FACTORS INFLUENCING CARBON SEQUESTRATION IN NORTHERN GREAT PLAINS GRASSLANDS

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ABSTRACT

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Soil development is influenced by the five soil forming factors; parent material, climate, landscape, organisms and time. This study was designed to examine the effects of landscape and organisms (vegetation) on carbon (C) in Conservation Reserve Program (CRP), restored grasslands, and undisturbed grasslands across the northern Great Plains of the U. S. using statistical methods. The effects of vegetation, slope, and aspect on C sequestered in the surface 30 cm of the soil for 997 sites sampled across portions of Iowa, Minnesota, Montana, and North and South Dakota were evaluated. A Partial F-test was used to evaluate models to determine the significance of factors and their interaction effects. For the vegetation component of these models, cool season grasses with or without legumes showed higher levels of soil organic C than warm season grasses with or without legumes or mixed cool and warm season grass regimes. When slopes were evaluated, slopes less than 3 % showed higher levels of sequestered C than slopes greater than 3 %. Southern and western aspects showed higher soil C levels than other aspects.

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INTRODUCTION

The Conservation Reserve Program (CRP) was established by the US government in 1985 in order to convert cultivated lands into grasslands with the idea of reducing agricultural over production as well as soil erosion while improving the soil organic matter content although it was not the original intent of CRP. The process of accumulating soil organic carbon (or soil organic matter) which is termed as carbon sequestration was later proposed as a means to biologically reduce CO_2. It is estimated that approximately17 million ha of erodable and environmentally sensitive croplands, when converted to grasslands would sequester about 45% of the 38.1 million tons of carbon released annually into the atmosphere from US agriculture (Gebhart et al. 1994). In addition to carbon sequestration, many other benefits like erosion control and increasing wildlife habitat are the advantages of restored grasslands. Factors that influence the sequestration of soil organic carbon (SOC) include vegetation, soil properties, grassland age class, climate and topography (Franzmeier et al. 1985). The process of carbon sequestration varies between different regions due to the interplay of many of these factors. Understanding the factors responsible for increasing soil carbon sequestration has many implications for reducing the effects of global warming through increasing the acreage of restored grasslands.

The effects and influence of these factors varies among different regions. By studying the factors responsible for higher soil organic carbon, one can improve the estimates and provide knowledge about improving carbon accumulation in different land management systems (Liebig et al. 2005). This preliminary study seeks to understand the influence of vegetation, slope, landscape and aspect on SOC relationships in restored grasslands in the north central Great Plains of the U.S.

LITERATURE REVIEW

Carbon Cycle

Carbon (C) is the fourth most abundant element in the universe and is one of the building blocks of life. It is the central element of all organic substances, from fossil fuels to DNA. On Earth, the carbon cycle occurs with the land, ocean, and atmosphere, all playing a part in the cycle. The global carbon cycle is divided into two categories: geological cycling (terrestrial cycling), which operates over large time scales (millions of years), and the biological/physical, which operates over shorter time scales (days to thousands of years). In the geological process, atmospheric carbon combines with water during rainfall or reacts with surface waters to form carbonic acid. This ultimately reaches the oceans in dissolved state and includes magnesium and calcium. This further reacts with the dissolved CO_2 to form carbonate minerals and are deposited in the ocean. Through the process of subduction, the inorganic carbon minerals reach the mantle of earth. This carbon can then be released into the atmosphere in the form of CO_2 through eruption of volcanoes after the mantle rocks are molten.

Living organisms play a vital role in the carbon cycle through the processes of photosynthesis and respiration both on the land and in the ocean. All forms of life on earth depend directly or indirectly on plants for the sugars produced through the process called photosynthesis. During the process of photosynthesis, green plants absorb solar energy, remove carbon dioxide from the atmosphere and produce carbohydrates (sugars) with the help of water. Plants and animals including humans effectively burn these carbohydrates and their derived products through the process of respiration. Respiration releases the energy necessary for the metabolism and survival of the organisms. During this process,

CO_2 is again released back into the atmosphere. On the other hand, plants and plant products when dead decompose releasing CO_2 back into the atmosphere. It is estimated that the process of photosynthesis accounts for up to 16% of the atmospheric CO_2 being moved from the atmosphere to the biosphere annually (Malhi, 2002). The total amount of carbon taken up by photosynthesis and released by respiration each year is ~1,000 times greater than the amount of carbon that moves through the geological cycle on an annual basis.

Global Warming

Global warming is one of the serious problems that is affecting human societies worldwide. Global warming is an increase of average world temperatures as a result of the greenhouse effect. Certain gases like CO_2, methane, nitrous oxide, chlorofluorocarbons (CFCs) in the atmosphere act like glass in a greenhouse, allowing sunlight to pass through it to heat the earth's surface but traps the heat as it radiates back into the space. As greenhouse gases increase in the atmosphere the earth temperature also increases.

It is estimated that the global average temperature has increased about 0.7 to 1.4 degrees F (0.4 to 0.8 degrees C) since the 1800's (Mastrandrea and Schneider, 2005; NASA's Goddard Institute for Space Studies, 2010, [http://www.giss.nasa.gov/research /news/20110112/]). It is also predicted that the average temperature will rise an additional 2.5 to 10.4 degrees F (1.4 to 5.8 degrees C) by 2100, a rate much larger than in the past (Mastrandrea and Schmeider, 2005). This increase in temperature cannot readily be adapted by many organisms. Human societies and natural ecosystems may have difficulty in adapting to this increase in temperature in the future. The last two decades were the hottest in 400 years and possibly the warmest for several millennia. According to United

Nations Intergovernmental Panel on Climate Change (Solomon, 2007), 11 of the past 12 years are among the dozen warmest years since 1850.

Global Warming- Causes

Global warming is mainly caused by disruption of the natural carbon cycle by increasing the concentration of CO_2 in the atmosphere mainly due to increased human activities. These contribute to global warming by enhancing earth's natural greenhouse effect. The human activities that contribute to global warming are the burning of fossil fuels (coal, oil, and natural gas) and the clearing of land. Modernization and industrialization have played a significant role in increasing the atmospheric CO_2 accumulation (Adger and Brown, 1995). Burning of fossil fuels occurs in automobiles, in factories, and in electric power plants. Clearing the land and deforestation contributes to the build up of CO_2 levels in the atmosphere and by the decomposition of dead vegetation (dry wood is about 50 percent carbon).

Since the 1990s, yearly emissions have gone up to 6 billion metric tons of "CO_2 equivalent" worldwide, more than a 20% increase (Solomon, 2007). It is estimated that the burning of C-containing fossil fuels contributes about 230 gigatons of carbon (GtC), and deforestation that leads to the burning of trees contributes about 122 ± 40 GtC (Schimel et al., 1995), and soil tillage contributes about 68 GtC (reviewed by Augustin, 2009). Another estimate predicts that fossil fuel burning releases ~ 5.5 GtC per year into the atmosphere and land-use changes such as deforestation contribute ~ 1.6 GtC per year. Of this total amount of 7.1 GtC released per year by human activities, approximately 3.2 GtC remain in the atmosphere, resulting in an increase in the atmospheric carbon dioxide concentration.

Combustion of fossil fuels has caused mean concentrations of CO_2 in the atmosphere to reach and exceed 380 lmol mol^{-1}, a level that is about 0.32 times greater than that in pre-industrial times (Keeling and Whorf, 2000). Predictions of future atmospheric CO_2 concentration in the year 2100 range between 540 and 970 lmol mol^{-1} (Houghton and Ding, 2001). Additional inputs of C to the atmosphere will produce further warming (Houghton and Ding, 2001) and contribute to the occurrence of more intense, more frequent and longer spells of high temperatures than expected (Meehl and Tebaldi, 2004).

Agriculture occupies a larger portion of global land area (about 35%) than any other human activity (Betts et al., 2007). Because of its large scale and intensity, agriculture emits a large quantity of greenhouse gases into the atmosphere (Salinger, 2007). It presently accounts for about 25% of the CO_2, 50% of the CH_4 and 70% of the N_2O released globally via human sources.(Hutchinson et al., 2007). However, it also has a potential to be a sink.

Global Warming Problems

Global warming contributes to an increase in temperature and affects living organisms in many ways including disrupting the ecosystem required for their survival. In addition it affects human life in many ways. According to Solomon (2007) predictions, the sea level could rise between 7 and 23 inches (18 to 59 centimeters) by century's end (by 2100) which would affect millions of people worldwide living in coastal areas due to flooding and loss of wetlands. Also, glaciers around the world could melt, causing sea levels to rise and create a fresh water shortage. Millions of species will become extinct due to disappearing habitat and a changing environment. All of these patterns force plants and animals into new habitats. The drastic effects may also include an increase in the frequency of storms, hurricanes, floods and drought.

Limiting Global Warming

The two human driven activities to limit global warming are (i) limiting CO_2 emissions and (ii) carbon sequestration that either prevents carbon dioxide from entering the atmosphere or removing CO_2 already present. Effective techniques for limiting CO_2 emissions are to replace fossil fuels with energy sources that do not emit CO_2, and use fossil fuels more efficiently. Alternative energy sources like wind, sunlight, nuclear energy do not emit CO_2. Even though the alternative sources of energy are more expensive than fossil fuels, increasing research on alternative energy would reduce their cost.

The International Panel on Climate Change (Solomon, 2007) distinguished three main options for the mitigation of atmospheric CO_2 concentrations by the agricultural sector: (i) reduction of agriculture-related emissions, (ii) creation and strengthening of C sinks in the soil, and (iii) production of biofuels to replace fossil fuels (Batjes, 1998).

Carbon Sequestration

Carbon sequestration is the act of capturing atmospheric carbon and storing it in a stable form in the soil to reduce the amount of CO_2 in the atmosphere (Burras et al., 2001). Carbon sequestration could also take the form of underground / underwater storage and storage in living plants (Reichle et al., 1999). Soil C sequestration means increasing the SOC and SIC levels through judicious land use and recommended management practices (Lal, 2004).

Underground or underwater storage involves injecting industrial emissions of CO_2 into underground geologic formations such as natural reservoirs of oil and gas or into the ocean bed. This method is generally much cheaper than other methods employed for carbon sequestration (Mastrandrea and Schneider, 2005). This method is termed as direct carbon sequestration (Reichle et al., 1999)

The important biological strategy to sequester atmospheric CO_2 into non gaseous forms in the soil is through plants (Schimel et al., 1995), and is termed as indirect carbon sequestration (Reichle et al., 1999). Green plants absorb CO_2 from the atmosphere and convert it into carbohydrate materials during the process of photosynthesis. After plants are harvested or after the death, the plant parts decay and release CO_2, which is a slow process. However, a portion of decay products condense into more stable organic compounds that have lifetime of years to centuries to millennia.Through these natural processes, more and more CO_2 can be fixed by the plants and stored in the soil. Ecosystems like forests and even croplands, could sequester more carbon due to more photosynthesis followed by lesser decay of the plant material.

The global soil carbon (C) pool of 2500 gigatons (Gt) includes about 1550 Gt of soil organic carbon (SOC) and 950 Gt of soil inorganic carbon (SIC) (Lal, 2004). The soil C pool is 3.3 times the size of the atmospheric pool (760 Gt) and 4.5 times the size of the biotic pool (560 Gt) (Lal, 2004). The SOC pool to 1-m depth ranges from 30 Mg/ha in arid climates to 800 Mg/ha in organic soils in cold regions, with a predominant range of 50 to 150 Mg/ha. (Lal, 2004).

The main reasons for C losses from the soil carbon pool include destruction of ecosystems like forests, and native grasslands followed by intense agricultural practices. It is estimated that on an average, agricultural practices result in ~ 75% loss of soil nitrogen and ~ 89% loss of soil carbon loss (Knops and Tilman, 2000). Recovery of soil C to 95% of the pre-agricultural levels is predicted to require ~180 yr for nitrogen and ~230 yr for carbon (Knops and Tilman , 2000). Agriculture and land-use changes contribute ~ 20% of the total emissions of CO_2 to the atmosphere (Dumanski and Lal, 2004). Average soil organic carbon (SOC) in the top 30 cm of native soil worldwide is approximately 15 Mg ha^{-1}. When cultivated, ~ 20–30% of this carbon is released to the atmosphere in temperate regions and 50–75% in the tropics within the first 20 years of conversion (Dumanski and Lal, 2004). In the US, an estimated 35.4 million metric tons C is released to the atmosphere every year from the use of agricultural fossil fuels and the manufacture of nitrogen fertilizers. These numbers indicate that agricultural activities are significant contributors of CO_2 gas to the atmosphere (Gebhart et al.,1994).

Another estimate is that the average loss of soil organic carbon (SOC) in the top 1m is between 15 to 40% within 2–8 years following conversion of native tropical vegetation to agriculture (Ingram and Fernandes, 2001). Average relative loss of SOC was 12.1 ± 7.9 g C kg^{-1} soil for soil depths ≤ 30 cm. Previous estimates of SOC loss in agricultural lands in the central and northern Great Plains ranged from 20 to 53% (Cihacek and Ulmer, 1995; Liebig et al., 2005).

Need for Sequestration

Carbon sequestration has the potential to offset fossil fuel emissions by 0.4 to 1.2 gigatons of carbon per year, or ~5 to 15% of the global fossil-fuel emissions (Lal, 2004). With this, the main option for greenhouse gas (GHG) mitigation identified by the Solomon is the sequestration of carbon in soils (Hutchinson et al., 2007). The U.S. Department of

Energy (DOE) established a Carbon Sequestration Program within the Office of Fossil Energy (FE) in 1997, and a basic research program in the Office of Science (OS) in 1999 (http://fossil.energy.gov/sequestration/overview.html). Both these programs seek to move sequestration technology forward so that its potential can be realized and improved. These programs also implement the President's Global Climate Change Initiative, and several National Energy Policy goals directly targeting the development of new technologies, market mechanisms, and increasing the international collaboration to reduce greenhouse gas intensity and greenhouse gas emissions (Reichle et al., 1999).

The main factors available to increase the SOC content in soil through the sequestration process is by efficient land use management practices and restoration of native grasslands and ecosystems. Efficient management practices include no-till farming, cover crops, nutrient management, manuring and sludge application, improved grazing, water conservation and harvesting, efficient irrigation, agroforestry practices, growing energy crops on spare lands, and the use of deep rooted, fast-growing tree and grass species in the tropics (Lal, 2004). It is estimated that an increase of 1 ton in the soil carbon pool of degraded cropland soils can increase crop yield by 20 to 40 kilograms per hectare (kg/ha) for wheat, 10 to 20 kg/ha for maize, and 0.5 to 1 kg/ha for cowpeas (Lal, 2004).

Estimates of the total potential of C sequestration in world soils vary between 0.4 to 1.2 Gt C/year (Lal, 2004). Paustian et al. (1997) estimated that over the next 50–100 years, the global estimates of C sequestration capacity in agricultural soils are in the order of 20–30 Pg C. It is estimated that C accumulation rates in tropical agroforestry systems range from 4 to 9 Mg C ha^{-1} $year^{-1}$, with more potential above ground than in the soil (Hutchinson et al., 2007). Over a normal agroforestry rotation of 20–25 years, C accumulations above-ground can be ~ 50 Mg C ha^{-1}, and in the soil can be ~ 50 Mg C ha^{-1} (Hutchinson et al., 2007)

The estimates of carbon sequestration costs are in the range of $100 to $300/ton of carbon emissions avoided using the present day technologies (Reichle et al., 1999). The ultimate goal of research is to achieve sequestration with less than $10/ton CO_2 avoided. Achieving this goal would save the United States and other countries trillions of dollars and carbon sequestration in soil can be a cost effective means of lowering CO_2 emissions to the atmosphere (Reichle et al., 1999).

Praire and Grasslands

Prairie is an ecologically important ecosystem in North America that once stretched from Canada to Mexico. Prairie ecosystems are dominated by grass species as the primary producer of energy within the ecosystem. Currently, tallgrass prairie has declined to about 1% of its historic extents (Samson and Knopf, 1994).

Grasslands are an ecosystem with highly varying characteristics and occur with wide ranges in temperature, precipitation, age, geographic location, and other properties. Prairies and grasslands are considered as a major sink for SOC because of high photosynthesis and soil biomass. These are considered superior sinks in comparison to forests with similar environmental characteristics (Seastedt and Knapp, 1993). With the goal to reduce CO_2 concentration in the atmosphere, there is a quick need to restore the unused land or cultivated land into grassland.

When grasslands that have been cultivated are planted back into perennial cover, the SOC levels within the soil can shift towards the original levels of the native grassland.

Recently, much research focus has been placed on grasslands to understand grassland relationships with accumulation of SOC and the potential management practices that can improve SOC levels within the soil. The soil organicC sequestration potential is estimated to be 0.01–0.3 Gt C year^{-1} on the 3.7 billion ha of permanent pasture worldwide (Lal, 2004). Thus soil organicC sequestration by the world's permanent pastures could potentially offset proportionately up to 0.04 of the global emissions of greenhouse gases.(Soussana and Luscher, 2007).

Grassland covers about 70% of the world's agricultural area (Soussana and Luscher, 2007). Higher atmospheric CO_2 enhances the growth of plants. So, a rise in atmospheric CO_2 concentrations are likely to affect several important aspects of grasslands, like the quantity and quality of the herbage produced, plant species composition, soil fertility and the potential to sequester carbon, potential to mitigate the rise in atmospheric CO_2 concentrations (Soussana and Luscher, 2007).

Properties of native, undisturbed ecosystems like grasslands can serve as the target properties for ecosystem restorations (Zedler and Lindig-Cisneros, 2000; Brye et al., 2008). Thus soil C sequestration rates observed and documented in native prairies serve as guidelines for expected rates of soil C sequestration or as an indication of soil C sequestration potential in Grassland restorations (Brye, 2009).

CRP Program

The conservation reserve program (CRP) is a government sponsored program started in 1985 that has evolved with the idea of increasing the carbon sequestration by increasing the natural habitat (i.e converting cultivated and unused lands into grasslands).

This program provides technical and financial assistance to eligible farmers and ranchers to address soil, water, and related natural resource concerns on their lands. In addition to increasing the amount of carbon sequestered, this program helps reduce soil erosion, protects the Nation's ability to produce food and fiber, reduces sedimentation in streams and lakes, improves water quality, establishes wildlife habitat, and enhances forest and wetland resources. This program encourages farmers to convert highly erodible cropland or other environmentally sensitive acreage to vegetative cover, such as native grasses, wildlife plantings, trees, etc. (Brye, 2009). Farmers receive an annual rental payment for the multi-year contract. The contracts normally are for a 10-year period with an option to extend the contract for additional 10-year terms upon expiration of original contract.

Different types of studies have been used to investigate the dynamics of SOC after converting agricultural fields into grasslands. These include repeated sampling of old fields (Knops and Tilman, 2000), paired sites (Baer et al., 2000), and unreplicated chronosequences (Jastrow, 1987). Most of these studies focus on the changes in surface soils during the first decade after agricultural abandonment, and estimated rates of SOC accumulation vary significantly (McLauchlan et al., 2006).

The reversion of agricultural land to grassland usually results in an increase in SOC (Nelson et al. 2008).Numerous studies have predicted a wide range of annual SOC sequestration rates in restored grasslands ranging from annual losses (Follett et al., 2001b) to gains on a global scale of 3.04 Mg C ha-1 yr-1 (Conant et al., 2001). It is estimated that the C sequestration can be as high as 1.01 Mg C ha^{-1} year^{-1} when converted from cultivated land to grassland (Conant et al., 2001). This estimate is larger than the one estimated by Smith et al. (2000) using the CENTURY model for Canada, a value of 0.62 Mg C ha^{-1} year^{-}

^{1} (Hutchinson et al., 2007). Another estimate showed that SOC contents of the surface 0 to 25 cm (0 to 10 in) of converted to perennial grasses were similar to that of adjacent native rangelands following about 50 years of recovery (Gebhart et al., 1994).With an average of 1. 1 tons C $ha^{-1}yr^{-1}$ accumulation of SOC, 17 million hectares of land enrolled in CRP have the potential to sequester about 45% of the 38.1 million tons of carbon released annually into the atmosphere from US. agriculture (Gebhart et al.,1994).

The regional specific rates varied among different regions based on the interplay of the many factors responsible for carbon sequestration. In the northwestern U.S. and western Canada, the estimate is 0.94 ± 0.86 Mg C ha-1 yr-1 of SOC (Liebig et al., 2005). In Saskatchewan, grassland restoration was reported to result in the sequestration of 0.6 to 1.2 Mg C ha-1 year-1 in the top 15 cm of soil over a period of approximately ten years (Nelson et al., 2008). Post and Kwon (2000) estimated that short-term soil accumulation rates are 30 to 60 g $C{\cdot}m^{-2}{\cdot}yr^{-1}$ on former agricultural fields when converted to grasslands. In Texas, Kansas and Nebraska, 5 years after restoration to a perennial grass cover, 21% of the soil carbon lost during decades of intensive tillage had been replaced in the top 40cm of soil (Gebhart et al., 1994). Blevins et al. (1983) showed that after 10 years of abandonment of corn production, SOC in the surface 30 cm (12 in) was increased by 25%.

Policies promoting wetland conservation and restoration, such as the U.S. Department of Agriculture (USDA) Conservation Reserve Program (CRP) and Wetlands Reserve Program (WRP), have led to the restoration of approximately 2.2 million ha (5.4 million acres) of wetland and grassland habitats in the Prairie Pothole Region of the United States (Gleason et al., 2008).

Approximately 5.5 million acres in the United States have been enrolled in the North Dakota Farmer's Union (NDFU) Carbon Credit Program. The estimate of CO_2 sequestered by no-till practices in North Dakota is 0.4 Mg CO_2 acre^{-1} year^{-1} and the grassland planting estimate is 1.0 Mg CO_2 acre^{-1} year^{-1}(Augustin, 2009). It should also be noted that it takes sequestration of 3.66 Mg CO_2 ha^{-1} to equal 1 Mg of soil organic carbon (Augustin, 2009).

The main reasons for the increase in SOC after converting agricultural lands to grasslands are elimination of tillage, reduced soil erosion, and accumulation of surface litter (Gebhart et al., 1994). Quantifying the rate of increase in soil organic matter (SOM) is important for understanding global C cycles, the long-term fertility of agricultural systems and restored grasslands, and how long it takes grassland systems to recover from agriculture (McLauchlan et al., 2006). With the potential for economic gain from grassland restorations, the number of restorations will increase in the future as a result of their soil carbon sequestration potential (Brye, 2009).

Factors Effecting SOC in Soil

Carbon sequestration is variable among different regions and varies with many factors like soil properties, temperature, precipitation, vegetation etc. Soil organic carbon levels within the soil are related to parent material, organisms, topography, climate and time (Franzmeier et al., 1985). Many studies have described the soil organic carbon (SOC) sequestration factors in local areas.

Soil Depths and Horizons

Studies have looked at different depths or soil horizons (Reeder et al., 1998) to determine SOC storage levels. It is found that the majority of the SOM is located in the top 5 cm of the soil profile (Ganjegunte et al., 2005), while other studies suggest that deep sampling is needed as different plant types and other situations influence SOC content in the deeper soil horizons in the soil profile (Davidson and Ackerman, 1993, Baker et al., 2007). It is estimated that CRP lands have gained an average of 0.8 metric tons C ha^{-1} yr^{-1} (700 lbs C ac-' yr') to a depth of 40 cm (16 in) and 1.1 metric tons C ha^{-1}yr^{-1} (1000 lbs C ac^{-1} y r^{-1}) to a depth of 300 cm (120 in) in Texas, Kansas and Nebraska (Gebhart et al., 1994).

Vegetation

Grassland vegetation affects soil properties through several mechanisms related to plant tissue chemistry, productivity, and morphology (McLauchlan et al., 2006). Even though C_4 grasses have lower N concentrations in the tissues and higher lignin concentrations than C_3 grasses (Sage and Monson, 1999), recent works suggests this may not be universally true (McLauchlan et al., 2006). It is found that due to presence of legumes such as alfalfa (*Medicago sativa*), that C_3 vegetation had higher percentage lignin, as well as N, than C_4 vegetation.

It is suggested that vegetation characteristics, like Net Primary Productivity (NPP), tissue N concentration, and lignin concentration, are uncorrelated with SOC content, SOC accumulation rates, or proportion of recalcitrant C formed (McLauchlan et al., 2006). McLauchlan et al. (2006), based on his results suggested that vegetation characteristics are not very important in determining the rate of SOC increase but are important in determining the equilibrium amounts, even though some SOC models such as CENTURY model assume that plant characteristics like lignin concentration affect the rate and type of SOC formation (Parton et al., 1987, Ogle et al., 2004), and have been verified in agricultural systems (Paustian et al., 1992). Knops and Tilman (2000) indicated that carbon accumulations are controlled by nitrogen accumulation in the soil and are positively correlated by the presence of legumes and C_4 grass species and negatively correlated by forbs and C_3 grass species. Raven et al. (2005) indicates that C_4 plants are better suited to higher temperatures than C_3 plants, use carbon dioxide more efficiently, use less water, and use nitrogen more efficiently.

Burke et al. (1995a) suggest that it is difficult to isolate the role of the vegetation factor in many previous studies because vegetation colonization and succession dynamics are unavoidable in the abandoned agricultural fields. Ogle et al. (2004) suggested that certain invasive grasses have the potential to alter SOC content differently than native plants. Christian and Wilson (1999) compared converted and abandoned grasslands and found that different vegetation composition effects ecosystem properties, including SOC, between the two different grassland types. Another factor contributing to observed differences in SOC between CRP and cropland may have been the greater root biomass associated with the establishment of perennial grasses(Gebhart et al., 1994). Plant root characteristics vary greatly by species, climate, soil, and time. Modeling C sequestration requires detailed understanding of a soil's root system.

Management Practices

Management practices have an influence on the rate of SOC accumulation in restored grasslands (Riopel, 2009). It has been shown that when SOM levels change, nutrient availability affects SOM accumulation in the short term (Burke et al., 1995 a;b).

Soil organic carbon losses due to cultivation averaged 61% in the surface 0 to 5 cm (0 to 2 in) increment and gradually declined with depth (Gebhart et al., 1994). Cultivation also increases soil temperature and microbial activity with better aerated conditions which increase the rate of organic matter decomposition resulting in subsequent SOC loss (Gebhart et al., 1994).

The effect of several management factors have been studied extensively. A few factors that contribute to changes in SOC include nutrient additions via fertilizer (USA) (Lal et al., 1998), nutrient additions via manure (USA) (Follett, 2001a), converting cultivated lands to grasslands (Canada) (Smith et al., 2001), adopting conservation tillage (USA) (Lal, 2001), conversion from conventional till to no-till (USA) (Lal, 1997), improved grassland management (USA) (Lal 2001), and reduction of summer fallow (Lal 2001)

Better management practices such as an increase in cropping frequency (reducing bare fallow), increasing use of forages in crop rotations, reducing tillage intensity and frequency, better crop residue management, and adopting agroforestry, improved fertility of cropland/pasture, woodland regeneration, nutrient management, manuring and sludge application, improved grazing, water conservation and harvesting, efficient irrigation, growing energy crops on spare lands increase SOC content (Lal, 2004; Hutchinson et al., 2007).

Soil Properties

Many research projects have studied the relationships of soil physical properties like texture, structure, bulk density and soil temperature to predict soil C accumulation or sequestration. (Sims and Nielsen, 1986; Burke et al., 1989; Amato and Ladd, 1992;

Franzluebbers et al., 1996; Young and Hammer, 2000a,b; Kahle et al., 2003; Hao and Kravchenko, 2007). Some have also observed and developed soil C models by observing soil colors (Konen et al., 2003; Wills et al., 2007). It is estimated that CRP soils gained an average of 7.9 metric tons C ha^{-1} to a depth of 40 cm and 9.0 metric tons C ha^{-1} to a depth of 300 cm, whereas sandy textured CRP soils gained 1.7 metric tons C ha^{-1} to a depth of 40 cm and 4.3 metric tons C ha^{-1} to a depth of 300 cm (Reviewed by Augustin, 2009). Only about 15% of the total carbon gain for loamy textured CRP soils occurred below a depth of 40 cm, while 60% of the total carbon gain for sandy textured CRP soils occurred below 40 cm in depth.(Gebhart et al., 1994). Finer textured soils generally have higher SOC contents than coarse textured soils when supplied with the same amount of organic inputs.(Ingram and Fernandes, 2001).

Climate and Other Factors

The effect of climate on SOC has been an important area of study since the 1950’s (Burke et al., 1989). It has been found that SOC increases as mean annual precipitation (MAP) increases up to values of 700–850 mm yr^{-1} (Guo et al., 2006). It has also been observed that C and N storage in the soil is a function of climate (Post et al., 1982; Nichols, 1984; Burke et al., 1989). Burke et al. (1989) found that organic C increased with increasing precipitation and decreased with increasing temperature.Other factors have been studied include landscape position (Schimel et al., 1985, Yonker et al., 1988), and topography (Chaplot et al., 2001) from local to regional levels (Guo et al., 2006).

Organic Carbon in North Central Great Plain Soils

Soil organic carbon levels within soils can vary greatly between different regions in the north central Great Plains (Riopel, 2009). Soil organic carbon accumulation rates may be dependent on different factors like nitrogen accumulation (Knops and Tilman, 2000), landscape slope (Franzmeier et al., 1985), landscape position (Yonker et al., 1988), parent material (Franzmeier et al., 1985), and vegetation (Franzmeier et al., 1985). Other factors include grazing and haying (Franzluebbers et al., 2000; Liebig et al., 2005), burning (Fynn et al., 2003), and plant type (Knops and Tilman, 2000; Fornara and Tilman, 2008), tillage, nitrogen, and cropping systems (Halvorson et al., 2002).

Statistical Analysis Used in Previous Studies

Most of the previous research is based on comparing the different levels of factors using a traditional ANOVA or effect of the factors using a stepwise regression. Burke et al.(1995a) used nested analysis of variance using a Scheffe procedure to look at the effects of sites (native, abandoned, and cultivated), depths and microsites. To analyze SOC accumulation rates in restored grasslands, multiple stepwise regression was used to correlate SOC to factors like soil texture and climate (Burke et al., 1989), vegetation variables, field age and their interaction (McLauchlan et al., 2006) . Gleason et al. (2008) also used analysis of variance to observe the differences between SOC and vegetation organic carbon (VOC) among land-use treatments (drained, hydrologically non drained and non drained restored and native prairie) and used native grasslands and/or cultivated fields to compare the levels of SOC with the restored grasslands. Burke et al. (1989) used all possible subset regression analysis in a full quadratic model to find the best predictive equation for soil organic C and N. Draper and Smith (1966) chose the best regression equation based on adjusted R^2 and Cp statistics. Nelson et al. (2008) used a two sample t-test with unequal variances. For significant main effects and interaction terms, specific ANOVA tests using contrast statements were applied to the observed levels of random effects (Gleason et al., 2008). McLauchlan et al. (2006) used a multiple stepwise regression using backward elimination with $p < 0.05$ as the cutoff for significance was used to test the effects of several potential predictor variables on SOC content. Gebhart et al. (1994) used paired t-tests to separate land use means within each location.

Proposed Statistics

Multiple regression is similar to simple linear regression with a greater number of variables and the tests can be performed under the assumption that the random error terms, ε_i , are normally and independently distributed with a mean of zero and variance of σ^2 (Abraham and Ledolter, 2006).

Three types of hypothesis tests can be performed for multiple linear regression models (Abraham and Ledolter, 2006): 1) Test for significance of regression, which checks for the significance of the whole regression model, 2) t-tests, which check the significance of individual variables adjusted for all the other variables being in the model and 3) Partial F-tests, to simultaneously check the significance of a number of variables with other variables being in the model.

1) Test for Significance of Regression

The test for significance of regression in multiple linear regression analysis is performed using the analysis of variance. This test is used to check if a linear statistical relationship exists between the response variable (Y) and at least one of the predictor variables (X_1, X_2, X_3,....X_k). The model and hypotheses statements are

$$Y = \beta_o + \beta_1 X_1 + \beta_2 X_2 + \cdots + \beta_k X_k + \varepsilon$$

$$H_0: \beta_1 = \beta_2 = ... = \beta_k = 0$$

$$\text{and } H_1: \beta_j \neq 0 \text{ for at least one } j$$

The test for H_0 is performed using the following statistic: $F_0 = MS_R / MS_E$ where MS_R is the regression mean square and MS_E is the error mean square. If the null hypothesis (H_0), is true then the statistic F_0 follows the F distribution with 'k' degrees of freedom in the numerator and 'n-(k+1)' degrees of freedom in the denominator. The null hypothesis, H_0, is rejected if the calculated statistic, $F_0 > f_{\alpha,k,n-(k+1)}$

Calculation of The Statistic F_0

To calculate the statistic F_0, the mean squares MS_R and MS_E must be known. The mean squares (MS) are obtained by dividing the sum of squares (SS) by their degrees of freedom. The total mean square, MS_T, is obtained as follows: $MS_T = SS_T / df$, where SS_T is the total sum of squares and 'df' is the number of degrees of freedom associated with SS_T, n-1. In multiple linear regression, SS_T is calculated by $SS_T = \sum (y_i - ӯ)^2$, where n is the total number of observations, y_i is the vector of observations, ӯ is the mean of the observations.

The regression mean square, MS_R, is obtained by dividing the regression sum of squares, SS_R, by the respective degrees of freedom (df) , as follows: $MS_R = SS_R / df$. The regression sum of squares, SS_R, is calculated using the following equation: $SS_R = \sum (\hat{y}_i - ӯ)^2$. The number of degrees of freedom associated with SS_R, is k, where k is the number of predictor variables in the model. Knowing SS_R and df, the regression mean square, MS_R, can be calculated. The error mean square, MS_E, is obtained by dividing the error sum of squares, SS_E, by the respective degrees of freedom (df). The error sum of squares, SS_E, is calculated using the following equation: $SS_E = \sum (y_i - \hat{y}_i)^2$. The number of degrees of freedom associated with SS_E, is n-k+1, where n is the total number of observations and k is the number of predictor variables in the model. The error mean square is an estimate of the variance, σ^2, of the random error terms, ε_i. $\sigma_{hat}{}^2 = MS_E$.

2) Test on Individual Regression Coefficients (t-test)

The t- test is used to check the significance of individual regression coefficients in the multiple linear regression models. Adding a significant variable to a regression model makes the model more effective, while adding an unimportant variable doesn't add anything to the model. The hypothesis statements to test the significance of a particular regression coefficient β_j, are: H_0: $\beta_j = 0$ and H_1: $\beta_j \neq 0$. The test statistic for this test is based on the t statistic, $T_0 = \beta'_j / se(\beta'_j)$ where the standard error ($se(\beta'_j)$), is obtained from above. We do not reject the null hypothesis if the test statistic lies in the acceptance region, $-t_{\alpha/2, n-2} < T_0 < t_{\alpha/2, n-2}$.

This test measures the contribution of a variable while the remaining variables are included in the model. For the model

$Y = \beta_o + \beta_1 X_1 + \beta_2 X_2 + \beta_3 X_3 + \varepsilon$, if the test is performed for β_1, then it will check the significance of excluding the variable X_1 in the model that contains X_2 and X_3 (i.e. the model $Y = \beta_o + \beta_2 X_2 + \beta_3 X_3 + \varepsilon$). Hence the test is also referred to as a *partial* or *marginal test*.

3) Test on Subsets of Regression Coefficients (Partial F- test)

A partial F-test is performed on subsets of variables. There are two methods of calculating extra sums of squares, i) A partial sum of squares and ii) sequential sum of squares.

The partial F- test can be considered to be the general form of the t- test described earlier. This is because the test simultaneously checks the significance of including many (or even one) regression coefficients in the multiple linear regression model with a given set of variables in model. Adding a variable to a model increases the regression sum of squares (SS_R). The test is based on this increase in the regression sum of squares. The increase in the regression sum of squares is known as the *extra sum of squares*.

Assume that the vector of the regression coefficients, β for the multiple linear regression model, $Y = X\beta + \varepsilon$, is partitioned into two vectors with the second vector, β_2, containing the last r regression coefficients, and the first vector, β_1, containing the first (k+1- r) coefficients as follows: $\beta = [\beta_1, \beta_2]'$ with $\beta_1 = [\beta_0, \beta_1 \ldots \beta_{k-r}]'$ and $\beta_2 = [\beta_{k-r+1}, \beta_{k-r+2} \ldots \beta_k]'$. The hypothesis statements to test the significance of adding the variables $X_{k-r+1}, X_{k-r+2}, \ldots X_k$ to a model containing the variables $X_1, X_2, \ldots X_{k-r}$ are written as

$$H_0: \beta_2 = 0 \text{ and } H_1: \beta_2 \neq 0$$

The test statistic for this test follows the F distribution and is calculated as

$$F_0 = [(SSE_R - SSE_C)/k\text{-}g] / MS_E$$

where SSE_R - SSE_C is the increase in the regression sum of squares when the variables corresponding to the coefficients in β_2 are added to a model already containing $X_1, X_2, \ldots X_{k-r}$, and MS_E is obtained as described earlier.

The null hypothesis, H_0, is rejected if $F_0 > f_{\alpha,\, r,\, n-(k+1)}$. Rejection of H_0 leads to the conclusion that at least one of the variables in $X_{k-r+1},\ X_{k-r+2} \ldots X_k$, contributes significantly to the regression model.

The extra sum of squares can be calculated using either the partial (or adjusted) sum of squares or the sequential sum of squares. The type of extra sum of squares used in the model affects the calculation of the test statistic described above.

Partial Sum of Squares

The partial sum of squares for a variable is the extra sum of squares when all variables, except the variable under consideration, are included in the model. For example, consider the model:

$$Y = \beta_0 + \beta_1 X_1 + \beta_2 X_2 + \beta_3 X_3 + \varepsilon$$

For example, the partial sum of squares for X_2 is the increase in the regression sum of squares when X_2 is added to the model. This increase is the difference in the regression sum of squares for the full model and the model that includes all variables except X_2. These variables are X_1, and X_3. The model that contains these terms is:

$Y = \beta_0 + \beta_1 x_1 + \beta_3 X_3 + \varepsilon$ (reduced model). The partial sum of squares for X_2 can be represented as

$SS_R(\beta_2|\beta_0, \beta_1, \beta_3) = SS_R$ for full model – SS_R for reduced model.

$$= SS_R(X_1, X_2, X_3) - SS_R(X_1, X_3)$$

Sequential Sum of Squares

The sequential sum of squares for a variable is the extra sum of squares when variables are added to the model in a sequence. For example, consider the model:

$$Y = \beta_0 + \beta_1 X_1 + \beta_2 X_2 + \beta_{12} X_1 X_2 + \beta_3 X_3 + \beta_{13} X_1 X_3 + \beta_{23} X_2 X_3 + \beta_{123} X_1 X_2 X_3 + \varepsilon$$

The sequential sum of squares for X_1X_3 is the increase in the regression sum of squares when X_1X_3 is added to the model. This extra sum of squares can be obtained by taking the difference between the regression sum of squares for the model after X_1X_3 was added and the regression sum of squares for the model before X_1X_3 was added to the model. The model after X_1X_3 is added is as follows

$$Y = \beta_0 + \beta_1 X_1 + \beta_2 X_2 + \beta_{12} X_1 X_2 + \beta_3 X_3 + \beta_{13} X_1 X_3 + \varepsilon$$

This is because to maintain the sequence of previous equation all variables preceding X_1X_3 must be included in the model. These are the variables X_1, X_2, X_1X_2 and X_3.

Similarly the model before X_1X_3 is added must contain all coefficients of above equation except X_1X_3. This model can be obtained as follows,

$$y = \beta_0 + \beta_1 X_1 + \beta_2 X_2 + \beta_{12} X_1 X_2 + \beta_3 X_3 + \varepsilon$$

The sequential sum of squares for X_1X_3 can be calculated as follows:

SS_R for full model – SS_R for reduced model.

$$SS_R (X_1, X_2, X_1X_2, X_3, X_1X_3) - SS_R (X_1, X_2, X_1X_2, X_3)$$

The objective of this study is to study the influence of factors like vegetation, slope and aspect as well as their possible interactions on soil organic carbon content using multiple regression analysis, including overall and partial F-tests.

MATERIALS AND METHODS

Sampling Locations

The sampling locations for this project are located in five states of the north Central US plains, more specifically the prairie pothole region. A total of 7 locations were sampled in north eastern Montana, central North Dakota, north-eastern North Dakota, north-central South Dakota, central South Dakota, western Minnesota, north-central Iowa and southern Minnesota. The sites selected give a good representation of the Prairie Pothole Region of the northern Great Plains of the United States and are selected based on long-term wetland study locations conducted by the U.S. Geological Survey (Riopel, 2009). Although, the study sites were adjacent to many of the wetland study areas, they did not include wetlands. The site locations of this study varied geographically by Major Land Resource Areas (MLRA) (Table1), with a range in precipitation and temperature (Table 2). Comparisons of these geographic locations will be made with in MLRA for each area except for the Central North Dakota and Central South Dakota locations. At these locations, the sampling was done along the boundaries and can be considered as transition areas between the MLRA's with similar climatic conditions.

Sampling Procedure

The sites were selected for sampling based on the soil survey maps, aerial photographs, management conditions etc. Once a sampling site was selected a flag was placed as a marker and the GPS coordinates are noted for the site. The fields selected were a minimum of 60 acres and a maximum of 160 acres and one sampling site represented 10 acres (Table 3).

Table 1. Sampling regions and geographic locations (Riopel, 2009)

Regions	Time Sampled	Latitude & Longitude	Major Land Resource Area
Montana	Jun 2008	48°46’ N, 104°34’ W	SSC
Central North Dakota[†]	Jun 2006	47°29’ N, 100°26’ W	55B
Central North Dakota[†]	Jun 2006		53B
North Eastern North Dakota	Jul 2008	48°06’ N, 98°50’ W	55A
North Central South Dakota	Jul 2007	45°27’ N, 98°27’ W	53B
Central South Dakota[†]	July & Aug 2007	44°20’ N, 98°12’ W	55C
Central South Dakota[†]	July & Aug 2007		53C
Western Minnesota	July & Oct 2007	46°48’ N, 95°50’ W	57
North Central Iowa & South Central Minnesota	July & Aug 2008	43°24’ N, 94°49’ W	103

† These regions were sampled along the boundary of two MLRA’s

Table 2. Average precipitation and temperature at the sampling regions (Riopel, 2009)

Regions	Avg Annual Precipitation (mm)	Avg Annual Temperature (°C)	Avg freeze free days
Montana	300-350	3-5°C	110-125
Central North Dakota[†]	400-500	4-7°C	120-140
Central North Dakota[†]	350-425	1-7°C	110-130
North-Eastern North Dakota	375-450	3-4°C	100-120
North-Central South Dakota	350-425	1-7°C	110-130
Central South Dakota[†]	450-525	7-9°C	130-155
Central South Dakota[†]	425-475	7-9°C	130-150
Western Minnesota	525-675	3-6°C	100-120
North-Central Iowa & South-Central Minnesota	625-850	6-9°C	130-160

† These regions were sampled along the boundary of two MLRA’s

Table 3. Number of fields and acres sampled (Riopel, 2009)

Regions	# of fields sampled	# of sample Sites	# of sampled acres
Montana	11	129	1290
Central North Dakota	15	166	1660
North-Eastern North Dakota	11	103	1030
North-Central South Dakota	15	166	1660
Central South Dakota	19	211	2110
Western Minnesota	20	189	1890
North-Central Iowa & Southern Minnesota	18	199	1990

Five samples were collected in a circle within approximately five meters from the center flag. The first sample of the 5 samples was collected on the north of the flag and the remaining four samples were sampled at equal distances in a circular fashion moving clockwise around the flag. A 12 inches deep (30 cm) sample was collected using a stainless steel hood probe and the top 6 inches (0-15cm) of the sample and bottom 6 inches (15-30 cm) of the sample were combined and bagged separately. After soil samples were collected at each location, information on vegetation present within 10 meters of the center of flag, landscape position, slope, aspect, slope shape, GPS coordinates, and other factors were recorded in a field notebook.

The collected samples were dried, quenched and then soil carbon content was estimated by high temperature combustion (1000° C) using a Skalar Primacs™ carbon analyzer as described by Nelson and Sommers (1996). Inorganic carbon was determined by acid treatment to dissolve soil carbonates, CO_2 evolved and measured by the same instrument. Organic carbon levels were determined by subtracting inorganic carbon from the total carbon.

Classification of Factors

Initially a general linear model (GLM) was run to find significant differences between the levels within each factor, for classification of factors.

The seven classes of vegetation are

- Cool season grasses(C)
- Cool season grasses and Legumes (CL)
- Cool + Warm Season grass (CW)
- Cool + Warm Season grass + Legumes (CWL)
- Legumes (L)
- Warm season grasses (W)
- Warm season grasses + Legume (WL)

The six classes of slope are

- 0-3%
- 3-6%
- 6-9%
- 9-12%
- 12-15%
- 15-18%

and the nine classes of aspect are

- No Aspect
- North
- Northeast
- East

- Southeast
- South
- Southwest
- West
- Northwest

Grouping based on least square difference (LSD) was used to combine classes within each factor (Appendix, Table A1).

The seven classes of vegetation were combined into four classes

i. Cool season grasses (C)
ii. Cool season grasses and legumes (CL)
iii. Legumes (L), warm season grasses (W), and warm season grasses & legume(WL)
iv. Cool & Warm Season grass (CW), and cool season grass+ warm season grass + legumes (CWL).

Six classes of slope were combined into two classes,

i. Slope of 0-3% and
ii. Slope > 3%.

Nine classes of aspect were reduced to five classes,

i. No Aspect.
ii. North & Northeast (N+ NE)
iii. South & Southwest (S+ SW)
iv. East & Southeast (E+ SE) and
v. West & Northwest (W+ NW).

Hypothesis testing was conducted by using multiple regressions to find the significance of a factor followed by calculating the effect of the factor including its interaction effects. This analysis differed from other traditional approaches, i.e. ANOVA and stepwise regression because of the number of levels of each factor.

Statistical Analysis

Initially we checked for assumptions of regression, constant variance and normality of residuals. If the multiple regression analysis using 3 variables was found to be significant, regression analysis was performed for two variables at once to find the best model. If a reduced model was found to be non-significant, a full model was considered for further analysis. On the other hand, if the reduced model was found to be significant, a partial F- test was performed to find the better model. If the partial F-test was significant then this reduced model was the best model compared to the full model. In case the partial F-test was non-significant then the full model was the best model.

Level 1 Analysis

Full models were first tested for the significance of interaction effects against a reduced model with no interaction effect

$$y = a^{*}\text{ vegetation} + b^{*}\text{ Slope} + c^{*}\text{ Aspect} + d^{*}\text{ Slope}^{*}\text{Aspect} + \varepsilon + \text{constant, and}$$

$$y = a^{*}\text{ vegetation} + b^{*}\text{ Slope} + c^{*}\text{ Aspect} + e^{*}\text{ Slope}^{*}\text{Vegetation} + \varepsilon + \text{constant}$$

While vegetation included 3 dummy variables, slope included one dummy variable and aspect had 4 dummy variables. The model with Vegetation * Aspect was not tested due to insufficient data for some of the classes.

For the regions where the interaction effects were not significant, a full model,

$$y = a^{*}\ \text{vegetation} + b^{*}\ \text{Slope} + c^{*}\text{Aspect} + \varepsilon + \text{constant}$$

was tested for the significance against reduced models with one of the factors excluded.

Level 2 Analysis

Full models with only two factors at a time were tested for significance in each region independently against a reduced model with only one factor.

RESULTS AND DISCUSSION

Partial F-test - Level 1

Central South Dakota

The partial F-test p-values for the interaction terms in the full model were 0.67 and 0.78 for slopeXaspect and slopeXvegetation, respectively (Table 4). Since the p-values were > 0.05, this suggests that there are no significant interaction effects. The full model was further tested with all 3 variables against reduced models with only 2 variables. The variable "vegetation" is significant when the other 2 variables are present (p-value < 0.001). The variable slope was not significant with the other two variables in the model (p-value = 0.63). Likewise, the variable aspect was not significant with the other two variables in the model (p-value = 0.83).

North Central South Dakota

The partial F-test p-values for the full model with interaction terms were 0.94 and 0.89 for slopeXaspect and slopeXvegetation, respectively. Since the p-values were greater than 0.05, there were no significant interaction effects. We further tested the full model with all 3 variables against reduced models with only two variables. We found vegetation was significant with slope and aspect in the model (p-value < 0.0001). We also found slope was significant with vegetation and aspect in the model (p-value < 0.0001). Aspect was not significant with vegetation and slope already in the model (p-value = 0.47).

Montana

The partial F-test p-values for the full model with interaction terms were 0.16 and 0.77 for slopeXaspect and slopeXvegetation, respectively. Thus there were no significant interaction effects. The full model with all 3 variables was tested against reduced models

Table 4. P-values from a partial F-test. This is a level 1 test including all three variables – vegetation, slope and aspect

Regions	Vegetation	Slope	Aspect	Slope*Aspect	Slope*Vegetation
Central South Dakota	0.00***	0.63	0.83	0.67	0.78
North central South Dakota	0.00***	0.00***	0.47	0.94	0.89
Montana	0.00***	0.88	0.03*	0.16	0.77
Western Minnesota	0.06	0.17	0.00***	0.25	0.74
Iowa	0.53	0.15	0.21	0.16	0.22
Southern Minnesota	0.26	0.00***	0.04*	0.70	0.60
Central North Dakota	0.02*	0.74	0.12	0.99	0.28
North eastern North Dakota	NA	NA	NA	NA	NA

*$p < 0.05$, *** $p < 0.001$, NA – Full and Reduced Models are not significant. So no further testing was done

with only 2 variables. Vegetation was significant with slope and aspect in the model (p-value < 0.0001). Aspect was significant with slope and vegetation in the model (p-value = 0.03). Slope was not significant with aspect and vegetation in the model (p-value = 0.88).

Western Minnesota

The partial F-test p-values for the full model with interaction terms were 0.25 and 0.74 for slopeXaspect and slopeXvegetation, respectively and showed no significant interaction effects. The full model was further tested with all 3 variables against reduced models with only 2 variables. Aspect was significant with slope and vegetation in the model (p-value < 0.0001). Vegetation was marginally significant with slope and aspect in the model (p-value = 0.06). Slope was not significant with aspect and vegetation in the model (p-value = 0.17).

Iowa

The partial F-test p-values for the full model with interaction terms were 0.16 and 0.22 for slopeXaspect and slopeXvegetation, respectively. Since the p-values were greater than 0.05, this suggests that there are no significant interaction effects. The full model was further tested with all 3 variables against reduced models with only 2 variables. None of the variables were significant (Table 4) with the other two variables in the model.

Southern Minnesota

The partial F-test p-values for the full model with interaction terms were 0.70 and 0.60 for slopeXaspect and slopeXvegetation, respectively.So there were no significant interaction effects. The full model was further tested with all 3 variables against reduced models with only 2 variables. Slope was significant with vegetation and aspect in the model (p-value < 0.0001). Aspect was significant with vegetation and slope in the model (p-value = 0.04). Vegetation was not significant with slope and aspect in the model (p-value = 0.26).

Central North Dakota

The partial F-test p-values for the full model with interaction terms were 0.99 and 0.28 for slopeXaspect and slopeXvegetation, respectively. Since the p-values were greater than 0.05, we conclude that there are no significant interaction effects. The full model was further tested with all 3 variables against reduced models with only 2 variables. Vegetation was significant with slope and aspect in model (p-value = 0.02). Neither slope nor aspect were significant with the other two variables in the model (p-values = 0.74 and 0.12 respectively).

North Eastern North Dakota

For this region we did not conduct partial F-tests, as the full model with all 3 variables and interaction terms were not significant when conducting an overall F-test. Therefore, there was not sufficient evidence to indicate vegetation, slope, or aspect is helping to explain the variability of carbon content of soil in this region.

Partial F-test - Level 2

Central South Dakota

Since the models with only 2 variables were significant (vegetation and slope; vegetation and aspect), we further conducted partial F-tests on models with both variables compared to models with just one variable. Vegetation was found to be significant with slope already in the model (p-value < 0.0001; Table 5). It was also found to be significant with aspect already in the model (p-value < 0.0001). Neither slope nor aspect were found to be significant with vegetation already in the model (p-values = 0.44 and 0.76, respectively).

Table 5. P-values from a partial F-test. This is a level 2 test that includes only two variables

Regions	Vegetation	Slope	Aspect
Central South Dakota	0.00***	0.44	
	0.00***		0.76
North Central South Dakota	0.00***	0.00***	
Montana	0.00***		0.02*
Western Minnesota	0.03*		0.00***
Iowa		0.06	0.28
	0.32		0.04*
	0.70	0.01*	
Southern Minnesota		0.00***	0.02*
Central North Dakota	0.00***		0.10
	NA	NA	

*p<0.05, *** p<0.001, NA – Full and Reduced Models are not significant.

North Central South Dakota

The model with only the 2 variables, vegetation and slope was significant. We further tested this model against models containing only one of the variables. Vegetation was found to be significant with slope in the model (p-value < 0.0001). Slope was found to be significant with vegetation already in the model (p-value < 0.0001).

Montana

The model with only the two variables, vegetation and aspect was significant. We further tested this model against models containing only one of the variables. Vegetation was found to be significant with aspect in the model (p-value < 0.0001). Aspect was found to be significant with vegetation already in the model (p-value = 0.02).

Western Minnesota

The model with only the two variables, vegetation and aspect was significant. We further tested this model against models containing only one of the variables. Vegetation was found to be significant with aspect in the model (p-value = 0.03). Aspect was found to be significant with vegetation already in the model (p-value < 0.0001).

Iowa

In level 1 testing, none of the variables were found to be significant with the other two variables in the model. We next considered testing for the significance of each variable given that one of the other two variables was already in the model. Aspect was found to be significant when vegetation was in the model (p-value = 0.04). Slope was found to be significant or marginally significant when either vegetation or aspect was in the model (p-values = 0.01 and 0.06 respectively). Nothing else was found to be significant.

Southern Minnesota

The model with only the two variables, slope and aspect was significant. We further tested this model against models containing only one of the variables. Slope was found to be significant with aspect in the model (p-value < 0.0001). Aspect was found to be significant with slope already in the model (p-value = 0.02).

Central North Dakota

Since the models with only 2 variables were significant (vegetation and aspect), we further conducted partial F-tests on models with both variables compared to models with just one variable. Vegetation was found to be significant with aspect already in the model (p-value < 0.0001). Aspect was not significant with vegetation already in the model (p-value = 0.10). Since the model with the two variables, vegetation and slope was not significant, no further tests were conducted.

Significant Parameter Estimates

Central South Dakota

Vegetation was found to be significant in predicting soil organic carbon content in Central South Dakota. There were four levels of vegetation with the base level being cool season grasses. Three indicator variables were placed in the model comparing the other vegetation types to cool season grasses. Since the parameter estimates of the indicator variables were all negative (Table 6), this indicates cool season grasses retain more carbon content in soil than other grasses.

North Central South Dakota

Vegetation and Slope are significant variables. The variable estimates are negative for two indicator variables. This indicates cool season grasses retain more carbon content in soil than these grasses. One level of vegetation has a small positive estimate. This means that warm, warm legumes and legumes is estimated to retain as much as may be little more carbon content as the base level. There were two levels of slope with 0- 3% being the reference. Negative coefficient associated with the indicator variable for a slope greater than 3% indicates that a slope less than 0-3% retain more carbon content in soil.

Montana

Vegetation and aspect were found to be significant in predicting soil organic carbon content in Montana. There were four levels of vegetation with the base level being cool season grasses. Three indicator variables were placed in the model comparing the other vegetation types to cool season grasses. Since the variable estimates of the indicator variables were all negative (Table 6), this indicates cool season grasses retain more carbon content in soil than other grasses. There are five levels of aspect with the base level being no aspect. Four indicator variables were placed in the model comparing the other aspects to no aspect. Since the parameter estimates of the indicator variables were all negative, this indicates no aspect retains more carbon content in soil than other aspects.

Western Minnesota

Vegetation and aspect were found to be significant in predicting soil organic carbon content in western Minnesota. There were four levels of vegetation with the base level being cool season grasses. Three indicator variables were placed in the model comparing the other vegetation types to cool season grasses. Since the parameter estimates of the indicator variables were all negative (Table 6), this indicates cool season grasses retain more carbon content in soil than other grasses. There are five levels of aspect with the base level being no aspect. Four indicator variables were placed in the model comparing the other aspects to no aspect. The estimated coefficient associated with one of the indicator variables is a small positive number, which indicates that the North/North east aspect retains as much as carbon content in soil as no aspect. The variable estimates of the other three indicator variables were negative. This indicates no aspect retains more carbon content in soil than these aspects.

Iowa

Slope and aspect were found to be significant in predicting soil organic carbon content in Iowa. There were two levels of slope with 0- 3% being the reference. A negative

Table 6. Parameter estimates of factors that are significant

Regions	Vegetation			Slope	Aspect			
	Veg2	Veg3	Veg4		Asp2	Asp3	Asp4	Asp5
Central South Dakota	-1.38	-1.47	-1.4					
North Central South Dakota	-1.48	-1.37	0.15	-0.49				
Montana	-1.58	-1.1	-2.77		-3.43	-2.49	-2.65	-3.39
Western Minnesota	-1.7	-0.45	-0.49		0.18	-0.67	-0.5	-1.03
Iowa				-1.11	-0.25	-1.77	-1.16	-1.58
Southern Minnesota				-1.46	-3.19	-2.5	-2.78	-2.17
Central North Dakota	-0.78	-0.58	NS					
North Eastern North Dakota	NA	NA	NA	NA	NA	NA	NA	NA

NS-Variables not significant at level 2 of partial F-test, NA- Not Applicable (Full model is not significant) value at a slope > 3% indicates that slope < 0-3% retains more carbon content in soil than the other level of slope. There are five levels of aspect with the base level being no aspect. Four indicator variables were placed in the model comparing the other aspect levels to no aspect. Since the variable estimates of the indicator variables were all negative, this indicates no aspect retains more carbon content in soil than the other aspects.

Southern Minnesota

Slope and aspect were found to be significant in predicting soil organic carbon content in southern Minnesota. There were two levels of slope with 0- 3% being the reference. A negative value of the coefficient associated with the indicator variable for a slope greater than 3% indicates that a slope of 0-3% retains more carbon content in soil. There are five levels of aspect with the base level being no aspect. Four indicator variables were placed in the model comparing the other aspects to no aspect. Since the variable estimates of the indicator variables were all negative, this indicates no aspect retains more carbon content in soil than other aspects.

Central North Dakota

Vegetation was found to be significant in predicting soil organic carbon content in Central North Dakota. There were four levels of vegetation with the base level being cool season grasses. Three indicator variables were placed in the model comparing the other vegetation types to cool season grasses. Since the variable estimates of the indicator variables were all negative, this indicates cool season grasses retains more carbon content in soil than other grasses.

CONCLUSIONS

In general, no interactions were observed between slope and aspect or slope and vegetation. The relationship between vegetation type and SOC was significant in most of the regions. The relationship between slope and SOC or aspect and SOC were significant in fewer regions.

- **Vegetation**: Overall, we found a higher SOC is associated with cool season grasses. This is indicated by the significance of the vegetation variable and the negative coefficients associated with the indicator variables when cool season grasses are used as a reference.
- **Slope:** In the three regions where slope was significant, the reference 0-3% slope showed higher SOC levels than when the slope was > 3%".
- **Aspect:** In the four regions where aspect was significant, it was generally found that higher SOC levels were associated with no aspect compared to other aspects. This is indicated by the negative estimated coefficients associated with aspect levels when no aspect was used as a reference.

From this data analyses, we conclude that the greatest SOC accumulations occur under cool-season grasses on slopes of <3% that tend to have no defined aspect. Future research could include studying more factors that affect SOC levels.

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APPENDIX

Table A1. P-values obtained from GLM when all levels were considered

Regions	Vegetation	Slope	Aspect
Central South Dakota	0.00	0.87	0.45
North Central South Dakota	0.00	0.21	0.89
Montana	0.01	0.86	0.05
Western Minnesota	0.14	0.27	0.01
Iowa	0.16	0.04	0.07
Southern Minnesota	0.03	0.01	0.45
North Eastern North Dakota	0.28	0.98	0.20
Overall	0.00	0.00	0.01

Table A2. Descriptive statistics for soil organic carbon values in the 0-30 cm depth for each region.

Regions	Sample size	Mean	Standard Deviation	Minimum	Maximum
Central South Dakota	209	7.43	1.51	3.37	12.27
North Central South Dakota	159	7.78	1.68	4.57	15.17
Montana	128	5.58	2.08	1.34	11.57
Western Minnesota	188	9.10	1.84	3.39	13.94
Iowa	96	9.35	2.06	4.38	13.51
Southern Minnesota	96	8.94	2.09	3.73	14.23
Central North Dakota	158	7.19	2.03	2.42	12.57
North Eastern North Dakota	98	6.34	2.34	2.16	11.28