

Evaluating the Effects of Long-Term No-Till and Crop Rotations in Soil Health and Corn
Productivity

Thesis

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By

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Abstract

Conservation practices, such as no-till and diversifying crop rotations are known for their capacity to reduce soil erosion and improve soil properties. However, the impact of these management practices on emerging soil health tests and the ability of these tests to reflect active organic matter dynamics and nutrient cycling, and corn productivity have not been explored. This project focused on determining the effects of half a century of continuous tillage treatments (moldboard plow, chisel till, and no-till) and crop rotations (continuous corn, corn-soybean, and corn-forage-forage) on soil health indicators and its relationship with crop productivity. The forages were alfalfa in Wooster, and red clover and oats in Northwest. Soil labile carbon (C) and nitrogen (N) temporal dynamics were quantified with permanganate oxidizable C (POXC), mineralizable carbon (Min C), and soil protein at six key stages in corn (*Zea mays*) development: before planting (around three weeks before planting), V5, V10, R1, R4, R6 in the 2017 and 2018 growing seasons. Corn leaf chlorophyll, aboveground plant biomass, nutrient uptake, and grain yield were also quantified. The soil health indicators (POXC, Min C, soil protein) and crop parameters (leaf chlorophyll, total nitrogen uptake and total aboveground biomass) were higher in reduced tillage (chisel and no-till) compared to moldboard plow and higher in the most diverse crop rotation (corn-forage-forage) compared to corn-soybean. Corn yields were not significantly different between tillage treatments but were higher in

the more diverse rotations (corn-soybean and corn-forage-forage) compared to corn monoculture. Although the treatment effects varied by site and year, rotation had a consistently larger effect on soil health indicators and corn productivity than tillage, highlighting the importance of including crop rotations in corn production. We conclude that Ohio soils under half a century of continuous tillage and rotation treatments have higher soil health and corn productivity in no-till and reduced tillage soils compared to moldboard plow. We also conclude that soil health indicators and maize productivity were higher in rotations including two years of forages compared to monoculture and corn-soybean rotations, especially in no-till soils.

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Chapter 1. Evaluating the effects of long-term no-till and crop rotations on soil health

1.1 Introduction

Soil Health, Tillage, and Crop Rotations

Soil health and quality are increasingly topics of interest by scientists, farmers and the general community. With rapid population increase and drastically changing climate, maintaining good quality soils is essential for today's and the future's sustainable agriculture. The USDA NRCS defines soil health as, 'the capacity of soil to function as a vital living ecosystem that sustains plants, animals, and humans.' Therefore, a healthy soil is believed to be productive and resilient to unfavorable conditions (Doran and Parkin, 1994; Kibblewhite et al., 2008; Roper et al., 2017).

Soil health encompasses soil physical, chemical and biological properties together because these aspects often influence one other in the soil ecosystem. For instance, the loss or depletion of soil organic matter (SOM) can reduce the soil aggregation, water infiltration, moisture and nutrient content, availability and cycling (Martínez et al., 2016; Alhameid et al., 2017). In fact, SOM is an essential component for studying soil quality because of its impact on soil productivity (ability to sustain crops sustainably). For instance, research has shown that soil productivity is reduced with soil organic matter loss even when soils are well fertilized (Aref and Wander, 1998; Oldfield et al., 2019).

Although the total soil organic matter is a good indicator of soil quality (Gregorich et al., 1994; Haynes, 2005; Wander and Drinkwater, 2000), it is the active pool (5-20%) of the total soil organic matter that functions as a better index of studying soil rapid response and changes to management like nutrient cycling and availability, soil aggregation, and potential soil carbon accumulation or loss (Cambardella and Elliott, 1992; Grandy and Robertson, 2007; Schmidt et al., 2011; Six et al., 1998; Weil and Magdoff, 2004; Wander, 2004; Haynes, 2005; Lewis, Kaye, Jabbour, & Barbercheck, 2011). Studying the soil labile organic matter pools may help us obtain a better understanding of rapid soil response or changes by soil management in long-term managed agricultural soils and improve management recommendations.

The soil health concept focuses on the soil dynamic quality, that is, the properties that change as a result of soil use and management by humans (Fine et al., 2016) The USDA NRCS considers the following management practices to improve soil health: reducing soil disturbance, increasing plant diversity, keeping soils covered year-round, and maintaining soils with living plants. Many soil conservation practices have been shown to improve overall soil quality compared to conventional practices. Two of the most common soil health-building practices are reduced tillage intensity (Melero et al., 2009; Karlen et al., 2013a; Lal, 2015; Nunes et al., 2018), and diverse crop rotations (Altieri, 1999; Karlen et al., 2006, 2013a; Mcdaniel et al., 2014).

For many decades, moldboard plow has been a successful tool in agriculture, especially in organic farming, for controlling pests, weeds, and incorporating the vegetative residues as it inverts the soil to a depth of 20 to 30 cm. It has also been used in

temperate regions for stimulating soil warming after the cold spring and improving soil conditions for seed emergence. However, the long-term effects of intensive tillage can promote higher soil erosion (Choudhary et al., 1997; Reicosky et al., 2011; Baumhardt et al., 2015), and increase soil compaction in the subsurface (Kinoshita et al., 2017) compared to less intensive tillage or no-till practices. After the Dust Bowl in the USA and the development of herbicides, reduced tillage and no-tillage adoptions have increase as a way to reduce soil erosion and degradation.

Chisel and no-till are examples of reduced tillage, also known as conservation tillage. The chisel till consists of incorporating part of the residues and leaving around 30% of these residues on the soil surface and the no-till consists of zero soil disturbance, leaving all crop residues on the soil surface. Previous research has shown that long-term chisel till and no-till produce less soil erosion compared to conventional plow (Choudhary et al., 1997). Recent research has shown that long-term no-till resulted in higher soil quality in diverse soils of New York compared to moldboard plow (Nunes et al., 2018).

Adding crop rotations in agronomic farming is another soil conservation management tool that can improve the soil health. The diversification of roots and vegetative residues may promote soil microbial diversity and activity, and nutrient cycling (Mcdaniel et al., 2014). Continuous long-term monoculture exposes the soil to non-diverse root type, depth, and density, and may decrease the availability of nutrients in the soil and increase the risk of pests and diseases attacks to crops (USDA NRCS a).

In the US Midwest, the most common agronomic rotation is a two-year corn-soybean rotation. Soybeans have the ability to fix nitrogen in the soil and therefore reduce the requirement and application of nitrogen fertilization. Previous research has shown higher yields in corn under a corn-soybean rotation compared to corn monoculture (Karlen et al., 2013b). Alfalfa and red clover are other perennial legumes that can fix nitrogen in the soil, often resulting in higher soil quality over corn monoculture in the US Midwest (Karlen et al., 2006) and in Ontario, Canada (Congreves et al., 2015).

Many studies recommend the long-term implementation of no-till or reduced tillage and crop rotations for increasing soil quality/health in temperate agricultural soils (Karlen et al., 2013b; Congreves et al., 2015; Nunes et al., 2018). However, many of these studies have evaluated soil C and N under different tillage intensities and crop rotations but have utilized methods that do not measure the labile fractions, and the other studies that do measure the labile fractions utilize expensive and/or more time-consuming methods. Some examples of these relatively expensive and time-consuming methods that estimate labile C and N pools are particulate organic matter (POM) or particulate organic C (POC) (Bongiorno et al., 2019), carbon-metabolizing enzymes like β -glucosidase, microbial biomass nitrogen (MBN), potentially mineralizable nitrogen (PMN)(Mahal et al., 2018), water soluble or water extractable nitrogen (WEOC-N). Many studies have utilized these methods to measure or estimate soil changes or responses to management, especially nutrient cycling in agroecosystems.

Ohio No-Till sites C and N studies Overview

The Triplett – Van Doren No-tillage Experimental Plots were established in the early 1960's to evaluate the long-term impact of different tillage intensities and crop rotations on soil properties and crop productivity (Dick et al., 2013) in Ohio, USA. To date, there has been a number of studies that have taken advantage of the unique treatment design and long-term nature to address agroecological and agronomic research questions. In this section we will focus on studies in these plots that have evaluated soil carbon and nitrogen.

Research conducted after 18 and 19 years of no-till, chisel till and moldboard plow in the Northwest and Wooster sites showed that organic C and N concentrations were higher in the soil surface (0-15cm) of the no-till soils compared to tilled soils (Dick, 1983). Soil type (clay content) influenced the changes in organic C and N concentrations in soil profiles where no-till practices had been maintained. When compared to the beginning of this experiment, results after 18 and 19 years of no-tillage showed that organic C concentrations remained constant in Northwest and decreased 11% in Wooster. On the chisel till and moldboard plowed soils, organic C decreased 12 and 14% respectively in Northwest and decreased 23 and 25 % in Wooster (Dick, 1983).

Another study conducted by Dick et al (1986) showed that biological activity and enzyme activities were higher in the no-till soils, especially in the surface layer and under the more diverse rotations, compared to conventional tillage. Also, this research showed that changes in soil properties were more rapid in the first ten years of the study by

comparing data from 1980 to 1971, and that the more residue returned, the greater the soil organic carbon content (Dick et al., 1986).

In 1991, Dick et al (1991) reported that no-till sites showed stratification of organic matter, nutrients, and soil enzymes relative to chisel tilled or moldboard plowed plots (Dick et al., 1991). Another research conducted after 28 years of continuous treatments showed significant effects of tillage, rotation and their interactions on soil organic carbon in Wooster. Also, organic C was higher for the continuous corn rotation compared to more diverse rotations (Lal et al., 1994).

Results of recent study results showed that no-till treatment increased C and N stock in the top 20 cm in Northwest, but there were no differences observed at the Wooster site (Burgos Hernández et al., 2019). A historical analysis of organic carbon in the no-till treatment in Wooster showed no significant increase in the top 10 cm from 1993-2013 in no-till soils; however, organic C did increase in the 10–30 cm depth over that 20 year period in no-till soils (Dick et al., 2013) (Burgos Hernández et al., 2019).

Our study proposes to evaluate soil active carbon and nitrogen pools with methodologies that are fast, easy, inexpensive, and sensible to management like POXC (Weil et al., 2003; Culman et al., 2012), soil protein (Hurisso et al., 2018b), and soil C mineralization (Franzluebbers et al., 2000; Haney et al., 2001). We aim to evaluate these labile C and N pools throughout corn growing seasons to better understand C and N seasonal dynamics and nutrient cycling on soils in one of the longest tillage and crop rotations experimental sites of America, the Triplett and Van-Doren No-tillage Experimental plots at two sites in Ohio, USA. Specifically, we aimed to (i) evaluate the

effects of different tillage intensities (moldboard plow, chisel till, and no-till) and crop rotation diversity (corn monoculture, corn-soybean, and corn-forage-forage) on labile C and N soil health indicators after 55+ years and (ii) examine below ground C and N temporal dynamics over a corn (*Zea mays*) growing season under these long-term management histories in soils of Ohio, USA. We hypothesize that both crop rotation and no-till practices will significantly impact soil health properties. For instance, we expect that soil health will be impacted primarily by tillage, with the no-till having greater soil health and moldboard plow having the lowest soil health. We also expect that soil health will be strongly influenced by the rotation, with the 3-year rotation having greater soil health and continuous corn the least favorable soil health. Finally, we expect to observe seasonal variation of soil health indicators throughout the corn growing season.

1.2 Materials and Methods

Sites Description

This study was conducted in the Triplett-Van Doren Long-Term Tillage and Crop Rotation Trials of The Ohio State University, located at Wooster and Hoytville Ohio, United States. A description of the soil series in each site is provided in Table 1. The Wooster site (established in 1962) is located in the northeast region of Ohio on a Wooster silt loam. This silt loam is a well-drained deep soil with a slope range of 2 to 6 percent and a low or no shrink-swell potential (Soil Survey Staff, 2019a). The Hoytville site (established in 1963) is located in the northwest region of Ohio on a Hoytville clay loam. This clay loam is a very deep and poorly drained soil with a slope range of 0 to 1 percent and high shrink-swell potential (Soil Survey Staff, 2019b). Subsurface tile drainage was installed in 1952 in the Hoytville site to improve the soil drainage (Dick et al., 2013).

Table 1. 1. Description of soils in the Northwest and Wooster sites

Sites	Location	Series	Taxonomic Class
Northwest	Custar, Wood County, OH. (41°13' N, 83°45' W)	Hoytville Clay Loam	Fine, illitic, mesic Mollic Epiaqualfs
Wooster	Wooster, Wayne County, OH. (40°45' N, 81°54' W)	Wooster Silt Loam	Fine-loamy, mixed, active, mesic Oxyaquic Fragiudalfs

Experimental Design and Treatments

Both sites were arranged as a full factorial, randomized complete block design with three intensities of tillage and three intensities of crop rotations, with three replicates each. The tillage treatments were i) no-tillage (NT) (lowest intensity), ii) chisel tillage (CH), and iii) moldboard plow (MP) (highest intensity). The NT consists of zero tillage and the residue from the previous year crop was left on the field. Tillage in the CH and MP treatments was done during the spring or previous fall each year. The CH treatment left approximately 30 percent of the residues from the previous crop. The MP treatment inverted the soil to a depth of 20-25 cm, incorporating the majority of the residues. The crop rotations were i) continuous corn (*Zea mays*) (CC), ii) corn-soybean (*Glycine max*) (CS) two-year rotation, and iii) a three-year corn-forage-forage (CFF) rotation in both sites. The forage crops in the 3-year rotation consist of oats (*Avena sativa*)-red clover (*Trifolium pretense*) in Northwest and alfalfa (*Medicago sativa*) in Wooster. Plots were 22.3 m by 4.3 m at the Wooster site and 30.5 m by 6.4 m at the Hoytville site. Corn was planted between mid and late May for both sites and years. All weather data was provided by the Ohio Agricultural Research and Development Center (OARDC) weather system. Weather data presented is from years 2017, 2018 and the average of from 2009 to 2018 (10-year average) growing seasons.

Plots Fertility Management

Starter Fertilizer was applied in the corn plots (34-45 kg ha⁻¹) during the pre-plant stage in spring. For both sites, plots under the CS and CFF were fertilized during the V5 corn stage with 202 kg ha⁻¹ of nitrogen only in plots where corn is planted. In the CC rotations, 235 kg ha⁻¹ of nitrogen were applied during the corn V5 stage every year. Soil phosphorus and potassium were applied as necessary depending on soil tests results.

Soil Samplings

For this study, the corn phase of all nine treatments (3 tillage treatments over 3 rotations) were sampled. Soils were sampled to a depth of 20 cm during 2017 and 2018 growing seasons using a soil probe (2.5cm diameter core) in the ¼ row position (around 15 cm) from the corn rows (around 15 centimeters apart from the corn plants). Soils were sampled at 6 key corn growth stages during each growing season (Table 1.2). These corn growth stages mark different development processes in the plant that require different water and nutrient use that may impact belowground dynamics. Eight cores per plot were taken from the pre-plant stage and six cores per plot were taken for the rest of the stages across the entire plot. Soil samples were composited per plot, field-moist samples were sieved to 8 mm and mixed until homogeneous. The soil samples were air dried for seven

days in 2017 and dried on a desiccator at 40°C for 2 days on 2018. After dried, soil

Sampling	Plant Stage	--Northwest Site--		--Wooster Site--	
		2017	2018	2017	2018
Pre-Plant	-	May 3	May 8	Apr 28	May 7
Fifth collared leaf	V5	Jun 21	Jun 19	Jun 27	Jun 18
Tenth collared leaf	V10	Jul 19	July 18	Jul 26	Jul 19
Silking	R1	Aug 8	Aug 2	Aug 9	Aug 1
Dough	R4	Aug 31	Aug 30	Sep 7	Sep 5
Physiological Maturity	R6	Oct 2	Oct 15	Oct 16	Oct 4

samples were ground and sieved to 2mm.

Table 1. 2. Soil samplings corresponding corn development stages during the 2017 and 2018 growing seasons.

Soil Labile C and N Pools

Rapid soil analyses focusing on labile soil C and N pools were quantified on all sampled soils over the two-year study.

Permanganate oxidizable carbon (POXC) was measured following the methods described by Weil et al. (2003) with minor modifications described in Culman et al. (2012). For each sample 2.5 g of soil was reacted with 20 mL of a 0.02 M potassium permanganate (KMnO₄) solution in 50 mL centrifuge tubes. The tube was shaken horizontally for two minutes (180 strokes per minute), allowed to settle vertically for 10 minutes and then 0.5 mL of supernatant was transferred into a second 50 mL centrifuge tube and mixed with 49.5 mL of deionized water. Finally, the sample absorbance was read in a 96 well plate reader spectrophotometer at 550 nm

Soil microbial respiration or mineralizable carbon upon rewetting was measured following the Franzluebbers et al. (2000), Haney et al. (2001) and Franzluebbers et al. (2016) methods. The 50% water holding capacity was determined empirically based on difference in weight between 10g of dried and sieved (2mm) soil samples that were saturated with deionized water and allowed to drain for 35 minutes, and the weight after drying overnight in an oven at 105°C. Briefly, 10 g of 2 mm ground, dry soil soil was rewetting to 50% water-holding capacity with deionized water in a 50 mL polypropylene centrifuge tube. After the rewetting of soils, tubes were capped tightly with lids containing rubber septum, sealed with parafilm and incubated at 25°C for 24 hours. After the 24 hours of incubation, 1 mL of headspace air was extracted per sample using a

syringe and injected into a LI-820 infrared gas analyzer (LI-COR, Biosciences, Lincoln, NE) to determine the concentration of CO₂. Finally, mineralizable C was calculated as the difference between a sample and a blank control using the ideal gas law (Zibilske, 1994) and the headspace volume.

Soil protein was extracted using a sodium citrate buffer (pH 7) following Hurisso et al. (2018). Twenty-four mL of 0.02 mol L⁻¹ sodium citrate solution was added to 3 g soil in a 50 mL glass centrifuge tube. Samples were shaken horizontally for 5 minutes (180 strokes per minute) and autoclaved for 30 minutes at 121°C. Samples were left to cool to room temperature for approximately 40 minutes and shaken horizontally for 3 minutes at 180 strokes per minute. After shaking, 1.5 mL were transferred to 2 mL centrifuge tubes and centrifugated at 10,000 x g for 3 minutes. Quantification of protein was done by using colorimetric bicinchoninic-acid (BCA) assay (Thermo Scientific, Pierce, Rockford, IL) in a 96-well spectrophotometric plate reader at 562nm. The sample absorbance readings were calibrated using a standard curve of bovine serum albumin. Autoclaved citrate extractable soil protein content was calculated as the protein concentration in samples multiplied by the volume of the extractant used and divided by the weight of the soil.

Other Soil Measurements

Soil moisture was determined gravimetrically at all stages from the fresh composite soil samples for both years by oven drying 10 g of fresh soils (8mm sieved)

and measuring mass loss. Routine soil nutrient and chemical properties analysis was conducted on samples on the first sampling (2017 Pre-plant) by a commercial laboratory (Spectrum Analytics, OH).. These measurements included soil organic matter by loss on ignition in a muffle furnace at 360 °C for two hours (Combs and Nathan, 1998), soil pH by a glass electrode in a 1:1 soil:water mixture (Peters et al. 2012), extractable P and K by Mehlich-3 extraction (Mehlich, 1984) and analyzed with inductively coupled plasma spectrometer (Vanhaecke et al., 1998). CEC was estimated by summation of base cations (Ca^{2+} , Mg^{2+} , and K^{+})(CECsum). Finally, soil penetration resistance was measured to a depth of 20 cm at stage V5 using a SpotOn® Digital Soil Compaction Meter (Innoquest, Inc., Woodstock, IL).

Statistical Analyses

Analysis of variance was performed for each site and year combination (i) for each individual stage, and (ii) for all stages sampled in a growing season using stage as a repeated measure, with the PROC MIXED procedure in SAS v.9 (SAS Institute, Cary, NC). Tillage and crop rotation were treated as fixed effects and rep (block) as a random effect with significant differences determined at $\alpha = 0.1$. Means separations were performed with an adjusted Tukey's pairwise comparison ($\alpha = 0.1$) using SAS v.9. Graphs were created using the ggplot2 (Wickham, 2009) package in RStudio (RStudio Team, 2016).

1.3 Results

Weather

At Northwest, monthly precipitation during 2017 was above the 10-year average from May to August and below average during September. In 2018, monthly precipitation was below the 10-year average from April to July in Northwest (Table 1.3; Figure 1.1). In Wooster, monthly precipitation in 2017 and 2018 were relatively close to the 10-year average values in except for the months of August and September of 2017 where rainfall in 2017 was drastically below the average, and September 2018 was above average (Table 1.3; Figure 1.1). Overall, monthly temperatures for both sites and years were similar to the 10-year average temperatures (Table 1.4)

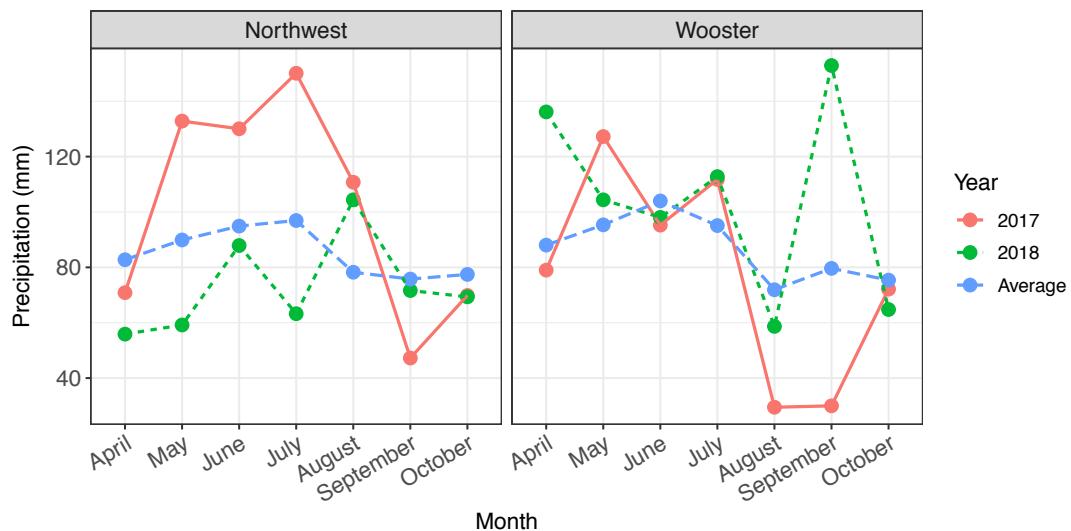


Figure 1. 1. Monthly precipitation for the Northwest and Wooster sites during the 2017 (red solid line) and 2018 (green dashed line) growing seasons and the past 10-year average (blue dotted line).

Table 1. 3. Monthly precipitation during 2017 and 2018 growing seasons and for the past 10-year average at the Wooster and the Northwest sites.

Site	Year	Sum of monthly Precipitation (Apr-Oct)							
		Apr	May	Jun	Jul	Aug	Sep	Oct	
		-----mm-----							
Northwest	2017	712	71	133	130	150	111	47	70
	2018	512	56	59	88	63	104	72	69
	10-Year Avg	596	83	90	95	97	78	76	77
Wooster	2017	544.8	79.0	127.3	95	112	30	30	72
	2018	727.7	136.1	104.4	98	113	59	153	65
	10-Year Avg	609.4	88.0	95.4	104	95	72	80	75

Table 1. 4. Monthly average temperature for the 2017 and 2018 growing seasons and the past 10-year average at the Wooster and the Northwest sites.

Site	Year	Average Temperature (Apr-Oct)							
		Apr	May	Jun	Jul	Aug	Sep	Oct	
		-----°C-----							
Northwest	2017	17.8	12.8	15.1	21.7	22.6	20.1	18.6	14.0
	2018	18.0	6.1	19.4	22.0	22.8	22.8	20.4	12.2
	10-Year Avg	17.9	10.0	17.3	21.8	23.2	22.0	18.6	12.2
Wooster	2017	17.4	13.0	14.7	20.6	22.2	20.2	17.8	13.3
	2018	17.5	6.6	19.1	20.9	22.6	21.6	20.2	11.7
	10-Year Avg	17.3	10.1	16.7	20.7	22.4	21.4	18.0	11.6

Soil Chemical Properties

Rotation had the strongest influence on soil chemical properties for both sites (Table 1.5; Table 1.6). For both sites, soil pH was lowest in the CC rotation compared to the CS rotation, but there were no differences between the CFF rotation and the other rotations. Total nitrogen and total organic carbon were highest in the CFF rotation; however, in Wooster there were no significant differences between CFF and CC rotations. The CEC was highest with increased rotation in Wooster, but no significant differences were observed in Northwest. On the other hand, CFF and CC had the highest OM% in Northwest, and there were no significant differences in OM% in Wooster.

Tillage impacted more the Northwest chemical properties than the Wooster site. Total nitrogen was highest in the no-till soils compared to other tillage, but there were no differences between no-till and moldboard plow in Wooster (Table 1.5; Table 1.6). All the other soil chemical properties were not significantly impacted by the different tillage treatments (Table 1.6). On the other hand, CEC and TOC were highest at the no-till soils, OM% was highest in soils with reduced tillage intensity, and pH was lowest in no-till soils compared to moldboard plow for both sites (Table 1.5).

Table 1. 5. Pre-plant 2017 soil chemical properties for Northwest (mean \pm standard error). Different letters within the same column of treatment values represent significantly different treatments ($\alpha = 0.1$). F-statistics and significance from analysis of variance for each soil chemical property.

Northwest Site							
Variable	pH	CEC (Meg/100g)	OM %	TN %	TOC%	P (ppm)	K (ppm)
Rotation							
CC	6.2 \pm 0.2 b	19.6 \pm 0.8	3.1 \pm 0.1 ab	0.20 \pm 0.01 b	2.1 \pm 0.08 b	39.6 \pm 7.0 a	203.2 \pm 6.0
CS	6.8 \pm 0.1 a	19.5 \pm 0.9	2.7 \pm 0.2 b	0.19 \pm 0.01 b	2.0 \pm 0.04 b	29.9 \pm 1.7 b	219.7 \pm 15.6
CFF	6.5 \pm 0.1 ab	19.5 \pm 0.6	3.2 \pm 0.1 a	0.23 \pm 0.01 a	2.3 \pm 0.09 a	40.1 \pm 3.0 a	231.1 \pm 5.9
Tillage							
MP	6.7 \pm 0.1 a	18.6 \pm 0.5 b	2.7 \pm 0.1 b	0.20 \pm 0.01 b	1.9 \pm 0.04 b	28.1 \pm 1.5 c	213.0 \pm 4.1
CH	6.5 \pm 0.1 ab	19.0 \pm 0.8 ab	3.1 \pm 0.1 a	0.20 \pm 0.01 b	2.1 \pm 0.05 b	34.9 \pm 4.3 b	217.6 \pm 7.1
NT	6.3 \pm 0.2 b	20.9 \pm 0.7 a	3.3 \pm 0.1 a	0.22 \pm 0.01 a	2.3 \pm 0.10 a	46.6 \pm 5.2 a	223.4 \pm 17.0
ANOVA F statistics and significance							
Rotation (R)	9.02 **	2.82 .	3.08 .	11.85 ***	7.11 **	13.25 ***	2.34 NS
Tillage (T)	5.0 *	0.01 NS	5.63 *	6.73 **	12.47 ***	34.9 ***	0.33 NS
T x R	5.31 **	0.49 NS	0.54 NS	0.9 NS	1.6 NS	26.55 ***	1.13 NS

Significance level: p < 0.1 | * Significance level: p < 0.05 | ** Significance level: p < 0.01 | *** Significance level: p < 0.001 | "NS" Significance level: p > 0.1
 Rotation: CC = continuous corn; CS = corn-soybean; CFF = corn-forage-forage
 Tillage: MP = moldboard plow; CH = chisel till; NT = no-till
 CEC = cation exchange capacity; OM = organic matter; TN = total nitrogen; TOC = total organic carbon

Table 1. 6. Pre-plant 2017 soil chemical properties in Wooster (mean \pm standard error). Different letters within the same column of treatment values represent significantly different treatments ($\alpha = 0.1$). F-statistics and significance from analysis of variance for each soil chemical property.

		Wooster Site						
Variable	pH	CEC (Mec/100g)	OM %	TN %	TOC %	P (ppm)	K (ppm)	
Rotation								
CC	6.2 \pm 0.2 b	6.7 \pm 0.5 b	2.1 \pm 0.1	0.20 \pm 0.005 ab	1.9 \pm 0.07 a	47.2 \pm 2.9	128.4 \pm 6.2 b	
CS	6.5 \pm 0.1 a	8.1 \pm 0.3 a	1.9 \pm 0.2	0.19 \pm 0.003 b	1.7 \pm 0.04 b	49.6 \pm 3.8	132.0 \pm 8.0 b	
CFF	6.3 \pm 0.1 ab	7.5 \pm 0.3 a	2.1 \pm 0.1	0.23 \pm 0.004 a	1.9 \pm 0.03 a	50.1 \pm 3.3	147.1 \pm 11.3 a	
Tillage								
MP	6.4 \pm 0.1	7.9 \pm 0.3	2.1 \pm 0.1	0.20 \pm 0.005 ab	1.8 \pm 0.03	45.1 \pm 2.3 b	139.1 \pm 7.5 a	
CH	6.4 \pm 0.1	7.3 \pm 0.4	2.1 \pm 0.1	0.20 \pm 0.005 b	1.9 \pm 0.07	55.1 \pm 3.2 a	151.9 \pm 8.4 a	
NT	6.2 \pm 0.2	7.1 \pm 0.5	1.9 \pm 0.1	0.22 \pm 0.003 a	1.8 \pm 0.05	46.7 \pm 3.4 ab	116.6 \pm 6.7 b	
ANOVA F statistics and significance								
Rotation (R)	4.48 *	4.43 *	0.67 NS	5.08 *	4.32 *	0.29 NS	4.78 *	
Tillage (T)	1.49 NS	1.43 NS	1.71 NS	2.71 .	1.49 NS	3.63 .	15.58 ***	
T x R	1.26 NS	0.42 NS	0.34 NS	0.65 NS	1.45 NS	1.93 NS	2.52 .	

“.” Significance level: $p < 0.1$ | * Significance level: $p < 0.05$ | ** Significance level: $p < 0.01$ | *** Significance level: $p < 0.001$ | “NS” Significance level: $p > 0.1$

Rotation: CC = continuous corn; CS = corn-soybean; CFF = corn-forage-forage

Tillage: MP = moldboard plow; CH = chisel till; NT = no-till

CEC = cation exchange capacity; OM = organic matter; TN = total nitrogen; TOC = total organic carbon

Soil Moisture

Tillage influenced soil moisture more than crop rotation at Wooster, but the magnitude of effects from Tillage and Rotation on soil moisture were similar at Northwest (Table 1.7). Seasonal variation for soil moisture was inconsistent across sites and years (Table 1.8, 1.9, 1.10, 1.11), but was overall significant (Table 1.7) indicating changes in soil moisture between different stages of the growing seasons. In Northwest, soil moisture trends were inconsistent across stages and years, and treatments. Overall, soil moisture was higher in 2017 than 2018 (Figure 1.2). In general, moldboard plow had higher soil moisture before planting corn and during the corn early growth stage (V5). In later corn growth stages soil moisture was higher in the no-till soils and in some stages no-till did not vary from moldboard plow (Table 1.8; Table 1.9). Interestingly, the no-till under the CFF rotation had the highest and less temporally variable soil moisture in Northwest 2018 (Figure 1.2).

In Wooster, soil moisture was highest in the no-till soils and lowest in moldboard plow (Figure 1.3; Table 1.10; Table 1.11). Also, CC and CFF had the highest soil moisture of the rotation treatments. The 2017 and 2018 growing seasons had similar trends of soil moisture, where the vegetative stages had higher moisture and the reproductive stages had less soil moisture as the temperature increased in the summer and rainfall decreased during September (Figure 1.3).

Table 1. 7. Soil moisture F statistics and significance from repeated measures analysis of variance of all sampling stages (n=162 per site)

Effect	Northwest			
	2017		2018	
Tillage (T)	4.1	*	11.4	***
Rotation (R)	4.6	*	14.7	***
T x R	0.5	NS	2.6	*
Stage (S)	371.5	***	261.0	***
S x T	3.5	***	2.2	*
S x R	1.9	.	2.8	**
S x T x R	1.4	NS	3.8	***

Effect	Wooster			
	2017		2018	
Tillage (T)	20.2	***	25.3	***
Rotation (R)	7.3	**	7.1	**
T x R	0.4	NS	0.2	NS
Stage (S)	191.1	***	494.9	***
S x T	1.7	.	4.0	***
S x R	2.4	*	2.8	**
S x T x R	1.6	.	1.6	.

“.” Significance level: $p < 0.1$ | * Significance level: $p < 0.05$

** Significance level: $p < 0.01$ | *** Significance level: $p < 0.001$

“NS” Significance level: $p > 0.1$

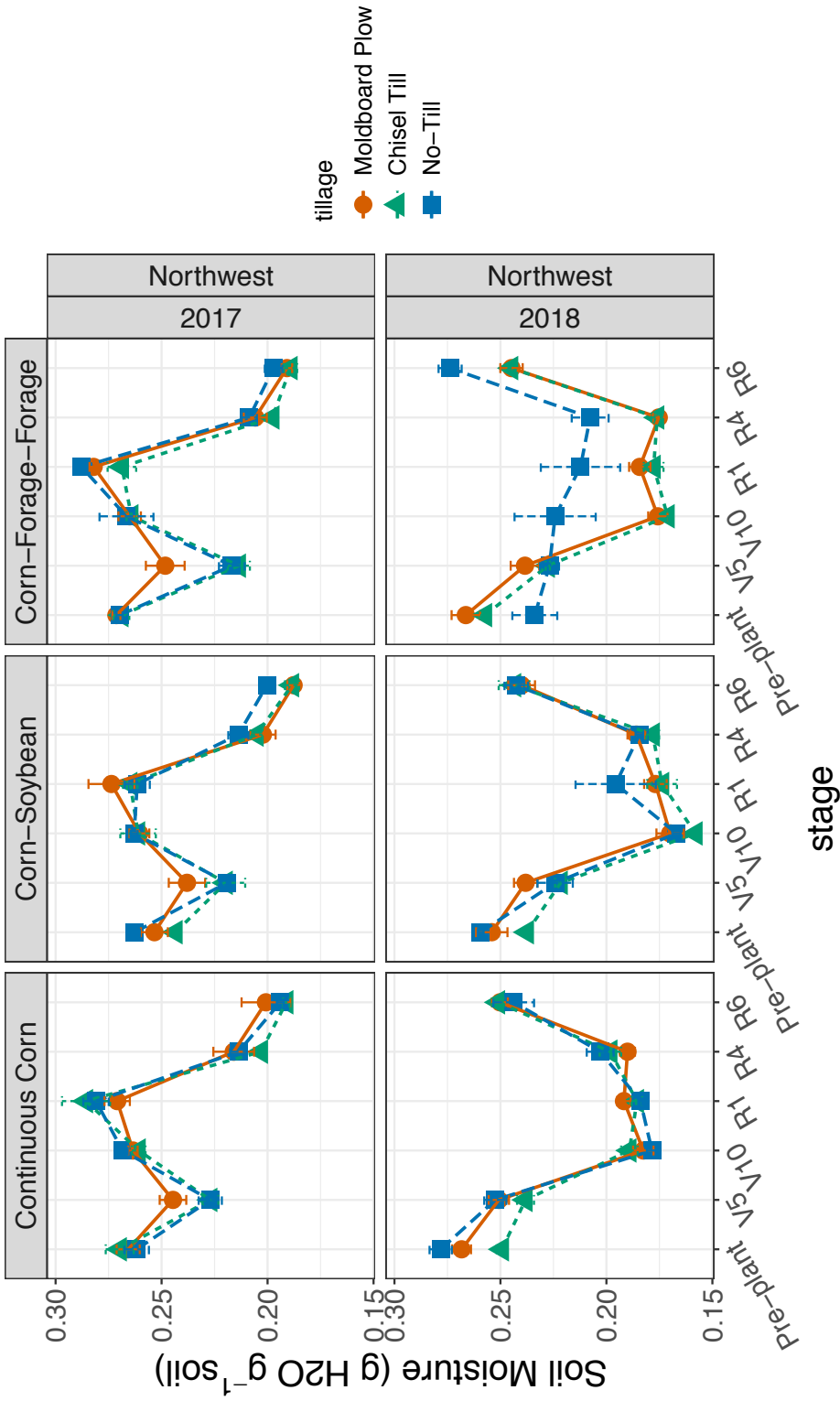


Figure 1. 2. Soil moisture means at Northwest in the 2017 and 2018 growing seasons for three tillage regimes in three crop rotations. Tillage practices include moldboard plow (red circles, solid line), chisel till (green triangles, dotted line), and no-till (blue squares, dashed line). Error bars represent one standard error of the mean.

Table 1. 8. Soil moisture at Northwest in 2017 by tillage and rotation treatments for each sampled crop developmental stage (mean \pm standard error). Tillage treatments were averaged over rotations and vice versa. Different letters within the same column of rotation or treatment values represent significantly different treatments ($\alpha = 0.1$). F-statistics and significance from analysis of variance for each crop stage.

Northwest Site 2017						
Variable	PP	V5	V10	R1	R4	R6
Soil Moisture (g H ₂ O kg ⁻¹ soil)						
Tillage						
MP	0.263 \pm 0.004	0.244 \pm 0.004 a	0.263 \pm 0.002	0.276 \pm 0.004	0.208 \pm 0.004 ab	0.193 \pm 0.004
CH	0.261 \pm 0.005	0.220 \pm 0.004 b	0.262 \pm 0.003	0.273 \pm 0.005	0.202 \pm 0.001 b	0.190 \pm 0.001
NT	0.265 \pm 0.002	0.221 \pm 0.003 b	0.266 \pm 0.004	0.277 \pm 0.005	0.212 \pm 0.002 a	0.197 \pm 0.002
Rotation						
CC	0.266 \pm 0.003 a	0.233 \pm 0.004	0.264 \pm 0.002	0.279 \pm 0.005 a	0.211 \pm 0.004	0.195 \pm 0.004
CS	0.253 \pm 0.003 b	0.226 \pm 0.005	0.262 \pm 0.003	0.267 \pm 0.004 b	0.207 \pm 0.003	0.192 \pm 0.002
CFF	0.270 \pm 0.001 a	0.226 \pm 0.007	0.265 \pm 0.004	0.280 \pm 0.003 a	0.204 \pm 0.003	0.192 \pm 0.002
Source	ANOVA F statistics and significance					
Tillage (T)	0.7 NS	12.3 ***	0.4 NS	0.2 NS	3.1 .	2.0 NS
Rotation (R)	11.7 ***	1.1 NS	0.3 NS	3.5 .	1.5 NS	0.4 NS
T x R	2.5 .	0.6 NS	0.1 NS	2.0 NS	0.8 NS	1.1 NS

“.” Significance level: $p < 0.1$ | * Significance level: $p < 0.05$ | ** Significance level: $p < 0.01$ | *** Significance level: $p < 0.001$ | “NS” Significance level: $p > 0.1$
 PP = pre-plant, V5 = fifth collared leaf, V10 = tenth collared leaf, R1 = silking, R4 = dough, R6 = physiological maturity
 Rotation: CC = continuous corn; CS = corn-soybean; CFF = corn-forage-forage | Tillage: MP = moldboard plow; CH = chisel till; NT = no-till

Table 1. 9. Soil moisture at Northwest in 2018 by tillage and rotation treatments for each sampled crop developmental stage (mean \pm standard error). Tillage treatments were averaged over rotations and vice versa. Different letters within the same column of rotation or treatment values represent significantly different treatments ($\alpha = 0.1$). F-statistics and significance from analysis of variance for each crop stage.

Northwest Site 2018						
Variable	PP	V5	V10	R1	R4	R6
Soil Moisture (g H ₂ O kg ⁻¹ soil)						
Tillage						
MP	0.263 \pm 0.004 a	0.242 \pm 0.004 a	0.176 \pm 0.003 b	0.184 \pm 0.003 ab	0.184 \pm 0.003 b	0.245 \pm 0.003
CH	0.248 \pm 0.003 b	0.229 \pm 0.003 b	0.173 \pm 0.005 b	0.179 \pm 0.003 b	0.184 \pm 0.004 b	0.247 \pm 0.003
NT	0.257 \pm 0.007 a	0.234 \pm 0.005 ab	0.190 \pm 0.010 a	0.197 \pm 0.009 a	0.198 \pm 0.005 a	0.253 \pm 0.006
Rotation						
CC	0.265 \pm 0.005 a	0.247 \pm 0.003 a	0.183 \pm 0.002 a	0.187 \pm 0.002	0.197 \pm 0.003 a	0.248 \pm 0.003 ab
CS	0.250 \pm 0.004 b	0.228 \pm 0.004 b	0.165 \pm 0.003 b	0.182 \pm 0.007	0.183 \pm 0.002 b	0.242 \pm 0.003 b
CFF	0.252 \pm 0.006 b	0.231 \pm 0.003 b	0.190 \pm 0.010 a	0.191 \pm 0.008	0.186 \pm 0.006 b	0.254 \pm 0.005 a
ANOVA F statistics and significance						
Source						
Tillage (T)	6.9 **	5.1 *	5.3 *	2.8 .	9.4 **	2.0 NS
Rotation(R)	8.1 **	12.0 ***	11.1 **	0.7 NS	7.4 **	4.0 *
T x R	9.4 ***	0.9 NS	7.3 **	1.2 NS	4.3 *	4.1 *

“.” Significance level: p < 0.1 | * Significance level: p < 0.05 | ** Significance level: p < 0.01 | *** Significance level: p < 0.001 | “NS” Significance level: p > 0.1
 PP = pre-plant, V5 = fifth collared leaf, V10 = tenth collared leaf, R1 = silking, R4 = dough, R6 = physiological maturity
 Rotation: CC = continuous corn; CS = corn-soybean; CFF = corn-forage-forage | Tillage: MP = moldboard plow; CH = chisel till; NT = no-till

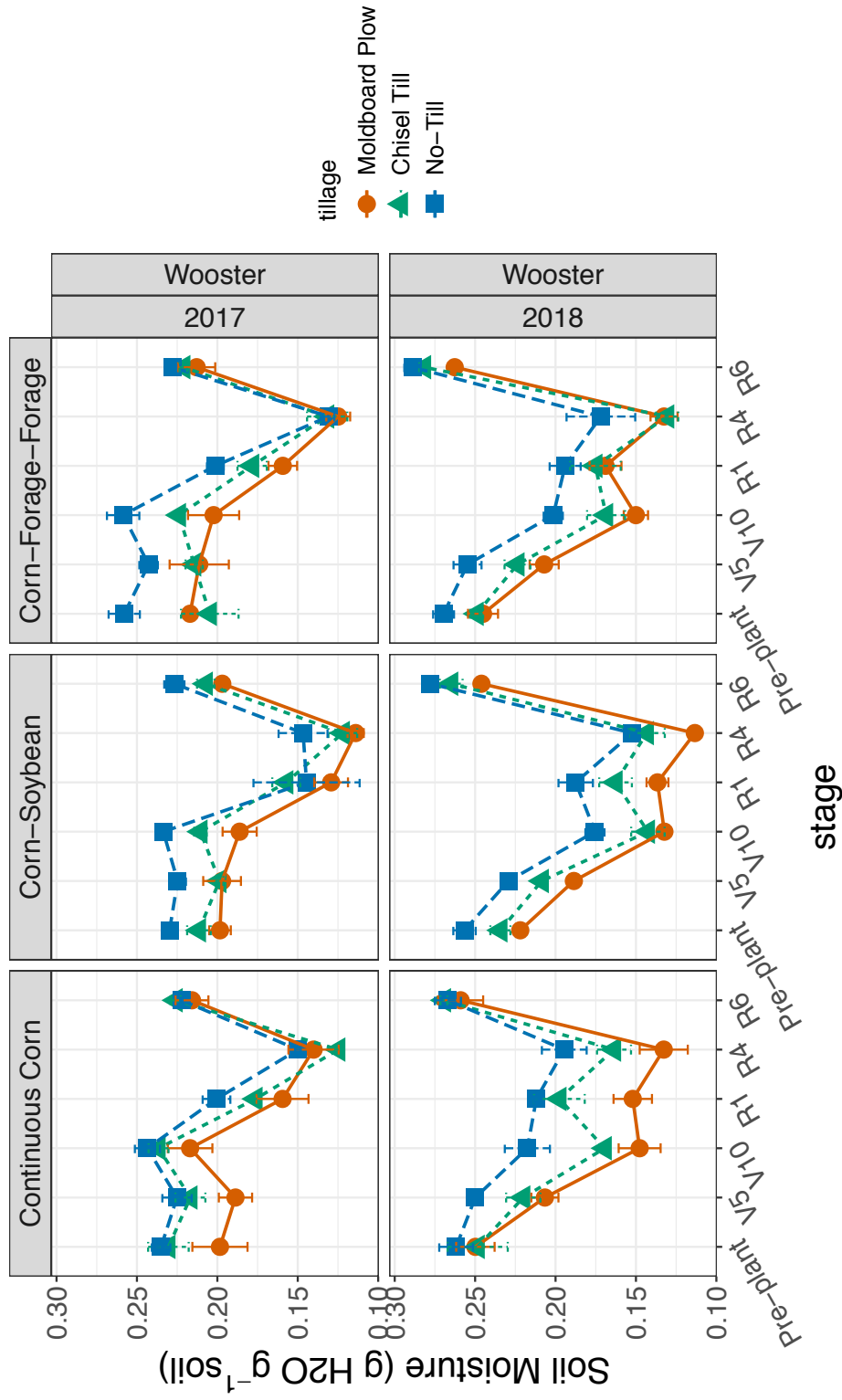


Figure 1. 3. Soil moisture means at Wooster in the 2017 and 2018 growing seasons for three tillage regimes in three crop rotations. Tillage practices include moldboard plow (red circles, solid line), chisel till (green triangles, dotted line), and no-till (blue squares, dashed line). Error bars represent one standard error of the mean.

Table 1. 10. Soil moisture at Wooster in 2017 by tillage and rotation treatments for each sampled crop developmental stage (mean \pm standard error). Tillage treatments were averaged over rotations and vice versa. Different letters within the same column of rotation or treatment values represent significantly different treatments ($\alpha = 0.1$). F-statistics and significance from analysis of variance for each crop stage.

Wooster 2017							
Variable	PP	V5	V10	R1	R4	R6	
Soil Moisture (g H ₂ O kg ⁻¹ soil)							
Tillage							
MP	0.205 \pm 0.006 b	0.199 \pm 0.008 b	0.202 \pm 0.008 c	0.149 \pm 0.008 b	0.126 \pm 0.006	0.209 \pm 0.005 b	
CH	0.216 \pm 0.008 b	0.210 \pm 0.004 b	0.224 \pm 0.004 b	0.171 \pm 0.005 ab	0.126 \pm 0.005	0.218 \pm 0.003 ab	
NT	0.241 \pm 0.006 a	0.231 \pm 0.005 a	0.245 \pm 0.005 a	0.182 \pm 0.014 a	0.143 \pm 0.005	0.226 \pm 0.002 a	
Rotation							
CC	0.221 \pm 0.009	0.210 \pm 0.007 ab	0.232 \pm 0.006 a	0.179 \pm 0.008 a	0.138 \pm 0.006	0.221 \pm 0.003 a	
CS	0.213 \pm 0.006	0.207 \pm 0.006 b	0.210 \pm 0.008 b	0.144 \pm 0.011 b	0.128 \pm 0.007	0.210 \pm 0.005 b	
CFF	0.227 \pm 0.010	0.223 \pm 0.008 a	0.228 \pm 0.010 a	0.180 \pm 0.007 a	0.129 \pm 0.004	0.221 \pm 0.004 ab	
ANOVA F statistics and significance							
Source							
Tillage (T)	10.5 **	11.9 ***	26.1 ***	5.2 *	2.9 .	6.3 **	
Rotation(R)	1.4 NS	3.2 .	7.8 **	7.9 **	1.1 NS	3.3 .	
T x R	1.9 NS	0.9 NS	1.3 NS	0.7 NS	1.1 NS	1.3 NS	

“.” Significance level: $p < 0.1$ | * Significance level: $p < 0.05$ | ** Significance level: $p < 0.01$ | *** Significance level: $p < 0.001$ | “NS” Significance level: $p > 0.1$
 PP = pre-plant, V5 = fifth collared leaf, V10 = tenth collared leaf, R1 = silking, R4 = dough, R6 = physiological maturity
 Rotation: CC = continuous corn; CS = corn-soybean; CFF = corn-forage-forage | Tillage: MP = moldboard plow; CH = chisel till; NT = no-till

Table 1. 11. Soil moisture at Wooster in 2018 by tillage and rotation treatments for each sampled crop developmental stage (mean \pm standard error). Tillage treatments were averaged over rotations and vice versa. Different letters within the same column of rotation or treatment values represent significantly different treatments ($\alpha = 0.1$). F-statistics and significance from analysis of variance for each crop stage.

Wooster 2018						
Variable	PP	V5	V10	R1	R4	R6
Soil Moisture (g H ₂ O kg ⁻¹ soil)						
MP	0.239 \pm 0.006 b	0.201 \pm 0.005 c	0.143 \pm 0.005 c	0.153 \pm 0.007 b	0.126 \pm 0.006 b	0.256 \pm 0.005 b
CH	0.243 \pm 0.007 b	0.217 \pm 0.004 b	0.160 \pm 0.006 b	0.178 \pm 0.009 a	0.146 \pm 0.007 b	0.272 \pm 0.003 a
NT	0.263 \pm 0.005 a	0.245 \pm 0.005 a	0.198 \pm 0.008 a	0.198 \pm 0.006 a	0.173 \pm 0.010 a	0.278 \pm 0.004 a
Rotation						
CC	0.253 \pm 0.007 a	0.226 \pm 0.008 a	0.178 \pm 0.012 a	0.187 \pm 0.011 a	0.164 \pm 0.011 a	0.265 \pm 0.005 b
CS	0.238 \pm 0.006 b	0.209 \pm 0.006 b	0.150 \pm 0.007 b	0.162 \pm 0.009 b	0.136 \pm 0.007 b	0.263 \pm 0.005 b
CFF	0.255 \pm 0.006 a	0.228 \pm 0.008 a	0.173 \pm 0.009 a	0.179 \pm 0.007 ab	0.145 \pm 0.010 ab	0.278 \pm 0.004 a
ANOVA F statistics and significance						
Source						
Tillage (T)	7.0 **	42.6 ***	29.5 ***	13.3 ***	12.3 ***	8.9 **
Rotation(R)	4.0 *	9.8 **	8.4 **	4.2 *	4.4 *	4.4 *
T x R	0.5 NS	0.3 NS	0.6 NS	1.1 NS	1.0 NS	0.9 NS

“.” Significance level: $p < 0.1$ | * Significance level: $p < 0.05$ | ** Significance level: $p < 0.01$ | *** Significance level: $p < 0.001$ | “NS” Significance level: $p > 0.1$
 PP = pre-plant, V5 = fifth collared leaf, V10 = tenth collared leaf, R1 = silking, R4 = dough, R6 = physiological maturity
 Rotation: CC = continuous corn; CS = corn-soybean; CFF = corn-forage-forage | Tillage: MP = moldboard plow; CH = chisel till; NT = no-till

Permanganate Oxidizable Carbon

Crop rotation had a stronger effect on POXC than tillage (Table 1.12), with the CFF rotation yielding the highest POXC values across both sites and years (Table 1.13; Table 1.14; Table 1.15; Table 1.16). Seasonal dynamics variation was significant for all sites and years, indicating crop development influences active carbon levels over the growing season. Also, there were significant tillage by rotation interactions in both sites and years (Table 1.12). In general, POXC values in no-till soils increased under the CFF rotation compared to less diverse rotations (Figure 1.4; Figure 1.5).

In Northwest, both tillage and rotation had a strong effect on POXC with no-till and CFF having higher POXC values across both years. Few differences were observed between no-till and chisel till in Northwest (Table 1.13; Table 1.14). Unlike Northwest, tillage had minimum effects on POXC in Wooster, with chisel till having higher POXC values than moldboard plow at very few vegetative stages (Table 1.15; Table 1.16). Rotation had a stronger effect on POXC in Wooster with the CFF rotation having higher POXC than CC and CS. Interestingly, in some stages there were no differences between CFF and CC rotations, and the CS rotation had consistently the lowest POXC values in Wooster (Table 1.15; Table 1.16).

Table 1. 12. F-statistics and significance of soil health indicators from repeated measures ANOVA of all six sampling stages by year (n=162 per site)

Effect	Permanganate Oxidizable C		Mineralizable C		Soil Protein	
	2017	2018	2017	2018	2017	2018
	Northwest site					
Tillage (T)	5.89 **	5.28 **	26.79 ***	36.75 ***	16.35 ***	77.73 ***
Rotation (R)	14.33 ***	15.01 ***	12.37 ***	9.1 ***	27.77 ***	59.48 ***
T x R	4.74 **	4.20 **	0.51 NS	2.27 .	2.61 *	12.55 ***
Stage (S)	24.46 ***	8.55 ***	8.20 ***	11.77 ***	71.49 ***	52.54 ***
S x T	1.12 NS	0.85 NS	1.07 NS	1.61 NS	1.8 .	1.31 NS
S x R	2.17 *	1.05 NS	1.49 NS	0.42 NS	2.11 *	0.83 NS
S x T x R	2.52 **	1.07 NS	1.76 *	0.59 NS	1.14 NS	0.41 NS
	Wooster site					
Tillage (T)	1.81 NS	2.13 NS	8.47 ***	4.48 *	2.48 .	1.93 NS
Rotation (R)	6.89 **	15.94 ***	6.43 **	12.19 ***	13.46 ***	10.45 ***
T x R	4.19 **	2.31 .	4.18 **	0.46 NS	0.47 NS	0.44 NS
Stage (S)	14.90 ***	23.70 ***	21.24 ***	15.44 ***	106.48 ***	18.17 ***
S x T	1.88 .	1.28 NS	0.96 NS	1.75 .	0.58 NS	0.72 NS
S x R	0.91 NS	0.54 NS	1.02 NS	2.35 *	1.26 NS	0.6 NS
S x T x R	0.67 NS	0.86 NS	0.96 NS	0.82 NS	1.2 NS	0.56 NS

“.” Significance level: p<0.1 | * Significance level: p < 0.05 | ** Significance level: p < 0.01 | *** Significance level: p<0.001 | “NS” Significance level: p>0.1

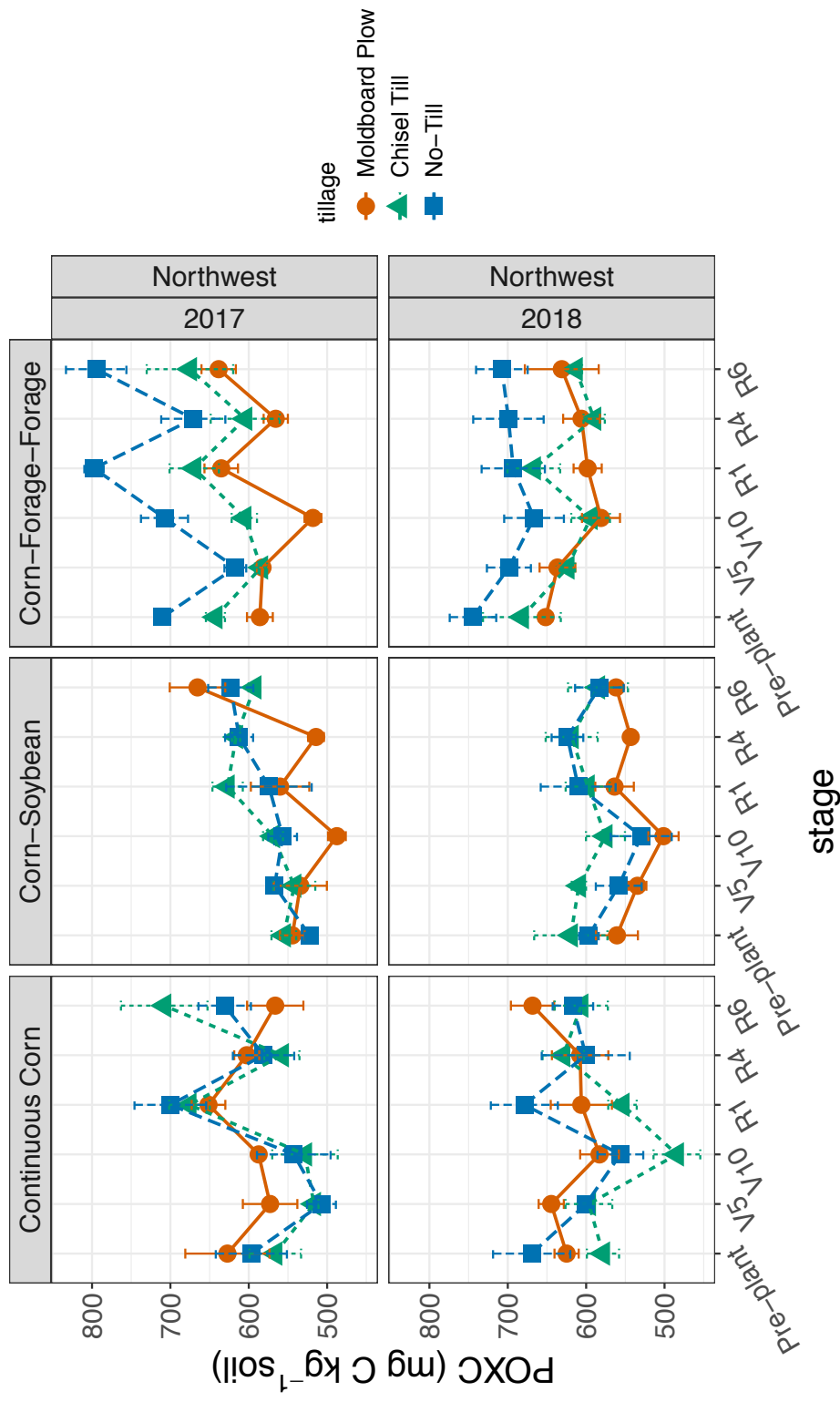


Figure 1. 4. Permanganate Oxidizable Carbon (POXC) mean values at Northwest in the 2017 and 2018 growing seasons for three tillage regimes in three crop rotations. Tillage practices include moldboard plow (red circles, solid line), chisel till (green triangles, dotted line), and no-till (blue squares, dashed line). Error bars represent one standard error of the mean

Table 1. 13. Northwest 2017 permanganate oxidizable carbon (POXC) tillage and rotations treatments means with standard errors for each sampling stage. Different letters within the same row represent significantly different treatments at $\alpha = 0.1$. F statistics and significance from analysis of variance for each stage.

Northwest Site 2017						
Variable	PP	V5	V10	R1	R4	R6
Tillage						
		POXC (mg C kg ⁻¹ soil)				
MP	585.8 ± 20.7	563.1 ± 16.0	531.0 ± 15.6 b	615.5 ± 19.7 a	560.8 ± 14.6	623.4 ± 21.7
CH	588.0 ± 17.7	547.7 ± 12.8	567.7 ± 17.5 ab	657.5 ± 15.1 b	592.9 ± 16.9	658.6 ± 28.4
NT	609.8 ± 30.4	563.8 ± 16.0	602.3 ± 15.6 a	690.6 ± 38.5 a	621.4 ± 21.6	682.8 ± 21.7
Rotation						
CC	596.7 ± 24.1 a	532.3 ± 15.4 b	552.7 ± 20.3 b	675.7 ± 17.8 a	580.5 ± 15.3	634.9 ± 29.6 b
CS	540.7 ± 8.2 b	547.7 ± 13.5 b	537.8 ± 14.6 b	587.1 ± 22.3 b	580.9 ± 17.8	627.1 ± 17.0 b
CFF	646.2 ± 19.1 a	594.6 ± 7.6 a	610.4 ± 29.2 a	700.7 ± 27.3 a	613.6 ± 23.6	702.7 ± 31.2 a

Source	ANOVA F statistics and significance					
Tillage (T)	0.7 NS	0.8 NS	5.8 *	4.0 *	4.9 *	1.9 NS
Rotation (R)	10.6 **	9.9 **	6.7 **	10.1 **	1.9 NS	3.7 *
T x R	2.9 .	2.6 .	6.1 **	2.3 NS	3.5 *	3.7 *

“.” Significance level: $p < 0.1$ | * Significance level: $p < 0.05$ | ** Significance level: $p < 0.01$ | *** Significance level: $p < 0.001$ | “NS” Significance level: $p > 0.1$
 PP = pre-plant, V5 = fifth collared leaf, V10 = tenth collared leaf, R1 = silking, R4 = dough, R6 = physiological maturity
 Rotation: CC = continuous corn; CS = corn-soybean; CFF = corn-forage-forage | Tillage: MP = moldboard plow; CH = chisel till; NT = no-till

Table 1. 14. Northwest 2018 permanganate oxidizable carbon (POXC) tillage and rotations treatments means with standard errors for each sampling stage. Different letters within the same row represent significantly different treatments at $\alpha = 0.1$. F statistics and significance from analysis of variance for each stage.

Northwest Site 2018						
Variable	PP	V5	V10	R1	R4	R6
Tillage						
	POXC (mg C kg ⁻¹ soil)					
MP	612.5 ± 16.3 b	619.0 ± 23.9	555.1 ± 17.7	589.3 ± 15.7 b	585.6 ± 16.4	620.4 ± 22.2
CH	626.7 ± 25.5 ab	609.9 ± 10.2	551.3 ± 21.6	606.2 ± 21.4 ab	612.5 ± 14.1	601.5 ± 15.5
NT	670.1 ± 27.3 a	602.3 ± 15.6	584.2 ± 27.5	660.7 ± 25.3 a	641.2 ± 26.2	635.8 ± 23.9
Rotation						
CC	624.3 ± 20.7 b	613.7 ± 12.6 b	541.2 ± 20.3 b	612.9 ± 25.2 ab	612.5 ± 21.2	630.5 ± 17.5 a
CS	592.2 ± 18.1 b	567.4 ± 14.5 c	535.5 ± 18.0 b	590.7 ± 18.8 b	595.2 ± 17.3	576.6 ± 14.7 b
CFF	692.7 ± 21.7 a	653.0 ± 15.8 a	614.0 ± 19.9 a	652.6 ± 21.4 a	631.6 ± 22.8	650.6 ± 22.1 a

Source	ANOVA F statistics and significance					
Tillage (T)	3.1 .	0.5 NS	1.4 NS	4.1 *	2.2 NS	1.1 NS
Rotation (R)	9.1 **	17.0 ***	8.2 **	2.9 .	1.0 NS	5.5 *
T x R	1.4 NS	5.6 **	3.7 *	1.4 NS	1.6 NS	1.8 NS

“.” Significance level: $p < 0.1$ | * Significance level: $p < 0.05$ | ** Significance level: $p < 0.01$ | *** Significance level: $p < 0.001$ | “NS” Significance level: $p > 0.1$
PP = pre-plant, V5 = fifth collared leaf, V10 = tenth collared leaf, R1 = silking, R4 = dough, R6 = physiological maturity
Rotation: CC = continuous corn; CS = corn-soybean; CFF = corn-forage-forage | Tillage: MP = moldboard plow; CH = chisel till; NT = no-till

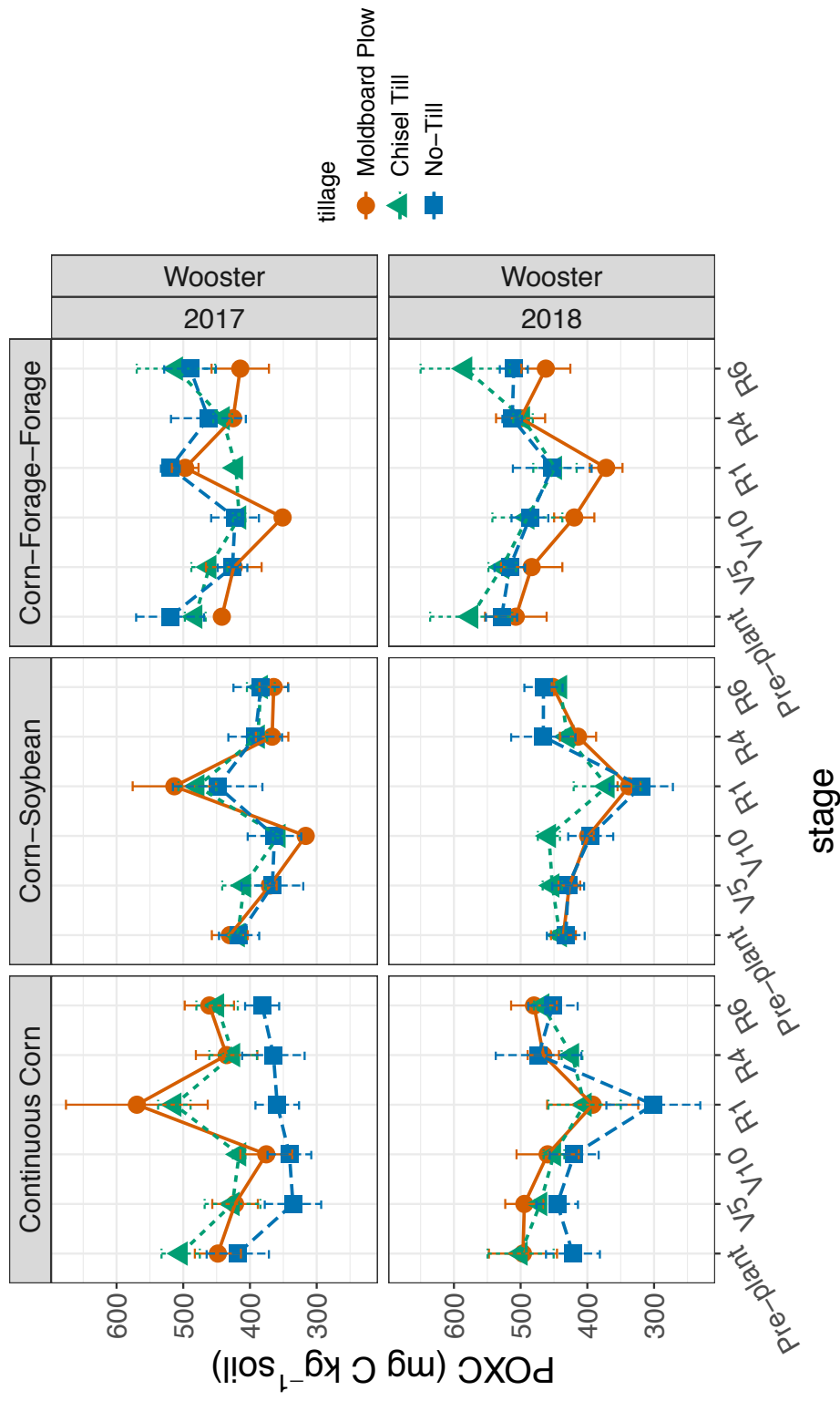


Figure 1. 5. Permanganate Oxidizable Carbon (POXC) mean values at Wooster in the 2017 and 2018 growing seasons for three tillage regimes in three crop rotations. Tillage practices include moldboard plow (red circles, solid line), chisel till (green triangles, dotted line), and no-till (blue squares, dashed line). Error bars represent one standard error of the mean

Table 1. 15. Wooster 2017 permanganate oxidizable carbon (POXC) tillage and rotations treatments averages with standard errors for each sampling stage. Different letters within the same row represent significantly different treatments at $\alpha = 0.1$. F statistics and significance from analysis of variance for each stage.

Wooster 2017						
Variable	PP	V5	V10	R1	R4	R6
Tillage						
			POXC (mg C kg ⁻¹ soil)			
MP	440.2 ± 12.9	405.6 ± 18.1ab	347.7 ± 14.5 b	526.7 ± 37.7	409.0 ± 18.5	413.3 ± 22.4
CH	468.1 ± 16.3	432.0 ± 18.8 a	396.6 ± 10.8 a	471.1 ± 17.3	418.1 ± 14.1	448.0 ± 27.2
NT	451.5 ± 27.6	376.1 ± 23.4 b	375.5 ± 21.9 ab	442.3 ± 31.9	406.4 ± 28.1	418.4 ± 25.3
Rotation						
CC	456.8 ± 22.5 ab	394.7 ± 24.7 b	377.6 ± 18.5 ab	480.7 ± 45.5	408.4 ± 24.2 ab	430.7 ± 20.0 a
CS	421.6 ± 12.4 b	381.8 ± 18.0 b	345.6 ± 14.2 b	480.2 ± 29.3	382.4 ± 14.7 b	377.3 ± 15.0 b
CFF	481.3 ± 19.0 a	437.1 ± 16.8 a	396.6 ± 15.8 a	479.2 ± 16.5	442.8 ± 17.6 a	471.8 ± 27.9 a

Source	ANOVA F statistics and significance					
Tillage (T)	1.2 NS	4.4 *	2.8 .	2.5 NS	0.2 NS	1.5 NS
Rotation (R)	5.3 *	4.7 *	3.1 .	0.0 NS	4.9 *	9.3 **
T x R	2.8 .	1.2 NS	1.4 NS	2.3 NS	1.7 NS	2.4 .

“.” Significance level: $p < 0.1$ | * Significance level: $p < 0.05$ | ** Significance level: $p < 0.01$ | *** Significance level: $p < 0.001$ | “NS” Significance level: $p > 0.1$
PP = pre-plant, V5 = fifth collared leaf, V10 = tenth collared leaf, R1 = silking, R4 = dough, R6 = physiological maturity
Rotation: CC = continuous corn; CS = corn-soybean; CFF = corn-forage-forage | Tillage: MP = moldboard plow; CH = chisel till; NT = no-till

Table 1. 16. Wooster 2018 permanganate oxidizable carbon (POXC) tillage and rotations treatments averages with standard errors for each sampling stage. Different letters within the same row represent significantly different treatments at $\alpha = 0.1$. F statistics and significance from analysis of variance for each stage.

Wooster 2018						
Variable	PP	V5	V10	R1	R4	R6
Tillage						
POXC (mg C kg ⁻¹ soil)						
MP	479.9 ± 23.4	468.3 ± 19.4	426.0 ± 18.5 b	367.1 ± 22.9	460.0 ± 19.4	464.6 ± 15.1
CH	496.5 ± 28.0	483.6 ± 13.6	465.9 ± 17.6 a	407.5 ± 26.0	449.9 ± 13.4	497.2 ± 29.4
NT	460.9 ± 23.2	463.4 ± 18.8	433.6 ± 21.3 ab	357.7 ± 38.2	484.0 ± 24.6	475.9 ± 17.2
Rotation						
CC	472.7 ± 26.9 b	470.1 ± 14.3 b	443.1 ± 18.7 ab	365.7 ± 36.2 ab	454.1 ± 21.7 b	466.4 ± 15.5 ab
CS	436.5 ± 10.2 b	436.1 ± 10.2 b	417.1 ± 15.0 b	342.2 ± 22.0 b	436.7 ± 17.8 b	452.8 ± 9.1 b
CFF	532.0 ± 22.8 a	509.1 ± 17.3 a	465.2 ± 22.3 a	424.3 ± 24.5 a	503.1 ± 12.4 a	518.5 ± 28.8 a

Source	ANOVA F statistics and significance					
Tillage (T)	2.3 NS	0.9 NS	3.6 .	1.8 NS	1.8 NS	1.0 NS
Rotation (R)	11.6 ***	10.2 **	4.7 *	4.6 *	6.8 **	4.2 *
T x R	1.3 NS	1.3 NS	2.5 .	1.6 NS	0.6 NS	1.9 NS

“.” Significance level: p < 0.1 | * Significance level: p < 0.05 | ** Significance level: p < 0.01 | *** Significance level: p < 0.001 | “NS” Significance level: p > 0.1
PP = pre-plant, V5 = fifth collared leaf, V10 = tenth collared leaf, R1 = silking, R4 = dough, R6 = physiological maturity
Rotation: CC = continuous corn; CS = corn-soybean; CFF = corn-forage-forage | Tillage: MP = moldboard plow; CH = chisel till; NT = no-till

Mineralizable Carbon

Both tillage and rotation had strong effects on soil mineralizable carbon (min C) in both sites (Table 1.12), with the CFF rotation and the low intensity tillage treatments having higher min C values (no-till in Northwest and chisel till in Wooster). As rotation diversity increased min C increased in the no-till soils (Figure 1.6; Figure 1.7). Seasonal variation was significant for all sites and years, indicating differences in microbial respiration (carbon mineralization from organic matter) during the different corn stages of the growing seasons (Table 1.12). While the trends in rotation effects were generally consistent across stages, sites and years, tillage effects on min C varied between sites.

In Northwest, no-till and the CFF rotation had the highest min C values consistently across all treatments. In Northwest mineralizable C increased with decreased tillage intensity and with rotations with two years of forages (Figure 1.6). Tillage had a stronger effect on min C than rotation in Northwest. During some stages, no-till and chisel till had no significant differences but had significantly higher values than the moldboard plow. Additionally, during early corn growth stages there were no differences between the CFF and the CC rotations (specifically in 2018) but were higher than the CS rotation.

In Wooster, chisel till and the CFF rotation had higher min C values in general across stages and years. Overall, min C increased in no-till soils with the most diverse rotation (CFF (Figure 1.7), but min C was lower in the no-till and moldboard plow compared to chisel till. Like Northwest, both tillage and rotation effects were significant

on min C in Wooster; however, trends varied for each year. In 2017, tillage had a stronger effect and in 2018 rotation had a stronger effect on min C (Table 1.12). Finally, the CS rotation had lower C min values.

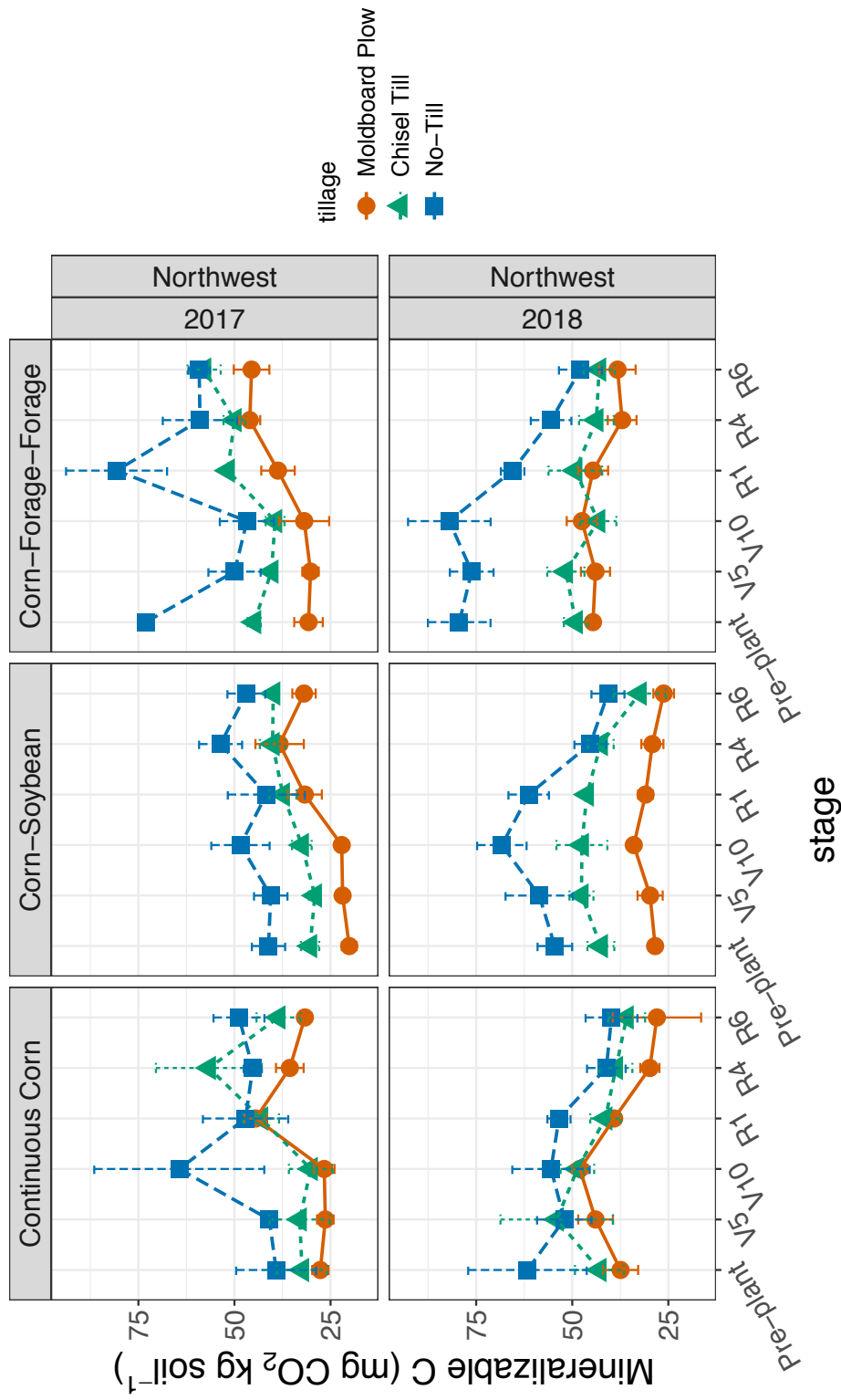


Figure 1. 6. Mineralizable carbon (min C) mean values at Northwest in the 2017 and 2018 growing seasons for three tillage regimes in three crop rotations. Tillage practices include moldboard plow (red circles, solid line), chisel till (green triangles, dotted line), and no-till (blue squares, dashed line). Error bars represent one standard error of the mean.

Table 1. 17. Northwest 2017 mineralizable carbon (min C) tillage and rotations treatments means with standard errors for each sampling stage. Different letters within the same row represent significantly different treatments at $\alpha = 0.1$. F statistics and significance from analysis of variance for each stage.

Northwest Site 2017						
Variable	PP	V5	V10	R1	R4	R6
Tillage						
			Min C (mg CO ₂ -C kg ⁻¹ soil)			
MP	26.2 ± 2.1 c	26.1 ± 1.5 c	26.9 ± 2.5 b	38.3 ± 2.7 b	40.0 ± 2.7 b	36.4 ± 2.8 b
CH	35.9 ± 3.1 b	34.2 ± 2.8 b	34.0 ± 2.4 b	44.1 ± 2.7 b	48.9 ± 4.8 ab	45.5 ± 3.8 a
NT	51.1 ± 6.4 a	43.8 ± 2.8 a	53.2 ± 7.6 a	56.5 ± 8.4 a	52.6 ± 3.9 a	51.6 ± 3.2 a
Rotation						
CC	33.1 ± 4.0 b	33.4 ± 3.2 b	40.4 ± 9.0	44.9 ± 3.6 b	45.8 ± 5.2	39.7 ± 3.6 b
CS	30.5 ± 3.4 b	30.5 ± 3.0 b	34.3 ± 4.5	36.9 ± 3.6 b	44.0 ± 3.6	39.6 ± 2.8 b
CFF	49.6 ± 6.3 a	40.2 ± 3.5 a	39.3 ± 3.6	57.0 ± 7.4 a	51.7 ± 3.6	54.2 ± 2.9 a
ANOVA F statistics and significance						
Source						
Tillage (T)	21.9 ***	18.5 ***	9.1 **	6.5 **	2.9 .	9.9 **
Rotation (R)	14.8 ***	5.8 *	0.5 NS	7.7 **	1.1 NS	11.8 ***
T x R	3.1 *	0.2 NS	0.9 NS	2.9 .	1.1 NS	0.3 NS

“.” Significance level: $p < 0.1$ | * Significance level: $p < 0.05$ | ** Significance level: $p < 0.01$ | *** Significance level: $p < 0.001$ | “NS” Significance level: $p > 0.1$
 PP = pre-plant, V5 = fifth collared leaf, V10 = tenth collared leaf, R1 = silking, R4 = dough, R6 = physiological maturity
 Rotation: CC = continuous corn; CS = corn-soybean; CFF = corn-forage-forage | Tillage: MP = moldboard plow; CH = chisel till; NT = no-till

Table 1. 18. Northwest 2018 mineralizable carbon (min C) tillage and rotations treatments means with standard errors for each sampling stage. Different letters within the same row represent significantly different treatments at $\alpha = 0.1$. F statistics and significance from analysis of variance for each stage.

Northwest Site 2018						
Variable	PP	V5	V10	R1	R4	R6
Tillage						
			Min C (mg CO ₂ -C kg ⁻¹ soil)			
MP	36.8 ± 2.7 b	39.2 ± 3.1 b	43.2 ± 2.7 b	38.2 ± 2.4 c	32.0 ± 2.0 b	30.8 ± 4.1 b
CH	44.9 ± 2.5 b	51.1 ± 4.6 a	46.5 ± 2.8 b	45.6 ± 2.6 b	41.6 ± 2.2 a	37.1 ± 3.1 ab
NT	65.2 ± 6.4 a	62.3 ± 5.1 a	68.6 ± 6.0 a	60.1 ± 2.6 a	47.3 ± 3.2 a	42.8 ± 3.1 a
Rotation						
CC	47.3 ± 6.2 ab	50.0 ± 5.1 ab	50.7 ± 3.4	44.6 ± 2.7 b	36.5 ± 2.7 b	34.5 ± 4.4
CS	41.8 ± 4.1 b	45.3 ± 5.1 b	49.9 ± 5.7	46.2 ± 4.7 b	39.0 ± 3.0 ab	33.1 ± 3.2
CFF	57.7 ± 6.0 a	57.3 ± 5.4 a	57.6 ± 7.1	53.1 ± 4.0 a	45.4 ± 3.5 a	43.0 ± 2.8
ANOVA F statistics and significance						
Source						
Tillage (T)	14.1 ***	9.5 **	14.2 ***	25.7 ***	10.9 **	2.8 .
Rotation (R)	4.3 *	2.6 NS	1.3 NS	4.2 *	3.8 *	2.3 NS
T x R	0.5 NS	1.5 NS	2.4 .	1.4 NS	0.5 NS	0.1 NS

“.” Significance level: $p < 0.1$ | * Significance level: $p < 0.05$ | ** Significance level: $p < 0.01$ | *** Significance level: $p < 0.001$ | “NS” Significance level: $p > 0.1$
 PP = pre-plant, V5 = fifth collared leaf, V10 = tenth collared leaf, R1 = silking, R4 = dough, R6 = physiological maturity
 Rotation: CC = continuous corn; CS = corn-soybean; CFF = corn-forage-forage | Tillage: MP = moldboard plow; CH = chisel till; NT = no-till

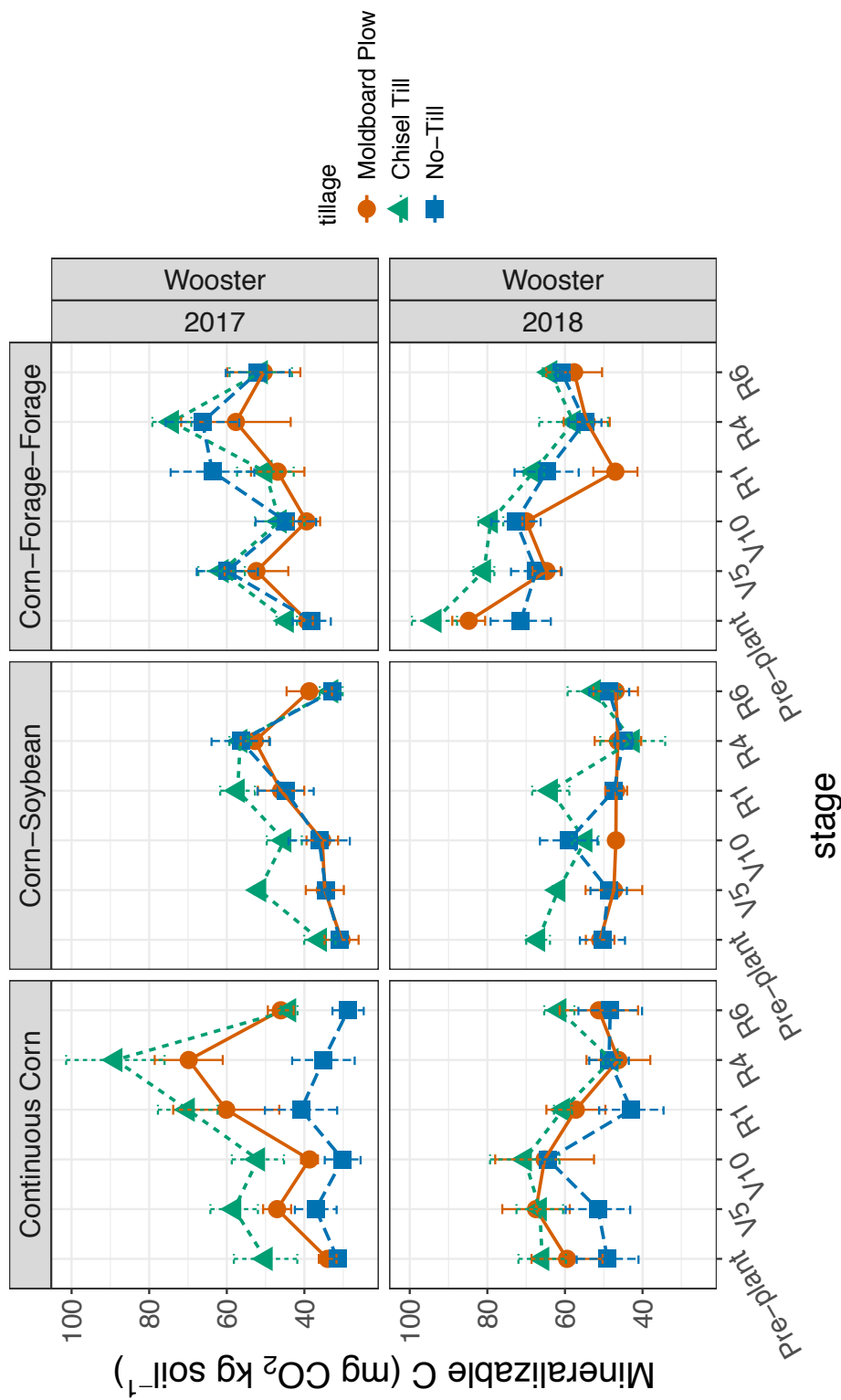


Figure 1. 7. Mineralizable carbon (min C) mean values at Wooster in the 2017 and 2018 growing seasons for three tillage regimes in three crop rotations. Tillage practices include moldboard plow (red circles, solid line), chisel till (green triangles, dotted line), and no-till (blue squares, dashed line). Error bars represent one standard error of the mean

Table 1. 19. Wooster 2017 mineralizable carbon (min C) tillage and rotations treatments means with standard errors for each sampling stage. Different letters within the same row represent significantly different treatments at $\alpha = 0.1$. F statistics and significance from analysis of variance for each stage.

Wooster 2017						
Variable	PP	V5	V10	R1	R4	R6
Tillage						
			Min C (mg CO ₂ -C.kg ⁻¹ .soil)			
MP	34.6 ± 2.0 b	44.7 ± 3.9 b	37.8 ± 1.8 ab	51.0 ± 5.3	60.1 ± 5.5 ab	45.1 ± 3.8
CH	43.5 ± 3.4 a	57.0 ± 2.9 a	47.8 ± 3.2 a	59.1 ± 4.4	73.1 ± 6.2 a	42.8 ± 3.7
NT	33.5 ± 1.9 b	43.8 ± 4.9 b	37.0 ± 4.1 b	49.8 ± 5.8	52.6 ± 6.2 b	37.8 ± 4.5
Rotation						
CC	38.5 ± 3.8 ab	47.4 ± 4.0 b	40.3 ± 4.0	57.0 ± 6.8	64.5 ± 9.3	39.6 ± 3.2 b
CS	32.4 ± 2.0 b	40.3 ± 3.2 b	38.9 ± 3.3	49.3 ± 3.6	55.2 ± 2.6	34.9 ± 2.2 b
CFF	40.6 ± 1.9 a	57.9 ± 4.0 a	43.5 ± 3.3	53.5 ± 5.0	66.0 ± 5.6	51.2 ± 4.3 a
ANOVA F statistics and significance						
Source						
Tillage (T)	5.7 *	6.3 **	3.5 .	1.1 NS	4.6 *	1.5 NS
Rotation (R)	3.4 .	9.0 **	0.5 NS	0.6 NS	1.5 NS	7.8 **
T x R	1.0 NS	1.3 NS	0.9 NS	1.9 NS	3.5 *	1.1 NS

“.” Significance level: $p < 0.1$ | * Significance level: $p < 0.05$ | ** Significance level: $p < 0.01$ | *** Significance level: $p < 0.001$ | “NS” Significance level: $p > 0.1$
 PP = pre-plant, V5 = fifth collared leaf, V10 = tenth collared leaf, R1 = silking, R4 = dough, R6 = physiological maturity
 Rotation: CC = continuous corn; CS = corn-soybean; CFF = corn-forage-forage | Tillage: MP = moldboard plow; CH = chisel till; NT = no-till

Table 1. 20. Wooster 2018 mineralizable carbon (min C) tillage and rotations treatments means with standard errors for each sampling stage. Different letters within the same row represent significantly different treatments at $\alpha = 0.1$. F statistics and significance from analysis of variance for each stage.

Wooster 2018						
Variable	PP	V5	V10	R1	R4	R6
Tillage						
			Min C (mg CO ₂ -C kg ⁻¹ soil)			
MP	65.1 ± 6.0 ab	59.9 ± 4.7 b	60.7 ± 5.1	50.3 ± 3.3 b	49.0 ± 3.7	51.9 ± 4.2
CH	73.2 ± 5.1 a	69.7 ± 3.5 a	68.1 ± 4.6	64.1 ± 2.0 a	49.4 ± 4.2	59.3 ± 2.9
NT	56.9 ± 5.1 b	55.9 ± 4.4 b	65.3 ± 3.5	51.7 ± 4.7 b	49.5 ± 2.5	52.6 ± 3.5
Rotation						
CC	58.1 ± 4.6 b	61.8 ± 4.7 b	66.7 ± 4.6 a	53.5 ± 4.3	47.6 ± 2.9	53.7 ± 4.4 ab
CS	56.1 ± 3.5 b	52.6 ± 3.4 c	53.5 ± 3.0 b	52.6 ± 3.2	44.6 ± 3.1	49.4 ± 3.1 b
CFF	82.0 ± 4.6 a	71.0 ± 3.4 a	73.9 ± 2.5 a	59.9 ± 4.4	55.7 ± 3.4	60.7 ± 2.4 a
ANOVA F statistics and significance						
Source						
Tillage (T)	5.4 *	7.1 **	1.1 NS	6.7 **	0.0 NS	1.3 NS
Rotation (R)	15.2 ***	11.8 ***	8.2 **	1.9 NS	2.6 NS	2.6 NS
T x R	0.4 NS	1.5 NS	0.4 NS	2.6 .	0.1 NS	0.2 NS

“.” Significance level: $p < 0.1$ | * Significance level: $p < 0.05$ | ** Significance level: $p < 0.01$ | *** Significance level: $p < 0.001$ | “NS” Significance level: $p > 0.1$
 PP = pre-plant, V5 = fifth collared leaf, V10 = tenth collared leaf, R1 = silking, R4 = dough, R6 = physiological maturity
 Rotation: CC = continuous corn; CS = corn-soybean; CFF = corn-forage-forage | Tillage: MP = moldboard plow; CH = chisel till; NT = no-till

Soil Protein

Tillage effects on soil protein values varied by site (Table 1.12); however, the rotation effects were consistent over sites, years and stages in general where the CFF and CC rotations had the highest soil protein values (Table 1.21; Table 1.22; Table 1.23; Table 1.24). Seasonal variation was significant for all sites and years, indicating differences in soil protein (labile organic nitrogen) during the different corn stages of the growing seasons (Table 1.12).

In Northwest, no-till soils had consistently the highest soil protein values across all crop rotations (Figure 1.8; Table 1.21; Table 1.22). Soil protein values were often lowest in the moldboard plow soils. Soil protein values were not different on the CC and CFF rotations and both of these rotations had higher soil protein values than the CS rotation (Figure 1.8; Table 1.21; Table 1.22). Although the effect of tillage on soil protein in Wooster was weak, chisel till occasionally had higher soil protein values than no-till in 2017 and higher values than moldboard plow in 2018 (Table 1.23; Table 1.24). Rotation had a stronger effect on soil protein in Wooster where values followed the same trends from Northwest.

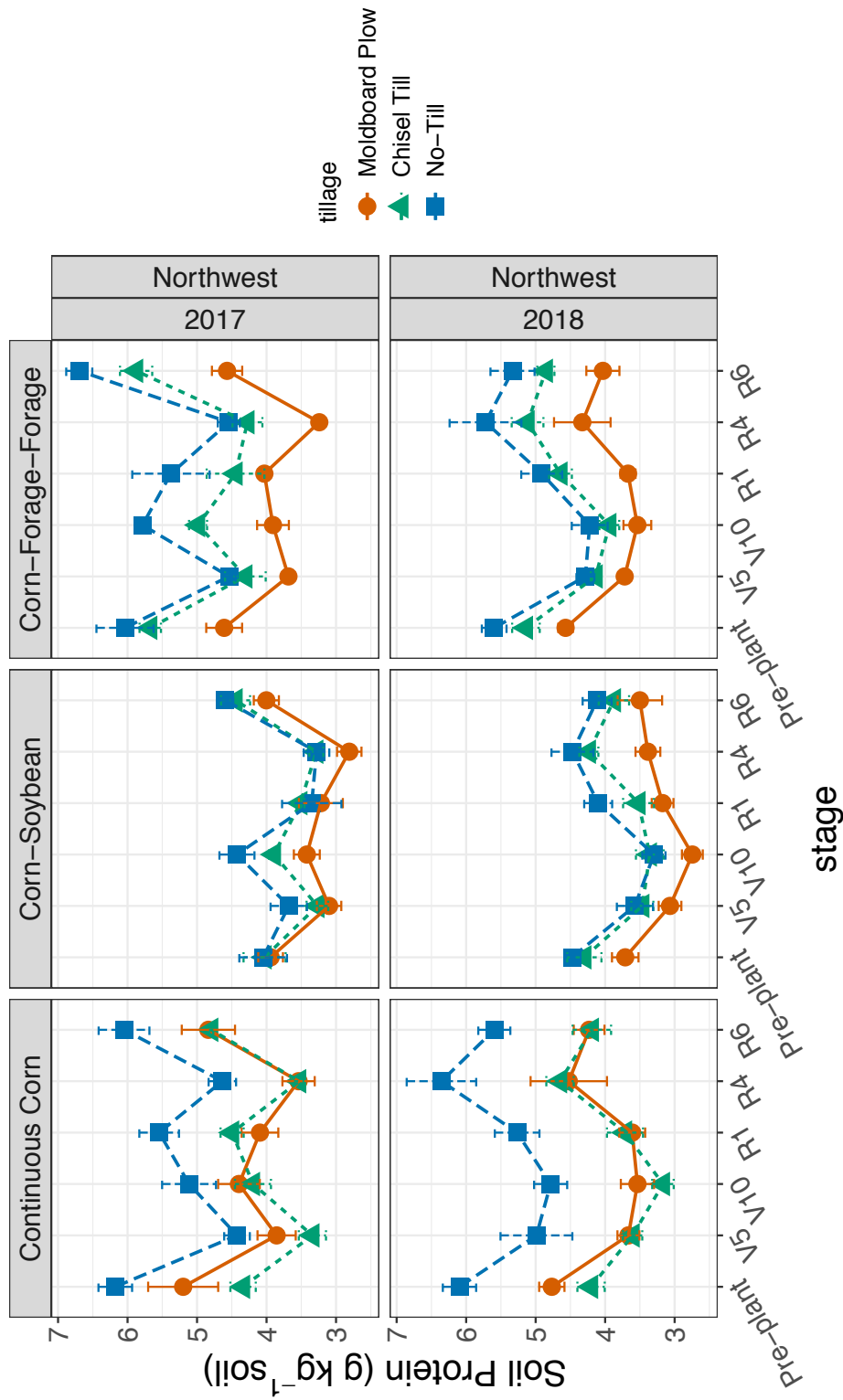


Figure 1. 8. Soil protein mean values at Northwest in the 2017 and 2018 growing seasons for three tillage regimes in three crop rotations. Tillage practices include moldboard plow (red circles, solid line), chisel till (green triangles, dotted line), and no-till (blue squares, dashed line). Error bars represent one standard error of the mean.

Table 1. 21. Northwest 2017 soil protein tillage and rotations treatments means with standard errors for each sampling stage. Different letters within the same row represent significantly different treatments at $\alpha = 0.1$. F statistics and significance from analysis of variance for each stage.

Northwest Site 2017						
Variable	PP	V5	V10	R1	R4	R6
Soil Protein (g kg ⁻¹ soil)						
Tillage						
MP	4.6 ± 0.2 b	3.5 ± 0.1 b	3.9 ± 0.2 c	3.8 ± 0.2 b	3.2 ± 0.1 c	4.5 ± 0.2
CH	4.7 ± 0.3 b	3.6 ± 0.2 b	4.4 ± 0.2 b	4.2 ± 0.2 b	3.7 ± 0.2 b	5.0 ± 0.2
NT	5.4 ± 0.4 a	4.2 ± 0.2 a	5.1 ± 0.2 a	4.8 ± 0.4 a	4.2 ± 0.2 a	5.8 ± 0.3
Rotation						
CC	5.2 ± 0.3 a	3.9 ± 0.2 b	4.6 ± 0.2 a	4.7 ± 0.2 a	3.9 ± 0.2 a	5.2 ± 0.3
CS	4.0 ± 0.1 b	3.3 ± 0.1 c	3.9 ± 0.2 b	3.4 ± 0.2 b	3.1 ± 0.1 b	4.3 ± 0.1
CFF	5.4 ± 0.3 a	4.2 ± 0.2 a	4.9 ± 0.3 a	4.6 ± 0.3 a	4.0 ± 0.2 a	5.7 ± 0.3
ANOVA F statistics and significance						
Source						
Tillage(T)	6.7 **	14.4 ***	20.6 ***	6.9 **	24.8 ***	0.3 NS
Rotation (R)	19.2 ***	19.2 ***	13.9 ***	16.2 ***	25.6 ***	0.5 NS
T x R	4.1 *	3.4 *	2.5 .	1.6 NS	4.8 *	0.3 NS

“.” Significance level: p < 0.1 | * Significance level: p < 0.05 | ** Significance level: p < 0.01 | *** Significance level: p < 0.001 | “NS” Significance level: p > 0.1
 PP = pre-plant, V5 = fifth collared leaf, V10 = tenth collared leaf, R1 = silking, R4 = dough, R6 = physiological maturity
 Rotation: CC = continuous corn; CS = corn-soybean; CFF = corn-forage-forage | Tillage: MP = moldboard plow; CH = chisel till; NT = no-till

Table 1. 22. Northwest 2018 soil protein tillage and rotations treatments means with standard errors for each sampling stage. Different letters within the same row represent significantly different treatments at $\alpha = 0.1$. F statistics and significance from analysis of variance for each stage.

Northwest Site 2018						
Variable	PP	V5	V10	R1	R4	R6
Soil Protein (g kg ⁻¹ soil)						
MP	4.3 ± 0.2 b	3.5 ± 0.1 b	3.3 ± 0.2 b	3.5 ± 0.1 c	4.1 ± 0.3 c	3.9 ± 0.2 c
CH	4.5 ± 0.2 b	3.7 ± 0.1 b	3.5 ± 0.1 b	4.0 ± 0.2 b	4.7 ± 0.2 b	4.3 ± 0.2 b
NT	5.4 ± 0.3 a	4.3 ± 0.3 a	4.1 ± 0.2 a	4.8 ± 0.2 a	5.5 ± 0.4 a	5.0 ± 0.3 a
Rotation						
CC	5.0 ± 0.3 a	4.1 ± 0.3 a	3.8 ± 0.3 a	4.2 ± 0.3 a	5.2 ± 0.4 a	4.7 ± 0.3 a
CS	4.2 ± 0.1 b	3.4 ± 0.1 b	3.1 ± 0.1 b	3.6 ± 0.2 b	4.0 ± 0.2 b	3.8 ± 0.2 b
CFF	5.1 ± 0.2 a	4.0 ± 0.1 a	3.9 ± 0.1 a	4.4 ± 0.2 a	5.1 ± 0.3 a	4.7 ± 0.2 a

Source	ANOVA F statistics and significance					
Tillage (T)	29.8 ***	11.8 ***	15.5 ***	30.1 ***	28.5 ***	21.7 ***
Rotation (R)	26.5 ***	11.6 ***	15.0 ***	12.8 ***	21.7 ***	18.1 ***
T x R	7.4 **	3.2 *	5.5 **	3.1 *	2.7 .	2.9 .

“.” Significance level: p < 0.1 | * Significance level: p < 0.05 | ** Significance level: p < 0.01 | *** Significance level: p < 0.001 | “NS” Significance level: p > 0.1
 PP = pre-plant, V5 = fifth collared leaf, V10 = tenth collared leaf, R1 = silking, R4 = dough, R6 = physiological maturity
 Rotation: CC = continuous corn; CS = corn-soybean; CFF = corn-forage-forage | Tillage: MP = moldboard plow; CH = chisel till; NT = no-till

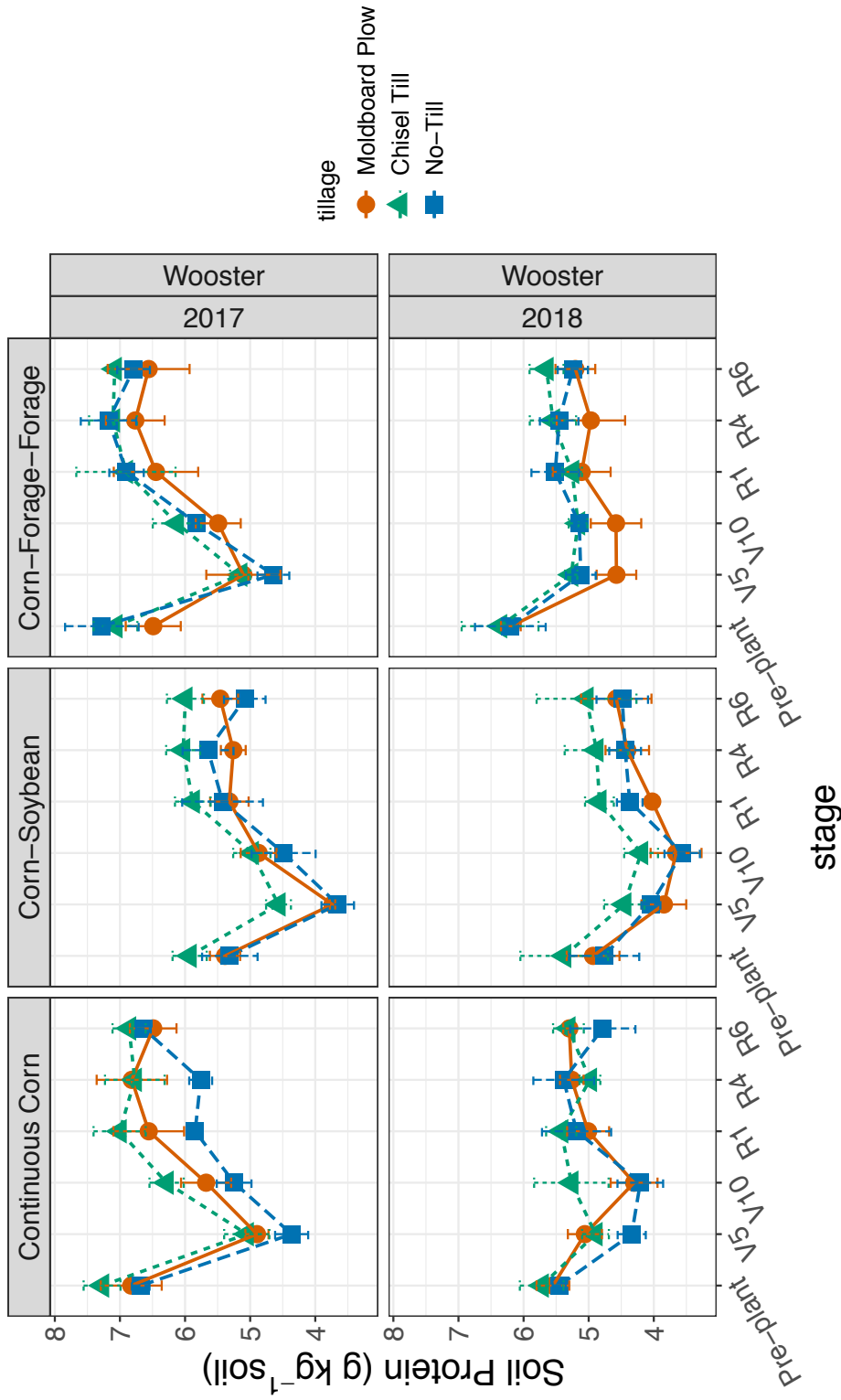


Figure 1. 9. Soil protein mean values at Wooster in the 2017 and 2018 growing seasons for three tillage regimes in three crop rotations. Tillage practices include moldboard plow (red circles, solid line), chisel till (green triangles, dotted line), and no-till (blue squares, dashed line). Error bars represent one standard error of the mean.

Table 1. 23. Wooster 2017 soil protein tillage and rotations treatments means with standard errors for each sampling stage. Different letters within the same row represent significantly different treatments at $\alpha = 0.1$. F statistics and significance from analysis of variance for each stage.

Wooster 2017						
Variable	PP	V5	V10	R1	R4	R6
Soil Protein (g kg ⁻¹ soil)						
Tillage						
MP	6.2 ± 0.3	4.6 ± 0.3 ab	5.3 ± 0.2 ab	6.1 ± 0.3	6.3 ± 0.3	6.2 ± 0.3
CH	6.8 ± 0.3	4.9 ± 0.2 a	5.8 ± 0.3 a	6.6 ± 0.3	6.6 ± 0.2	6.6 ± 0.2
NT	6.4 ± 0.4	4.2 ± 0.2 b	5.2 ± 0.3 b	6.1 ± 0.3	6.2 ± 0.3	6.2 ± 0.3
Rotation						
CC	6.9 ± 0.2 a	4.8 ± 0.2 a	5.7 ± 0.2 a	6.5 ± 0.3 a	6.4 ± 0.3 a	6.7 ± 0.1 a
CS	5.5 ± 0.2 b	4.0 ± 0.2 b	4.8 ± 0.2 b	5.5 ± 0.2 b	5.6 ± 0.2 b	5.5 ± 0.2 b
CFF	6.9 ± 0.3 a	5.0 ± 0.2 a	5.8 ± 0.2 a	6.8 ± 0.3 a	7.0 ± 0.2 a	6.8 ± 0.2 a
ANOVA F statistics and significance						
Source						
Tillage (T)	1.5 NS	4.6 *	2.8 .	1.2 NS	1.2 NS	2.2 NS
Rotation (R)	14.2 ***	10.0 **	9.5 **	5.2 *	10.0 **	14.8 ***
T x R	0.6 NS	0.6 NS	0.7 NS	0.5 NS	1.4 NS	0.4 NS

“.” Significance level: $p < 0.1$ | * Significance level: $p < 0.05$ | ** Significance level: $p < 0.01$ | *** Significance level: $p < 0.001$ | “NS” Significance level: $p > 0.1$
 PP = pre-plant, V5 = fifth collared leaf, V10 = tenth collared leaf, R1 = silking, R4 = dough, R6 = physiological maturity
 Rotation: CC = continuous corn; CS = corn-soybean; CFF = corn-forage-forage | Tillage: MP = moldboard plow; CH = chisel till; NT = no-till

Table 1. 24. Wooster 2018 soil protein tillage and rotations treatments means with standard errors for each sampling stage. Different letters within the same row represent significantly different treatments at $\alpha = 0.1$. F statistics and significance from analysis of variance for each stage.

Wooster 2018						
Variable	PP	V5	V10	RI	R4	R6
Tillage						
MP	5.6 ± 0.2	4.5 ± 0.2	4.2 ± 0.2 b	4.7 ± 0.2 b	4.9 ± 0.2	5.0 ± 0.2
CH	5.8 ± 0.3	4.9 ± 0.2	4.9 ± 0.3 a	5.2 ± 0.1 a	5.1 ± 0.2	5.3 ± 0.3
NT	5.5 ± 0.3	4.5 ± 0.2	4.3 ± 0.3 ab	5.0 ± 0.3 ab	5.1 ± 0.2	4.8 ± 0.2
Rotation						
CC	5.6 ± 0.1 ab	4.8 ± 0.2 a	4.6 ± 0.3 a	5.2 ± 0.2 a	5.2 ± 0.2 a	5.1 ± 0.2 ab
CS	5.0 ± 0.3 b	4.1 ± 0.2 b	3.8 ± 0.2 b	4.4 ± 0.1 b	4.6 ± 0.2 b	4.7 ± 0.3 b
CFF	6.2 ± 0.2 a	5.0 ± 0.2 a	5.0 ± 0.2 a	5.3 ± 0.2 a	5.3 ± 0.2 a	5.4 ± 0.2 a
ANOVA F statistics and significance						
Tillage (T)	0.3 NS	2.3 NS	3.5 .	2.6 NS	0.5 NS	2.0 NS
Rotation (R)	6.5 **	10.2 **	9.0 **	11.2 ***	4.4 *	3.7 *
T x R	0.3 NS	1.9 NS	0.7 NS	0.7 NS	0.7 NS	0.3 NS

“.” Significance level: $p < 0.1$ | * Significance level: $p < 0.05$ | ** Significance level: $p < 0.01$ | *** Significance level: $p < 0.001$ | “NS” Significance level: $p > 0.1$
 PP = pre-plant, V5 = fifth collared leaf, V10 = tenth collared leaf, RI = silking, R4 = dough, R6 = physiological maturity
 Rotation: CC = continuous corn; CS = corn-soybean; CFF = corn-forage-forage | Tillage: MP = moldboard plow; CH = chisel till; NT = no-till

Soil Penetration Resistance

Soil penetration resistance values were inconsistent across years and between sites (Table 1.25). Overall, the 2018 year showed more differences between treatments than the 2017 growing season. In Northwest, penetration resistance was higher in chisel and no-till soils compared to moldboard plow. Effects of rotation on soil penetration resistance were only observed in 2018 for Northwest where the CFF rotation had higher values than CC. In Wooster, differences between treatments were only observed during the 2018 growing season where chisel till, CC and CFF had higher soil penetration resistance.

Table 1. 25. Soil Penetration resistance for tillage and rotations treatments means with standard errors for the V5 stage. Different letters within the same column represent significantly different treatments at $\alpha = 0.1$. F statistics and significance from analysis of variance for each stage.

Variable	Northwest Site		Wooster Site	
	2017	2018	2017	2018
Soil Penetration Resistance (MPa)				
Tillage				
MP	1.75 ± 0.1 b	1.75 ± 0.1 b	3.35 ± 0.1	2.55 ± 0.1 b
CH	2.12 ± - a	2.37 ± 0.1 a	3.34 ± 0.2	2.93 ± 0.2 a
NT	2.29 ± 0.1 a	2.61 ± 0.2 a	3.20 ± 0.2	2.57 ± 0.1 b
Rotation				
CC	2.04 ± -	2.07 ± 0.1 b	3.42 ± 0.1	2.77 ± 0.1 a
CS	2.02 ± 0.1	2.26 ± 0.1 ab	3.38 ± 0.2	2.89 ± 0.1 a
CFF	2.09 ± 0.1	2.40 ± 0.3 a	3.09 ± 0.2	2.39 ± 0.1 b
ANOVA F statistics and significance				
Source	Northwest Site		Wooster Site	
Tillage(T)	19.93 ***	22.62 ***	0.23 NS	6.22 *
Rotation(R)	0.3 NS	3.2 .	1.01 NS	9.97 **
T x R	0.44 NS	7.16 **	0.14 NS	1.88 NS

Significance levels: “.” = $p < 0.1$ | * = $p < 0.05$ | ** = $p < 0.01$ | *** = $p < 0.001$ | NS = $p > 0.1$

Rotation: CC = continuous corn; CS = corn-soybean; CFF = corn-forage-forage

Tillage: MP = moldboard plow; CH = chisel till; NT = no-till

Table 1. 26. Summary of soil measurements results from Chapter 1

Soil Measurements	Northwest	Wooster
Soil Moisture	<ul style="list-style-type: none"> • Minimum differences between treatments, trends were inconsistent across years and stages. The NT CFF had the highest soil moisture in 2018. • Significant seasonal variation. 	<ul style="list-style-type: none"> • Tillage had stronger effect: as tillage intensity decreased, soil moisture increased. The CC and CFF rotations had highest soil moisture. • Significant seasonal variation.
POXC	<ul style="list-style-type: none"> • Crop Rotation had strongest effect, the CFF had highest POXC values. • POXC values are highest in NT under CFF rotation compared to CC and CS rotations, and to CH and MP tillage. • Significant seasonal variation. 	<ul style="list-style-type: none"> • Crop Rotation had strongest effect, the CFF had highest POXC values. • POXC values increase in NT soils under CFF compared to CS and CC rotations. POXC values are lowest in NT soils under CC rotation. • Significant seasonal variation.
C Min	<ul style="list-style-type: none"> • The CFF rotation and the NT soils, separately, promoted the highest C min values across all tillage and rotations. • The No-till soils under CFF rotation had the highest C min values • Significant seasonal variation. 	<ul style="list-style-type: none"> • The CFF rotation and the conservation tillage (CH and NT) treatments, separately, promoted higher C min values • No-till soils under CC had lowest Min C values, but as values increased in NT soils under CFF rotation • Significant seasonal variation.
Protein	<ul style="list-style-type: none"> • No-till soils had highest soil protein values across all rotations • CFF and CC rotations had the highest soil protein values • Significant seasonal variation. 	<ul style="list-style-type: none"> • Minimum tillage effects were observed in few stages where CH had higher values than NT in 2017 and than MP in 2018 • Rotation had stronger effect on soil protein where the CFF and CC rotations had the highest soil protein values • Significant seasonal variation.

Soil Chemical Properties	<ul style="list-style-type: none"> • Rotation had stronger effect where OM%, TN and TOC had higher values in the CFF rotation • pH was lower in the CC rotation and in NT soils • CEC, OM%, are higher in NT and CH compared to MP • TN and TOC are higher in NT soils compared to CH and MP 	<ul style="list-style-type: none"> • Rotation had stronger effect where CEC was higher in CFF and CC, TN was higher in CFF compared to CS, and TOC was higher in CFF and CC. • pH was lower in the CC rotation • No significant differences were observed in OM% on tillage or rotation effects. • Tillage did not affect TOC, pH, and CEC.
Penetration Resistance	<ul style="list-style-type: none"> • Tillage had stronger effect where NT and CH had higher values than MP • Rotation effects only significant in 2018 where CFF values were higher than CC. 	<ul style="list-style-type: none"> • Tillage effects only observed in 2018 where CH had higher values than MP and NT • Rotation effects only significant in 2018 where CC and CS values were higher than CFF.

Rotation: CC = continuous corn; CS = corn-soybean; CFF = corn-forage-forage

Tillage: MP = moldboard plow; CH = chisel till; NT = no-till

POXC = permanganate oxidizable carbon; Min C = mineralizable carbon; Protein = soil protein

1.4 Discussion

Soil Health Temporal Dynamics

One of our objectives was to examine soil C and N temporal dynamics over a corn growing season under 55+ years of tillage (no-till, chisel till, moldboard plow) and rotations (corn monoculture, corn-soybean, corn-forage-forage) histories. Our results indicate that the significant temporal variability of the soil health indicators under these long-term treatments might suggest that active organic matter stabilization and mineralization, and nutrient cycling throughout the growing season changes as the plants nutrient requirements change. Variability in temporal dynamics of POXC and Min C have also been reported in Culman et al., 2013 where a more diverse rotation (corn-soy-wheat) had higher values across plant growth stages than continuous corn. Differences between crop growth stages in all soil temporal measurements were consistent and strongly significant ($p < 0.001$) suggesting the time of year when soil is sampled can significantly impact the value measured.

The F statistics from analysis of variance with stage as repeated measures can provide a measure of relative magnitude of variability across measured variables. Soil moisture was the most temporally variable soil measurement followed by soil protein, and POXC and Min C (Soil moisture > Protein > POXC = Min C). We expected soil moisture to be significantly different within stages in both sites as different corn growth stages require different levels of water, as air temperature increases, precipitation varies,

and evapotranspiration rates change in the growing season. However, we expected that soil protein would have the least temporal variability out of the soil health indicators measured due to findings in (Hurisso et al., 2018a) where soil protein had the least temporally variable results over different stages of a growing season in sites in the same regions studied in this project. Nevertheless, our study had more stages sampled in the Hurisso et al (2018a) study and may provide a better or a more detailed insight into soil health indicators temporal dynamics over a corn growing season. High variability in soil protein across the growing season in both sites may be due to changes in active organic matter mineralization and nitrogen loss or uptake by plants at different stages and may suggest that plants are taking nitrogen from the soil protein pool after mineralization. Results for Min C and POXC were less variable between stages compared to protein; however, trends were highly variable by year and site. Inconsistent trends in these soil health indicators temporal dynamics may be due to differences in rainfall, soil moisture, soil texture (with Northwest having more clay than Wooster), or rhizodeposition and root turnover across the growing season. Variable temporal trends for soil protein, POXC and min C are also observed in Hurisso et al (2018a) study between sites.

Tillage Effects on Soil health indicators

The first component of our first objective was to evaluate the effects of different tillage intensities (moldboard plow, chisel till, and no-till) on labile C and N soil health indicators after 55+ continuous years. In general, no-till and chisel tillage (conservation

tillage) had higher values for the soil health indicators (POXC, Respiration and Protein) compared to tilled soils, suggesting that organic matter stabilization or accumulation and mineralization are higher in less intensive tillage and zero tillage compared to intensive tillage (moldboard plow). Hurisso et al (2016) proposed that POXC better reflects soil-conservation-oriented practices suggesting that reduced soil disturbance influenced POXC more than Min C compared to conventional tillage (Hurisso et al., 2016). The higher levels of labile carbon and nitrogen pools on less reduced tillage and/or no-till soils compared to intensive tillage have been reported in other long-term studies (Idowu et al., 2009; Karlen et al., 2013a; Van Eerd et al., 2014; Zuber and Villamil, 2016).

Tillage had more consistent effects in Northwest where NT had higher values for all the soil health indicators (POXC, Respiration and Protein) compared to tilled soils. Our results suggest that soils with higher clay in Northern Ohio might have a greater capacity for accumulating and mineralizing organic matter, especially in non-disturbed soils. Also, although the Northwest soil is tile drained it still has naturally poor drainage which can slow the organic matter decomposition rate and allow higher organic matter accumulation compared to a well-drained soil like Wooster. We can see in the soil chemical properties as well that Northwest had higher CEC, TOC and TN in no-till soils compared to Wooster. Although rotation had a stronger effect in POXC, we found in concordance with Hurisso et al (2016) that reduced tillage promoted higher POXC values in Northwest and that tillage had a stronger effect in min C (Table 1.12); however, unlike Hurisso et al (2016), no-till promoted the highest C mineralization in Northwest and not tillage. Zuber et al., (2016) summarized that no-till soils had similar microbial respiration to chisel

tilled soils and explains that eventually, in long term systems microbes (which mineralize the soil organic matter) in no-till soils may become as active as those in chisel tilled soils.

The high soil health values in NT soils may also be due to well-developed soil structure and aggregation. Previous studies show that long-term no-till or minimum tillage promote higher soil aggregate stability compared to intensive tillage (Karlen et al., 1994; Congreves et al., 2015). Although we did not measure soil structure and/or aggregation, our soil penetration resistance results showed that less disturbed soils (CH and NT) had higher soil penetration resistance compared to moldboard plow in a 0-20 cm depth. These values concur with (Burgos Hernández et al., 2019) findings where NT soils were more compact than MP and CH in the same Northwest site. These results might indicate better soil aggregation or structure and soil natural compaction processes (like shrinking and swelling) during the corn early stages (V5) in NT and CH compared to MP soils that may still be loose from the inversion tillage.

The soil health indicators values in Wooster were generally higher for the chisel till, especially Min C and soil protein, but higher POXC was documented in the no-till soils. Like Northwest, the higher POXC in the no-till soils might suggest that zero disturbance of the soil promotes organic matter stabilization or accumulation, but in this case not necessarily the mineralization of that active pool or the organic matter in NT soils. This may suggest that active organic matter is not getting mineralized as much as it is getting accumulated or stabilized in the no-till soils in the Wooster silt loam. Also, this might suggest that the physical disturbance of the chisel till helps in breaking down protected

organic matter and easing the accessibility of the labile C and N pools to microorganisms to mineralize, making the chisel till have more organic matter mineralization and soil protein and less active organic matter stabilization. These findings match with Hurisso et al (2016) study where tillage promotes enrichment of mineralizable carbon, and higher POXC values in the no-till soils reflect accumulation or stabilization of the soil organic matter and potentially long-term carbon sequestration. Also, similar results were summarized in Zuber and Villamil, (2016) where a higher soil microbial metabolic quotient, which is the respiration to microbial biomass ratio and indicator of microbial activity, was documented in chisel till soils compared to no-till soils.

The effects of the organic matter accumulation are observed in the soil moisture in Wooster, where soil moisture was higher in the no-till soils across all rotations in this naturally well drained soil. This organic matter accumulation may result in increased soil moisture retention in well drained soils like Wooster especially in corn later growth stages (V10, R1) as evapotranspiration rates increase, and monthly rainfall can be highly variable during the growing season. Although POXC was higher in NT, no significant differences in OM% and TOC were observed between tillage treatments in Wooster. Interestingly, the soil CH had higher penetration resistance than NT and MP in 2018 and no differences between tillage treatments were observed in 2017 in Wooster. Although we expected for CH to have higher soil penetration resistance values than MP at corn V5 stage, we were not expecting for NT and MP to have similar values since soils are tilled during the spring and expected to be less compacted than non-disturbed soils in the crop early stages. These findings are different from (Burgos Hernández et al., 2019) and may

suggest that adjustments in soil penetration resistance may be done for looking at the bare effect of the treatments on the soil compaction by accounting for the soil moisture since soil water content strongly influences soil penetration resistance (Hamza and Anderson, 2005).

Rotation effects on soil health indicators

Part of our first objective was to evaluate the effects of different crop rotations (continuous corn (CC), corn-soybean (CS), and corn-forage-forage (CFF)) on labile C and N soil health indicators after half a century of continuous treatments. In general, the Corn-Forage-Forage and the Continuous Corn rotations had higher values for the labile C and N pools (POXC, Respiration and Protein). These results suggest that active organic matter accumulation or stabilization, mineralization and potential N availability are higher in rotations including two years of forages like oats, alfalfa and red clover, and in corn monoculture compared to a Corn-Soybean rotation. Similar results of POXC and Min C increasing with increasing rotation diversity are observed in Culman et al., (2013).

We expected the CC rotation to have the lowest C min results for both sites since these plots are fertilized yearly with nitrogen and N fertilization has been shown to minimize or inhibit carbon mineralization in soils (Mahal et al., 2019). However, there were no significant differences between the CFF and CC rotations in C mineralization for the majority of the stages sampled. An increase of active organic matter stabilization, mineralization, and potential N availability in the CFF rotation may indicate a more

biologically based activity since this rotation receives less fertilizer input than the other rotations. The high soil protein values in the CC may be due to yearly corn nitrogen fertilization. Interestingly, the corn-soybean rotation, which is one of the most common rotations in the US Midwest, had consistently the lowest soil health values compared to the other rotations, indicating that this rotation promotes lower active organic matter accumulation and mineralization compared to corn monoculture and corn-forage-forage. Similar results were found in Karlen et al., (2006) study where rotation including forage crops in the Midwest corn belt area had higher soil quality compared to corn-soybean rotation. Another study found similar results in rotations which included alfalfa or winter wheat had higher soil health than soils under corn monoculture and corn-soybean rotations (Congreves et al., 2015).

Overall, rotation effects on all soil properties measured were stronger and more consistent than tillage effects, except for min C that was more influenced by tillage. The consistently higher soil health values in the CFF rotation might be due to higher root density, rhizodeposition and turnover. The accumulation or stabilization of active organic matter primarily in the CFF and some times in the CC rotation may influence soil moisture since soil moisture values were higher under these rotations. Significant differences in soil penetration resistance between rotations were only observed in 2018 and were overall inconsistent. Higher values for soil penetration resistance were observed in NW for the CFF rotation compared to CC, but Wooster showed higher values for CC and CS rotations compared to CFF rotation.

Tillage and Rotation interactions effects on soil health

On our objective to evaluate 55+ continuous years of different tillage intensities and crop rotation diversity in soil active organic matter dynamics, we have to not only study the treatments individual effects but their interactions as well. Although trends were clearer in Northwest, in both sites the no-till soils under the CFF rotation had higher POXC (Figure 1.5; Figure 1.6), Min C (Figure 1.7; Figure 1.8), and soil protein (Figure 1.9; Figure 1.10) compared to NT under corn monoculture and corn-soybean. These results may suggest that under zero tillage, having two years of perennial forages like alfalfa and red clover for 55+ years increases soil active organic matter stabilization or accumulation, nutrient mineralization and potential availability (active labile C and N pools, nutrient cycling) compared to corn monoculture and corn-soybean rotation. Another study also found that increasing crop rotation diversity in no-till soils increased soil organic carbon and total nitrogen compared to less diverse rotations (Alhameid et al., 2017). On the contrary, eliminating or reducing crop diversity in no-till soils demonstrated to promote lower soil health indicators than tilled soils, suggesting less active organic matter accumulation and mineralization across the growing season. Higher POXC and mineralization values in NT compared to tilled soils were also observed in other studies with crop rotations including legumes (Morrow et al., 2016). However, another study reported POXC to be not significantly different in NT under diverse crop

rotation compared to corn monoculture after long-term continuous treatments (Van Eerd et al., 2014). Additionally, in the current study in the CFF rotation, Moldboard plow often had the lowest soil health values compared to chisel and no-till suggesting long-term intensive tillage is not beneficial for the soil dynamic quality.

1.5 Conclusions

Maintaining good soil health in agroecosystems is essential for agricultural and environmental sustainability. In this study, we evaluated the effects of half a century of continuous and different tillage intensities and crop rotation diversity. Overall, temporal variability of the soil health indicators under these long-term treatments might suggest active organic matter stabilization and mineralization, and nutrient cycling throughout the growing season. In general, no-till and chisel tillage had higher values for the soil health indicators compared to tilled soils, and Corn-Forage-Forage and the Continuous Corn rotations had higher values for the labile C and N pools (POXC, Respiration and Protein) suggesting that organic matter stabilization or accumulation and mineralization are higher in reduced and zero tillage compared to intensive tillage and in systems including two years of forages like alfalfa and red clover, and corn monoculture compared to a Corn-Soybean rotation. Our results suggest that implementing less intensive or zero tillage and

including forages like alfalfa, oats and red clover in similar soils helps in improving soil health and in the long-term in corn cropping systems.

Chapter 2. Does soil health relate to crop temporal dynamics and productivity in soils under half-century of no-till and crop rotations?

2.1 Introduction

Corn (*Zea mays*) is one of the most important crops in the world and its development under different management histories have been extensively studied. Tillage and crop rotations are examples of these managements that have been known to impact corn development and productivity. For instance, moldboard plow is a type of intensive tillage that was commonly used in temperate soils for incorporating the vegetative residues from the past harvest and controlling weeds, and for accelerating soil warming and evaporation in the spring, ultimately with the goal of improving soil conditions for seed emergence. Although this type of tillage was very successful at the beginning, research has found that in the long term, plowing lowers the quality of the soil negatively affecting crop production (Reicosky et al., 2011; Baumhardt et al., 2015). Reduced soil quality, including increased soil erosion and reduction of the soil organic matter, are factors known to decrease crop development and productivity even in well fertilized soils (Oldfield et al., 2019). Therefore, reduced tillage and zero tillage are practices that have been proposed for improving or at least maintaining the quality of the soil and prevent it from negative effects, as part of efforts for soil conservation.

Studies have claim that implementing reduced and, especially no-tillage in corn agronomic production with crop rotations can not only improve the soil quality, but also produce higher, equal or very similar yields as tilled soils and monoculture (Daigh et al., 2018; Nunes et al., 2018). However, the long term impact of no-till and diverse crop rotations on corn productivity are highly variable by region, soil type (Nunes et al., 2018), and climate (Toliver et al., 2012). Also, many studies contradict no-till having higher or equal crop productivity compared to tilled soils (Ogle et al., 2012; Toliver et al., 2012; Pittelkow et al., 2015). Some studies have found that no-till usually has lower yields in the first years of implementation but that productivity increases with time (Dick et al., 1986). Different findings on the effect of no-till in soil quality and corn yields complicates the understanding and recommendations of soil management for the scientific and agricultural communities.

Although many studies have been evaluated the effects of no-till and rotations on corn productivity and results have been variable, not all studies have evaluated these treatments in long-term trials and gaps still remain on understanding no-till and crop rotation interaction effects. Our study aimed to evaluate the effects of 55+ years of continuous tillage intensities (moldboard plow (most intense), chisel till, and no-till (least intense)) and crop rotation diversity (continuous corn, corn-soybean, and corn-forage-forage) on corn temporal dynamics and productivity to better understand these long-term effects in corn productivity in soils of Ohio. Also, our study aimed to describe the relationship between soil health indicators and corn productivity to potentially answer the question: are soil health indicators related to corn productivity? We hypothesized that

corn leaf chlorophyll, total plant biomass, total plant nitrogen uptake, and yields will be higher in no-till soils under the 3-year corn-forage-forage rotation. We also hypothesized that corn total biomass, total nitrogen uptake, and corn yield will be strongly related to soil health parameters.

2.2 Materials and Methods

Plant Samplings and Measurements

All corn samplings and measurements coincided with the same dates as soil samplings starting at the V5 stage and ending with the R6 stage (Table 2.1). Corn plants were staged using the leaf collar method (Abendroth et al., 2011). Corn whole plant aboveground biomass was collected by cutting at the base of the plant immediately above the brace roots. Five plants were collected per plot during the vegetative stages (V5, V10) and four plants were collected from the reproductive stages (R1, R4, R6) (Abendroth et al., 2011). After collection, plant samples were oven-dried at 40°C for 1.5 to 2 weeks. During the R4 and R6 stage samplings, the corn ears were separated from the vegetative part and quantified separately. After dried, the vegetative parts of the samples were weighted and chipped and a subsample was sent to a commercial laboratory (Spectrum Analytics, OH) where subsamples were ground and total N was quantified via dry combustion. The corn ears were managed separately after drying, grain was separated

from the husks and cobs, weighed and analyzed for total N. The plant biomass in kg ha^{-1} was estimated for each plot by using the following calculation:

$$(\text{weight of 1 plant kg}) \times (79074 \text{ plants/ha})$$

The 79074 plants/ha number represent an estimate of the mean plants planted per acre. The plant biomass was calculated separately for the vegetative and reproductive parts. The total plant biomass was calculated by the summation of the vegetative and the grain biomass. Total plant nitrogen uptake was estimated with the following calculation:

$$(\% \text{ total N} \times \text{total plant biomass}) / 100$$

Leaf chlorophyll content was measured indirectly by using a SPAD-502 meter (Konica Minolta, Ramsey, NJ) on the uppermost fully collared leaf for the V5 and V10 stages and on the leaf above the primary ear for the R1 and R4 samplings. The average of 10 randomly selected leaves were measured between the central vein and the edge of the leaf for each plot. Corn grain from the middle 2 plots were harvested with a harvest combine and adjusted to 15.5% moisture.

Table 2.1. Soil and corn samplings corresponding development stages during the 2017 and 2018 growing seasons.

Sampling	Plant Stage	Northwest Site		Wooster Site		Measurements Taken
		2017	2018	2017	2018	
Fifth leaf	V5	21-Jun	19-Jun	27-Jun	18-Jun	Soil, whole plant aboveground biomass, leaf chlorophyll
Tenth leaf	V10	19-Jul	July 18	26-Jul	19-Jul	Soil, whole plant aboveground biomass, leaf chlorophyll
Silking	R1	8-Aug	Aug 2	9-Aug	Aug 1	Soil, whole plant aboveground biomass, leaf chlorophyll
Dough	R4	31-Aug	Aug 30	Sep 7	Sep 5	Soil, whole plant aboveground biomass, leaf chlorophyll
Physiological Maturity	R6	2-Oct	Oct 15	16-Oct	Oct 4	Soil, whole plant aboveground biomass

2.3 Results

Chlorophyll

Although the effects of tillage and rotation varied between years and sites (Table 2.2), overall mean leaf chlorophyll values were higher in no-till and chisel till soils compared to moldboard plow and in the CFF rotation compared to the CC and CS rotation (Table 2.3, Table 2.4, Table 2.5, Table 2.6). Results were clearer in Northwest whereas rotation diversity increased, and tillage intensity decreased, leaf chlorophyll values slightly increased (Figure 2.1). Most leaf chlorophyll values peak at corn stage V10 in Northwest. In Wooster, leaf chlorophyll values were less consistent between treatments, but overall no-till soils had lower leaf chlorophyll values in the CC rotation compared to other tillage in 2017, and the CFF rotation had greater chlorophyll values compared to the CC rotation in 2018 (Figure 2.2). Similar to Northwest, leaf chlorophyll values peak at corn stage V10 for 2017 in Wooster; however, peaks are observed in corn R1 for 2018.

Correlation coefficients between leaf chlorophyll and the soil health indicators were highly variable across stages, sites and years (Table 2.7). Looking at all the sites and years, correlation coefficients for leaf chlorophyll and the soil health indicators (POXC, Min C, soil protein) were commonly stronger (positive) during the V10 stage. In Northwest 2018, correlation coefficients between leaf chlorophyll and the soil health indicators were all stronger (positive) during the V10 and R1 stages, but some results

varied for 2017 where Min C had stronger values during V5, and soil protein during V5 and R6. In Wooster 2017 all correlation coefficients were higher during the V10 stage, but for 2018 values varied with POXC having strong correlation also during V5, Min C similar values between all stages, and soil protein having a higher result during V5.

Table 2.2. F-statistics and significance of leaf chlorophyll, total plant aboveground biomass and total plant nitrogen uptake from repeated measures ANOVA of all five sampling stages by year (n=108 per site)

Effect	Leaf Chlorophyll		Total Plant Aboveground Biomass		Total Plant Nitrogen Uptake	
	2017	2018	2017	2018	2017	2018
Tillage (T)	3.2 *	7.3 **	1.7 NS	0.8 NS	3.3 *	0.5 NS
Rotation (R)	18.9 ***	37.9 ***	11.8 ***	12.0 ***	16.9 ***	11.2 ***
T x R	1.1 NS	3.1 *	1.5 NS	3.0 *	2.2 .	0.8 NS
Stage (S)	75.3 ***	199.7 ***	214.5 ***	485.3 ***	262.7 ***	279.0 ***
S x T	10.3 ***	2.5 *	1.0 NS	0.7 NS	1.5 NS	0.2 NS
S x R	3.6 **	6.4 ***	3.1 **	1.9 .	5.1 ***	1.8 .
S x T x R	0.8 NS	9.0 ***	0.6 NS	0.9 NS	0.9 NS	0.5 NS
Northwest site						
Wooster site						
Tillage (T)	1.5 NS	3.0 .	7.8 ***	12.3 ***	6.9 **	0.8 NS
Rotation (R)	0.9 NS	4.6 *	2.7 .	13.9 ***	3.4 *	29.6 ***
T x R	3.3 *	0.4 NS	0.0 NS	3.0 *	0.2 NS	1.8 NS
Stage (S)	691.9 ***	209.0 ***	482.4 ***	1203.9 ***	537.9 ***	1196.9 ***
S x T	5.3 ***	0.8 NS	1.8 .	2.1 *	2.2 *	1.9 .
S x R	1.1 NS	0.9 NS	1.0 NS	1.4 NS	1.0 NS	2.7 *
S x T x R	1.8 .	1.7 .	1.4 NS	0.9 NS	1.2 NS	1.4 NS

“.” Significance level: P < 0.1 | * Significance level: P < 0.05 | ** Significance level: P < 0.01 | *** Significance level: P < 0.001 | “NS” Significance level: p > 0.1

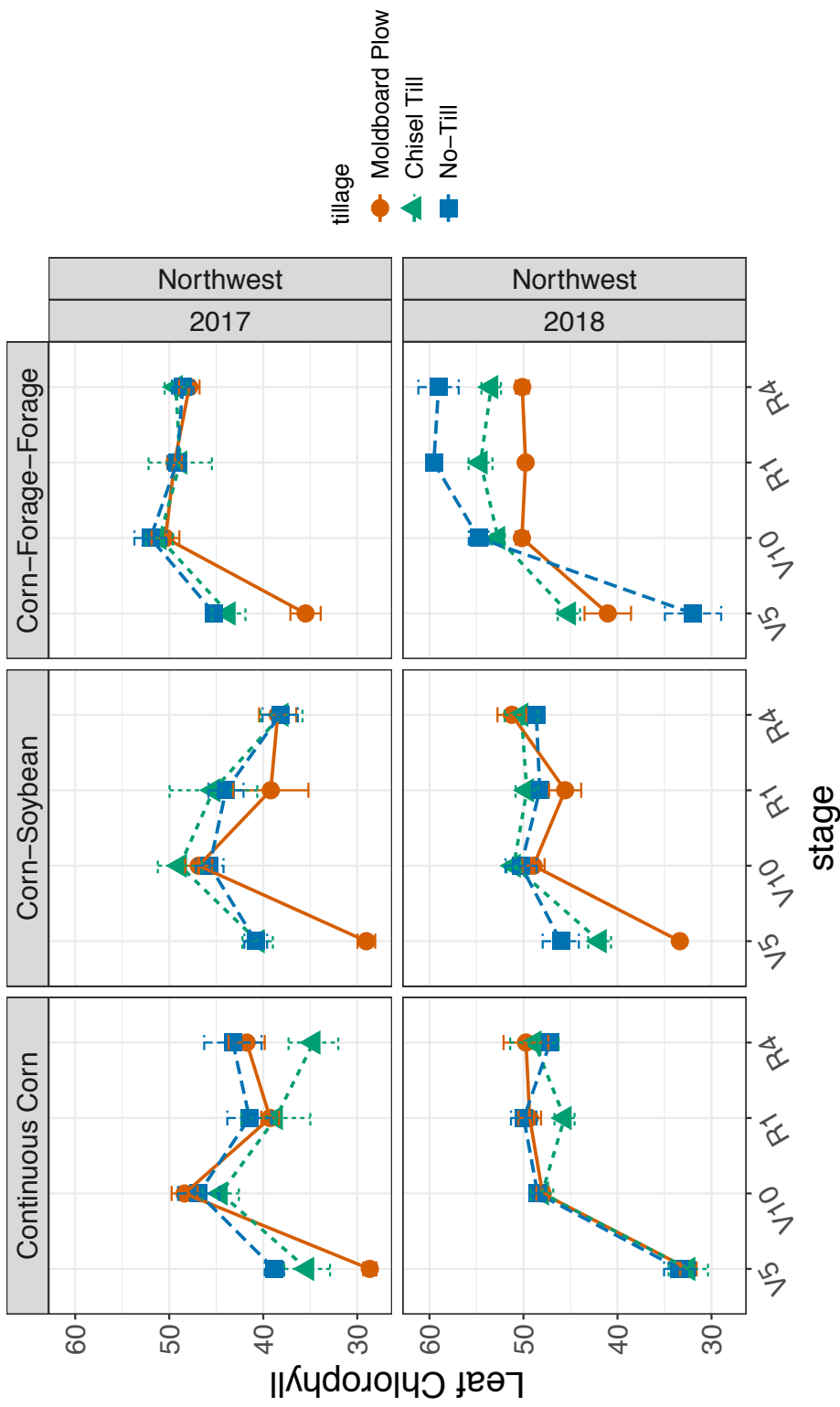


Figure 2.1. Leaf Chlorophyll (SPAD units) means at Northwest in the 2017 and 2018 growing seasons for three tillage regimes in three crop rotations. Tillage practices include moldboard plow (red circles, solid line), chisel till (green triangles, dotted line), and no-till (blue squares, dashed line). Error bars represent one standard error of the mean.

Table 2.3. Leaf chlorophyll at Northwest in 2017 by tillage and rotation treatments for each sampled crop developmental stage (mean \pm standard error). Different letters within the same column of rotation or treatment values represent significantly different treatments ($\alpha = 0.1$). F-statistics and significance from analysis of variance for each crop stage.

Northwest 2017				
Variable	V5	V10	R1	R4
Tillage				
MP	31.1 \pm 1.2 b	48.5 \pm 0.9	42.6 \pm 2.1	42.7 \pm 1.6
CH	39.9 \pm 1.6 a	48.1 \pm 1.4	44.3 \pm 2.5	40.7 \pm 2.5
NT	41.6 \pm 1.1 a	48.2 \pm 1.3	44.9 \pm 1.5	43.3 \pm 1.8
Rotation				
CC	34.3 \pm 1.7 c	46.6 \pm 1.1 b	39.8 \pm 1.4 b	39.9 \pm 1.8 b
CS	36.8 \pm 2.0 b	47.1 \pm 1.0 b	42.8 \pm 2.1 b	38.2 \pm 1.0 b
CFF	41.5 \pm 1.7 a	51.1 \pm 0.8 a	49.1 \pm 1.0 a	48.6 \pm 0.6 a
ANOVA F statistics and significance				
Tillage (T)	52.4 ***	0.1 NS	0.5 NS	1.4 NS
Rotation (R)	21.8 ***	6.1 *	8.3 **	22.9 ***
T x R	0.9 NS	1.1 NS	0.5 NS	2.0 NS

“,” Significance level: $p < 0.1$ | * Significance level: $p < 0.05$ | ** Significance level: $p < 0.01$ | *** Significance level: $p < 0.001$ | “NS” Significance level: $p > 0.1$
V5 = fifth collared leaf, V10 = tenth collared leaf, R1 = silking, R4 = dough | Rotation: CC = continuous corn; CS = corn-soybean; CFF = corn-forage-forage
Tillage: MP = moldboard plow; CH = chisel till; NT = no-till

Table 2.4. Leaf chlorophyll at Northwest in 2018 by tillage and rotation treatments for each sampled crop developmental stage (mean \pm standard error). Different letters within the same column of rotation or treatment values represent significantly different treatments ($\alpha = 0.1$). F-statistics and significance from analysis of variance for each crop stage.

Northwest 2018				
Variable	V5	V10	R1	R4
Tillage				
	Leaf Chlorophyll (SPAD units)			
MP	35.6 \pm 1.6 b	49.0 \pm 0.5 b	48.2 \pm 0.9 b	50.4 \pm 0.9 b
CH	39.8 \pm 2.1 a	50.6 \pm 0.8 ab	49.9 \pm 1.4 b	50.8 \pm 1.2 b
NT	37.1 \pm 2.5 ab	51.2 \pm 1.1 a	52.6 \pm 1.8 a	51.6 \pm 2.0 a
Rotation				
CC	32.8 \pm 0.8 b	48.2 \pm 0.4 c	48.3 \pm 0.9 b	48.6 \pm 1.1
CS	40.4 \pm 2.0 a	50.1 \pm 0.7 b	47.8 \pm 0.9 b	50.0 \pm 0.8
CFF	39.4 \pm 2.3 a	52.5 \pm 0.8 a	54.6 \pm 1.5 a	54.2 \pm 1.5
ANOVA F statistics and significance				
Source				
Tillage (T)	4.8 *	3.8 *	12.7 ***	0.4 NS
Rotation (R)	17.9 ***	14.7 ***	37.7 ***	9.2 **
T x R	12.8 ***	1.4 NS	8.3 ***	4.0 *

“.” Significance level: $p < 0.1$ | * Significance level: $p < 0.05$ | ** Significance level: $p < 0.01$ | *** Significance level: $p < 0.001$ | “NS” Significance level: $p > 0.1$
V5 = fifth collared leaf, V10 = tenth collared leaf, R1 = silking, R4 = dough | Rotation: CC = continuous corn; CS = corn-soybean; CFF = corn-forage-forage
Tillage: MP = moldboard plow; CH = chisel till; NT = no-till

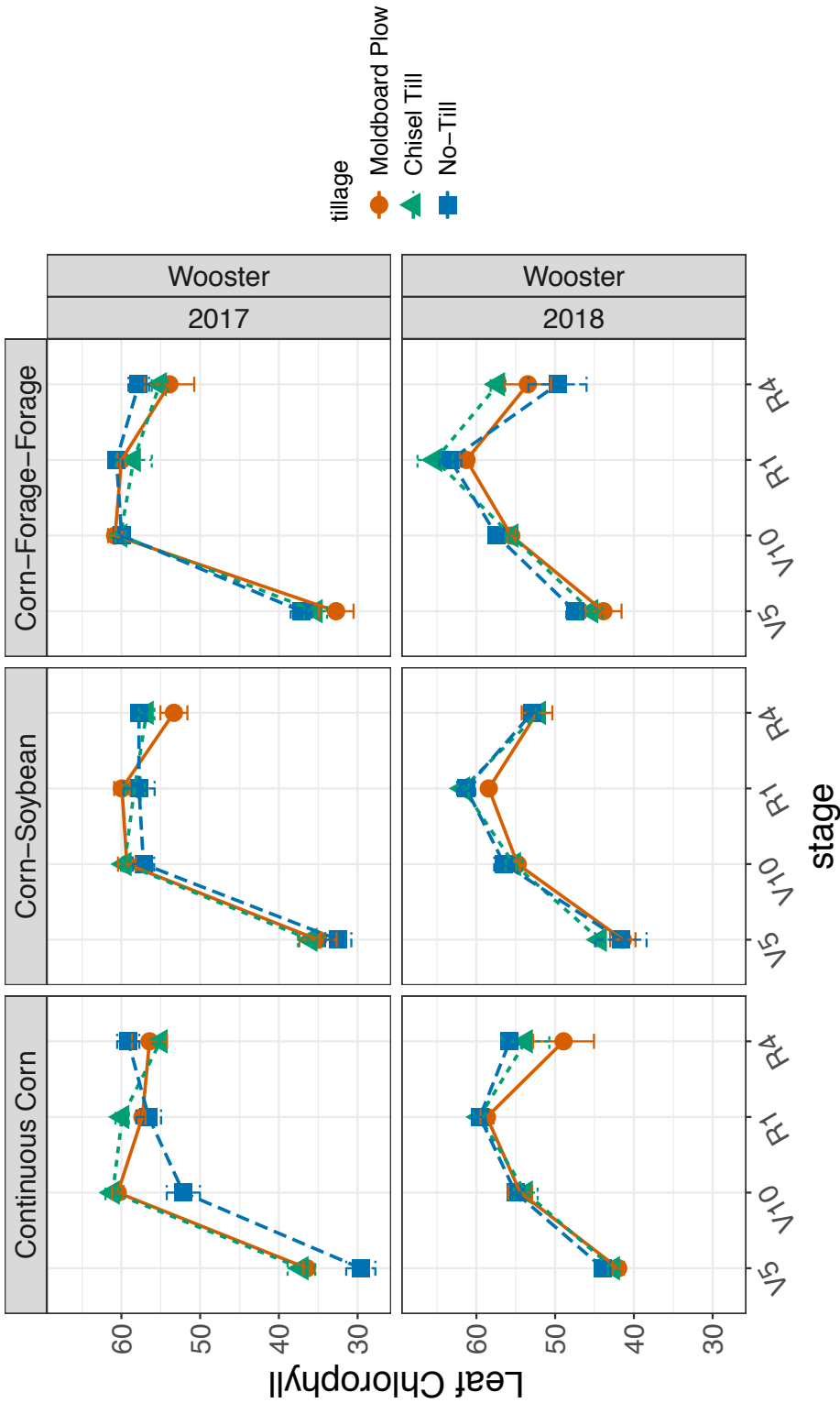


Figure 2.2. Leaf Chlorophyll (SPAD units) means at Wooster in the 2017 and 2018 growing seasons for three tillage regimes in three crop rotations. Tillage practices include moldboard plow (red circles, solid line), chisel till (green triangles, dotted line), and no-till (blue squares, dashed line). Error bars represent one standard error of the mean.

Table 2.5. Leaf chlorophyll at Wooster in 2017 by tillage and rotation treatments for each sampled crop developmental stage (mean \pm standard error). Different letters within the same column of rotation or treatment values represent significantly different treatments ($\alpha = 0.1$). F-statistics and significance from analysis of variance for each crop stage.

Wooster 2017				
Variable	V5	V10	R1	R4
Tillage				
MP	34.7 \pm 1.1	60.2 \pm 0.5 a	59.1 \pm 0.6	54.6 \pm 1.3 b
CH	36.2 \pm 0.8	60.3 \pm 0.4 a	58.9 \pm 0.8	55.7 \pm 0.5 ab
NT	33.1 \pm 1.4	56.4 \pm 1.4 b	58.3 \pm 1.0	58.2 \pm 0.7 a
Rotation				
CC	34.4 \pm 1.4	57.9 \pm 1.6 b	57.9 \pm 0.7	56.9 \pm 1.0
CS	34.5 \pm 1.1	58.7 \pm 0.6 ab	58.7 \pm 0.8	56.0 \pm 0.9
CFF	35.1 \pm 1.1	60.3 \pm 0.3 a	59.7 \pm 0.8	55.6 \pm 1.2
ANOVA F statistics and significance				
Tillage (T)	2.4 NS	13.1 ***	0.3 NS	3.9 *
Rotation (R)	0.1 NS	4.1 *	1.3 NS	0.5 NS
T x R	3.2 *	5.7 **	1.5 NS	0.6 NS

“.” Significance level: $p < 0.1$ | * Significance level: $p < 0.05$ | ** Significance level: $p < 0.01$ | *** Significance level: $p < 0.001$ | “NS” Significance level: $p > 0.1$
V5 = fifth collared leaf, V10 = tenth collared leaf, R1 = silking, R4 = dough | Rotation: CC = continuous corn; CS = corn-soybean; CFF = corn-forage-forage
Tillage: MP = moldboard plow; CH = chisel till; NT = no-till

Table 2.6. Leaf chlorophyll at Wooster in 2018 by tillage and rotation treatments for each sampled crop developmental stage (mean \pm standard error). Different letters within the same column of rotation or treatment values represent significantly different treatments ($\alpha = 0.1$). F-statistics and significance from analysis of variance for each crop stage.

Wooster 2018				
Variable	V5	V10	R1	R4
Tillage				
Leaf Chlorophyll (SPAD units)				
MP	42.4 \pm 0.9	54.9 \pm 0.5 b	59.5 \pm 0.6 b	51.6 \pm 1.7
CH	44.1 \pm 0.6	54.8 \pm 0.6 b	62.2 \pm 1.1 a	54.4 \pm 1.2
NT	44.3 \pm 1.3	56.3 \pm 0.6 a	61.4 \pm 0.8 ab	52.8 \pm 1.4
Rotation				
CC	42.9 \pm 0.5 ab	54.4 \pm 0.7 b	59.3 \pm 0.4 b	52.8 \pm 1.8
CS	42.5 \pm 1.2 b	55.5 \pm 0.5 ab	60.5 \pm 0.7 b	52.4 \pm 0.6
CFF	45.6 \pm 1.0 a	56.2 \pm 0.4 a	63.3 \pm 1.0 a	53.5 \pm 1.8
ANOVA F statistics and significance				
Tillage (T)	1.4 NS	4.7 *	4.6 *	1.0 NS
Rotation (R)	3.6 .	5.6 *	9.8 **	0.1 NS
T x R	0.7 NS	0.4 NS	0.6 NS	1.8 NS

“.” Significance level: $p < 0.1$ | * Significance level: $p < 0.05$ | ** Significance level: $p < 0.01$ | *** Significance level: $p < 0.001$ | “NS” Significance level: $p > 0.1$
V5 = fifth collared leaf, V10 = tenth collared leaf, R1 = silking, R4 = dough | Rotation: CC = continuous corn; CS = corn-soybean; CFF = corn-forage-forage
Tillage: MP = moldboard plow; CH = chisel till; NT = no-till

Table 2.7. Correlation coefficients from the relationships between leaf chlorophyll and soil health indicators

Stage	2017			2018		
	POXC	Min C	Protein	POXC	Min C	Protein
Northwest						
V5	0.239	0.785	0.406	-0.164	0.000	-0.196
V10	0.414	0.099	0.225	0.402	0.463	0.246
R1	0.111	0.144	-0.017	0.582	0.529	0.641
R4	0.334	0.137	0.446	0.308	0.335	0.216
Wooster						
V5	0.139	0.173	0.065	0.519	0.352	0.397
V10	0.366	0.520	0.463	0.521	0.376	0.087
R1	0.238	0.344	0.149	0.269	0.343	0.283
R4	0.004	0.020	0.116	0.046	0.321	0.072

POXC = permanganate oxidizable carbon; Min C = mineralizable carbon; Protein =soil protein
V5 = fifth collared leaf, V10 = tenth collared leaf, R1 = silking, R4 = dough, R6 = physiological maturity

Total Plant Biomass

Overall total plant biomass was higher in reduced or conservation tillage treatments (NT and CH) compared to intensive tillage and greater in the more diverse rotations (CS and CFF) compared to corn monoculture (Tables 2.8-2.11). Crop rotation had the strongest effect on total plant biomass in both sites. Interactions between tillage and rotation were only observed in 2018. In Northwest, total plant biomass values peak at R4 corn stage for 2017, but at R6 for 2018. In Wooster, total plant biomass values peak at corn stage R6.

Correlation coefficients between total biomass and soil health indicators were highly variable between years, sites, and stages (Table 2.12). In Northwest 2017, correlations between total plant biomass and soil health indicators were positively stronger during the V5 and R6 stages. On the other hand, in Northwest 2018, all correlations coefficients were negative during the V5 stage, and only soil protein had a stronger correlation during the R6 stage. In Wooster, the majority of correlation coefficients were positive and higher during the V10 and R1 stages. In Wooster 2018, there are also higher correlation coefficients during the V5 stage, and on the R4 stage for Min C only.

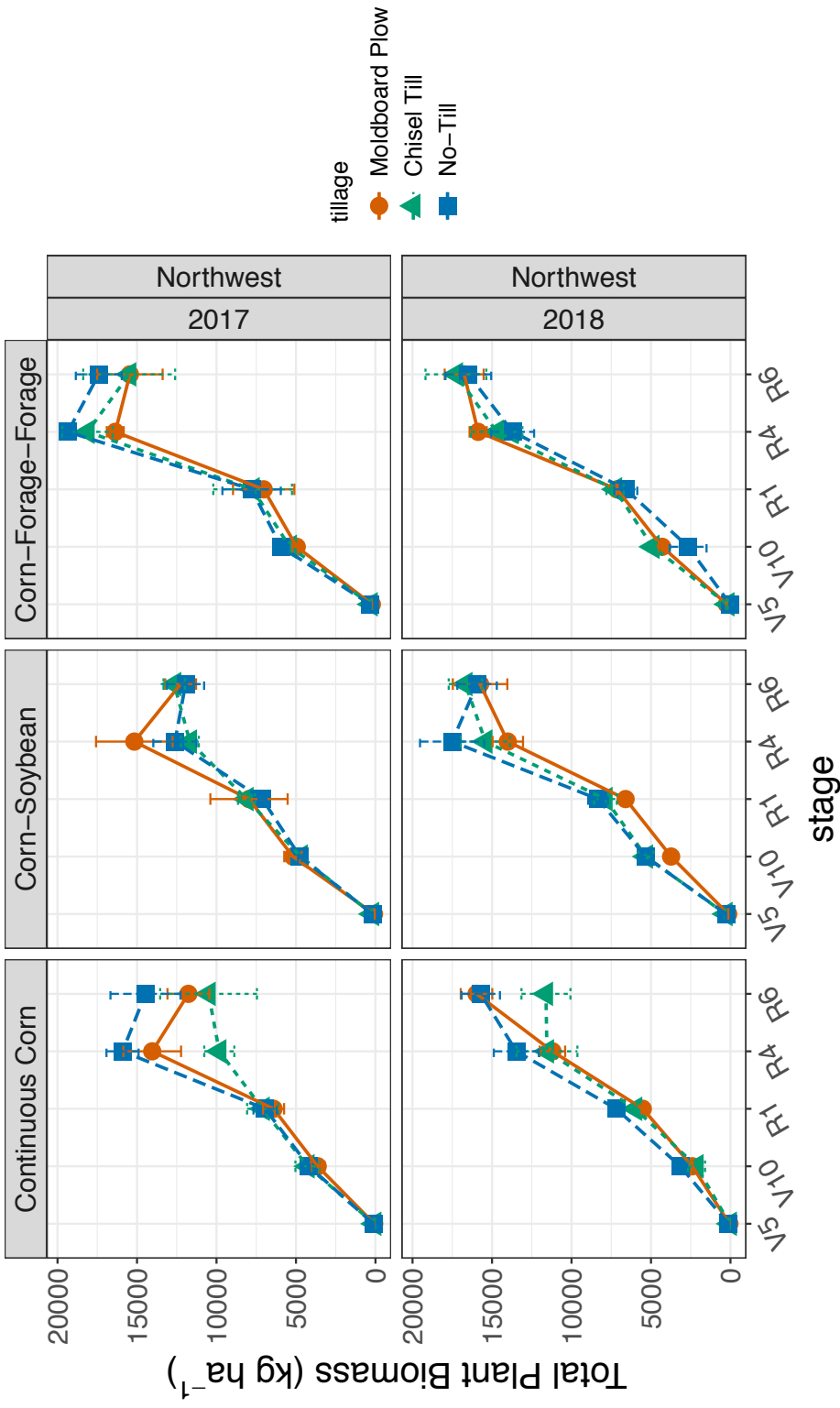


Figure 2.3. Total Plant Biomass (kg ha⁻¹) means at Northwest in the 2017 and 2018 growing seasons for three tillage regimes in three crop rotations. Tillage practices include moldboard plow (red circles, solid line), chisel till (green triangles, dotted line), and no-till (blue squares, dashed line). Error bars represent one standard error of the mean.

Table 2.8. Total plant aboveground biomass at Northwest in 2017 by tillage and rotation treatments for each sampled crop developmental stage (mean \pm standard error). Different letters within the same column of rotation or treatment values represent significantly different treatments ($\alpha = 0.1$). F-statistics and significance from analysis of variance for each crop stage.

Northwest 2017						
Variable	V5	V10	R1	R4	R6	
Tillage						
			Plant Total Biomass (kg ha ⁻¹)			
MP	133.4 \pm 27.7 b	4593.1 \pm 323.3	7132.4 \pm 943.0	15193.4 \pm 945.6 ab	13156.4 \pm 953.4	
CH	196.8 \pm 36.7 a	4799.8 \pm NA	7645.3 \pm 797.5	13203.7 \pm 1372.9 b	12884.9 \pm 1424.4	
NT	221.0 \pm 33.3 a	4961.8 \pm 297.7	7282.7 \pm 589.5	15959.2 \pm 1091.0 a	14606.6 \pm 1145.9	
Rotation						
CC	95.1 \pm 16.4 c	4033.8 \pm 270.7 b	6816.5 \pm 414.4	13260.3 \pm 1115.5 b	12245.0 \pm 1289.7 b	
CS	166.9 \pm 23.5 b	4903.8 \pm NA	7727.1 \pm 738.1	13152.6 \pm 967.8 b	12288.5 \pm 490.1 b	
CFF	289.3 \pm 22.4 a	5428.6 \pm 217.6 a	7516.9 \pm 1056.8	17943.4 \pm 650.8 a	16114.4 \pm 1152.3 a	
ANOVA F statistics and significance						
Tillage (T)	7.4 **	0.6 NS	0.1 NS	3.4 .	1.3 NS	
Rotation (R)	35.0 ***	9.0 **	0.3 NS	12.6 ***	7.2 **	
T x R	1.4 NS	1.0 NS	0.1 NS	2.6 .	0.7 NS	

“.” Significance level: $p < 0.1$ | * Significance level: $p < 0.05$ | ** Significance level: $p < 0.01$ | *** Significance level: $p < 0.001$ | “NS” Significance level: $p > 0.1$
V5 = fifth collared leaf, V10 = tenth collared leaf, R1 = silking, R4 = dough, R6 = physiological maturity
Rotation: CC = continuous corn; CS = corn-soybean; CFF = corn-forage-forage
Tillage: MP = moldboard plow; CH = chisel till; NT = no-till

Table 2.9. Total plant aboveground biomass at Northwest in 2018 by tillage and rotation treatments for each sampled crop developmental stage (mean \pm standard error). Different letters within the same column of rotation or treatment values represent significantly different treatments ($\alpha = 0.1$). F-statistics and significance from analysis of variance for each crop stage.

Northwest 2018						
Variable	V5	V10	R1	R4	R6	
Tillage						
MP	145.4 \pm 25.1 b	3477.7 \pm 299.9	6418.1 \pm 253.8 b	13694.2 \pm 784.5	16159.0 \pm 689.1	
CH	181.7 \pm 40.2 a	4119.4 \pm 546.0	7039.0 \pm 379.0 ab	13890.6 \pm 1038.3	15183.5 \pm 1184.2	
NT	140.9 \pm 38.0 b	3720.3 \pm 550.2	7368.8 \pm 376.9 a	14895.7 \pm 1042.5	16045.9 \pm 660.1	
Rotation						
CC	78.8 \pm 13.3 c	2564.8 \pm 223.0 b	6234.3 \pm 254.0 b	12072.8 \pm 807.2 b	14430.0 \pm 949.0 b	
CS	243.3 \pm 26.3 a	4798.9 \pm 338.2 a	7596.6 \pm 385.1 a	15646.1 \pm 934.8 a	16120.8 \pm 696.7 ab	
CFF	145.9 \pm 35.2 b	3953.6 \pm 484.5 a	6995.0 \pm 281.1 ab	14761.6 \pm 702.2 a	16837.5 \pm 784.9 a	
ANOVA F statistics and significance						
Source						
Tillage (T)	4.9 *	1.2 NS	3.4 .	0.6 NS	0.5 NS	
Rotation (R)	66.5 ***	14.9 ***	6.9 **	5.1 *	2.9 .	
T x R	33.6 ***	4.2 *	2.4 .	1.1 NS	1.7 NS	

“.” Significance level: $p < 0.1$ | * Significance level: $p < 0.05$ | ** Significance level: $p < 0.01$ | *** Significance level: $p < 0.001$ | “NS” Significance level: $p > 0.1$
V5 = fifth collared leaf, V10 = tenth collared leaf, R1 = silking, R4 = dough, R6 = physiological maturity
Rotation: CC = continuous corn; CS = corn-soybean; CFF = corn-forage-forage
Tillage: MP = moldboard plow; CH = chisel till; NT = no-till

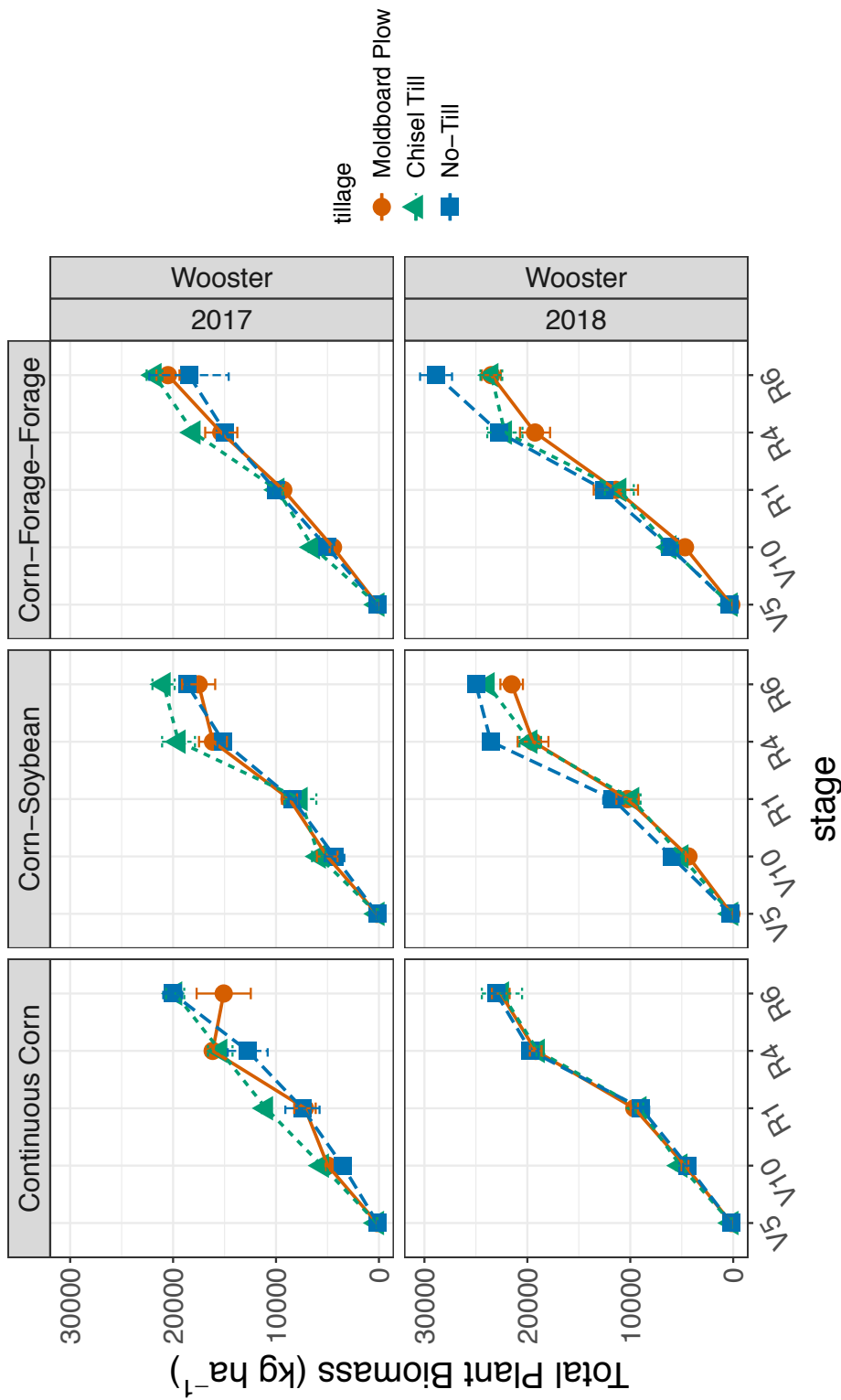


Figure 2.4. Total Plant Biomass (kg ha⁻¹) means at Wooster in the 2017 and 2018 growing seasons for three tillage regimes in three crop rotations. Tillage practices include moldboard plow (red circles, solid line), chisel till (green triangles, dotted line), and no-till (blue squares, dashed line). Error bars represent one standard error of the mean.

Table 2.10. Total plant aboveground biomass at Wooster in 2017 by tillage and rotation treatments for each sampled crop developmental stage (mean \pm standard error). Different letters within the same column of rotation or treatment values represent significantly different treatments ($\alpha = 0.1$). F-statistics and significance from analysis of variance for each crop stage.

Wooster 2017						
Variable	V5	V10	R1	R4	R6	
Tillage						
MP	175.9 \pm 14.4	4818.6 \pm 324.2 b	8389.9 \pm 483.3	15829.3 \pm NA. ab	17698.9 \pm 1228.8	
CH	160.8 \pm 23.3	5933.0 \pm 263.5 a	9485.5 \pm 693.8	17636.5 \pm 826.7 a	20843.9 \pm 543.0	
NT	151.0 \pm 13.9	4287.1 \pm 434.7 b	8615.4 \pm 633.1	14262.8 \pm 698.9 b	19042.7 \pm 1173.2	
Rotation						
CC	170.8 \pm 15.4	4678.0 \pm 379.4	8531.8 \pm 843.0	14601.8 \pm NA b	18307.8 \pm 1167.7	
CS	165.1 \pm 15.9	5059.4 \pm 493.8	8230.9 \pm 526.9	16902.0 \pm 909.9 a	19025.7 \pm 753.5	
CFF	151.8 \pm 21.6	5301.2 \pm 357.0	9728.1 \pm 233.1	16088.4 \pm 666.6 ab	20252.1 \pm 1277.0	
ANOVA F statistics and significance						
Source						
Tillage (T)	0.5 NS	6.7 **	1.2 NS	6.5 **	2.3 NS	
Rotation (R)	0.3 NS	0.9 NS	2.2 NS	2.6 NS	0.9 NS	
T x R	0.7 NS	0.9 NS	2.4 .	0.8 NS	1.0 NS	

“.” Significance level: $p < 0.1$ | * Significance level: $p < 0.05$ | ** Significance level: $p < 0.01$ | *** Significance level: $p < 0.001$ | “NS” Significance level: $p > 0.1$
V5 = fifth collared leaf, V10 = tenth collared leaf, R1 = silking, R4 = dough, R6 = physiological maturity
Rotation: CC = continuous corn; CS = corn-soybean; CFF = corn-forage-forage
Tillage: MP = moldboard plow; CH = chisel till; NT = no-till

Table 2.1.1. Total plant aboveground biomass at Wooster in 2018 by tillage and rotation treatments for each sampled crop developmental stage (mean \pm standard error). Different letters within the same column of rotation or treatment values represent significantly different treatments ($\alpha = 0.1$). F-statistics and significance from analysis of variance for each crop stage.

Wooster 2018						
Variable	V5	V10	R1	R4	R6	
Plant Total Biomass (kg ha ⁻¹)						
Tillage						
MP	180.0 \pm 11.4 c	4572.7 \pm 143.4 b	10424.0 \pm 749.5	19296.5 \pm 625.9 b	22536.4 \pm 559.3 b	
CH	208.0 \pm 15.5 b	5455.2 \pm 314.7 a	9995.7 \pm 558.9	20296.5 \pm 755.0 ab	23277.0 \pm 674.7 b	
NT	265.7 \pm 18.2 a	5542.8 \pm 385.0 a	11046.2 \pm 686.5	21984.6 \pm 650.1 a	25628.5 \pm 1008.3 a	
Rotation						
CC	187.2 \pm 10.2 b	4755.4 \pm 214.6 b	9219.0 \pm 284.9 b	19286.9 \pm 297.1 b	22688.3 \pm 660.8 b	
CS	212.4 \pm 17.5 b	5129.7 \pm 332.0 ab	10589.0 \pm 552.6 ab	20902.5 \pm 853.1 ab	23464.7 \pm 622.4 ab	
CFF	254.1 \pm 22.0 a	5685.7 \pm 358.8 a	11657.9 \pm 809.3 a	21388.3 \pm 852.7 a	25288.8 \pm 1080.2 a	
ANOVA F statistics and significance						
Source						
Tillage (T)	25.1 ***	4.2 *	0.7 NS	5.2 *	6.8 **	
Rotation (R)	15.0 ***	3.2 .	3.7 *	3.4 .	4.7 *	
T x R	2.7 .	1.7 NS	0.3 NS	1.6 NS	2.2 NS	

“.” Significance level: $p < 0.1$ | * Significance level: $p < 0.05$ | ** Significance level: $p < 0.01$ | *** Significance level: $p < 0.001$ | “NS” Significance level: $p > 0.1$
V5 = fifth collared leaf, V10 = tenth collared leaf, R1 = silking, R4 = dough, R6 = physiological maturity
Rotation: CC = continuous corn; CS = corn-soybean; CFF = corn-forage-forage
Tillage: MP = moldboard plow; CH = chisel till; NT = no-till

Table 2.12. Correlation coefficients on the relationships between total plant biomass and soil health indicators

Stage	2017			2018		
	POXC	Min C	Protein	POXC	Min C	Protein
Northwest						
V5	0.450	0.515	0.360	-0.328	-0.232	-0.233
V10	0.500	0.146	0.333	0.018	0.068	-0.142
R1	-0.001	-0.144	0.018	-0.079	0.190	0.115
R4	0.161	0.038	0.432	0.012	0.182	-0.138
R6	0.406	0.501	0.397	0.133	0.022	0.325
Wooster						
V5	-0.140	-0.229	-0.127	0.412	0.326	0.257
V10	0.252	0.366	0.314	0.125	0.328	0.258
R1	0.251	0.523	0.297	0.455	0.164	0.312
R4	0.204	0.269	0.226	0.250	0.334	0.193
R6	-0.016	-0.007	0.082	-0.032	0.028	0.093

POXC = permanganate oxidizable carbon; Min C = mineralizable carbon; Protein =soil protein
V5 = fifth collared leaf, V10 = tenth collared leaf, R1 = silking, R4 = dough, R6 = physiological maturity

Total Nitrogen Uptake

Although nitrogen uptake values varied significantly by stage, rotation had the most significant effect on total plant nitrogen uptake out of the treatments (Table 2.2) where values were higher in the CFF rotation compared to corn monoculture which had the lowest values (Figure 2.5; Figure 2.6). Although tillage had minimum effects on nitrogen uptake, values were higher in less intense tillage treatments (CH and NT) compared to intensive tillage (MP) (Table 2.13-2.16). In Northwest, no-till had higher N uptake values compared to tilled soils, and there was a tillage by rotation interaction in 2017. Also, total plant nitrogen uptake values peak at R4 in 2017 and at R6 for 2018 in Northwest. On the other hand, Wooster results for nitrogen uptake varied by year, where chisel had higher values in 2017, but no-till had higher values compared to the other tillage treatments in 2018. Peaks in total plant nitrogen uptake occur in corn R4 and R6 in 2017 and at R6 in 2018.

Correlation coefficients between total plant N uptake and soil health indicators were highly variable between years, sites, and stages. In Northwest 2017, correlation coefficients were higher during the V5 and R6 stages, and higher values are also observed during V10 for POXC and R4 for soil protein. Also, during 2017 in Northwest the lowest values for correlations are observed during R1. During 2018, higher results for correlations were observed during R1 for Min C and Protein, during R4 for POXC and Min C, and R6 for soil protein in Northwest. In Northwest 2018, the lowest values for correlations were observed during the V5 stage. In Wooster 2017, higher values for the

correlation coefficients were observed during the V10, R1 and R4 stages for all soil health indicators, and the lowest values resulted in the V5 stage. For Wooster 2018, values varied between the soil health indicators correlation coefficients with total biomass. For instance, correlations with POXC were higher during the V5 and R1 stages; correlations between total plant biomass and Min C were stronger during the V5 and R4 stages; and correlations between total plant biomass and soil protein were very similar across stages except for the R6 where it was the lowest.

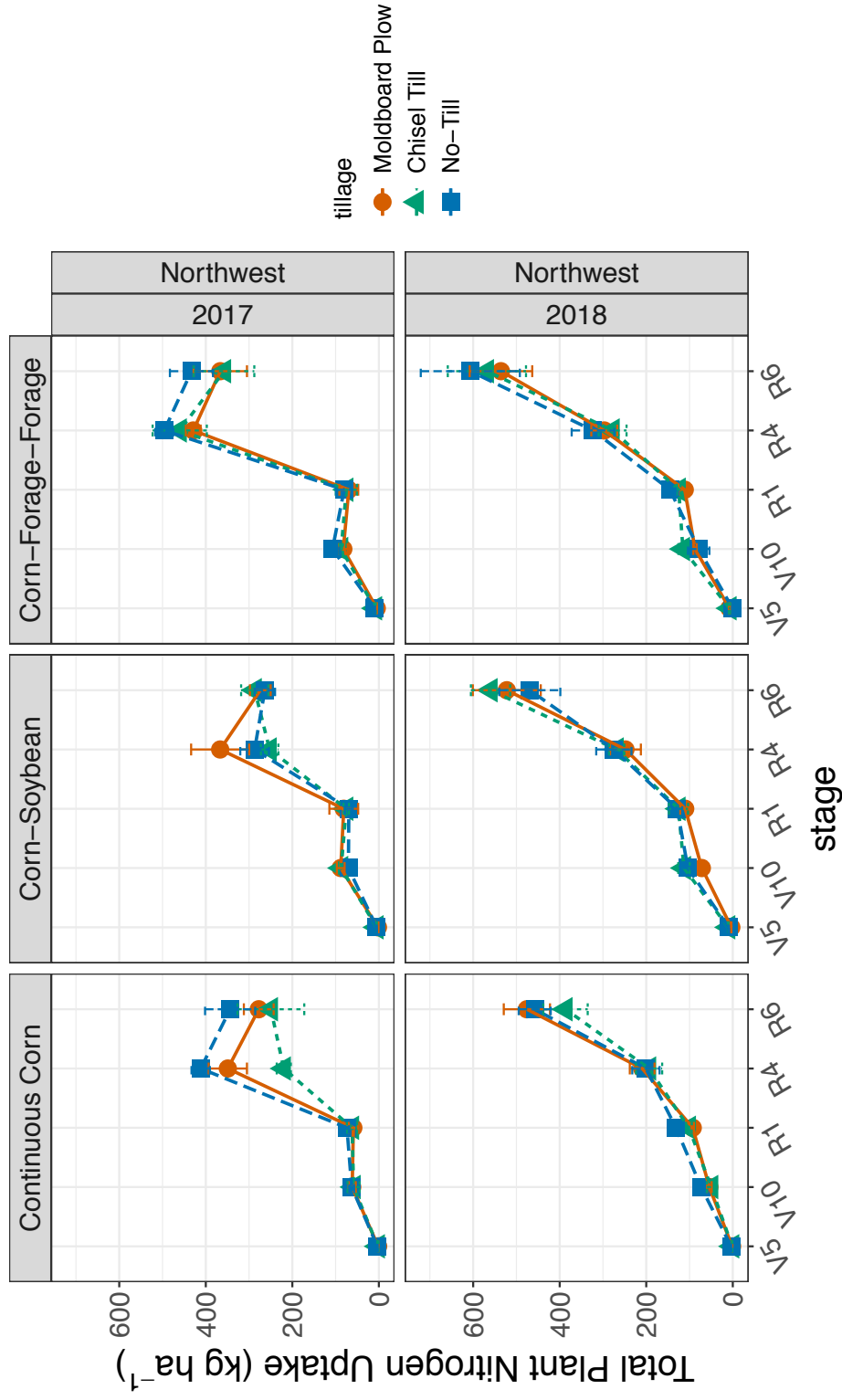


Figure 2. 5. Total Plant Nitrogen Uptake (kg ha^{-1}) means at Northwest in the 2017 and 2018 growing seasons for three tillage regimes in three crop rotations. Tillage practices include moldboard plow (red circles, solid line), chisel till (green triangles, dotted line), and no-till (blue squares, dashed line). Error bars represent one standard error of the mean.

Table 2. 13. Total corn nitrogen uptake at Northwest in 2017 by tillage and rotation treatments for each sampled crop developmental stage (mean \pm standard error). Different letters within the same column of rotation or treatment values represent significantly different treatments ($\alpha = 0.1$). F-statistics and significance from analysis of variance for each crop stage.

Northwest 2017						
Variable	V5	V10	R1	R4	R6	
Tillage						
MP	2.9 \pm 0.6 b	77.3 \pm 6.1	69.8 \pm 12.3	381.6 \pm 26.6 a	306.0 \pm 26.3	
CH	5.5 \pm 1.1 a	76.3 \pm NA	70.9 \pm 8.3	310.8 \pm 42.4 b	298.1 \pm 35.2	
NT	5.9 \pm 0.9 a	80.1 \pm 8.3	74.4 \pm 6.3	398.3 \pm 32.9 a	346.8 \pm 33.7	
Rotation						
CC	2.5 \pm 0.5 c	60.9 \pm 5.0 b	64.9 \pm 5.2	326.9 \pm 32.0 b	290.3 \pm 32.7 b	
CS	4.2 \pm 0.6 b	81.2 \pm NA a	75.3 \pm 10.2	302.1 \pm 28.0 b	274.8 \pm 13.7 b	
CFF	7.7 \pm 0.9 a	92.1 \pm 6.7 a	75.0 \pm 11.0	461.7 \pm 21.9 a	385.8 \pm 32.8 a	
ANOVA F statistics and significance						
Source						
Tillage (T)	10.3 **	0.1 NS	0.1 NS	4.4 *	1.4 NS	
Rotation (R)	26.0 ***	9.3 **	0.3 NS	15.1 ***	7.5 **	
T x R	1.1 NS	1.6 NS	0.2 NS	2.6 .	0.7 NS	

“.” Significance level: $p < 0.1$ | * Significance level: $p < 0.05$ | ** Significance level: $p < 0.01$ | *** Significance level: $p < 0.001$ | “NS” Significance level: $p > 0.1$
V5 = fifth collared leaf, V10 = tenth collared leaf, R1 = silking, R4 = dough, R6 = physiological maturity
Rotation: CC = continuous corn; CS = corn-soybean; CFF = corn-forage-forage
Tillage: MP = moldboard plow; CH = chisel till; NT = no-till

Table 2.14. Total corn nitrogen uptake at Northwest in 2018 by tillage and rotation treatments for each sampled crop developmental stage (mean \pm standard error). Different letters within the same column of rotation or treatment values represent significantly different treatments ($\alpha = 0.1$). F-statistics and significance from analysis of variance for each crop stage.

Northwest 2018						
Variable	V5	V10	R1	R4	R6	
Total Plant Nitrogen Uptake (kg ha ⁻¹)						
Tillage						
MP	4.2 \pm 1.0 b	71.0 \pm 5.7	104.5 \pm 5.5 b	251.5 \pm 20.2	511.3 \pm 35.6	
CH	6.5 \pm 1.4 a	92.6 \pm 12.1	117.3 \pm 7.8 b	246.5 \pm 19.1	504.9 \pm 44.1	
NT	4.7 \pm 1.3 b	85.3 \pm 10.0	135.1 \pm 7.0 a	266.2 \pm 27.2	511.2 \pm 46.8	
Rotation						
CC	2.3 \pm 0.4 c	59.1 \pm 5.0 b	108.5 \pm 7.4 b	201.2 \pm 15.3 b	440.1 \pm 27.5 b	
CS	7.8 \pm 1.2 a	96.0 \pm 9.2 a	121.3 \pm 8.4 ab	263.8 \pm 16.9 a	517.1 \pm 35.7 ab	
CFF	5.3 \pm 1.3 b	93.8 \pm 9.5 a	127.1 \pm 7.1 a	299.3 \pm 20.2 a	570.3 \pm 48.1 a	
ANOVA F statistics and significance						
Source						
Tillage (T)	9.2 **	2.3 NS	8.7 **	0.3 NS	0.0 NS	
Rotation (R)	48.7 ***	8.1 **	3.4 .	6.4 **	4.9 *	
T x R	30.5 ***	2.0 NS	0.6 NS	0.2 NS	1.0 NS	

“.” Significance level: $p < 0.1$ | * Significance level: $p < 0.05$ | ** Significance level: $p < 0.01$ | *** Significance level: $p < 0.001$ | “NS” Significance level: $p > 0.1$
V5 = fifth collared leaf, V10 = tenth collared leaf, R1 = silking, R4 = dough, R6 = physiological maturity
Rotation: CC = continuous corn; CS = corn-soybean; CFF = corn-forage-forage
Tillage: MP = moldboard plow; CH = chisel till; NT = no-till

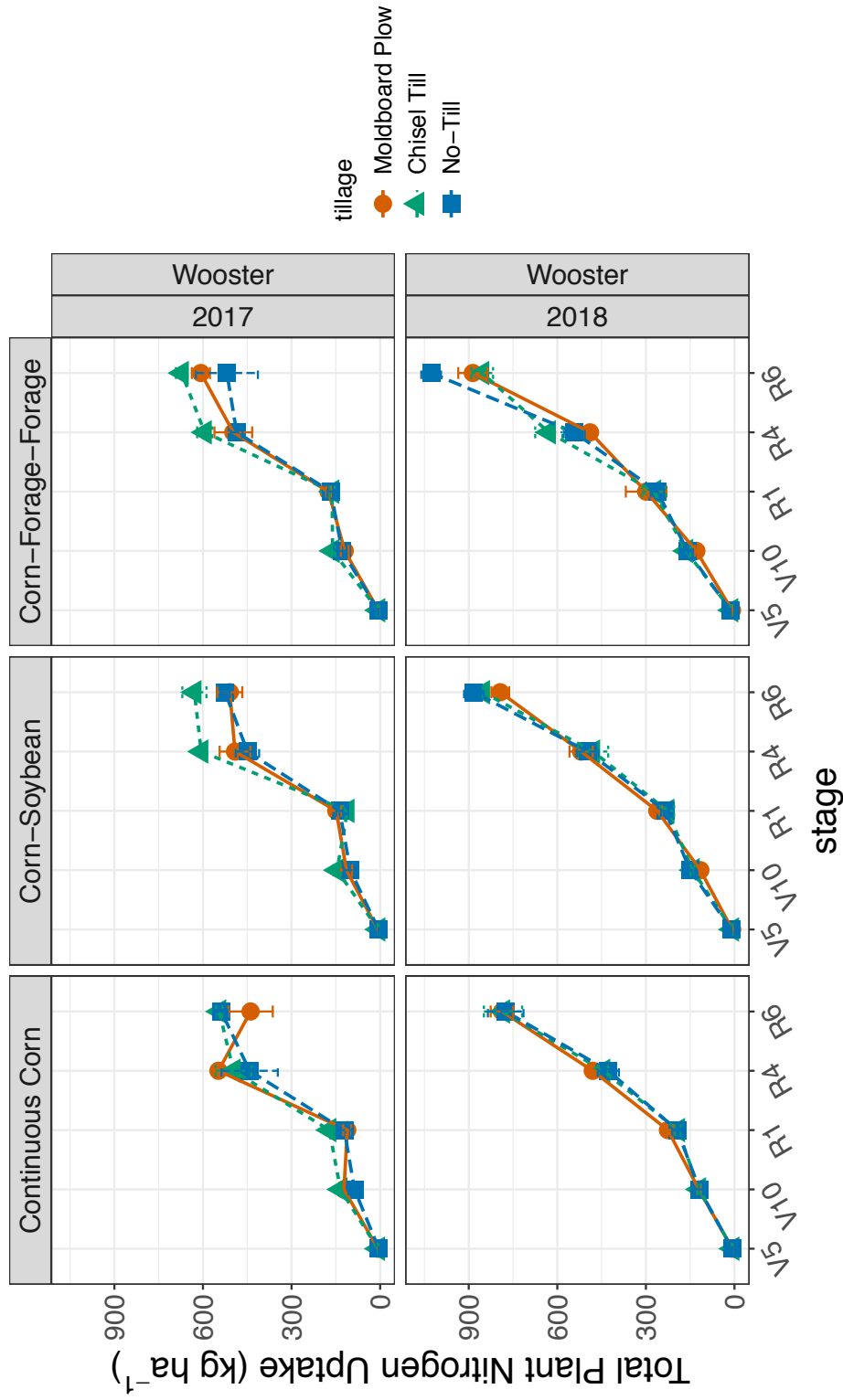


Figure 2. 6. Total Plant Nitrogen Uptake (kg ha^{-1}) means at Wooster in the 2017 and 2018 growing seasons for three tillage regimes in three crop rotations. Tillage practices include moldboard plow (red circles, solid line), chisel till (green triangles, dotted line), and no-till (blue squares, dashed line). Error bars represent one standard error of the mean.

Table 2. 15. Total corn nitrogen uptake at Wooster in 2017 by tillage and rotation treatments for each sampled crop developmental stage (mean \pm standard error). Different letters within the same column of rotation or treatment values represent significantly different treatments ($\alpha = 0.1$). F-statistics and significance from analysis of variance for each crop stage.

Wooster 2017						
Variable	V5	V10	R1	R4	R6	
Tillage						
MP	7.2 \pm 0.6	118.3 \pm NA b	144.9 \pm 11.5	507.6 \pm NA ab	518.3 \pm 36.0 b	
CH	6.7 \pm 1.0	147.1 \pm 6.0 a	148.5 \pm 11.1	565.5 \pm 25.8 a	615.4 \pm 23.0 a	
NT	5.3 \pm 0.5	105.4 \pm 11.5 b	141.8 \pm 11.3	459.0 \pm 31.1 b	527.1 \pm 31.3 ab	
Rotation						
CC	6.5 \pm 0.7	113.5 \pm NA	134.9 \pm 13.1 b	488.9 \pm NA	507.2 \pm 28.4 b	
CS	6.5 \pm 0.7	120.3 \pm 11.2	132.0 \pm 9.3 b	515.5 \pm 30.1	554.3 \pm 25.7 ab	
CFF	6.2 \pm 0.9	136.6 \pm 9.0	168.1 \pm 5.1 a	525.8 \pm 26.9	599.4 \pm 39.0 a	
ANOVA F statistics and significance						
Tillage (T)	1.5 NS	7.2 **	0.2 NS	5.8 *	3.5 .	
Rotation (R)	0.1 NS	2.2 NS	5.6 *	0.9 NS	2.6 NS	
T x R	0.7 NS	0.5 NS	3.4 *	0.9 NS	1.0 NS	

“.” Significance level: $p < 0.1$ | * Significance level: $p < 0.05$ | ** Significance level: $p < 0.01$ | *** Significance level: $p < 0.001$ | “NS” Significance level: $p > 0.1$
V5 = fifth collared leaf, V10 = tenth collared leaf, R1 = silking, R4 = dough, R6 = physiological maturity
Rotation: CC = continuous corn; CS = corn-soybean; CFF = corn-forage-forage
Tillage: MP = moldboard plow; CH = chisel till; NT = no-till

Table 2. 16. Total corn nitrogen uptake at Wooster in 2018 by tillage and rotation treatments for each sampled crop developmental stage (mean \pm standard error). Different letters within the same column of rotation or treatment values represent significantly different treatments ($\alpha = 0.1$). F-statistics and significance from analysis of variance for each crop stage.

Wooster 2018						
Variable	V5	V10	R1	R4	R6	
Tillage						
MP	7.8 \pm 0.6 b	121.7 \pm 3.2	261.9 \pm 23.5	496.2 \pm 15.0	822.2 \pm 25.6	
CH	8.9 \pm 0.7 b	141.8 \pm 7.9	230.3 \pm 18.0	517.5 \pm 34.9	830.1 \pm 24.4	
NT	11.2 \pm 0.9 a	142.5 \pm 10.5	229.7 \pm 13.3	489.1 \pm 24.6	894.8 \pm 42.7	
Rotation						
CC	7.6 \pm 0.4 c	119.7 \pm 4.9 b	203.4 \pm 9.4 b	450.6 \pm 14.8 b	782.0 \pm 27.9 b	
CS	9.1 \pm 0.7 b	135.7 \pm 7.6 ab	241.1 \pm 10.3 ab	498.6 \pm 22.3 ab	842.5 \pm 18.4 b	
CFF	11.1 \pm 1.0 a	150.6 \pm 8.8 a	277.4 \pm 24.6 a	553.6 \pm 26.9 a	922.6 \pm 33.4 a	
ANOVA F statistics and significance						
Source						
Tillage (T)	18.8 ***	3.1 .	1.2 NS	0.6 NS	2.7 .	
Rotation (R)	19.8 ***	5.4 *	4.6 *	7.4 **	8.6 **	
T x R	3.0 .	1.0 NS	0.0 NS	2.5 .	1.7 NS	

“.” Significance level: $p < 0.1$ | * Significance level: $p < 0.05$ | ** Significance level: $p < 0.01$ | *** Significance level: $p < 0.001$ | “NS” Significance level: $p > 0.1$
V5 = fifth collared leaf, V10 = tenth collared leaf, R1 = silking, R4 = dough, R6 = physiological maturity
Rotation: CC = continuous corn; CS = corn-soybean; CFF = corn-forage-forage
Tillage: MP = moldboard plow; CH = chisel till; NT = no-till

Table 2. 17. Correlation coefficients for total plant nitrogen uptake and soil health indicators for all five sampling stages by year (n=135 per site).

Stage	2017			2018		
	POXC	Min C	Protein	POXC	Min C	Protein
Northwest						
V5	0.379	0.594	0.348	-0.240	-0.142	-0.163
V10	0.464	0.193	0.240	0.271	0.216	0.054
R1	-0.003	-0.113	0.041	0.287	0.423	0.415
R4	0.119	0.005	0.467	0.370	0.367	0.070
R6	0.401	0.505	0.417	0.228	0.208	0.355
Wooster						
V5	-0.077	-0.111	-0.002	0.441	0.356	0.257
V10	0.287	0.398	0.365	0.268	0.214	0.255
R1	0.222	0.392	0.303	0.424	0.132	0.250
R4	0.319	0.319	0.313	0.173	0.386	0.264
R6	0.042	0.084	0.077	0.029	0.014	0.120

POXC = permanganate oxidizable carbon; Min C = mineralizable carbon; Protein =soil protein
V5 = fifth collared leaf, V10 = tenth collared leaf, R1 = silking, R4 = dough, R6 = physiological maturity

Grain Yield

Rotation was the main treatment that significantly impacted yields where the CFF rotation had higher values compared to the CC rotation (Table 2.18; Table 2.19). When averaging over rotations, no significant differences ($p < 0.1$) were observed between tillage treatment for both years and both sites except for Wooster 2018 where yields were higher in no-till soils compared to tilled soils. Significant interactions ($p < 0.1$) between tillage and rotation treatments were only observed in the 2018 growing season. In Northwest, yield increased in the no-till soils with increasing crop rotation diversity except for 2018 where no-till soils yield dropped in the CFF rotation (Figure 2.8). In Wooster, the no-till soils had higher yields under the CS and CFF rotations compared to corn monoculture (Figure 2.8). In general, results showed moderate positive correlation coefficients between corn yield and soil health indicators except for Northwest soil protein (both years and 2018 POXC which showed negative weak to moderate correlation coefficients (Table 2.20). Correlation coefficients were stronger in 2017 where Min C had the highest coefficients.

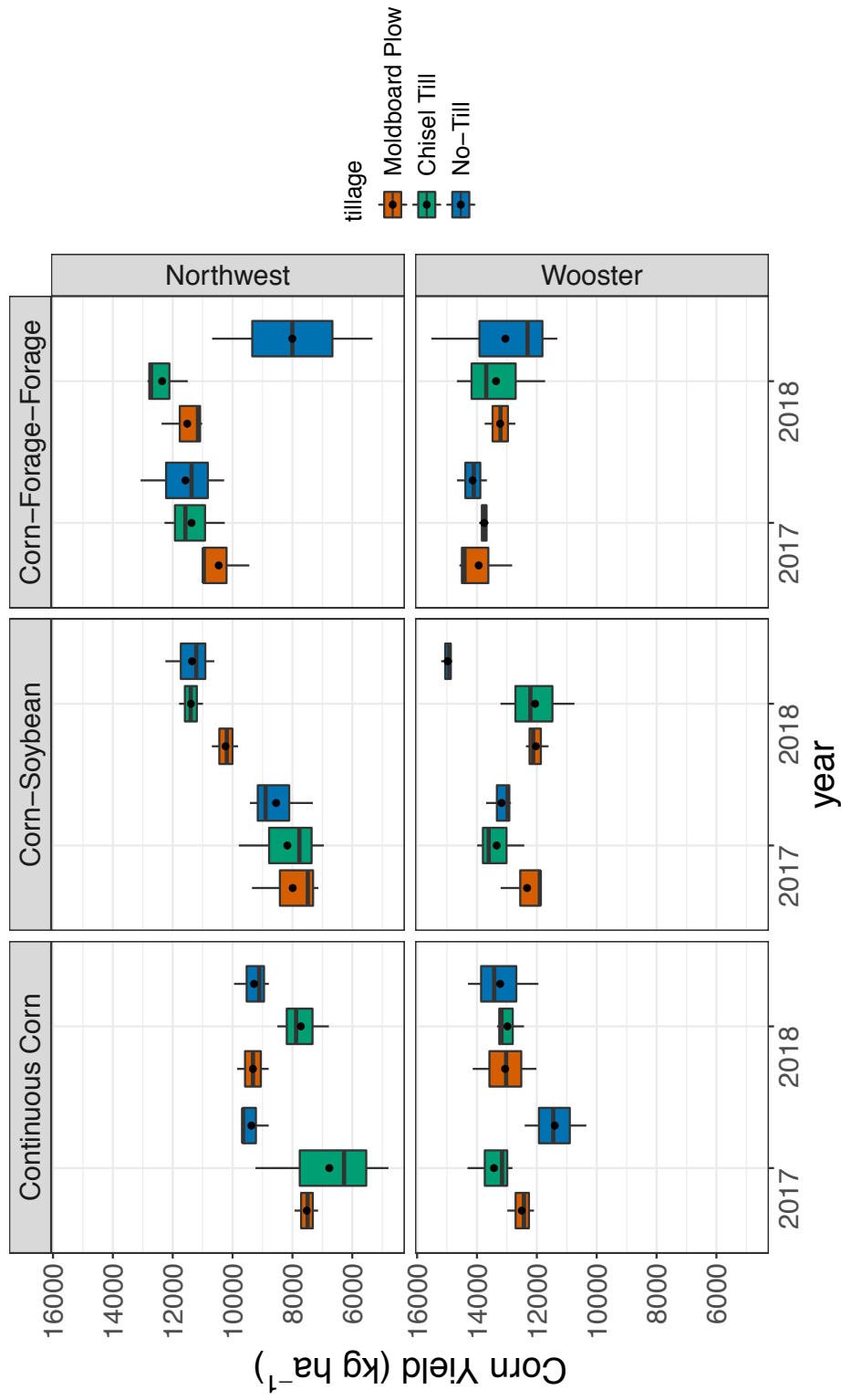


Figure 2. 7. Corn grain yield (kg ha^{-1}) mean values at Wooster and Northwest in the 2017 and 2018 growing seasons for three tillage regimes in three crop rotations. Tillage practices include moldboard plow (red), chisel till (green), and no-till (blue). Error bars represent one standard error of the mean.

Table 2. 18. Corn yield at Northwest in 2017 and 2018 by tillage and rotation treatments for each sampled crop developmental stage (mean \pm standard error). Different letters within the same column of rotation or treatment values represent significantly different treatments ($\alpha = 0.1$). F-statistics and significance from analysis of variance.

Northwest Site		
Variable	2017	2018
	Grain Yield (kg ha ⁻¹)	Grain Yield (kg ha ⁻¹)
Tillage		
MP	8658.8 \pm 523.0	10356.4 \pm 360.4
CH	8769.8 \pm 833.4	10489.0 \pm 734.2
NT	9831.1 \pm 547.4	9740.6 \pm NA
Rotation		
CC	7887.6 \pm 550.5 b	8774.8 \pm 329.0 b
CS	8235.5 \pm 372.5 b	10994.0 \pm 254.9 a
CFF	11136.6 \pm 367.5 a	10951.8 \pm NA a
ANOVA		
Source		
Tillage (T)	2.4 NS	1.7 NS
Rotation (R)	18.3 ***	10.0 **
T x R	0.9 NS	5.9 **

“.” Significance level: $P < 0.1$ | * Significance level: $P < 0.05$ | ** Significance level: $P < 0.01$

*** Significance level: $P < 0.001$ | “NS” Significance level: $p > 0.1$

Rotation: CC = continuous corn; CS = corn-soybean; CFF = corn-forage-forage

Tillage: MP = moldboard plow; CH = chisel till; NT = no-till

Table 2. 19. Corn yield at Wooster in 2017 and 2018 by tillage and rotation treatments for each sampled crop developmental stage (mean \pm standard error). Different letters within the same column of rotation or treatment values represent significantly different treatments ($\alpha = 0.1$). F-statistics and significance from analysis of variance.

Wooster Site		
Variable	2017	2018
	Grain Yield (kg ha ⁻¹)	Grain Yield (kg ha ⁻¹)
Tillage		
MP	12924.2 \pm 337.4	12772.3 \pm 278.9 b
CH	13511.1 \pm 201.8	12799.6 \pm 386.6 b
NT	12908.6 \pm 451.5	13751.5 \pm 519.2 a
Rotation		
CC	12445.4 \pm 372.0 b	13088.2 \pm 280.1
CS	12947.7 \pm 255.9 b	13023.2 \pm 536.3
CFF	13950.9 \pm 192.6 a	13212.1 \pm 454.1

Source	ANOVA	
Tillage (T)	2.1 NS	3.4 .
Rotation (R)	10.4 **	0.1 NS
T x R	3.0 .	3.6 *

“.” Significance level: P < 0.1 | * Significance level: P < 0.05 | ** Significance level: P < 0.01 *** Significance level: P < 0.001 | “NS” Significance level: p > 0.1
 Rotation: CC = continuous corn; CS = corn-soybean; CFF = corn-forage-forage
 Tillage: MP = moldboard plow; CH = chisel till; NT = no-till

Table 2. 20. Correlation coefficients for total corn yield and soil health indicators for the 2017 and 2018 growing seasons.

Site	Year	POXC	Min C	Protein
Northwest	2017	0.437	0.629	0.529
	2018	-0.01	0.10	-0.11
Wooster	2017	0.319	0.525	0.262
	2018	0.38	0.26	-0.36

Soil health indicators: POXC = permanganate oxidizable carbon; Min C = mineralizable carbon; Protein =soil protein.

2.4 Discussion

Corn Temporal Dynamics

Significant seasonal variability of all the plant measurements under these long-term treatments suggests plants are requiring and obtaining different levels of nutrients from the soil at different growth stages which was expected. For instance, variability in leaf chlorophyll and similar results have also been reported in Culman et al (2013), where leaf chlorophyll also peaks at corn V10 and R1 in all treatments, indicating higher nitrogen uptake potentially from inorganic fertilization done at V5. It was expected to see changes in total plant aboveground biomass and nitrogen uptake especially during the V10 and R1 stages since these are the stages where the plants growth increases dramatically and is in preparation to transition to the reproductive stages. Temporal changes in biomass and nitrogen uptake are also observed in (Bender et al., 2013) and (Osterholz et al., 2018) where values increase drastically during the V10 to R1 corn stages.

Tillage effects on plant measurements

One component of our objectives was to study the effects of 55+ continuous years of different tillage intensities (moldboard plow, chisel till, no-tillage) on corn seasonal dynamics and productivity. When averaged over rotations, plant measurements including total biomass, total nitrogen uptake and leaf chlorophyll were all higher in reduced tillage

treatments (NT and CH) compared to intensive plow, and no significant differences between tillage treatments were observed for corn yield. These results might suggest that reduced tillage promotes better corn plant growth and health and yields not different than intensive tilled soils. A meta-analysis by Pittelkow et al., (2015) and a literature review by Ogle et al., (2012) where no-till soils have generally lower yields in cooler regions with higher precipitation. However, other studies results are consistent with our findings that reduced tillage has higher or similar yields compared to intensive tillage (Phillips et al., 1980; DeFelice et al., 2006; Sindelar et al., 2015; Daigh et al., 2018). In fact, studies have reported that on no-till first years or even decades of implementation, yields tended to be lower on poor-drained soils compared to well-drained soils (Dick et al., 1986) (DeFelice et al., 2006). However, our results suggest that after 56 years of no-till, grain yields become equal or similar the poorly drained soils (Northwest) under no-till compared to tilled soils.

Rotation effects on plant measurements

A second component of our objectives was to study the effects of half a century of different crop rotations (continuous corn, corn-soybean and corn-forage-forage) on corn seasonal dynamics and productivity. When averaging over tillage, all plant measurements including total biomass, total nitrogen uptake, leaf chlorophyll and yields were higher in soils under more diverse rotations (CS and CFF) compared to corn monoculture,

suggesting that increasing crop rotations in corn growing systems benefit plant growth and nitrogen uptake and promotes higher yields compared to corn monoculture. Higher values for leaf chlorophyll, plant total nitrogen, and grain yield in diverse crop rotation compared to corn monoculture were also reported in Culman et al., (2013). Other studies have documented the increase on corn yields with increasing crop rotations compared to corn monoculture (Karlen et al., 2013b; Sindelar et al., 2015; Jarecki et al., 2018) and to a corn-soybean rotation especially when incorporating forages like alfalfa (Osterholz et al., 2018).

Tillage and rotation interaction effects on plant measurements

Although tillage and rotation interactions vary per site and year, and were mostly significant for 2018, plant measurements including leaf chlorophyll, total biomass and total N uptake values in no-till soils increase with increasing crop rotations, especially the CFF rotation in Northwest, compared to corn monoculture. These results might suggest that integrating two years of forages improves corn plant growth and nitrogen uptake in no-till soils compared to tilled soils and other rotations. However, significant interactions in corn yield were only observed in 2018 where yields were higher in the no-till soils under the corn-soybean rotation in Wooster, but lower in the CFF rotation in Northwest. During 2017, there were no significant tillage and rotation interactions in Northwest; however, a trend of yield increasing in the no-till soils with increasing rotation is

observed, which concurs with the other plant measurements findings. These results may be due to changes in abiotic factors between years. Other studies have also reported higher corn yields in no-till soils with increased crop rotation diversity compared to continuous corn monoculture (Halvorson et al., 2002; Daigh et al., 2018) and similar results in no-till soils with increased rotations compared to tilled soils (Dick and Van Doren, 1985). Other meta analyses state that generally no-till produces lower yields compared to tilled soils (Pittelkow et al., 2015); however, that these effects vary by latitude and yields may be similar to tilled soil when including well management like crop rotations and residue management (Pittelkow et al., 2014).

What is the relationship between soil health indicators and corn development and productivity?

Our final objective for this study was to describe the relationship between soil health indicators and corn development and productivity. When performing correlations by site, year and stage, the relationship between soil health indicators and plant measurements were highly variable and inconsistent across sites, years and stages. These data might suggest that there is a complex relationship between temporal soil health indicators and plant growth measurements that needs to be better and more intensively explored (i.e. use different statistical analyses methods for studying these relationships). Overall, correlation coefficients were mostly positive and varied between -0.2 to 0.7.

A general trend found when studying all the correlations between soil health indicators and plant measurements in Northwest 2017 was that higher values were found for the corn early growth stages and the R6 stage, suggesting that on stages where corn growth and nutrient uptake is higher (V5, V10) and during corn physiological maturity, active organic matter stabilization and mineralization may be promoted more compared to other stages. However, different trends are observed during Northwest 2018 where there are negative and/or weak correlations during the corn early growth stages, and stronger and positive correlations during corn silking stage (R1) which may be due to changes in soil moisture and precipitation per year (2017 had more precipitation and soil moisture than 2018 in Northwest). In Wooster 2017, correlation between soil health indicators and plant measurements were stronger and positive during corn V10 and R1 stages in general, and during 2018 we can see additional higher correlations during corn V5 (although some correlations results vary between stages). These correlation results might indicate active organic matter stabilization and mineralization being promoted during corn stages where nutrient and water requirements are the highest for growth and development.

When focusing on the corn total biomass, total nitrogen uptake and grain yields in the physiological maturity stage, correlation coefficients were higher in the 2017 growing season especially in Northwest where Min C had also the strongest relationship of the soil health indicators. These results might suggest that Min C more strongly related to corn biomass, nitrogen uptake and yield than the other soil health indicators (POXC and soil protein). Similar results were found in Culman et al., (2013), where Min C had a stronger

correlation to corn biomass and grain yield out of the soil health indicators. As studying the correlations between the soil health and plants across different corn growth stages was complex, studying these correlations across stages and not by each stage, and sub-setting the data separately for each of the treatments may help to better understand these relationships better.

When observing trends and not correlations coefficients, soil health indicators and plant measurements were higher in reduced tillage treatments except for yields where no significant differences were observed between tillage treatments except in Wooster 2018 where yields were higher in no-till soils compared to tilled soils. These results might suggest that after half a century of treatments, not only soil health increases in reduced tillage soils (chisel and no-till), but plant biomass, chlorophyll, and total plant nitrogen increase as well and no differences in yield are observed in soils of Ohio. This also suggests that after more than 50 years of no-till, yields are equal or not different from tilled soils which are characterized for having higher yields than no-till. Rotation effects, on the other hand, were stronger for both soils and plant measurements, emphasizing the importance of including rotations in corn cropping systems, especially two years of perennial forages that have the ability to fix nitrogen like alfalfa and red clover. The importance of including diverse crops in rotations are also highlighted in (Osterholz et al., 2018) where cropping systems with more diverse rotations and nitrogen inputs increased corn nitrogen uptake and grain yield; but interestingly, these results were not primarily due to the soil N dynamics in these diverse systems but they suggest these results may be due to plant-microbe interactions and/or soil physical properties. The only

inconsistency among results were observed in Northwest yields in 2018 being significantly lower in the no-till soils; however, this result may be due to abiotic factors such as precipitation which was lower than the past 10-year average. In summary, plant measurements and soil health indicators were overall higher in reduced tillage soils, especially in no-till, and in the corn-forage forage rotation suggesting that including diverse rotations with perennial and nitrogen fixing forages in no-till soils may increase soil health and plant quality, and produce similar or no different yields than tilled soils under less diverse rotations especially continuous corn, in soils of Ohio.

2.5 Conclusion

Our study evaluated the effects of different tillage intensities and crop rotation diversity in corn temporal dynamics and productivity in one of the oldest no-till and crop rotations research plots of America. Overall, total biomass, total nitrogen uptake and leaf chlorophyll were all higher in reduced tillage treatments compared to intensive plow, and no significant differences between tillage treatments were observed for corn yield during the time of this study. Also, crop rotation had a stronger effect on plant measurements where all values were higher in the more diverse rotations compared to continuous corn suggesting that increasing crop rotations in corn growing systems benefit plant growth and quality and promotes higher yields compared to corn monoculture. Interactions of treatments were mostly non-significant across sites and sampling years. The relationship

between soil health indicators and the different corn measurements in this study were highly variable; however, when focusing on corn physiological maturity stage, especially 2017, soil health indicators showed a moderate to strong correlation with plant measurements, especially Mineralizable C.

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