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Modeling Soil Organic Carbon in Select Soils of Southeastern South Dakota

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MODELING SOIL ORGANIC CARBON IN

SELECT SOILS OF SOUTHEASTERN SOUTH DAKOTA

BY

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

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This thesis is dedicated to my son, Little Paul.

I hope you are always curious, kind, and nurture a good sense of humor. So far, so good.

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ABBREVIATIONS

α	Alpha: Pertains to the Confidence Interval Set for the Study
°C	Degrees Centigrade
δC	Change in Carbon
µg	Microgram (0.001mg)
µm	Micrometer (0.001mm)
1-L	One-Liter
1:1 clay	One Tetrahedral Layer to One Octahedral Layer per Clay Mineral (e.g. kaolinite)
2:1 clay	Two Tetrahedral Layers to One Octahedral Layers per Clay Mineral (e.g. smectite)
AB	Five-Year Rotation with One Year of Hay or Root Crop
AeroGRID	European Satellite Imagery Database
AF	Five-Year Rotation with Two Years of Fallow
ANOVA	Analysis of Variance
BBS	Back Backslope
BCE	Before the Common Era
BF	Base Factor
BFS	Back Footslope
BMP's	Best Management Practices
BS	Backslope
C	Carbon
C:N	Carbon to Nitrogen Ratio
^{12}C	Natural Carbon
^{13}C	Radioactive Carbon Isotope
^{13}C‰	Parts per Thousand of Radioactive Carbon
CA	California

CEC	Cation Exchange Capacity
CENTURY	Plant-Soil Nutrient Cycling Model Produced by Colorado State Univeristy
CF	Chinese Fir Plantation
cm	Centimeter
CO_2	Carbon Dioxide
COMET-Farm	Carbon Footprint Model for Industrial Operations
CON	Continuous Corn
cPOM	Coarse Particulate Organic Matter
CRP	Conservation Reserve Program
CT	Conventional Tillage
DayCENT	Submodel of the CENTURY Model which Operates on a Daily Timescale
DI	Deionized
DIC	Dissolved Inorganic Carbon
DNA	Deoxyribonucleic Acid
DOC	Dissolved Organic Carbon
DRFIT	Diffuse Reflectance Infrared Fourier Transform spectroscopy
EC	Electrical Conductivity
ESRI	Environmental Systems Research Institute- Satellite Imagery Software Developers
e.g.	For Example
et. al.	And Others
FAME	Fatty Acid Methyl Ester
FL	Farm Land
FLF	Free Light Fraction
fPOM	Fine Particulate Organic Matter
FS	Footslope
FYM	Farm-Yard Manure

g	Gram
GEOeye	Supplier of Satellite Imagery
GIS	Geographic Information System
H	Climate Region
H, h	Land Use and Management Identification
ha, ha^{-1}	Hectare, Per Hectare
HCl	Hydrochloric Acid
HF	Minimal Associated Heavy Fraction
HT	Heated Weight
i.e	In OtherWords
IC	Inorganic Carbon
IF	Input Factor
IG	Ignition Weight
IGN	French National Geographic Institute
IPCC	Intergovernmental Panel on Climate Change
K	Potassium
kg, kg^{-1}	Kilogram, Per Kilogram
km	Kilometer
LA	Land Area
LBS	Lower Backslope
LC3	Three-Year Grass-Clover Rotation
LC8	Eight-Year Grass-Clover Rotation
Ley	Perennial Crop
LFOC	Light Fraction Organic Carbon
LN3	Three-Year Grass with Nitrogen Fertilizer
LN8	Eight-Year Grass with Nitrogen Fertilizer

LOC	Labile Soil Carbon
LSD	Least Significant Difference
Lu	Alfalfa
m, m^2	Meter, Square Meter
MBC	Microbial Biomass Carbon
Mg	Megagram
MID	Mid-Intensity Rotation
MinOM	Mineral Organic Matter
mL	Milliliter
mm	Millimeter
MLR	Multiple Linear Regression
MLRA	Major Land Resource Area
mm	Millimeter
MNDA	Minnesota Department of Agriculture
MWD	Mean Weight Diameter
N	Normality
°N	Degrees North
N, ^{14}N	Nitrogen
^{15}N	Radioactive Nitrogen Isotope
NF	Native Forest
NO_3^{-1}-N	Nitrate Nitrogen
NPK	Nitrogen, Phosphorus, Potassium
NPP	Net Primary Production
NRCS	Natural Resources Conservation Service
NT	No-Till
NTVG	Native Grass, Native Vegetation

NW	Native Woodlots
OH	Ohio, USA
OLF	Occluded Light Fraction
OM	Organic Matter
p	*p*-value: Probability-value
P	Phosphorus
PET	Potential Evapotranspiration
PFLA	Phospholipid Fatty Acid
Pg	Petagram
pH	Soil Acidity
PM	*P. massoniana* plantaion
POC	Particulate Organic Carbon
POM	Particulate Organic Matter
ppm	Parts Per Million
Q_{10}	Soil Organic Matter Reaction to Temperature
QIIME	Quantitative Insights into Microbial Ecology
r^2	Coefficient of Determination
R^2	Multiple Correlation Coefficient
RC	Reference Carbon Stock
RCP(2.6; 8.5)	Representative Concentration Pathway
RDF	Recommended Dose of Fertilizer
RothC	Carbon Cycling Model Created by Rothamsted Research, United Kingdom
RP	*Robinia pseudoacacia* L.
RSE	Reference Surface Elevation fo Soil
S	Sulfur
SCS	Soil Conservation Service

SEM	Structural Equation Modeling
SH	Shoulder Hillslope Position
SHB	Shoulder Backslope
SIC	Soil Inorganic Carbon
SM	Summit
SOC	Soil Organic Carbon
SOM	Soil Organic Matter
SPADE2	Soil Profile Analytical Database for Europe
SYI	Sustainable Yield Index
t, t a^{-1}, t ha^{-1}	Ton, Ton per Acre, Ton per Hectare
TC	Total Carbon
TC:TN	Total Carbon to Total Nitrogen Ratio
TF	Tillage Factor
TK	Total Potassium
TN	Total Nitrogen
TOC	Total Organic Carbon
TP	Total Phosphorus
TS	Toeslope
UBS	Upper Backslope
USA	United States of America
USDA	United States Department of Agriculture
USGS	United States Geological Survey
°W	Degrees West
WF	Wheat-Fallow Rotation
yr, yr^{-1}	Year, Per Year

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ABSTRACT

MODELING SOIL ORGANIC CARBON IN
SELECT SOILS OF SOUTHEASTERN SOUTH DAKOTA

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Soil organic matter (SOM) is composed of living biomass, dead plant and animal residues, and humus. Humus is a class of complex, organic molecules that are largely responsible for improving soil water holding capacity, nutrient mineralization, nutrient storage, and other critical soil functions. Soil organic carbon (SOC) accounts for approximately 60 percent of SOM and thus SOC is recognized as a strong indicator of soil health. Land use changes and intense cultivation of arable soils in the United States over the past century have led to large decreases in SOM.

The objective of this research was to develop a multiple linear regression model to predict SOC levels in select southeastern South Dakota soils and the region. Conventional Till (CT), No-Till (NT), and Native Grass (NTVG) management systems were studied within South Dakota Major Land Resource Area 102B, 102C, and McCook County, South Dakota. It was hypothesized that NTVG treatments would have the highest SOC levels, followed by NT treatments, and CT treatments would have the least. Samples were analyzed for pH, electrical conductivity (EC), total nitrogen (TN), total carbon (TC), SOM, soil inorganic carbon (SIC), particle size, color, and water stable aggregates.

Management was found to have a significant effect on soil pH, EC, TN, TC, and SOC compared to native conditions ($p<0.05$). Multiple linear regression (MLR) was used

to build the full SOC prediction model which was then reduced using stepwise selection in R 3.5.0. The final reduced model that was produced by stepwise selection is defined as SOC = 3.25 - 0.811(Conventional Tillage) - 0.939(No-Tillage) - 0.548(10-20 Depth) - 0.918(20-40cm Depth) + 0.0396(Moisture) - 0.288(Temperature). Although this model did not result in an acceptable Shapiro-Wilk *p*-value, the model did not have multicollinearity issues, and approximately 67% of the variation in SOC was explained by the model.

To create a model that includes all management variables, filtering the data set to include only specific data points before running MLR analysis is an option. One proposed filtered model incorporates No-Till management and Corn-Soybean rotation data points. The resulting filtered model is defined as SOC = -0.0885 - 0.473(10-20cm Depth) - 0.082(20-40cm Depth) + 0.067(Moisture) - 0.267(Temperature) + 0.156(pH). This model produced an acceptable Shapiro-Wilk *p*-value (p=0.969), displayed approximately normal residuals, and did not exhibit multicollinearity. Approximately 64% of the variation in SOC was explained by the model. Based upon these results, filtering the data set is an appropriate method for data analysis and model construction. Developing an electronic application for use via website or mobile device as a means of sharing this information with producers is a viable option.

Additional data is needed to improve the models, to meet the assumptions of multiple linear regression when using stepwise selection, and to increase the applicability of the model to producers in southeastern South Dakota and the region before the electronic application is constructed. Furthermore, more data points are needed to validate the proposed models.

CHAPTER 1: INTRODUCTION AND LITERATURE REVIEW

Introduction:

Soil is a complex medium that is responsible for the anchoring of plants, storage of water and nutrients, support of soil microbial life, and many other tasks that are essential to crop production (Brady and Weil, 2017). The health of the soil is critical to successful and sustainable crop production. Soil health is, “the capacity of soil to function as a vital living system, within ecosystem and land-use boundaries, to sustain plant and animal productivity, maintain or enhance water and air quality, and promote plant and animal health” (Doran and Zeiss, 2000). Physical and chemical properties, such as bulk density, aggregate stability, electrical conductivity (EC), soil organic matter (SOM), and soil organic carbon (SOC) serve as indicators of soil health (NRCS, 2015). Soil organic matter is a fundamental component of the soil matrix in terms of water holding capacity, cation exchange capacity (CEC), nutrient storage, microbial health and other primary soil functions (Brady and Weil, 2017). Aside from living biomass and decaying plant and animal tissues (Fenton, et.al, 2008), SOM is composed of humus; a complex, organic molecule that is largely responsible for the critical soil functions mentioned above (deHaan, 1977). Nearly 60 percent of humus is composed of SOC and is a strong indicator of soil health (Soil Survey Staff, 1999).

A brief tour through human history solidifies the importance of soil health to the security of human civilization. Salinization of arable ground from salt-laden irrigation water reduced Sumerian harvests to one third of original production between 3000 Before the Common Era (B.C.E.). and 1800 B.C.E. (Montgomery, 2007). Plato of Ancient

Greece wrote, “The rich, soft soil has all run away leaving the land nothing but skin and bone” concerning the effect of erosion on Greece’s soil (Montgomery, 2007). Man-induced erosion has worn the Jordan Valley, a once fertile agricultural center in the Middle East, down to bedrock (Lowdermilk, 1948). The Dust Bowl in the American Great Plains during the late 1800’s through the 1930’s was another historical period of soil degradation as intensive cultivation of fine-textured soils and removal of perennial vegetation severely damaged soil quality (Montgomery, 2007). It is estimated that this period in U.S. history led to a 40-50% reduction in SOM (Clay et. al, 2017). The Dust Bowl led to the creation of the Soil Conservation Service (SCS) that has since become the Natural Resources Conservation Service (NRCS) (Helms, 1992). Events like the Dust Bowl have shaped our nation and serve as powerful reminders of the consequences of poor land stewardship.

Soil is a finite resource and yet 2.7 tons per acre per year ($t\ a^{-1}\ yr^{-1}$) of soil are lost in the United States (Nearing et. al., 2017). Land use change from native prairie to row crop can result in a 90% reduction in labile SOC within a century of cultivation (Brady and Weil, 2017). An estimated 20-43% reduction in SOC has been witnessed around the world as native forest is converted to farm ground (Wei et. al., 2014b). This literature review will address the role of SOC in the global carbon cycle, abiotic factors and agricultural management systems that impact SOC levels, and offer a brief synopsis of the parameters and applications of select current SOC models.

Global Carbon Cycle:

Earth's carbon (C) exists in three primary reservoirs; aquatic ecosystems, terrestrial ecosystems, and the atmosphere (Post et. al., 1990). Carbon is constantly in flux between these reservoirs due to additions and losses from the overall carbon system. Of the primary C reservoirs, oceans serve as the largest sink for C in the form of dissolved inorganic carbon (DIC), dissolved organic carbon (DOC), and particulate organic carbon (POC) (Post et. al., 1990). Oceans and lakes store 40,000 petagrams (Pg) of carbon (Brady and Weil, 2017). Terrestrial ecosystems serve as the second largest C sink at almost 3,000 Pg of C storage between Earth's vegetation and soil (Brady and Weil, 2017). Above ground vegetation is responsible for storing 550 Pg of C yr^{-1} while soils store 2,450 Pg C yr^{-1} (Brady and Weil, 2017). The atmosphere historically holds the least amount of carbon at 760 Pg of C (Brady and Weil, 2017). Burning of fossil fuels, land use changes, and increasing human populations around the globe have led to loss of equilibrium in the C cycle by disproportionately adding carbon dioxide (CO_2) to the atmosphere (Lal et. al., 1998). In 2017, it was estimated that there was 404 parts per million (ppm) of CO_2 in Earth's atmosphere as compared to the first reading of 315ppm CO_2 at the Mauna Loa Research Facility in Hawaii in 1958 (NOAA, 2017). Restoring soil organic carbon (SOC) in degraded soils by 0.01% has the potential to sequester the same amount of CO_2 in the soil as is released annually to the atmosphere (Lal et. al., 1998).

Soil Organic Carbon

Soil organic matter can be divided into three fractions; coarse particulate organic matter (cPOM), fine particulate organic matter (fPOM), and mineral associated organic matter (MinOM) (Benbi et.al, 2014). Each fraction of SOM is composed of different materials and represents different pools of C. Coarse POM consists of labile C in the form of living biomass, plant litter/residues, and organic biomolecules (Clay et. al., 2017; Benbi et. al., 2014). This pool is subject to change quickly with new additions or losses of organic materials into the soil (Clark et. al, 2017). Fine fPOM is associated with the slow C pool, which has a turnover rate of years to decades (Clay et. al., 2017; Benbi et. al., 2014). Humus is a stabilized material which makes up MinOM, or the recalcitrant C pool, and is composed of highly decomposed and no longer identifiable tissues as well as biomolecule conglomerates (Benbi et. al., 2014; Brady and Weil, 2017). Humus is highly resistant to change over short periods of time and persists in the soil for hundreds to thousands of years (Clay et. al, 2017). Soil organic matter pools are influenced by additions and losses of SOC. Biomass removal, erosion, soil respiration, and mineralization are all modes of C transport from the soil matrix (Rumpel et. al., 2015). Above and belowground biomass production and organic amendments serve as C additions to the soil matrix (Curtin, 2012; Rumpel et. al., 2015).

Losses of Soil Organic Carbon:

Predominant losses of SOC include crop removal, erosion, and soil respiration (Rumpel et. al., 2015). Crop removal directly impacts SOC levels by reducing aboveground biomass available for C cycling due to the harvesting of grain, silage, and/or stover (Wilhelm et. al., 2004). Erosion is another direct loss of SOC with serious ramifications in many areas of soil health. In the United States, erosion accounts for the loss of 1.67 billion tons of soil each year (NRCS, 2015). Soil respiration and SOM mineralization account for large fluxes of CO_2 back to the atmosphere (Brady and Weil, 2017). Soil respiration is the process by which microbial, plant root, and mycorrhizal organisms decompose organic materials while emitting CO_2 (Brady and Weil, 2017). This decomposition of organic materials leads to the mineralization of inorganic nutrients from complex organic compounds (Brady and Weil, 2017). Soil respiration is estimated to attribute 75 Pg C yr^{-1} back into the atmosphere (Schelsinger and Andrews, 2000). Soil respiration accounts for a larger flux of C into the atmosphere than net primary production (NPP) sinks into the soil, because of CO_2 production from microbial and root respiration (Rumpel et. al., 2015; Schelsinger and Andrews, 2000). Various environmental factors, such as soil temperature, soil moisture, SOC content, and root density impact the rate of soil respiration (Guo et. al., 2016).

Guo et. al (2016) compared soil respiration rates across three forest treatments; 200-year old native forest (NF), 36-year old Chinese fir (*Cunninghamia lanceolata*) (CF) plantation, and 36-year old *P. massoniana* (PM) plantation. Microbial biomass carbon (MBC), pH, texture, SOC, nitrogen (N), and phosphorus (P) were measured in the top 0-10 cm. Soil respiration was measured biweekly from October 2010 to September 2012. Soil organic carbon, total nitrogen (TN), and MBC were significantly higher in the NF treatment than the CF or PM. The NF treatment additionally had significantly more litter fall than either plantation treatment. Soil respiration was highly correlated to SOC (R^2= 0.918) which led to significantly higher levels of soil respiration in the NF treatment than in the plantation treatments (Guo et. al., 2016). The higher levels of SOC and fine root biomass in the NF treatment were responsible for increased soil respiration rates (Guo et. al., 2016). These results indicate that SOC serves as a fuel for soil respiration and for the release of CO_2 from the soil matrix.

Additions to Soil Organic Carbon:

Primarily, plant material (litter) is responsible for the return of organic matter (OM) to soil and to the formation of SOM (Kogel-Knabner, 2002). Litter composition is important to microbial decomposition, mineralization, and formation of SOM (Rumpel et. al, 2015). The carbon to nitrogen ratio (C:N) is a critical component of the decomposition formula. Infertile, coniferous forest soils have three times the amount of C on the forest floor as compared to fertile, deciduous forest floors (Buol et. al., 2011). High C levels in the surface litter cause microbes that are responsible for organic matter (OM) decomposition to use all available nutrients (nitrogen (N), phosphorus (P), sulfur (S), etc.) in the soil to complete the decomposition process. This is called the “priming effect” and can lead to microbial mineralization of stable humus products (Fontaine et. al., 2011). This impacts the amount of C that can be transferred to SOC (Kirkby et. al., 2014). Additionally, management of the soil resource has a large impact on additions of C to the soil.

Factors that Impact Soil Organic Carbon:

A wide range of abiotic and biotic factors affect the amount of SOC that accumulates in the soil within a given time frame and location. Warm, wet environments have the potential to sequester more C than dry climates, but are also more susceptible to

C losses than more temperate regions (Ogle et. al., 2005). Sandy soils on steep slopes cannot accumulate the same levels of SOC as finer textured soils in footslope (FS) positions (Aguilar and Heil, 1988; VandenBygaart, 2016). Microbial activity defines soil respiration rates and microbial decomposition adds to SOM (Miltner et. al, 2012; Guo et. al., 2016). Land use change from native conditions to cultivated systems results in significant decreases in SOM and SOC (Cambardella and Elliott, 1992; Wei et. al., 2014a). Agriculture has vast impacts on the soil system as a whole, and different management practices such as residue removal, rotation, fertilizer amendments, and tillage impact SOC storage (Janzen et. al., 1992).

Temperature and Moisture:

Temperature affects organic matter fractions to different degrees. The impact of increasing temperature on cPOM, fPOM, and MinOM fractions was measured with varying organic matter inputs in a rice (*Oryza sativa*)-wheat (*Triticum aestivum*) rotation (Benbi et. al., 2014). Plots either received annual inputs of rice straw and NPK (nitrogen, phosphorus, potassium) fertilizer, solely NPK fertilizer, farmyard manure, or no organic inputs. Surface samples were collected from the 0-15cm depth after the wheat crop was harvested. Samples were sieved to determine the cPOM, fPOM, and MinPOM fractions. Microbial biomass carbon was measured on non-sieved samples via incubation and chloroform fumigation extraction. The influence of temperature on mineralization was assessed for both sieved and un-sieved samples by estimating mineralization coefficients at 15°C, 25°C, 35°C, and 45°C and using the Q_{10} function [SOM sensitivity to temperature (Fang et. al., 2005)] in five different models. Data on mean C stocks and mineralization rates by temperature were subjected to analysis of variance (ANOVA) (Freund et. al., 2010).

Results showed that cPOM (labile C) composed the smallest portion of whole soil samples while MinOM (stable C) composed the largest portion. Although cPOM made up the smallest fraction of organic matter, the most C was mineralized from this fraction regardless of temperature. At all temperature increments, MinOM appeared to be the least responsive to temperature changes when compared to cPOM and fPOM. The authors concluded that while cPOM composes the smallest portion of whole soil, cPOM is responsible for the largest amount of C mineralization in the soil. Increases in temperature had a significant effect on cPOM decomposition. Mineral associated organic matter accounts for the largest portion C in the soil system, but is the least susceptible to decomposition. Temperature increases did not have a significant effect on MinOM decomposition. Results such as this indicate that temperature plays a critical role in decomposition of the labile C pool, but the stabilized C pool is relatively resilient to temperature increases (Benbi et. al., 2014).

Climate is an important factor in SOC dynamics. The effect of various land management scenarios on SOC in different climate zones was studied via meta-analysis of existing data (Ogle et. al., 2005). Researchers hypothesized that long-term cultivation, tillage, cropping intensity, residue production, organic amendments, and set-aside land (such as the NRCS Conservation Reserve Program (CRP)) would all have significant impacts on SOC in tropical and temperate climates with moist and dry moisture regimes. The Intergovernmental Panel on Climate Change (IPCC) method (Equations 1.1 and 1.2) was used on 126 data sets pertaining to the above parameters to analyze changes in SOC levels over a minimum 20-year time frame. The change in C (δC) as defined by the IPCC model is the sum of SOC in last year of the data set (SOC_1(h)) minus the SOC in the first year of the data set (SOC_{1-20}(h)) by climate region (H). SOC(h) is calculated by multiplying the Reference Carbon Stock (RC), Base Factor (BF), Tillage Factor (TF), Input Factor (IF), and Land Area (LA) (see Equation 1.2). Reference carbon stocks represent carbon stocks under native conditions, BF values estimate the change in SOC from native after long-term cultivation, TF values estimate the effect of tillage on SOC stocks, IF values estimate the effect of cropping intensity/input levels on SOC stock, and Land Area (LA) represents the area for a particular land use and management (h).

Eq 1.1: $\delta C = \sum_{h=1}^{H} \left(SOC_1(h) - SOC_{1-20}(h)\right)$ (IPCC, 1997)

Eq 1.2: $SOC(h) = RC * BF * TF * IF * LA$ (IPCC, 1997)

The data sets were divided by climate region and moisture regime in which Tropical sites were defined as areas with mean annual temperature greater than 20°C (Ogle et. al., 2005). Sites were defined as Temperate if the mean annual temperature was less than 20°C (Ogle et. al., 2005). Moist Tropical sites were defined as locations where the mean annual rainfall was greater than 1000mm and Dry were locations where mean annual rainfall was less than 1000mm (Ogle et. al., 2005). To define Moist and Dry temperate sites, the precipitation to potential evapotranspiration (PET) ratio (P:PET) was used. Ratios greater than one were defined as Moist and ratios less than one were defined as Dry (Ogle et. al., 2005). Linear mixed-effect models were implemented to incorporate fixed and random effects into the response variable. The response variable was the ratio of SOC per management parameter to SOC level in native conditions (BF) (Ogle et. al., 2005). All data sets were limited to the top 30cm soil depth.

From the 126 data sets gathered, 80 pertained to tillage, 31 pertained to rotation and cropping intensity, 30 pertained to long-term cultivation, and 17 pertained to land restoration. The largest changes in SOC were seen in Tropical, Moist regions and the smallest changes were seen in Temperate, Dry regions. Long-term cultivation caused the largest decrease in Tropical, Moist locations with 58% ± 12% retention in SOC compared to native conditions (Ogle et. al., 2005). The Temperate, Dry locations were found to have 82% ± 4% retention compared to native conditions (Ogle et. al., 2005). These results indicate that only 58% of the native SOC was still present under long-term cultivation management in Tropical, Moist climates whereas Temperate, Dry climates retained 82% of the native SOC (Ogle et. al., 2005). Implementing no-till from conventional tillage resulted in a positive increase in SOC by a factor of 1.10 ± 0.03 (equal to a 10% increase in SOC) in Temperate, Dry regions and a factor of 1.23 ± 0.05 in Tropical, Moist regions (Ogle et. al., 2005). Increasing the cropping intensity by using higher yielding varieties and/or cover crops led to an increase in SOC with a factor of 1.07 ± 0.05 in dry climates and 1.11 ± 0.05 in moist climates (Ogle et. al., 2005). Additions of organic amendments in high intensity systems led to an increase in SOC with a factor of 1.34 ± 0.08 in dry climates and 1.38 ± 0.06 in moist climates.

The data suggests that temperature and moisture play a large role in the reduction or accumulation of SOC across multiple management practices. Tropical, moist regions in this study appeared to decline in SOC levels more rapidly with long-term cultivation than did Temperate, Dry regions (Ogle, et. al, 2005). The authors conclude that, while

Tropical, Moist climates accumulate more SOC, they are also more susceptible to SOM decomposition than Temperate, Dry regions due to changes in management (Ogle et. al., 2005).

<u>Texture, Soil Taxonomy, and Topography:</u>

Texture, soil classification, and topographic location are related to SOC levels in the soil landscape. Coarse and fine textured soils affect SOC in different ways. Chemical protection is the chemical binding of SOM to silt and clay particles (Six et. al., 2002). Soil clay content influences C concentration of a soil as the structure of clay is composed of negatively charged binding sites which provide protection for SOM (VandenBygaart, 2016). Texture is the limiting factor for C accumulation in fine-textured soils (e.g.: clay, silty clay, sandy clay, etc.) whereas organic inputs are the limiting factor for C accumulation in coarse-textured soils (e.g.: sand, loamy sand, etc.) (Hassink, 1997, Johnston et. al., 2017).

Six et. al. (2002) analyzed the influence of chemical protection, physical protection, and biochemical stabilization on SOC content within SOM. Existing surface soil data (0-10cm) was compiled from cultivated, grassland, and forest ecosystem studies around the globe. Two soil particle size ranges were analyzed within these studies; clay (0-20µm) and clay with silt (0-50µm). Regressions between C associated with clay and silt (g C kg^{-1} soil) and proportion of silt and clay in the soil (g of silt g^{-1} of soil and g of clay g^{-1} of soil) were performed for all ecosystems and particle sizes. Six et. al. (2002) found, like Hassink (1997), that all regressions were significant indicating a strong positive relationship between SOC and silt and clay content. However, Six et al. also found that the y-intercept for the 0-50µm silt and clay size fraction was significantly higher than the 0-20µm size fraction. Greater levels of C were associated with the 0-50µm size fraction due to larger aggregates binding together and trapping more C (Six et.al., 2002). The authors went further to divide the data into 1:1 and 2:1 clays to assess any potential significance from chemical composition of clay on C content. The data indicated significantly lower C stabilization in 1:1 clays (e.g. kaolinite) when compared to 2:1 clays (e.g. smectite). While there are distinct differences in cation exchange capacity (CEC) and surface area between 1:1 and 2:1 clays, the authors also noted that most of the 1:1 data was collected in tropical or subtropical regions in which temperature and moisture would accelerate the decomposition of SOM (Six et. al., 2002). In summary, Six et. al found that there was a direct link between SOC content and the combined silt/clay fraction of the soil. They postulated that this information could be used to determine the point at which soils would be saturated with C and unable to store more.

VandenBygaart (2016) tested the hypothesis that the C saturation point in a degraded soil could be calculated by relating clay content to SOC. A total of 433 data sets from well-drained Ap horizons under numerous management systems, landscape positions, textures, and soil types in southern Ontario, Canada were analyzed for C content and clay content (0-20µm). The data set did not include silt information so the statistical analysis was limited to clay. A third of the data sets used the Walkley-Black method to measure SOC while the remaining two thirds used dry combustion. To account for the differences in SOC values, VandenBygaart (2016) multiplied Walkley-Black values by a conversion factor of 1.25 to equate them to the dry combustion method.

Upper boundary analysis was done by dividing clay levels (the independent variable) into increments of 50g clay kg^{-1} soil and fitting the maximum values of C (response variable) within the increments. The upper boundary, or C saturation point, was determined via regression analysis.

From the available data, VandenBygaart (2016) found that C saturation for soils with less than 19% clay is approximately 3.5% SOC. In soils with greater than 19% clay, C saturation is texture dependent. The author acknowledges that although this research shows 3.5% SOC as the upper boundary for soils with less than 19% clay, management of sandy and low-clay soils is important for building of SOM and SOC (VandenBygaart, 2016). Organic amendments can add SOM to the soil profile but without the clay and silt particles to aid in chemical protection, the SOM from organic amendments will be highly susceptible to decomposition (VandenBygaart, 2016). Additionally, VandenBygaart (2016) mentions that the potential of high clay soils to hold large amounts of C may not be realized due to the nature of elevated clay content restricting plant growth and limiting C inputs.

The world's soils are amazingly diverse and are products of the region and parent material from which they were formed. Soil Taxonomy is the system that soil scientists in the United States use to classify soil relationships as well as the processes that formed them (Soil Survey Staff, 1999). Work done by Westin and Buntley (1967) illustrate the diversity of soils within a geographic region, and the effect of taxonomy on soil properties. Mean annual soil temperature in South Dakota is approximately 8.3°C along the northern edge of Kingsbury and Brookings Counties. Frigid soil temperature regimes [mean annual soil temperature is less than 8°C (Soil Survey Staff, 1999)] are located in soils north of this line whereas mesic soil temperature regimes [mean annual soil temperature is greater than 8°C but less than 15°C (Soil Survey Staff, 1999)] are located in soils south of this line. Within this study, a 260km transect was laid from Day County to Clay County in South Dakota. Eight profiles were sampled north of the 8.3°C line on frigid soils and eight were selected south of the line on mesic soils. All sites were formed from the same parent material and had been under similar agronomic management. The profiles were sampled and analyzed for P, N, and C.

Westin and Buntley (1967) determined that in South Dakota soils, there are significant changes in soil P, N, and C according to soil taxonomy. Moving from north to south resulted in reductions in aluminum and calcium associated P, whereas iron (Fe) and reductant P [iron-bound P (Chang and Jackson, 1958)] increased. Both C and N declined in the A horizon from north to south, but increased in the B horizons. Lower C and N in the A horizon in southern counties could be due to the increases in temperature and precipitation leading to higher mineralization rates (Westin and Buntley, 1967). Increases in C and N in the B horizons was determined to be due to the thicker horizons present in southern counties as compared to the northern counties (Westin and Buntely, 1967). Across the soil transect, the changes in soil properties were gradual. However, the greatest differences were observed at sites that bordered the mean annual soil temperature line along Kingsbury County and Brookings County where soils changed from frigid soils in the north to mesic soils in the south (Westin and Buntely, 1967). These results illustrate the effect of differences in soil classification on soil properties and offer insight into conclusions that can be drawn from taxonomic information.

Topography is another important abiotic factor when assessing SOC dynamics. Soil solum and horizon structure change with landscape position (e.g. shoulder (SH) versus toeslope (TS)). An experiment done by Aguilar and Heil (1988) aimed to show the importance of toposequence and parent material on SOC content. Soils with sandstone, siltstone, and shale parent materials under pasture and cultivation were located adjacent to each other in Grant County, ND. The sandstone toposequence was divided into summit (SM), shoulder (SH), upper backslope (UBS), lower backslope (LBS), footslope (FS), back backslope (BBS), and back footslope (BFS) landform positions. The siltstone toposequence was divided into SM, two shoulders (SH & SHB), UBS, LBS, FS, and toeslope (TS) landform positions. The shale toposequence was divided into SM, SH, BS, and FS landform positions. Each horizon per toposequence was sampled to a depth of 120cm or to a lithic contact if before 120cm. Bulk density was taken via the core method, pH was done by saturated paste, SOC was calculated by the Walkley-Black method, TN assessed via Kjeldahl, and total/organic P were determined by acid extraction and combustion.

Overall, the sandstone contained the least amount of C for all landscape positions, followed by siltstone, and shale had the largest amount of C at all landscape positions. The summit positions statistically had the lowest SOC concentrations whereas the footslope positions statistically had the greatest. Nitrogen and P followed this trend also. The authors credited this to greater OM production in footslope positions as well as to deposition from upper landscape positions (Aguilar and Heil, 1988). This research shows how location in the toposequence and parent material impact SOC concentrations.

Soil Organisms:

Soil is home to billions of living creatures; there are more microorganisms in a teaspoon of healthy soil than there are people on the planet (Herring, 2010). Soil microorganisms include diverse species of bacteria, actinomycetes, fungi, nematodes, and more (Brady and Weil, 2017). Earthworms are important soil macro-organisms that modify soil structure via ingestion and excretion of soil, effectively mix surface litter deeper into the soil profile, and impact microbial mineralization (Barre et. al., 2009; Fahey et. al., 2013). Microbial communities influence the types and quantities of C pools in the soil (Zhao et. al., 2018). Soil organisms are important to the formation, accumulation, and mineralization of SOC.

Miltner et. al. (2012) hypothesized that microbial biomass accounts for a significant portion of particulate SOM. To test this, ^{13}C labeled *Escherichia coli* (*E.coli*) was added to a rye (*Secale cereale*) cropping system on a Haplic Phaeozem (Typic Hapludoll) soil. The *E.coli* accounted for 26% of the naturally occurring MBC in the soil. Incubation at 20°C lasted up to 224 days depending on treatment. At the end of the incubation period, it was found that 56% of the ^{13}C labeled MBC had been mineralized and the remaining 44% was still in the soil. Further analysis of proteins indicated that 75% of the ^{13}C labeled proteins that had been initially added to the soil were converted to SOM. Given the assumption that 50% of microbial biomass is composed of proteins (Miltner et. al, 2012), approximately 37.5% of MBC was transferred into SOM. The authors conclude that roughly 40% of microbial biomass is transformed into SOM (Miltner et. al, 2012). Research such as this illustrates the importance of healthy microbial communities on SOC accumulation.

As was noted earlier, SOC exists in different pools. Zhao et. al. (2018) studied the impact of microbial diversity on different SOC pools under afforestation. They monitored SOC and microbial communities of three *Robinia pseudoacacia* L. (RP) stands and one farmland (FL) treatment in Anasi County, China. Tree stands ranged in age from 42 years, 27 years, or 17 years since afforestation. Prior to replanting with RP, the ground had been farmland. Each treatment was replicated three times, each replicate was divided into three plots, and each plot was divided into two subplots. Soil samples were collected from the 0-10cm depth after carefully removing the surface litter layer and analyzed for bacterial and fungal species composition, MBC, and carbon fractions. Soil organic carbon was determined via potassium dichromate oxidation, MBC was determined via fumigation, light fraction organic carbon (LFOC) was determined by sodium iodide extraction, particulate organic carbon (POC) was determined by sieving, labile soil carbon (LOC) via potassium permanganate extraction, and DOC via total organic carbon (TOC) analyzer. Microbial DNA was extracted and quantified by polymerase chain reaction and using Quantitative Insights into Microbial Ecology (QIIME) software. Relationships between C fractions, bacterial diversity, abundance, and phyla present in soil samples was assessed via Spearman's rank coefficient (Zhao et. al., 2018).

Zhao et. al. (2018) conclude that time, land use, and microbial community have significant impacts on C pools. Coarse and fine POM, LOC, POC, MBC, and LFOC were all significantly greater in the 42 year old RP stand when compared to all other treatments. *Robinia pseudoacacia* L. (RP) carbon fractions ranged from 27.2% to 94.1% and were all significantly higher than FL carbon fractions. Bacterial and fungal diversities were significantly greater in each RP treatment than in the FL treatment. The dominant bacterial phylum in the RP treatments was *Proteobacteria*, whereas in the FL treatment *Actinobacteria* was dominant. Dominant fungal phyla included *Ascomycota*, *Zygomycota*, and *Basidiomycota* across all treatments but *Zygomycota* was the largest in afforested treatments whereas *Ascomycota* were predominant in FL. Each phylum consists of many different classes of bacteria or fungi, but specific phyla have a positive or negative relationship with carbon. *Proteobacteria* and *Zygomyota*, the predominant bacterial and fungal phyla in RP treatments, have a positive correlation with SOC. *Ascomycota*, the predominant fungal phylum in the FL treatment, are negatively correlated to SOC. The authors note that microbial populations are hinged on C and N inputs into the soil, as dictated by land use, due to the effect those nutrients have on lignin decomposition. The main highlight of this research is that as microbial diversity increases in a soil ecosystem, C also increases. Specific bacterial and fungal phyla also play an important role in C fraction dynamics, and different forms of management dictate which phyla will be most prevalent in the soil ecosystem (Zhao et. al., 2018).

Vegetative and Residue Cover:

The type of vegetative cover and amount of plant residue covering a soil will impact SOC accumulation. Conversion of forestland to agricultural ground leads to significant reductions in SOC (Lemenih et. al., 2005; Wei et. al., 2014a; 2014b). Similarly, cultivating native prairie soils initiates significant decreases in SOM (Cambradella and Elliott, 1992; Malo, 2005; Smith, 2008). Removal of crop residues after harvest is also directly related to decreases in SOM (Wilhelm et. al., 2004; Ruis et. al., 2018). Aside from directly removing C-laden stover from agricultural fields, secondary impacts of residue removal, such as increased erosion, will compound reductions in SOC content (Mann et. al., 2002).

Converting native forest to cultivated systems has detrimental impacts on SOC. Wei et. al. (2014a) studied the effect of vegetation conversion from native forest to cultivated land on SOC levels. Research was conducted in the Huanglongshan Forest in Shaanxi Province, China on a Cambisol (Inceptisol). The region is semi-humid with a temperate climate. Four sample locations were paired by soil type and management practices. Each location encompassed native forest, 4 year-, 50 year-, and 100 year-long cultivation treatments within 3km of each other. The native forest was composed of minimum 200 year-old oak (*Quercus liaotungensis* Koidz) and birch (*Betula platyphylla* Sukaczev) stands with bunge needlegrass (*Stipa bungeana* Trinius) established on the forest floor. Cultivated sites had been sown to a millet (*Setaria italica* L.), maize (*Zea mays* L.), and potato (*Solanum tuberosum* L.) rotation (Wei et. al., 2014a). Bulk density, SOC, and N samples were collected at 0-10cm and 10-20cm depths. Laboratory analysis included separating the light and heavy SOM fractions and measuring the natural abundance of ^{13}C and ^{15}N isotopes.

Wei et. al. (2014a) determined that there were significant decreases in SOM after conversion from native forest to cropland with the largest decreases witnessed in the 0-10cm depth. SOC declined by 19% in the first four years of cultivation. Cultivation had led to a 33% reduction compared to native forest after 50 years of cultivation in the top 0-10cm depth. In the 10-20cm depth, Wei et. al. (2014a) determined a 3% decrease in SOC after four years of cultivation and a 16% decrease in SOC after 50 years of cultivation. Length of cultivation did not significantly impact SOC between 50 and 100 years of cultivation. Additionally, light-fraction OM (LFOM) significantly decreased (34%) after four years of cultivation in the 0-10cm depth only and was not significantly different between 50 and 100 years of cultivation. The authors conclude that the most dramatic losses of SOC occur before 36 years of cultivation on ground converted from native forest (Wei et. al., 2014).

Cambardella and Elliott (1992) were interested in how changes in cultivation impact POM in a grassland ecosystem. Native sod, bare-fallow, stubble mulch, and no-till were all sampled from the 0-20cm depth on a fine-silty, mixed, mesic Pachic Haplustoll in a temperate climate near Sidney, Nebraska. Mineral-associated and water-soluble C and N, as well as POM, were separated in the lab. One-way ANOVA was used to statistically analyze differences between treatments and Fisher's LSD (Freund et. al., 2010) was used to test pairwise comparisons.

Results indicated a significantly higher amount of SOC in the native sod than for any of the tillage treatments. The stubble mulch and no-till treatments exhibited statistically similar SOC and both had significantly more SOC than the bare-fallow treatment. Native sod also had significantly higher POM than any of the tillage treatments. The authors conclude that cultivation leads to reductions in the labile C pool and reduces overall SOC compared to native conditions (Cambardella and Elliott, 1992). This research highlights the positive impact of perennial vegetation on SOC stocks.

Crop residues are important sources of C in the soil system. Field residues are sometimes removed via livestock grazing or are baled for use as animal bedding (Ruis et. al., 2018) or cellulosic ethanol (Wilhelm et. al., 2007). Significant reductions in C stocks can result from the removal of crop residues. Ruis et. al. (2018) hypothesized that removal of corn stover as bales or via grazing would significantly reduce C stocks in a central Great Plains, prairie-derived ecosystem. Ruis et. al. (2018) tested this hypothesis over a three-year period on a fine-silty, mixed, superactive, mesic, Cumulic Haplustoll near Gothenberg, Nebraska. Treatments were arranged in a split-strip, split-strip randomized complete block design. Residue removal treatments were broken into no-removal, livestock grazing, and stover baling. Each removal treatment took place on one of two irrigation treatments; full-irrigation and limited-irrigation. Each removal treatment also took place on two tillage practices; strip-till (20cm deep by 25cm wide tilled in April) and No-Till. Nitrogen and P fertilizer were applied to all treatments each year. The University of Nebraska Corn Stalk Grazing Calculator (Stockton and Wilson, 2013) was used to determine optimal stocking rate for grazing treatments in which the cattle grazed stalks for five days in the winter. Surface residue was measured using quadrats each spring and composite soil samples were collected from 0-5cm, 5-10cm, and 10-20cm depths. The geometric mean diameter of dry and of wet aggregates was analyzed to assess wind and water erosion potential. Soil organic carbon was measured by combustion, bulk density was measured by the core method, POM was separated into cPOM and fPOM via sieving, fatty acid methyl ester (FAME) was used to analyze microbial community composition, and sorptivity was measured by the single ring method. Soil organic carbon stocks were calculated using SOC concentration and bulk density values. For analysis, the fixed effects were residue removal, irrigation level, and tillage level. The authors tested for correlation at the irrigation and tillage level to avoid confounding results by unintended interaction.

Data indicated that, over the three-year study, baling stalks removed 66% of residue and grazing removed 24% of residues when compared to the no-removal treatments. Neither irrigation level nor tillage level had a significant impact on SOC concentration. However, when analyzing the effect of tillage on SOC stocks, tillage had a significant impact at the 0-5cm depth (Ruis et. al., 2018). In 2014 there was no significant difference between grazing and baling treatments, and both had significantly less residue than the no-removal treatments. However, in 2015 and 2016, baling removed significantly more residue than the grazing or no-removal treatments, which were statistically equal. Baling decreased C concentration by 27% in the 0-5cm depth and 12% in the 10-20cm depth (Ruis et. al., 2018). Baling reduced SOC stocks by 31% over both tillage systems in the 0-5cm depth when compared to no-removal treatments. Grazing did not have a significant impact on SOC stocks. No-Till grazing showed 31% greater SOC stocks than strip-till. Particulate organic matter was significantly decreased in the 0-5cm depth with residue removal, but no additional interaction between tillage or irrigation was found. Residue removal reduced cPOM by 82% and fPOM by 27%, whereas grazing reduced cPOM by 17% and fPOM by 5% when compared to no-removal. These reductions are likely caused by limited labile C additions to the soil system (Ruis, et. al., 2018). Additionally, the data indicated that residue removal significantly altered microbial biomass and community composition, but grazing and no-removal were generally statistically the same. Baling residue significantly increased wind and water erosion potential as compared to grazing and no-removal (Ruis et. al., 2018). From this research, it is evident that crop residues play an important role in C additions to the soil profile and, inversely, residue removal leads to reductions in SOC.

Rotation and Cropping Intensity:

Diverse crop rotations are a means by which producers can spread economic risk over different commodities, break disease and pest cycles, and improve soil quality (Zotarelli et. al., 2005; Lakhran et. al., 2017; Manns and Martin, 2017; Jarecki et. al., 2018). Not all plants have the same potential for building SOC. Some plant species, such as oats (*Avena sativa*), will enrich SOC levels in the surface horizon whereas others, such as tall fescue (*Lolium arundinaceum*), can enrich SOC down to 60cm (Manns and Martin, 2017). Research has shown that diversifying crop rotations to include legumes and perennial grasses is an efficient method of increasing SOC even on low OM soils (Johnston et. al., 2017; Jarecki et. al., 2018).

Although sandy soils are not optimal candidates for building SOC, proper management and diverse rotations can help maintain current levels and potentially build SOC levels over time (Johnston et. al., 2017). It was hypothesized that adding perennial crops (termed "ley crops" within this study) into the crop rotation would build SOC (Johnston et. al., 2017). This research took place at Rothamsted Farm in Harpenden, UK on the Woburn ley-crop experiment site, which began in 1938 and is located on a Cambic Arenosol (Psamment). Ley-crop treatments consisted of either three years safronin (*Onobrychis viciifoilia)* or alfalfa (*Medicago sativa*) (Lu), three years of grass with N fertilizer (LN3), 8 years of grass with clover (*Trifolium spp.*) (LC8), or 8 years of grass with nitrogen fertilizer (LN8). In 1972, safronin and alfalfa were replaced by grass with clover (LC3). Ley-crop treatments were followed by two years of arable crops which were tested for yield differences between ley-crop treatments and all-arable crop treatments. Treatments without a multi-year ley crop in the rotation consisted of a five-year rotation with one year of a hay or root crop (AB) or a five-year rotation in which two years were bare fallow (AF). Until the 1960's, farm-yard manure was added every fifth year to the treatments. Above ground biomass was removed and ley-crops were generally cut twice a season. Soil samples were collected from 0-23cm and 23-46cm beginning in 1938 and were regularly collected beginning in 1960. Soil was tested for Olsen P, exchangeable K, exchangeable magnesium (Mg), and pH to ensure nutrients were not limiting to crop growth. Total carbon (TC) and N were analyzed by combustion. A conversion factor of 1.72 was used to convert SOC to SOM (Johnston et. al., 2017). SOC turnover was estimated using the RothC model (Rothamsted Research, 2017).

Initial SOC at this site before starting the rotation study was 0.98% across all plots. By the early 1960's, the LN3 treatments exhibited a 1.27% SOC increase compared to the AB and AF treatments (Johnston et. al, 2017). Safronin or alfalfa (Lu) ley-crops did not result in an SOC increase. After changing those plots to grass with clover (LC3), there was a 1.24% increase in SOC which was comparable to the (LN3) treatment (Johnston et. al, 2017). The AB treatment declined in SOC from 0.98% to 0.91% and the AF treatment declined from 0.98% to 0.80% (Johnston et. al, 2017). Soil organic carbon in the LN8 and LC8 rotations increased from 0.98% to 1.42% and 1.40% respectively (Johnston et. al, 2017). Carbon inputs over the 70-year study period were modeled using the RothC model. Model predictions for the 70-year period estimated that the AB treatment had a C input of 140 t/ha, the AF treatment had 122t/ha, LN3 had 189t/ha, and LC3 had 134t/ha of C input. The model also predicted that most of the C inputs from the AB factor had been lost due to limited belowground biomass, all C would was lost from the AF treatment, 96% was lost from the LN3 treatment, and 98% was lost from the LC3 treatment (Johnston et. al, 2017). Overall, the RothC model optimistically estimated that 5% of C inputs would be retained in the soil from year to year, but more realistically 3% or less would remain in this sandy clay loam soil (Johnston et. al, 2017). Although C retention was low, the authors noted the beneficial impact of ley-crops significantly increasing SOC over time on a sandy textured soil.

Semi-arid landscapes also are limited in their ability to build SOC. Work done by Rosenzweig et. al. (2018) in the semi-arid Great Plains suggests, however, that intensifying the crop rotation can significantly build SOC. Typical crop rotations in the semi-arid Great Plains include wheat (*Triticum aestivum*)-fallow system, mid-intensity rotations, and continuous cropping. Rosenzweig et. al. (2018) hypothesized that increasing crop rotation intensity would lead to significant increases in SOC, fungal biomass, and soil aggregation. Research took place in western Nebraska and eastern Colorado on a variety of soil types. Wheat-fallow (WF), mid-intensity (MID), and continuous cropping (CON) were the three rotations assessed in this research. Wheat-fallow rotations are defined as rotations in which winter wheat is raised from September to July, then the ground is left fallow for 14 months until the next wheat crop is planted. Mid-Intensity rotations are defined as management systems in which corn (*Zea mays*), sorghum (*Sorghum bicolor*), peas (*Pisum sativum*), sunflowers (*Helianthus annuus*), or another crop well suited to the region is planted in conjunction with wheat. This reduces the frequency of fallow. Lastly, CON rotations contain no fallow years and a crop was raised every growing season. Samples were collected on a total of 96 fields that were under dryland, No-Till management, without manure or compost additions. Of the 96 fields, 54 were located on working farms and the remaining 42 were located at long-term experiment stations. A total of 27 fields were under WF management, 37 were under MID management, and 26 were under CON management. For comparison purposes only, six 30 year-old CRP perennial grass plots were also sampled. Potential evapotranspiration increased along a gradient from 1368 mm per year in Nebraska to 1975 mm per year in southeastern Colorado.

Samples were collected from 0-10cm and 10-20cm in the fall of 2015 and again in the spring of 2016. Fall samples were analyzed for SOC, texture, and pH. Spring 2016 samples were analyzed for water-stable aggregates (mean weight diameter), bulk density, SOC, total nitrogen, and phospholipid fatty acids (PLFA) (Rosenzweig et. al., 2018). Multiple linear regression and backward selection were used to measure the relationship between SOC, C concentration, aggregate stability, and microbial PLFA (Rosenzweig et. al., 2018). Analysis of variance and Tukey Multiple Comparison tests (Freund et. al., 2010) were used to identify pairwise differences between C and C:N ratios within different aggregate size classes. This final regression model was tested for fit (significance) through structural equation modeling (SEM) and improved by adding back in previously removed covariates until significance was achieved.

Results indicate that SOC increased by 17% in CON rotations over WF at the 0-10cm depth (Rosenzweig et. al., 2018). There was no difference between CON and MID treatments. When comparing the whole sample depth (0-20cm), there was a 16% increase in SOC concentrations for CON treatments over MID treatments. Continuous and MID treatments reflected approximately 80% and 70%, respectively, of the SOC found in the 30-year old CRP fields. Based on the model, cropping intensity explained roughly 4% of the SOC variability at both sampling depths, whereas clay content explained approximately 17% of the variability and PET explained approximately 14% (Rosenzweig et. al., 2018). Mean weight diameter (MWD) of aggregates was twice as large in CON treatments as in WF treatments after accounting for the role of PET and clay content on aggregate stability. Mean weight diameter (MWD) of CRP aggregates were four times larger than CON aggregates and eight times larger than WF aggregates. Microbial community structure plays a large role in SOC accumulation (Zhao et. al., 2018). Rosenzweig et. al. (2018) found that microbial communities were significantly impacted by increasing cropping intensity. Continuous treatments had three times higher fungi:bacteria ratios compared to WF treatments while MID treatments were not significantly different from other treatments. Additionally, aggregate stability linearly increased with fungal biomass and SOC linearly increased with aggregate stability. For these reasons, increasing cropping intensity directly (greater levels of crop residue additions) and indirectly (effects on aggregate stability) increases SOC. One more benefit the authors note concerning increasing cropping intensity is that they did not observe an increase in N fertilizer application, but did observe an increase in crop production (Rosenzweig et. al., 2018). This research highlights the importance of vegetative cover on crop ground and the benefit of increasing the cropping intensity.

Fertilizer Amendments:

Direct and indirect effects on SOC are witnessed with the use of organic and synthetic fertilizer materials. Organic amendments, such as manure, directly add C to the soil (Ryals et. al., 2014). Use of organic and/or synthetic fertilizers increases above and belowground biomass, which indirectly enhances SOC through greater plant additions (Ryals et. al., 2014). Research in different global regions and under different vegetation has shown that fertilizer amendments build SOC (Srinivasarao et. al., 2012; Ryals et. al., 2014).

Ryals et. al. (2014) assessed the impact of compost on C and N storage on two California grassland soils under different biomes over a period of three years. In this study, the first location focused on valley grasslands at the Sierra Nevada foothills in Brown Valley, CA on Xerochrept and Haploeralf soils. The second location focused on coastal grasslands in Nicasio, CA on Haploxeroll and Argixeroll soils. Seasonally, Brown Valley is hot and dry, whereas Nicasio experiences milder seasons along the coast. Two treatments were studied at both sites; a one-time compost amendment and a non-amended control. Compost was analyzed for C and N content, C:N ratio, and particle size classes before being applied to a thickness of approximately 1.3cm over compost plots. Compost was applied in December of 2008. Soil samples were collected before compost application and in mid-summer (corresponding to water years) for three years. Water years pertain to surface waters and are based on a twelve month period from October 1 through September 30 (United States Geological Survey, 2016). Sample depths in valley grasslands were 0-10, 10-30, and 30-50cm and sample depths in coastal grasslands were 0-10, 10-30, 30-50, and 50-100cm. Bulk density was collected via the core method. Carbon and N were analyzed via CN Analyzer, texture was conducted via the hydrometer method, and pH was measured on a 1:2 soil:water solution. At year three, 0-10cm samples were collected at all plots and the organic matter was broken into three fractions; free light fraction (FLF), occluded light fraction (OLF), and mineral-associated heavy fraction (HF). Occluded light fraction associated C is protected within soil aggregates. Organic matter decomposition was calculated using ratios derived from Diffuse

Reflectance Infrared Fourier Transform spectroscopy (DRFIT) (Ryals et. al., 2014). Statistical analysis included ANOVA to test for differences in C and N concentrations and pools for each site. Repeated measures ANOVA analysis was used to identify interactions between variables.

Soil texture and pH did not vary significantly between the valley and coastal grasslands, soil bulk density significantly increased with depth at each site, and the compost amendment had no significant impact on bulk density values at either site. Prior to compost application, there were no significant differences in C and N concentrations between treatment plots and C and N significantly declined with depth across all plots. At the valley site, compost application led to significant increases in C and N concentrations compared to the control. The increase in C and N were sustained through the three-year trial. Although C and N increased numerically over the three-year study with compost application at the valley site, the increase was not significant. At both sites, compost significantly increased the C content of the FLF organic matter fraction in the 0-10cm depth. Valley grasslands saw a 26% increase in FLF associated SOC and coastal grasslands saw a 37% increase in FLF associated SOC over the three year study. Occluded light fraction organic matter trended upward at both sites but only significantly increased at the coastal site. Heavy fraction organic matter was unaffected by treatment and had the lowest C concentration of the three OM fractions, however, it accounted for most of the C present on a mass basis. Nitrogen concentration was significantly increased in all OM fractions at the valley site and in the FLF and OLF fractions on the coastal site. Overall, the C:N ratio decreased with compost addition at both sites. Ryals et. al. (2014)

conclude that organic amendments not only increase soil C and N concentrations, but also protect C and N through the formation of aggregates over a short time period.

Work by Srinivasarao et. al. (2012) assessed the effect of organic and synthetic fertilizer on SOC sequestration, the relationship of SOC to the sustainable yield index (SYI), and the amount of C inputs was required to maintain SOC levels. Research took place on an Udic Ustochrept soil in subhumid, tropical Varanasi, India over a 21-year period. Seven treatments were studied under rice (*Oryza sativa* L.)-lentil (*Lens esculenta* Moench) rotation in which rice was grown in the rainy season (June-September) and lentil was grown in the post rainy season (October-December). Treatments were no-fertilizer control, 100% recommended dose (i.e. recommended application rate) mineral fertilizer (RDF), 50% RDF, 100% organic Farm-Yard Manure (FYM), 50% RDF with 50% RDF-foliar, 50% FYM with 50% RDF, and Farmer's Practice of 20kg N ha^{-1}. Each treatment had three replicates. Farm-Yard Manure was applied at a rate of approximately 10.7 Mg ha^{-1} and the RDF in the region was a 60-50-30 (N-P-K) blend. All fertilizer was applied to the rice crop and the legume crop used residual fertility. Three 0.2m long samples were collected from each plot to a depth of one meter. Samples were analyzed for pH, carbonates, CEC, texture via hydrometer, and plant available N, P, and K. Bulk density was collected separately. Above and belowground biomass were measured after harvest had removed the grain. Carbon stocks were calculated by multiplying SOC concentration by the soil bulk density, then by the sample depth, and then by a factor of 10. Sequestered C was calculated by the equation: $\mathrm{SOC}_{\mathrm{current}} - \mathrm{SOC}_{\mathrm{initial}}$. The sustainable yield index was calculated by subtracting the average crop yield from the estimated standard deviation, and dividing by maximum yield. The Duncan Multiple

Range test (Duncan, 1955) was used to measure differences between means for each treatment and treatment-wise regression models for rice and lentil were used to determine the effect of fertilizer on SOC.

Carbon inputs in the control treatment were 1.1 Mg C ha^{-1} yr^{-1} whereas the 50% FYM with 50% RDF treatment had significantly more C input at 2.4 Mg C ha^{-1} yr^{-1}. All FYM treatments resulted in an additional 1.77 Mg C ha^{-1} yr^{-}1 to 3.55 Mg C ha^{-1} yr^{-1} when compared to other treatments. Early in the 21-year study period, yields across mineral and organic inputs were statistically similar. As the experiment progressed, the 50% FYM with 50% RDF treatment had the highest yield followed by 100% mineral and 100% FYM. Additionally, treatments with FYM saw better water retention which improved the SYI values for those treatments. Bulk density was lower in treatments with FYM when compared to mineral fertilizer or the control treatment. Carbon concentration in the control treatment declined most significantly in the 0-20cm and 20-40cm depths. Farm-Yard Manure was able to increase SOC down to 80cm. Overall, SOC stocks were greatest in the 100% FYM treatment, followed by 50% FYM with 50% RDF, and then the 100% RDF treatment. The 50% RDF with 50% RDF-foliar treatment had statistically the same SOC stocks as the 50% RDF and Farmer's Practice, but significantly more than the control treatment. In this study, Srinivasarao et. al (2012) calculated that 13.4% of the C directly applied in the form of FYM was converted into stable SOC stock. Control treatments saw the largest reduction in SOC stocks (-3.1 Mg C ha^{-1}) and the 100% RDF treatment did not supply enough C to prevent decreases in SOC stocks (-0.4 Mg C ha^{-1}). From their calculations, Srinivasarao et. al. (2012) conclude that 2.47 Mg of C ha^{-1} need to be added to tropical Inceptisols each year to maintain SOC stocks. Fertilizer amendments can help directly and indirectly add C to the soil to maintain SOC stocks in agricultural land.

Tillage:

Tillage breaks soil structure and protective aggregates, promotes wind and water erosion, and accelerates microbial decomposition of labile C (Angers et. al., 1997; Balesdent et. al., 2000; Mann et. al., 2002; Metay et. al., 2007; Manns and Martin, 2017; Maillard et. al., 2018). Long-term tillage can lead to substantial reductions in SOC content compared to SOC under native conditions (Malo et. al., 2005; Franzluebbers and Stuedemann, 2008). Certain tillage management practices, such as No-Till, have shown promise in restoring SOC with time (Blanco-Canqui and Lal, 2008; Mishra et. al., 2010; Engel et. al., 2017).

Prairie-derived soils in the Northern Great Plains are young, fertile soils that are naturally high in OM and C (Liebig et. al., 2004). Work done by Malo et. al. (2005) sought to quantify the effect of long-term tillage on various soil properties in the Northern Great Plains. Research took place on Argiustolls, Natrustolls, Argialbolls, Calciustolls, Haplustolls, Argiaquolls, Endoaquolls, and Fluvaquents in Beadle, Minnehaha, McCook, and Union counties in South Dakota. Sample locations coincide with unpublished data collected between 1919 and 1922 by J.G. Hutton in which Hutton analyzed TC, SOC, soil inorganic carbon (SIC), TN, total phosphorus (TP), total potassium (TK), pH, and bulk density on cultivated and uncultivated paired soils. Malo et. al. (2005) returned to these sample locations 75 years later in 1996 and 1997, and repeated Hutton’s sample procedure to gather new samples. Some locations that had been uncultivated at the time of Hutton's work had since been cultivated when Malo et. al. (2005) returned. In these areas, uncultivated ground on the same soil type was located within 100m of the original sample site when possible. Based on information from Hutton's study, it is known that the cultivated sites had been tilled for greater than 80 years. Sample depths were 0-15, 15-50, and 50-100cm and samples were analyzed for the same soil properties as Hutton completed in 1922. Bulk density was not collected in 1996/1997 but was estimated based off bulk density values provided by the USDA-NRCS.

Results from Malo et. al. (2005) indicate significant changes in nitrate nitrogen (NO_3^{-1}-N), extractable P, and extractable K with sample depth and treatment. Nitrate levels were higher in cultivated treatments compared to uncultivated due to repeated N fertilizer additions, uncultivated treatments had significantly more P in the 0-10cm depth than cultivated treatments due to harvest removal in cultivated treatments, and uncultivated treatments had significantly higher extractable K than cultivated treatments at all depths due to harvest removal (Malo et. al., 2005). Organic matter is the only natural source of N in the soil (Brady and Weil, 2017). Total nitrogen significantly decreased with depth in response to declining humus levels. Additionally, cultivated soils experienced 20-30% reductions in TN compared to uncultivated; even with fertilizer additions, high amounts of TN have leached or been harvested. Depth had a significant impact on TC as well. TC was greatest in the 0-15cm depth for uncultivated soils, but TC was greatest in the 50-100cm depth for cultivated soils due to carbonate rich parent material. Effect of parent material on TC is evidenced by SIC measurements in which the 50-100cm depth has significantly greater SIC than either the 0-15 or 15-50cm depths.

Cultivated treatments had significantly less SOC than uncultivated at the 0-15 and 15-50cm depths, but were statistically the same at the 50-100cm depth. A 15-30% reduction in SOC between cultivated and uncultivated treatments was determined. Changes in bulk density can have large impacts on a soil's reference surface elevation (RSE). When bulk density values increase, soils essentially "sink" from a reduction in pore space. Therefore, if sampling protocol does not account for changes in RSE corresponding to changes in bulk density, the potential for incorrect sampling depth increases. Malo et. al. (2005) corrected the data for these changes in RSE. While the cultivated and uncultivated treatments had similar TC:TN ratios in the 0-15cm depth, uncultivated treatments had significantly lower TC:TN ratios at deeper depths than the cultivated treatments. The TC:TN ratio in uncultivated treatments did not change from 0-15cm to 15-50cm, but that ratio significantly increased in the 50-100cm depth due to the presence of carbonates. The TC:TN ratio in cultivated treatments increased with each depth. Decomposition of humus is evidenced by the SOC:TN ratio, as it significantly decreased in the 0-15cm and 15-50cm depths for both cultivated and uncultivated treatments. No major changes in SOC:TN were noted in the 50-100cm depth. Many of the sites were located on landscape positions that are susceptible to wind and water erosion. Although wind erosion was minimal due to the soil texture, there was a high risk of water erosion. For this reason, Malo et. al. (2005) hypothesize that SOC loss in cultivated treatments could in part be due to soil erosion in this area. Malo et. al. (2005) conclude that C losses from harvest removal, decomposition of OM, and erosion have caused the greatest losses to SOC in this study.

Cultivation is known to result in lower SOC levels, weaker aggregates, and higher erosion potential when compared to native systems (Malo et. al., 2005). Work done by Mishra et. al. (2010) aimed to quantify changes in SOC as land was converted from forest to crop ground, ascertain the effects of tillage on SOC sequestration, and quantify the rate of SOC sequestration under different tillage practices in the Corn Belt of Ohio, USA. Mishra et. al. (2010) hypothesized that reducing soil disturbance with No-Till practices would lead SOC levels to surpass those of conventional tillage systems. This study was implemented on three university-conducted long-term tillage experiments and three producer operated farms in Ohio, USA. The first long-term experiment was located near Coshocton, OH on a Ultic Hapludult, the second was near South Charleston, OH on a Aeric Ochraqualf, and the third was near Hoytville, OH on a Mollic Ochraqualf. Producer locations were near Delaware, OH on an Aquic Hapludalf, near Coshocton, OH on a Typic Hapludult, and near Hoytville, OH on a Mollic Epiaqualf. A total of three treatments were studied; native woodlots (NW), chisel plow/conventional tillage (CT) and no-till (NT). Level of tillage in CT treatments, duration of NT, and crop rotation varied from location to location. The long-term experiment sites were replicated four times and the producer-managed sites were analyzed as pseudo-replicates by pairing samples at each treatment level by soil type and topography.

Soil samples were collected from each treatment in summer of 2006. Bulk samples and core samples were collected from 0-10cm, 10-20cm, 20-30cm, and 30-40cm at each treatment. Due to a warm regional climate, organic horizons do not form under Ohio forests and fresh litter was not included in soil samples of NW or NT treatments (Mishra et. al., 2010). Samples were analyzed for soil moisture content, bulk density, TC, TN, and isotopic carbon concentration (^{13}C). Relative abundance of ^{13}C was determined by dividing the ratio of sample ^{13}C and ^{12}C to a reference value. This was subtracted from 1 and multiplied by 1000 to attain ^{13}C ‰. SOC and N pools (stocks) were calculated with bulk density values and sample depth. One-way ANOVA was used to determine the effect of tillage on SOC and N concentration, SOC and N stocks, bulk density, and carbon isotopes at each sample depth (Mishra et. al., 2010).

Bulk density was only mildly impacted by tillage, but was significantly lower in NW treatment down to 30cm. Native Woodlot bulk density significantly increased in the 30-40cm depth. Tillage had no significant impact on bulk density at the long-term tillage experiment locations and results were not consistent at the producer locations. At all six locations, SOC and N levels at the 0-10cm depth were greater in the NW treatment than in CT or NT treatments. Four of six locations had more SOC and N in the 0-10cm depth in NT treatments than in CT treatments. There were no significant differences among SOC and N concentrations at the 10-20cm depth between tillage treatments. Soil organic carbon and N stocks were significantly greater under NW treatments at all locations. There was a significant difference between NT and CT for SOC and N stocks at the long-term experiment locations where NT had been in practice for over 50 years. There was a lack of SOC and N stock response at the producer locations which the authors note is on par with similar pseudo-replicate research. Mishra et. al. (2010), postulate that the lack of response in SOC and N stocks is due to the shorter duration of on-farm NT as compared to the long-term experiment locations. Based on the overall data from long-term experiment sites, the authors conclude that converting from CT to NT can lead to a 0.57 Mg ha^{-1} increase in C annually over the course of 15-20years. From the ^{13}C data, Mishra et. al. (2010) calculated the loss of SOC due to deforestation to range between 26% and 55%. However, by converting management practices from CT to NT, some of that SOC can be replaced in the soil profile (Mishra et. al., 2010).

Building SOC in the Agricultural Landscape:

Best Management Practices (BMPs) strive to economically, feasibly, and effectively improve crop production while considering the integrity of surrounding environmental ecosystems (MNDA, 2017). Best Management Practices can be targeted toward pest management, erosion management, water quality, soil health, and other important agronomic issues. Various possible BMPs for increasing SOC accumulation have already been mentioned in this review such as reducing tillage, increasing cropping intensity, and maintaining surface residues (Ogle et. al, 2005; Johnston et. al., 2017; Ruis et. al., 2018). Based on a global review by West and Post (2002), data suggests that conversion from Conventional Tillage to No-Till can increase C sequestration rates by 57 g C m^{-2} yr^{-1}. Additionally, increasing rotation intensity can increase C sequestration rates by 20 g C m^{-2} yr^{-1} (West and Post, 2002). The use of cover crops to increase the length of vegetative cover and increase surface residue within the growing season is another potential BMP. Another review by Blanco-Canqui et. al. (2015) highlights how the use of cover crops can increase SOC due to reduced erosion, increased biomass, and improving other soil properties to facilitate soil aggregation (i.e. limiting soil compaction).

The efficacy of BMPs is hinged on numerous factors. Climate and inherent soil properties may inhibit SOC accumulation in certain regions. For instance, in research mentioned earlier in this paper (Johnston et. al., 2017), the cool climate and sandy texture of the soil in the ley crop rotation limited SOC accumulation potential even with diverse crop rotations. Results of that research concluded that there was still a significant increase in SOC over time, but a majority of the carbon inputs were not retained in that system. Time is another restraint on many BMPs in that the positive effects may take many years to become evident (Mishra et. al., 2010). One study included earlier (Srinivasarao et. al., 2012), focused on the effect of mineral and organic amendments on SOC. The authors note that significant differences between mineral and organic amendments on SOC were minimal until later in the 21-year study period. The benefits of cover crops on SOC are often not seen in the first few years of implementation (Blanco-Canqui et. al., 2015). Producers may resist committing to a BMP that will not produce positive results in initial years of implementation. No-Till is a popular BMP for reducing erosion and improving soil health, although it is noted that significant changes may only be seen in surface horizons (West and Post, 2002; Blanco-Canqui and Lal, 2008; Rosenzweig et. al., 2018). Other problems with BMP implementation exist, such as financial burden of lost productivity or investment in new equipment, but governmental programs are available to defray the cost (Dayer et. al., 2018).

Current Carbon Models:

Carbon models have been used in scientific research since the 1940's and model development and utilization has been steadily growing with every decade (Campell and Paustian, 2015). The CENTURY and RothC models account for a majority of SOM modeling citations and publications (Campbell and Paustian, 2015). The CENTURY model was developed by the Natural Resource Ecology Laboratory (NREL) at Colorado State University. Weather data, soil texture, plant nutrient content, lignin content in plant materials, N inputs from the soil and atmosphere, and initial C and N levels are the necessary inputs for running the CENTURY model which operates on a monthly time sequence (NREL, 2012). Additional submodels for C, water, grasslands/crop systems, and forest systems exist to further separate model inputs into more specific groups with varying turnover rates (NREL, 2012). The CENTURY model has been expanded into the DayCENT model as well. DayCENT uses similar parameters to CENTURY, but operates on a daily time sequence (NREL, 2012). The RothC model was developed by Rothamsted Research in the United Kingdom. Monthly rainfall, monthly evaporation, monthly mean air temp, clay content, estimated decomposition rate, soil cover, monthly plant inputs, monthly manure inputs, and soil sample depth are the parameters required for the RothC model which also operates on a monthly time sequence (Rothamsted Research, 2017). CENTURY and RothC are complex models that have been utilized for research experiments focused on long-term impacts of agricultural management or of climate change on SOC.

Work done by Dintwe and Okin (2018) used the CENTURY model to assess the impact of anthropogenic climate change on SOC in southern Africa on the Kalahari savannahs. Four study sites were selected in this region where C, plant biomass, and nutrient cycling data had been collected in the past. Previously collected soil data, weather models, and Representative Concentration Pathway (RCP) 2.6 and RCP8.5 [future climate conditions as outlined by the Intergovernmental Panel for Climate Change (Dintwe and Okin, 2018)] were used to run the CENTURY model. The RCP2.6 scenario assumes that greenhouse gas emissions peak between the years 2010 and 2020 (IPCC, 2017). The RCP8.5 scenario assumes that emissions will continue to rise throughout the

21st century (IPCC, 2017). Simulation began in the year 4000BCE and ended in 2100. Model results indicate that warming climates are associated with higher concentrations of CO_2 in the atmosphere which will increase plant production. SOC will be negatively impacted, however, due to large losses of CO_2 from microbial respiration that outpace plant biomass carbon additions to the soil (Dintwe and Okin, 2018).

The RothC model was utilized to assess the effect of compost amendments on SOC levels in Italitan agricultural soils over a century of use (Mondini et. al., 2012). The study area encompassed all of Italy, soil data was collected from the Soil Geographical Database of Europe (Van Liedekerke and Panagos, 2012), bulk density was estimated through the Soil Profile Analytical Database for Europe (SPADE2) (Hollis et. al., 2006), a total of 12 different climate scenarios were defined based on data from the IPCC, and the RothC model was used to simulate changes in SOC from the years 2001 to 2100. Compost simulations were run on mineral-soils suited for agricultural use; approximately 60% of all soil map units in Italy were included in the compost analysis. The RothC model predicted a general decline in SOC stocks in association with climate change scenarios (Mondini et. al., 2012). However, by the addition of compost amendments SOC stocks could actually increase throughout those same scenarios (Mondini et .al., 2012).

Both the CENTURY and RothC model are well suited for scientific research that is focused on long-term impacts of management changes or changes in climate. For the average producer who is interested in potential SOC levels on their operation, however, the functionality and accessibility of these models is limited. COMET-Farm is a farm-scale model built by the United State Department of Agriculture that is based on Colorado State's DayCENT model (USDA, 2018). COMET-Farm is geared for producers who want to quantify the C footprint of their operation (USDA, 2018). Although this model is intended for producers, it can be cumbersome and results are not clearly explained. For this reason, COMET-Farm may be better served for producers to utilize with the guidance of a local NRCS or Extension Agent to help interpret results. Contrastingly, models in other disciplines exist that are producer-focused and very easy to use. One such model is the Fusarium Head Blight Prediction Center model (USWBI, 2008; De Wolf et. al., 2018) which consists of one web-page with three drop down menus. Producers need only to select their geographic region on the available map, select their hybrid's level of resistance to Fusarium head blight, and the length of time in hours they would like the projection to focus on. Results are concise and clearly illustrated. Access to a SOC model of similar simplicity would be useful for producers in South Dakota and the region who are interested in quickly quantifying their operation's potential SOC levels.

Conclusion:

Soil is a limited and valuable natural resource. Maintaining soil health for future generations will be a major challenge as the global population continues to grow and the demand for food and fiber follow (Lal, 2006). Soil organic carbon is one of three primary C pools within the global C cycle and maintaining SOC levels can sustain crop production over time (Post et. al., 1990; Lal, 2006). Numerous factors impact SOC levels including climate, physical soil properties, soil organisms, plant cover, residue removal, tillage, and more. Cultivation leads to reduction in SOC due to biomass removal, destruction of aggregates, erosion, and accelerated microbial respiration (Malo et. al., 2005; Rumpel et. al., 2015; Brady and Weil, 2017). Certain management practices show promise in returning C to the soil and increasing overall soil health (Assmann et. al., 2014; NRCS, 2017).

Currently, C models are complex and produce information above and beyond what is required at a farm-scale for producers in South Dakota and the region. For this reason, there is a need for a simple and straight-forward SOC prediction model that is similar in scope to the Fusarium Head Blight Risk Assessment Tool from USWBI. Construction of this user-friendly model would assist producers in setting relative C level goals for their operation. With a clear goal in mind, deciding which BMPs to incorporate into future management decisions will be less intimidating and more useful.

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CHAPTER 2: MATERIALS AND METHODS

Introduction and Objectives:

This research was conducted to create a simple soil organic carbon (SOC) model that would be accessible and relevant for agricultural producers in southeastern South Dakota and the region. Currently, available C models are cumbersome and do not offer straight forward information to assist producers in making management changes to enhance SOC storage on their farm. It was hypothesized that by utilizing information that is readily available from commercial soil testing labs as well as from public sources, such as Web Soil Survey (Soil Survey Staff, 2018), it would be possible to build a simple multiple linear regression model to predict SOC levels at the farm scale. In order to ascertain the effect of management practices on SOC at a farm scale, three treatments were assessed for SOC content; Native Grass (NTVG), No-Tillage (NT), and Conventional Tillage (CT). The NTVG treatments served as a control treatment as the NTVG sites represented an ecosystem in equilibrium with the majority of C preserved in the system. No-Tillage is a conservation-based BMP that has the potential to sequester 57g C m^{-2} yr^{-1} when ground is converted from CT management (West and Post, 2002). Therefore, the secondary hypothesis was that NTVG would contain the greatest SOC content, NT would fall in the middle of the spectrum, and CT would have the least SOC.

Location Descriptions:

Data for this study was collected in the summer of 2017 in South Dakota Major Land Resource Area (MLRA) 102B, 102C, and McCook County in MLRA 55C (USDA-NRCS, 2005). MLRA's 102B, 102C, and 55C are located in southeastern South Dakota

(Figure 2.1). The state of South Dakota was acquired as part of the Louisiana Purchase in 1803 and South Dakota was granted statehood in November of 1889 (Fodness, 1994). Since then, the southeastern region of South Dakota has been dedicated to intensive row crop production (Fodness, 1994). Due to long term, heavy cultivation in MLRA 102B, 102C, and in McCook County, the area was a strong candidate for measuring SOC losses and gains over time as a product of management practices.

Location 1:

Location 1 was in Lincoln County near Beresford, South Dakota (approx.: 43.125°N, -96.847°W). All treatments were located on Egan silty clay loam soil with 3 to 6% slopes. Mean annual precipitation for Location 1 is 58.4-66.0cm, mean annual air temperature is 6.10-8.90°C, and there on an average of 135-160 frost-free days (temperature basis is 0°C) (NRCS, 2018). Table 2.1 offers a summary of all the soil types included in this study and their soil classification. Specific rotation, fertilization, tillage, and yield information can be found in Appendix A.

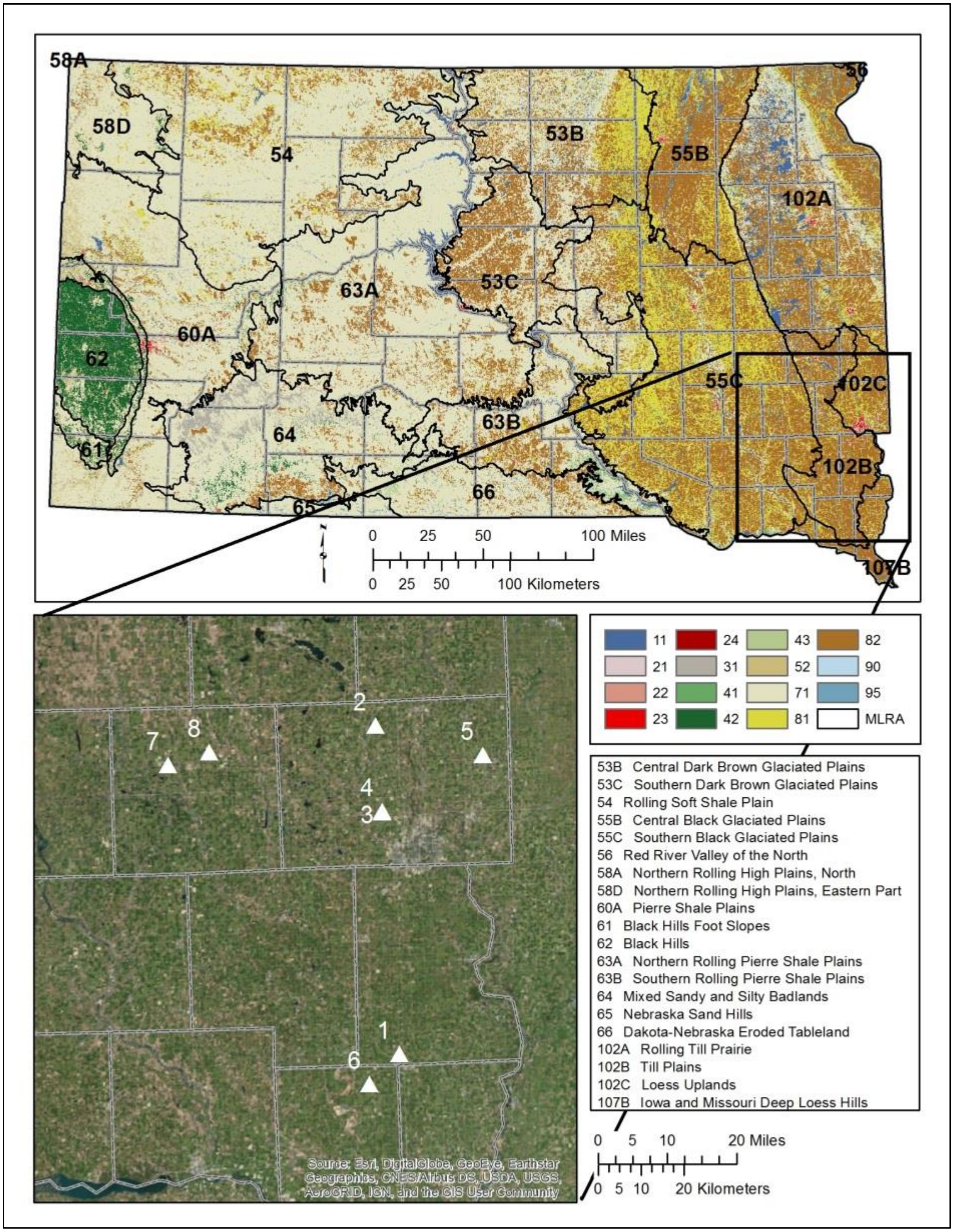

Figure 2.1: MLRA information and sample locations. Credit- Dr. Bruce Millett, SDSU Geography Department. Map Source: Esri, DigitalGlobe, GeoEye, Earthstar Geographics, CNesAirbus DS, USDA, USGS, AeroGRID, IGN, and the GIS User Community.

Location 2:

Location 2 was in Minnehaha County near Dell Rapids, South Dakota (approx.: 43.828°N, -96.712°W). All treatments were located on Dempster silt loam soil with 0 to 2% slopes. Mean annual precipitation at this location is 58.4-76.2cm, mean annual air temperature is 6.10-10.0°C, and there are on average 140-160 frost-free days (temperature basis is 0°C) (NRCS, 2018). Specific rotation, fertilization, tillage, and yield information can be found in Appendix A.

Location 3 and 4:

Locations 3 and 4 were in Minnehaha County near Sioux Falls, South Dakota (approx.: 43.546°N, -96.744°W). Treatments were located on Moody-Nora Complex soil with 2 to 6% slopes at Location 3. The predominant soil sampled was Moody. Mean annual precipitation is 61.0-78.7cm, mean annual air temperature is 6.10-11.1°C, and there are on average 140-180 frost-free days (temperature basis is 0°C) (NRCS, 2018). At Location 4, treatments were located on the Nora-Crofton Complex with 6 to 9% slopes. The predominant soil sampled was Nora. Mean annual precipitation is 61.0-78.7cm, mean annual air temperature is 6.10-11.1°C, and there are on average 140-180 frost-free days (temperature basis is 0°C) (NRCS, 2018). Specific rotation, fertilization, tillage, and yield information for both locations can be found in Appendix A.

Location 5:

Location 5 was in Minnehaha County near Garretson, South Dakota (approx.: 43.715°N, -96.5024°W). All treatments were located on Moody-Nora Complex soils. The predominant soil sampled was Moody. Mean annual precipitation is 61.0-78.7cm, mean annual air temperature is 6.10-11.1°C, and there are on average 140-180 frost-free days (temperature basis is 0°C) (NRCS, 2018). Specific rotation, fertilization, tillage, and yield information can be found in Appendix A.

Location 6:

Location 6 was in Clay County near Beresford, South Dakota (approx.: 43.077°N, -96.779°W). All treatments were located on Egan-Trent silty clay loam. The predominant soil sampled was Egan. Mean annual precipitation is 58.4-66.0cm, mean annual air temperature is 6.10-8.90°C, and there are on average 135-160 frost-free days (temperature basis is 0°C) (NRCS, 2018). This Location is missing data for a NTVG site. All potential NTVG sites in the area had been worked at one point in time or another. Specific rotation, fertilization, tillage, and yield information can be found in Appendix A.

Location 7:

Location 7 was in McCook County near Salem, South Dakota (approx.: 43.725281°N, -97.388832°W). All treatments were located on Clarno-Crossplain Complex. The predominant soil sampled was Clarno. Mean annual precipitation is 50.8-68.6cm, mean annual air temperature is 6.10-11.1°C, and there are on average 130-160 frost-free days (temperature basis is 0°C) (NRCS, 2018). Specific rotation, fertilization, tillage, and yield information can be found in Appendix A.

Location 8:

Location 8 was in McCook County near Salem, South Dakota (approx.: 43.725°N, -97.389°W). All treatments were located on Clarno-Crossplain Complex. The predominant soil sampled was Clarno. Mean annual precipitation is 50.8-68.6cm, mean annual air temperature is 6.10-11.1°C, and there are on average 130-160 frost-free days (temperature basis is 0°C) (NRCS, 2018). Specific rotation, fertilization, tillage, and yield information can be found in Appendix A.

Table 2.1. Soil sample general location, map unit, soil composition, and classification of the soils included in study.

Location	MLRA, County	Map Unit	Map Unit Name	Major Components	Soil Classification
1	102B, Lincoln	EaB	Egan silty clay loam, 3-6% slope	Egan (80%) Minor Components (20%)	Fine-silty, mixed, superactive, mesic, Udic Haplustolls
2	102C, Minnehaha	DmA	Dempster silt loam, 0-2% slope	Dempster (80%) Minor Components (20%)	Fine-silty over sandy or sandy-skeletal, mixed, superactive, mesic Udic Haplustolls
3	102C, Minnehaha	MnB	Moody-Nora Complex, 2-6% slope	**Moody (50%)** Nora (30) Minor Components (20%)	**Moody: Fine-silty, mixed, superactive, mesic Udic Haplustolls** Nora: Fine-silty, mixed, superactive, mesic Udic Haplustolls
4	102C, Minnehaha	NcC	Nora-Crofton complex, 6-9% slope	**Nora (60%)** Crofton (30%) Minor Components (10%)	**Nora: Fine-silty, mixed, superactive, mesic Udic Haplustolls** Crofton: Fine-silty, mixed, superactive, calcareous, mesic Udic Ustorthents
5	102C, Minnehaha	MnB	Moody-Nora complex, 2-6% slope	**Moody (50%)** Nora (30) Minor Components (20%)	**Moody: Fine-silty, mixed, superactive, mesic Udic Haplustolls** Nora: Fine-silty, mixed, superactive, mesic Udic Haplustolls
6	102B, Clay	EhA	Egan-Trent silty clay loam, 0-2% slope	**Egan (50%)** Trent (30%) Minor Components (20%)	**Egan: Fine-silty, mixed, superactive, mesic, Udic Haplustolls** Trent: Fine-silty, mixed, superactive, mesic Pachic Haplustolls
7	55C, McCook	Co	Clarno-Crossplain, 0-2% slope	**Clarno (45%)** Crossplain (35%) Minor Components (20%)	**Clarno: Fine-loamy, mixed, superactive, mesic Typic Haplustolls** Crossplain: Fine, smectitic, mesic Typic Argiaquolls

Location	MLRA, County	Map Unit	Map Unit Name	Major Components	Soil Classification
8	55C, McCook	C0	Clarno-Crossplain, 0-2% slope	**Clarno (45%)** Crossplain (35%) Minor Components (20%)	**Clarno: Fine-loamy, mixed, superactive, mesic Typic Haplustolls** Crossplain: Fine, smectitic, mesic Typic Argiaquolls

*Soil series and classification in bold print indicate the major soil sampled at that particular location.

**Soil map units, map unit names, major components, and soil classification information was attained from Web Soil Survey (USDA-NRCS, 2017).

Experimental Design:

This experiment was completed at the farm-scale, utilizing paired samples across treatments without making changes to producers' management programs.

Sample Collection:

At each site, a representative area was sampled based on soil survey maps from Web Soil Survey, field topography, and other indicators such as consistent residue cover and relation to wheel tracks. Four replicate samples were collected at each site. Replicates were collected in a triangular pattern where replicate one was in the center of the triangle, replicate two was the northernmost point, replicate three was the southwestern point, and replicate four was the southeastern point. Replicates two through four were spaced approximately 30m apart from Rep 1 at the center of the triangle (Figure 2.2). Latitude and longitude were collected at each replicate and photographs of the surrounding topography were taken at each site.

From each replicate, bulk samples were collected between the crop row at 0-10cm, 10-20cm, and 20-40cm using a spade. A 2mm sieve was used to collect aggregates in the surface 0-5cm. Soil samples for N analysis were collected at each depth in air-permeable bags to prevent N conversion to a nitrogen gas (e.g. N_2 or N_2O) from the anaerobic conditions that would arise in an airtight bag. An attempt was made to collect bulk density using a 6cm probe, but the ground was too hard and too dry to attain a sample with a hand probe.

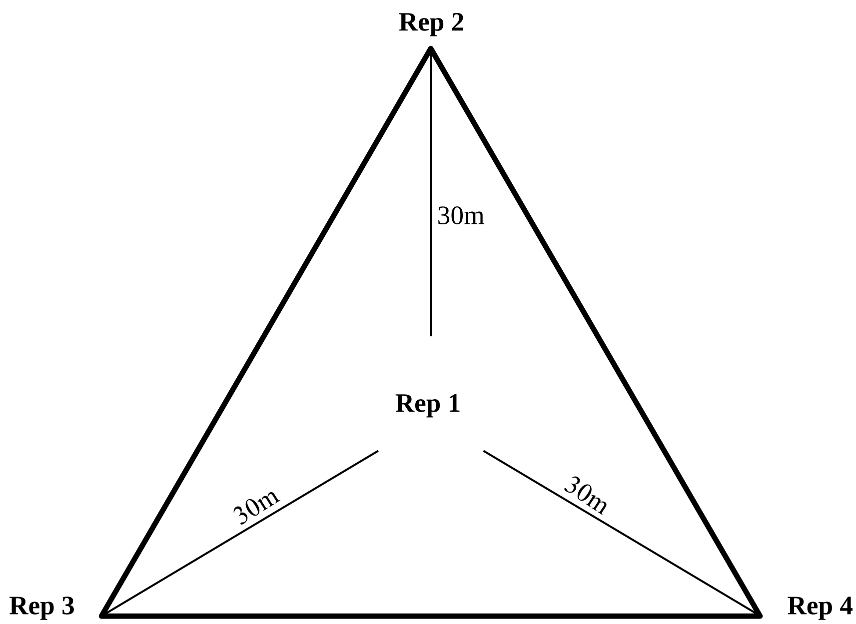

Figure 2.2. Illustration of design used for replicate collection.

Laboratory Analysis:

Rock fragments were removed from bulk samples and soil was ground to pass through a 2mm sieve using a soil grinder (Dynacrush Soil Crusher). Laboratory analyses included electrical conductivity (EC), pH, soil organic matter (SOM), soil inorganic carbon (SIC), total carbon (TC), total nitrogen (TN), water stable aggregates, particle size, and color.

Electrical Conductivity and pH

Soil EC was measured on a 1:1 soil:water basis following the protocol from The Recommended Chemical Soil Testing Procedures for the North Central Region publication (Whitney, 1998). For this analysis, 20 ± 0.1mL of water was added to 20.0 ± 0.1 grams (g) of ground soil in a 88.7mL Dixie cup. The soil solution was thoroughly stirred with a glass stir rod and allowed to rest for 30-35 minutes. After that period, the solution was stirred again and immediately measured using an EC meter. The reading was recorded after the meter stabilized and the number was consistent. Directly after reading EC, pH was measured with the same sample using a pH probe. The reading was recorded after the probe stabilized and the number was consistent. Two replicates of each sample were completed.

Soil Organic Matter

SOM was measured via loss on ignition as per the SDSU Soil Testing and Plant Analysis Laboratory protocol (2006). Three 50-sample racks of 10mL crucibles were weighed prior to analysis. Using a scale with three significant figures accuracy, 5.00 ± 0.1g of ground soil was weighed into the 10mL crucibles. Check soils were interspersed every 12 samples to ensure even heating in the furnace. Samples were placed in a muffle furnace (Lindberg Hevi-Duty BPC) at 100°C for two hours to remove water content. Crucibles were removed from the furnace and allowed to cool for 10 minutes to prevent heat from inhibiting scale function. Crucibles were then weighed to 0.001g to calculate weight loss due to moisture content. When analyzing one to two racks, samples were returned to the muffle furnace for 2 hours and 10 minutes at 375°C. When analyzing three racks, samples were returned to the muffle furnace for 2 hours and 10 minutes at 400°C. At the end of the second heating cycle, the furnace door was cracked open to allow heat to escape slowly which prevented cracking of crucibles due to sudden temperature changes. Once removed from the furnace, samples were allowed to cool to room temperature and were weighed to 0.001g. Using Equations 2.1 to 2.3, SOM was determined. A conversion factor of 1.724 was used to convert SOM to SOC (Pribyl, 2010).

Equation 2.1a:

$$\text{Empty Crucible Weight (g)} - \text{Crucible Weight after } 100°\text{C (g)} = \text{Heated Weight (g) (HT)}$$

Equation 2.1b:

$$\text{Empty Crucible Weight (g)} - \text{Crucible Weight after } 375°\text{C or } 400°\text{C (g)} = \text{Ignition Weight (g) (IG)}$$

Equation 2.2a:

$$\text{HT} - \text{IG} = \text{Weight Loss (g)}$$

Equation 2.2b:

$$\left(\frac{\text{Weight Loss (g)}}{\text{HT}}\right) \times 100 = \%\ \text{Weight Loss}$$

Equation 2.3:

$$a + b\ (\%\ \text{Weight Loss}) = \%\ \text{SOM}$$

Note: Coefficients “a” and “b” are predetermined coefficients built into the SOM spreadsheet offered by the SDSU Soil Testing and Plant Analysis Laboratory.

Soil Inorganic Carbon

Soil inorganic carbon (SIC) was measured via gravimetric loss of carbonate ion (CO_3^{-2}) as per the SDSU Soil Testing and Plant Analysis Laboratory protocol (2006). Forty 125mL Erlenmeyer flasks were numbered from 1through 40 and weighed with a correspondingly numbered rubber stopper. To each flask, 10mL of 3 N (3N) HCL was added and the flask was reweighed. In a plastic weigh boat, 5.00 ± 0.01g of soil was weighed and the weight was recorded. Once all initial weights were recorded, the soil was added to the flask and plugged with the rubber stopper. After one hour, the samples were opened to the air and swirled for 12 seconds to allow CO_2 to dissipate. Samples were weighed, re-stoppered, and allowed to react overnight. The following day, flasks were weighed and the $CaCO_3$ equivalent was calculated using Equations 2.4 to 2.6.

Equation 2.4:

$$\text{Initial weight (g)} - \text{Final weight (g)} = \text{Weight of } CO_3{}^{-2} \text{ (carbonate ion) broken down}$$

Equation 2.5:

$$\text{Weight of } CO_3{}^{-2} \text{ evolved x } \frac{\text{Atomic weight of carbon}}{\text{Atomic weight of } CO_3{}^{-2}} = \text{Weight of carbon evolved}$$

Equation 2.6:

$$\frac{\text{Weight of carbon evolved}}{\text{Weight of sample}} = \text{\% soil inorganic carbon}$$

Total Carbon and Total Nitrogen

Ward Labs in Kearney, Nebraska analyzed all samples for TC and TN by CN Analyzer.

Water Stable Aggregates

The following protocol is based on work done by Yoder (1936) concerning wet sieving for aggregate stability analysis. Upon returning to the laboratory from the field, aggregates were gently transferred to aluminum pans that were open to the air.

Aggregates were dried at room temperature overnight. Four grams of air-dry samples were placed in a copper cylinder with a 0.25mm mesh sieve soldered to the bottom. The cylinders were labeled with a letter and a number from one through eight (i.e. A1, A2, A3…A8). Samples were gently tapped on the lab bench surface to remove fine, non-aggregate materials without damaging the soil aggregates. Correspondingly, labeled metal tins were filled approximately three quarters full with deionized (DI) water. Plastic 50mL beakers were filled with 35mL of DI water.

Samples were then placed in a humidifier to saturate. Aggregates were properly saturated once a shiny sheen was visible on the sample surface. Cylinders with saturated aggregates were transferred to a tray which suspended them above the metal tins filled with water. A motor gently submerged the copper sieves at a rate of 40 revolutions per minute for five minutes. Metal tins were transferred to the oven to dry at 105°C overnight; the soil collected in the tins represented the unstable aggregates. The sieves containing the stable aggregates were gently placed in the plastic 50mL beaker. Using a 550 Sonic Dismembrator (Fisher Scientific), samples were homogenized for 20 seconds. Sieves were removed and soil was gently washed into the 50mL beaker. Beakers were transferred to the oven to dry at 105°C overnight.

Once soil in the tins and beakers were completely dry, the samples were removed to a desiccation box to cool without risking the samples absorbing moisture from the air. Tins and beakers were allowed to cool for 10 minutes and then were weighed. Percent stable aggregates were calculated using Equation 2.7.

Equation 2.7:

$$\frac{\text{Stable Aggregates (g)}}{\text{Stable+Unstable Aggregates (g)}} \times 100 = \text{ \% Stable Aggregates}$$

Particle Size

Prior to determining textural class via the hydrometer method, organic matter was removed from the samples with concentrated 30% hydrogen peroxide (H_2O_2) (Malo et. al., 2014). To remove organic matter, 60.0 ± 0.1g of soil was weighed into a pre-weighed 1-L Pyrex bottle and placed under the fume hood. Between 10 and 20 drops of n-octanol were added to the soil surface to control foaming during the oxidation reaction. Using an automatic dispenser, 20mL of 30% H_2O_2 was added to the 1-L Pyrex bottle and closely monitored to prevent the solution from frothing over the rim. Once the reaction slowed, another 20mL of H_2O_2 was added to the 1-L Pyrex bottle. At this point, six to eight drops of glacial acetic acid were added to the 1-L Pyrex bottle and the 1-L Pyrex bottle was then placed on a hot plate set at 350°C. The bottles were monitored closely to prevent frothing over of the soil:H_2O_2 solution and to prevent soil from burning to the bottom of the bottle. Periodically, H_2O_2 was added in 15mL increments until all SOM had been removed (observed by color change of sample). Samples were heated and maintained this way for one to two hours or until the reaction had gone to completion (Malo et. al., 2014). The bottles were then transferred to the oven to dry at 105°C overnight.

The following day, 1-L Pyrex bottles were removed from the oven, placed in desiccation chambers to cool for approximately 30 minutes, and weighed. After recording the weight, 30mL of dispersion solution (sodium hexametaphosphate and sodium carbonate) was added to the Pyrex bottles and allowed to react for 30 min to one hour.

From here, the bottles were filled to the 500mL line with DI water, tightly capped, placed in cloth bags to prevent damage to the glass, and arranged on a reciprocating shaker (40 cycles $min^{-}1$) to shake overnight (Malo et. al., 2014).

Samples were transferred to 1-L graduated cylinders the next day. Soil texture was measured via hydrometer as per the SDSU Testing and Plant Analysis Laboratory protocol (2006). Once all soil particles were rinsed from the Pyrex bottle and transferred to the cylinder, the volume was filled to the one liter line with DI water. A wooden plunger fitted with a plastic disc at the bottom was used to homogenize the sample for 60 seconds before beginning the hydrometer readings. The exact time was recorded as soon as the plunger was removed. A glass hydrometer was inserted into the cylinder and the first reading was taken 40 seconds after plunging. The hydrometer was then removed and an instant read thermometer was used to take the temperature of the solution. The solution was then plunged again for 60 seconds and the exact time was recorded. After two hours, a second hydrometer reading was taken and temperature was again recorded. Using a 53µm sieve, very fine to coarse sands were retained from the texture samples. Sands were rinsed with DI water and placed in the oven to dry at 105°C overnight. The following day, the sand was weighed and used to calculate the percent sand within the sample.

Sand, silt, and clay fractions were calculated using Equations 2.8 to 2.17. A soil textural triangle (Soil Science Division Staff, 2017) was used to determine texture class. When textures fell on a textural class boundary, (i.e. loam and silt loam boundary) the finer texture was used.

Equation 2.8:

$$40 \text{ Second Measured Temperature } (°\text{C}) - 20\ °\text{C} = $$
$$40 \text{ Second Temperature Difference } (°\text{C})$$

Equation 2.9:

$$40 \text{ Second Temperature Difference } (°\text{C}) \times 0.36 = $$
$$40 \text{ Second Temperature Correction Factor}$$

Equation 2.10:

$$\text{Hydrometer Reading} + 40 \text{ Second Temperature Correction Factor} = $$
$$\text{Silt and Clay (g)}$$

Equation 2.11:

$$\text{Sample weight (g)} - \text{Silt and Clay (g)} = \text{Sand (g)}$$

Equation 2.12:

$$\frac{\text{Sand (g)}}{\text{Sample weigt (g)}} \times 100 = \ \%\ \text{Sand}$$

Equation 2.13:

$$2 \text{ Hour Measured Temperature } (°\text{C}) - 20\ (°\text{C}) = $$
$$2 \text{ Hour Temperature Difference } (°\text{C})$$

Equation 2.14:

$$2 \text{ Hour Temperature Difference } (°\text{C}) \times 0.36 = $$
$$2 \text{ Hour Temperature Correction Factor } (°\text{C})$$

<u>Equation 2.15:</u>

$$\text{2 Hour Hydrometer Reading} - \text{2 Hour Temperature Correction Factor (°C)} = \text{Clay (g)}$$

<u>Equation 2.16:</u>

$$\frac{\text{Clay (g)}}{\text{Sample Weight (g)}} \times 100 = \text{ \% Clay}$$

<u>Equation 2.17:</u>

$$100\% - \text{\% Sand} - \text{\% Clay} = \text{\% Silt}$$

<u>Soil Color</u>

Soil color was analyzed with a Munsell Color Book as per the USDA-NRCS protocol (Soil Survey Staff, 2009). Color was assessed for dry and moist soils. For the surface depth, samples were mixed whereas for the 10-20 cm and 20-40 cm depth a large ped was broken in half and the predominant ped interior color was determined. With natural sunlight, dry samples were compared to color chips beginning on the 10YR page. Once the sample was paired with the chip most closely resembling its color on the 10YR page, it was also compared to the 7.5YR and 2.5Y pages to determine the best match. The most closely matching Hue, Value, and Chroma were recorded for the ped or mixed sample. The process was repeated again after using a spray bottle to gently moisten the soil. Mollic colors for dry soil are defined as Values less than or equal to five with Chromas less than or equal to three (Soil Survey Staff, 1999). Mollic colors for moist soil are defined as Values and Chromas that are less than four (Soil Survey Staff, 1999).

Statistical Analysis:

Descriptive statistics were completed using R 3.5.0 (R Core Team, 2018). One-way Analysis of Variance (ANOVA) and Tukey Multiple Comparison Tests were completed in JMP (SAS Institute Cary, NC). One-way ANOVA was used to validate the data with published literature and to identify factor-level significance. The three assumptions that must be met for ANOVA are that the data was collected from an independent and random sample, there is equal variance between treatments, and the data is approximately normally distributed (Freund et. al., 2010). To assess these assumptions, the Shapiro-Wilk Normality Test was completed and the residuals were plotted to ensure equal variance (Freund et. al., 2010).

One-way ANOVA will identify the presence of significant differences within factor-levels. To determine specific pairwise significant differences, the Tukey Multiple Comparison Test was used. Assumptions for the Tukey Multiple Comparison Test are the same as for ANOVA (Freund et. al., 2010). Post-hoc tests which simultaneously compare all pairwise combinations, such as the Tukey Multiple Comparison Test, reduce the risk of committing a Type 1 error in which the null hypothesis (no significance) would be improperly rejected (Berry, 2007).

Multiple linear regression (MLR) analysis was conducted in R 3.5.0 utilizing the “lm” function in Base R (R Core Team, 2018). The Variance Inflation Factor was assessed using the “car” package (Fox and Weisberg, 2011) and stepwise model selection was completed using the “olsrr” package (Hebbali, 2018). The “dplyr” (Wickham et. al., 2018) package was used to filter the data to create the filtered models. The MLR model is defined as $y = \beta_0 + \beta_i X_i \ldots + \beta_k X_k + \varepsilon$ which is explained below (Freund et. al., 2010). Assumptions for MLR include approximately normally distributed error terms, lack of outliers in the data set, and equal variance of the residuals (Freund et. al., 2010). For all statistical analysis, alpha was set at 0.05.

y = Response Variable

β_0 = Intercept. This is the value of the line when all independent variables are equal to zero. Although it is not realistic to expect all independent variables to ever equal zero, the intercept is necessary to define the model.

$\beta_i X_{\mathrm{i}}$ = Partial Regression Coefficient x Corresponding Independent Variable. The partial regression coefficient pertains to the slope associated with a unit increase in the independent variable when all other variables in the model are held constant. i = 1, 2 …k.

$\beta_k X_k$ = Denotes the same relationship above for slope multiplied by a unit increase in the independent variable through variable “k”

ε = Error. The error terms are assumed to have a normal distribution, with a mean of zero, and equal variance.

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CHAPTER 3: RESULTS AND DISCUSSION

Descriptive Statistics:

One-way Analysis of Variance (ANOVA) and Tukey Multiple Comparison Tests (Freund et. al., 2010) were conducted to ascertain significant differences between factors. The main interactions evaluated were the effect of depth and treatment on soil properties. All data was analyzed at the 95% confidence interval (α=0.05; p<0.05). Lab data and metadata concerning management practices that were included in this study are available in Appendix A. Boxplots illustrating the distribution of SOC data related to the management practices included in this study can be found in Appendix B.

Effect of Depth and Treatment on Soil Properties

The soil properties included in this analysis include soil pH, EC, TN, TC, and SOC. Aggregate stability was analyzed by treatment alone as those samples were collected from the surface depth only. Figure 3.1 illustrates the effect of depth and treatment on soil pH. Soil pH was significantly more acidic in the 0-10cm depth than the 20-40cm depth (p=0.0004), but there was no significant difference between the 10-20cm and 20-40cm depths. No-Till (NT) treatments were significantly more acidic than native conditions (NTVG) at each depth ($p_{0\text{-}10cm}$=0.0008, $p_{10\text{-}20cm}$=0.0008, $p_{20\text{-}40cm}$=0.0012). There was no significant difference between conventional tillage (CT) and either the NT or NTVG treatment at any depth. Over time, No-Till management leads to increases in soil acidity, likely to due to nitrogen fertilizer additions and increased microbial activity (Belvins et. al., 1977; Dick, 1983), which was similarly observed in this data set.

Electrical conductivity was not affected by depth, but treatment did have a significant effect on EC (Figure 3.2). All soils exhibited a low EC which is typical of upland parent materials commonly found in southeastern South Daktoa (Malo et. al., 2005). There was no significant difference in EC between NT and NTVG treatments at any depth. At the 0-10cm depth, CT treatments had significantly higher EC values than NT (p=0.0021) or than NTVG (p=0.0005). Similarly at the 10-20cm depth, CT treatments had significanlty higher EC values than NT (p=0.0009) or NTVG (p=0.0028). Again, at the 20-40cm depth, EC was significanlty higher in CT treatments than in NT treatments (p=0.0088) or NTVG (p=0.0058). Greater EC levels in the CT treatments could be due to erosion processes removing topsoil and exposing salts from less weathered soil parent materials that were originally located deeper in the profile (Malo et. al., 2005).

Depth and treatment had significant effects on TN (Figure 3.3). Each increase in depth resulted in a significant decrease in TN (p<0.0001). At all depths, NTVG had significantly higher levels of TN than CT or NT (p<0.0001). There is no significant difference between CT and NT at the 0-10cm depth or the 20-40cm depth. Total nitrogen was greater in CT treatments than NT treatments at the 10-20cm depth (p=0.0018). Nitrogen is a fuel source for soil microbial activity and can lead to accelerated decomposition of C when in abundant supply (Guo et. al., 2016). There may have been less TN at the 10-20cm depth in NT treatments due to increased microbial respiration, less residue incorporation, and higher consumption of existing organic residues, and less mixing of N fertilizers in the NT when compared to the CT treatment (Zhao et. al., 2018).

Total carbon was significantly affected by depth and by treatment (Figure 3.4). Similar to TN, with each increase in depth, there was a significant decline in TC ($p<0.0001$). Total carbon was significantly higher in NTVG treatments than CT or NT at the 0-10cm depth ($p<0.0001$). At the 0-10cm depth, CT treatments had significantly higher levels of TC than NT ($p=0.0021$). Again, NTVG had significantly greater levels of TC than CT or NT at the 10-20cm depth ($p<0.0001$). Also, CT had significantly greater levels of TC than NT at the 10-20cm depth ($p=0.0012$). In the 20-40cm depth, NTVG had significantly greater levels of TC than CT ($p=0.0006$) or NT ($p<0.0001$). At the 20-40cm depth, there was no significant difference in TC between CT and NT. It was interesting to find that the NT treatments had significantly less TC than CT treatments at 0-10cm and 10-20cm. Potentially, the higher levels of TN observed in the 0-10cm depth would lead to higher levels of microbial decomposition of SOC and result in lower TC at the 0-10cm depth (Zhao et. al., 2018).

Soil organic carbon was significantly affected by depth and by treatment (Figure 3.5). Each increase in depth led to a decline in SOC ($p<0.0001$). At each depth, NTVG had significantly greater levels of SOC than CT or NT ($p<0.0001$). There was no significant difference between CT and NT treatments at any depth. It is worth noting that SIC was not significantly affected by either depth or by treatment. Although it was hypothesized that there would be significantly more SOC in NT treatments than CT, that was not observed by this data. Converting ground to NT management may not be enough to build SOC levels past that observed in conventional tillage management (Blanco-Canqui and Lal, 2008). Bulk density is a very important factor when assessing SOC concentrations or stocks, and the lack of bulk density information could explain some of the relationship observed here (Ruis et. al., 2018).

Aggregate stability was not significantly different between the three treatments. There was not enough data for aggregate stability to be included in model construction. The relationship between SOC and various soil properties was also analyzed. No strong relationships amongst the soil properties tested (texture, color, etc.) and SOC existed in this data set. For instance, the r^2 for SOC and clay content was 0.107 which does not support the strong linear relationship between clay and SOC that Six et. al. (2002) found in their research. Given the narrow range of clay content within the study data set (24 to 40%), the likelihood of finding similar results to Six et. al. (2002) may have been reduced. Total nitrogen was the one soil property that did have a strong linear relationship with SOC (r^2=0.811). Total nitrogen is an integral component of organic matter composition. Therefore, TN was not included in the SOC prediction models as that would have inherently introduced multicollinearity into the model.

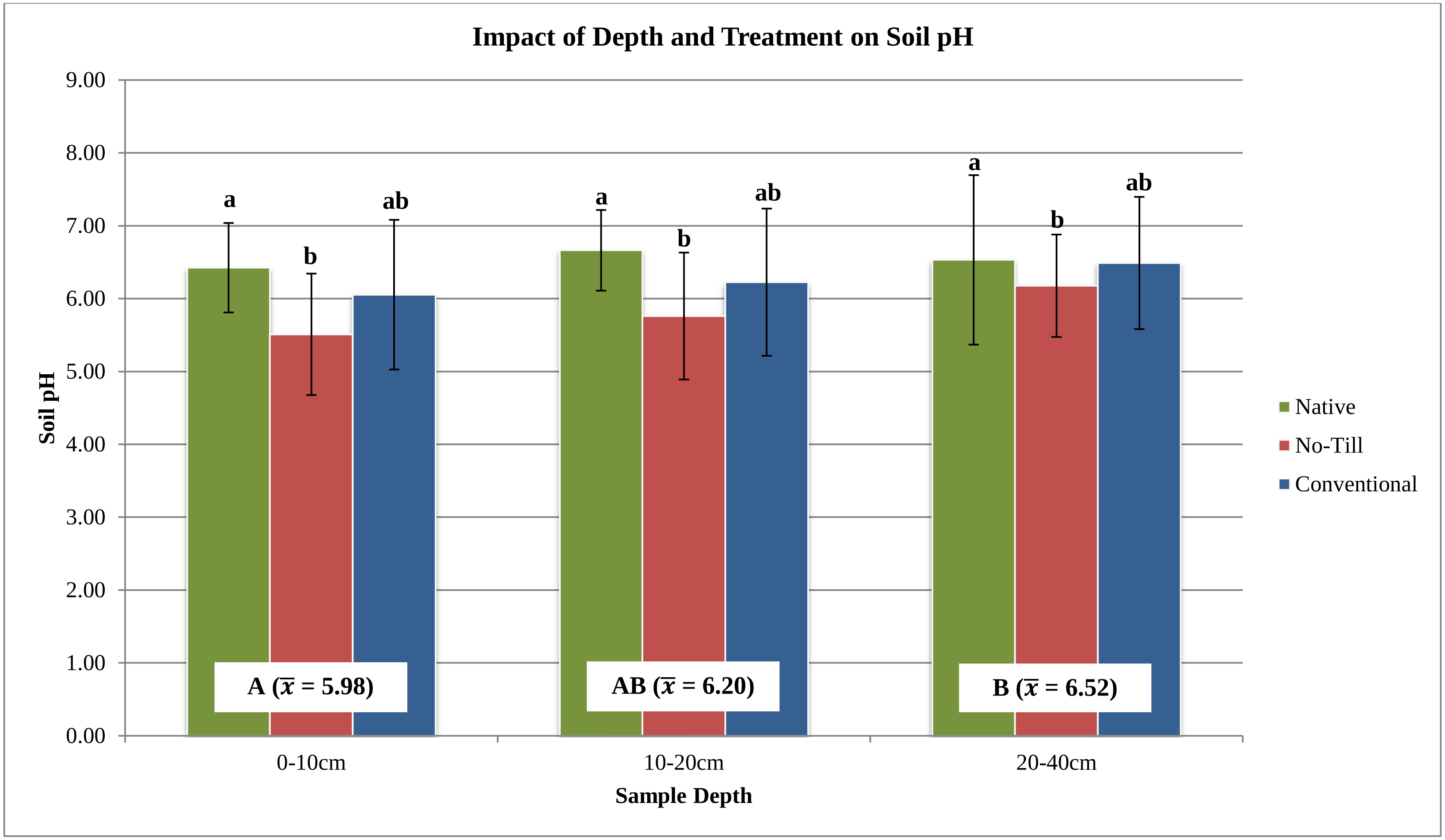

Figure 3.1: Effect of depth and treatment on soil pH. Capital letters at the base of the bars denote significance due to depth ($\alpha = 0.05$). Lowercase letters above bars indicate significance between treatments at a specific depth ($\alpha = 0.05$). Black bars below lowercase letters illustrate the standard deviation of the data. At each depth, n=28 for NTVG, n=32 for NT, and n=32 for CT. Soils included in this analysis were Haplustolls.

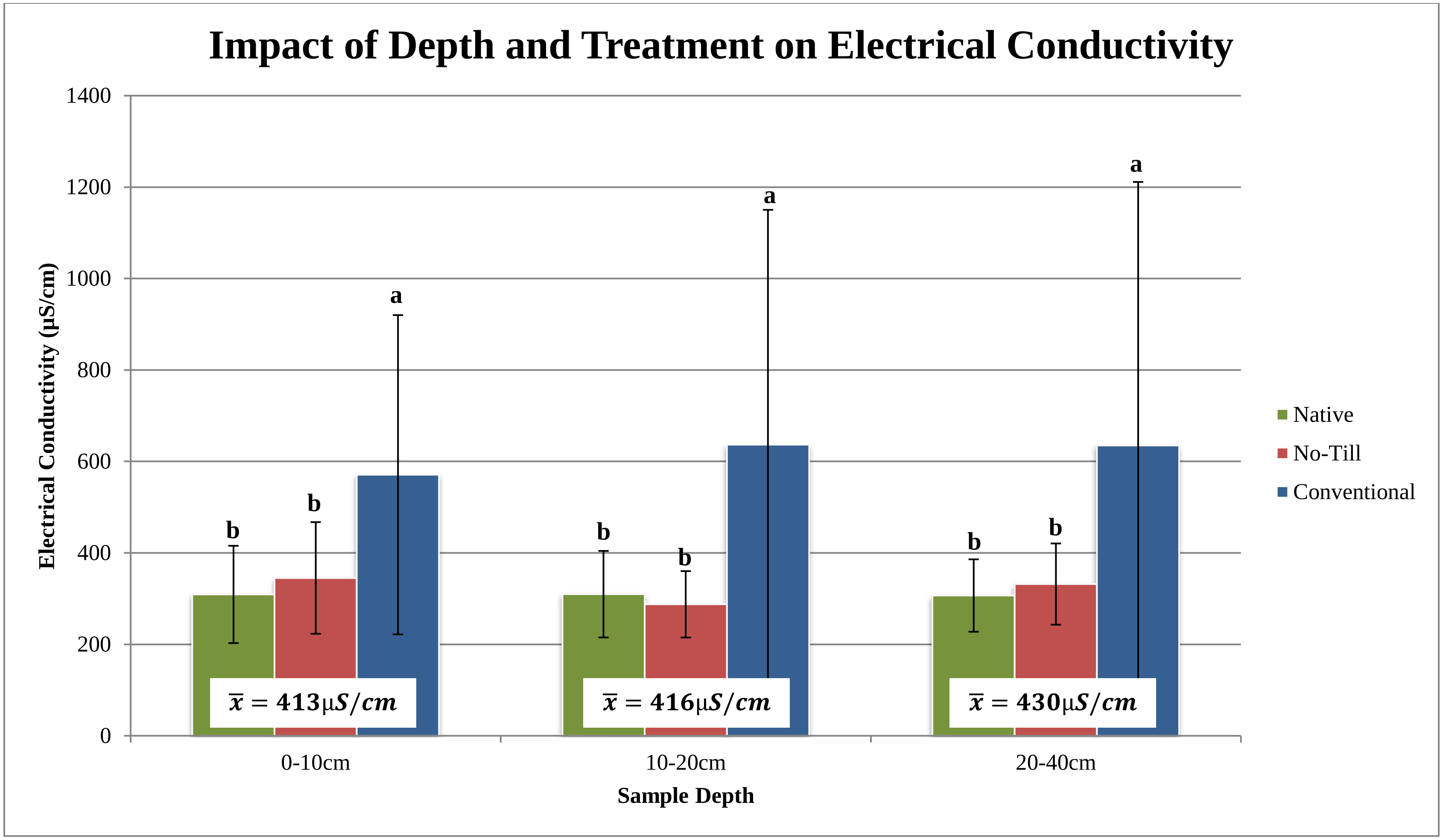

Figure 3.2: Effect of depth and treatment on soil EC. Depth did not have a significant effect on electrical conductivity. Lowercase letters above bars indicate significance between treatments at a specific depth when $\alpha = 0.05$. Black bars below lowercase letters illustrate the standard deviation of the data. At each depth, n=28 for NTVG, n=32 for NT, and n=32 for CT. Soils included in this analysis were Haplustolls.

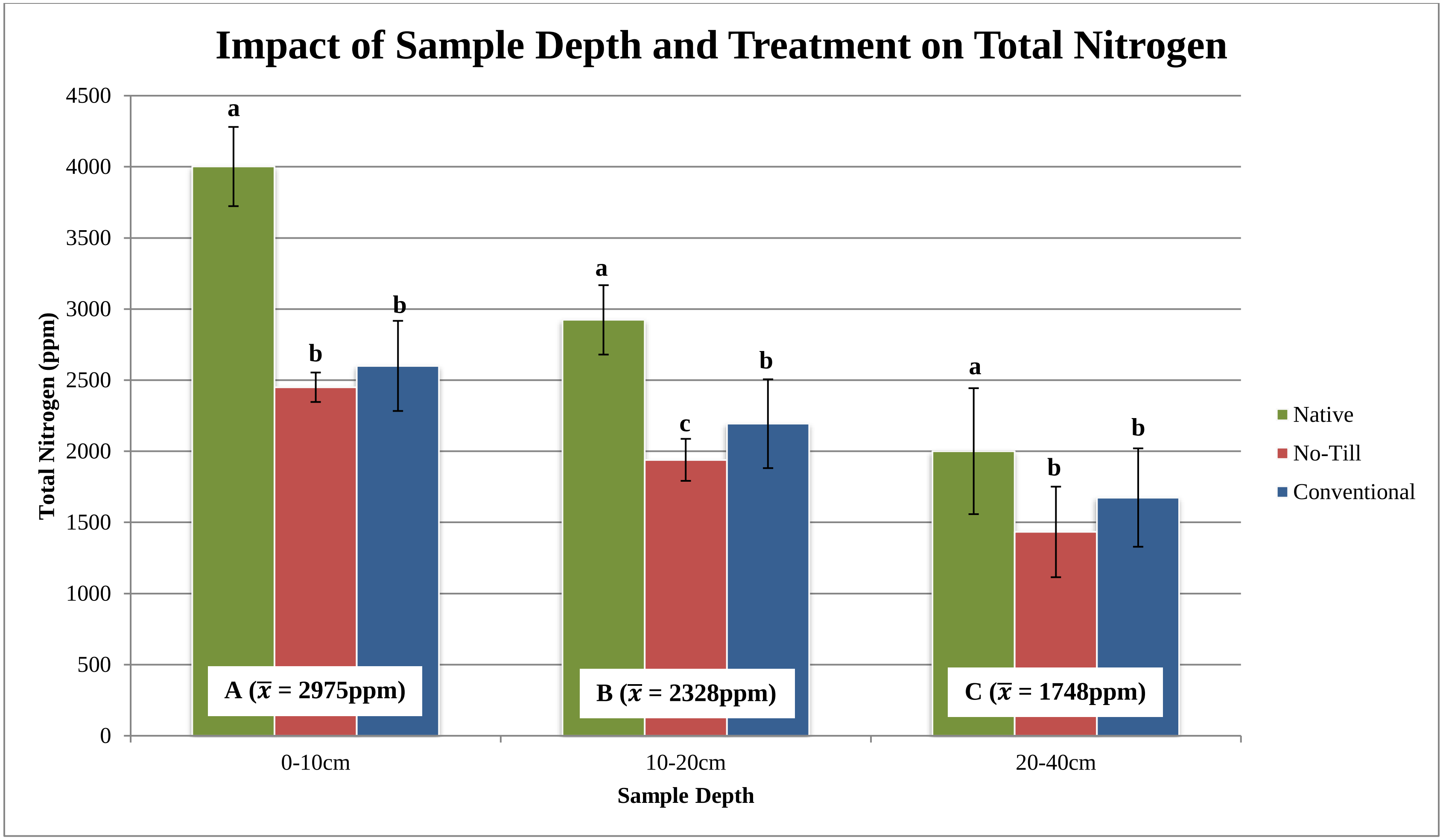

Figure 3.3: Effect of depth and treatment on total nitrogen. Capital letters at the base of the bars denotes significance due to depth at α = 0.05. Lowercase letters above bars indicate significance between treatments at a specific depth when α = 0.05. Black bars below lowercase letters illustrate the standard deviation of the data. At each depth, n=28 for NTVG, n=32 for NT, and n=32 for CT. Soils included in this analysis were Haplustolls.

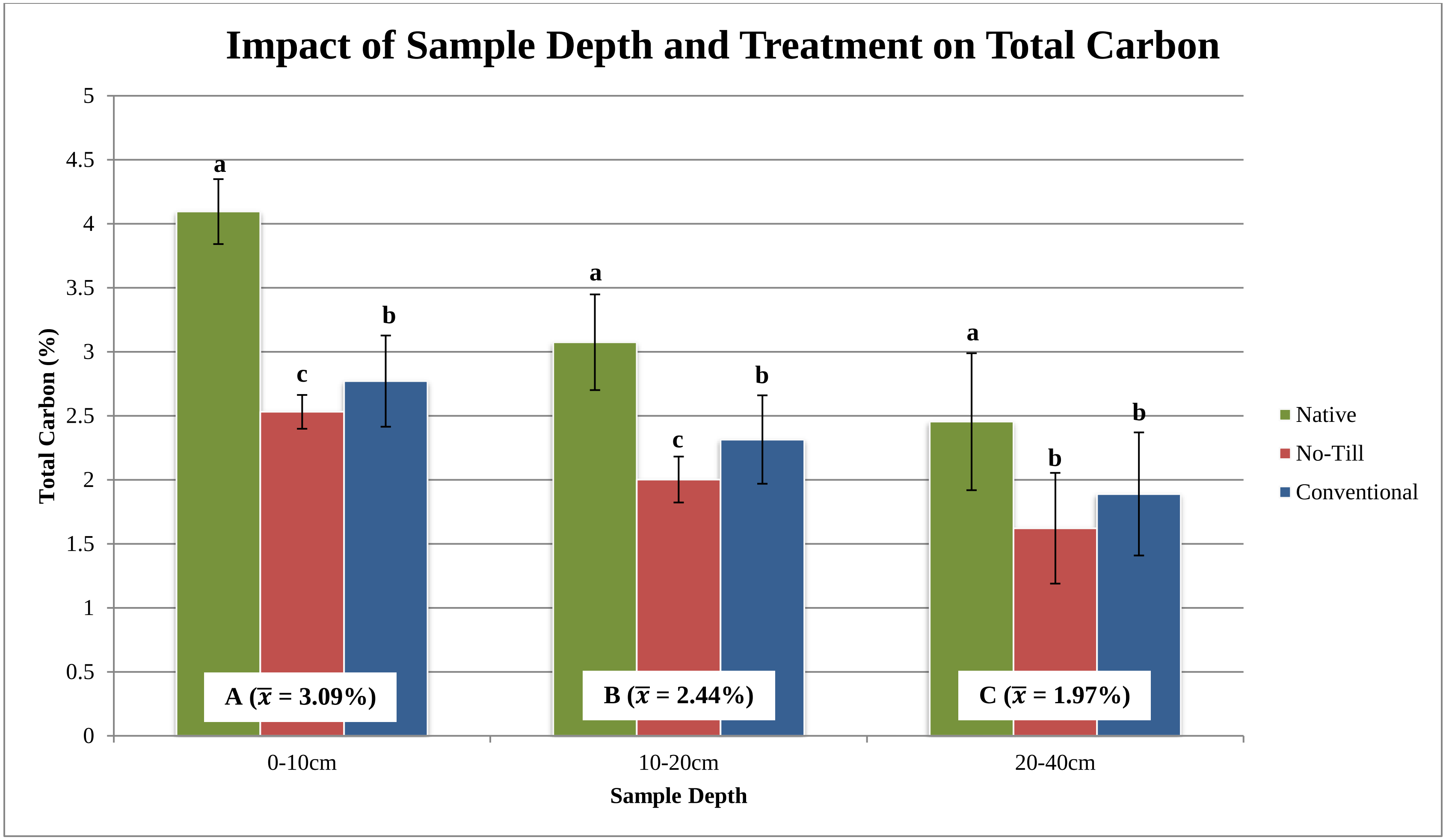

Figure 3.4: Effect of depth and treatment on total carbon. Capital letters at the base of the bars denotes significance due to depth at α = 0.05. Lowercase letters above bars indicate significance between treatments at a specific depth when α = 0.05. Black bars below lowercase letters illustrate the standard deviation of the data. At each depth, n=28 for NTVG, n=32 for NT, and n=32 for CT. Soils included in this analysis were Haplustolls.

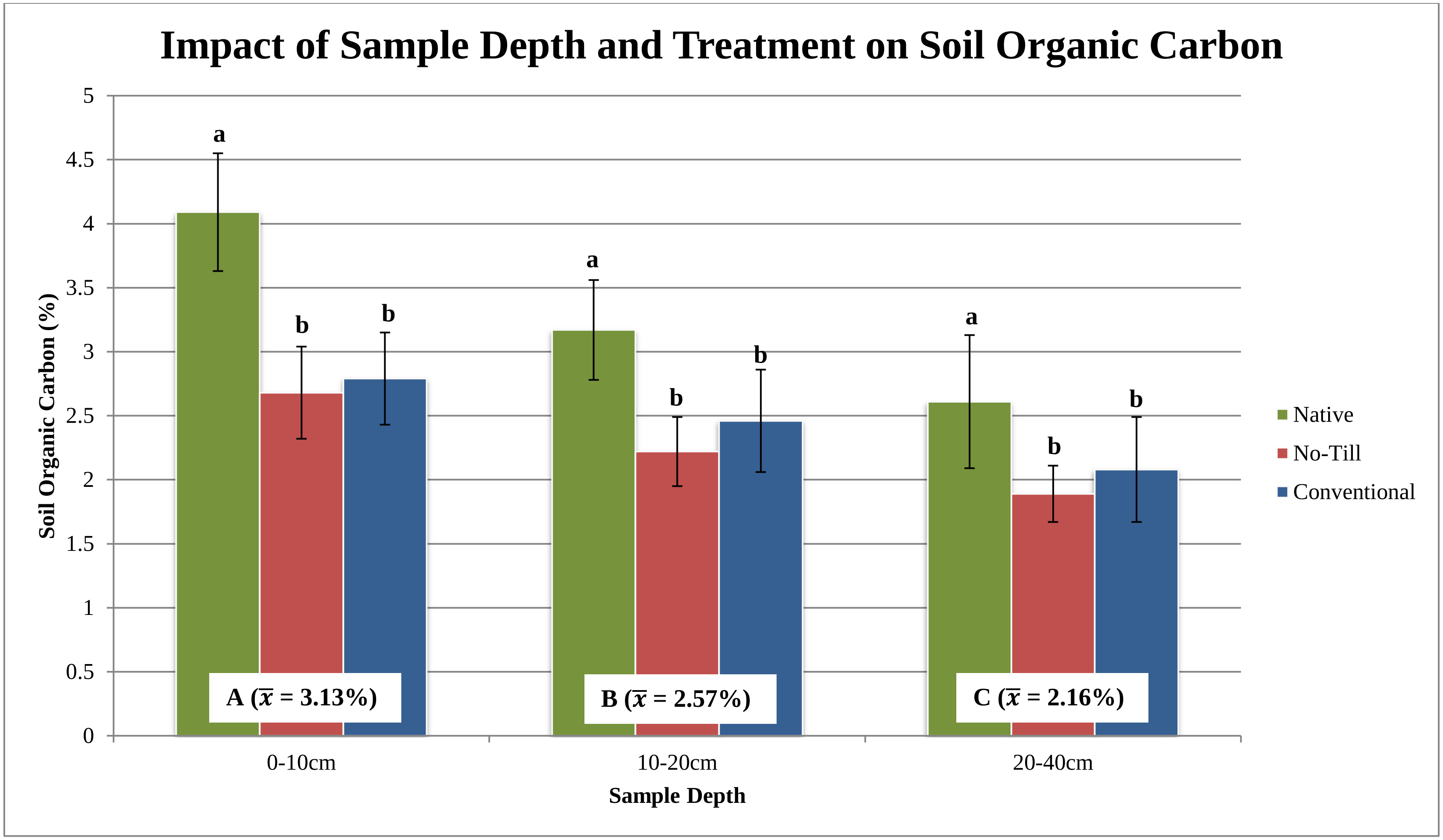

Figure 3.5: Effect of depth and treatment on soil organic carbon. Capital letters at the base of the bars denotes significance due to depth at α = 0.05. Lowercase letters above bars indicate significance between treatments at a specific depth when α = 0.05. Black bars below lowercase letters illustrate the standard deviation of the data. At each depth, n=28 for NTVG, n=32 for NT, and n=32 for CT. Soils included in this analysis were Haplustolls.

Model Construction:

Multiple linear regression (MLR), stepwise selection, and data filtration were used to produce the included SOC prediction models. Assumptions for MLR require approximately normally distributed error terms, lack of outliers in the data set, and equal variance of the residuals (Freund et. al., 2010). Variables included in the MLR full model include Depth (defined as a factor), Treatment (factor), Tillage (factor), Rotation (factor), Years in NT (numeric/integer), Dry Soil Color (factor), Moist Soil Color (factor), Mean Annual Moisture (numeric/integer), Mean Annual Temperature (numeric/integer), Sand Content (numeric/integer), Clay Content (numeric/integer), Soil pH (numeric/integer), Electrical Conductivity (numeric/integer), and Soil Inorganic Carbon (numeric/integer).

Multiple Linear Regression, Stepwise Selection, and Filtered Models

Using the linear model (lm) function in R 3.5.0 (R Core Team, 2018), all variables were assessed via MLR. The resulting model from this analysis is expressed in Equation 3.1. The Shapiro-Wilk normality test is a method of testing the data set for irregularities; if the p-value from the test is less than that set for the study, the assumptions for MLR are not met (Freund et. al., 2010). In this study, the Shapiro-Wilk p-value had to be greater than 0.05. For the full model, the Shapiro-Wilk test was acceptable (p=0.077) and the residuals were approximately normal (see Appendix B). Therefore, it was determined that the data set met the necessary assumptions of MLR. The adjusted R^2 value was equal to 0.678 which indicates that approximately 68% of the variation in SOC was explained by the model. The Variance Inflation Factor (VIF) is a method of detecting multicollinearity in which one of the independent variables included in the model is correlated to another independent variable (Freund et. al., 2010). The VIF is defined below in Equation 3.2 (Craney and Surles, 2002). According to Craney and Surles (2002), the generally accepted VIF value is ≤10. The resulting VIF for the full model ranged between 1.05 and 12.50. Based on the VIF, it is evident that the full model may have multicollinearity issues.

Equation 3.1: Full model as determined by MLR in R 3.5.0.

SOC = 4.078 – 1.95(Conventional Tillage) – 0.551(10-20cm Depth) – 0.926(20-40cm Depth) – 0.562(Tillage) + 0.168(Rotation) + 0.052(Moisture) – 0.300(Temperature) – 0.259(Soil Inorganic Carbon).

Equation 3.2: Variance Inflation Factor (VIF)

$$\mathrm{VIF} = \frac{1}{1-r_i^2}$$

r_i^2 is the Pearson Correlation Coefficient between two independent variables

Due to potential multicollinearity in the full model, stepwise variable selection was utilized to reanalyze the data set. Stepwise selection is the process by which all variables are entered into a statistical software program and the program systematically determines which relationships and variables are most significant to the overall model (Freund et. al., 2010). Results are presented in Appendix B for the stepwise regression of all the variables. There are multiple methods by which the most robust model can be chosen from the results of stepwise selection (Freund et. al., 2010). For this study, the combination of variables with the largest adjusted R^2 value and the smallest Akaike Information Criteria (AIC) (Freund et. al., 2010) was selected as the best model for the current data set. Equation 3.3 represents the reduced model predicted by stepwise selection in R 3.5.0. The Shapiro-Wilk test did not yield acceptable results (p=0.0398), the plotted residuals indicated potential outliers, and the VIF ranged between 1.02 and 11.37 (Appendix B). For the reduced model, the adjusted R^2 was 0.682 which indicates that approximately 68% of the variation in SOC was explained by the model. Tillage was the factor with an unacceptable VIF of 11.37. Due to the issues with this model, Tillage was removed and the linear model function was run on the data again. Equation 3.4 represents the amended reduced model. The Shapiro-Wilk test again resulted in an unacceptable p-value (p=0.0230) and the plotted residuals suggested potential outliers in the data (Appendix B). The VIF ranged between 1.01 and 2.27 which was acceptable. The Adjusted R^2 was 0.674 which indicates that approximately 67% of the variation in SOC was explained by this model. Given the current data set, this was the best stepwise model that could be constructed without multicollinearity when assessing the entire data set.

<u>Equation 3.3:</u> Reduced model produced by stepwise regression in R 3.5.0.

SOC = 4.94 - 2.16(Conventional Tillage) - 0.972(No-Tillage) - 0.547(10-20cm Depth) -0.920(20-40cm Depth) - 0.703(Tillage) + 0.205(Rotation) + 0.0482(Moisture) -0.234(Temperature) - 0.215(Soil Inorganic Carbon)

<u>Equation 3.4:</u> Reduced model produced by stepwise regression after removing Tillage. Model produced by R 3.5.0.

SOC = 3.25 - 0.811(Conventional Tillage) - 0.939(No-Tillage) - 0.548(10-20 Depth) -0.918(20-40cm Depth) + 0.0396(Moisture) - 0.288(Temperature)

The amended reduced model is more inclusive of the management variables assessed in this study. However, the overall objective of this research was to study the effect of management on SOC. Therefore, the model would be of more use if all management variables were included. For this reason, the data was filtered in specific combinations to account for all management variables. An example of a filtered model is represented in Equation 3.5 in which only data from NT treatments under a corn-soybean crop rotation were included in the MLR analysis. For this filtered model, the Shapiro-Wilks normality test was acceptable (p=0.969), the residuals were approximately normal (Appendix B), and the VIF ranged between 1.14 and 2.32 for all variables. The adjusted R^2 was 0.641 indicating that approximately 64% of the variation in SOC was explained by the model. This filtered model met all assumptions of MLR and did not exhibit multicollinearity. Other potential filters could be conventional tillage with fall/spring tillage management under corn-soybean rotation or perennial cover with No-Tillage under native conditions. Such filters would allow producers to tailor the application to their operation and account for all management combinations, as well as compare the results of their management to native conditions.

Equation 3.5: Filtered model utilizing only No-Till management and Corn-Soybean rotation data points.

SOC = -0.0885 - 0.473(10-20cm Depth) - 0.082(20-40cm Depth) + 0.067(Moisture) -0.267(Temperature) + 0.156(pH)

Electronic Application Development:

Information included in the following section was based on a personal conversation with Mr. Ahmed El-Magrous and Dr. Gary Hatfield in the South Dakota State University Mathematics and Statistics Department. Two kinds of electronic applications can be created for this data. Applications that are accessible by website and by mobile phone compose one category of electronic applications, while applications that are only accessible by mobile phone compose the other (El-Magrous, 2018). Websites are presented in a larger visual format, are accessible to more people, and may be less intimidating than strictly mobile applications (Billi et. al., 2010). For this reason, generating a website and mobile phone compatible application is the best option for this particular data set.

Electronic applications are a three step process beginning with data input, moving into data processing, and ending with data output (Figure 3.6). Data input can be constant or variable, meaning that the model components would change from use to use (El-Magrous, 2018). For this study, data input pertains to constant variables. Additionally, input can be completed automatically from an external website such as Web Soil Survey, manually with all inputs coming directly from the user, or a combination of both (El-Magrous, 2018). This application would utilize semi-automatic data input. Producers would select their geographic location on a map, and data from Web Soil Survey would automatically populate moisture, temperature, and texture information into the model. At this point, producers would have the option to review and verify the automatically filled variables. If they are unsatisfied with the information coming from Web Soil Survey, they can override the application and enter that information manually. Other components of the model would be manually entered by the producer such as management practices, pH, N, etc.

For data processing, the final application would only consist of one model (e.g. a suite of filtered models to account for all management variables or one stepwise model) that would be used on all queries. All processing occurs in the background using programs like R 3.5.0 and Visual Studio (Microsoft, 2018) (El-Magrous, 2018). The variables included in the model would be made clear to the producer so that they can see which components were significant in producing the results.

The data output would be a simple percentage with the option to convert SOC into SOM using a conversion factor of 1.724 (Pribyl, 2010). Another potential component to add to this application would be to give producers the option of using the model as a user or as a guest. Users would have a login/password and the application would store the results of their queries over time (El-Magrous, 2018). In this way, producers could revisit predictions the model made before they changed their management and assess the accuracy of the model over time. If producers did not want their data stored within the application, they could use the application as a guest and not maintain their results on that platform (El-Magrous, 2018).

Information pertaining to this study (i.e.: sample collection, laboratory analysis, statistical analysis) would be available within the application if producers were interested in how the model was created. Additional information to make available would include hyperlinks to Extension articles concerning best management practices, research conducted in the state of South Dakota, and general soil health information from various sources (e.g. NRCS). The goal is for this application to be very user-friendly, provide easy to interpret results, and offer follow-up information on various management changes producers could adopt to improve SOC levels on their operation.

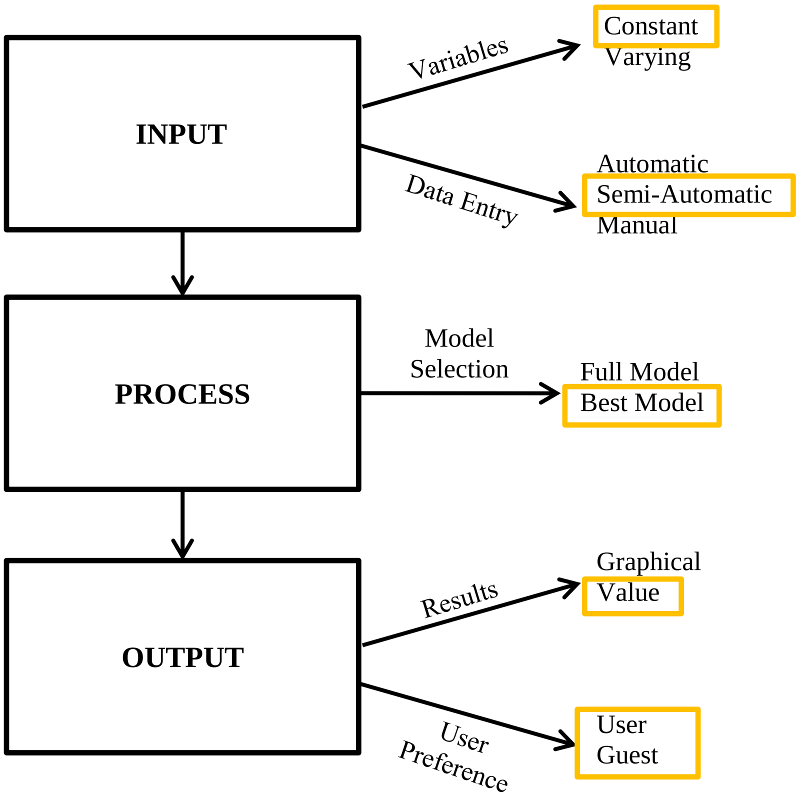

Figure 3.6: Visual representation pertaining to the steps involved in electronic application development and their components. (El-Magrous, 2018). Application components that are outlined in yellow boxes indicate the options that would be selected for this particular data set.

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CHAPTER 4: CONCLUSION

Conclusion:

Soil organic carbon is critical to the overall health and function of the soil ecosystem. Current models are research focused and difficult to interpret at a production scale. This research identified significant effects of management on SOC and on other soil properties in southeastern South Dakota. Given that management is a significant factor in SOC development, a simple SOC prediction model would be beneficial for producers in southeastern South Dakota and the surrounding region. Results from the model could assist producers in deciding to make significant changes to their management for the benefit of SOC accumulation on their operation.

Stepwise selection yielded a model that was free of multicollinearity and explained 67% of the variation in SOC, but analysis suggested potential outliers in the data set. The variables included in this model include Conventional Tillage, No-Tillage, Depth at 10-20cm and 20-40cm, Mean Annual Precipitation (Moisture), and Mean Annual Temperature. Filtered models have been proposed as a means to assess the impact of all management variables on SOC. One proposed filtered model utilizes No-Till management and corn-soybean rotation data points. The resulting model included Depth, Moisture, Temperature, and pH. This model produced an acceptable Shapiro-Wilk *p*-value, displayed approximately normal residuals, and did not exhibit multicollinearity. Approximately 64% of the variation in SOC was explained by the model. Based upon these results, filtering the data set is an appropriate method for data analysis and model construction.

Additionally, producing a user-friendly mobile application for the dissemination of this data is highly feasible. Working with experts in the South Dakota State University Mathematics and Statistics Department has yielded a rough roadmap toward the development of an electronic application. Producers would be responsible for delineating their approximate geographic location, inputting management information, and information from soil sample results to receive SOC information for their operation. Results will be expressed in percent SOC and could easily be converted to SOM within the application. Another component of the app would be including hyperlinks to Extension and NRCS articles outlining SOC and SOM research completed in South Dakota and general soil health information. Background information concerning this study and the work that created the models would also be available within the application.

The most important conclusion to be made, however, is that more data is needed to improve this study. Many management combinations (i.e.: deep tillage with continuous corn) were under-sampled and there was insufficient information for model development. More sample locations need to be found and sampled to expand the data set, increase model accuracy, and verify the models produced by this data. Including more data points would improve the overall models proposed in this research and more adequately meet the assumptions for normality. Furthermore, resampling the eight locations included in this study over time could lead to the creation of different model types, such as equilibrium rate constants, rather than multiple linear regression models. Much work is needed to improve model accuracy and expand applicability before being made available to producers.

APPENDIX A

Supplemental Table A1: Specific management information for study sites.

Loc	Trt	Tillage: Type and Time	Crop Rotation	Years in No-Till	Manure	Residue Removal	Fertilizer Rec N-P-K-S	Tile Drain	Average Yield (bu/a)
1	CT	Field finisher in spring	Corn Soy	N/A	N	N	150-50-50-10 11-11-0-1	N	178 58
	NT	N/A	Corn Soy	10	N	N	140-80-60-15 0-60-60	N	195 70
	NTVG	N/A	N/A	100+	N	N	N/A	N	N/A
2	CT	Fall Disk & Spring Cultivator	Corn Soy	N/A	N	Cut hay	150-0-0-15 18-46-60	N	160 50
	NT	N/A	Corn Soy	10	N	N	150-0-0-15 18-46-60	N	160 50
	NTVG	N/A	N/A	100+	N	Cut for hay	N/A	N	N/A

Loc	Trt	Tillage: Type and Time	Crop Rotation	Years in No-Till	Manure	Residue Removal	Fertilizer Rec N-P-K-S	Tile Drain	Average Yield (bu/a)
3 & 4	CT	Fall Deep Tillage Spring Cultivator	Continuous Corn	N/A	N	N	150-50-40-20 & 20gallons 28%	N	203
	NT	N/A	Corn Soybean	29	N	N	Unknown	N	Unknown
	NTVG	N/A	N/A	100+	N	Grazed	N/A	N	N/A
5	CT	All spring: Corn years- disk twice, SB years cultivate once	Corn Soy	N/A	N	N	Two year blend: 150-50-50-10-2	Y	165 45
	NT	N/A	Corn Soy	21	N	N	Two year blend: 160-60-90-2	N	185 42
	NTVG	N/A	N/A	100+	N	N	N/A	N	N/A
6	NT	N/A	Corn Soy	26	N	N	Highly variable	Y	140 47
	CT	N/A	Corn Soy	26	N	N	Highly variable	Y	146 45

Loc	Trt	Tillage: Type and Time	Crop Rotation	Years in No-Till	Manure	Residue Removal	Fertilizer Rec N-P-K-S	Tile Drain	Average Yield (bu/a)
7	CT	Unknown	Corn Soy	N/A	N	N	Unknown	N	Unknown
	NT	N/A	Corn Soy	7	Y	N	Two year blend: 140-30-0	N	190 55
	NTVG	N/A	N/A	N/A	N/A	Cut for hay	N/A	N	N/A
8	CT	Fall Disk & Spring Cultivate	Corn Soy	N/A	N	N	150-50-40-10	N	200 60
	NT	N/A	Corn Oat Soy	4	Y	Grazed	150-70-70 80-50-50 45-50-50	N	170 125 50
	NTVG	N/A	N/A	100+	N	Grazed	N	N	N/A

CT: Conventional Tillage
NT: No-Tillage
NTVG: Native Grass
Soils included in this analysis were Haplustolls.

N-P-K-S: Percent Nitrogen, Phosphorus, Potassium, and Sulfur in fertilizer blend
N/A: Not Applicable

Supplemental Table A2: Sample mean, standard deviation (σ), and Coefficient of Variation (C.V.) values between replicates for sand content (%), clay content (%), electrical conductivity (EC, μS/cm), and soil pH by depth. Small letters following mean values indicate significant differences at α=0.05.

Location	Treatment	Depth	Sand Content			Clay Content			Soil EC			Soil pH		
			Mean	**σ**	**C.V.**	**Mean**	**σ**	**C.V.**	**Mean**	**σ**	**C.V.**	**Mean**	**σ**	**C.V.**
1	**CT**	0-10	29.00	3.74	12.90	34.75	0.96	2.76	366.00	32.56	8.90	7.69	0.50	6.44
		10-20	28.50	4.12	14.47	33.50	0.58	1.72	319.25	54.30	17.01	7.75	0.34	4.36
		20-40	30.25	6.65	21.99	34.75	4.99	14.36	339.43	74.13	21.84	8.11	1.06	13.03
	NT	0-10	21.00	4.24	20.20	34.25	0.50	1.46	381.26	55.33	14.51	7.15	0.30	4.21
		10-20	20.25	5.56	27.46	36.25	2.06	5.69	352.48	81.56	23.14	7.24	0.41	5.65
		20-40	26.00	10.95	42.13	37.00	1.41	3.82	423.13	92.01	21.74	7.29	0.48	6.60
	NTVG	0-10	24.75	9.00	36.34	31.25	2.06	6.60	363.50	59.75	16.44	6.74	0.21	3.16
		10-20	25.75	9.18	35.65	31.50	2.08	6.61	376.75	81.48	21.63	7.01	0.36	5.18
		20-40	27.50	7.23	26.31	31.50	3.00	9.52	363.05	112.31	30.94	6.91	0.52	7.59
2	**CT**	0-10	25.25	4.03	15.96	26.00	4.97	19.10	534.25	67.38	12.61	7.49	0.29	3.87
		10-20	24.75	3.77	15.25	26.25	4.79	18.24	535.00	63.92	11.95	7.67	0.12	1.54
		20-40	26.25	9.78	37.24	27.75	4.57	16.48	512.50	63.54	12.40	7.80	0.17	2.20
	NT	0-10	39.00	6.06	15.53	21.75	3.95	18.15	191.92	25.89	13.49	5.80 a	0.21	4.22
		10-20	38.25	5.56	14.54	26.25	2.22	8.45	216.73	57.25	26.42	5.03 b	0.11	2.26
		20-40	39.25	1.50	3.82	22.75	4.50	19.78	229.23	26.12	11.40	4.91 b	0.41	7.15
	NTVG	0-10	36.50	2.38	6.52	29.50	2.52	8.53	473.00	57.98	12.26	7.32	0.35	4.82
		10-20	37.25	3.30	8.87	28.50	2.65	9.28	468.75	51.91	11.07	7.52	0.17	2.24
		20-40	37.75	2.87	7.61	29.00	2.58	8.90	429.50	52.30	12.18	7.77	0.25	3.28

Location	Treatment	Depth	Sand Content			Clay Content			Soil EC			Soil pH		
			Mean	σ	C.V.	Mean	σ	C.V.	Mean	σ	C.V.	Mean	σ	C.V.
3	CT	0-10	3.50	4.04	115.47	33.00 b	0.00	0.00	569.00	73.66	12.95	5.28 a	0.14	2.99
		10-20	8.25	1.26	15.25	32.75 b	0.50	1.53	565.50	82.57	14.60	4.57 b	0.28	6.11
		20-40	8.25	1.71	20.70	37.25 a	2.87	7.71	459.00	101.35	22.08	4.55 b	0.24	4.59
	NT	0-10	7.25	0.50	6.90	30.50 b	1.91	6.28	328.18 a	37.54	11.44	4.95 a	0.44	8.98
		10-20	7.75	1.50	19.35	34.00 ab	1.14	4.16	228.43 b	7.01	3.07	4.79 a	0.27	5.59
		20-40	7.50	1.29	17.21	35.50 a	3.00	8.45	280.83 ab	42.30	15.06	5.23 a	0.26	5.01
	NTVG	0-10	13.00	1.41	10.88	24.25	2.99	12.31	223.18	53.91	24.15	5.60 b	0.44	7.90
		10-20	11.00	0.82	7.42	27.50	2.08	7.57	204.93	32.01	15.62	6.14 ab	0.25	4.15
		20-40	12.00	3.74	31.19	27.50	3.70	13.44	200.73	4.65	2.31	6.72 a	0.23	3.38
4	CT	0-10	10.75	2.87	26.72	31.50	1.91	6.08	376.00	53.37	14.19	5.84	0.37	6.39
		10-20	9.50	1.73	18.23	32.75	1.50	4.58	433.50	191.48	44.17	6.00	0.36	5.99
		20-40	9.00	2.94	32.71	33.25	3.86	11.62	342.65	63.55	18.55	6.18	0.40	6.53
	NT	0-10	10.00 a	0.00	0.00	35.25	2.06	5.85	253.08	32.30	12.76	4.95 b	0.20	3.94
		10-20	8.75 b	0.50	5.71	34.00	2.94	8.66	220.38	22.58	110.25	5.33 b	0.33	6.14
		20-40	7.00 c	0.82	11.66	35.75	2.87	8.03	212.15	39.70	18.71	5.86 a	0.18	3.12
	NTVG	0-10	14.50	2.65	18.25	25.50	3.87	15.19	247.70	98.75	39.87	5.57	0.78	14.09
		10-20	16.75	1.89	11.30	31.00	2.83	9.12	287.65	95.81	33.31	6.01	0.60	9.96
		20-40	16.00	2.31	14.43	22.50	8.85	39.34	316.50	78.16	24.70	6.91	0.69	9.93
5	CT	0-10	10.25 a	0.96	9.34	32.50 b	0.58	1.78	162.23	9.84	6.06	5.46	0.28	5.18
		10-20	8.75 ab	1.50	17.14	33.75 b	1.26	3.73	160.43	21.75	13.56	5.62	0.33	5.80
		20-40	7.25 b	1.50	20.69	35.50 a	0.58	1.63	169.60	10.32	6.08	5.97	0.25	4.18
	NT	0-10	16.00 a	1.41	8.84	29.50 b	0.58	1.96	195.58 b	36.75	18.79	4.69 b	0.28	6.01
		10-20	14.25 ab	2.22	15.56	31.75 b	1.71	5.38	216.98 b	33.92	15.64	5.06 ab	0.18	3.54
		20-40	9.75 b	2.99	30.63	36.50 a	3.70	10.13	289.43 a	31.88	11.02	5.45 b	0.28	5.17
	NTVG	0-10	4.75	2.22	46.68	33.50	4.43	13.24	413.75	65.19	15.76	6.71	0.40	5.91
		10-20	5.50	1.91	34.82	35.50	1.91	5.39	349.08	40.28	11.54	7.20	0.43	5.98
		20-40	7.25	2.06	28.44	33.00	1.15	3.50	331.60	93.77	28.28	7.33	0.41	5.53

Location	Treatment	Depth	Sand Content			Clay Content			Soil EC			Soil pH		
			Mean	σ	C.V.	Mean	σ	C.V.	Mean	σ	C.V.	Mean	σ	C.V.
6	**CT**	0-10	7.25	0.50	6.90	34.50	1.00	2.90	864.25	234.56	27.14	5.92	0.25	4.23
		10-20	7.25	0.96	13.21	36.75	0.96	2.61	905.25	601.38	66.43	6.13	0.46	7.54
		20-40	5.75	1.50	26.09	38.25	0.50	1.31	943.50	336.52	35.67	6.31	0.35	5.54
	NT	0-10	7.25	1.26	17.36	36.75	1.71	4.65	491.00	79.97	16.29	5.22 b	0.28	5.27
		10-20	7.00	1.41	20.20	38.50	0.58	1.50	359.05	92.06	25.64	5.56 ab	0.11	2.02
		20-40	7.75	1.50	19.35	38.50	2.08	5.41	385.00	84.25	21.88	6.00 a	0.42	7.07
7	**CT**	0-10	23.00	2.58	11.23	32.25	0.50	1.55	420.48	140.21	33.35	5.10 b	0.21	4.10
		10-20	22.50	3.42	15.18	32.75	2.99	9.12	384.00	79.95	20.82	5.53 a	0.21	3.80
		20-40	24.00	6.78	28.26	34.25	1.50	4.38	362.98	81.53	22.46	5.90 a	0.12	2.07
	NT	0-10	26.25	3.30	12.59	34.25	1.26	3.67	442.00	52.58	11.90	5.47 b	0.36	6.60
		10-20	25.75	3.59	13.96	35.00	2.16	6.17	375.25	15.65	4.17	6.34 a	0.56	8.88
		20-40	27.50	9.26	33.66	35.75	4.19	11.73	429.00	85.63	19.96	6.87 a	0.34	5.02
	NTVG	0-10	26.25	3.40	12.97	33.50	1.91	5.72	204.05	32.33	15.85	6.18	0.38	6.15
		10-20	30.25	4.43	14.63	35.00	0.82	2.33	265.68	78.17	29.42	6.12	0.28	4.66
		20-40	33.25	6.95	20.89	35.25	1.71	4.84	281.20	15.81	5.62	6.19	0.24	3.92
8	**CT**	0-10	25.25	4.27	16.92	34.75	1.71	4.91	1275.50	681.06	53.40	6.38	0.43	6.66
		10-20	24.00	3.56	14.83	35.25	0.96	2.72	1789.00	1006.59	56.27	6.55	0.52	7.95
		20-40	22.75	5.62	24.70	36.75	1.71	4.65	1947.75	1130.14	58.02	6.37	0.37	5.75
	NT	0-10	35.75	1.50	4.20	35.25	0.96	2.72	478.50	87.36	18.26	6.63	0.22	3.35
		10-20	34.25	1.26	3.67	35.75	1.50	4.20	330.85	85.06	25.71	6.85	0.19	2.73
		20-40	37.50	7.77	20.71	36.50	1.73	4.75	405.48	150.43	37.10	6.91	0.54	7.87
	NTVG	0-10	22.25	3.30	14.85	34.50	3.00	8.70	238.85	79.82	33.42	6.85	0.89	12.98
		10-20	23.50	1.91	8.15	35.75	3.69	10.31	215.05	63.80	29.67	6.65	0.75	11.29
		20-40	25.00	2.58	10.33	37.75	6.65	17.62	224.43	61.33	27.33	6.90	0.45	6.46

CT: Conventional Tillage, NT: No-Tillage, NTVG: Native Grass. Soils included in this analysis were Haplustolls.

Supplemental Table A3: Sample mean, standard deviation (σ), and Coefficient of Variation (C.V.) values between replicates for soil organic carbon (SOC, %), soil inorganic carbon (SIC, %), total carbon (TC, %), and total nitrogen (TN, ppm) by depth. Small letters following mean values indicate significant differences at α=0.05.

Location	Treatment	Depth	SOC			SIC			TC			TN		
			Mean	σ	C.V.	Mean	σ	C.V.	Mean	σ	C.V.	Mean	σ	C.V.
1	CT	0-10	2.23 a	0.10	4.70	0.65	1.01	156.56	2.42	0.22	9.07	2246.75 a	121.26	5.40
		10-20	1.78 b	0.07	3.72	0.91	1.18	129.72	1.89	0.12	6.44	1727.50 b	118.70	6.87
		20-40	1.58 b	0.36	22.80	0.84	1.00	120.17	2.11	0.68	32.32	1395.25 b	414.83	29.73
	NT	0-10	2.97 a	0.23	7.88	0.10	0.03	29.82	2.46	0.11	4.66	2321.50 a	118.13	5.09
		10-20	2.33 b	0.21	8.89	0.13	0.10	77.42	1.78	0.04	2.12	1650.75 b	99.25	6.01
		20-40	1.96 b	0.32	16.47	1.05	1.15	108.78	2.08	0.75	36.27	1163.25 c	229.92	19.77
	NTVG	0-10	4.86 a	0.40	8.13	0.12 a	0.04	36.31	4.59 a	0.38	8.35	4298.50 a	314.64	7.32
		10-20	3.81 b	0.27	7.11	0.13 b	0.08	64.59	3.73 b	0.09	2.36	3367.75 b	115.52	3.43
		20-40	3.50 b	0.15	4.20	0.22 b	0.13	57.01	3.37 b	0.19	5.64	2730.25 c	185.15	6.78
2	CT	0-10	3.42	0.22	6.48	0.22 a	0.04	20.06	3.18	0.19	5.99	2838.75 a	148.54	5.23
		10-20	3.13	0.36	11.40	0.23 ab	0.04	18.45	2.90	0.11	3.87	2599.00 ab	194.01	7.46
		20-40	2.94	0.41	14.07	0.38 a	0.13	33.96	2.82	0.29	10.30	2315.75 b	368.46	15.91
	NT	0-10	2.90	0.46	15.88	0.00	0.00	0.00	2.54 a	0.31	12.17	2407.00 a	236.29	9.82
		10-20	2.44	0.30	12.10	0.00	0.00	0.00	2.21 ab	0.23	10.35	2078.00 ab	204.00	9.82
		20-40	2.19	0.34	15.63	0.03	0.06	200.00	1.86 a	0.33	17.64	1711.50 b	294.99	17.24
	NTVG	0-10	4.03 a	0.30	7.42	0.33	0.19	57.64	4.02 a	0.15	3.77	3706.75 a	158.29	4.27
		10-20	3.44 b	0.15	4.44	0.33	0.16	47.93	3.37 b	0.16	4.68	2995.00 b	141.11	4.71
		20-40	2.81 c	0.27	9.60	0.36	0.19	51.21	2.72 c	0.40	14.58	2243.75 c	268.06	11.95

Location	Treatment	Depth	SOC			SIC			TC			TN		
			Mean	σ	C.V.	Mean	σ	C.V.	Mean	σ	C.V.	Mean	σ	C.V.
3	**CT**	0-10	2.97 a	0.16	5.38	0.00	0.00	0.00	2.68 a	0.10	3.71	2653.00 a	32.73	1.23
		10-20	2.85 a	0.33	11.45	0.00	0.00	0.00	2.42 a	0.25	10.25	2412.50 a	212.98	8.83
		20-40	2.03 b	0.04	1.81	0.00	0.00	0.00	1.42 b	0.08	5.83	1406.50 b	75.08	5.34
	NT	0-10	3.04 a	0.08	2.53	0.00	0.00	0.00	2.70 a	0.09	3.45	2636.25 a	91.30	3.46
		10-20	2.58 b	0.06	2.50	0.00	0.00	0.00	2.12 b	0.23	11.07	2113.50 b	227.07	10.74
		20-40	1.85 c	0.15	7.94	0.00	0.00	0.00	1.13 c	0.19	16.42	1109.75 c	181.71	16.37
	NTVG	0-10	4.15 a	0.53	12.65	0.02	0.04	200.00	4.06 a	0.66	16.28	4179.50 a	697.97	16.70
		10-20	3.14 b	0.10	3.29	0.02	0.05	200.00	2.90 b	0.29	9.86	2878.50 b	257.09	8.93
		20-40	2.48 b	0.39	15.60	0.06	0.01	17.48	2.03 b	0.46	22.63	1932.00 b	427.09	22.11
4	**CT**	0-10	2.65 a	0.29	10.94	0.00	0.00	0.00	2.30 a	0.20	8.72	2304.00 a	169.56	7.36
		10-20	2.48 a	0.24	9.64	0.00	0.00	0.00	1.95 ab	0.31	15.65	1995.00 a	326.64	16.37
		20-40	1.84 b	0.42	22.63	0.04	0.05	119.16	1.27 b	0.63	49.43	1211.25 b	530.34	43.78
	NT	0-10	3.05 a	0.23	7.53	0.00	0.00	0.00	2.58 a	0.23	8.80	2486.50 a	138.45	5.57
		10-20	2.41 b	0.06	2.49	0.00	0.00	0.00	1.92 b	0.14	7.27	1892.25 b	130.26	6.88
		20-40	1.93 c	0.26	13.31	0.00	0.00	0.00	1.31 c	0.34	25.77	1261.75 c	290.63	23.03
	NTVG	0-10	4.16 a	0.66	15.94	0.03	0.05	200.00	4.05 a	0.88	21.67	4285.75 a	950.48	22.18
		10-20	2.87 b	0.18	6.37	0.12	0.24	200.00	2.55 b	0.22	8.80	2577.50 b	150.23	5.83
		20-40	2.05 c	0.08	3.75	0.48	0.50	104.78	1.86 b	0.36	19.42	1536.75 b	64.48	4.20
5	**CT**	0-10	3.10 a	0.05	1.63	0.00	0.00	0.00	2.70 a	0.05	2.01	2641.00 a	62.19	2.35
		10-20	2.70 b	0.15	5.38	0.00	0.00	0.00	2.15 b	0.19	8.86	2150.50 b	210.19	9.77
		20-40	2.41 c	0.19	7.99	0.02	0.04	200.00	1.81 b	0.31	17.22	1772.75 b	333.40	18.81
	NT	0-10	2.46 a	0.30	12.09	0.00	0.00	0.00	2.31 a	0.34	14.60	2325.25 a	256.11	11.01
		10-20	2.06 b	0.06	2.68	0.00	0.00	0.00	1.81 b	0.09	4.79	1872.00 b	86.15	4.60
		20-40	1.61 c	0.14	8.39	0.00	0.00	0.00	1.13 c	0.18	15.91	1154.50 c	176.30	15.27
	NTVG	0-10	4.22 a	0.23	5.57	0.12	0.07	60.30	4.14 a	0.33	8.03	4091.50 a	331.79	8.11
		10-20	3.18 b	0.38	11.94	0.08	0.02	28.07	3.03 b	0.37	12.30	3018.50 b	414.68	13.74
		20-40	2.86 b	0.61	21.34	0.12	0.12	100.81	2.83 b	0.68	24.20	2716.50 b	572.22	21.06

Location	Treatment	Depth	SOC			SIC			TC			TN		
			Mean	σ	C.V.	Mean	σ	C.V.	Mean	σ	C.V.	Mean	σ	C.V.
6	**CT**	0-10	2.38 a	0.10	4.28	0.02	0.03	200.00	2.88 a	0.08	2.73	2496.50 a	112.93	4.52
		10-20	2.01 ab	0.26	13.04	0.03	0.06	200.00	2.27 b	0.33	14.35	1993.25 b	257.61	12.92
		20-40	1.97 b	0.20	10.39	0.04	0.04	117.15	2.07 b	0.22	10.76	1843.50 b	138.29	7.50
	NT	0-10	2.30 a	0.21	9.30	0.00	0.00	0.00	2.70 a	0.15	5.56	2433.00 a	116.04	4.77
		10-20	1.95 b	0.06	2.82	0.00	0.00	0.00	2.25 b	0.05	2.21	1988.75 b	49.29	2.48
		20-40	2.04 ab	0.12	6.08	0.04	0.04	121.59	2.28 b	0.14	6.33	2004.25 b	99.14	4.95
7	**CT**	0-10	2.69 a	0.19	6.98	0.00	0.00	0.00	3.35 a	0.25	7.54	3216.50 a	229.97	7.15
		10-20	2.31 a	0.27	11.51	0.00	0.00	0.00	2.68 b	0.15	5.77	2591.75 b	147.38	5.69
		20-40	1.61 b	0.27	16.81	0.00	0.00	0.00	1.65 c	0.30	18.15	1631.25 c	239.18	14.66
	NT	0-10	2.13 a	0.20	9.55	0.00 b	0.00	0.00	2.45 a	0.10	4.07	2478.50 a	99.53	4.02
		10-20	1.80 ab	0.05	2.63	0.09 a	0.07	75.71	2.02 b	0.13	6.64	2030.50 b	92.30	4.57
		20-40	1.54 b	0.25	16.38	0.08 a	0.01	9.67	1.47 c	0.20	13.54	1490.75 c	161.97	10.86
	NTVG	0-10	3.32 a	0.43	12.81	0.03	0.05	200.00	4.08 a	0.46	11.20	3817.50 a	373.48	9.78
		10-20	2.57 b	0.32	12.46	0.19	0.04	200.00	2.95 b	0.17	5.65	2821.50 b	123.75	4.39
		20-40	2.00 b	0.22	11.04	0.03	0.05	200.00	2.16 c	0.21	9.53	2056.25 c	214.44	10.43
8	**CT**	0-10	2.40 a	0.19	8.01	0.06	0.04	74.97	2.66 a	0.21	7.99	2405.25 a	156.32	6.50
		10-20	2.05 ab	0.08	3.75	0.05	0.04	74.09	2.25 b	0.15	6.84	2081.25 b	109.55	5.26
		20-40	1.95 b	0.29	14.94	0.05	0.03	74.34	1.98 b	0.22	11.12	1820.75 b	179.54	9.86
	NT	0-10	2.63 a	0.17	6.41	0.09	0.03	32.64	2.52 a	0.03	1.32	2513.50 a	48.97	1.95
		10-20	2.17 b	0.16	7.47	0.08	0.03	40.64	1.9 b	0.07	3.81	1892.25 b	71.64	3.79
		20-40	1.98 b	0.13	6.37	0.17	0.15	90.44	1.71 c	0.08	4.41	1568.00 c	148.18	9.45
	NTVG	0-10	3.87 a	0.35	9.04	0.06	0.01	12.59	3.74 a	0.28	7.46	3633.25 a	277.00	7.62
		10-20	3.15 b	0.26	8.40	0.07	0.01	18.43	3.01 b	0.25	8.25	2808.00 b	224.09	7.98
		20-40	2.57 b	0.40	15.55	0.08	0.02	32.07	2.21 c	0.35	15.87	2124.75 c	324.26	15.26

CT: Conventional Tillage, NT: No-Tillage, NTVG: Native Grass. Soils included in this analysis were Haplustolls.

Supplemental Table A4: Sample mean, standard deviation (σ), and Coefficient of Variation (C.V.) values between replicates for sand content (%), clay content (%), electrical conductivity (EC, μS/cm), and soil pH by treatment. Small letters following mean values indicate significant differences at α=0.05.

Location	**Depth**	**Treatment**	**Sand Content**			**Clay Content**			**Soil EC**			**Soil pH**		
			Mean	**σ**	**C.V.**	**Mean**	**σ**	**C.V.**	**Mean**	**σ**	**C.V.**	**Mean**	**σ**	**C.V.**
1	**0-10cm**	CT	29.00	3.74	12.90	34.75 a	0.96	2.76	366.00	32.56	8.90	7.69 a	0.50	6.44
		NT	21.00	4.24	20.20	34.25 a	0.50	1.46	381.26	55.33	14.51	7.15 ab	0.30	4.21
		NTVG	24.75	9.00	36.34	31.25 b	2.06	6.60	363.50	59.75	16.44	6.74 b	0.21	3.16
	10-20cm	CT	28.50	4.12	14.47	33.50 ab	0.58	1.72	319.25	54.30	17.01	7.75 a	0.34	4.36
		NT	20.25	5.56	27.46	36.25 a	2.06	5.69	352.48	81.56	23.14	7.24 ab	0.41	5.65
		NTVG	25.75	9.18	35.65	31.50 b	2.08	6.61	376.75	81.48	21.63	7.01 b	0.36	5.18
	20-40cm	CT	30.25	6.65	21.99	34.75	4.99	14.36	339.43	74.13	21.84	8.11	1.06	13.03
		NT	26.00	10.95	42.13	37.00	1.41	3.82	423.13	92.01	21.74	7.29	0.48	6.60
		NTVG	27.50	7.23	26.31	31.50	3.00	9.52	363.05	112.31	30.94	6.91	0.52	7.59
2	**0-10cm**	CT	25.25 b	4.03	15.96	26.00	4.97	19.10	534.25 a	67.38	12.61	7.49 a	0.29	3.87
		NT	39.00 a	6.06	15.53	21.75	3.95	18.15	191.92 b	25.89	13.49	5.03 b	0.21	4.22
		NTVG	36.50 a	2.38	6.52	29.50	2.52	8.53	473.00 a	57.98	12.26	7.32 a	0.35	4.82
	10-20cm	CT	24.75 b	3.77	15.25	26.25	4.79	18.24	535.00 a	63.92	11.95	7.67 a	0.12	1.54
		NT	38.25 a	5.56	14.54	26.25	2.22	8.45	216.73 b	57.25	26.42	4.91 b	0.11	2.26
		NTVG	37.25 a	3.30	8.87	28.50	2.65	9.28	468.75 a	51.91	11.07	7.52 a	0.17	2.24
	20-40cm	CT	26.25 b	9.78	37.24	27.75	4.57	16.48	512.50 a	63.54	12.40	7.80 a	0.17	2.20
		NT	39.25 a	1.50	3.82	22.75	4.50	19.78	229.23 b	26.12	11.40	5.80 b	0.41	7.15
		NTVG	37.75 ab	2.87	7.61	29.00	2.58	8.90	429.50 a	52.30	12.18	7.77 a	0.25	3.28

Location	Depth	Treatment	Sand Content			Clay Content			Soil EC			Soil pH		
			Mean	σ	C.V.	Mean	σ	C.V.	Mean	σ	C.V.	Mean	σ	C.V.
3	0-10cm	CT	3.50 b	4.04	115.47	33.00 a	0.00	0.00	569.00 a	73.66	12.95	4.55 b	0.14	2.99
		NT	7.25 b	0.50	6.90	30.50 a	1.91	6.28	328.18 b	37.54	11.44	4.95 ab	0.44	8.98
		NTVG	13.00 a	1.41	10.88	24.25 b	2.99	12.31	223.18 b	53.91	24.15	5.60 a	0.44	7.90
	10-20cm	CT	8.25 b	1.26	15.25	32.75 a	0.50	1.53	565.50 a	82.57	14.60	4.57 b	0.28	6.11
		NT	7.75 b	1.50	19.35	34.00 a	1.14	4.16	228.43 b	7.01	3.07	4.79 b	0.27	5.59
		NTVG	11.00 a	0.82	7.42	27.50 b	2.08	7.57	204.93 b	32.01	15.62	6.14 a	0.25	4.15
	20-40cm	CT	8.25	1.71	20.70	37.25 a	2.87	7.71	459.00 a	101.35	22.08	5.28 b	0.24	4.59
		NT	7.50	1.29	17.21	35.50 a	3.00	8.45	280.83 b	42.30	15.06	5.23 b	0.26	5.01
		NTVG	12.00	3.74	31.19	27.50 b	3.70	13.44	200.73 b	4.65	2.31	6.72 a	0.23	3.38
4	0-10cm	CT	10.75 ab	2.87	26.72	31.50 a	1.91	6.08	376.00	53.37	14.19	5.84	0.37	6.39
		NT	10.00 b	0.00	0.00	35.25 a	2.06	5.85	253.08	32.30	12.76	4.95	0.20	3.94
		NTVG	14.50 a	2.65	18.25	25.50 b	3.87	15.19	247.70	98.75	39.87	5.57	0.78	14.09
	10-20cm	CT	9.50 b	1.73	18.23	32.75	1.50	4.58	433.50	191.48	44.17	6.00	0.36	5.99
		NT	8.75 b	0.50	5.71	34.00	2.94	8.66	220.38	22.58	110.25	5.33	0.33	6.14
		NTVG	16.75 a	1.89	11.30	31.00	2.83	9.12	287.65	95.81	33.31	6.01	0.60	9.96
	20-40cm	CT	9.00 b	2.94	32.71	33.25 ab	3.86	11.62	342.65 a	63.55	18.55	6.18 ab	0.40	6.53
		NT	7.00 b	0.82	11.66	35.75 a	2.87	8.03	212.15 b	39.70	18.71	5.86 b	0.18	3.12
		NTVG	16.00 a	2.31	14.43	22.50 b	8.85	39.34	316.50 ab	78.16	24.70	6.91 a	0.69	9.93
5	0-10cm	CT	10.25 b	0.96	9.34	32.50	0.58	1.78	162.23 b	9.84	6.06	5.46 b	0.28	5.18
		NT	16.00 a	1.41	8.84	29.50	0.58	1.96	195.58 b	36.75	18.79	4.69 c	0.28	6.01
		NTVG	4.75 c	2.22	46.68	33.50	4.43	13.24	413.75 a	65.19	15.76	6.71 a	0.40	5.91
	10-20cm	CT	8.75 b	1.50	17.14	33.75 ab	1.26	3.73	160.43 b	21.75	13.56	5.62 b	0.33	5.80
		NT	14.25 a	2.22	15.56	31.75 b	1.71	5.38	216.98 b	33.92	15.64	5.06 b	0.18	3.54
		NTVG	5.50 b	1.91	34.82	35.50 a	1.91	5.39	349.08 a	40.28	11.54	7.20 a	0.43	5.98
	20-40cm	CT	7.25	1.50	20.69	35.50	0.58	1.63	169.60 b	10.32	6.08	5.97 b	0.25	4.18
		NT	9.75	2.99	30.63	36.50	3.70	10.13	289.43 a	31.88	11.02	5.45 b	0.28	5.17
		NTVG	7.25	2.06	28.44	33.00	1.15	3.50	331.60 a	93.77	28.28	7.33 a	0.41	5.53

Location	Depth	Treatment	Sand Content			Clay Content			Soil EC			Soil pH		
			Mean	σ	C.V.	Mean	σ	C.V.	Mean	σ	C.V.	Mean	σ	C.V.
6	0-10cm	CT	7.25	0.50	6.90	34.50	1.00	2.90	864.25	234.56	27.14	5.92	0.25	4.23
		NT	7.25	1.26	17.36	36.75	1.71	4.65	491.00	79.97	16.29	5.22	0.28	5.27
	10-20cm	CT	7.25	0.96	13.21	36.75	0.96	2.61	905.25	601.38	66.43	6.13	0.46	7.54
		NT	7.00	1.41	20.20	38.50	0.58	1.50	359.05	92.06	25.64	5.56	0.11	2.02
	20-40cm	CT	5.75	1.50	26.09	38.25	0.50	1.31	943.50	336.52	35.67	6.31	0.35	5.54
		NT	7.75	1.50	19.35	38.50	2.08	5.41	385.00	84.25	21.88	6.00	0.42	7.07
7	0-10cm	CT	23.00	2.58	11.23	32.25	0.50	1.55	420.48 a	140.21	33.35	5.10 b	0.21	4.10
		NT	26.25	3.30	12.59	34.25	1.26	3.67	442.00 a	52.58	11.90	5.47 b	0.36	6.60
		NTVG	26.25	3.40	12.97	33.50	1.91	5.72	204.05 b	32.33	15.85	6.18 a	0.38	6.15
	10-20cm	CT	22.50 b	3.42	15.18	32.75	2.99	9.12	384.00	79.95	20.82	5.53 b	0.21	3.80
		NT	25.75 ab	3.59	13.96	35.00	2.16	6.17	375.25	15.65	4.17	6.34 a	0.56	8.88
		NTVG	30.25 a	4.43	14.63	35.00	0.82	2.33	265.68	78.17	29.42	6.12 ab	0.28	4.66
	20-40cm	CT	24.00	6.78	28.26	34.25	1.50	4.38	362.98 ab	81.53	22.46	5.90 b	0.12	2.07
		NT	27.50	9.26	33.66	35.75	4.19	11.73	429.00 a	85.63	19.96	6.87 a	0.34	5.02
		NTVG	33.25	6.95	20.89	35.25	1.71	4.84	281.20 b	15.81	5.62	6.19 b	0.24	3.92
8	0-10cm	CT	25.25 b	4.27	16.92	34.75	1.71	4.91	1275.50 a	681.06	53.40	6.38	0.43	6.66
		NT	35.75 a	1.50	4.20	35.25	0.96	2.72	478.50 b	87.36	18.26	6.63	0.22	3.35
		NTVG	22.25 b	3.30	14.85	34.50	3.00	8.70	238.85 b	79.82	33.42	6.85	0.89	12.98
	10-20cm	CT	24.00 b	3.56	14.83	35.25	0.96	2.72	1789.00 a	1006.59	56.27	6.55	0.52	7.95
		NT	34.25 a	1.26	3.67	35.75	1.50	4.20	330.85 b	85.06	25.71	6.85	0.19	2.73
		NTVG	23.50 b	1.91	8.15	35.75	3.69	10.31	215.05 b	63.80	29.67	6.65	0.75	11.29
	20-40cm	CT	22.75 b	5.62	24.70	36.75	1.71	4.65	1947.75 a	1130.14	58.02	6.37	0.37	5.75
		NT	37.50 a	7.77	20.71	36.50	1.73	4.75	405.48 b	150.43	37.10	6.91	0.54	7.87
		NTVG	25.00 b	2.58	10.33	37.75	6.65	17.62	224.43 b	61.33	27.33	6.90	0.45	6.46

CT: Conventional Tillage, NT: No-Tillage, NTVG: Native Grass. Soils included in this analysis were Haplustolls.

Supplemental Table A5: Sample mean, standard deviation (σ), and Coefficient of Variation (C.V.) values between replicates for soil organic carbon (SOC, %), soil inorganic carbon (SIC, %), total carbon (TC, %), and total nitrogen (TN, ppm) by treatment. Small letters following mean values indicate significant differences at α=0.05.

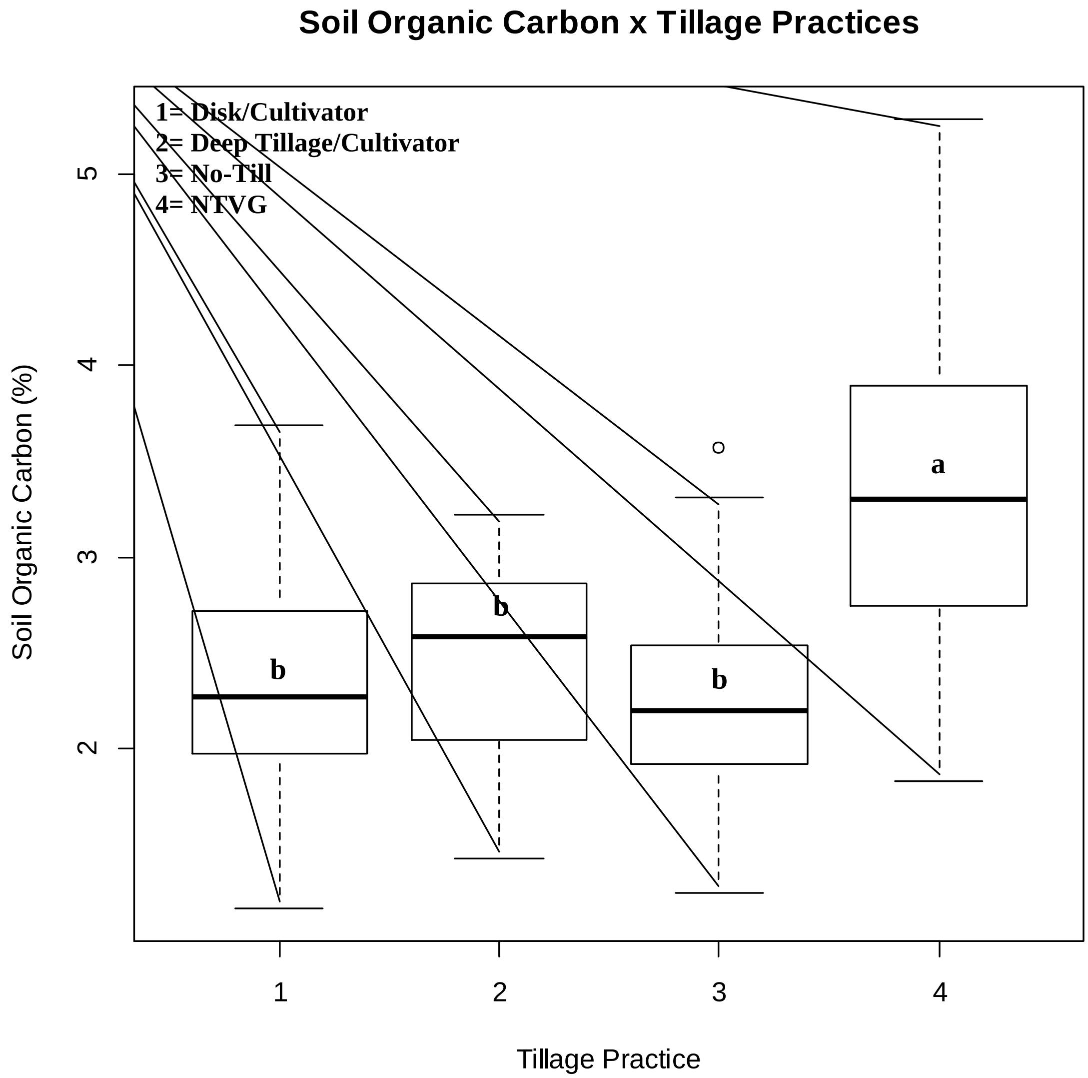
Location	Depth	Treatment	SOC			SIC			TC			TN		
			Mean	**σ**	**C.V.**	**Mean**	**σ**	**C.V.**	**Mean**	**σ**	**C.V.**	**Mean**	**σ**	**C.V.**
1	**0-10cm**	CT	2.23 b	0.10	4.70	0.65	1.01	156.56	2.42 b	0.22	9.07	2246.75 b	121.26	5.40
		NT	2.97 b	0.23	7.88	0.10	0.03	29.82	2.46 b	0.11	4.66	2321.50 b	118.13	5.09
		NTVG	4.86 a	0.40	8.13	0.12	0.04	36.31	4.59 a	0.38	8.35	4298.50 a	314.64	7.32
	10-20cm	CT	1.78 c	0.07	3.72	0.91	1.18	129.72	1.89 b	0.12	6.44	1727.50 b	118.70	6.87
		NT	2.33 b	0.21	8.89	0.13	0.10	77.42	1.78 b	0.04	2.12	1650.75 b	99.25	6.01
		NTVG	3.81 a	0.27	7.11	0.13	0.08	64.59	3.73 a	0.09	2.36	3367.75 b	115.52	3.43
	20-40cm	CT	1.58 c	0.36	22.80	0.84	1.00	120.17	2.11 b	0.68	32.32	1395.25 b	414.83	29.73
		NT	1.96 b	0.32	16.47	1.05	1.15	108.78	2.08 b	0.75	36.27	1163.25 b	229.92	19.77
		NTVG	3.50 a	0.15	4.20	0.22	0.13	57.01	3.37 a	0.19	5.64	2730.25 a	185.15	6.78
2	**0-10cm**	CT	3.42 ab	0.22	6.48	0.22 ab	0.04	20.06	3.18 b	0.19	5.99	2838.75 b	148.54	5.23
		NT	2.90 b	0.46	15.88	0.00 b	0.00		2.54 c	0.31	12.17	2407.00 c	236.29	9.82
		NTVG	4.03 a	0.30	7.42	0.33 a	0.19	57.64	4.02 a	0.15	3.77	3706.75 a	158.29	4.27
	10-20cm	CT	3.13 a	0.36	11.40	0.23 a	0.04	18.45	2.90 b	0.11	3.87	2599.00 b	194.01	7.46
		NT	2.44 b	0.30	12.10	0.00 b	0.00		2.21 c	0.23	10.35	2078.00 c	204.00	9.82
		NTVG	3.44 a	0.15	4.44	0.33 a	0.16	47.93	3.37 a	0.16	4.68	2995.00 a	141.11	4.71
	20-40cm	CT	2.94 a	0.41	14.07	0.38 a	0.13	33.96	2.82 a	0.29	10.30	2315.75	368.46	15.91
		NT	2.19 b	0.34	15.63	0.03 b	0.06	200.00	1.86 b	0.33	17.64	1711.50	294.99	17.24
		NTVG	2.81 ab	0.27	9.60	0.36 a	0.19	51.21	2.72 a	0.40	14.58	2243.75	268.06	11.95

Location	Depth	Treatment	SOC			SIC			TC			TN		
			Mean	σ	C.V.	Mean	σ	C.V.	Mean	σ	C.V.	Mean	σ	C.V.
3	**0-10cm**	CT	2.97 b	0.16	5.38	0.00	0.00		2.68 b	0.10	3.71	2653.00 b	32.73	1.23
		NT	3.04 b	0.08	2.53	0.00	0.00		2.70 b	0.09	3.45	2636.25 b	91.30	3.46
		NTVG	4.15 a	0.53	12.65	0.02	0.04	200.00	4.06 a	0.66	16.28	4179.50 a	697.97	16.70
	10-20cm	CT	2.85 ab	0.33	11.45	0.00	0.00		2.42 b	0.25	10.25	2412.50 b	212.98	8.83
		NT	2.58 b	0.06	2.50	0.00	0.00		2.12 b	0.23	11.07	2113.50 b	227.07	10.74
		NTVG	3.14 a	0.10	3.29	0.02	0.05	200.00	2.90 a	0.29	9.86	2878.50 a	257.09	8.93
	20-40cm	CT	2.03 ab	0.04	1.81	0.00 b	0.00		1.42 b	0.08	5.83	1406.50 ab	75.08	5.34
		NT	1.85 a	0.15	7.94	0.00 b	0.00		1.13 b	0.19	16.42	1109.75 b	181.71	16.37
		NTVG	2.48 b	0.39	15.60	0.06 a	0.01	17.48	2.03 a	0.46	22.63	1932.00 a	427.09	22.11
4	**0-10cm**	CT	2.65 b	0.29	10.94	0.00	0.00		2.30 b	0.20	8.72	2304.00 b	169.56	7.36
		NT	3.05 b	0.23	7.53	0.00	0.00		2.58 b	0.23	8.80	2486.50 b	138.45	5.57
		NTVG	4.16 a	0.66	15.94	0.03	0.05	200.00	4.05 a	0.88	21.67	4285.75 a	950.48	22.18
	10-20cm	CT	2.48 b	0.24	9.64	0.00	0.00		1.95 b	0.31	15.65	1995.00 b	326.64	16.37
		NT	2.41 b	0.06	2.49	0.00	0.00		1.92 b	0.14	7.27	1892.25 b	130.26	6.88
		NTVG	2.87 a	0.18	6.37	0.12	0.24	200.00	2.55 a	0.22	8.80	2577.50 a	150.23	5.83
	20-40cm	CT	1.84	0.42	22.63	0.04	0.05	119.16	1.27	0.63	49.43	1211.25	530.34	43.78
		NT	1.93	0.26	13.31	0.00	0.00		1.31	0.34	25.77	1261.75	290.63	23.03
		NTVG	2.05	0.08	3.75	0.48	0.50	104.78	1.86	0.36	19.42	1536.75	64.48	4.20
5	**0-10cm**	CT	3.10 b	0.05	1.63	0.00 b	0.00		2.70 b	0.05	2.01	2641.00 b	62.19	2.35
		NT	2.46 c	0.30	12.09	0.00 b	0.00		2.31 b	0.34	14.60	2325.25 b	256.11	11.01
		NTVG	4.22 a	0.23	5.57	0.12 a	0.07	60.30	4.14 a	0.33	8.03	4091.50 a	331.79	8.11
	10-20cm	CT	2.70 b	0.15	5.38	0.00 b	0.00		2.15 b	0.19	8.86	2150.50 b	210.19	9.77
		NT	2.06 c	0.06	2.68	0.00 b	0.00		1.81 b	0.09	4.79	1872.00 b	86.15	4.60
		NTVG	3.18 a	0.38	11.94	0.08 a	0.02	28.07	3.03 a	0.37	12.30	3018.50 a	414.68	13.74
	20-40cm	CT	2.41 a	0.19	7.99	0.02	0.04	200.00	1.81 b	0.31	17.22	1772.75 b	333.40	18.81
		NT	1.61 b	0.14	8.39	0.00	0.00		1.13 b	0.18	15.91	1154.50 b	176.30	15.27
		NTVG	2.86 a	0.61	21.34	0.12	0.12	100.81	2.83 a	0.68	24.20	2716.50 a	572.22	21.06

Location	Depth	Treatment	SOC			SIC			TC			TN		
			Mean	σ	C.V.	Mean	σ	C.V.	Mean	σ	C.V.	Mean	σ	C.V.
6	0-10cm	CT	2.38	0.10	4.28	0.02	0.03	200.00	2.88	0.08	2.73	2496.50	112.93	4.52
		NT	2.30	0.21	9.30	0.00	0.00		2.70	0.15	5.56	2433.00	116.04	4.77
	10-20cm	CT	2.01	0.26	13.04	0.03	0.06	200.00	2.27	0.33	14.35	1993.25	257.61	12.92
		NT	1.95	0.06	2.82	0.00	0.00		2.25	0.05	2.21	1988.75	49.29	2.48
	20-40cm	CT	1.97	0.20	10.39	0.04	0.04	117.15	2.07	0.22	10.76	1843.50	138.29	7.50
		NT	2.04	0.12	6.08	0.04	0.04	121.59	2.28	0.14	6.33	2004.25	99.14	4.95
7	0-10cm	CT	2.69 b	0.19	6.98	0.00	0.00		3.35 b	0.25	7.54	3216.50 b	229.97	7.15
		NT	2.13 b	0.20	9.55	0.00	0.00		2.45 c	0.10	4.07	2478.50 c	99.53	4.02
		NTVG	3.32 a	0.43	12.81	0.03	0.05	200.00	4.08 a	0.46	11.20	3817.50 a	373.48	9.78
	10-20cm	CT	2.31 a	0.27	11.51	0.00 b	0.00		2.68 a	0.15	5.77	2591.75 a	147.38	5.69
		NT	1.80 b	0.05	2.63	0.09 a	0.07	75.71	2.02 b	0.13	6.64	2030.50 b	92.30	4.57
		NTVG	2.57 a	0.32	12.46	0.19 ab	0.04	200.00	2.95 a	0.17	5.65	2821.50 a	123.75	4.39
	20-40cm	CT	1.61	0.27	16.81	0.00 b	0.00		1.65 b	0.30	18.15	1631.25 b	239.18	14.66
		NT	1.54	0.25	16.38	0.08 a	0.01	9.67	1.47 b	0.20	13.54	1490.75 b	161.97	10.86
		NTVG	2.00	0.22	11.04	0.03 ab	0.05	200.00	2.16 a	0.21	9.53	2056.25 a	214.44	10.43
8	0-10cm	CT	2.40 b	0.19	8.01	0.06	0.04	74.97	2.66 b	0.21	7.99	2405.25 b	156.32	6.50
		NT	2.63 b	0.17	6.41	0.09	0.03	32.64	2.52 b	0.03	1.32	2513.50 b	48.97	1.95
		NTVG	3.87 a	0.35	9.04	0.06	0.01	12.59	3.74 a	0.28	7.46	3633.25 a	277.00	7.62
	10-20cm	CT	2.05 b	0.08	3.75	0.05	0.04	74.09	2.25 b	0.15	6.84	2081.25 b	109.55	5.26
		NT	2.17 b	0.16	7.47	0.08	0.03	40.64	1.90 c	0.07	3.81	1892.25 b	71.64	3.79
		NTVG	3.15 a	0.26	8.40	0.07	0.01	18.43	3.01 a	0.25	8.25	2808.00 a	224.09	7.98
	20-40cm	CT	1.95 b	0.29	14.94	0.05	0.03	74.34	1.98 ab	0.22	11.12	1820.75 ab	179.54	9.86
		NT	1.98 ab	0.13	6.37	0.17	0.15	90.44	1.71 b	0.08	4.41	1568.00 b	148.18	9.45
		NTVG	2.57 a	0.40	15.55	0.08	0.02	32.07	2.21 a	0.35	15.87	2124.75 a	324.26	15.26

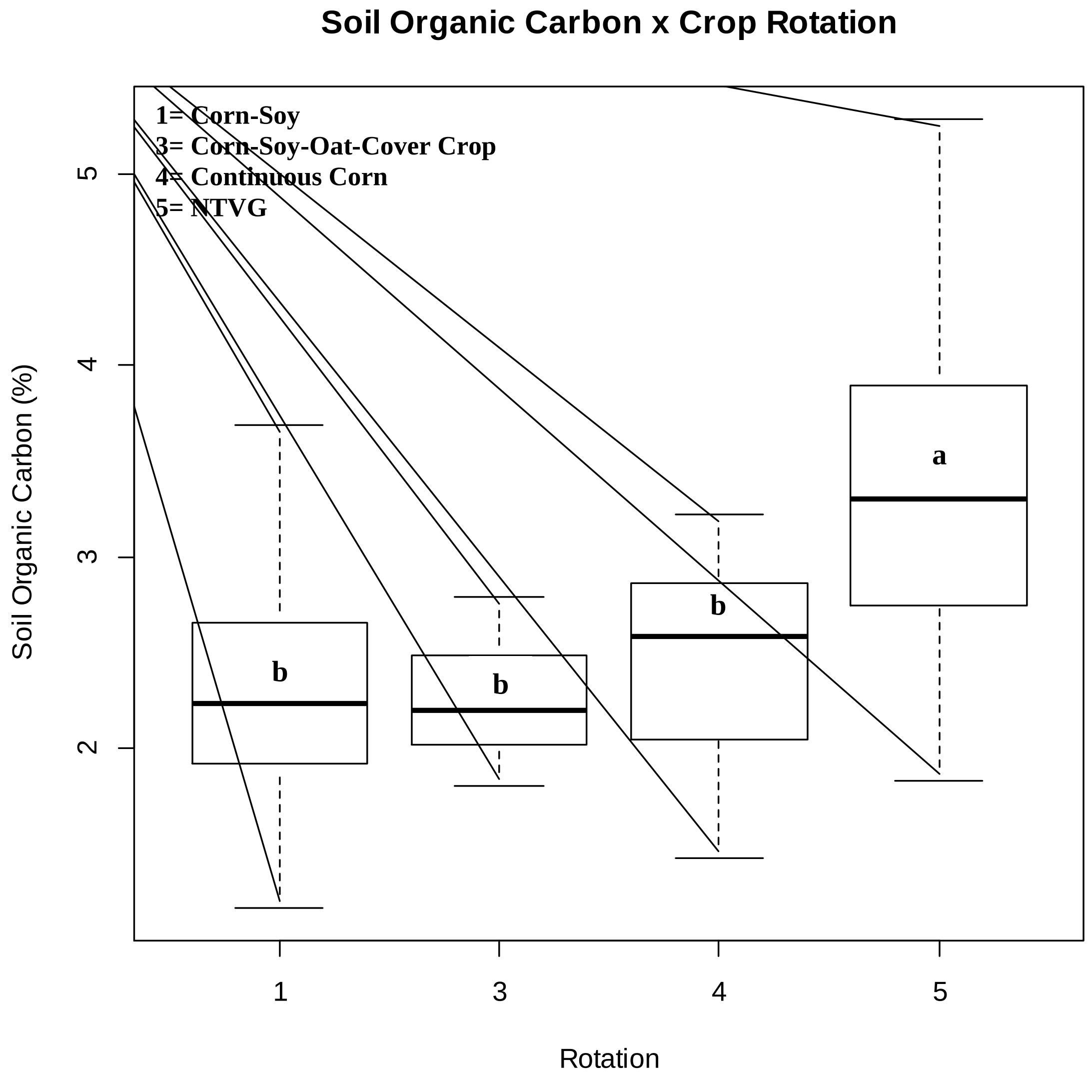
CT: Conventional Tillage, NT: No-Tillage, NTVG: Native Grass. Soils included in this analysis were Haplustolls.

APPENDIX B



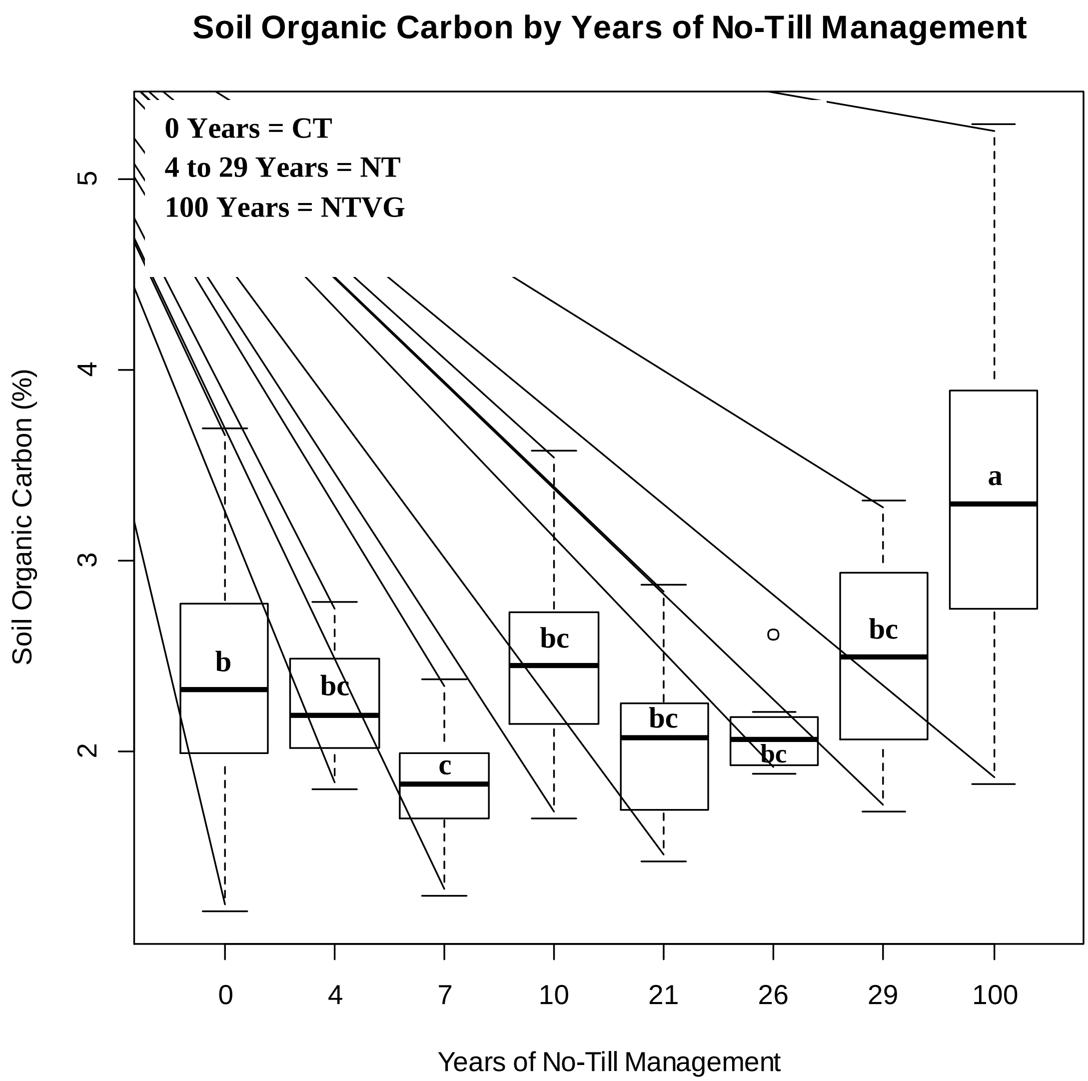
Supplemental Figure B1: Boxplot illustrating the distribution of soil organic carbon (SOC) data by different tillage practices. Native conditions were significantly higher in SOC than any of the cultivated treatments ($p<0.0001$). There was no significant difference in Disk/Cultivator, Deep Tillage/Cultivator, or No-Tillage practices. Boxplot generated in R 3.5.0.

*The top whisker identifies the maximum value of the data, the top of the box represents the third quartile, the bold black bar represents the median data value, the bottom of the box represents the first quartile, and the bottom whisker identifies the minimum value of the data.



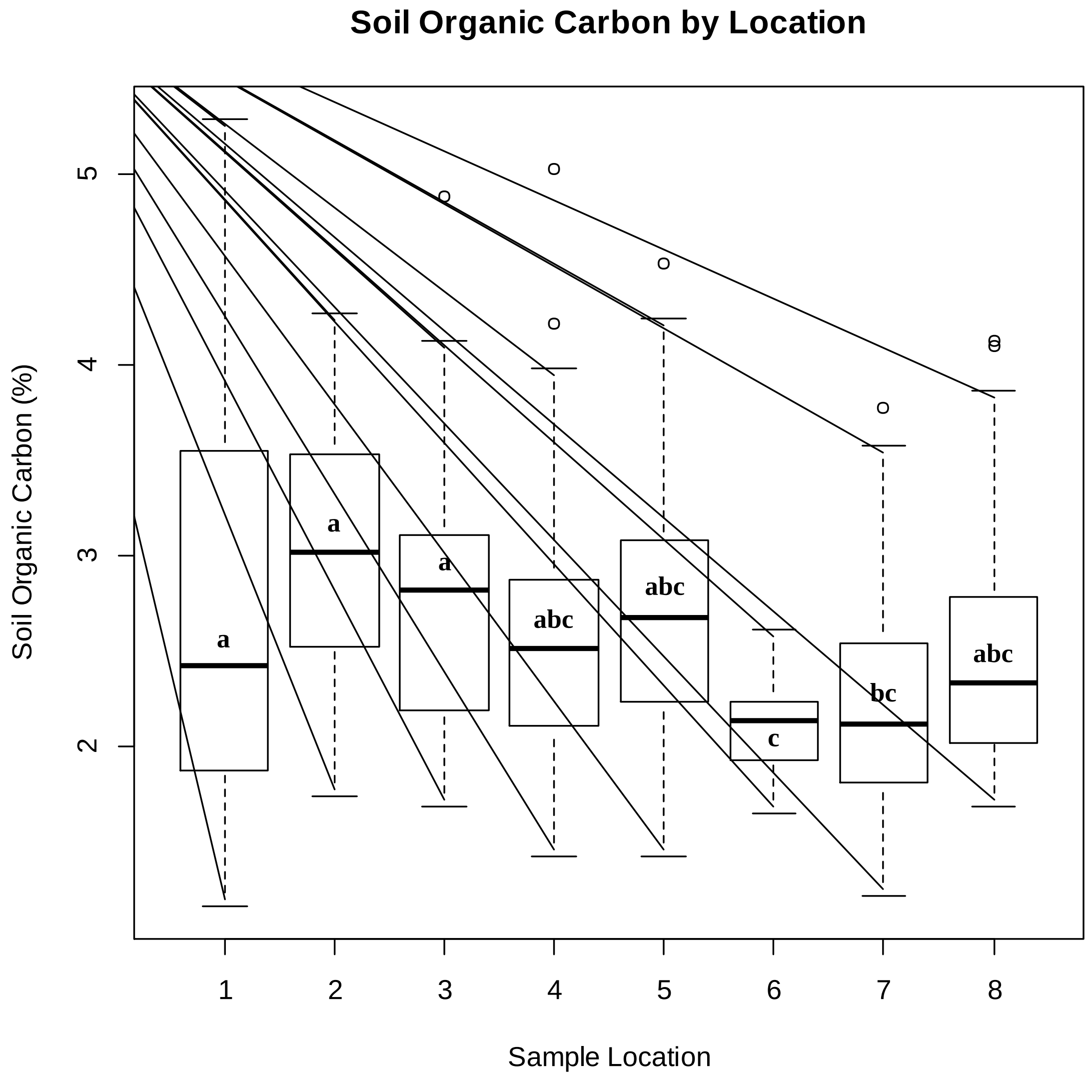
Supplemental Figure B2: Boxplot illustrating the distribution of SOC data by different crop rotations. Native conditions were significantly higher in SOC than any of the cultivated crop rotations included in this study ($p<0.0001$). There was no significant difference between Corn-Soybean, Corn-Soybean-Oats-Cover Crop, or Continuous Corn data. Boxplot generated in R 3.5.0.

*The top whisker identifies the maximum value of the data, the top of the box represents the third quartile, the bold black bar represents the median data value, the bottom of the box represents the first quartile, and the bottom whisker identifies the minimum value of the data.



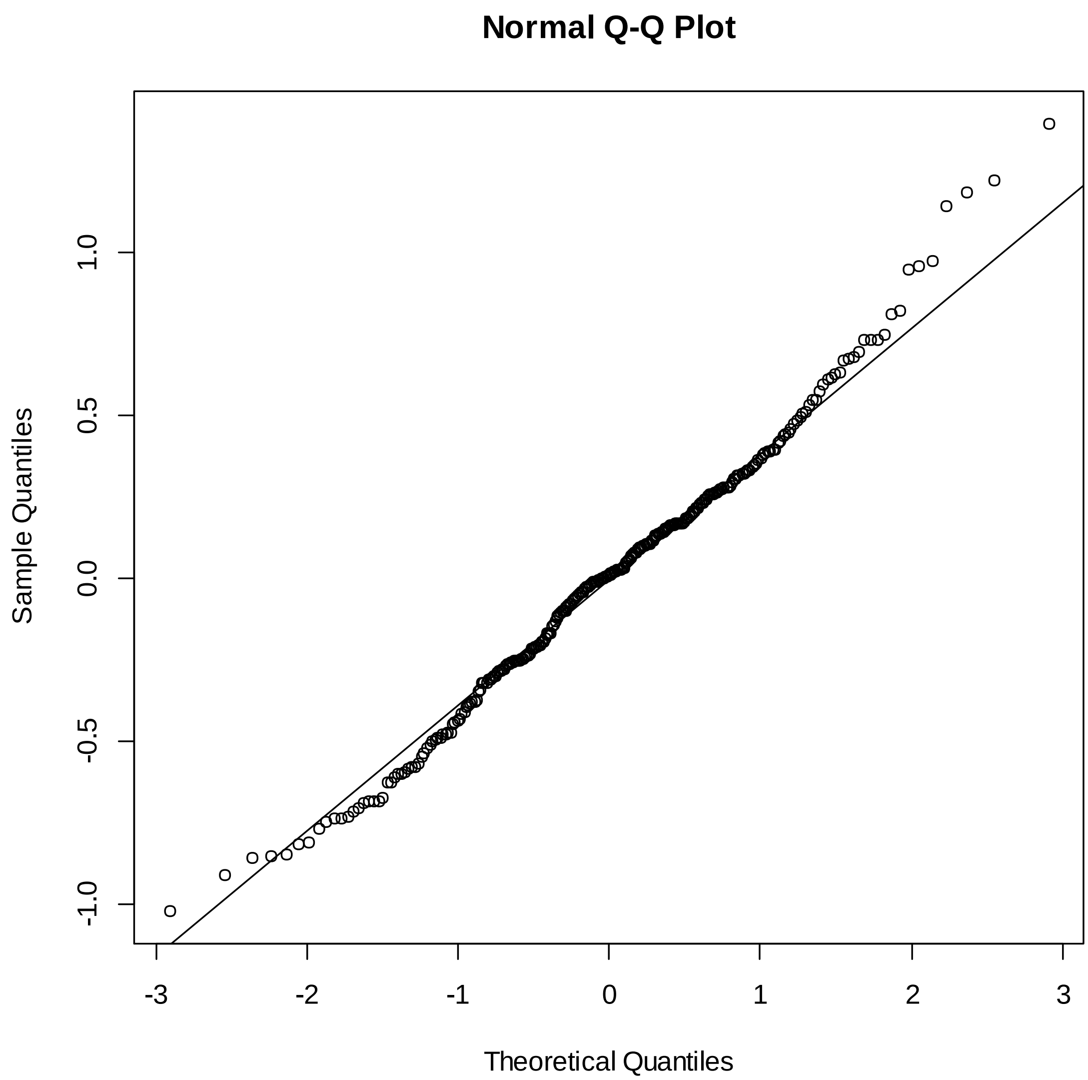
Supplemental Figure B3: Boxplot illustrating the distribution of SOC data by different periods of time under No-Till management. When α=0.05, Native Grass (NTVG) had significantly higher levels of SOC than any of the cultivated treatments. The site that had been under No-Till (NT) management for 7 years had the least SOC, but was not significantly different from 4, 10, 21, 26, or 29 years of NT management. In this boxplot, zero years represent the Conventional Tillage (CT) treatments in this study. Statistically, CT had the same SOC as 4, 10, 21, 26, and 29 years of NT. Boxplot generated in R 3.5.0.

*The top whisker identifies the maximum value of the data, the top of the box represents the third quartile, the bold black bar represents the median data value, the bottom of the box represents the first quartile, and the bottom whisker identifies the minimum value of the data.

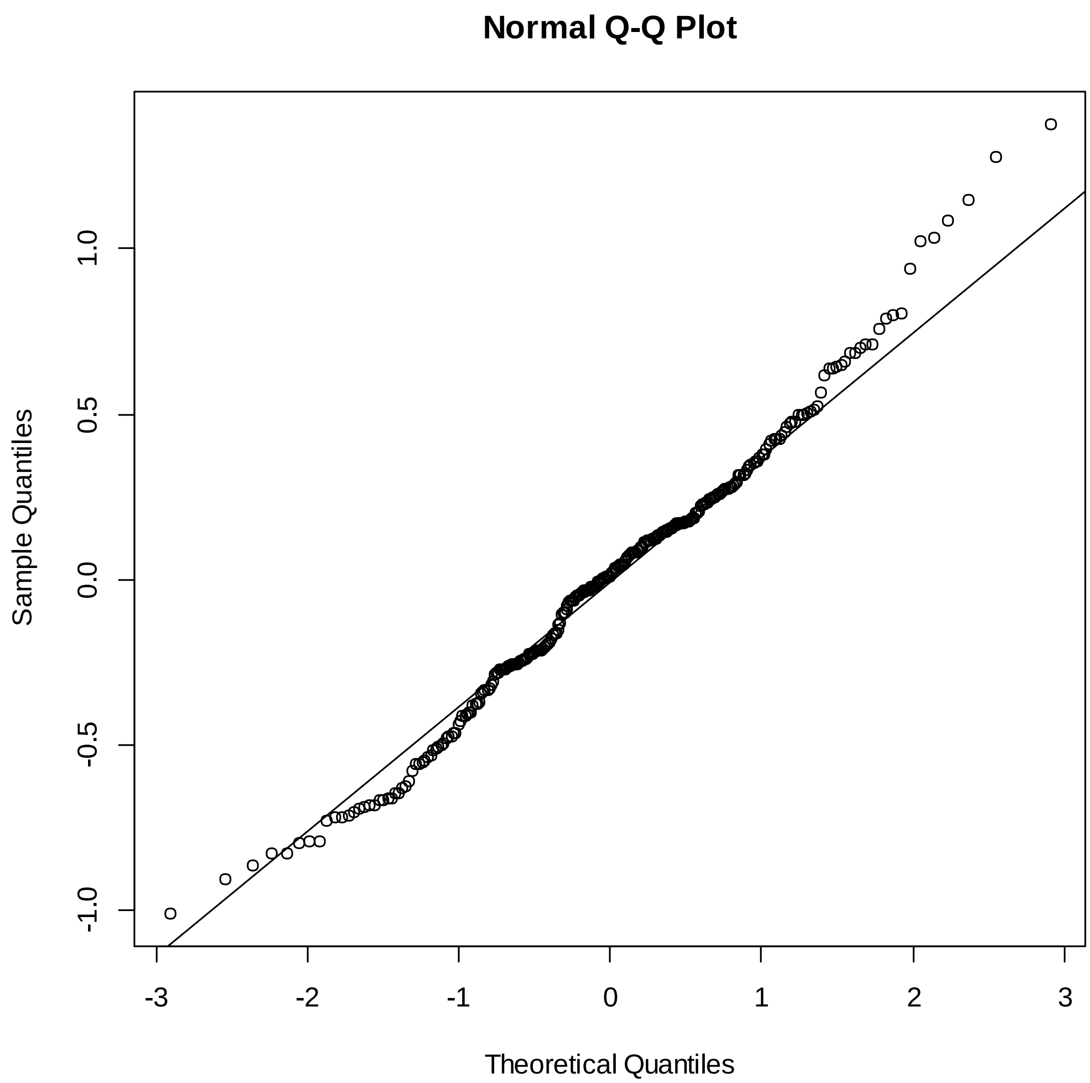


Supplemental Figure B4: Boxplot illustrating the distribution of SOC data by Sample Location. At α=0.05, Location 6 statistically had the least SOC, but was not significantly different from Locations 4, 7, or 8. Locations 4, 5, 7, and 8 were all statistically similar for SOC. Locations 1, 2, and 3 had the greatest levels of SOC of all locations in this study, but were not significantly different from Locations 4, 5, or 8. Boxplot generated in R 3.5.0.

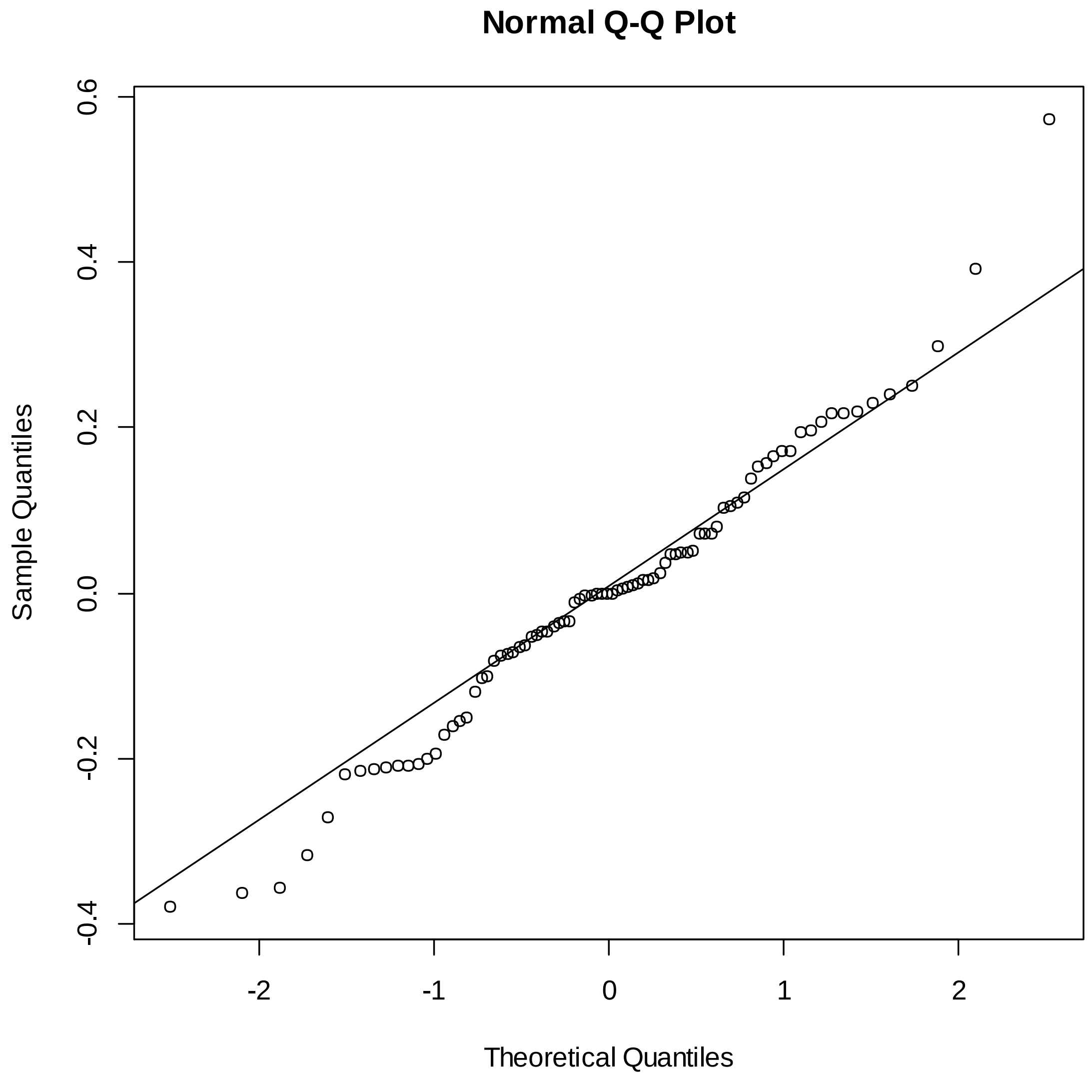
*The top whisker identifies the maximum value of the data, the top of the box represents the third quartile, the bold black bar represents the median data value, the bottom of the box represents the first quartile, and the bottom whisker identifies the minimum value of the data.



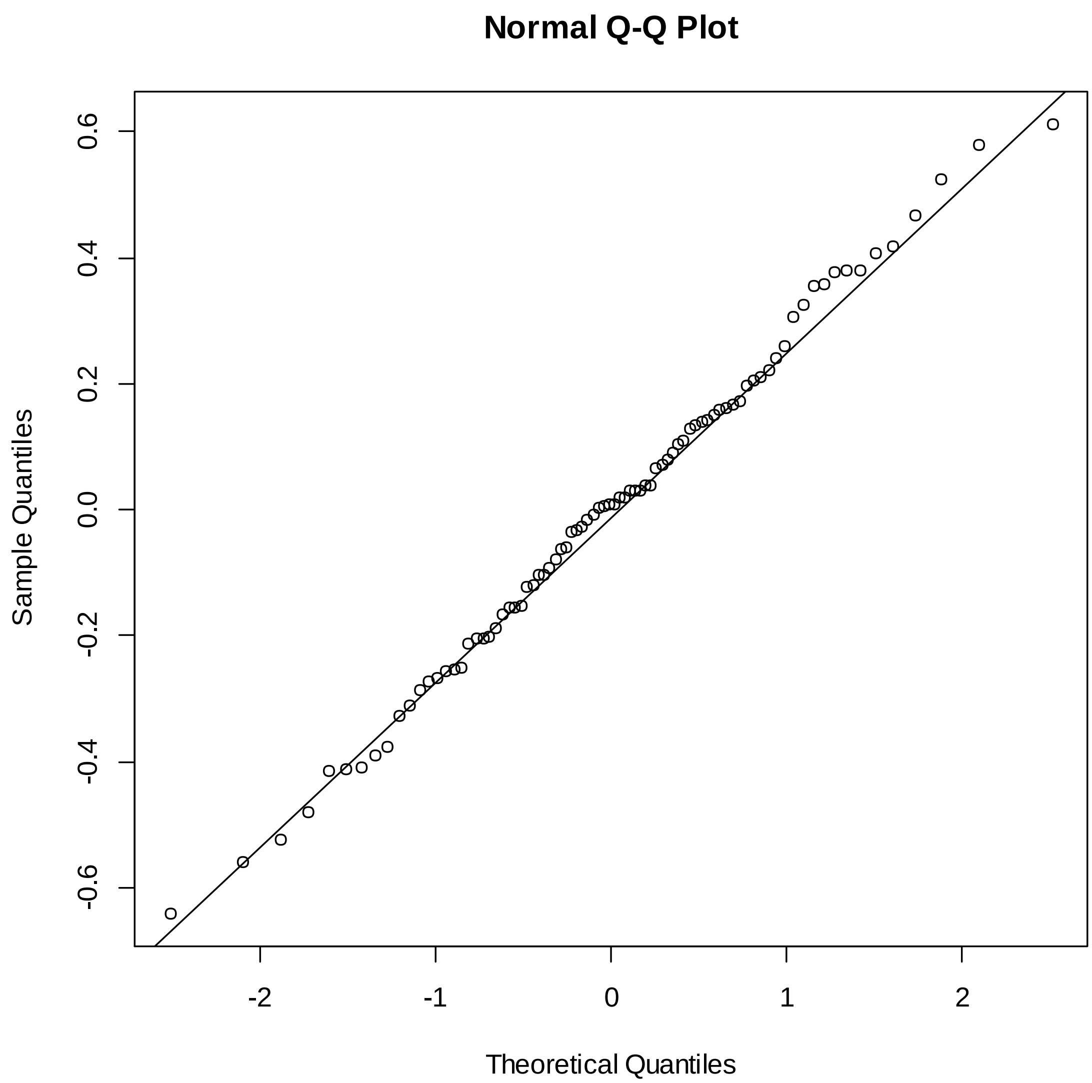
Supplemental Figure B5: Plotted residuals for the full model expressed in Equation 3.1. Plot generated in R 3.5.0.



Supplemental Figure B6: Plotted residuals for the reduced model expressed in Equation 3.3. Plot generated in R 3.5.0.



Supplemental Figure B7: Plotted residuals for the reduced model expressed in Equation 3.4 after Tillage had been removed. Plot generated in R 3.5.0.



Supplemental Figure B8: Plotted residuals for the filtered model expressed in Equation 3.5. Plot generated in R 3.5.0.

Supplemental Table B1: Example of results for stepwise selection in R 3.5.0. This data refers to the initial reduced model expressed in Equation 3.3. Model 8 was the model selected for the reduced model due to a high Adjusted R^2 and low AIC value.

Subsets Regression Summary

Model	Adj. R-Square	Pred R-Square	R-Square	C(p)	AIC	SBIC	SBC	MSEP	FPE	HSP	APC
1	0.3465	0.3417	0.3316	286.0579	518.6571	-269.1296	533.1387	0.3793	0.3793	0.0014	0.6631
2	0.6207	0.6151	0.6064	53.9137	372.5170	-415.6683	394.2394	0.2226	0.2226	8e-04	0.3877
3	0.6553	0.6490	0.64	26.3106	348.0648	-439.6947	373.4076	0.2037	0.2037	7e-04	0.3548
4	0.6720	0.6647	0.6544	14.1005	336.4063	-450.9945	365.3696	0.1953	0.1953	7e-04	0.3401
5	0.6802	0.6718	0.6609	9.1222	331.4431	-455.6847	364.0267	0.1919	0.1918	7e-04	0.3341
6	0.6852	0.6757	0.6626	6.8589	329.1009	-457.7827	365.3049	0.1903	0.1902	7e-04	0.3312
7	0.6912	0.6807	0.6695	3.6997	325.7532	-460.7589	365.5777	0.1881	0.1879	7e-04	0.3272
8	**0.6940**	**0.6825**	**0.6697**	**3.2972**	**325.2271**	**-461.0205**	**368.6719**	**0.1878**	**0.1875**	**7e-04**	**0.3266**
9	0.6954	0.6827	0.6678	4.1026	325.9624	-460.0767	373.0276	0.1883	0.1881	7e-04	0.3275
10	0.6961	0.6822	0.6665	5.5414	327.3663	-458.5007	378.0519	0.1893	0.1890	7e-04	0.3292
11	0.6966	0.6815	0.6656	7.1217	328.9196	-456.7797	383.2256	0.1905	0.1901	7e-04	0.3310
12	0.6966	0.6804	0.6611	9.0401	330.8326	-454.7277	388.7590	0.1919	0.1914	7e-04	0.3333
13	0.6967	0.6792	0.6593	11.0165	332.8075	-452.6192	394.3543	0.1933	0.1928	7e-04	0.3357
14	0.6967	0.6780	0.6576	13.0000	334.7899	-450.5037	399.9571	0.1948	0.1942	7e-04	0.3382

AIC: Akaike Information Criteria
SBIC: Sawa's Bayesian Information Criteria
SBC: Schwarz Bayesian Criteria
MSEP: Estimated error of prediction, assuming multivariate normality
FPE: Final Prediction Error
HSP: Hocking's Sp
APC: Amemiya Prediction Criteria

Supplemental Table B2: Model Index 8 was the model selected for the reduced model expressed in Equation 3.3 based on information in Table B2 due to a high Adjusted R^2 and low AIC value.

Best Subsets Regression

Model Index	Predictors
1	Trt
2	Trt Depth
3	Trt Depth Moisture
4	Trt Depth Moisture Temp
5	Trt Depth Moisture Temp SIC
6	Trt Depth Moisture Temp pH SIC
7	Trt Depth Tillage Rotation Moisture Temp SIC
8	**Trt Depth Tillage Rotation Moisture Temp Dry SIC**
9	Trt Depth Tillage Rotation Moisture Temp Dry pH SIC
10	Trt Depth Tillage Rotation Moisture Temp Sand Dry pH SIC
11	Trt Depth Tillage Rotation Moisture Temp Sand Dry Wet pH SIC
12	Trt Depth Tillage Rotation Moisture Temp Sand Clay Dry Wet pH SIC
13	Trt Depth Tillage Rotation Moisture Temp Sand Clay Dry Wet pH EC SIC
14	Trt Depth Tillage Rotation Yrs_NT Moisture Temp Sand Clay Dry Wet pH EC SIC

Abbreviations- Trt: Treatment, Yrs_NT: Years in No-Till management, Temp: Temperature (°C), Sand : Sand Content (%), Clay: Clay Content (%), Dry: Dry Mollic Color, Wet: Moist Mollic Color, EC: Electrical Conductivity (μS/cm), SIC: Soil Inorganic Carbon (%)