instability (Ljungqvist et al., 2021). The Middle Eastern Arab Spring in 2011 has also been linked to climate change with low crop yields and increased food prices, which again led to social upheaval (Zurayk, 2016). To reduce the risks of future agricultural failures, sustainable intensification needs to be implemented to optimize production and simultaneously decrease agriculture's environmental impact (Lindblom et al., 2017). Sustainable intensification requires enhanced management that can be delivered in part using DSSs (Lindblom et al., 2017).

3.0. Low adoption of DSSs

While some components of Precision Agriculture (PA), such as autosteer and yield monitors, were quickly adopted, to date variable rate fertilizers, another PA tool, has not been widely adopted (Rossi et al., 2013; Winter, 2018; Baumeister et al., 2015; Lindblom et al., 2017, McCown et al., 2002). The discrepancy between the adoption of some PA tools and not others suggests that there are barriers to long term use of PA tools. Many barriers have been suggested and may include financial constraints, a hesitancy to change from a known trusted decision processes to an unknown decision system, uncertainty of data ownership and privacy, and workforce availability (USDA 2022; Mitchell et al., 2021; Erickson et al., 2018).

In addition to these barriers, it is important to understand that fully integrating data collection, processing, and implementing is difficult, and requires diverse knowledge, skills, and abilities. For example, processing remote sensing data into useful information may require a human pilot to collect the data, a geographic information specialist to create a map, an agronomist to confirm yield limiting factor(s), machines that can perform these tasks, knowledgeable applicators to apply the treatments at the right time, place, and rate, and follow-up with economists and agronomists to determine if savings, yield, or other tangible outcomes have occurred. It is important to point out that currently each step requires time, and that skipping steps that often require human involvement can reduce the value of the information (Priya and Ramesh, 2020).

4.0 Barriers to adoption of DSSs

4.1. Financial constraints

Agricultural retailers have been surveyed about producer attitudes at least every other year since the mid-1990s (Erickson & Lowenberg-Deboer, 2022). These surveys provide insights into the adoption barriers. In 2021 about 37% of the dealers reported that economic limitations was one of the most important barriers. However, this barrier fluctuates with crop prices and is reduced when prices are high and increased when prices are low. For greater use of these technologies, uncertainties must be balanced with an increased return on investment (Baldin et al., 2021; Rinaldi et al., 2014).

4.2. Financial constraints of small farms

Farm size also influences the amount of capital that can be invested in DSSs and PA tools that can be used by DSSs (Baldin et al., 2021; Akaka et al., 2021). In general, large farms have more capital and manpower to test and implement new practices (Akaka et al., 2021). A good example of this barrier was discussed by Ashworth et al. (2018), where partial budgeting determined that the break-even farm size for auto-guidance was between 10 to 50 ha. The implication of this analysis is that the cost of the equipment per hectare decreases with increasing farm size. For example, if the equipment costs $100,000 and the farm size is 100 ha, then the cost/ha is $1,000. However, if the farm size is 1000 ha, then the cost/ha is $100. This difference in cost/ha may result in lower adoption PA adoption rates on small farms (Denmark Statistics, 2022; Thompson et al., 2021). The difference in price/ha between small and large farms results in small farms taking longer to return a profit than large farms. Another barrier is that DSS recommendations do not provide protection from uncertainty (Ara et al., 2021). Uncertainty results from any given treatment having a chance that it may or may not be effective.

Solving cost differences between small and large farms (financial equity) is a complex problem, but solutions have been proposed to overcome this adoption barrier. One potential solution is for DSS designers and manufacturers to provide free or inexpensive trial periods or monthly subscriptions to DSS services. A free trial period would allow a farmer to see if the DSS fits their operation and provides useful actionable recommendations (Akaka et al., 2021). Small farms could work together to spread technology costs over many producers. In addition, university extension services could provide training that reduces the investment costs or federal agencies and/or industry could provide financial incentives to use DSSs (McFadden et al., 2023).

4.3. Farmers hesitancy to change the decision process

On many farms, agricultural decisions result from one-on-one discussions between the producer and their trusted advisor. Many producers do not feel comfortable replacing the trusted advisor with a DSS (Gardezi et al., 2022). This discomfort or lack of trust may be attributed to poor communication between the users and DSS developers, who may or may not have agriculture experience. This often manifests as recommendations that are acceptable to a developer but not considered acceptable or actionable by the end-user. For example, if the recommendation is to apply water, but the field is not, or cannot be, irrigated, the recommendation is useless. The difference between a recommendation and what is possible, is called the paradox of acceptability (Hochman et al., 1994).

Trust can also be lost due to miscommunication between the development team and the end-user. One form of miscommunication is the selection of an DSS tool that assists in short-term planning (tactical) when a tool for long-term (strategic) planning is required (Ara et al., 2021). A DSS could combine strategic and tactical systems, which would provide the initial short-term decision and aide in long term strategic planning (Ara et al., 2021).

To improve trust in DSSs, producers and their advisors should carefully consider the benefits and deficiencies of the various options before purchase. Similarly, the marketing of a DSS as tactical, strategic, or both, needs to be made clear to the end-user. The failure to select an appropriate system can result in a general distrust of PA (Lindblom et al., 2017; Rossi et al., 2013; Ara et al., 2021). A milestone is reached when a farmer decides to trust a DSS recommendation because each decision has economic implications that can be devastating. For example, a farmer may not have the income to make land payments, purchase farm inputs for the next cropping season, and/or cover their personal expenses. These financial concerns, especially when combined with distrust, can result in farmers deciding to keep a trusted decision process that has worked reasonably well in the past (McCown et al., 2002). However, trust in DSSs can be cultivated by providing demonstrations, training, and clear examples on how to successfully integrate the technology into their operations (Akaka et al., 2021).

4.4. Questions over data ownership and privacy

Many producers and lawmakers are concerned about data ownership and privacy of farm data (Erickson & Lowenberg-DeBoer, 2022). Examples of on-farm data include current and historical yield, seeding rates, applied soil nutrient rates, and remote sensing (Ellixson, 2022). When these data are combined with public information through models connected to “the cloud” or the internet, the models may produce sensitive information. Unlike a bushel of corn that is tangible, data are intangible, easily transmitted long distances, stored in “the cloud”, and can be subject to security breaches. Additionally, farm data ownership is legally difficult to protect because it is considered both unrivaled, meaning one person’s access does not prevent another’s, and because of its uncertain excludability (the right of the owner to deny another’s access) (Goeringer, 2016; Jouanjean, 2020; Kaur et al., 2022).

Farm data ownership is akin to a FICO credit score. FICO collects data on an individual and calculates their score. Although FICO does not own the data, it does own the credit score because it is newly generated data (Goeringer, 2016). It is currently legally, and ethically ambiguous, whether a third-party technology provider has the right to generate new data from technology owned by individual farmers, and then sell the "new" data back to them after it has been combined with data from the same geographical region (Jouanjean, 2020). This problem is further confounded by efforts to make programs and data collected by 'smart machinery' freely available as "open source" (Rinaldi et al., 2014) to other companies, which then take the free data and profit by selling it back to farmers in DSS technologies.

Laws are often slow to catch up with technology, and currently laws that protect farmers and their data are limited (Ellixson, 2022; Kaur et al., 2022; Goeringer, 2016). Moreover, the term "ownership" itself is only the tip of the iceberg. Legally speaking, "ownership" is a relatively weak protection for farm data and does not necessarily mean the kind of control that farmers seek. Conditions and rights are often specified separately in individual contracts; therefore, there is no such thing as an all-inclusive data ownership law (Jouanjean, 2020). Determining where to draw the line is a challenge that law makers and stakeholders must define together, highlighting many new career opportunities for those interested in the legal protection of farm data as well as opportunities to bridge the gap between companies and farmers through legal communication and advocacy of farmer's data rights.

4.5 Limited trained workforce

A 2015 survey asked agriculture retailers about their minimum education requirements for careers in PA such as equipment operator, agronomist, equipment technician, technical support, and PA specialist (Erickson et al., 2018; Fausti et al., 2021). Results at that time revealed that most of the PA workforce met or exceeded the education expectations of employers by completing either a 2-yr associate degree or a 4-

year bachelor's degree. However, employers indicated difficulty in locating and recruiting qualified applicants. Additionally, necessary skills, such as data intensive thinking, the ability to understand statistical standards to produce means and standard deviations, as well as their ability to install, calibrate, troubleshoot, and repair PA hardware and equipment were lacking (Erickson et al., 2018; Fausti et al., 2021). Further analysis suggested that there was a mismatch between the training received and the training required to proficiently perform the job.

To help PA employees meet the job expectations, professional societies and higher education institutions must commit to curricula that will close this gap. For example, the American Society of Agronomy and Ag*IDEA have created PA certificate programs (Erickson et al., 2018). Those trained in both the technical and human dimension of DSSs can act as important communicators between program developers and the end-users (Lundström & Lindblom et al., 2018). Additionally, communicators between education institutions and industry professionals, such as retail dealerships, can help to align academic programs for students pursuing PA positions with the qualifications required by industry professionals. This will aid in creating employees proficient in the knowledge, skills, and abilities in math and statistical skills required by the PA industry (Fausti et al., 2021).

5.0. Lessons learned

While there are a range of barriers slowing DSS adoption, it is also important to evaluate why some succeed (e.g. Vite.net and Pigs2Win) and others fail (e.g. FEEDMAN). Vite.net® has been adopted successfully in both small and large European vineyards since 2013 (Rossi et al., 2013). Vite.net uses real-time data from sensors placed around a vineyard to produce recommendations for pest (disease, weed) control, fertilization, and irrigation management decisions that have increased the overall vineyard productivity (Lindblom et al., 2017; Rossi et al., 2013). For example, in 2016, organic producers who used Vite.net saved about €195 ($205) per hectare (ha) relative to their usual practices. The savings were attributed to the DSS making site-specific recommendations about the application of non-organic herbicides, pesticides, and fertilizers. Specifically, growers using Vite.net reduced copper application by 37% when compared to producers who did not use the DSS (TpOrganics, 2016).

Along with observable savings, Vite.net designers focused on communication with end-users with feedback throughout development. Vite.net also focused on specific vineyard problems that were identified by vineyard managers. The communication between developers and users was deliberate and allowed the developers to understand how to best convey and make recommendations through the system's user interface. This communication/training had value because it improved end-user trust.

Another example of a successful DSS is Pigs2win, which improves swine production and reduces environmental impact (smell and nutrient losses) of the operation (Lindblom et al., 2017; Meensel et al., 2012). These decisions were traditionally made using key performance indicators such as productivity costs, labor income, and feed conversion (kg of feed per kg of live weight). However, the Pigs2Win development team worked closely with the farmers and stakeholders to ensure that results are aligned with the expectations of the end-user. Objectives were defined by the development team and farmers and stakeholders identified how an objective could be met by using a DSS. For example, Pigs2Win was built using Excel® as a framework because it was easily accessed and understood by end-users. Several prototypes were presented, and farmers were encouraged to provide feedback and familiarize themselves with the system. A benefit of this open communication process was that it built trust by demonstrating transparency (Meensel et al., 2012).

FEEDMAN, a feed to dollars beef management package, in contrast to Vite.net and Pigs2Win, was an unsuccessful DSS. FEEDMAN was designed in 1998 to help farmers and farm managers make strategic and tactical decisions about feeding options, animal performance, market options, and economic decisions for livestock (Newman et al., 2000). While the development team understood the need to make the system user-friendly, users found it to be easy to use but not useful. Many users indicated that they were reluctant to take recommendations from DSS that did not provide clear benefits. Users also cited abandoning the program due to a lack of maintenance. These issues likely could have been avoided by communicating with the end-users throughout the process such as in the case of Vite.net and Pigs2Win. Potential users and stakeholders were brought into the process at the end of FEEDMAN development, instead of being allowed to shape the DSS in the process.

Vite.net, Pigs2Win, and FEEDMAN are different DSSs, however, communication is a common thread linking their successes and failures. Vite.net and Pigs2Win worked with the end-users throughout the process to ensure that the software would provide real value, whereas FEEDMAN was marketed as a complete system that did not provide real value to the users. Additionally, Pigs2Win took steps to appeal to smaller farms by ensuring that the users did not need to purchase additional software, building part of the system in Excel® to reduce the financial burden. While this represents only a few of the many examples of successful and not successful DSSs, it is important to learn from what has and has not worked.

6.0. The Future of DSSs and the importance of improved communication

Decision tools are being developed to provide more accurate recommendations to farmers. Future DSSs that are empowered by ML algorithms will have the capacity to collect and process enormous amounts of site-specific information (Priya and Ramesh, 2020). However, because research scientists tend to work very narrowly and silo themselves, our ability to address “big questions” may take a long time. Dr. Cynthia Rosenzweig, the 2022 World Food Prize winner, addressed the scope of these problems and stated, “Climate change is so challenging: We must solve it; but no one group or discipline or sector of society is going to solve it on their own. -- Such ‘silos’ do not work for finding solutions to climate change” (Coyne, 2022).

To improve communication among scientists, Dr. Rosenzweig created a program called AgMIP (Agriculture Model Intercomparison and Improvement Project). AgMIP brings together an interdisciplinary and international team of stakeholders and experts to estimate and predict how climate change will produce new risks and vulnerabilities in global agriculture, while also providing risk mitigation and adaptation suggestions (AgMIP, 2022). The model’s framework focuses on four dimensions: adaptation, mitigation, food security, and agriculture policy (Rosenzweig et al., 2017). The user interface provides this information at three different levels of expertise. The first level is the regional summary provided and demonstrated using individual stories about farmers who have benefited. The second level is the spatial dashboard using maps and data to make comparisons among various regions This approach was designed in response to the

United Nations (United Nations Framework Convention on Climate Change) request for information about, and implications of, constraining the global temperature increase to 1.5℃ and 2.0℃. While there is still much uncertainty in how much the climate will change, the model identified vulnerabilities and uncertainties in managing the future risk to agriculture (Rosenzweig et al., 2017).

Another agriculture DSS that stepped out of its silo and into a broad regional system is DAWN (Dashboard for Agricultural Water use and Nutrient management). DAWN is supported by the USDA through NIFA's Agriculture and Food Research Initiative, with the goal of informing row crop producers about water and nutrient management decisions. The system couples a crop growth model with existing regional climate systems and links them with data about land and water use, agroecology, hydrology economics and human intervention. The DSS then uses these models to help producers with field level decisions (DAWN, 2022). This system, produced for both farmer and researcher use, can run scenarios about specific problems and predict outcomes based on soil maps, historical climate data, estimated crop yields, and more. These scenarios look to optimize economic return and minimize environmental impacts. From these various predictions, the producer can choose the most preferred scenario. This tool can help producers explore, risk free, their options for different crops, and various irrigation strategies. Even without an irrigation system, this DSS can be helpful by providing land and yield information that relate to potential water quantity and quality needs such as evapotranspiration rates, average precipitation and air temperature, soil moisture, crop yield potential, nutrient loss, runoff, and drainage potential.

The user interface of DAWN is also being co-developed by stakeholders to ensure that the intended users/communities will have their needs met. DAWN will do this by providing information through both text and graphics, allowing users to explore options and strategies easily and quickly, collect statistics for the user about location and crop type, evaluate the producers' needs, and finally provide education materials such as extension publications and research summaries to keep users informed on the decisions they make. This new and exciting DSS will provide insights into new strategies for sustainable agriculture as well as opportunities for graduate students and early career researchers to work on creating/improving farm management.

FRST (the Fertilizer Recommendation Support Tool) also seeks to take DSSs to a broader scale by being a USDA operated, national fertilizer recommendation database and tool in the United States. This project takes inspiration from a national soil recommendation project produced in Australia in 2013 called The Australian Better Fertilizer Decisions for Cropping Systems Project (Conyers et al., 2013). FRST is a calibration and correlation study currently focused on potassium and phosphorus but will expand into nitrogen and micronutrient recommendations (Lyons et al., 2020). This collaborative project includes over 30 land grant universities, USDA-ARS and USDA-NRCS and aims to address the inconsistent information on fertilizer recommendation across state lines based on local site characteristics rather than state boundaries (McCauley, 2020). The dataset is being populated with historic data from 29 states that meet appropriate criteria and includes crop yield, grain moisture, rainfall, air and soil temperatures, seasonal stresses, and the production system used (Speirs et al., 2013). The web-based, user-friendly platform allows users to select input variables such as soil test methods, geographic locations, yield levels, and crop types to assist in recommendation decisions. This tool is expected to be continuously updated and hosted on a neutral internet space with common access and author attribution for as long as possible. The project leaders are encouraging more collaborative research on soil fertility among researchers and provide new career opportunities for graduate students to work with large historical, as well as current, datasets to produce transparent useful information to producers. We believe that programs such as AgMIP, DAWN, and FRST can help graduate students and professionals cultivate career opportunities, expand their horizons, see how their research can help answer the "big" questions, and move out of their narrow research path.

7.0. CONCLUSION

In this unprecedented era of environmental crisis and high-tech digital agriculture, which can collect information at high spatial and temporal resolutions, DSS product developers, researchers, and end-users need to collaborate and broaden their scope to solve critical issues facing agriculture at multiple scales. When implemented, we believe that DSSs are powerful tools that can help humans make connections in large datasets and find patterns that are crucial to making more sustainable and resilient crop production decisions. While DSS tools demonstrate an exciting future, it is important to recognize five aspects that have implications for PA adoption.

First, while many producers want to increase sustainability of their production, it is difficult to be cognizant of all factors that create the most profitable and sustainable outcomes. Second, the replacement of process or empirical models with machine learning algorithms within a DSS has the potential to improve accuracy and reduce uncertainty.

Third, policy makers must continue to reduce the financial burden of acquiring the technology through strengthening programs that provide financial incentives to smaller farms to adopt DSSs. Fourth, industry professionals need to increase opportunities for farmers to engage with technology to build confidence in appropriate DSS products while lawmakers must begin to address serious questions about data equity such as the legal definition of data ownership.

Fifth, communication barriers between DSS designers and stakeholders also still need to be broken down, and additionally, appropriate training must be provided at higher education levels for those pursuing careers in PA. Once these hurdles have been overcome, DSSs can work as tools to aid farmers in making decisions that will fully allow us to meet both the current and future agronomic needs for food, feed, fuel, and fiber in a sustainable manner.

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CHAPTER II
DOES SPLITTING THE NITROGEN RATE REDUCE CARBON EQUIVALENTS?

ABSTRACT

Climate smart practices are site-specific approaches focused on combating the challenges of climate change through sustainably increasing agricultural production, employing site-specific strategies to increase resiliency, and utilizing strategies to reduce greenhouse gas (GHG) emissions. Climate smart practices can work within management frameworks such as the 4R nutrient management model; an approach that encourages farmers to apply the right fertilizer source at the right rate at the right location and at the right time. However, the 4R model was not specifically designed to be a climate smart practice. The goal of this paper is to partially fill this gap. Therefore, the objective was to determine if splitting half the urea application between pre-emergence (VE) and half between corn (*Zea mays*) V6 growth stage could be considered a climate smart practice. This replicated two-year study contained three N treatments including 0, 157 split between pre-emergence and V6, and a single application of 157 kg N/ha at pre-emergence. The GHGs were determined using a near continuous sampling and measurement system; LI-COR LI-8100-104 long-term opaque chambers. Across the years and treatment interaction, splitting the N fertilizer compared to the pre-emergence application did not impact N_2O-N emissions, however, CO_2-C and CO_{2e} was reduced. Results indicated that the split application should be considered as a climate smart practice.

INTRODUCTION

Climate smart practices are approaches to agriculture that transform and reorient agriculture systems to deal with the effects of climate change. These practices do this through focusing on the three pillars: increasing crop and soil productivity, improving soil resiliency through management adaptation, and reducing greenhouse gas emissions (GHG) where possible (Campbell, 2017). Because management, soils, genetics, and climate impact an agricultural system's ability to achieve these goals, specific practices can vary from location to location. For example, at sites subject to drought, the climate smart practices may focus on maintaining soil moisture through reduced tillage or leaving residue on the field. However, at another location subject to flooding, the climate smart practices used may include a change in crop type, creating grass waterways, or building vegetated riparian buffers.

Agricultural systems in the U.S and globally are extremely variable, these climate smart strategies seek to work within the framework of sustainable agriculture with a strong focus on the climate change dimensions of agriculture. Therefore, these strategies can be used within the 4R framework model. The conceptual basis for 4R nutrient management is to apply the right source of fertilizer, at the right rate, at the right time, and at the right location to optimize plant productivity while minimizing the impact on the environment (Fixen, 2020). Adopting 4R nutrient management can involve many different strategies, including the use of soil sampling to determine residual soil N, splitting the fertilizer rate, and using urease and nitrification inhibitors to reduce N losses to the atmosphere and groundwater (Clay et al., 1990a; Venterea et al., 2016). Research suggests that appropriate 4R nutrient management strategies are dependent on crops, soil, and environmental conditions. However, the impacts on greenhouse gas (GHG) emissions were not initially considered upon conception of the 4R nutrient management strategies. A 4R strategy that has the potential to be widely adopted is splitting the N rate. However, splitting the N application or delaying the application of the N fertilizer to match plant uptake requirements may increase GHG emissions (Thies et al., 2020; Venterea and Coulter, 2015). Thies et al. (2020) reported that applying urea in the fall after soil temperatures had decreased to less than 10℃ minimized Nitrous oxide (N_2O) emissions, whereas delaying the fertilizer from spring to early summer increased fertilizer derived N_2O emissions over the 21 days following the application. In studies conducted in Minnesota, Venterea and Coulter, (2015) and Venterea et al. (2016) reported that when N was applied as a single application at planting when compared to a split application, splitting the application either increased or had no impact on N_2O emissions over the entire season. Differences in emissions were partially attributed to climate and moisture variability. Venterea and Coulter (2015) suggested that a prolonged dry period before the split application followed by a large rainfall event resulted in higher N_2O emissions from the split application compared to the single application. Additionally, Venterea and Coulter (2015) and Venterea et al. (2016) indicated that years with wetter soils are more likely to produce higher levels of N_2O. This may be due to the increase in microbial activity and respiration in wetter soil compared to dry soil (Bogati & Walczak, 2022). However, Venterea and Coulter, (2015) and Venterea et al. (2016) did not report the carbon dioxide (CO_2), or carbon equivalents (CO_{2e}) and they used a measurement method that can have very high variability (Thies et al., 2020). The CO_{2e} is the summation of the emitted greenhouse gases of CO_2-C and N_2O-N that have been converted to CO_2.

One of the goals of 4R is to apply fertilizer at the right time. Matching when the fertilizer is applied to when the plant takes up the nutrient. This strategy has the potential to improve fertilizer use efficiency in many soils. However, splitting the N rate does not consider the potential impact to N_2O emissions. Urea applied in the early summer has demonstrated increased GHG emissions compared to urea applied in the fall. The lower N_2O emissions in the early winter than early summer were attributed to lower fall soil temperatures which increased N_2O water solubility and reduced microbial activity (Joshi et al., 2022; Reicks et al., 2021; Clay et al., 2015; Clay et al., 2012). The hypothesis of this experiment was that a 50/50 split urea (157 kg N/ha) as a broadcast application will not reduce and may increase N_2O-N and CO_2-C emissions compared to a single (157 kg N/ha) broadcast application Therefore, the objective was to determine if splitting the N rate increased N_2O-N, CO_2-C, and CO_{2e} emissions.

MATERIALS AND METHODS

Site location and Climate

The study was conducted at the SD Aurora Research Farm that was located at 44° 18’ 20.0448” N, 96° 40’ 12.5004” W. The soil at the site was a Brandt silty clay loam (Fine-silty, mixed super active, frigid Calcic Hapludolls) (Thies et al., 2019; Clay et al., 2015; Soil Survey Staff , 2023) and the parent materials were loess over glacial outwash. The soil surface horizon contains 110g sand, 580 g silt, and 310 g clay. The soil organic carbon in the surface 15 cm was approximately 2.21 g SOC-C kg^{-1}, (Clay et al., 2015). The surface soil water contents at field capacity and the wilting point were 0.315 g/g and

0.1777 g/g, respectively (Thies et al., 2020). The surface 15 cm had a bulk density of 1.27 g/cm^3 in 2021 and 2022. Additional information on this site has been reported in Clay et al., (1996, 2005, 2015). In 2021 and 2022 years, the site had been in no-tillage for one year and the previous crop was soybean (*Glycine max* L.). The experiment site is found within a semi-arid moisture regime on the eastern side of South Dakota with a hot summer continental climate (Dfa) as the Koppen climate region. Rainfall and air temperatures were obtained from the South Dakota Mesonet (2023).

Experimental Design and Treatments

The study used a completely randomized design (CRD), three N rates (0, 157 kg N/ha, and 78.5 kg N/ha that was applied twice), with two replicates and it was repeated in 2021 and 2022. For the pre-emergence 157 kg N/ha treatment, N was broadcast late due to funding complications on 1 June 2021 and several days after planting 17 May 2022. For the split N application, the second broadcast application (78.5 kg N/ha) was applied at the corn V6 growth stage. In 2021 and 2022 the applications were made on 28 June 2021 and 22 June 2022, respectively.

Corn was planted on 12 May in 2021, and 11 May 2022 at a seeding rate of 79,000 plants/ha. It was harvested on 15 Oct 2021 and 11 Oct 2022. The corn rows were separated by 76 cm. The impacts of splitting the N rate on corn yields and N budgets are beyond the scope of this paper and will be reported in subsequent papers.

Greenhouse gas emissions and soil moisture and temperature measurements

Assays of GHG emissions were initiated on 17 May 2021 before urea application and 25 May 2022, 8 days after urea application and terminated on 25 Oct 2021 and 11 Oct 2022. GHG emissions were measured with LI-COR LI-8100-104 long-term opaque chambers that had a diameter of 20 cm (8100-104 LI-COR, Lincoln, NE). The chamber consisted of a base with a bottom PVC ring that was pushed approximately 5 cm into the soil, and a pivoting top that seals itself by pivoting itself to cover the base. The chambers had a surface area of 317 cm^2. Each chamber measured emissions six times daily between 0000 to 0230 h, 0400 to 0630 h, 0800 to 1030 h, 1200 to 1430 h, 1600 to 1830 h, and 2000 to 2230 h (LI-COR, 2019). At each sampling interval, gas concentrations (CO_2, and N_2O) were measured every second for 15 minutes by a Picarro® Cavity Ringdown Spectrometer (model G2508; Picarro Inc., Santa Clara, CA). N_2O-N emissions were calculated with data obtained between 45 to 900 seconds, while CO_2-C emissions were most accurately calculated between 45 and 165 seconds. The software 4.01 LI-COR SoilFluxPro™ (v. 4.01; LICOR, Lincoln, NE) was used in the calculations. At the initiation and completion of the studies, measured GHG values were compared with known standard gases (Airgas USA LLC, Cinnaminson, NJ) .

Soil temperature and moisture for the surface 5 cm were measured simultaneously with LI-COR LI-8150-205 Soil Moisture Probes (LI-COR, Lincoln, NE) and LI-COR LI-8150-203 Soil Temperature Probes (LI-COR, Lincoln, NE). During the two-year experiment there were machine errors and power failures that resulted in short gaps in measurement of gas, soil temperature, air temperature, and soil moisture which were replaced with time-appropriate information from each chamber. The chambers were placed in the center between the corn rows.

Soil sampling

Soil samples were collected at pre-plant and post-harvest in 2021 and 2022. Initial soil samples from the 0 to 15, 15 to 30, and 30 to 60 cm depths were analyzed for soil moisture, EC, pH, soil organic matter (SOM) and inorganic N. Following soil moisture determination, samples were dried, ground, and sieved. Inorganic nitrogen in the soil sample was extracted with 1M KCl using a 10 to 1 solution to soil ratio. This mixture was shaken for one hour and filtered. The extracted solution was analyzed for NO_3-N and NH_4-N using spectrophotometry (Astoria-Pacific ™). The soil properties and chemical conditions are shown in Table 2.1. Soil organic matter was determined using loss on ignition, and $pH_{1:1}$ and $EC_{1:1}$ was determined using 10 g soil and 10 mL of water. After harvest, soil samples were collected on 29 Oct 2021 and 21 Oct 2022 from 0 to 60 cm depth. These samples were analyzed for NO_3-N and NH_4-N using the above method.

Phospholipid Fatty Acid Analysis (PLFA)

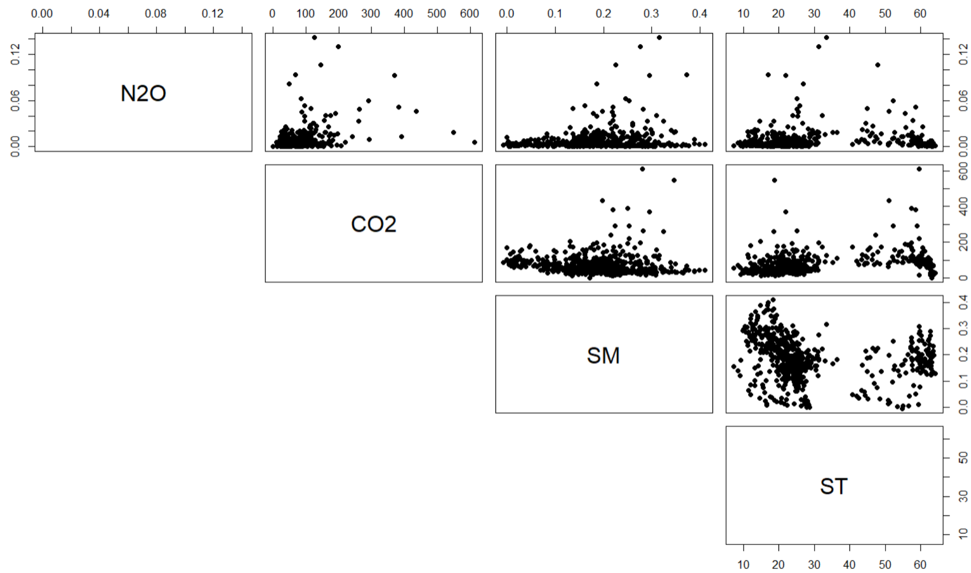
The soil microbial community plays a crucial role in N_2O and CO_2 emission pathways through denitrification, nitrification, and microbial respiration. Composite soil samples were collected from the surface 15 cm on 19 July 2021 and 15 July 2022. Following collection, the samples were placed in a sterile bag that was placed on dry ice. Once out of the field, the samples were stored in a -80℃ freezer until analysis. To prepare the samples for analysis they were dried for approximately 8 hours using vacuum pressure and centrifugation in the Thermo Scientific Savant SC250EXP SpeedVac Concentrator. Once dried, 1.5g of soil was weighed into a tube to complete the four steps of PLFA: Extraction, Separation, Chromatography, and Transesterification. Further PLFA methodology is detailed in the appendix. The samples were then analyzed using gas chromatography via GC-2010 Plus High-end GC and the peaks were analyzed using MIDI Sherlock™ Chromatographic Analysis System (CAS).

Statistical analysis

ANOVA was conducted to determine significant differences among the treatments, years, and years by treatment (R core team, 2020). Years and treatments were considered fixed effects. On GHG emissions there was a significant interaction between years and treatments ($p = < 0.01$). An LSD test was used to determine treatment differences (R Core Team, 2020).

Spearman correlation, through R software, was used to determine a correlation across years between the measured gases, soil temperature, and soil moisture. Spearman correlation was used instead of Pearson correlation due to the monotonic relationship between the variables. Pearson is used when the variables show either a positive or negative linear relationship, this can be tested through a scatterplot. However, this GHG data was monotonic showing both positive and negative relationships with variables moving in the same direction but not at a constant rate (Figure 2.1). When visually inspected, the scatterplots did not produce a constant positive or negative linear graph, but instead some variables had the same strength and direction without a constant rate, producing a non-linear graph. P-values for each correlation were also determined through the R software.

Figure 2.1. Scatterplot of N_2O-N (kg/ha), CO_2-C (kg/ha), soil moisture (SM) (cm^3/cm^3) and soil temperature (ST) (℃) over total 2021 and 2022 growing season. In 2021, the sampling period was 17 May to 25 October. In 2022, the sampling period was 25 May to 11 October.



RESULTS AND DISCUSSION

Table 2.1. Rainfall cm for both experimental years from planting to split application and after split application to harvest, (South Dakota Mesonet, 2023).

Year	Date	Planting to Split (V6)	Date	Split to Harvest
		Rainfall cm		Rainfall cm
2021	12 May- 28 Jun	1.96	29 Jun- 19 Oct	16.8
2022	11 May- 22 Jun	10.6	23 Jun- 12 Oct	17.7

Table 2.2. Rainfall cm and Air Temperature℃ for both experimental years by month and 100-year (1901-2000) average by month from May to October data was collected from South Dakota Mesonet.

	Average Rainfall cm						Average Air Temperature℃					
Year	May	Jun	Jul	Aug	Sept	Oct	May	Jun	Jul	Aug	Sept	Oct
2021	4.5	1.7	5.8	6.4	5.4	8.6	14	23	24	23	19	12
2022	7.2	5.3	6.7	3.0	1.0	1.0	14	20	24	23	19	10
1901-2000	8.1	9.3	6.9	6.3	5.1	3.8	14	20	23	22	17	10

Total growing season (1 May -30 Sept) in 2021 and 2022 was 32.5 and 26.2 cm, respectively. Rainfall in 2021 was 24.1 cm while 2022 had 20% greater rainfall with a total of 30 cm. At Aurora South Dakota, the long-term 100-year precipitation mean (1901- 2000) at planting (May) is 8 cm (NOAA, 2023) and in June the 100-year average is 9.2 cm. The 100-year annual precipitation mean (1901-2000) is 55 cm and the 100-year precipitation mean (1901-2000) from 1 May to 30 September is 35 cm (NOAA, 2023). In 2021, low rainfall during May and June resulted in 1.03 cm of surface soil moisture (5 cm) on 28 June 2021. The split application date in 2022, 22-June had more soil moisture at the soil surface (1.57 cm). 2021 and 2022 both had higher accumulated growing degree days (GDD) (1 May – 31 Oct) than long term 20-year seasonal GDD of 1376 (NOAA, 2023). Base temperature used for corn was 10℃ and maximum temperature was 30℃.

Table 2.3. Initial soil properties (1 May- 31 May) pre-V6 in the surface 0-15cm depth in 2021 and 2022 experimental years.

		Pre-V6
2021	NO_3 kg N/ha	32.5
	NH_4 kg N/ha	14
	pH $_{1:1}$	5.6
	EC $_{1:1}$ dS/m	0.14
	Soil Organic Matter %	0.46
	Bulk Density 15cm g/cm3	1.27
2022	NO_3 kg N/ha	26.2
	NH_4 kg N/ha	15.4
	pH $_{1:1}$	6.5
	EC $_{1:1}$ dS/m	0.17
	Soil Organic Matter %	0.49
	Bulk Density g/cm3	1.27

Initial Soil Properties and Climate Conditions

Prior to planting, the amount of NO_3^- in the surface 15 cm in 2021 and 2022 was 32.5 kg-N/ha and 26.2 kg-N/ha, respectively (Table 2.1). These results suggest that in both years the soil contained a moderate amount of NO_3^-. The amount of inorganic N concentrations contained in the soil are important. For example, Thomas et al. (2017) suggested that the transformation of N_2O into N_2 increases when NO_3^- concentration decreased below 6 ppm (11 kg N/ha). This is lower than the initial soil NO_3 levels in both experimental years indicating more N_2O is being lost to the atmosphere because the microorganisms in the soil are less likely to reduce N_2O into N_2. Weier et al. (1993) reported that when the microbial community is N limited adding N may stimulate both denitrification and CO_2 emissions.

Nitrous oxide emissions

Table 2.4. The N_2O-N loss during the 2021 and 2022 growing season with first and second application compared separately and both years analyzed together showing the Treatment: Year interaction significance. Means with different letters are different at the 5% level. In 2021, the sampling periods were pre- split, before V6 from 17 May to 27 June and post-split after V6 from 28 June to 25 October. In 2022, the sampling periods were from 25 May to 21 June and from 22 June to 11 October.

N Treatment	Year	Pre-Split	N rate kg N/ha	Post-Split	Total N applied	Total
kg N/ha		kg N_2O-N/ha	kg N/ha	kg N_2O-N/ha	kg N/ha	kg N_2O-N/ha
0	2021	0.03a	0	0.04a	0	0.07a
78.5	2021	0.05a	78.5	0.13bc	157	0.18a
157	2021	0.05a		0.08a	157	0.13a
0	2022	0.15a	0	0.19bc	0	0.34a
78.5	2022	0.28a	78.5	1.67c	157	1.95b
157	2022	0.71b	0	0.60b	157	1.32b
p value		0.04		< 0.01		0.02
	2021	0.04a		0.08a		0.13a
	2022	0.38b		0.82b		1.20b
p value		< 0.01		< 0.01		< 0.01
0		0.09a	0	0.11a	0	0.21a
78.5		0.16a	78.5	0.90b	157	1.06b
157		0.38b	0	0.34a	157	0.72b
p value		0.03		< 0.01		0.01

Table 2.5. The CO_2-C loss during the 2021 and 2022 growing season with first and second application compared separately and both years analyzed together showing the Treatment: Year interaction significance. Means with different letters are different at the 5% level. In 2021, the sampling periods were pre- split, before V6 from 17 May to 27 June and post-split after V6 from 28 June to 25 October. In 2022, the sampling periods were from 25 May to 21 June and from 22 June to 11 October.

N Treatment	Year	Pre-Split	N rate kg N/ha	Post-Split	Total N applied	Total

kg N/ha		kg CO2-C/ha	kg N/ha	kg CO2-C/ha	kg N/ha	kg CO2-C/ha
0	2021	199a	0	847a	0	1045a
78.5	2021	255a	78.5	1,001a	157	1256a
157	2021	294a		888a	157	1183a
0	2022	700a	0	1,460a	0	2161a
78.5	2022	1130a	78.5	8,425b	157	9556b
157	2022	3029b	0	11,245c	157	14274c
p value		0.02		< 0.01		< 0.01
	2021	249b		912b		1162b
	2022	1620a		7044a		8664a
p value		< 0.01		< 0.01		< 0.01
0		449b	0	1153c	0	1603c
78.5		693b	78.5	4713b	157	5406b
157		1662a	0	6067a	157	7727a
p value		0.01		< 0.01		<0.01

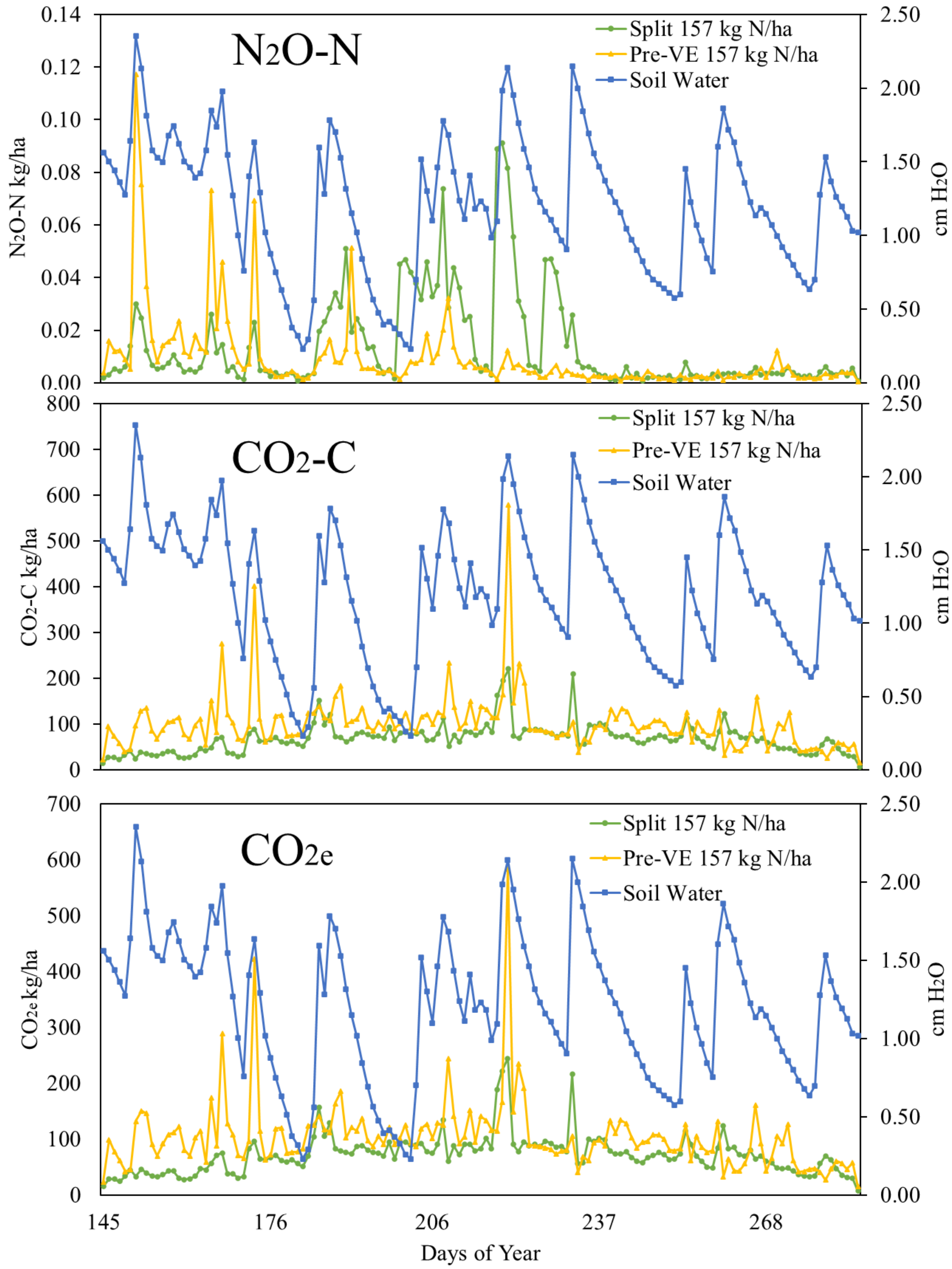
Table 2.6. The carbon equivalents (CO_{2e}), as impacted from fertilizer treatment and year. Due to both CO_2-C and N_2O-N. Both years during the 2021 and 2022 growing season with first and second application compared separately and both years analyzed together showing the Treatment: Year interaction significance. Means with different letters are different at the 5% level. In 2021, the sampling periods were pre- split, before V6, from 17 May to 27 June and post-split, after V6, from 28 June to 25 October. In 2022, the sampling periods were from 25 May to 21 June and from 22 June to 11 October.

N Treatment	Year	Pre-Split	N rate kg N/ha	Post-Split	Total N applied	Total
kg N/ha		kg CO_{2e}/ha Intensity	kg N/ha	kg CO_{2e}/ha Intensity	kg N/ha	kg CO_{2e}/ha Intensity
0	2021	209a	0	859a	0	1067a
78.5	2021	269a	78.5	1040a	157	1310a
157	2021	309a		913a	157	1222a
0	2022	745a	0	1517a	0	2262a
78.5	2022	1213a	78.5	8922b	157	10136b
157	2022	3242b	0	11424c	157	14667c

p value		0.02		< 0.01		< 0.01
	2021	1733b		937a		1199a
	2022	262a		7288b		9021b
p value		< 0.01		< 0.01		< 0.01
0		477a	0	1187a	0	1665a
78.5		741a	78.5	4981b	157	5723b
157		1775b	0	6168c	157	7944c
p value		0.01		< 0.01		< 0.01

Nitrous oxide emissions

Figure 2.2. N_2O-N, CO_2-C and CO_{2e} emissions with soil moisture (cm H_2O) at 0 to 5 cm at post V6 mid- season 15-July to 28-Aug 2021 and 2022.



Across the years, adding urea increased N_2O-N emissions. These results were expected because N_2O emissions are produced during nitrification and denitrification. Prior to the split urea application, the lowest N_2O emissions rates were for the 0 and 78.5

kg N/ha treatment in 2022 while in 2021 the 0, 78.5, and 157 kg N/ha treatments were all low. The lack of increase in emissions in 2022 between the 0 and 78.5 kg N/ha treatment was attributed to N_2O emissions following an S-shaped curve rather than a linear curve. In an S-shaped curve, N_2O emissions may not be increased by small increases in organic N when the amount of inorganic N in the soil is either low or high (Joshi et al., 2022; Kima et al., 2013). After the split application the highest N_2O emissions losses for 2021 and 2022 were in the 78.5 kg N/ha treatment. These results were attributed to reduced solubility of the gases with increasing temperature (i.e. gases are emitted at higher temperatures). Thies et al. (2020) had similar results and reported that fertilizer-induced N_2O-N emissions were highest when applied on 12 June and lowest when applied on 1 November. The impact of the sampling date on N_2O-N losses could be attributed to several factors including higher temperatures and moisture differences across the growing season. The lowest N_2O-N emissions were associated with the 0 N/ha treatment. The split while numerically higher was not significantly different from the application of 157 kg N/ha early in the growing season. These results indicate that splitting the fertilizer rate did not reduce N_2O emissions. Venterea et al. (2015) had the same results and reported that splitting the N rate increased N_2O -N emissions in one of the two years. They attributed their results to rainfall fluctuations that resulted in a prolonged dry period prior to the fertilizer application followed by a large rainfall event after fertilizer application. Others have reported a mixed impact of a split application, on N_2O (Burton et al., 2008; Del Grosso et al., 2009; Phillips et al., 2009). For example, De Grosso et al. (2009) used the DAYCENT model to assess the impact of different management scenarios on CO_{2e} and reported that the split N had higher yields of corn than baseline management but similar CO_{2e} values. Burton et al. (2008) reported that splitting the fertilizer application decreased cumulative N_2O emissions in 2003, but not in 2002.

Across years, the split and preplant fertilizer application treatments emitted < 1% of the fertilizer derived N (Fertilizer – 0 N) (DeKlein et al., 2006). For the split N rate the % decrease was 46% [=100*(1- 0.58/1.57)] and for the preplant N the decrease was 68 % [=100*(1-0.51/1.57)]. Del Grosso et al. (2007) using the DAYCENT model had similar results, and reported that emissions can be lower than IPCC predicted values.

Lower than predicted values may be due to not accounting for N immobilization/fixation. In related research conducted at this site, Thies et al. (2020) reported that 54% of the applied fertilizer was either immobilized by the soil microorganisms or fixed on the clay exchange sites. Microbial immobilization or clay fixation contradicts the perception that 100% nitrogen is available and subject to leaching, nitrification, and denitrification. Others have reported that immobilization and/or fixation can affect inorganic N concentrations. For example, Nieder and Benbi (2008) reported that NH_4^+ fixation can range from 10 to 60 mg N/kg in sandy soils and from 90 to 460 mg N/kg in clay soils. Clay et al. (1990a) reported that 18 days after applying ^{15}N-labeled urea to sandy loam soil, 35% was either immobilized/fixed into pools that were not extracted by 1 M KCl. Of the immobilized/ fixed N, 13.9% and 66% were in 6 M HCl hydrolysable amino acid and NH_4^+ pools, respectively. They also reported that the immobilized/ fixed-N mineralized 5 to 10 times faster than the non-labeled organic N. Therefore, while a significant portion of N is lost as N_2O-N to the atmosphere in agriculture, a much larger portion of N is likely fixed to clay particles or immobilized by microorganisms. It is important to consider the possibility that due to environmental conditions, 2021 had greater fixation and immobilization of the N compared to 2022, producing greater N_2O-N emissions in 2022. The higher soil moisture recorded in 2022, compared to in 2021, produced higher emissions likely due to microbial and plant metabolic functions being more able to actively work with the addition of water while a dry soil will have slowed these metabolic functions in the soil significantly (Bogati & Walczak, 2022).

Carbon dioxide emissions

When urea [$CO(NH_2)_2$] is applied to soil, it is hydrolyzed by the extracellular urease enzyme. Hydrolysis transforms [$CO(NH_2)_2$] into NH_3 and CO_2. Through secondary reactions, CO_2 reacts with water to form H_2CO_3, and NH_3 reacts with H^+ to form $NH_4^{+.}$ However, H_2CO_3 is not a stable molecule and will quickly decompose into CO_2 and H_2O (DeKlein et al., 2006). CO_2 is also released via microbial respiration when soil organic carbon is mineralized (Clay et al., 2012). The NH_3 component simultaneously reacts with H^+ to form NH_4^+, which when taken up by soil microorganisms can stimulate microbial respiration if they are N-limited.

The year-by-treatment interaction showed that CO_2-C emissions can change depending on the year (Table 2.5). Others have reported large temporal variation in CO_2-C emissions. For example, Joshi et al. (2022) reported that in 2019 and 2020 on a similar soil, CO_2-C emissions were 3935 and 5691 kg CO_2-C/ha, respectively. Lower CO_2-C emissions in 2019 than 2020 were attributed to lower growing degree days that were 1266 in 2019 and 1436 in 2020, and higher growing season rainfall that was 61 cm in 2019 and 32 cm in 2020. At our study site in 2021 and 2022, the growing degree days were almost identical to the past studies in both years, however precipitation was lower in

2021 than 2022 (Table 2.1). Lower soil moisture most likely contributed to reduced CO_2-C emissions (Fig. 2.2).

In addition to a large annual difference in CO_2-C emissions, splitting the fertilizer had a different impact on CO_2-C emissions in 2021 than 2022. For example, in 2021, CO_2-C emissions were not influenced by the fertilizer application, whereas in 2022 CO_2 emissions were increased by urea application. Differences between years most likely can be attributed to less microbial respiration in 2021 than in 2022, which resulted in a lower N requirement in 2021 than 2022. Others have reported that applying N fertilizer can increase CO_2-C emissions (Zhang et al., 2019; Fiedler et al., 2021). Fiedler (2021) showed that when averaging over soils and years, applying 224 kg N/ha increased CO_2-C emissions. Thies et al. (2020) had similar results and reported that urea can have a mixed impact on CO_2-C emissions and increased CO_2-C emissions when applied in early winter and early spring. Additionally, Thies et al. (2020) also reported that CO_2-C emissions did not increase emissions when applied in early fall, mid-fall, mid-spring, and early summer.

N_2O and CO_2 relationship to soil moisture and temperature

Table 2.7. Correlation coefficients between N_2O-N, CO_2-C emission rates as impacted by soil moisture and soil temperature contents. Years were analyzed together across the growing season. In 2021, the sampling periods reported in correlation include 17 May to 25 October. In 2022, the sampling periods reported in correlation include from 25 May to 11 October. Bolded correlation coefficients were significant at the 5% level.

Treatment	Date		N_2O-N			CO_2-C	
2021 and 2022		N rate	SM	ST	CO_2	SM	ST
		kg N/ha	r	r	r	r	r
Control	17-May to 27-Jun	0	0.08	**-0.47**	**0.65**	**0.22**	**-0.53**
Control	28-Jun to 25-Oct	0	**-0.19**	**-0.41**	**0.46**	**-0.43**	**-0.35**
Control	17-May to 25-Oct	0	**-0.19**	**-0.39**	**0.49**	**-0.33**	**-0.39**
Urea	17-May to 27-Jun	157	**0.36**	0.04	**0.51**	0	**0.38**
Urea	28-Jun to 25-Oct	157	-0.04	**0.25**	**0.33**	**-0.3**	**0.29**
Urea	17-May to 25-Oct	157	0.04	**0.27**	**0.28**	**-0.24**	**0.27**

Split Urea	17-May to 27-Jun	78.5	0.01	**-0.25**	**0.29**	-0.14	**0.41**
Split Urea	28-Jun to 25-Oct	78.5	**0.1**	**-0.13**	**0.42**	**-0.26**	**0.26**
Split Urea	17-May to 25-Oct	157	**0.08**	**-0.15**	**0.36**	**-0.32**	**0.33**

The urea fertilizer influenced the relationship between soil moisture, soil temperature, N_2O-N, and CO_2-C emissions. N_2O -N emissions in the 157 kg N/ha pre-emergence and 78.5 kg N/ha split treatments had weak positive relationships with soil moisture; however, the 0 kg N/ha treatment there was a negative relationship ($r = -0.19$ $p<0.05$) This difference in moisture likely had an impact on denitrification and nitrification in the soil and therefore, an impact on N_2O-N emissions. Nitrification is an aerobic process, whereas denitrification is an anaerobic process. The increased soil moisture and precipitation in 2022 most likely increased the likelihood of N_2O-N being lost from denitrification compared to nitrification.

The fertilizer treatments also affected the relationship between N_2O-N emissions and temperature. The 157 kg N/ha pre-emergence treatment had a positive relationship while the 0 kg N/ha treatment was negatively correlated to soil temperature, ($r= -0.39$, p-value< 0.05). Differences in correlation relationships in the unfertilized and fertilized treatments to soil temperature were also observed for CO_2-C emissions (Table 2.7). CO_2-C emissions from the 0 kg N/ha control had a negative correlation to temperature while 78.5 kg N/ha split application and 157 kg N/ha pre-emergence had positive correlations. These data indicated that soil temperature plays an important role in the gas' solubility, influencing the N_2O-N and CO_2-C emissions lost to the atmosphere.

Many studies have measured over the growing season and have reported that N_2O-N emissions are event based with soil temperature and soil moisture playing important roles (Fujinuma et al., 2011; Venterea et al., 2015; Thies et al., 2020). In this experiment, soil temperature and soil moisture differences in the two experimental years agree with the literature that N_2O-N loss is variable due to environmental fluctuations such as soil moisture and temperature. These environmental characteristics influence the solubility equilibrium and gas diffusion rate (Thies et al., 2019). Henry's Law is used to determine the amount of gas that will be retained by the soil. It is a temperature-dependent equation that relates gas solubility to temperature (Blackmer et al., 1982).

N_2O-N loss to the atmosphere also depends on its ability to be emitted from the soil. The release of gases from the soil depends on soil moisture which was different between 2021 and 2022 experiment years. Fick's law states that an increase in soil moisture will slow the release of oxygen gas in the soil via diffusion, the soil-gas diffusion constant coefficient. This can be applied to other gases in the soil solution as well such as N_2O and CO_2 (Thies et al., 2019; Clay et al., 1990a; Desutter et al., 2008). The 2021 experiment year had relatively low soil moisture from May to October whereas in 2022 moisture was higher (Table 2.1 and Figure 2.2).

A positive relationship was observed between N_2O-N and soil moisture in the 78.5 kg N/ha split and 157 kg N/ha pre-emergence treatments, disagreeing with Fick's law. This indicates that gas diffusion is not the single guiding principle to determining emission rates. Soil moisture influences gas fluxes, but also nitrification and denitrification, microbial processes. These processes are ultimately influenced by microbial activity playing a crucial role influencing emissions (Di et al., 2014). Venterea et al. (2015) agree, also finding a positive trending relationship between N_2O emissions and soil moisture.

However, it is also important to consider that N_2O and CO_2 emissions can be limited by many factors, including N and C limitations. When N is limited, adding N will increase respiration and GHG emissions, and when C is limited adding N may not increase GHG emissions. Variable correlations between soil temperature, soil moisture, N_2O-N, and CO_2-C emissions indicate that soil can switch back and forth from being N- and C-limited.

When organic matter decays, the carbon components can be integrated into soil organic carbon or mineralized into CO_2 by microorganisms (Clay et al., 2012; Clay et al., 2015; Chang et al., 2017). Because CO_2 is released from microbial respiration, it follows a diurnal cycle, similar to N_2O (Thies et al., 2019). These cycles throughout the day are based on soil temperature phases (Weerden et al., 2013; Blackmer et al., 1982; Chang et al., 2017; Chang et al., 2016). CO_2-C demonstrated a positive trend with soil temperature in the 157 kg N/ha pre-emergence and 78.5 kg N/ha split treatments, because microbial activity increases with temperature, whereas gas solubility decreases with temperature (Drake et al., 2013). Using phospholipid fatty acid analysis (PLFA), the mid-season 0-15 cm surface soil samples in 2021 and 2022 were analyzed for microbial biomass (Table 2.8). Microbial biomass among the treatments is statistically similar mid-season in July 2021. Figure 2.2 demonstrates how soil moisture likely impacted the microbial community. This reduced microbial respiration and nitrification, resulting in lower emissions in 2021 compared to 2022.

Table 2.8. The total soil microbial biomass ug C/ g soil, fungi ug C/ g soil and bacteria ug C/ g soil in the mid-season post V6 19 July 2021 and 15 July 2022.

N treatment	N rate	Date	Year	Total Biomass	Fungi	Bacteria	Fungi: Bacteria

	kg N/ha			ug C/g soil	ug C/g soil	ug C/g soil	ug C/g soil
Control	0	19-Jul	2021	11.8	1.30	10.49	20.0
Split N Rate	157	19-Jul	2021	10.9	0.70	10.20	13.8
Pre-emergence	157	19-Jul	2021	12.0	1.10	10.90	12.75
Control	0	15-Jul	2022	10.6	0.53	10.10	9.0
Split N Rate	157	15-Jul	2022	9.1	0.51	8.58	9.25
Pre-emergence	157	15-Jul	2022	9.7	0.52	9.20	10.3
p value				0.9	0.7	0.9	0.2
			2021	11.5	1.02	10.3	15.5b
			2022	9.8	0.52	9.30	9.25a
p value				0.19	0.07	0.3	0.03
Control	0			11.2	0.90	10.3	14.5
Split N Rate	157			1.0	0.60	9.5	11.5
Pre-emergence	157			10.9	0.81	10.0	11.5
p value				0.7	0.7	0.8	0.4

Using phospholipid fatty acid analysis (PLFA), the post-V6 was analyzed for microbial biomass and microbial community structure (Table 2.8). Fungi in this analysis included arbuscular mycorrhizal fungi (AMF) and saprophytes, while bacteria included Gram negative unicellular bacteria, Gram positive unicellular bacteria, and actinomycetes. Analysis of the interaction between the years and the treatments was nonsignificant. However, numerically it is clear that 2021 had a higher total microbial biomass than 2022. Analysis of years showed that there was a higher Fungi: Bacteria ratio in 2021 compared to 2022. This supports the low N_2O-N and CO_2-C emissions found in 2021. Fungi have a higher C:N ratio, increasing the amount of carbon that must be collected and stored in the soil to allow fungi to grow and reproduce. Studies utilizing PLFA and RNA genome sequencing support this by indicating that fungi are linked to higher carbon storage potential (Malik et al., 2016). Analysis of treatments also did not indicate that the split application changed the microbial community. This indicated that nitrogen application did not play as significant a role as the environmental conditions within the year.

Findings Relative to IPCC N_2O default value of 1% of applied N

Agriculture is the main source of anthropogenic N_2O loss to the atmosphere. It is estimated that agriculture is responsible for more than 60% of global N_2O emissions (Myhre et al., 2013, Syakila et al., 2011, Adair et al., 2019). Analysis of the interaction between years and treatments indicated that 157 kg N/ha pre-emergence had a higher percent of N lost as N_2O before V6 in 2022; however, after V6 in the same year, the 78.5 kg N/ha split application appeared to lose a higher percent of N as N_2O instead. Over the total growing season, there was no difference between the treatments. In 2021 there was no difference between the treatments before or after corn's V6 growth stage. Analysis of years indicated that overall, 2022 had the highest percent N lost as N_2O. Analysis of treatments indicated that pre-emergence 157 kg N/ha had a higher percentage of N lost as N_2O before the V6 growth stage. After V6, the split application produced the highest loss, but the total over the growing season demonstrated no significant difference.

While agriculture is a main source of N_2O, its loss from applied synthetic fertilizer has demonstrated to be low. Analysis of treatments shows that 75% percent of treatments lost less than 1% of N from fertilizer as N_2O (Table 2.9). This is below the IPCC Tier 1 emission factor method used. Across years, the split and pre-emergence fertilizer application treatments emitted < 1% of the fertilizer derived N (Fertilizer – 0 N) (DeKlein et al., 2006). For the split N rate, the % decrease was 46% [=100*(1-0.58/1.57)] and for the pre-emergence N the decrease was 68 % [=100*(1-0.51/1.57)].

These results agree with past studies and suggest a re-evaluation of lifecycle analysis policies aimed at agriculture depending on climatic variables such as the year's soil moisture content instead of a single static default value. Dry years appear to lose less N as N_2O compared to wet years (Thesis et al., 2019, Halvorson & Del Grosso 2013; Maharjan et al., 2013).

Table 2.9. The % of N applied as urea fertilizer lost as N_2O-N during the 2021 and 2022 growing season with first and second application compared separately and both years analyzed together. The 0 kg N/ha control is subtracted out to determine the effect of the fertilizer. Means with different letters are different at the 5% level. In 2021, the sampling periods were pre- split, before V6, from 17 May to 27 June and post-split, after V6, from 28 June to 25 October. In 2022, the sampling periods were from 25 May to 21 June and from 22 June to 11 October.

N treatment	Year	Pre-Split	N rate kg N/ha	Post-Split	Total N applied	Total
kg N/ha		% N	kg N/ha	% N	kg N/ha	% N
78.5	2021	0.005a	78.5	0.06a	157	0.09a
157	2021	0.004a	0	0.027a	157	0.06a
78.5	2022	0.15a	78.5	1.04b	157	1.21b
157	2022	0.43b	0	0.36a	157	0.81b
p value		0.07		0.04		0.3
	2021	0.005a		0.04a		0.07a
	2022	0.3b		0.70b		1.0b
p value		< 0.01		< 0.01		< 0.01
78.5		0.08a	78.5	0.55b	157	0.65a
157		0.22a	0	0.19a	157	0.43a
p value		0.07		0.03		0.22

Carbon equivalents

In carbon equivalent calculations, all GHG emissions are converted to an equivalent amount of CO_2-C using appropriate conversion factors. CO_2, because of its molecular geometry, absorbs a wide range of wavelengths including infrared wavelengths. This is approximately half of the absorbed energy reradiated toward earth.

As more CO_2 is added to the atmosphere the ability to trap energy (heat) increases. This heats up the earth like a blanket, or the inside of a car left in the sun (Stips et al., 2016). Different gases have various abilities to trap energy. For example, 1 kg N_2O traps as much heat as 298 kg of CO_2 (DeKlein et al., 2006). Therefore, 1 kg of N_2O is equivalent to 298 kg of CO_2 (DeKlein et al., 2006). This produced the carbon-equivalent (CO_{2e}) results before the split application and post-split application, as well as total CO_{2e} over the 2021 and 2022 growing seasons (Table 2.6).

Analysis of the years and treatments interaction shows that the 157 kg N/ha pre-emergence had a higher CO_{2e} compared to the 78.5 kg N/ha split application. This appears to be true especially in the year with higher soil moisture, 2022. However, in 2021 all values were similar. Analysis of the years indicates that 2022 had the higher CO_{2e} over the entire growing season compared to 2021. In 2021 and 2022, the 0 kg N/ha treatment had the lowest CO_{2e} and the 157 kg N/ha pre-emergence treatment had the highest CO_{2e}. These findings indicate that while a split N rate may not reduce N_2O-N emissions, it may reduce the CO_{2e}. Therefore, because CO_{2e} was reduced by the split N application, it should be considered a climate smart practice. However, because prior research did not determine CO_{2e}, additional research is needed. It is important to consider that N_2O and CO_2 are unavoidable products of the important chemical and biological processes associated with producing food. Given our current technologies, we do not have the capacity to eliminate these losses. Therefore, a realistic and prudent goal is reduction, not elimination. For example, during the production of corn, a portion of the fixed soil C is respired to produce CO_2, and a portion of the nitrogen fertilizer applied is lost to N_2O through nitrification or denitrification (Thies et al., 2019; Clay et al., 2012).

In many situations, our ability to accurately assess the impact of 4R nutrient strategies such as urea fertilizer timing has been limited by technology. While the concepts of the 4R model are extremely important, it is necessary to improve our understanding of the theory and application (Clay et al., 1990; Thies et al., 2020, Thies et. al., 2019; Dunsenbury et al., 2008; Reicks et al., 2021, Bouwman et al., 2002). For example, research shows that GHG emissions are lost from soils by diurnal cycles influenced by soil temperature and moisture (Thies et al., 2019; Thies et al., 2020; Weerden et al., 2013). Past recommendations suggested that the diurnal cycle could be minimized by sampling in mid-morning (Parkin & Venterea, 2010). However, Theis et al. (2019) showed that this sampling protocol may or may not accurately estimate emissions.

This technology has been used by many studies for example, in samples collected every two weeks from 7 May 2018 to 3 July 2018 at 12:48 PM the average N_2O-N emissions were 2.18 ± 3.54 g N_2O-N /(ha×4 h). When samples were collected every 4 hours over the same period, the average emissions were 1.48 ± 0.22 g N_2O-N /(ha×4 h). It is important to note that the two sampling protocols had different means and confidence intervals. Improvements in technology allow for continuous measurement throughout the growing season, as well as the simultaneous measurement of soil physical properties, providing a greater understanding of the system to reduce environmental impact.

CONCLUSION

Climate smart agriculture focuses on site specific approaches to achieving its three pillars of increasing soil productivity and resilience while reducing GHG emissions. The IPCC has predicted that increasing the earth's warming to 2℃ could reduce yields globally by

15% (Masson-Delmotte et al., 2018; Campbell, 2017) (EPA, 2023). To reduce this risk, GHG emissions need to be reduced. One approach to help achieve GHG emission reduction is to adopt climate smart practices. However, techniques used in the past have relied on methods with very high measurement variability. To resolve these fundamental issues and to assess the potential more accurately for the reduction of GHG emissions using 4R nutrient management, additional research was conducted. The results showed that N_2O was not impacted by the split application and may be increased by the split. However, the split application reduced CO_2 and CO_{2e} emissions in wet years. Therefore, the 4R nutrient framework has demonstrated that it can be used as a climate smart practice to reduce GHG emissions and can act as one of many the site-specific approaches to combat the negative effects of climate change (Galloway et al., 2003; Wolfe & Patz, 2002). Similar research should be conducted on other 4R nutrient management strategies.

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FINAL STATMENT

Agriculture is a large producer of GHG emissions such as N_2O and CO_2 which can result in persistent environmental effects (Galloway et al., 2003; Cavigelli et al., 2012). These GHG emissions absorb energy in the form of photons in the earth's atmosphere, warming the earth and increasing the effects of global warming, which include loss of biodiversity, decreased ecosystem resiliency and a reduced ability to grow food (De Klein et al., 2006; Weil & Brady, 2017). Climate Smart Practices focus on strategies to mitigate these effects. These practices work within management frameworks to focus on an assortment of site-specific and locally based strategies to increase soil and crop productivity, increase resiliency in the agriculture ecosystem, and to mitigate the effects of climate change by reducing GHG emissions. Currently, technology is unable to entirely prevent the release of these GHG emissions from the soil because they are emitted through necessary soil processes such as nitrification, denitrification, and microbial respiration. Therefore, the goal is to reduce loss of GHG emissions to the atmosphere.

This can be achieved using Climate Smart Practices effectively implemented through DSSs that are combined with AI and ML algorithms. DSSs as a PA technology are effective tools that can find patterns in large datasets and synthesize the information into a sustainable and profitable recommendation. The 4R management model is a framework that can be utilized by DSSs. This framework includes climate smart strategies focusing on applying fertilizer at the right time, right rate, right place, and right source. New technologies are changing agriculture practices and research broadly. An example of this change is in GHG measurement. Initially, GHG emission measurement methods that have been used to make important policy decisions for agriculture were measured with much variability. The emissions were measured once daily or weekly through static chambers using approximately five syringes to collect the gas from the headspace over equally spaced intervals of time (for example, collected at 0, 0.5, 1, and 1.5 hours) for each chamber. This data would be averaged over the entire growing season (Venterea et al., 2015; Dusenbury et al., 2008; Thomas et al., 2017). However, by using new technology such as near-continuous measurement systems, an example being LI-COR LI-8100-104 long-term opaque chambers (8100-104 LI-COR, Lincoln, NE), researchers can more accurately measure and determine the impact of the 4R model on GHG emissions (Thies et al., 2019; Weerden et al., 2013; Thies et al., 2020; Blackmer et al., 1982). This has increased our understanding of how various aspects of the 4R framework may impact GHG emissions. Technology is changing rapidly and providing new information to agriculture researchers and managers. Therefore, management strategies in agriculture must also change. This thesis has discussed the importance of, and barriers to, widespread use of technology such as DSSs and machine learning that can help evolve agriculture management strategies and achieve climate smart goals. Finally, the second chapter of this thesis evaluated fundamental concepts in management practices and, by utilizing new technology in a two-year study, has determined that split N timing, as described within the 4R framework can be considered a climate smart practice due to the reduction of GHG emissions such as CO_2-C and CO_{2e}. Future research may compare nitrogen stabilizers such as urease inhibitors to non-stabilized fertilizer. These nitrogen stabilizers may change peak emission time which may produce inaccurate results when comparing GHG emission peaks for nitrogen stabilizers to the non-stabilized fertilizer. For example, when a fertilizer amendment is applied such as NBPT the peak emission is slightly later than its untreated urea counterpart (Clay et. al., 1990). If the GHG measurement is taken once a day or once weekly, the measurement could pick up the peak of NBPT but miss the much larger peak from untreated urea that occurred more quickly after application. These results would conclude that NBPT increases GHG emissions compared to untreated urea. Therefore, our understanding of the 4R model in terms of nitrogen stabilizers as well as other aspects of the framework like fertilizer timing, requires new technology and data collection techniques such as the near continuous measurement technology.

Additionally, this project will continue to move forward with evaluating components of the 4R management strategy, examining 'the right source' using nitrogen stabilizers to determine their potential as Climate Smart Practices. The nitrogen stabilizers that will be included will be urease inhibitor N-butyl-thiophosphoric-triamide (NBPT) treated urea also known as Factor or Agrotain commercially, and slow-release polymer-coated urea fertilizers known as Environmentally Smart Nitrogen (ESN). This will focus on measuring the impact of NBPT- treated urea and polymer-coated urea (ESN) on GHG emissions utilizing the long-term opaque chambers. Additionally, analysis of crop yield, tissue RNA expression, and the soil microbial community including PLFA and DNA analysis will also be performed.

Previous studies have indicated that NBPT treated urea can reduce Ammonia emissions 100 times more than its untreated counterpart (Clay et al., 1990). This is because NBPT delays the urease enzyme from breaking down the urea molecule. Originally, the urea molecule would be drawn into the urease enzyme's active site by the two Ni^{2+} ions inside the site (Franzen, 2017). However, with NBPT treated urea, the NBPT molecule will act as a competitive inhibitor to urea for approximately 10 days, making it more difficult for urea to be broken down (Franzen, 2017). This will more accurately synchronize the timing of nitrogen release to nitrogen demand, making NH_3 less likely to be lost through volatilization when applied to the soil surface. Many studies have demonstrated the effectiveness of NBPT. A meta-analysis on use of NBPT reviewed literature from 1990 to 2014. It used regression analysis to determine that NBPT is capable of reducing NH_3 volatilization loss by 52% across all soil texture classes, pH classes, soil organic carbon (SOC) classes and nitrogen rates. NBPT was also observed to delay 50% of nitrogen loss by 8.3 days compared to the untreated urea, as well as produce a 5.3% crop yield increase (Silva et al., 2017). Future work will create a cost benefit analysis of using NBPT compared to untreated urea. This will examine how corn yield is affected from using a split application of NBPT compared to a single pre-emergence application. Additionally, future work will examine how NBPT may affect the plant at the transcription level by examining RNA expression from tissue samples. The impact of using NBPT compared to an untreated urea on the soil microbial community will also be evaluated using PLFA and DNA sequencing of the soil to gain greater understanding of how soil biology can be impacted by NBPT.

Polymer-coated urea (ESN) slows the release of nitrogen by waiting for an increase in soil moisture and soil temperature (Franzen, 2017). ESN slows the release of urea by first allowing soil moisture to move into the polymer membrane, then the urea granule forms a nitrogen solution with the soil water which, is released from the membrane as the soil temperature increases. The N is released when physiological development is most likely to occur, increasing the likelihood that the N will be taken up and used by the plant.

When considering GHG emissions and climate smart practices of this nitrogen stabilizer, Parkin and Hatfield et al. (2013) reported that ESN was less effective at reducing N_2O emissions in a dry, rain-fed region when compared to conventional fertilizers UAN, and UAN treated with Agrotain. However, Halvorson and Del Grosso et al. (2013) reported that ESN applied to irrigated fields was effective at reducing N_2O by 34% to 57% compared to untreated urea. This indicates that ESN is more effective in an environment with higher soil moisture and is more likely to reduce N_2O loss due to denitrification than from nitrification. Additionally, future work will examine how ESN may affect the plant at the transcription level by examining RNA expression from tissue samples. The impact of using ESN compared to an untreated urea on the soil microbial community will also be evaluated using PLFA and DNA sequencing of the soil to gain greater understanding of how soil biology can be impacted by ESN.

Overall, future works will focus on exploring the network of soil biology within the agroecosystem, nutrient-use efficiency, and yield to understand how the 4R model affects all aspects of the agroecosystem, and the potential for each component to be a climate smart practice.

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APPENDIX

Introduction to Phospholipid Fatty Acid Analysis

The procedure was completed in 4 steps using the Buyer & Sasser extraction method (Buyer & Sasser, 2016). These steps included: extraction and separation of the lipids from the rest of the organic material, isolation of the phospholipids through lipid chromatography, and finally the production of Fatty Acid Methyl Esters (FrAMEs) through the process of transesterification. These FrAMEs are analyzed for peaks using flame ionization chromatography (FID-GC) to determine microbial community. These peaks were interpreted by the MIDI Sherlock™ Chromatographic Analysis System (CAS) Software. The microbial categories identified are based on the structural components of the fatty acid chain through nomenclature as follows: A:BωC. "A" indicates the number of carbon atoms in the fatty acid position, "B" indicates the number of double bonds, and "C" indicates the carbon atom from the aliphatic end before the double bond. Prefixes and suffixes also indicated cis or trans configuration as well as iso and anteiso. Additionally, the point of branching and point of hydroxy groups were also considered as the software identified the microbial community within each sample.

Preparation Details

Once soil was collected it was immediately placed on dry ice and stored in a -80℃ freezer. The sample was later freeze dried through vacuum pressure and centrifugation using the Thermo Scientific Savant SC250EXP SpeedVac Concentrator. This drying process occurred for approximately 5 hours. The method of extraction used in this procedure is the Buyer & Sasser extraction method (Buyer & Sasser, 2016) which uses the Bligh and Dyer method to optimize lipid extraction. Additionally, an internal standard 1,2-dinonadecanoyl-sn-glycero-3-phosphochloline (19:0 PC) was added at the beginning of extraction to assess overall extraction method. Lipid chromatography was performed using a silica gel plate to produce the separation of phospholipids from neutral lipids and glycolipids (Quideau et al., 2016; Zelles, 1999).

List of Reagents and Equipment Used in Procedure

1. Screw caps and glass test tubes 16x100
2. Disposable glass Pasture Pipettes
3. 50 mM PO_4 Buffer pH 7.4 (8.7g K_2HPO_4 per liter of deionized water)
4. Bligh-Dyer Extractant (100ml of 50 mM PO_4 Buffer, 500ml methanol, 125ml chloroform)
5. Internal standard 1,2-dinonadecanoyl-sn-glycero-3-phosphochloline (19:0 PC) purchased from Avanti Polar Lipids Catalog #850367P as a white powder.
6. Thermo Scientific Savant SC250EXP SpeedVac Concentrator
7. Silica gel SPE 96-well plate from Phenomenex part #8E-S012-DGB
8. Microplate E & K Scientific Part # EK-99238
9. Transesterification reagent (0.561g KOH, 75ml methanol, 25ml toluene)
10. Thomas Scientific 2ml autosampler GC vial Part # 2702-A01
11. Agilent 250 ul limited volume insert Part #5183-2085
12. Thomas scientific screw caps for GC vials with PTFE/Silicone/PTFE Septa part #2702-A68

13. GC-2010 Plus High-end GC
14. MIDI Sherlock™ Chromatographic Analysis System (CAS) Software

The Procedure was Completed as Follows:

Extraction and Separation

1. 1.5g of freeze-dried soil, 4 ml of Bligh-Dyer extractant, 2 uL of internal standard 19:0 PC was added to a sterile glass 16x100 test tube.
2. The sample was capped and vortexed.
3. The sample was sonicated for 10 minutes and vortexed with this step being immediately repeated.
4. The sample was centrifuged for 11 minutes at 3700 rpm.
5. The supernatant was transferred to a new sterile glass tube.
6. 1 ml chloroform was added to the glass tube containing the sample.
7. 1 ml of deionized water was added to the glass tube containing the sample.
8. The sample was capped and vortexed.
9. The sample was centrifuged for 15 minutes at 3700 rpm.
10. The bottom lipid layer was transferred to a new sterile glass tube.
11. The sample was freeze dried using SpeedVac Concentrator for 1 hour.

Lipid Chromatography

1. The freeze-dried sample was dissolved in 1 ml of chloroform.
2. The sample was capped and vortexed.
3. 1 ml of methanol was added to the wells of a silica gel plate. The methanol was allowed to gravity drain into the waste container underneath the wells for 1 minute and then was drained through vacuum pressure.

4. This step with methanol was repeated twice more.
5. This step with 1 ml of chloroform was preformed three times.
6. The sample extract was applied to the wells, the liquid was gravity drained for 1 minute and then was vacuum drained into the waste container.
7. 1 ml of chloroform was added to the wells containing the sample, the liquid was gravity drained for 1 minute and then was vacuum drained into the waste container.
8. 1 ml of acetone was added to the wells containing the sample, the liquid was gravity drained for 1 minute and then was vacuum drained into the waste container.
9. The waste container underneath the wells was replaced by microplate to collect the phospholipid extract from each sample.
10. 0.5 ml of extract containing a ratio of 5 ml methanol :5 ml chloroform: 1 ml deionized water was applied to the wells containing the sample, the liquid was gravity drained for 1 minute and then was vacuum drained into the small sterile glass tubes.
11. The collected liquid containing the extracted phospholipids in the small tubes was decanted into large sterile tubes.
12. The sample was freeze dried using SpeedVac Concentrator for 1 hour.

Transesterification

1. The freeze-dried sample was dissolved in 0.2 ml of transesterification reagent.
2. The sample was capped and vortexed.
3. The sample was placed in a 37℃ oven for 15 minutes.

4. 0.4 ml of 0.075M acetic acid was added to the sample.
5. 0.4 ml of chloroform was added to the sample.
6. The sample was capped and vortexed.
7. The sample was placed at room temperature and the layers were allowed to separate for 15 minutes.
8. The bottom fatty acid methyl ester layer was transferred to a sterile glass 2ml GC vial.
9. The sample within the vial was freeze dried using SpeedVac Concentrator for 20 minutes.

Gas Chromatography Analysis

1. The freeze-dried sample was dissolved in 75ul of hexane using a glass syringe.
2. The dissolved sample was transferred to a glass limited volume insert that was then inserted back into the GC vial.
3. The sample GC vials were placed in order within the autosampler attached to the gas chromatography instrument.
4. The peaks were analyzed using flame ionization gas chromatography and using MIDI Sherlock™ Chromatographic Analysis System (CAS).

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