

## Table of Contents

---

Clay, David - #1828 - Influence of nitrogen stabilizers and application dates in no-tillage corn production on nitrogen use efficiency and N losses to the atmosphere and groundwater .....	1
Interim Report - Due July 1, 2022 .....	1
Interim Report - Due July 1, 2022 .....	3

# Progress Report

## 2022 South Dakota Nutrient Research and Education Council Invited Proposals

<b>Progress Report Title:</b>	Interim Report - Due July 1, 2022
<b>Applicant Name:</b>	David Clay
<b>Application Title:</b>	Influence of nitrogen stabilizers and application dates in no-tillage corn production on nitrogen use efficiency and N losses to the atmosphere and groundwater
<b>Application ID:</b>	1828
<b>Review Deadline:</b>	07/1/2022 11:59 PM

## Interim Report - Due July 1, 2022

### Project

	Start Date	End Date
<b>Start and End Dates of Funding:</b>	01/1/2022	12/31/2022
<b>Title of Project:</b>	Building a South Dakota Corn No-tillage N Recommendation Algorithm that Considers Improvements in Soil Health	
<b>Project Description:</b>	<p><b>Research goal:</b></p> <p>The goal of this project is to improve South Dakota N recommendation for reduced tillage systems. The objectives are to: 1) build a corn reduced tillage N recommendation algorithm for South Dakota; and 2) as recommended by the board, assess if similar changes are needed for P and K fertilizers.</p> <p><b>Introduction:</b></p> <p>Over the three-years of this experiment, research has been conducted on over 20 sites to determine the effects of soil health on N recommendations in no-tillage systems. Soil from these sites have been analyzed for plant and soil health characteristics and tissue samples have been analyzed for N and C. The next step in this project is to develop N recommendation algorithms that consider soil health.</p> <p>I believe that the changes in soil health will improve nutrient efficiency for three reasons. First, higher soil organic matter contents and soil residue cover in reduced tillage systems increase precipitation and nutrient uptake efficiency. Second, higher soil organic matter contents and no-tillage leads to increased microbial diversity, which accelerates the movement of nutrients from less to more readily available forms. Third, increased fungal biomass increases the ability to take up both water and soil nutrients. No-tillage should increase fungal concentrations, which by itself should increase nutrient and water uptake.</p> <p>Benefits from a diverse microbial community can be integrated into fertilizer recommendations through multiple mechanisms including creating system recommendations (for example, tillage-based recommendations in North Dakota) or basing the recommendation on changes in a measured soil property (carbon recommendation in Nebraska). We will explore both techniques in the creation of an algorithm that considers how interactions between management and soil biology affects N cycling. We believe that integrating soil health into the N recommendation will improve accuracy and reduce costs.</p>	

---

## Publications

**Publication Title:** none to date  
**Publication Date:** 06/30/2022  
**Status:** none to date  
**Publication Description:** Working on a thesis that will be converted into publications

**NREC – 2022**  
**Building a South Dakota Corn No-tillage N Recommendation Algorithm that Considers  
Improvements in Soil Health**  
**Prepared by David Clay and Dwarika Bhattarai**  
**Report – Work Progress**  
**06/29/22**

**Research goal:**

The goal of this project is to improve South Dakota N recommendation for reduced tillage systems. The objectives are to: 1) build a corn reduced tillage N recommendation algorithm for South Dakota; and 2) as recommended by the board, assess if similar changes are needed for P and K fertilizers.

**Introduction:**

Over the three-years of this experiment, research has been conducted on over 20 sites to determine the effects of soil health on N recommendations in no-tillage systems. Soil from these sites have been analyzed for plant and soil health characteristics and tissue samples have been analyzed for N and C. The next step in this project is to develop N recommendation algorithms that consider soil health.

We believe that the changes in soil health will improve nutrient efficiency for three reasons. First, higher soil organic matter contents and soil residue cover in reduced tillage systems increase precipitation and nutrient uptake efficiency. Second, higher soil organic matter contents and no-tillage leads to increased microbial diversity, which accelerates the movement of nutrients from less to more readily available forms. Third, increased fungal biomass increases the ability to take up both water and soil nutrients. No-tillage should increase fungal concentrations, which by itself should increase nutrient and water uptake.

Benefits from a diverse microbial community can be integrated into fertilizer recommendations through multiple mechanisms including creating system recommendations (for example, tillage-based recommendations in North Dakota) or basing the recommendation on changes in a measured soil property (carbon recommendation in Nebraska). We will explore both techniques in the creation of an algorithm that considers how interactions between management and soil biology affects N cycling. We believe that integrating soil health into the N recommendation will improve accuracy and reduce costs.

**Site description and experimental design:**

In years 1, 2, and 3 experiments were conducted at seven sites each year. Table 1 shows the list of farmers who provided their field to conduct our experiment. At this point, all sites have been harvested from year 3 and yields have been determined. Soil and plant samples are in the process of analysis. As discussed, these studies were conducted in a wide range of conditions, we will conduct a rigorous analysis of these data sets. The findings will be shared annually with the individual producers, neighbors, and agronomists. Blogs will be prepared and distributed through social media and non-technical guides published in the South Dakota Corn Best Management Practice Manual. The potential impacts include improved profitability resulting from more accurate recommendations, and findings that will determine if P and K recommendation warrant similar modifications.

**Table 1.** List of different farmer's field, their location, and year of experiment.

SN	Farmer's name	# Of field	Years of experiment	Location
1	Bryan Jorgensen	3	2019-2021	Ideal
2	Bob Speck	2	2019 and 2021	St. Lawrence
3	Dan Forgey	4	2019-2021 (2 field in 2021)	Gettysburg
4	Dennis Hoyle	2	2019-2020	Roscoe
5	Dakota Lakes Dryland	3	2019-2021	Pierre
6	Dakota Lakes Irrigated	3	2019-2021	Pierre
7	Scott Carlson	3	2019-2021	Badger

The experiment has six different N treatments (0, 40, 80, 120, 160 and 200 lbs. N acre<sup>-1</sup>), only four treatments in 2019, replicated four times and arranged in RCB design. Urea (46:0:0) fertilizer (treated with urease inhibitor) were broadcasted as the source of N before corn V4 growth stage. Each site was applied with recommended rate of P and K fertilizers based on soil test results.

Soil samples from four depths (0-2", 2-6", 6-12", and 12-24") were collected from each block prior to the application of fertilizers and from each plot after corn harvest. These samples were analyzed for 24-hr microbial respiration (0-2"), soil nitrate-N and ammonium-N (all depths), soil pH and EC (all depths), total N and total C analysis (all depths). In addition, soil samples (0-2") were taken from each block before planting corn and from each plot at various corn growth stages for phospholipid fatty acid (PLFA) analysis. The respiration test was performed using Solvita test kit. Soil nitrate-N and ammonium-N is extracted with 1M KCl (10:1). Soil PLFA is extracted following a modified Buyer and Sasser (2012) protocol.

#### **Climatic information:**

Across the years, weather was never similar. The first year of our experiment, 2019, was a wet year followed by a normal wet year in 2020. The last year, 2021, was extremely dry year and resulted in poor corn yield in several locations. According to Applied Climate Information System (ACIS), in 2021, the cumulative rainfall during the corn growing season (May-October) ranged from around 4 inches to 16 inches across sites; precipitation during September and October covers more than 50% of the total rainfall during the season.

#### **Preliminary N analysis on corn yield**

Corn yield and corn N response across the experimental sites and years were different that might be because of various locations and weather conditions across years. For example, corn yield in 2020 ranged between 61 to 282 bu a<sup>-1</sup> whereas in 2021 the range was between 41 and 236 bu a<sup>-1</sup>. This information will be re-analyzed using strong machine learning algorithms to determine the corn N response.

At this point, we are reviewing several articles to get familiar with different machine learning approaches that were used in similar research and using different sets of predicting variables. We will publish the reviewed article in a renowned journal and move forward from there to analyze our data using machine learning approaches.

## Analysis between measured values.

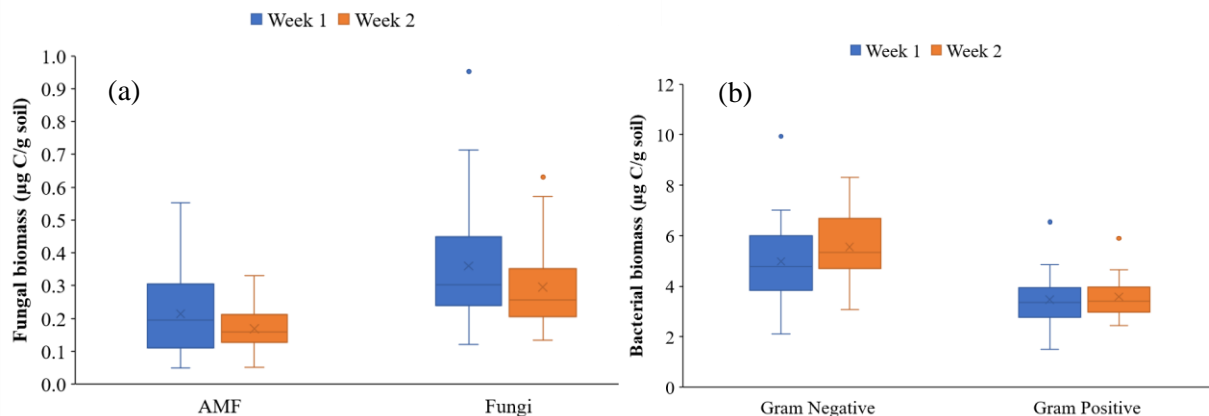
Initial analysis of data collected in 2019, which was a high rainfall fall year showed that infiltration rate was very important (Table 2). Site with high infiltration had high yields. This data set also showed that sites with high electrical conductivity had low yields. The high correlation between yields and water infiltration during the high rainfall year was expected. It will be interesting to compare these results with different climatic conditions.

**Table 2.** Correlation coefficients between the different measurement

	Microbial biomass				Water	Solventa	
	NO <sub>3</sub> -N	NH <sub>4</sub> -N	pH	EC	infiltration	CO <sub>2</sub>	
Yield	-0.23	-0.15	-0.23	-0.55	-0.67	0.82	-0.37

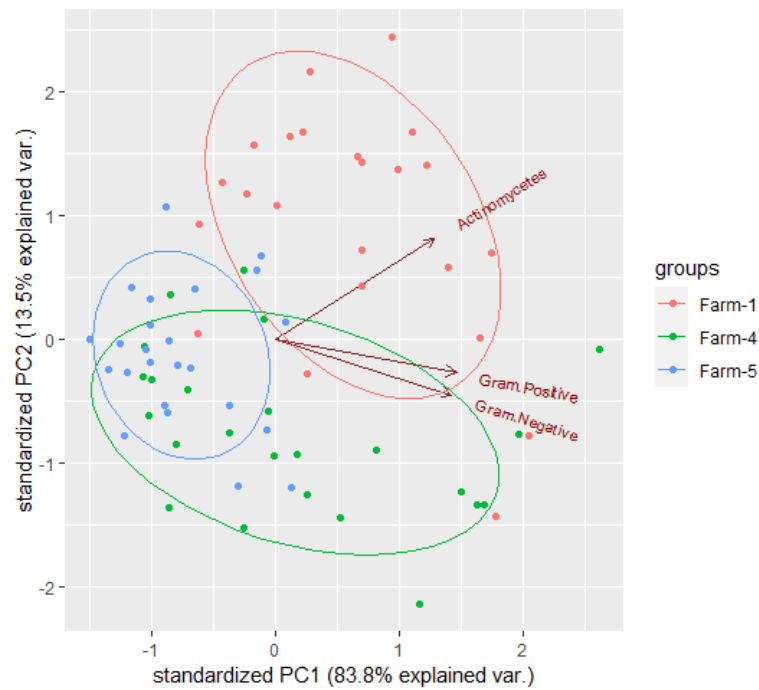
## Update on microbial community analysis

Soil microbial communities are important components of healthy soil because different microbes perform different tasks. A comparison on spring 2021 soil microbial biomass data from consecutive weeks (last week of April and first week of May) showed reduced fungal and mycorrhizal biomass in the later week whereas the bacterial biomass slightly increased (Figure 1). This implies that as the soil temperature increases, bacterial population starts increasing and thus improves mineralization. Fungi are known to improve nutrients uptake through their hyphal network and are favored at lower temperatures.

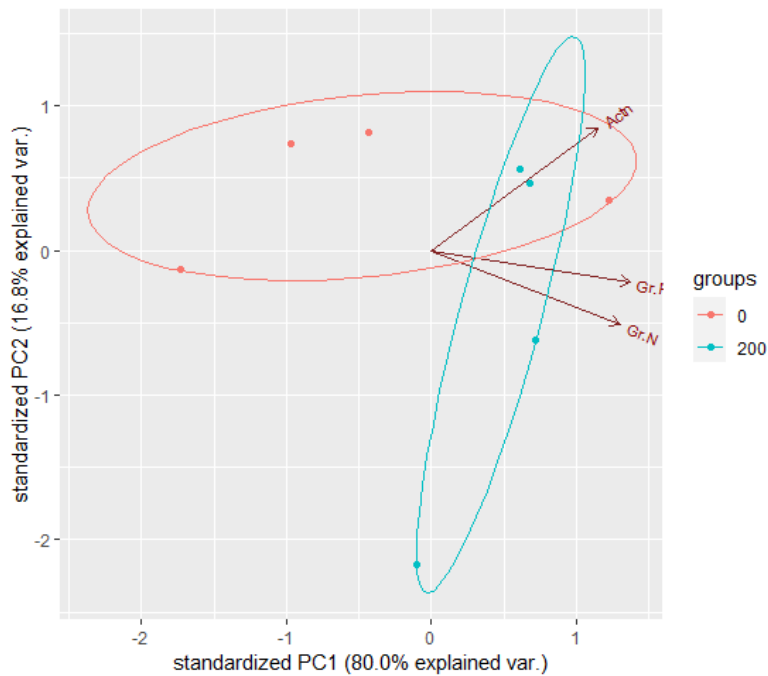


**Figure 1.** Boxplots showing the biomass of (a) Arbuscular Mycorrhizal Fungi (AMF), and Fungi, and (b) gram-positive and gram-negative bacteria (µg C/g soil) from the experiment sites in Spring 2021 before planting corn. In the figure, blue box represents the first sampling week (last week of April), and the orange box represents the second sampling week (first week of May). The data is averaged across experimental sites.

In addition, we have analyzed the soil samples collected before and after the application of fertilizer until harvest. We will compare spatial, temporal, and treatment difference in the soil microbial data. Figure 2 shows the variation across three farms in the biomass of gram-positive bacteria, gram-negative bacteria, and actinomycetes from the samples collected after fertilizer application. All three farms have similar soil type with varying weather and management practices.



**Figure 2.** Principal Component Analysis (PCA) showing the variation of soil bacteria and actinomycetes across three different farms from the samples collected two weeks after fertilizer application in 2021.



**Figure 3.** Principal Component Analysis (PCA) showing the variation of soil bacteria and actinomycetes between control and 200 lbs N a<sup>-1</sup> in Farm-1 from the samples collected two weeks after fertilizer application in 2021.

Figure 3 shows the variation in the biomass of bacteria and actinomycetes between control (0 lb N a<sup>-1</sup>) and 200 lbs N a<sup>-1</sup> applied plots in Farm-1. Due to the variation across sites, the analysis of soil microbial data in response to fertilizer applied and averaged across sites was not helpful to determine the impact of fertilizers on soil microbes. Management practices and weather pattern could be more strong predictors of soil microbial biomass.

We have extracted soil DNA from one of our baseline samples from spring 2021. We have used DNA extraction kits from MP Bio company (Cat.NO.116560200, MP Biomedicals, Solon, Ohio). Microbial biomass determined using PLFA method had fair positive correlation (0.29) with the DNA yield. However, the DNA yield was highly correlated with the biomass of AMF (0.57) and fungi (0.4) (Figure 2).



**Figure 2.** Correlation matrix showing the correlation coefficients across various soil microbial biomass and soil DNA yield.

Our previous results from Figure 1 have shown that the fungal biomass is higher in spring samples, which could have resulted in greater DNA from fungi and AMF. This information gives insight into determining the gene expression from the extracted DNA that will determine the proportion of different genes.

Further, we have been working on quantifying the relative amount of nitrifying and denitrifying genes across the sites using qPCR analysis. We conducted the analysis for all sites; however, we were not able to obtain accurate results. We are, currently, working on troubleshooting the issue by optimizing the analysis protocol in the lab.

## Summary



In this project, research was conducted for 3 site years with 7 sites each year. Each site year has very different soil and climatic conditions. The laboratory analysis has been completed for the first two years and we are towards the end in the analysis for year 3. We have found differences in corn yield and corn N response across sites. We performed preliminary data analysis from the first two years of data, which showed that soil health had a large effect on corn yields. We have extracted soil DNA from baseline samples, in addition to the PLFA analysis. Analyzing gene expression might make it easier to interpret the microbial activities; we analyzed them once but need to optimize the protocol as the results obtained from the first run were not accurate. By the end of this year, our goal is to complete this analysis and publish the findings at appropriate locations.