

INHERENT SOIL PROPERTIES, FARM MANAGEMENT,  
AND BIOLOGICAL SOIL HEALTH IN ORGANIC GRAIN SYSTEMS

By

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## **CHAPTER 1: INHERENT SOIL PROPERTIES AND BIOLOGICAL SOIL HEALTH**

### **ABSTRACT**

Besides soil texture and pH, inherent soil properties have been explored minimally as determinants of soil health as they are unmanageable constraints under relevant timescales. However, identifying inherent soil properties that are the most influential to soil health is necessary for developing regionally specific critical values for assessments. Publicly accessible soil information from the Natural Resources Conservation Service (NRCS) Web Soil Survey is a relatively untapped resource for determining potential co-variables for soil health assessment. We used soils of similar texture collected from 124 fields across 16 certified organic grain farms in the Driftless Region of Wisconsin to analyze for effects of NRCS soil properties on soil health. The main pools of carbon and nitrogen (soil organic matter (SOM), total organic carbon (TOC), and total nitrogen (TN)), along with indicators of biological soil health (permanganate oxidizable carbon (POXC), mineralizable carbon (minC), potentially mineralizable nitrogen (PMN), and autoclaved-citrate extractable protein (ACE)) were measured. Simple linear regression and analysis of variance were utilized to determine effects of soil properties and indices on biological soil health indicators. The most influential NRCS soil properties and indices differed by the elemental cycle and process associated with the indicator. Differences in soil taxonomy, bulk density, soil surface sealing, whole soil erodibility, the fragile soil index, and organic matter depletion affected multiple indicators. Soil water properties were solely influential for TOC with wetter soils containing higher SOM and TOC values. These properties deserve further exploration as potential co-variables within

larger regional and national datasets for developing soil health assessments. Inclusion of identified inherent soil properties as co-variates is critical for comparing soils among relevant peers to properly inform management decisions.

## **INTRODUCTION**

Soil performs a multitude of functions in agricultural systems that affect the productivity of our lands for food, feed and fiber as well as their capacity to mitigate climate change and environmental degradation. An increase in public awareness of soil's potential to provision services for the betterment of humanity has heightened interest in soil health. According to the United States Department of Agriculture-Natural Resources Conservation Service (USDA-NRCS), soil health is "the capacity of the soil to function as a vital living ecosystem that supports plants, animals, and humans" (Stott, 2019). This all-encompassing definition allows soil health assessment to adjust to different societal contexts based on societal priorities, values, and needs. For example, the soil functions required for engineering purposes is much different than those for agricultural production. It is important to incorporate these contexts in soil health assessment for optimal decision-making.

Primarily, three components of soil health are measured to assess soil functioning: soil biological, physical, and chemical properties. Soil chemical and physical properties have been studied much longer than biological properties; their measurements are well developed while researchers are still debating on how to measure biological properties. Recently, indirect measures of soil functions, known as indicators, are receiving strong

support due to their high application potential outside of research. Indicators being developed for soil health assessment based on their ability to inform land management for farmers, natural area managers, among others. Generally, a strong indicator is cost-effective, easy to use, sensitive to management, and can timely deliver information to implement best management practices relevant to that soil (Morrow et al., 2016; Stott, 2019). A combination of indicators is necessary to evaluate biological soil health. For agricultural production, major attention to the biological processes involved in carbon (C) and nitrogen (N) cycling is needed for assessing nutrient management. Different soil health measurement packages have been proposed by the Soil Health Institute, USDA's Soil Management Assessment Framework (SMAF), and Cornell's Comprehensive Assessment of Soil Health (CASH) (Schindelbeck et al., 2016; Stott, 2019). Determination of the most informative indicators for decision-making is key to the development of benchmarks for regional and national soil health assessments. In 2019, NRCS released indicator recommendations and standardized protocols for measuring C and N cycling functions. Our study utilized several indicators that evaluate C and N cycling components vital to agricultural production as well as the provision of crucial ecosystem services such as the protection of water quality through nutrient retention.

For evaluating organic matter cycling and C sequestration, measurement of soil total organic carbon (TOC) via dry combustion is recommended and can be modified for use on alkaline soils (Sherrod et al., 2002; Harris et al., 2001). Soil TOC content provides information on a multitude of soil functions such as water-holding capacity and nutrient

retention, but it does not provide information on the bioavailability of C and N within the organic matter (Stott, 2019). In addition, the effect of management on TOC content is undetectable for three or more years, and its rate of change is highly affected by climate (Sikora et al., 1996). Thus, it is necessary to include other measures that are more sensitive to management and can evaluate the quality and availability of organic matter constituents as it relates to important biological functions.

Bioavailability of C and N can be evaluated through chemical extraction or biological incubation. Permanganate oxidizable C (POXC) uses 0.02 M  $\text{KMnO}_4$  to chemically oxidize the most readily available fraction of soil TOC. Since soil microorganisms use oxidative enzymes to decompose the organic matter available to them, chemical oxidation mimics biological oxidation (Loginow et al., 1987). This notion is well-supported as POXC is positively correlated with many biological properties such as TOC (Bongiorno et al., 2019; Morrow et al., 2016; Culman et al., 2012; Plaza-Bonilla et al., 2014; Weil et al., 2003), particulate organic C (Bongiorno et al., 2019; Culman et al., 2012), microbial biomass C (Bongiorno et al., 2019; Culman et al., 2012), soil respiration (Bongiorno et al., 2019; Weil et al., 2003), soluble carbohydrates (Weil et al., 2003) and substrate-induced respiration (Weil et al., 2003). Despite its connection to biological activity and properties, Romero et al. (2018) determined that C outside of the readily bioavailable C pool is detected with POXC. In fact, POXC is more closely associated with a highly processed fraction of TOC as well as organic matter building and stabilization (Culman et al., 2012; Hurisso et al., 2016). Regardless of the evolving interpretation on POXC, it is considered a useful tool for informing soil organic matter

management and fulfills many of the criteria for a robust soil health indicator (Stott, 2019; Morrow et al., 2016; Fine et al., 2017).

Potentially mineralizable C (minC) is an incubation method that measures soil respiration via CO<sub>2</sub> evolution after rehydrating air-dried soil to ~50% water-filled pore space (WFPS). Maximum minC values are obtained when WFPS is between 53 to 66% (Franzluebbers, 1999). As a soil health indicator, it represents general microbial activity but is associated with nutrient bioavailability (Hurisso et al. 2016). Since short-term C mineralization is more representative of growing conditions and avoids substrate depletion, it is the preferred method over long-term C mineralization (Stott, 2019). Schindelbeck et al. (2016) recommends a 4-day incubation to minimize variation between replicates. However, 24-hr incubations have been shown to be as indicative of microbial activity as 24-day incubations and are more applicable to high-throughput commercial lab settings (Haney et al., 2004).

Potentially mineralizable N (PMN) is another incubation method that represents the capacity of the soil microbiome to provide plant-available N through mineralization. It can be measured under aerobic or anaerobic conditions. Anaerobic conditions simplify PMN measurement by preventing nitrification so that ammonium is the only mineralization product as well as circumventing management of soil water content during incubations (Drinkwater et al., 1996). Potentially mineralizable N is a well-established measurement but has limited potential for high-throughput commercial labs due to the week-long or longer

duration of its incubations. However, Hurisso et al. (2018b) suggested a chemical extraction for soil protein as an indicator of bioavailable N.

Autoclaved-citrate extractable protein (ACE) measures soil protein through adaptation of a chemical extraction for glomalin-related soil protein. Rosier et al. (2006) and Hurisso et al. (2018b) demonstrated that this method captures more than arbuscular mycorrhizal fungi protein (glomalin), but instead captures a range of proteins. Since protein is the largest organic N fraction in soil and depolymerization of protein to amino acids is the rate-limiting step for N mineralization, it is a viable proxy for bioavailable N (Nannipieri & Eldor, 2009; Schimel & Bennet, 2004; Hurisso et al., 2018b).

Research has utilized long-term trials to develop an understanding of soil health threshold values and the effects of management on these indicators across geospatial regions and edaphic conditions (e.g. Culman et al., 2012; Hurisso et al., 2016; Diederich et al., 2019; Norris et al., 2020). However, the effect of soil properties beyond pH and texture have been given less consideration regardless of public access to NRCS soil data for greater than 95% of U.S. counties (Soil Survey Staff, n.d.). Despite accounting for soil textural differences in soil health assessment, there are regional differences in mean indicator values (Moebius-Clune et al., 2016). Subsequently, soil health scoring functions were adjusted to convey these differences. Identifying unaccounted sources of variation in regional soil health values may improve the efficacy of soil health assessment through improving or adding co-variates.

As part of its mission to “produce and deliver scientifically based soil information to help society understand, value, and wisely manage global resources,” the NRCS Web Soil Survey (WSS) provides publicly accessible information for the spatial distribution of U.S. soils, and their properties, suitabilities and limitations (USDA-NRCS, 2019). Field observations and mapping are utilized to identify patterns in soil distributions to generate soil maps. Distinct areas defined by their soils in a survey area are regarded as map units and have associated soil information. Due to the soil survey mapping scale, soil property and index information of map units are not site-specific, but are instead representative of typical soil profiles and laboratory measurements of map units. The scale of mapping along with map unit design considerations are the most likely causes of error on soil maps (Soil Science Division Staff, 2017). Although the soil information is not a direct representation of site conditions, exploration of NRCS soil properties and indices effects on soil health indicator values may discover novel connections or influential factors for potential co-variates in soil health assessment.

Not all soils have the same intrinsic potential for soil health. The most influential soil properties can be used to differentiate soils to ensure comparison among their peers. Soil health assessments that include these factors as co-variates would provide better interpretation of soil health status and better inform management decisions. The need for co-variates to account for inherent differences in soil health may depend on whether an assessment is national or regional. Understanding underlying soil health variation within a region would provide first-indications if co-variates are essential for regional-based soil

health assessments. Overall, this research aims to identify key determinants of soil health for use as potential co-variables in assessment through the evaluation of inherent soil property effects. The specific objectives of this study are to: 1) evaluate the relationship between NRCS soil information with soil health indicators (SOM, TOC, TN, POXC, minC, PMN and ACE) and 2) to evaluate the relationships among soil health indicators.

Inherent soil properties disproportionately affect soil function as they are derived as a result of soil formation. We expect dominant soil properties, such as pH, texture, drainage class, and clay content, to effect soil health as they have greater effects on the dynamic properties of soil (temperature and moisture content) and on the soil's capacity to store organic matter (Brady and Weil, 2010). Since soils are classified by primarily their dominant soil properties (Soil Survey Staff, 1999), it is expected that soils with more similar classifications (i.e. orders, suborders, great groups, subgroups, and soil series) will have more similar soil health values than dissimilar classifications. In addition, we hypothesize that TOC, POXC and minC will have positive relationships to each other, and that ACE and PMN will have a positive relationship.

## **MATERIALS AND METHODS**

### **Field Descriptions and Soil Sampling**

Soil samples were collected from 124 fields of 16 certified organic grain farms in the southwest Driftless Region of Wisconsin (Figure 1.1). Farms were selected from a list of Wisconsin farms that are members of the organic cooperative Organic Valley based on the



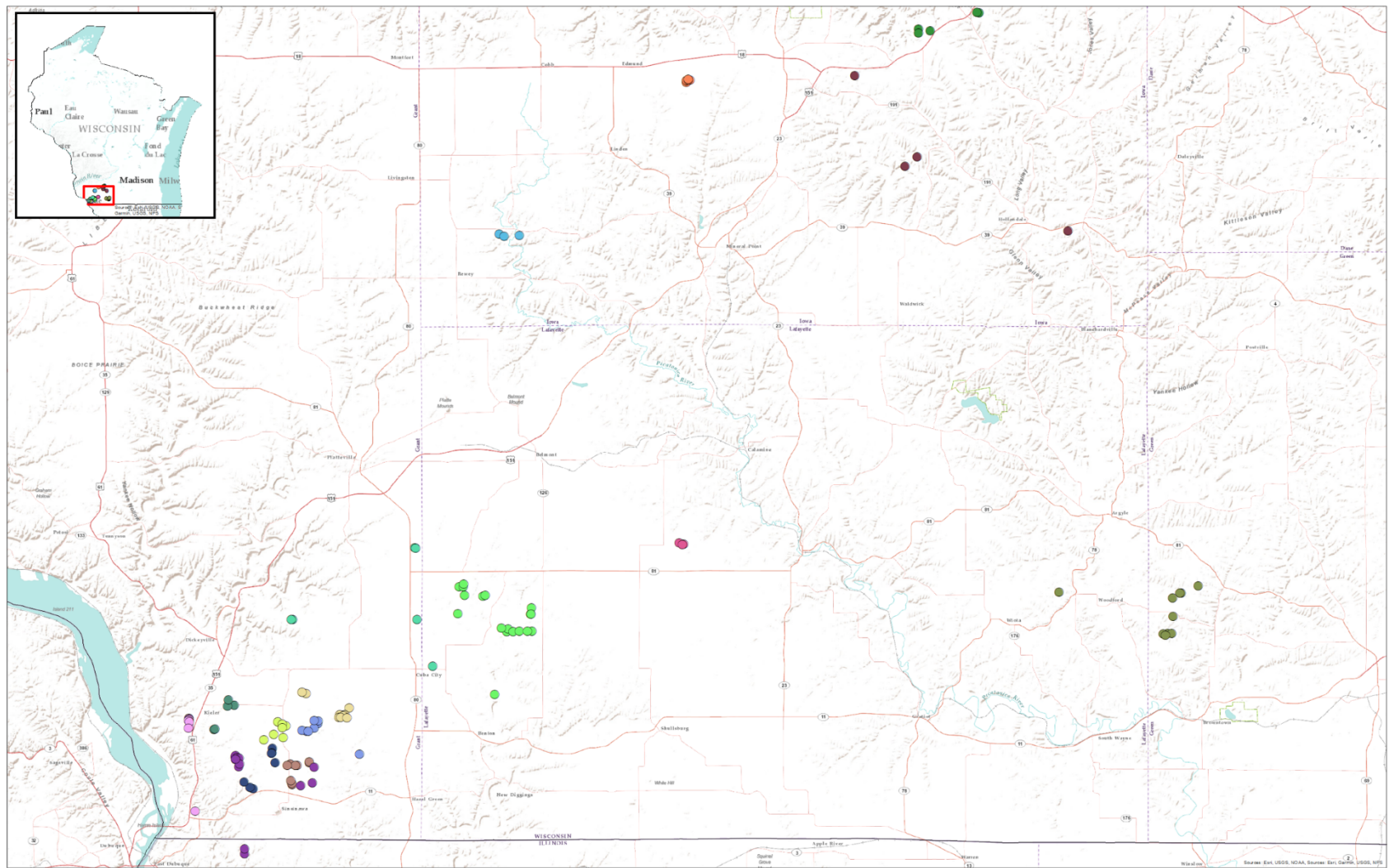


Figure 1.1. Map of 2018 and 2019 field sites. Each color represents a field managed by the same farmer.

following criteria: 1) grain production was a major part of their farm operations, 2) farms were based in the Driftless Region of Wisconsin, and 3) willingness to participate in this research. Locations were limited to the Driftless Region because of the high density of organic grain farms in that region and to minimize the variation in inherent soil properties and climate. Fields were located in the Wisconsin counties of Grant (n = 73), Lafayette (n = 23), Iowa (n = 17), and Green (n = 9), and the Illinois county of Jo Daviess (n = 2).

Soil samples were collected between 18 May to 2 July in 2018 (n = 59) and 22 April to 13 May in 2019 (n = 65), prior to planting of the next crop. Soil samples were solely collected in fields where the previous year's crop was corn. For each field, ten soil cores were obtained in a circular array with a radius of approximately 2 meters. GPS coordinates noted the center of the sampling circle. The soil cores were taken from a depth of 0 to 15 cm by a probe with an internal diameter of 2.0 cm, and then were composited and well-mixed. Upon immediate return from field locations, composited samples were air-dried using a force-air drier at 32 °C for 3 days, then ground to pass through a 1-mm sieve and stored at room temperature until analysis.

### **Soil Analysis**

According to procedures set and standardized by the North Central Regional Extension & Research Activity (2015), air-dried soils were analyzed for soil pH, reserve acidity, and SOM. Soil pH was measured using a 1:1 ratio of soil to deionized water. For soils with a pH  $\leq$  6.5, reserve acidity was measured using Sikora buffer (Sikora, 2006). Soil was

heated for 2 hours at 360 °C and the mass lost on ignition was used to calculate SOM content.

Soil total C and N content were obtained through dry combustion of 8–10 mg of soil in a tin capsule, where the soil was previously ground to a flour-like consistency. Total C and N were measured with the Flash EA 1112CN Automatic Elemental Analyzer (Thermo Finnigan, Milan, Italy). If the coefficient of variation (CV) of a soil sample's total N exceeded 20% and N was between 0.1 to 0.2 %, the mass of the soil sample was raised to 24-25 mg in order to improve detection. Presence of soil inorganic C was determined visually from effervescence using 5% HCl. Soils without inorganic C were assumed to have TOC values equivalent to measured total C values. For soils testing positive for inorganic C, TOC was obtained using HCl fumigation to remove inorganic C prior to dry combustion (Harris et al. 2001). For both C and N cycling, a chemical extraction and incubation method were utilized as indicators for important soil processes of each element.

Permanganate oxidizable C was measured according to Culman et al. (2012), a modification of Weil et al. (2003). In triplicate,  $2.5 \pm 0.01$  g of soil were combined in a 50 mL polypropylene centrifuge tube with 18 mL of deionized water and 2 mL of 0.2 M  $\text{KMnO}_4$ . Upon addition, samples were immediately capped and shaken at 240 rpm for 2 minutes, then uncapped and placed in a dark space to settle for 10 minutes. The supernatant was diluted 100-fold by transferring 0.5 mL of supernatant to 49.5 mL of deionized water. In triplicate, 275  $\mu\text{L}$  of the diluted supernatant was pipetted into a 96-cell plate, along with three sets of a positive control (silt loam soil with established POXC values), deionized water

blank, and standard curve (5, 10, 15 and 20 mM KMnO<sub>4</sub>). A spectrophotometer (Omega, BMG LABTECH GmbH, Ortenberg, Germany) measured the absorbance of each cell at wavelength 550 nm. POXC was calculated using the following equation:

$$\text{POXC (mg C kg}^{-1} \text{ soil)} = [0.02 \text{ mol L}^{-1} - (a + b(\text{Abs}))] * (9000 \text{ mg C mol}^{-1}) \\ \times (0.02 \text{ L solution} \times \text{Wt}^{-1}) \quad [\text{Eq. 1}]$$

where 0.02 mol L<sup>-1</sup> is the initial concentration of KMnO<sub>4</sub> solution; *a* is the intercept of the standard curve; *b* is the slope of the standard curve; Abs is the absorbance of the unknown soil sample; 9000 mg is the amount of C oxidized by 1 mol of MnO<sub>4</sub><sup>-</sup>; 0.02 L is the volume of KMnO<sub>4</sub> solution reacted with soil; and Wt is the kg of air-dried soil utilized in the reaction.

Mineralizable C was obtained using a modification of the procedures described by Franzluebbers et al. (2000) and Haney et al. (2004). In triplicate, 10 ± 0.01 g of soil was placed into a 59-mL plastic dish and carefully transferred to a 0.932-L glass jar. In a fume hood, deionized water was pipetted onto each soil sample to achieve 60% water-filled pore space (WFPS). A WFPS of 60% was selected as it is within the range of moisture content that maximizes microbial respiration while mitigating the risk of observing the steep decline that occurs below 50% WFPS (Franzluebbers et al., 1999). Water-filled pore space was calculated using an estimated bulk density as well as an assumed particle density of 2.65 g mL<sup>-1</sup>, the density of quartz (Eq.2).

$$60\% \text{ WFPS} = (1 - (\text{bulk density} / 2.65 \text{ g mL}^{-1})) \times (0.6) \times (10 \text{ g}) \quad [\text{Eq. 2}]$$

Upon wetting, the jars were capped tightly and incubated at 25 °C for 24 hours. An infrared gas analyzer (LiCor Li-820, LI-COR Biosciences, Lincoln, NE, USA) with a flow-through reactor was used to measure the concentration of CO<sub>2</sub> produced in each jar. Incubated along the soil samples was an empty jar containing solely a plastic dish, and another jar containing a silt loam soil with established minC value; the empty jar was used to blank the analyzer to control for the atmospheric CO<sub>2</sub> concentrations within the fume hood while the known minC soil was used as a positive control. Mineralizable C was calculated using the following equation:

$$\text{minC} = [(C_v \times M \times P) / (R \times T) \times (H/Wt)] \times (0.001/0.0001) \quad [\text{Eq. 3}]$$

where  $C_v$  is the average concentration of CO<sub>2</sub> in ppm;  $M$  is the molecular weight of C (12 µg/µmol);  $P$  is the barometric pressure (1 atm);  $R$  is universal gas constant (0.0820575 L · atm · °K · mole);  $T$  is the incubation temp in °K (298.15);  $H$  is the volume of the incubation chamber (0.932 L); and  $Wt$  is the mass of the soil used in the incubation (10 g). The volume of the connecting lines attached to the Li-Cor Li-820 was calculated and not included in the total volume of the incubation chamber as its volume was negligible.

Potentially mineralizable N (PMN) was assessed using a 7-day, 40 °C anaerobic incubation according to the methods of Drinkwater et al. (1996). Initial soil ammonium-N concentration was determined from the extraction of  $5 \pm 0.01$  g of soil with 40 mL of 2 M KCl while shaking for 1 hour at 240 rpm. Following extraction, initial samples were centrifuged at 1320 rpm for 5 minutes and supernatant was filtered through 2.7 µm filter paper into 30 mL scintillation vials and immediately frozen and stored at -20 °C. In duplicate,

7-day ammonium-N was determined from the incubation of  $5 \pm 0.01$  g of soil with 10 mL of deionized water at 40 °C. Following the incubation, samples were immediately extracted by adding 30 mL of 2.5 M KCl and shaking for 1 hour at 240 rpm. Supernatant was obtained, filtered, and stored to identical methods for initial ammonium-N samples. N mineralization was determined from ammonium-N concentration of thawed extracts measured using a Lachat Flow Injection System. Potentially mineralizable N was calculated as the difference in average ammonium-N concentration between the two incubated samples and initial sample.

Autoclaved-citrate extractable protein (ACE) was measured according to the procedures of Hurisso et al. (2018b). In triplicate,  $3 \pm 0.01$  g of soil was combined with 24 mL of 0.02 M sodium citrate pH 7 solution in a glass screw-top tube, and then shaken at 180 rpm for 5 minutes. Following shaking, tubes were autoclaved at 121 °C for 30 minutes with the caps loosened and left atop of tubes. Tubes were then allowed to cool before resuspending via shaking for 3 minutes at 180 rpm. Into a 2.2 mL microcentrifuge tube, 1.75 mL of extract was transferred and then centrifuged at 10,000x g for 3 minutes. Protein content of the supernatant was quantified with the bicinchoninic acid assay using bovine serum albumin (BSA) as a standard (0, 125, 250, 500, 750, 1500, and 2000  $\mu\text{g mL}^{-1}$ ).

### **Dataset Construction & Statistical Analysis**

Publicly available soil property and indices data for each field was obtained and aggregated from the NRCS Soil Survey Geographic Database (SSURGO) via the Web Soil Survey (WSS) (Soil Survey Staff, n.d.). The soil map was used to determine the soil series and

its map unit code for each sampling location based on soil sample georeferenced coordinates. Using the map unit code in Soil Data Explorer, soil properties and qualities as well as soil suitabilities and limitations data was collected with cropland as a land use filter to avoid incorporating data for other land uses. Depth of A horizon and landform position information were added from the map unit description for each soil series. Tables 1.1 and 1.2 are a truncated list of the data collected including only soil data explorer subsections pertinent to crop production, soil health, soil classification, and inherent soil properties. From this list, soil properties and indices were selected for analysis based on the following criteria: 1) they are related to grain and forage systems, 2) they are developed for Wisconsin or apply to this region, and 3) they do not capture information for climate and/or engineering purposes as these are not relevant to the localized area and land use considerations. The remaining soil properties and indices were aggregated to construct a dataset to evaluate the relationships between NRCS soil information and soil health indicators (Tables 1.3 - 1.5).

Descriptive statistics were used to explore the distribution of NRCS and response variables (SOM, TOC, TN, POXC, minC, PMN, and ACE). The *describe* function from the *Psych* package in R was used to perform univariate statistics, such as mean, median, skewness, etc., on both continuous NRCS and response variables. Categorical NRCS variable distributions were evaluated from the number of observations and percent of total observations per category. Categories containing less than three field observations within a categorical variable were excluded from analysis because meaningful standard deviations

cannot be computed for  $n < 3$ . To exclude NRCS variables from analysis that did not provide sufficient information due to lack of or excess variation, NRCS soil properties and indices were removed from the dataset if the following criteria were met: 1)  $<80\%$  of fields had a value reported, 2)  $>90\%$  of fields belong to a single category, and/or 3) categorical variables contained greater than ten categories that were highly unbalanced. NRCS variables that were computed by applying a factor to another NRCS variable were excluded since they would supply redundant information and exhibit high collinearity.

Of the remaining NRCS variables, the effect of individual inherent soil properties and indices on soil health indicators was assessed in R using analysis of variance (ANOVA) with a Fisher's least significant difference (LSD) test for categorical explanatory variables and simple linear regression for continuous explanatory variables ( $\alpha = 0.05$ ). The *aov* and *summary* functions were used for ANOVA while *lm* and *summary* functions were used for simple linear regression. Simple linear regression assumptions of linearity, constant variance, and normality were evaluated using residuals versus fitted values plots and Q-Q plots.

## RESULTS AND DISCUSSION

### Relationships Between Indicators

Soil health indicator values were within the range of those recorded across diverse cropping systems and geographic regions of the United States (e.g. Drinkwater et al., 1995; Marinari et al., 2006; Culman et al., 2012; Hurisso et al., 2016; Fine et al., 2017). Mean indicator



Table 1.1. Soil information from the suitabilities and limitations, and map unit description sections of the NRCS WSS.

Section	Subsection	Soil Information
Suitabilities and Limitations	Land Classifications	Conservation Tree and Shrub Group
		Farmland Classification
		Hydric Rating by Map Unit
		Irrigated Capability Class
		Irrigated Capability Subclass
		National Commodity Crop Productivity Index (NCCPI)
		NCCPI (Corn)
		NCCPI (Small Grain)
		NCCPI (Soybean)
		Non-Irrigated Capability Class
		Non-Irrigated Capability Subclass
		Order of Soil Survey
		Soil Moisture Class
		Soil Moisture Subclass
		Soil Taxonomy Classification
		Soil Temperature Regime
	Soil Health	Agricultural Organic Soil Subsidence
		Farm and Garden Composting Facility - Surface
		Fragile Soil Index
		Organic Matter Depletion
		Soil Surface Sealing
		Soil Susceptibility to Compaction
		Suitability for Aerobic Soil Organisms
		Surface Salt Concentration
	Vegetative Productivity	American Wine Grape Varieties Site Desirability
		Wisconsin Commodity Crop Productivity Index
		Crop Productivity Index
		Hybrid Wine Grape Varieties Site Desirability
		Iowa Corn Suitability Rating (CSR2)
		Minnesota Crop Productivity Index
		Vinifera Wine Grape Site Desirability
		Yields of Irrigated Crops
		Yields of Non-Irrigated Crops
Map Unit Description	Typical Profile	Depth of A Horizon
	Setting	Slope Position (2D Landform Position)
		3D Landform Position

NRCS = Natural Resources Conservation Service; WSS = Web Soil Survey

Table 1.2. Soil information from the soil properties and qualities section of the NRCS WSS.

Subsection	Soil Information
Chemical Properties	Calcium Carbonate
	Cation-Exchange Capacity (CEC)
	Effective Cation-Exchange Capacity
	Electrical Conductivity (EC)
	Gypsum
	pH
	Sodium Adsorption Ratio (SAR)
Physical Properties	Available Water Storage
	Available Water Supply
	Bulk Density
	Linear Extensibility
	Liquid Limit
	Organic Matter
	Percent Clay
	Percent Sand
	Percent Silt
	Plasticity Index
	Saturated Hydraulic Conductivity
	Saturated Hydraulic Conductivity Standard Classes
	Surface Texture
	Water Content at 1500 and 3 kPa
Water Features	Depth to Water Table
	Flooding Frequency Class
	Ponding Frequency Class

Subsection	Soil Information
Erosion Factors	K Factor, Rock Free (Erodibility Factor)
	K Factor, Whole Soil (Erodibility Factor)
	T Factor (Tolerable Soil Loss)
	Wind Erodibility Group
	Wind Erodibility Index
Soil Health Properties	Available Water Capacity (AWC)
	Bulk Density
	Sodium Adsorption Ratio (SAR)
	pH
Soil Quality and Features	Surface Texture
	AASHTO Group Classifications
	AASHTO Group Index
	Depth to a Selected Soil Restrictive Layer
	Depth to Any Soil Restrictive Layer
	Drainage Class
	Frost Action
	Frost-Free Days
	Hydrologic Group
	Map Unit Name
	Parent Material Name
	Representative Slope
	Soil Slipping Potential
	Unified Soil Classification

AASHTO = American Association of State Highway Transportation Officials;  
 NRCS = Natural Resources Conservation Service; WSS = Web Soil Survey

Table 1.3. Univariate statistics for NRCS continuous variables.

Type	Variable	min	Q <sub>0.25</sub>	mean	median	Q <sub>0.75</sub>	max	range	sd	cv (%)	skewness	kurtosis
Properties	pH*	5.8	6.8	7.0	7.1	7.3	7.7	1.9	0.4	5.0	-0.76	0.60
	Clay (%)	15.0	19.5	21.7	21.0	24.0	31.0	16.0	3.6	16.6	0.35	0.18
	Silt (%)	62.0	67.6	70.8	68.9	75.0	77.0	15.0	3.9	5.5	-0.08	-1.09
	Sand (%)	4.0	4.0	7.6	6.9	9.7	14.0	10.0	3.2	42.0	0.48	-1.01
	Bulk Density (g cm <sup>-3</sup> )	1.23	1.36	1.39	1.40	1.42	1.60	0.37	0.07	4.7	-0.01	1.82
	CEC (cmol <sub>c</sub> kg <sup>-1</sup> )	12.4	16.7	19.2	17.8	21.7	33.0	20.6	4.1	21.4	1.71	3.65
	Depth of A Horizon (in)	6	7	9	8	10	18	12	2	28.7	1.35	1.48
	Representative Slope (%)	1.5	4.0	8.2	9.0	9.0	16.0	14.5	3.6	43.9	0.47	-0.08
	Water Content at 1500 kPa (%)	10.3	13.5	14.9	14.5	15.3	20.6	10.3	2.4	16.1	0.47	-0.33
	Water Content at 3 kPa (%)	25.3	28.7	29.6	29.4	30.0	32.9	7.6	1.7	5.7	0.11	-0.44
Indices	NCCPI (Small Grain)	0.246	0.484	0.518	0.510	0.540	0.668	0.422	0.060	11.6	-0.27	2.51
	NCCPI (Corn)	0.474	0.703	0.719	0.743	0.796	0.899	0.425	0.106	14.7	-0.82	-0.24
	NCCPI (Soybean)	0.346	0.557	0.604	0.614	0.663	0.880	0.534	0.126	20.8	-0.60	-0.18

\*pH was measured from field soil samples and not retrieved from WSS

NRCS = Natural Resources Conservation Service; CEC = Cation-exchange capacity at pH 7; NCCPI = National Commodity Crop Productivity Index;

min = minimum; Q<sub>0.25</sub> = 1<sup>st</sup> quartile; Q<sub>0.75</sub> = 3<sup>rd</sup> quartile; sd = standard deviation; cv = coefficient of variation

n = 123 for all NRCS variables

Table 1.4. NRCS soil property categorical variables and their distribution.

Fields (n)	Order	Fields (n)	Slope Position
79	Alfisols	44	Summit; Summit, shoulder;
3	Entisols		Summit, shoulder, backslope
41	Mollisols	59	Shoulder; Shoulder,
			backslope; Shoulder, summit;
Fields (n)	Suborder		Shoulder, toeslope
79	Udalfs	12	Backslope; Backslope,
3	Fluvents		shoulder
41	Udolls	5	Footslope
		3	Toeslope
Fields (n)	Great Group	Fields (n)	3D Landform Position
79	Hapludalfs	68	Interfluve
3	Udifluvents	29	Interfluve, side slope
34	Argiudolls	8	Side slope
7	Hapludolls	4	Base slope
		15	No Data
Fields (n)	Subgroup	Fields (n)	Depth to Restrictive Feature
70	Typic Hapludalfs	14	10 to 25 inches to strongly
9	Mollic Hapludalfs		contrasting textural
3	Aquic Udifluvents		stratification, 20 to 39 inches
34	Typic Argiudolls		to lithic bedrock; 10 to 25
6	Aquic Hapludolls		inches to strongly contrasting
2	No Data		textural stratification
Fields (n)	Soil Series		20 to 44 inches to lithic
3	Atterberry-Downs silt loams	9	bedrock; 20 to 39 inches to
10	Dodgeville silt loam		lithic bedrock; 24 to 48 inches
3	Dodgeville soils		to lithic bedrock; 16 to 55
6	Downs silt loam		inches to lithic bedrock
34	Fayette silt loam	22	36 to 72 inches to lithic
6	Muscatine silt loam		bedrock; 39 to 59 inches to
3	Newglarus Complex		lithic bedrock; 42 to 60 inches
14	Newglarus silt loam		to lithic bedrock
12	Palsgrove silt loam	78	More than 80 inches
6	Seaton silt loam		
18	Tama silt loam		
8	No Data		
Fields (n)	Texture		
120	silt loam		
3	silty clay loam		

Fields (n)	Parent Material	Fields (n)	Depth to Water Table
65	Loess	7	About 12 to 36", About 24 to 48 inches
34	Loess over clayey pedisegment derived from dolomite; Loess over clayey pedisegment over loamy residuum weathered from dolomite; Loess over brown clayey pedisegment over loamy residuum weathered from dolomite	5	About 48 to 72 inches
		111	More than 80 inches
		Fields (n)	Saturated Hydraulic Conductivity Class ( $\mu\text{m s}^{-1}$ )
		120	Moderately High (1 to 10)
		3	High (10 to 100)
6	Loess over calcareous loess over a landscape of residuum weathered from clayey shale	Fields (n)	Saturated Hydraulic Conductivity ( $\mu\text{m s}^{-1}$ )
		1	7.7
6	Loess over clayey pedisegment	7	8
3	Loess over maquoketa residuum weathered from calcareous shale	112	9
		1	9.17
4	Silty or dark slope alluvium	3	28
3	Silty loess over clayey pedisegment over residuum weathered from dolomite	Fields (n)	Available Water Storage
		21	Low
		11	Moderate
3	No Data	81	High
		10	Very High
Fields (n)	Soil Moisture Subclass	Fields (n)	Available Water Capacity
104	Typic	3	20%
9	Aquic	34	21%
11	No Data	17	22%
		69	23%
Fields (n)	Drainage Class	NRCS = Natural Resources Conservation Service All variables have missing data with n=1 unless otherwise stated. Variables with a single category were not listed.	
7	Somewhat poorly drained		
5	Moderately well drained		
111	Well drained		
Fields (n)	Flooding Frequency Class		
120	None		
3	Occasional		

Table 1.5. NRCS categorical indices and their distribution.

Fields (n)	Farmland Class	Fields (n)	Wind Erodibility Index (T A <sup>-1</sup> yr <sup>-1</sup> )
14	Not prime farmland	86	48
70	Farmland of statewide importance	37	56
39	Prime farmland if drained... (n= 1), All areas are prime farmland (n = 38)	Fields (n)	Run-off Class
		21	Low
		4	Medium
		24	High
		5	Very High
Fields (n)	WICCPI (Corn)	Fields (n)	OM Depletion
3	Low inherent productivity	29	Moderate
6	Moderately low inherent productivity	94	Moderately High
80	Moderate inherent productivity		
25	Moderately high inherent productivity	Fields (n)	Suitability to Aerobic Organisms
7	High inherent productivity	3	Not favorable
3	No Data	120	Somewhat favorable
Fields (n)	Hydrologic Group	Fields (n)	Soil Surface Sealing
25	B	19	Low
96	C	55	Moderate
3	No Data	49	High
Fields (n)	Non-irrigated Capability Class	Fields (n)	Tolerable Soil Loss (T A <sup>-1</sup> )
40	Class 2	21	2
73	Class 3	24	3
9	Class 4	78	5
2	No Data		
Fields (n)	Fragile Soil Index	Fields (n)	Whole Soil Erodibility Factor
85	Moderately Fragile	41	0.32, 0.37
15	Fragile	72	0.43
2	No Data	10	0.49, 0.55
Fields (n)	Wind Erodibility Group	All variables have missing data with n=1 unless otherwise stated. Variables with a single category were not listed. NRCS = Natural Resources Conservation Service; WICCPI = Wisconsin Commodity Crop Productivity Index	
37	Group 5		
86	Group 6		

values were primarily larger than in previous research, except for minC (Table 1.6). Mean minC was one-third of values reported by Hurisso et al. (2016). Mineralizable C is highest in the late summer months of June through August (Diederich et al., 2019), thereby the earlier sampling dates likely depressed minC values. Even within our study's sampling period of late April to Early July, minC was sensitive to temporal variation as sampling date had a strong positive linear relationship ( $R^2 = 0.20$ ) with minC (Table 2.5). Despite spatiotemporal variability similar to routine soil tests, direct comparisons of soil health values between studies are difficult due to indicator sensitivity to spatiotemporal variability (Hurisso et al., 2018a).

The strength of linear relationships varied largely between soil health indicators (Figure 1.2). Carbon-related soil health indicators (SOM, TOC, and POXC) had moderately strong positive relationships between each other ( $R^2 > 0.54$ ). The exception was minC, which had weak positive relationships with all other C-related indicators ( $R^2 < 0.15$ ). Similarly, prior research found positive relationships between TOC and POXC while indicator relationships with minC were more varied (Culman et al., 2012; Plaza-Bonilla et al., 2014; Hurisso et al., 2016; Morrow et al., 2016; Bongiorno et al., 2019). Mineralizable C had a positive relationship with TOC in only 7 out of 13 studies in a meta-analysis by Hurisso et al. 2016. Mineralizable C also differentiated from POXC ( $R^2 = 0.15-0.80$ ) as the indicators responded differently based on the management practice utilized. The lack of strong relationships between C indicators highlight that they reflect different pools of C and provide different information regarding C bioavailability. Multiple C cycling indicators may

Table 1.6. Univariate statistics for soil health response variables.

Variable	min	Q <sub>0.25</sub>	mean	median	Q <sub>0.75</sub>	max	range	sd	cv (%)	skewness	kurtosis
SOM (%)	1.7	3.0	3.3	3.2	3.6	7.0	5.3	0.6	18.2	1.81	10.27
TOC (mg kg <sup>-1</sup> )	10000	17400	20200	20000	22400	42200	32200	4220	20.9	1.20	4.97
TN (mg kg <sup>-1</sup> )	867	1690	2030	2030	2310	3750	2880	449	22.1	0.44	0.77
POXC (mg C kg <sup>-1</sup> )	390	656	726	734	803	1160	768	118	16.3	0.03	1.14
minC (mg C kg <sup>-1</sup> day <sup>-1</sup> )	54.0	84.4	99.1	95.3	110	154	100	20.7	20.9	0.44	-0.19
PMN (mg N kg <sup>-1</sup> )	39.6	77.2	90.8	89.3	102	177	138	20.5	22.5	0.70	1.68
ACE (mg protein kg <sup>-1</sup> )	3470	6210	6860	6725	7490	10200	6730	1090	15.9	-0.04	0.67

SOM = soil organic matter; TOC = total organic carbon; TN = total nitrogen; POXC = permanganate oxidizable carbon; minC = mineralizable carbon; PMN = potentially mineralizable nitrogen; ACE = autoclaved-citrate extractable protein; min = minimum; Q<sub>0.25</sub> = 1<sup>st</sup> quartile; Q<sub>0.75</sub> = 3<sup>rd</sup> quartile; sd = standard deviation; cv = coefficient of variation  
n = 124 for all response variables.



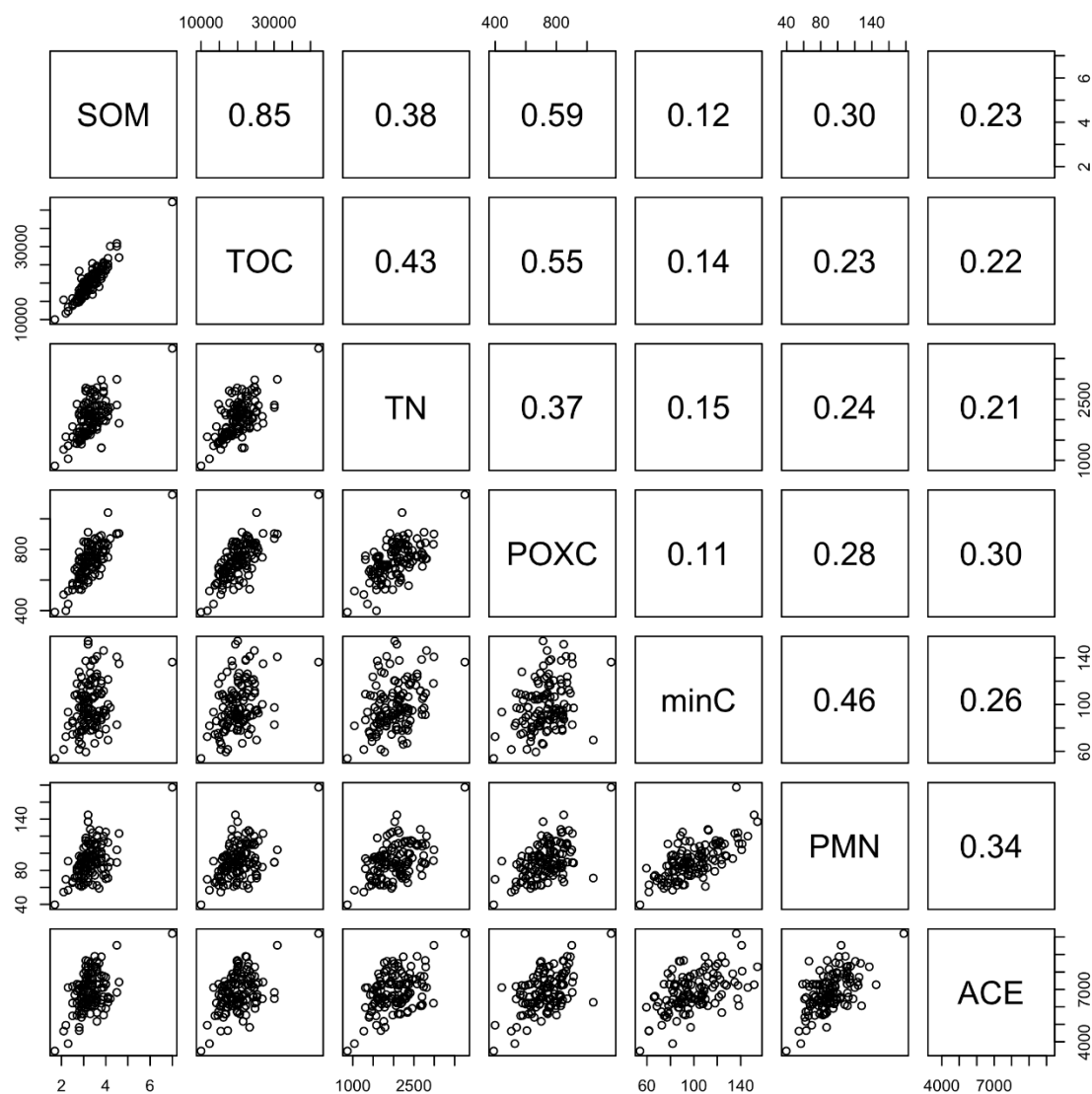


Figure 1.2. Linear relationships between soil health indicators and their R-squared value.

be required in soil health assessment to prioritize management strategies for desired soil functions.

Nitrogen-related soil health indicators (TN, PMN, and ACE) had weak positive relationships to each other ( $R^2 < 0.34$ ), and thereby represented distinct pools of N. In contrast, Geisseler et al. 2019 determined that ACE had a weak positive relationship with net N mineralization ( $R^2 = 0.21$ ), but had strong positive relationships with TN ( $R^2 = 0.86$ ) and POXC ( $R^2 = 0.84$ ). However, when soils were grouped according to low and high TN content, they concluded that soils with higher TN (2.71 to 12.48 g kg<sup>-1</sup> soil) exhibited stronger positive relationships between ACE and POXC as well as ACE and TN than soils with less TN (0.65 to 1.56 g kg<sup>-1</sup> soil). In our study, TN values (867 to 3750 mg kg<sup>-1</sup> soil) were similar to those observed in the low TN soils. Subsequently, weak relationships between ACE and POXC as well as ACE and TN were observed. Thus, soil TN content affects the strength of indicator relationships with ACE.

The biological incubation methods, minC and PMN, had the strongest relationship between each other than any other combination of the biological soil health indicators (POXC, minC, PMN, and ACE). Yet, coefficients of determination between minC and PMN were not large enough to select one indicator to represent both rates of C and N mineralization. This further supports the recommendation by NRCS for minC to represent general microbial activity while PMN and ACE represent bioavailable N (Stott, 2019).

### NRCS Variable Selection and Descriptive Statistics

From the WSS soil properties and indices, forty-one categorical and sixteen continuous explanatory variables were identified to fit the scope of our analysis (Tables 1.3 – 1.5). Eleven categorical variables (soil temperature regime, soil moisture class and subclass, agricultural organic soil subsidence, susceptibility to compaction, surface salt concentration, suitability to aerobic organisms, soil texture, flooding frequency class, ponding frequency class, and saturated hydraulic conductivity) were excluded from further analysis due to lack of variation (i.e. >90% of fields belonged to a single category). For all fields: soil temperature regime was mesic; soil moisture class was udic; agricultural organic soil subsidence was rated mineral; susceptibility to compaction was rated medium; and ponding frequency class was rated none. Surface salt concentration was rated low for all reported fields (n = 98). Since <80% of fields had a value reported, run-off class was excluded. Soil series was excluded due to imbalanced field distributions. For continuous variables, electrical conductivity was excluded because all fields had the same value of 0.0 dS/m (n = 98); yields for non-irrigated corn and soybean were excluded because they had observations for <80% of the fields with n = 27 and n = 29, respectively. Available water supply (0 - 25, 0 - 50, 0 - 100, 0 - 150 cm) is computed by multiplying the fixed factor of soil depth to available water capacity (AWC). Thus, available water supply was excluded because it provided redundant information already provided by available water capacity. Thirty-seven NRCS variables remained and were analyzed for significant effects (Table 1.7 – 1.9).

Normality of soil health indicator values is required for simple linear regression and ANOVA analysis to evaluate effects of NRCS soil information. West et al. (1996) described skewness as a measure of asymmetry where absolute values  $> 2.1$  is indicative of a non-normal distribution. Absolute “excess” kurtosis values  $> 4.1$  (i.e. kurtosis (proper)  $> 7.1$ ) is indicative of a non-normal peak shape. Only SOM and TOC presented any signs of non-normality as their kurtosis (excess) values indicated “peakedness” (Table 1.6). All continuous NRCS soil variables met conditions of normality (Table 1.3). For simple linear regression, evaluation of Q-Q and residuals versus fitted values plots concluded that the assumptions of normality, constant variance, and linearity were met for analysis of NRCS soil properties with soil health indicators. Thus, no transformations were implemented.

## **Effects of Inherent Soil Properties and Indices**

### **Soil Taxonomy**

Classification of soils in the U.S. is performed according to the principles outlined in USDA-NRCS’ *Soil Taxonomy*, which categorizes soils primarily by their composition, structure, and chemical and physical properties. Since the criteria used in classification affects many soil functions, biological soil health is likely to differ between more disparate soil classifications. Three soil orders (Alfisols, Entisols, and Mollisols) were represented in our dataset (Table 1.4). Fields with higher SOM and TOC occurred in Mollisols and soils containing Mollic epipedons (Figure 1.3 and 1.4). An inherent aspect of Mollisols and Mollic epipedons is a deep, SOM-rich surface horizon primarily formed from long-term accumulation under historical grassland cover (Brady and Weil, 2010). In addition, fields

that were Mollisols and Udolls had higher TN (Figure 1.5). The influence of soil classification on TN may be less important as TN was differentiated at a coarser resolution than SOM and TOC. Suborder was the finest resolution that was able to differentiate TN compared to subgroup for SOM and TOC. Soil order and suborder have identical distributions due to perfect collinearity in the dataset. Thus, the resolution for TN may be even coarser.

The higher baseline values of soil health in Mollisols highlight that soil taxonomy is an important consideration in soil health evaluation and the setting of benchmarks. However, none of the biological indicators of soil health were differentiated by soil taxonomy. Although there were not differences at this regional scale, a greater representation of soil taxonomic classes may be necessary for assessing biological soil health on a national scale. The NRCS and the USDA-Agricultural Research Service are cooperating on a meta-analysis spanning 38 states to evaluate the effect of 57 different soil series, 68 great groups, 28 suborders, and 9 different soil orders on SMAF soil health indicators and their response to management (Karlen et al., 2019).

### **Parent Material**

Although parent material had an effect on SOM, TOC, and TN, its effect was difficult to interpret with the limited differentiation between parent materials (Figure 1.3 to 1.5). The surface layer of fields were primarily windblown-deposited materials such as silt and loess (Table 1.4). Only four out of 124 fields had surface alluvial deposits. The primary differences between parent materials were in the subsurface layers and many categories had 6 or fewer observations. Narrowing the focus to the Driftless Region may have limited

Table 1.7. Continuous NRCS variable effects on SOM, TOC, and TN.

Type	Variable	SOM			TOC			TN		
		p	Rel.	R <sup>2</sup>	p	Rel.	R <sup>2</sup>	p	Rel.	R <sup>2</sup>
Properties	pH	NS			0.03	+	0.038	0.032	+	0.037
	Clay (%)	NS			NS			NS		
	Silt (%)	NS			NS			NS		
	Sand (%)	0.004	+	0.067	0.002	+	0.077	NS		
	Bulk Density	0.0002	-	0.11	0.0002	-	0.11	0.009	-	0.055
	CEC	0.045	+	0.033	NS			NS		
	Depth of A Horizon	0.046	+	0.032	0.044	+	0.033	NS		
	Representative Slope (%)	NS			NS			NS		
	Water Content at 1500 kPa	NS			NS			NS		
	Water Content at 3 kPa	NS			NS			NS		
Indices	NCCPI (Small Grain)	NS			0.02	-	0.044	NS		
	NCCPI (Corn)	NS			NS			NS		
	NCCPI (Soybean)	NS			NS			NS		

NRCS = Natural Resources Conservation Service; NS = not significant; CEC = Cation-exchange capacity at pH 7; NCCPI = National Commodity Crop Productivity Index; SOM = soil organic matter; TOC = total organic carbon; TN = total nitrogen

Table 1.8. Continuous NRCS variable effects on POXC, minC, PMN, and ACE.

Type	Variable	POXC			minC			PMN			ACE		
		p	Rel.	R <sup>2</sup>	p	Rel.	R <sup>2</sup>	p	Rel.	R <sup>2</sup>	p	Rel.	R <sup>2</sup>
Properties	pH	0.0009	+	0.086	0.014	-	0.048	NS			0.033	-	0.037
	Clay (%)	NS			NS			NS			NS		
	Silt (%)	NS			NS						0.036	+	0.036
	Sand (%)	NS			NS			NS			0.019	-	0.045
	Bulk Density	NS			NS			NS			NS		
	CEC	NS			NS			NS			NS		
	Depth of A Horizon	0.022	+	0.043	NS			NS			NS		
	Representative Slope (%)	NS			NS			NS			NS		
	Water Content at 1500 kPa	NS			NS			NS			NS		
	Water Content at 3 kPa	NS			NS			NS			NS		
Indices	NCCPI (Small Grain)	NS			NS			NS			NS		
	NCCPI (Corn)	NS			NS			NS			NS		
	NCCPI (Soybean)	NS			NS			NS			NS		

NRCS = Natural Resources Conservation Service; NS = Not significant; CEC = Cation-exchange capacity at pH 7; NCCPI = National Commodity Crop Productivity Index; POXC = permanganate oxidizable carbon; minC = mineralizable carbon; PMN = potentially mineralizable nitrogen; ACE = autoclaved-citrate extractable protein.

Table 1.9. Categorical NRCS variable effects on soil health indicators.

Type	Variable	p-value						
		SOM	TOC	TN	POXC	minC	PMN	ACE
Properties	Available Water Capacity	0.0008	0.007	NS	NS	NS	0.02	0.002
	Order	<0.0001	0.0001	0.034	NS	NS	NS	NS
	Suborder	<0.0001	0.0001	0.034	NS	NS	NS	NS
	Great Group	0.0001	0.0001	NS	NS	NS	NS	NS
	Subgroup	<0.0001	<0.0001	NS	NS	NS	NS	NS
	Drainage Class	NS	0.009	NS	NS	NS	NS	NS
	Parent Material	0.004	0.0009	0.035	NS	NS	NS	NS
	Depth to Restrictive Feature	0.012	NS	0.012	NS	NS	NS	NS
	3D Landform Position	NS	NS	NS	NS	NS	NS	0.042
	Depth to Water Table	NS	0.009	NS	NS	NS	NS	NS
	Available Water Storage	NS	NS	NS	NS	NS	NS	NS
	Saturated Hydraulic Conductivity	NS	NS	NS	NS	NS	NS	NS
Indices	Fragile Soil Index	NS	NS	0.004	NS	0.026	0.018	0.006
	Soil Surface Sealing	<0.0001	<0.0001	0.033	0.002	NS	NS	0.013
	OM Depletion	0.022	0.018	0.033	NS	NS	NS	NS
	WICCPI (Corn)	NS	0.0499	NS	NS	NS	NS	NS
	Whole Soil Erodibility Factor	<0.0001	0.0006	NS	NS	NS	NS	NS
	Tolerable Soil Loss	NS	NS	0.047	NS	NS	NS	NS
	Farmland Class	NS	NS	NS	NS	NS	NS	NS
	Hydrologic Group	NS	NS	NS	NS	NS	NS	NS
	Non-Irrigated Capability Class	NS	NS	NS	NS	NS	NS	NS
	Wind Erodibility Group	NS	NS	NS	NS	NS	NS	NS
	Wind Erodibility Index	NS	NS	NS	NS	NS	NS	NS

NRCS = Natural Resources Conservation Service; WICCPI = Wisconsin Commodity Crop Productivity Index;

NS = not significant; SOM = soil organic matter; TOC = total organic carbon; TN = total nitrogen;

POXC = permanganate oxidizable carbon; minC = mineralizable carbon; PMN = potentially mineralizable nitrogen;

ACE = autoclaved-citrate extractable protein



the capacity to accurately identify minute differences in parent materials. At a larger regional or national scale, parent material may have clearer effects on soil health as parent materials might diverge further in composition. Effects of parent material on microbial parameters have been found in natural and forested systems, but most effects are explained by soil properties, such as clay content and pH, resulting from soil development under different parent materials (Yarwood et al., 2014; Alfaro et al., 2017; Angst et al., 2018). It is unclear if parent material would be a useful component to soil health assessment or if it could be easily reflected by other properties.

### **Slope Position**

Representative slope percentage did not affect soil health values, but primarily summit and primarily back slope positions had higher SOM than other hillslope positions. Soil protein was highest in soil profiles found on both interfluvial and side slope positions. Interfluvial positions are analogous to summit positions while side slopes are analogous to back slopes (Schoeneberger and Wysocki, 2017). Due to parallel surface water flow, back slopes/side slopes generally have higher rates of erosion relative to accumulation compared to summit/interfluvial and toeslope positions. Thus, it is surprising that soils primarily residing on back slopes/side slopes would contain the highest SOM and ACE values due to their predisposition to erosion. Contour strip cropping was common on farms in our study area and may imply that preventative management was taken for areas with high susceptibility to water erosion. However, this was not captured in our study. Future

research may want to evaluate the relative importance of this management practice on varying slopes.

### **Depth of A Horizon**

As the thickness increased of a soil's A horizon (the top mineral layer of soil that is typically higher in OM than its lower layers), the amount of SOM, TOC, and POXC increased. A thicker A horizon may be indicative of soils that accumulated more OM due to the factors of soil formation. However, management such as tillage and cropping also affect the depth of soil horizons, but is not always reflected in soil surveys. Surface layers that are more frequently disturbed are more likely to change in composition and structure over time.

The recency of soil surveys may limit the ability to discriminate differences at the field level derived under intensive management and land use change. According to the USDA-NRCS, field mapping for the entire state of Wisconsin was completed in fall 2005 and soil survey information made available online in 2006. The latest modifications to the soil survey occurred in September of 2019 in an effort to continuously update WSS compatibility with GIS technology and modern standards of mapping. Explicit statements highlighting the soil information that was updated, when, and how is critical to interpretation of field observations. Updates on soil information from archived samples using modern methods compared to new observations from soil field mapping have very different considerations for interpretation. Nevertheless, Depth of A horizon can be directly measured by farmers and land managers with training, and thereby can circumvent the use of estimates from the WSS and provide a potential co-variate for soil health assessment.

## pH

Soil acidity is recognized as an important factor for microbial community composition and activity (Rousk et al., 2009), but the bounds of the pH range used in analysis may be an important consideration in evaluating effects. All indicators were affected by pH besides SOM and PMN (Table 1.7 and 1.8). Total organic C, TN, and POXC had positive relationships with pH while minC and ACE had negative relationships with pH. Most fields had pH values higher than the target optimum for corn, soybean, and alfalfa production with 50% of fields between a pH of 6.8 and 7.3 (Laboski and Peters, 2012) (Table 1.3). The higher density of neutral to alkaline fields may not capture pH effects in soils that span acidic to basic conditions. Field experiments with a more extensive range of pH values (as low as 3.5 to as high as 8.3) identified the opposite relationship between C mineralization and pH (Rousk et al. 2009 and Kemmit et al. 2006). Even with the direct measurement of pH (i.e. instead of an estimated pH from WSS), the limited range in pH values explained less than 5% of the variation in soil health indicators. Since pH is largely managed for optimizing production of specific crops, pH may be more useful for adjusting soil health expectations across different cropping systems than within similar cropping systems.

## Soil Texture, Cation-Exchange Capacity, and Bulk Density

Currently, multiple soil health assessments for agricultural systems (e.g. CASH and SMAF) utilize soil texture to adjust indicator scoring functions (Moebius-Clune et al., 2016; Stott, 2019). Other properties that relay soil textural information may provide further

understanding in biological soil health differences. Of the 124 fields, only three had a soil texture other than silt loam. Although variation in soil texture was limited, the amount of sand (one of the particle sizes that quantitatively defines texture) had a positive relationship with SOM, TOC, and ACE. Sand content was relatively low (4.0 to 14.0%). Typically, larger variation in sand content is necessary to affect soil function as it is relatively inert compared to the other solid components of soil, such as clay. Small changes in the proportion of clay-sized particles has large effects on soil properties and affects a soil's potential for accumulating SOM and TOC (Moebius-Clune et al., 2016). However, clay content did not affect any of the soil health indicator values in our study while soil protein was the only indicator responsive to silt content.

The Cation-Exchange Capacity (CEC) of soils is primarily a function of OM and clay mineralogy (Brady and Weil, 2010). Soil organic matter increases soil CEC through its high proportion of negatively charged sites that attract and exchange cations with the soil solution. As CEC and SOM are intrinsically related, soils with higher CECs coincided with higher SOM values. Cation-exchange capacity values from the WSS reflect antecedent SOM, which can be related by other soil information that is unlikely to change over relevant timescales (e.g. soil taxonomy).

Bulk density had stronger linear relationships with SOM and TOC than sand content or any other continuous soil property. Bulk density more accurately reflects soil composition and texture as it is a measure of the amount of soil solids within a given volume. It is a function of soil aggregation as well as sand, silt, clay, and OM content. Bulk

density estimates from the WSS may prove useful for adjusting soil health benchmarks or be a potential substitute for textural class.

### **Soil Water**

Total organic C was heavily influenced by soil water properties. Wetter soils contained more TOC (Table 1.9 and Figure 1.4). Aquic Hapludolls had higher TOC values than other subgroup classifications. Soils with aquic moisture subclass regimes experience saturated conditions more frequently than drier regimes (Soil Survey Staff, 1999). These conditions slow decomposition rates causing SOM and C to accumulate. In fact, soils with poorer drainage, shallower depths to water table, and higher AWC had higher TOC, but hydrologic group had no effect on TOC or any other soil health indicator (Figure 1.4 and Table 1.9).

Available water capacity is an estimate of plant-available water calculated as the difference in soil water content at 3 kPa (i.e. field capacity) and 1500 kPa (i.e. wilting point). Although TOC increased with increasing AWC, water content at 1500 kPa and 3 kPa did not affect any indicators and should not be used individually to define critical thresholds for TOC. Site-specific management is likely to cause divergence from WSS estimated AWC. Future research should evaluate direct measurement of AWC for use as a potential co-variate while other soil moisture information may be well-estimated from the WSS. Soil moisture is an important consideration for evaluating TOC potential and was identified as the primary environmental factor for storage of TOC in cropland (Wiesmeier et al., 2013).

### **Soil Erodibility and Tolerance to Soil Loss**

Soil organic matter and TOC were impacted by whole soil erodibility and tolerance to soil loss. Soil erodibility (i.e. K factor) is used to quantify the vulnerability of soil particles to detachment and movement by water. Increases in index values reflect increased susceptibility to water erosion. Soils with a whole soil erodibility factor less than 0.43 had higher SOM and TOC. Granted, erodibility is not the only component determining soil loss, but reduced susceptibility to water erosion may minimize the overall erosion rate leading to higher SOM and TOC values. Soil erodibility is determined mainly by soil structure, saturated hydraulic conductivity, and percentage of silt, sand, and SOM. Thus, its relationship with SOM and TOC may in part be the result of differences in antecedent SOM.

Only TN increased with increasing tolerance to soil loss and did not reflect SOM and TOC, which are usually depleted when soil is lost. Tolerable soil loss (i.e. T factor) is described as the “maximum rate of annual soil loss that will permit crop productivity to be sustained economically and indefinitely on a given soil” (Wischmeier and Smith, 1978). The inherent soil properties used to estimate tolerable soil loss are the depth to root and plant growth limiting layers and the severity of their physical and chemical properties (USDA-NRCS, 2019). Since tolerable soil loss is related to a soil’s inherent fertility, soils with higher tolerance may exhibit intrinsic conditions for superior crop growth, such as higher TN.

### **NRCS Soil Health Indices**

Several NRCS soil health indices depicted differences in most of the soil health indicators, especially the biological indicators (POXC, minC, PMN and ACE) (Figure 1.3 to 1.7). Soils with lower susceptibility to surface sealing had higher SOM, TOC, TN, and POXC, while ACE was highest in highly susceptible soils. Surface seals and crusts are layers with poor infiltration formed by the destruction of soil aggregates, which causes surface pores to be blocked or filled (USDA-NRCS, 2008). Soil susceptibility to surface sealing is related to soil texture, OM content, and sodium content. The fragile soil index is a measurement of a soil's susceptibility to degradation. It is characterized by low OM content, low aggregate stability, and weak soil structure. Soils that were less fragile had higher TN, minC, PMN, and ACE. Organic matter depletion is an index of soil vulnerability to OM losses estimated by antecedent OM, aeration, clay content, and land shape. Lower susceptibility to OM depletion had higher SOM, TOC, and TN. Many individual properties have not identified differences in biological soil health indicators, but NRCS soil health indices represented differences well. The NRCS soil health indices combine multiple properties to explain susceptibility of soil to various forms of degradation and require further exploration as potential co-variates for biological indicators.

### **NRCS Productivity Indices**

Farmland class and National Commodity Crop Productivity Indices (NCCPI) for corn and soybean did not affect any soil health indicators. The NCCPI for small grains had a negative relationship with TOC. It is an index based on inherent soil properties and modeled

with winter wheat. The index weighs heavily the negative effects to small grain production from saturated soil conditions. Since TOC increases under wetter conditions, the inverse relationship between TOC and NCCPI for small grains occurs despite the common association of TOC with enhanced crop productivity. The Wisconsin Commodity Crop Productivity Index (WICCPI) assesses the inherent capacity of land to produce non-irrigated crops through soil, landscape, and climate criteria for corn production. Fields ranked high according to WICCPI had higher TOC than less productive ratings. This index directly utilized antecedent OM in its evaluation, which may contribute to the differences in TOC between ratings. The influence of biological soil health is unrepresented in crop productivity indices, but recent work from Wade et al. (2020) found that improvements in biological soil health (POXC, minC, and ACE) increased fertilizer N use efficiency and corn grain yield. Biological soil health should be incorporated into crop productivity indices based on soil potential.

## **CONCLUSIONS**

Besides SOM and TOC, relationships between soil health indicators sufficiently diverged ( $R^2 < 0.60$ ) highlighting that they provide unique information regarding soil health and the bioavailability of C and N. The main pools of C and N (SOM, TOC, and TN) were most affected by soil taxonomy, textural-related soil information, and indices regarding soil depletion or erodibility. Total organic C was the only indicator affected by soil water properties and demonstrated the importance of soil moisture in C cycling. Biological indicators representing the bioavailability of C and N (POXC, minC, PMN, and ACE) were differentiated by fewer NRCS variables. However, several NRCS soil health indices (soil



surface sealing, OM depletion, and the fragile soil index) captured differences in many biological indicators as well as the larger C and N pools. Many NRCS properties and indices that utilized antecedent SOM content were associated with soil health indicators, especially SOM and TOC, but it was not consistent. It is important to incorporate and contextualize these differences in indicator response to soil properties when evaluating soil health within and across regional cropping systems. Identification of the most influential soil properties on individual soil health indicators is critical to the establishment of benchmarks that evaluate soils among their peers. Soil health assessments with improved criteria for benchmarking would optimize their ability to inform farm management decisions and enact change that improves agricultural production and the provision of ecosystem services.

Existing, publicly available soil information provided by the NRCS WSS was a useful tool to explore how inherent soil properties affected soil health in a given region. Analysis of soil health in the Driftless Region identified potential co-variables to include in soil health assessment despite minimization of variation in soil properties relative to inter-region variation. In particular, soil taxonomy, soil moisture, soil surface sealing, whole soil erodibility, the fragile soil index, and OM depletion were identified as potential co-variables for indicator scoring functions and benchmarks. Those properties and indices differentiated soil health values within the Driftless Region. This soil information should be collected and utilized in future soil health studies to understand their effects in other regions, climates, and cropping systems as well as to understand their applicability across spatial scales. Soil health assessments that incorporate essential co-variables when setting benchmarks may more accurately represent the health status of a given soil among its related peers as well

as inform management to improve crop production and the provision of ecosystem services.

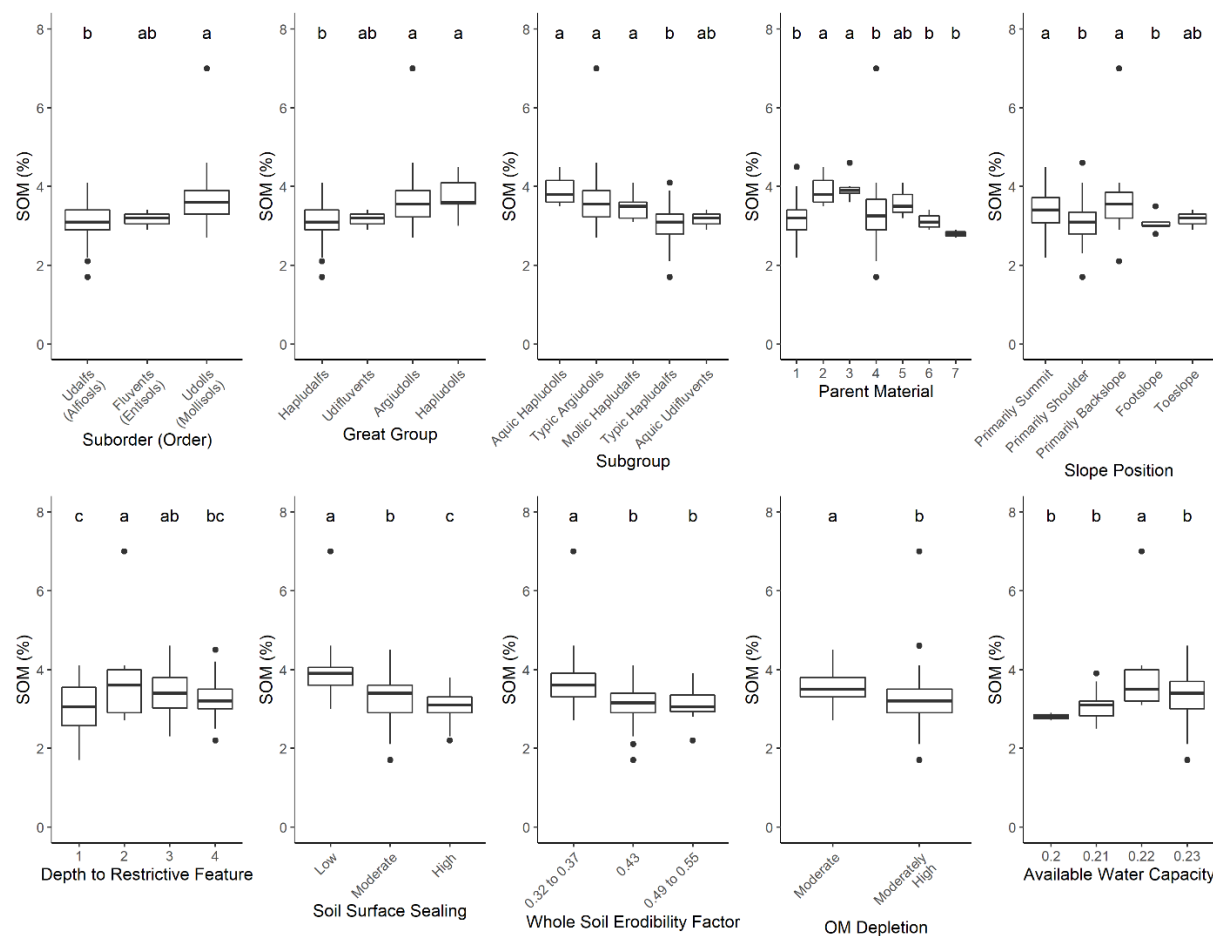


Figure 1.3. NRCS categorical variable effects on soil organic matter (SOM).

Parent Material: 1 = loess; 2 = loess over calcareous loess over a landscape of residuum weathered from clayey shale, 3 = loess over clayey pedisediment, 4 = loess over clayey pedisediments derived from dolomite or over residuums of dolomite; 5 = loess over maquoketa residuum weathered from calcareous shale; 6 = silty or dark slope alluvium, and 7 = silty loess over clayey pedisediment over residuum weathered from dolomite.

Depth to Restrictive Feature: 1 = 10 to 25 inches to strongly contrasting textural stratification, 20 to 39 inches to lithic bedrock; 2 = 20 to 44 inches to lithic bedrock; 20 to 39 inches to lithic bedrock; 24 to 48 inches to lithic bedrock; 16 to 55 inches to lithic bedrock; 3 = 36 to 72 inches to lithic bedrock; 39 to 59 inches to lithic bedrock; 42 to 60 inches to lithic bedrock; 4 = More than 80 inches.

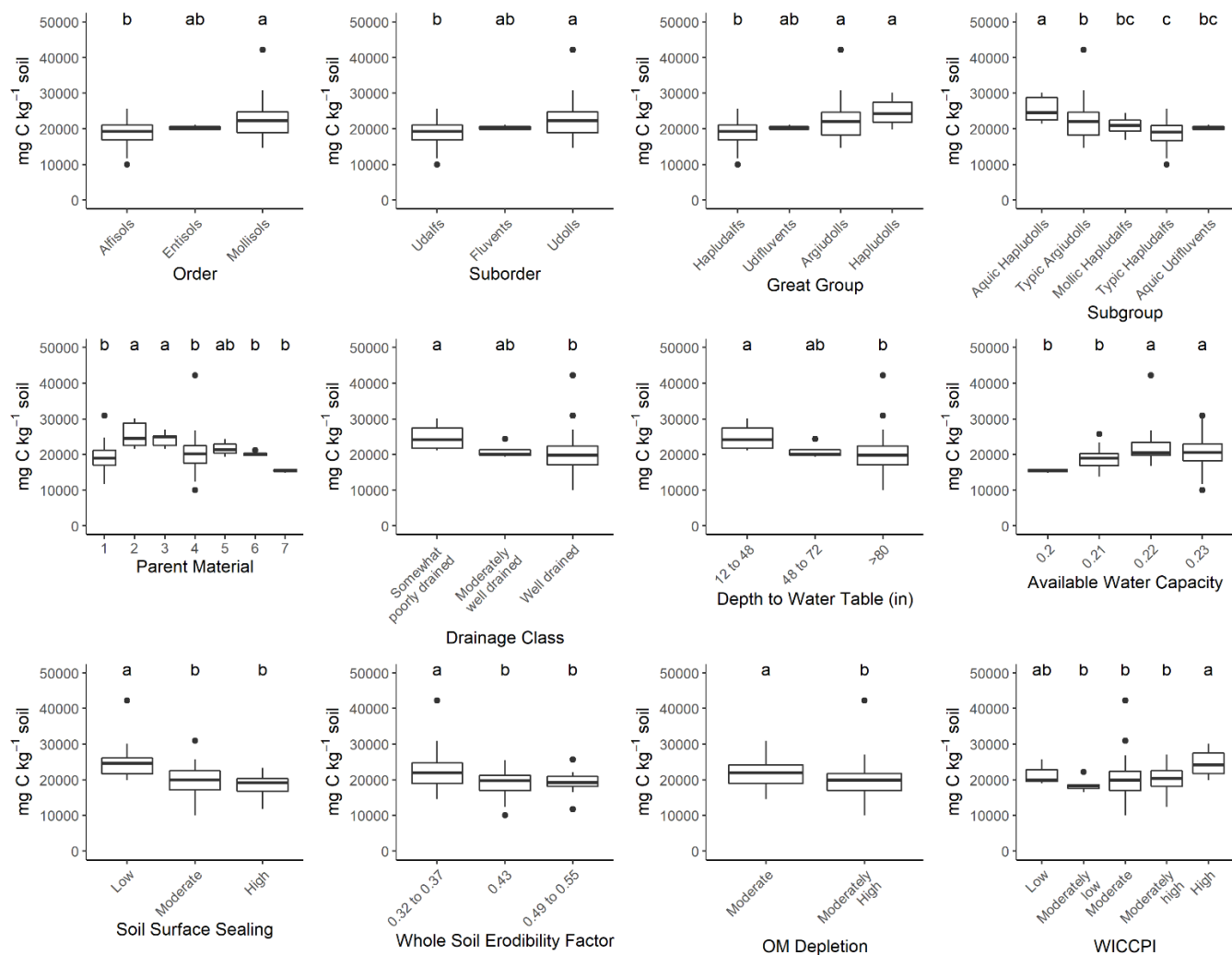


Figure 1.4. NRCS categorical variable effects on total organic carbon (TOC).

Parent Material: 1 = loess; 2 = loess over calcareous loess over a landscape of residuum weathered from clayey shale, 3 = loess over clayey pedisediment, 4 = loess over clayey pedisediments derived from dolomite or over residuums of dolomite; 5 = loess over maquoketa residuum weathered from calcareous shale; 6 = silty or dark slope alluvium, and 7 = silty loess over clayey pedisediment over residuum weathered from dolomite.

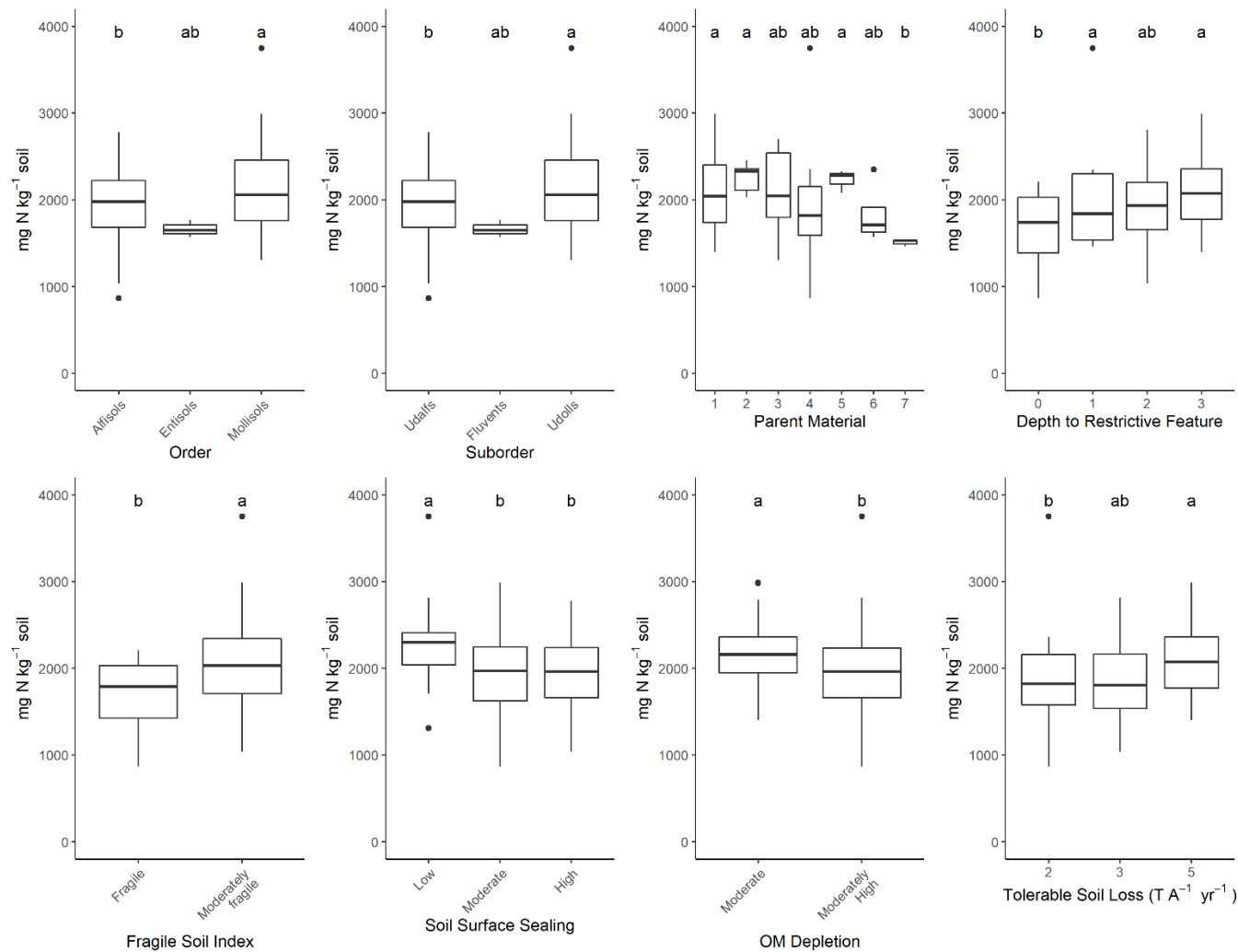


Figure 1.5. NRCS categorical variable effects on total nitrogen (TN).

Parent Material: 1 = loess; 2 = loess over calcareous loess over a landscape of residuum weathered from clayey shale, 3 = loess over clayey pedisegment, 4 = loess over clayey pedisegments derived from dolomite or over residuums of dolomite; 5 = loess over maquoketa residuum weathered from calcareous shale; 6 = silty or dark slope alluvium, and 7 = silty loess over clayey pedisegment over residuum weathered from dolomite.

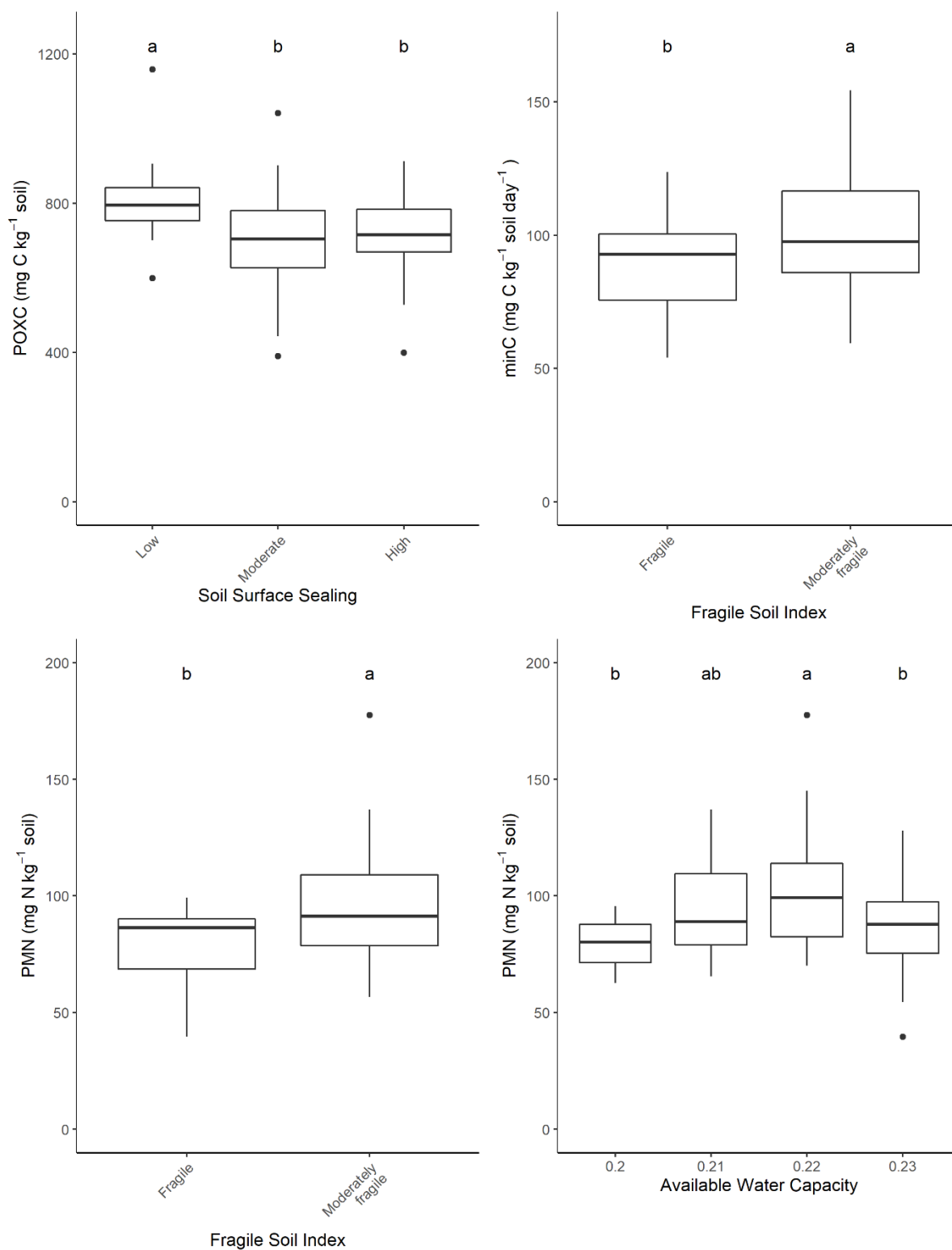


Figure 1.6. NRCS categorical variable effects on permanganate oxidizable carbon (POXC), mineralizable carbon (minC), and potentially mineralizable nitrogen (PMN).

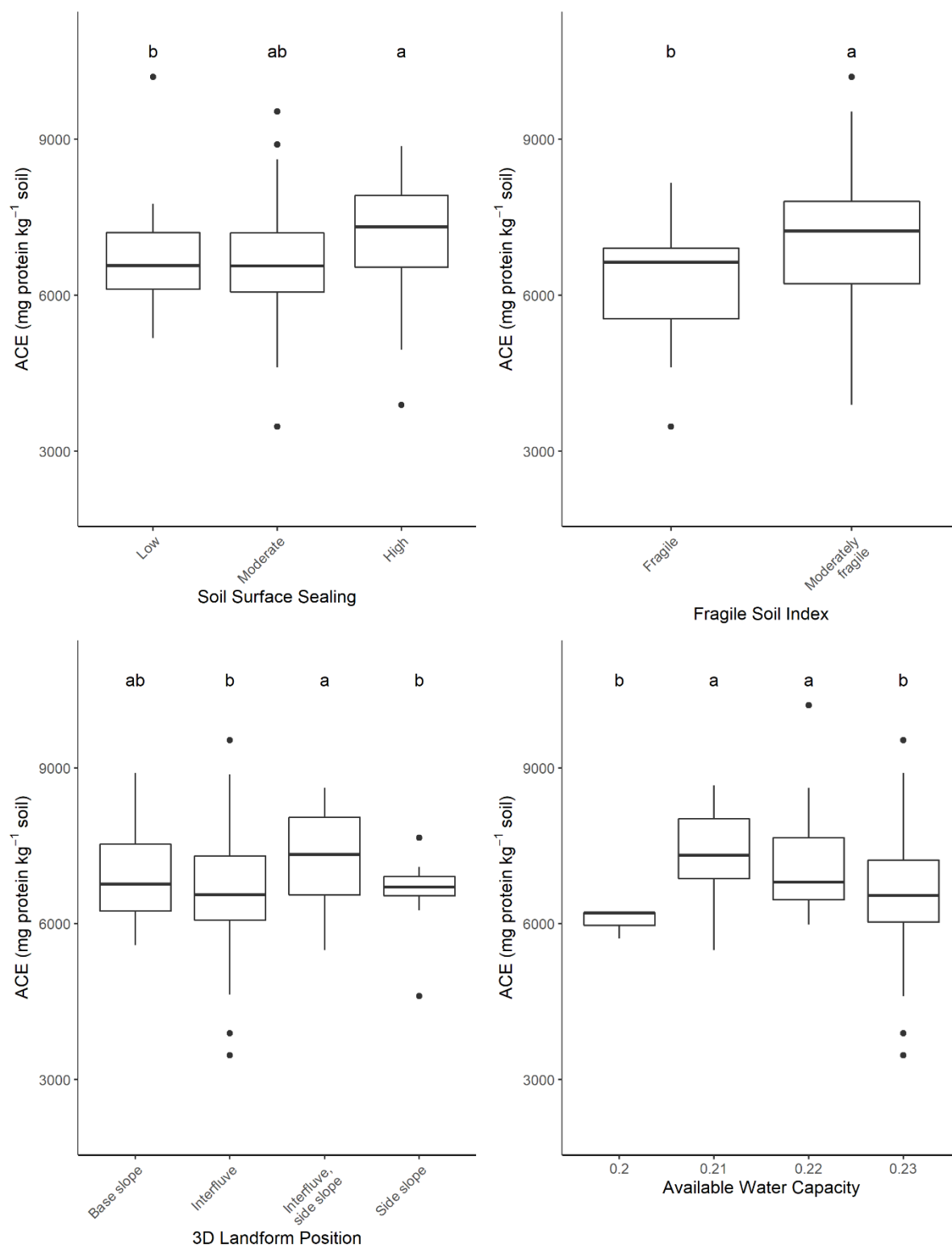


Figure 1.7. NRCS categorical variable effects on autoclaved-citrate extractable protein (ACE).

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## CHAPTER 2: FARM MANAGEMENT AND BIOLOGICAL SOIL HEALTH

### ABSTRACT

Understanding the effect of farm management and its interaction with inherent soil properties on soil health is critical to developing best management practice recommendations. To evaluate the effect of organic management practices and their duration on soil health, soils were obtained from 124 fields across 16 certified organic grain farms in the Driftless Region of Wisconsin. In spring of 2018 and 2019, soils were sampled to a depth of 15 cm and analyzed for soil health with an emphasis on biological health. The main pools of carbon and nitrogen (soil organic matter (SOM), total organic carbon (TOC), and total nitrogen (TN)) as well as biological soil health indicators (permanganate oxidizable carbon (POXC), mineralizable carbon (minC), potentially mineralizable nitrogen (PMN), and autoclaved-citrate extractable protein (ACE) were measured. Simple linear regression and analysis of variance were utilized to determine effects of management practices on soil health indicators while regression tree analysis was utilized to determine the relative importance of both inherent soil properties and management practices on soil health. Land use legacy, cropping sequence, manure management, and tillage management affected soil health values, but indicators responded to different management practices. Although SOM and TOC were affected by management, susceptibility to surface sealing was the most important determinant for SOM and TOC. Soils with lower susceptibility had higher SOM and TOC. Total nitrogen was most influenced by the inclusion of a perennial crop in the cropping sequence; exclusion led to decreased TN. Sampling time had the largest effect on

minC, thereby standardization of a sampling window is necessary for minC use as an indicator. Many of the newly recommended biological soil health indicators (POXC, PMN, and ACE) did not generate regression trees, but were affected by specific management practices. The selective response of soil health indicators to management highlights that many management strategies may be required to achieve soil health goals. Future research should utilize larger regional and national soil health datasets to improve selection of co-variates for soil health assessment, and generation of best management practice recommendations.

## **INTRODUCTION**

Agricultural systems rely on soil to accomplish a multitude of functions that affect food, feed, and fiber production as well as mitigation of climate change and environmental degradation. The ability of soil to provision services for the betterment of society has amplified public interest in soil health. According to the United States Department of Agriculture-Natural Resources Conservation Service (USDA-NRCS), soil health is “the capacity of the soil to function as a vital living ecosystem that supports plants, animals, and humans” (Stott, 2019). There are three components of soil health that regulate soil functioning: biological, physical, and chemical properties.

Organic agricultural systems heavily depend upon soil health’s biological component for crop productivity. Since synthetic fertilizers are prohibited, they rely on the activity of soil organisms to supply nutrients, especially nitrogen (N), through the processing of organic amendments, such as manure and compost. According to the 2016 Certified Organic Survey,

Wisconsin organic agriculture represents an over \$250 million industry with greater than 88,000 hectares in organic production (USDA-NASS, 2017). Over half of that area is in field crops, thereby making Wisconsin its second largest producer. Maintenance and improvement in biological soil health is critical to sustain organic agricultural systems. Many comparison studies found that biological metrics of soil health are improved in organic systems relative to conventional systems (Marinari et al., 2006; Suja, 2013; Surekha et al., 2013). However, to optimize soil function in organic systems, it is important to understand how variation in soil characteristics and management practices affect biological soil health on organic farms.

Debate over how to measure biological soil health has delayed standardization of methods and assignment of benchmarks for assessment. Relating the composition and abundance of a soil's microbial community to presumed functions has been one approach to understand the living component of soil (Lehman et al., 2015). Along with being cost prohibitive, microbial community composition does not yet have a clear interpretation for informing management (Stott, 2019). Indirect measurements known as indicators have been developed to quantify important biological processes with the specific aim to inform management. Robust indicators are easy to use, cost-effective, sensitive to management, well-suited to commercial labs, and able to supply timely information to adapt management (Morrow et al., 2016; Stott, 2019). With the 2019 NRCS endorsement of soil health indicators and release of standardized methods (Stott, 2019), prospects have improved for

coordinating regional and national efforts to develop informative soil health assessments (e.g. Norris et al., 2020).

For development of soil health assessments, long-term trials have been leveraged to develop an understanding of soil health threshold values and the effects of management across geospatial regions and edaphic conditions (e.g. Culman et al., 2012; Hurisso et al., 2016; Diederich et al., 2019; Norris et al., 2020). Individual long-term trials are able to observe changes in soil health from management while minimizing variation in inherent soil properties and climate. However, their capacity to evaluate multiple practices and their interactions are constrained by the land area available for such studies. Unstructured sampling designs on working farms present an opportunity to capture a large amount of variation in management that cannot be captured in long-term trials. Although a larger sampling size would be required to achieve sufficient statistical power, the potential to aggregate data from similar on-farm studies and long-term trials can be utilized to build large regional and national databases.

Another approach to handling a large number of predictor variables (e.g. inherent soil properties, climate, and management) relative to observations is to apply recursive partitioning, repeated splitting of observations into groups with similar response values. Classification and regression trees are methods of recursive partitioning well-suited for analysis of complex ecological data because of their ability to 1) analyze numerical and categorical values in response and explanatory variables, 2) identify linear and non-linear effects, 3) analyze high-order interactions, 4) handle missing values in response and



explanatory variables, and 5) present easily interpretable results (De'Ath and Fabricus, 2000; Strobl et al., 2009). The ability of classification and regression trees to identify and quantify the relative importance of numerous predictor variables is highly applicable to the evaluation of agroecosystems, which are the result of climatic, edaphic, and management factors.

Much research has focused on the effect of management on soil health and not on the effect of inherent soil properties beyond pH and texture despite public access to NRCS soil data for greater than 95% of U.S. counties (Soil Survey Staff, n.d.). While a majority of inherent soil properties are unalterable under relevant timescales, farm management is an important lever that can be utilized to optimize soil health. Nevertheless, inherent soil properties are determinants to an individual soil's potential; thereby it is critical to determine their effects on soil health and the potential interactions they may have with management. Regression tree analysis that includes both inherent soil properties and management as predictors may better inform our understanding of soil health potentials and effects at regional and national scales.

Overall, this research aims to determine the effect of organic management practices and their duration on the main soil pools of carbon (C) and N (soil organic matter (SOM), total organic carbon (TOC), and total nitrogen (TN) as well as biological indicators of soil health (permanganate oxidizable C (POXC), mineralizable carbon (minC), potentially mineralizable N (PMN), and autoclaved-citrate extractable protein (ACE) in order to develop best management practice recommendations for optimizing C and N cycling in organic grain

and forage production systems. The specific objectives of this study are to: 1) evaluate the individual effect of organic management practices on soil health indicators and 2) evaluate the relative importance of management and inherent soil properties on indicators of soil health.

Since variation within soil properties may be limited due to constraining field selection by region and cropping system, we hypothesize that the greatest variation in soil health indicator values will be explained by management. A meta-analysis of long-term research trials showed that POXC increases in systems utilizing reduced tillage and applications of processed C (e.g. compost) while minC increases in systems utilizing high-intensity tillage, cover crops, and manure (Hurisso et al. 2016). Mineralizable N was enhanced under reduced tillage, organic amendment and manure additions, and crop rotation diversity (Doran, 1987; Rasmussen et al. 1998; Mikha et al. 2006; Sanchez et al. 2001; Sharifi et al. 2008). Soil protein is similarly enriched under reduced tillage and organic amendment (Balota et al. 2016; Luna et al. 2016; Sandeep et al. 2016; Singh et al. 2016). We expect POXC, minC, PMN, and ACE to respond similarly to management on organic grain farms. Changes in SOM and TOC occur slowly and are unlikely to be detected in five-year management histories, but long-term management (e.g. duration of certified organic management and pre-organic land history) are likely to affect their values.

## **MATERIALS AND METHODS**

Field selection as well as soil sampling and analysis were performed according to the methods outlined in *Chapter 1: Inherent Soil Properties and Biological Soil Health*. Detailed

histories of the previous five years of management regarding cropping sequence, cover cropping, and tillage and fertility management as well as long-term field management information regarding organic management history were obtained directly from correspondence with each farmer.

In particular, the following management information for the past five years was obtained: the order, timing, and plant species used in the cropping sequence; the frequency, timing, and plant species used in cover cropping; the tillage implements used, the number of passes with each, and when they were used; and the application method, frequency, timing, source (i.e. animal), and physical state of manure and other applied amendments. The following information regarding the organic history of each field was obtained: first transition year, first organic certification year, duration under organic management (years), and land use prior to organic transition.

### **Statistical Analysis**

Descriptive statistics were used to explore the distribution of management and response variables (SOM, TOC, TN, POXC, minC, PMN, and ACE). The *describe* function from the *Psych* package in R was used to perform univariate statistics, such as mean, median, skewness, etc., on both continuous management and response variables. Histograms were used to visually review normality of continuous variables. Categorical management variable distributions were evaluated from the number of observations and percent of total observations per category. To exclude management variables from analysis that did not provide sufficient information due to lack of or excess variation, explanatory variables were removed from the dataset if either of the following criteria were met: 1) <80% of fields had

a value reported, 2) >90% of fields belong to a single category, and/or 3) categorical variables contained greater than ten categories that were highly unbalanced.

Of the remaining management variables, their effects on soil health indicators were assessed in R using analysis of variance (ANOVA) with a Fisher's least significant difference (LSD) test for categorical explanatory variables and simple linear regression for continuous explanatory variables ( $\alpha = 0.05$ ). The *aov* and *summary* functions were used for ANOVA while *lm* and *summary* functions were used for simple linear regression. Simple linear regression assumptions of linearity, constant variance, and normality were evaluated using residuals versus fitted values plots and Q-Q plots.

The remaining management variables were pooled with previously identified NRCS variables from *Chapter 1: Inherent Soil Properties and Biological Soil Health* to construct a dataset for regression tree analysis. The cumulative dataset of explanatory variables underwent analysis for multicollinearity. Contingency tables were used to evaluate multicollinearity between categorical variables while continuous variables were evaluated with simple linear regression. If perfect multicollinearity was identified between variables, the variable providing the most reliable highest resolution information was selected for inclusion in the regression tree model.

Regression tree analysis was used to evaluate the relative importance of inherent soil properties and management practices on soil health indicator values. For each soil health indicator, trees were manually generated 20-times under the following node termination criteria using *rpart*: 1) nodes must contain >10% of observations for a split to be attempted, 2) for a split to be included it must decrease the complexity parameter (cp) by a

minimum of 0.0001, and 3) the maximum branching depth of the tree is eight. Each manual generation of a tree was cross-validated with ten equally sized subsets of the original dataset. The cp for the branch depth that minimized the cross-validation error was recorded for each of the twenty manually generated trees. To avoid overfitting the data, a single regression tree was formed for each soil health indicator by pruning the original tree to the branch depth that most frequently ( $n = 20$ ) yielded the lowest cross-validation error. Primary and surrogate splits, and variable importance for pruned trees were obtained with *summary*. Variable importance is calculated as “the sum of the goodness of split measures for each split for which it was the primary variables, plus goodness  $\times$  (adjusted agreement) for all splits in which it was a surrogate” (Therneau & Atkinson, 2019).

## RESULTS AND DISCUSSION

### Descriptive Statistics

Nine categorical and eight continuous explanatory management variables were generated from farmer-provided field histories and records (Table 2.1 and 2.2). None of the management variables generated met any of the preliminary criteria for exclusion. Skewness and kurtosis values were indicative of normal distributions for continuous management variables according to West et al. (1996). Evaluation of residuals versus fitted values plots confirmed that all continuous management variables met the assumptions of constant variance, and linearity for simple linear regression with soil health indicators. However, Q-Q plots of continuous management variables did not always fulfill the assumption of normality; they typically exhibited minute deviations in regions with

relatively few observations. Log and exponential transformations did not improve normality. Consequently, simple linear regression and ANOVA proceeded without transformations for the evaluation of relationships between management factors and indicators of soil health (Table 2.3 and 2.4).

### **Sampling Time**

Mineralizable C and POXC were the only indicators affected by sampling day (Table 2.5). The linear relationship between the day of sampling and minC had the largest coefficient of determination ( $R^2$ ) with 0.20; it was nearly double the next largest  $R^2$  observed with any other indicator and explanatory variable (management or NRCS). As in our study, previous studies concluded that temporal variation was a limitation in soil health evaluation and comparison; soils sampled later in the growing season (June to August) tended to have higher minC, PMN, and POXC values than when sampled earlier (Diederich et al. 2019; Hurisso et al. 2018a). Unlike previous work, POXC had a weak inverse relationship with sampling day, but temporal variation in POXC values may have been more limited as we sampled over a narrower window. Still, standardizing a sampling period for soil health assessment may be essential for developing critical values and making meaningful interpretations for management recommendations, especially for minC.

### **Land Use Legacy**

Despite narrowing the focus of the study to certified organic grain and forage production systems, legacy effects of historical land use were a major contributor to soil health. All but one of the fields that were conventional pasture prior to organic certification

Table 2.1. Farm management categorical variables and their distribution.

Long-term Management		Short-term Management	
Fields (n)	Rotation Sequence	Fields (n)	Manure Use Year Prior
19	4-yr A	70	None
39	1-yr annual following 3-yr A	24	Spring
13	2-yr annual following 3-yr A	22	Fall
20	Rotations with 2-yr or 1-yr A	6	Winter
26	Annuals	2	No Data
6	Perennial lands transitioning to cropping	Fields (n)	Cover Crop Year Prior
Fields (n)	Manure State	68	No
8	None	55	Yes
54	Liquid		Till Type Year Prior
5	Solid	Fields (n)	(Most aggressive)
47	Combination	23	No-till
10	No Data	26	Min-till
Fields (n)	Pre-Organic Land History	56	Chisel/deep rip
97	Conventional dairy	4	Rotovator
7	CRP	7	Chisel and rotovator
7	Conventional Row Crop	6	Moldboard
6	Conventional Pasture	3	No Data
4	No Data	Fields (n)	Corn Residue Year Prior
	Till Type Past 5 Years	47	Silage
Fields (n)	(Most aggressive)	40	Grain or snaplage with stalks left
0	No-till	36	Grain with stalks removed
22	Min-till	1	No Data
38	Chisel/deep rip	Fields (n)	Crop Prior to Previous Corn Year
4	Rotovator	73	A or A-Ra
15	Chisel and rotovator	7	C
44	Moldboard	7	C-Rye
<p>All variables have missing data with n=1 unless otherwise stated.</p> <p>Min-till refers to either disk, cultivator, tine weeder, harrow, mulcher and finisher use.</p> <p>A = Alfalfa; C = Corn; S = Soybean; W = Winter Wheat; Ra = Tillage Radish; CRP = Conservation Reserve Program</p>		20	S
		13	W or other small grain with or without cover crop following
		3	CRP or Pasture

Table 2.2. Univariate statistics for farm management and sampling continuous variables.

Variable	n	min	Q <sub>0.25</sub>	mean	median	Q <sub>0.75</sub>	max	range	sd	cv (%)	skew	kurt
Sampling Time (Day of Year)	124	112	116	141	133	159	183	71	24	17	0.32	-1.25
No. of Last Fall to Spring Tillage Passes	121	0	1	3.05	3	4	10	10	2.58	85	0.95	0.47
Years Certified Organic	121	0	6	11.84	11	18	27	27	8.22	69	0.07	-1.03
No. of Different Crops*	116	2	3	3.77	3	4	7	5	1.17	31	0.87	0.32
No. of Years with Legume Cover*	123	0	2	2.76	3	3	4	4	0.97	35	-0.95	0.87
No. of Years with Perennial Cover*	123	0	2	2.38	3	3	4	4	1.38	58	-0.77	-0.76
No. of Years with a Winter Cover Crop*	123	0	0	0.89	1	1	5	5	1.03	117	1.24	1.57
No. of Manure Applications*	113	0	2	2.89	2	4	9	9	1.76	61	0.91	0.72

\*Frequency of event over five years prior to sampling.

No. = number; min = minimum; Q<sub>0.25</sub> = 1<sup>st</sup> quartile; Q<sub>0.75</sub> = 3<sup>rd</sup> quartile; max = maximum; sd = standard deviation; cv = coefficient of variation; skew = skewness; kurt = kurtosis

Table 2.3. Categorical farm management variable effects on biological soil health indicators.

Variable	p-value						
	SOM	TOC	TN	POXC	minC	PMN	ACE
Pre-Organic Land History	0.0002	0.003	0.002	<0.0001	NS	NS	NS
Rotation Sequence	0.053	NS	0.0007	0.022	NS	NS	NS
Manure State	0.039	NS	NS	NS	<0.0001	0.003	NS
Crop Prior to Previous Corn Year	NS	NS	0.009	NS	NS	NS	NS
Corn Residue Year Prior	0.005	0.005	NS	NS	NS	NS	NS
Till Type Past 5 Years	NS	NS	NS	NS	NS	NS	0.017
Manure Use Year Prior	NS	NS	NS	NS	0.0003	NS	NS
Cover Crop Year Prior	NS	NS	NS	NS	NS	NS	NS
Till Type Year Prior	NS	NS	NS	NS	NS	NS	NS

NS = not significant; SOM = soil organic matter; TOC = total organic carbon; TN = total nitrogen; POXC = permanganate oxidizable carbon; minC = mineralizable carbon; PMN = potentially mineralizable nitrogen; ACE = autoclaved-citrate extractable protein.



Table 2.4. Continuous farm management variable effects on SOM, TOC, and TN.

Variable	SOM			TOC			TN		
	p	Rel.	R <sup>2</sup>	p	Rel.	R <sup>2</sup>	p	Rel.	R <sup>2</sup>
Sampling Time (Day of Year)	NS			NS			NS		
Years Certified Organic	NS			NS			NS		
No. of Last Fall to Spring Tillage Passes	NS			NS			NS		
No. of Different Crops*	NS			NS			NS		
No. of Years with Legume Cover*	NS			NS			NS		
No. of Years with Perennial Cover*	NS			NS			0.004	+	0.067
No. of Years with a Winter Cover Crop*	NS			NS			NS		
No. of Manure Applications*	NS			NS			NS		

\* Frequency of event over five years prior to sampling.

NS = not significant; SOM = soil organic matter; TOC = total organic carbon; TN = total nitrogen

Table 2.5. Continuous farm management variable effects on POXC, minC, PMN, and ACE.

Variable	POXC			minC			PMN			ACE		
	p	Rel.	R <sup>2</sup>	p	Rel.	R <sup>2</sup>	p	Rel.	R <sup>2</sup>	p	Rel.	R <sup>2</sup>
Sampling Time (Day of Year)	0.049	-	0.032	<0.0001	+	0.20	NS			NS		
Years Certified Organic	0.032	+	0.038	NS			NS			0.031	+	0.038
No. of Last Fall to Spring Tillage Passes	NS		NS	NS			NS			0.027	-	0.04
No. of Different Crops*	0.011	+	0.055	NS			0.013	+	0.053	0.04	+	0.036
No. of Years with Legume Cover*	NS		NS	NS			NS			NS		
No. of Years with Perennial Cover*	NS		NS	NS			NS			NS		
No. of Years with a Winter Cover Crop*	NS		NS	NS			NS			NS		
No. of Manure Applications*	NS		NS	NS			NS			NS		

\* Frequency of event over five years prior to sampling.

NS = not significant; POXC = permanganate oxidizable carbon; minC = mineralizable carbon; PMN = potentially mineralizable nitrogen; ACE = autoclaved-citrate extractable protein

were converted over ten years ago. Despite this temporal distance in system, fields that were previously conventional pastures had higher SOM, TOC, TN, and POXC (Table 2.3 and Figure 2.1 to 2.4). Perennial pasture systems contribute a larger amount of belowground C to the soil compared to cropped land (Radrizzani et al., 2011; Sanford et al., 2012) while also stabilizing OM and C through soil aggregation and cover. Over a 20-year period, soil TOC in the upper 15 cm of Wisconsin Mollisols increased in perennial pasture systems, while other common Midwestern cropping systems lost TOC (Sanford et al., 2012). Identifying novel strategies to incorporate pasture into cropping systems of the Midwest may be a worthwhile approach to improve SOM and TOC content of our agricultural lands. In conventional tillage systems, Salvo et al. 2010 found that crop-pasture rotations (three years of annual crops followed by three years of pasture) led to higher TOC accumulation than rotations solely with annual crops.

Fields that were conventional row crop prior to initiating organic management were primarily in the transition phase and not yet organic certified. These fields along with fields previously enrolled in the Conservation Reserve Program (CRP) had lower TN values than conventional pasture and dairy systems. Since animal waste management is an intrinsic aspect of conventional pasture and dairy systems, fields with these prior uses were more likely to receive manure through application or grazing than conventional row crop systems and CRP. This may contribute to differences in TN values.

### **Rotation and Cropping Sequence**

Total N and POXC were lowest in fields with five years of consecutive annual cropping (Figure 2.3 and 2.4). A positive relationship between perennial cover and TN

reinforced an effect of perenniality (Table 2.4). Increasing system perenniality has been recognized as a strategy to increase biological soil health and SOM accumulation. Specifically, increases in system perenniality led to higher total C, TN, POXC, minC, and PMN in Wisconsin Mollisols (Diederich et al., 2019).

In addition, the cropping sequences and order of crops had an effect on N indicators. Increasing the diversity of plants utilized in five-year cropping sequences increased PMN and ACE (Table 2.4). This reinforces work that found that PMN was higher in rotations containing at least three different crops (Mahal et al., 2018). What crops were planted within the last two years affected TN. Total N was reduced when soybean was planted before corn compared to other crops before corn (Figure 2.3). Despite being an annual legume, soybean provides little N through biological fixation and additional N is required to supplement uptake from high N demanding crops. The differences in TN by crop sequence may also be a result of manure and N management decisions by crop not reflected in other management variables.

### **Duration of Organic Management**

Although greater than three years of organic management was shown to improve soil health (Drinkwater et al., 1995), most indicators were not affected by duration of organic management. Only 3.8% of the variation in POXC and ACE was explained by duration of organic management. Without the use of synthetic fertilizers, organic systems rely on soil microorganisms to process SOM in order to supply fertility (Doran et al., 1987) resulting in a tightly-coupled N cycle with higher turnover rates (Drinkwater et al., 1995). Many comparison studies have found that biological metrics of soil health are improved in

organic systems. Organic management increased SOC (Surekha et al., 2006), TN (Marinari et al. 2005), and PMN (Drinkwater et al., 1995; Doran et al., 1987; Gunapala and Scow, 1997) compared to conventional management. Surprisingly, these indicators did not respond to duration of organic management in our study despite containing fields that spanned zero years (in transition) to 27 years of certified organic management (Table 2.2).

### **Corn Harvest and Residue**

The type of corn harvest and residue management that occurred the year prior to soil sampling affected SOM and TOC (Figure 2.1 and 2.2). Most of the corn was harvested for grain ( $n = 74$ ), but 36 of the fields harvested the stalks for other on-farm uses while 38 of the fields left their stalks remaining as residue (Table 2.1). Snaplage, an alternative feed for dairy cattle made of ensiled corn ears, husks, and shanks, was utilized in two fields that left their stalks as residue (Akins et al., 2008). Fields that harvested corn for silage or left corn stalks as residue had higher SOM and TOC values. Corn silage harvest leaves little residue on the field as the whole corn plant is harvested in the process. Thus, it is unlikely that the same mechanism is responsible for the higher SOM and TOC values in fields harvesting for silage or grain with stalks remaining.

### **Manure Management**

Mineralizable C and PMN positively responded to manure application (Figure 2.4). Fields with manure applied the year prior to sampling had higher minC. When solely liquid manure was applied compared to none or other physical states of manure, minC and PMN values were elevated regardless of the animal(s) that provided the manure. Fields that did not receive manure over the past five years were primarily conventional pasture prior to

their organic transition. Soil organic matter was highest in these fields due to collinearity between previously conventional pasture fields and fields with no manure application. The land use legacy of pasture is likely the overriding factor in SOM values as manure and organic amendment application had been previously shown to increase minC and PMN in a diverse set of cropping systems and geographic regions (Hurisso et al., 2016; Mikha et al., 2006; Sharifi et al., 2008).

### **Tillage Management**

Increasing tillage frequency and intensity reduced N cycling indicators (Table 2.5 and Figure 2.4). The number of tillage passes completed in between the fall and spring before sampling had a negative relationship with ACE, but the relationship only explained 4% of the variation. Minimum tillage fields (fields utilizing a disk, cultivator, or other less disruptive tillage implement) and chisel-tilled fields had higher ACE values than fields that utilized a moldboard plow within the last five years. Previous work has found that PMN and ACE respond positively to reduced tillage systems (Mahal et al., 2018; Doran, 1987; Rasmussen et al., 1998; Nunes et al., 2018; Sandeep et al., 2016).

### **Regression Tree Analysis**

Regression tree analysis utilizing NRCS soil information and farm management information produced trees for SOM, TOC, TN, and minC. No trees were generated for POXC, PMN, and ACE. Soil organic matter and TOC yielded identically pruned trees with a single split at low susceptibility to soil surface sealing (Figure 2.5 and 2.6). According to USDA-NRCS (2008), surface seals and crusts are layers with poor infiltration as a result of blocked or filled surface pores from the destruction of aggregates. Soil surface sealing

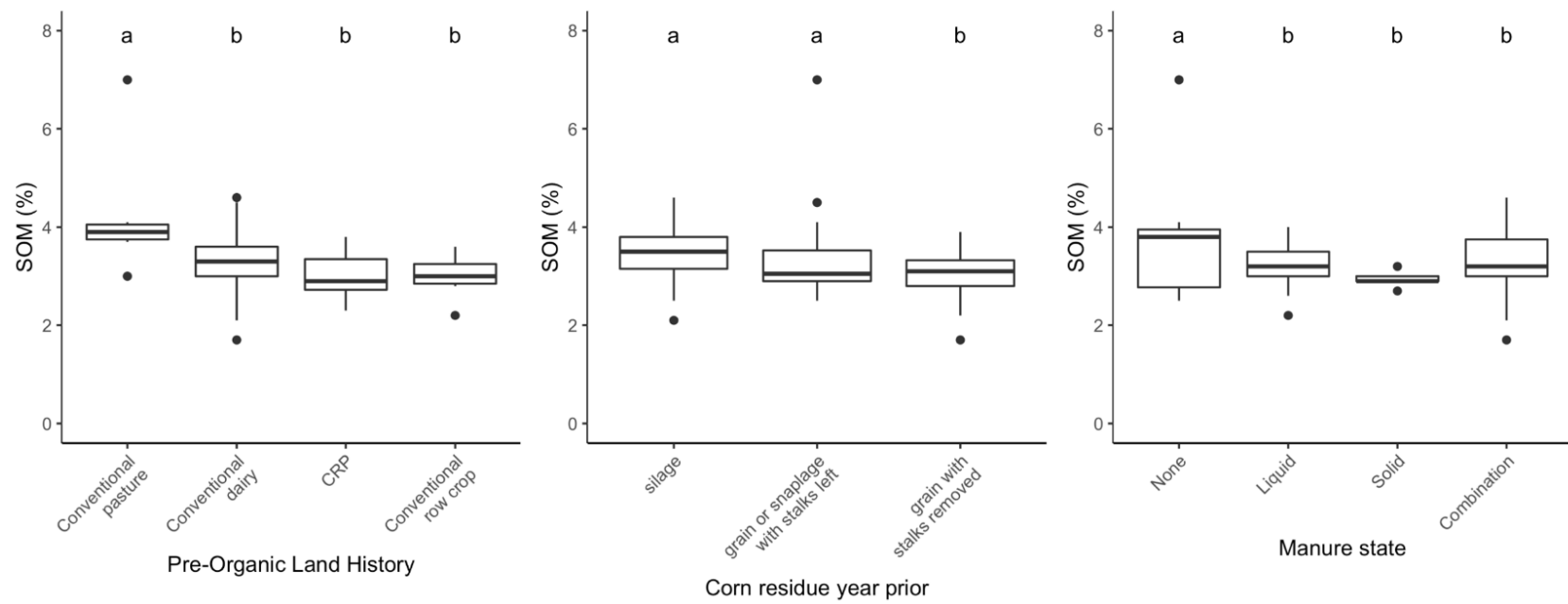


Figure 2.1. Categorical farm management variable effects on soil organic matter (SOM).

CRP = Conservation Reserve Program

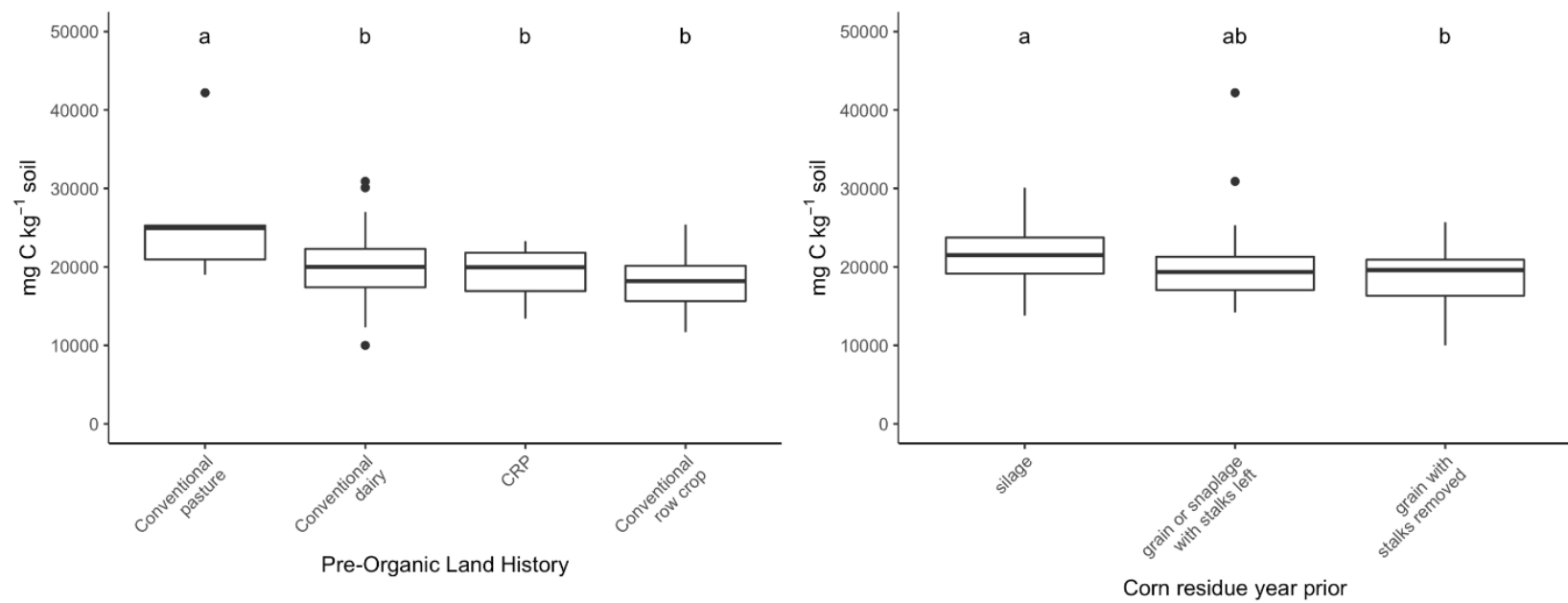


Figure 2.2. Categorical farm management variable effects on total organic carbon (TOC)

CRP = Conservation Reserve Program.

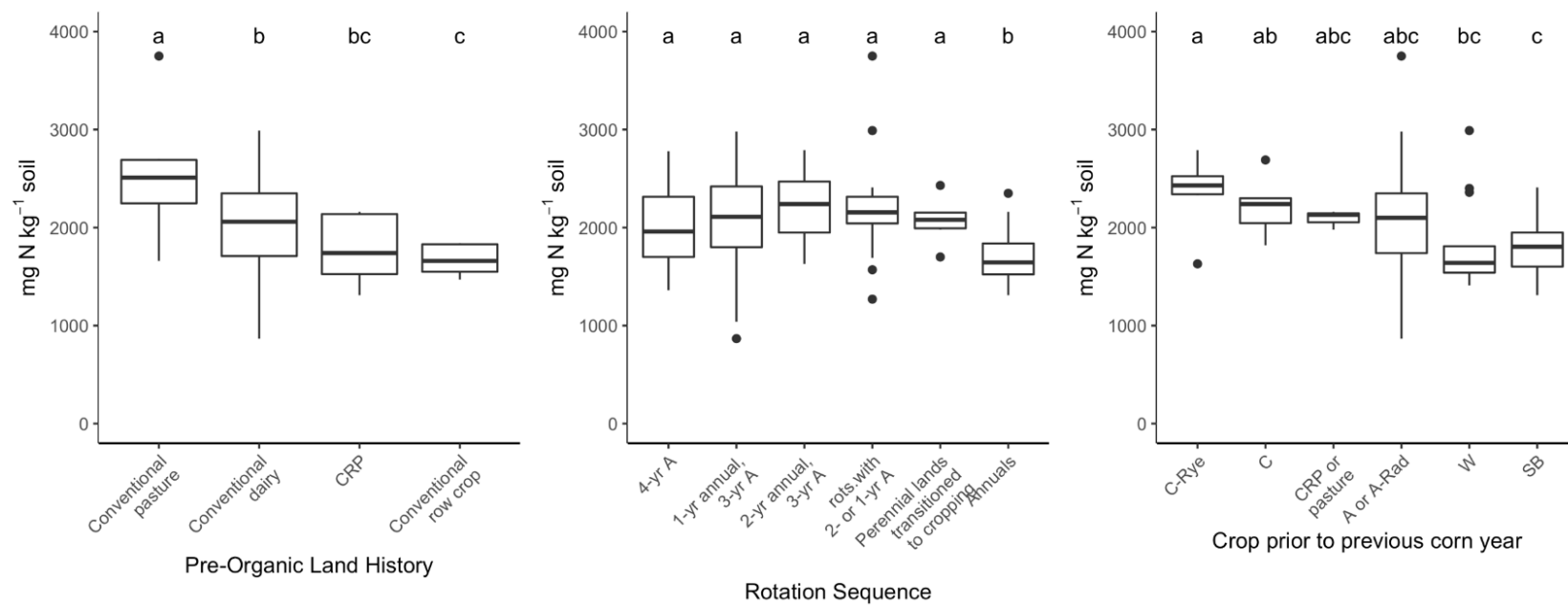


Figure 2.3. Categorical farm management variable effects on total nitrogen (TN).

A = alfalfa; CRP = Conservation Reserve Program; C = corn; Rad = Radish; W = wheat; SB = soybean



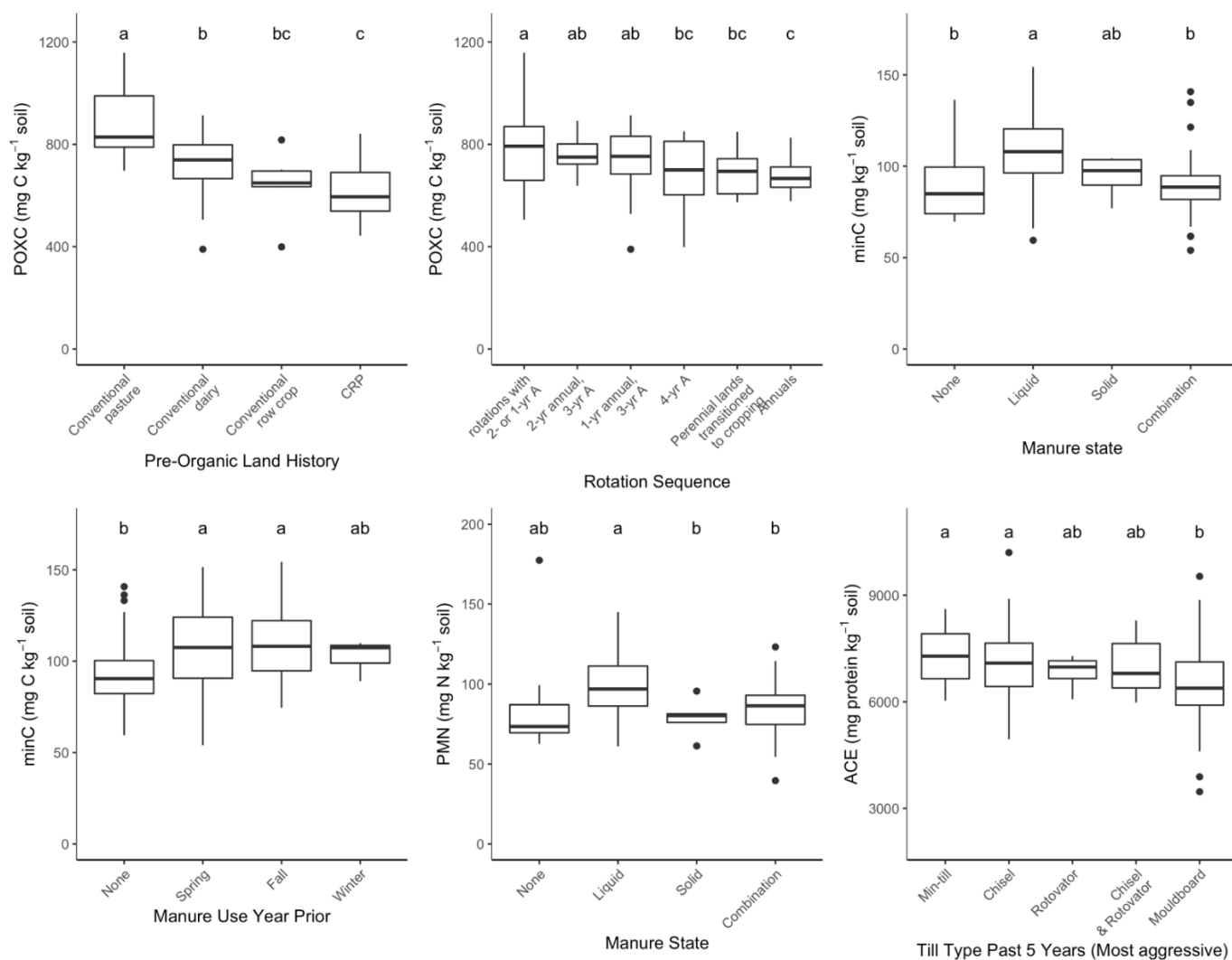


Figure 2.4. Categorical farm management variable effects on biological soil health indicators. A = alfalfa; POXC = permanganate oxidizable carbon; minC = mineralizable carbon; PMN = potentially mineralizable nitrogen; ACE = autoclaved-citrate extractable protein

susceptibility is related to soil texture, OM, and sodium content. Thereby, soil surface sealing intrinsically reflects differences in antecedent OM and TOC content between soils.

In further support of the standardization of sampling times for soil health analysis, minC was highest in samples obtained after May 26<sup>th</sup> (i.e. day 146) (Figure 2.7). Timing of sampling can greatly influence soil health values leading to misinterpretations of soil health status and management needs.

Total N differentiated fields by their inclusion of perennials within the past five years (Figure 2.8); consecutive annual cropping led to lower TN values. Over 70% of fields utilized alfalfa, a perennial forage, which replenishes soil N through biological fixation. Alfalfa can meet the N demands of first-year corn when sufficient stand plant density is met (Laboski and Peters, 2012), and older stands of alfalfa typically provide larger N credits than younger stands (Yost et al., 2018). Although many annually cropped fields incorporated soybean, the amount of N provided is over 7-times smaller than that of a good alfalfa stand. Thereby, annual cropping systems are more susceptible to soil TN depletion following corn if organic amendments do not supplement N uptake.

Regression trees were able to supply nuanced information despite containing only a single branch. The tree for minC explained more variation with its single primary split than simple linear regression of sampling time and minC (Figure 2.7). Regression trees are also able to indicate the amount of variation in indicator values explained by categorical primary variables through  $R^2$  values while ANOVA is unable to provide this information. The  $R^2$  for each tree was 0.24 for SOM and TOC, 0.15 for PMN, and 0.26 for minC. Regression tree analysis has the ability to identify linear and non-linear effects, thereby providing better

$$R^2 = 0.24$$

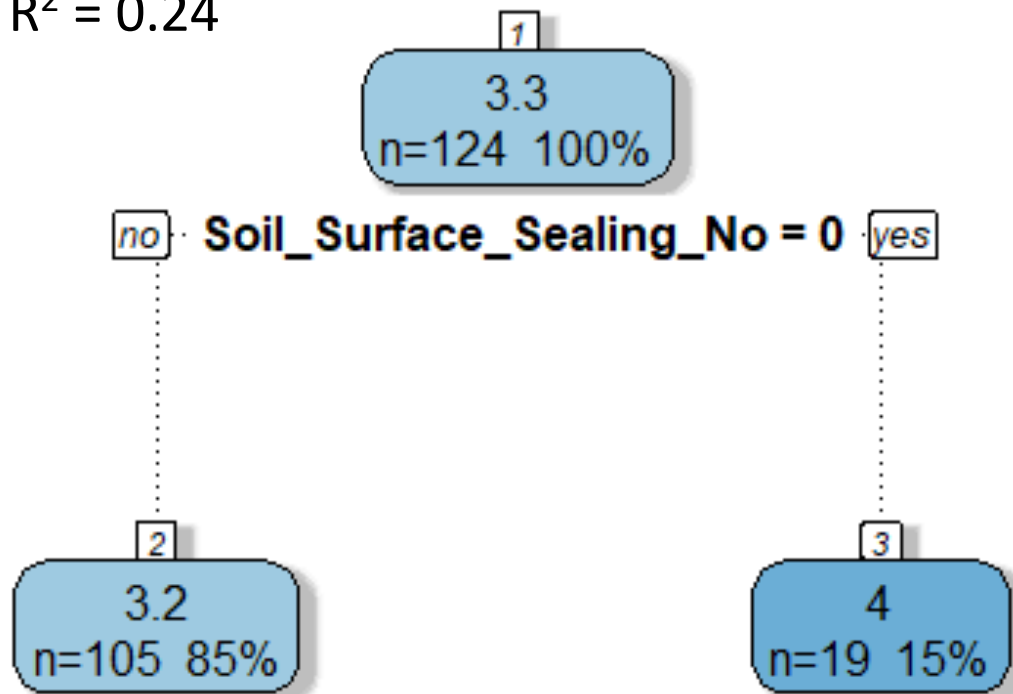


Figure 2.5. Pruned regression tree for soil organic matter (SOM) percentage.

Within each node, the top number is the average SOM value for soils in that node. The lower-left number is the number of fields in that node, and the lower-right number is the percentage of total fields in that node. Darker shading indicates higher SOM values. Soil\_Surface\_Sealing\_No (soil surface sealing): 0 = low; 1 = moderate; 2 = high.

$$R^2 = 0.24$$

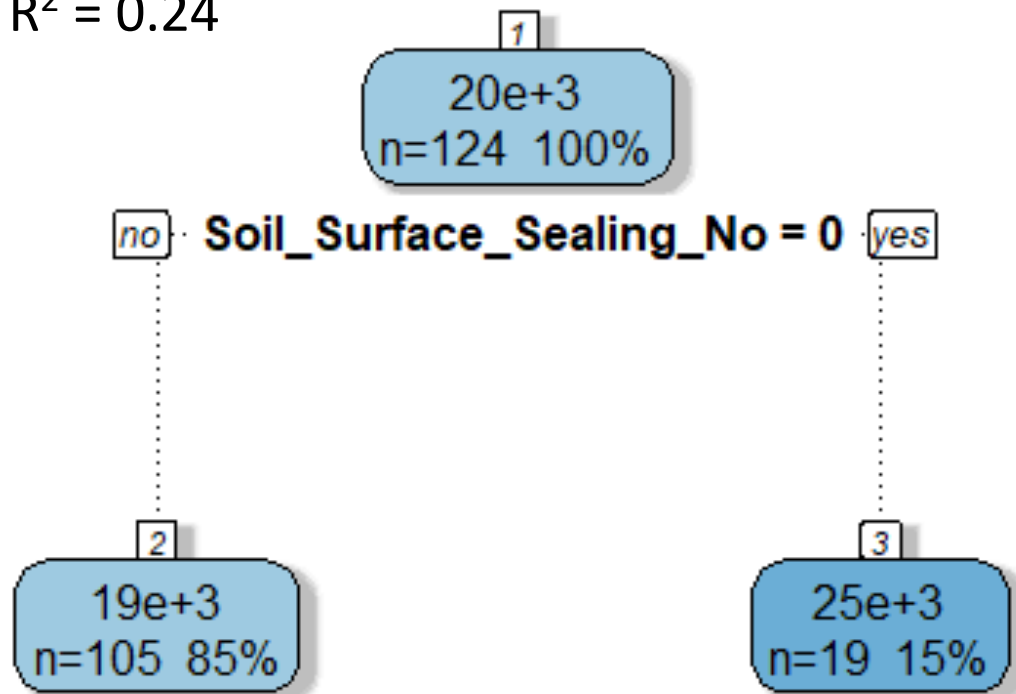


Figure 2.6. Pruned regression tree for total organic carbon (TOC) (mg C kg<sup>-1</sup> soil).

Within each node, the top number is the average TOC value for soils in that node. The lower-left number is the number of fields in that node, and the lower-right number is the percentage of total fields in that node. Darker shading indicates higher TOC values. Soil\_Surface\_Sealing\_No (soil surface sealing): 0 = low; 1 = moderate; 2 = high.

$$R^2 = 0.26$$

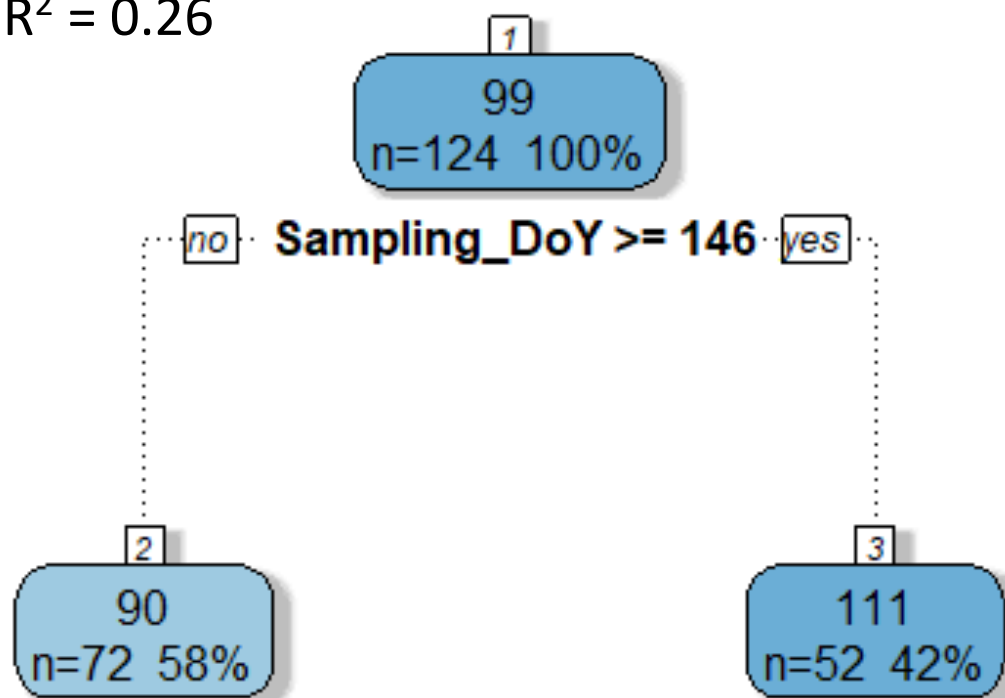


Figure 2.7. Pruned regression tree for mineralizable carbon (minC) (mg C kg<sup>-1</sup> soil day<sup>-1</sup>).

Within each node, the top number is the average minC value for soils in that node. The lower-left number is the number of fields in that node, and the lower-right number is the percentage of total fields in that node. Darker shading indicates higher minC values. Sampling\_DoY = day of year when soil samples were obtained.

$$R^2 = 0.15$$

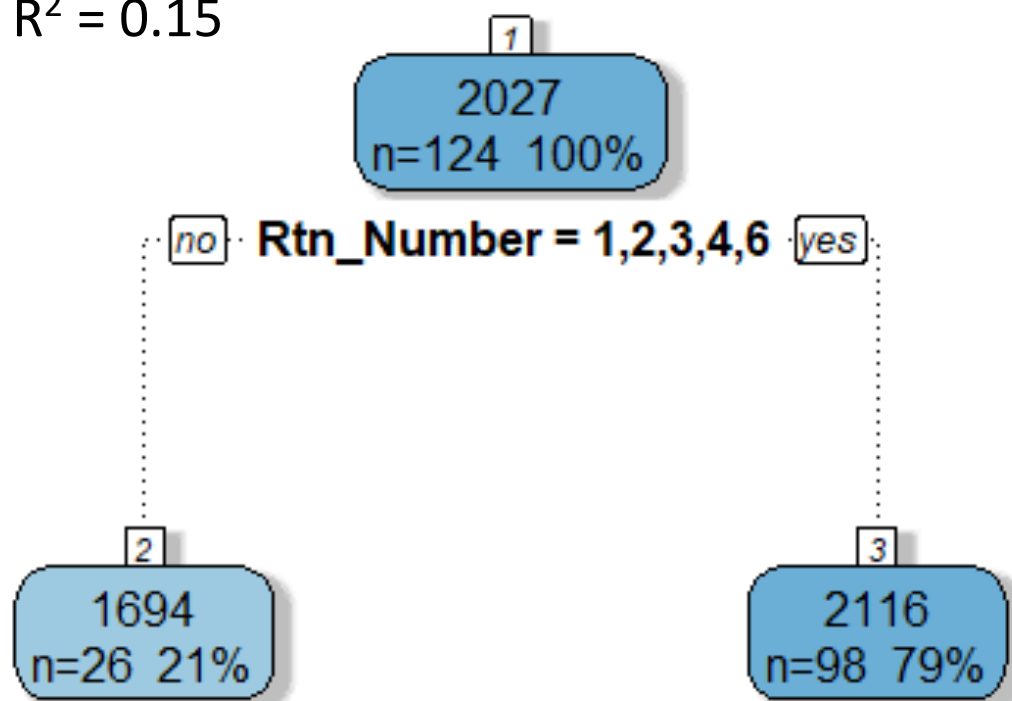


Figure 2.8. Pruned regression tree for total nitrogen (TN) ( $\text{mg N kg}^{-1}$  soil).

Within each node, the top number is the average TN value for soils in that node. The lower-left number is the number of fields in that node, and the lower-right number is the percentage of total fields in that node. Darker shading indicates higher TN values. Rtn\_Number (rotation sequence): 1 = 4-yr alfalfa (A); 2 = 1-yr annual following 3-yr A; 3 = 2-yr annual following 3-yr A; 4 = rotations with 2-yr or 1-yr A; 5 = annuals; 6 = perennial lands transitioning to cropping.

descriptions of relationships between indicators and explanatory variables than simple linear regression. Unfortunately, the ability is limited to variables solely included in the pruned trees.

Regression trees were produced using the variable that best segregated fields by their soil health values. Variable importance, a measure of a variable frequency as a primary and surrogate split while accounting for “goodness of split”, was highest for the variables selected for inclusion in indicator regression trees (Table 2.6). The best primary variable had improvement ratings 1.23 to 1.69 times greater than the variable with the next highest improvement rating (Table 2.7). Improvement is the difference in sum of squared errors between parent and child nodes, and is a measure of decreased impurity (i.e. increased homogeneity) (Therneau and Atkinson, 2019). For the TN regression tree, five-year rotation sequence and the number of years with perennial cover had equal importance and improvement values due to their identical splitting pattern; they split according to whether the past five years of cropping were all annual crops or not.

Surrogate variable agreement was sufficient for handling missing observations in primary variables. Agreement is the proportion of observations sent the correct direction when using a surrogate variable in place of the primary variable for an observation with missing data (Therneau and Atkinson, 2019). Besides minC, the best performing surrogate variable of each regression tree had agreement values greater than 94%. Mineralizable C utilized sampling time as its primary variable, which has no missing observations and does not require a surrogate split for regression tree formation. Therefore, all of the surrogate variables utilized in tree formation were able to accurately direct observations to the

Table 2.6. The top five most important variables (variable importance scores) by soil health indicator.

Rank	SOM	TOC	TN	minC
1	Surface Sealing (29)	Surface Sealing (29)	Rotation Sequence (25)	Sampling Time (41)
2	Parent Material (18)	Parent Material (18)	No. of Years with Perennial Cover* (25)	Manure State (16)
3	Depth of A Horizon (14)	Depth of A Horizon (14)	Crop Prior to Previous Corn Year (17)	pH (13)
4	Sand (14)	Sand (14)	No. of Years with Legume Cover* (15)	No. of Last Fall to Spring Tillage Passes (12)
5	Clay (14)	Clay (14)	Years Certified Organic (10)	Manure Use Year Prior (10)

\* Frequency of event over five years prior to sampling.

SOM = soil organic matter; TOC = total organic carbon; TN = total nitrogen; minC = mineralizable carbon; No. = number



Table 2.7. The top three primary and surrogate splits by soil health indicator.

Indicator	Primary Splits	Root to Leaf Node Improvement	Surrogate Splits	Agreement
SOM	Surface Sealing	0.24	Parent Material	0.943
	Subgroup	0.195	Sand or Depth of A Horizon	0.919
	Whole Soil Erodibility Factor, Suborder, or Great Group	0.158	Clay or Water Content at 1/3-bar	0.911
TOC	Surface Sealing	0.240	Parent Material	0.943
	Subgroup	0.142	Sand or Depth of A Horizon	0.919
	NCCPI Corn	0.138	Clay or Water Content at 1/3-bar	0.911
TN	Rotation Sequence	0.149	No. of Years with Perennial Cover*	1.00
	No. of Years with Perennial Cover*	0.149	Crop Prior to Previous Corn Year	0.927
	Crop Prior to Previous Corn Year	0.097	No. of Years with Legume Cover*	0.911
minC	Sampling Time	0.256	Manure State	0.742
	Manure State	0.191	pH	0.718
	Manure Use Year Prior	0.142	No. of Last Fall to Spring Tillage Passes*	0.702

\* Frequency of event over five years prior to sampling.

SOM = soil organic matter; TOC = total organic carbon; TN = total nitrogen; minC = mineralizable carbon; No. = number

appropriate nodes and produce informative trees for assessing relative effects on indicator values. Future research that aggregates soil health data to form larger regional and national datasets would be beneficial for developing soil health assessments by increasing the likelihood to identify important effects and interactions with regression tree analysis.

## **CONCLUSIONS**

While many inherent soil properties contribute to differences in soil health, farm management practices can improve or weaken the biological health of soils under agricultural production. Legacy land use and cropping sequences that utilized perennial cover had higher SOM, TOC, and TN. Nitrogen-cycling indicators were more affected by cropping sequence, crop diversity, and tillage. The incubation methods, minC and PMN, were influenced by the use of manure in the last year and the physical state of the manure. Mineralizable C was highly affected by sampling time and requires standardization of a sampling window for soil health comparisons. The disparate response of indicators to management indicates differences in sensitivity between indicators as well as soil elemental cycles. Therefore, multiple shifts in management may be required to support both healthy C and N cycling in agricultural systems. These results provide first-indications of best management practices for biological soil health in organic grain systems.

Overall, regression tree analysis produced trees for over half of the indicators. Perennial cover within rotation sequences was most important for differentiating TN while susceptibility to soil surface sealing was most important for differentiating SOM and TOC. Indicator regression trees were able to explain more variation in indicator values than simple linear regression and ANOVA for the explanatory variables included. Regression trees

are able to provide  $R^2$  values for categorical primary variables as well as capture linear and non-linear effects to generate better descriptions of indicator relationships with inherent soil properties and management practices. Despite minimizing variation in climate, geographical region, and production system, a higher number of field observations may be required to increase the likelihood of identifying important effects and explain more of the variation between field soil health values. Regardless, regression tree analysis was a useful tool for exploring the relative importance of management and inherent soil properties on farms as well as evaluating soil health indicator sensitivity and limitations.

Further research should coordinate larger studies or aggregate existing soil health data from long-term research trials and on-farm studies to improve detection and understanding of soil property and management effects on soil health. Identifying specific best management practices is crucial to improving nutrient use efficiency in agricultural production, the provision of ecosystem services, and the development of guidelines and policies to ensure it.

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