

ABSTRACT

ROPER, WAYNE ROBERT. Evaluating Soil Health in North Carolina. (Under the direction of Dr. Deanna Osmond and Dr. Joshua Heitman).

Soil health is the capacity for soils to function while sustaining life in ecosystems. This functional capacity may be maintained by stimulating chemical, physical, and biological soil properties, but land management practices that possibly improve soil health have not been evaluated in all environments. To evaluate how land management practices affect soil health in North Carolina (NC), we measured the properties of soils from long-term agronomic trials in the coastal plain (19 yr), piedmont (31 yr), and mountain (25 yr) regions of NC. Trials included various combinations of tillage (no-till, in-row subsoiling, chisel, disc, and moldboard) and management (conventional and organic). Our results showed that soil health tests were not consistently differentiating the effects of agronomic practices on overall soil health for any chemical or physical indicators, but differences were observed for biological indicators. Because soil organic carbon (SOC), is a major factor regulating soil biology, we measured SOC using four methods. Results from one method were not predictive of results from another method based on correlations, and agronomic practices did not have similar effects on SOC when comparing different methodologies. Cover cropping was added to the piedmont trial to improve soil physical resilience, but with the exception of aggregate stability, physical conditions of the piedmont soil were similar between winter-fallow and continuously covered soil after two years. Our study shows that agronomic practices have variable effects on soils in NC and that soil health evaluations should be interpreted with greater consideration of the intrinsic limitations of soils in different environments.

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Evaluating Soil Health in North Carolina

by
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DEDICATION

To my advisors, cohorts, and family who supported me throughout this process. I thank all of you for your kind words and helpful criticism.

BIOGRAPHY

Wayne was born in Detroit, Michigan in the United States of America. He grew up learning to appreciate compassion, intelligence, and brevity.

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Thank you to my wonderful friends, cohorts, and mentors in the Department of Crop and Soil Sciences at NC State. I appreciate how much you all have helped me grow and realize my potential as a great scientist.

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Chapter 1: Evaluating Soil Health in North Carolina

1.1 Introduction

Soil health is defined as ‘the capacity for soil to sustain life and function as part of an ecosystem’ (USDA-NRCS, 2015). This definition includes life within soil and the various ecosystem services it performs to assist ecological productivity (Karlen et al., 1997; Doran and Zeiss, 2000). Using the word ‘health’ to describe soils emphasizes vitality that is powered and sustained by different soil components. Healthy interaction between physical, chemical, and biological soil properties helps sustain the resources needed to keep soil functioning. In exchange for food, water, and nutrient resources, soil biota recycle nutrients (Delgado and Follett, 2002), filter water (Keesstra et al., 2012), and decompose organic residues (Carter, 2002). These same resources are also used for agriculture and other types of production that relies on soils. If the extraction of resources is greater than the capacity for soils to restore those resources, soil productivity declines (Dias et al., 2015), which is why sustaining life in soils is an important component of soil health management.

Although soil health is a metric for the condition of soils, it is not an independently measurable characteristic. Soil health is evaluated based on measurable chemical, physical, and biological soil properties that may be indicative of soil productivity (Doran and Parkin, 1994; Karlen et al., 1997; Doran and Zeiss, 2000). Soil properties used as indicators of soil health vary depending on perceptions of the significance that different properties have on ecosystem processes. The quality of soils has often been interpreted based on their potential to supply resources for agronomic management (Karlen et al., 2003; Lal, 2015), but soil health management expands that interpretation to consider how soils respond to various agronomic practices that are believed to improve natural ecosystem processes.

The major practices of soil health management are to reduce disturbance, provide residue cover, maintain living roots, and optimize functional diversity (Doran and Zeiss, 2000; USDA-NRCS,

2015). When combined, these practices may increase soil resilience by increasing soil carbon and nutrient cycling (Doran and Zeiss, 2000; Larkin, 2015; Corstanje et al., 2015), but variability in the productivity of natural systems may not always demonstrate these effects. Soil management without tillage (no-till) decreases erosion and increases structural stability compared to tilled soils (Langdale et al., 1979; K. C. McGregor and C. K. Mutchler, 1992; Nyakatawa et al., 2001). Despite some observed benefits of no-till, there are not always related improvements in crop yields (Johnson, 1994; Soane et al., 2012), carbon storage (López-Fando and Pardo, 2011; Powlson et al., 2014), or water capacity (Strudley et al., 2008; Minasny and Mcbratney, 2017), which indicates that the practice has limited effects on productivity in certain soils. In some environments, cover crops increase soil carbon (Blanco-Canqui et al., 2015), plant biomass (Finney et al., 2016; White et al., 2017), and nutrient cycling (Schomberg et al., 2006), but in other environments, cover crops were proven difficult to manage with no benefit to soil productivity (Smith et al., 2014; Kaspar and Bakker, 2015). Biological diversity in soils increases resistance to pests so that natural systems are not overwhelmed by disease, and may improve productivity if plant and microbial species are also functionally diverse (Larkin, 2015). Research suggests that soil health practices have variable and limited effects on soils, and these practices should be evaluated in different environments to learn how local climatic and edaphic factors impact soil health.

Many soil properties have been proposed as indicators of soil health (Doran and Zeiss, 2000; Picone et al., 2002; Haney et al., 2006; Moebius et al., 2007; Cardoso et al., 2013; de Paul Obade and Lal, 2016). Because it is laborious and expensive to consider every proposed soil property when evaluating soil health, developers of soil health tests have emphasized the most critical factors. Soil biological functionality is a major factor that differentiates soil health from other concepts, such as soil quality (Doran and Zeiss, 2000; Lehman et al., 2015), but biology is not the only focus of soil health. Physical and chemical soil properties also influence soil productivity, but consideration of

different types of soil properties depends on the evaluator, which makes it difficult to develop a uniform soil health assessment. Two recently developed soil health tests are the Haney soil health test (HSHT) (Haney et al., 2006, 2010) and the comprehensive assessment of soil health (CASH) (Idowu et al., 2008; Moebius-Clune et al., 2016). Both tests are configured so that measurements of multiple soil properties are used to calculate a soil health score, but the HSHT favors biological soil properties whereas the CASH includes an assortment of chemical, physical, and biological soil properties. In comparison to conventional soil testing methods, the soil health tests measure a broader assortment of soil properties, but the extent of their usefulness for guiding soil management decisions is unclear.

As soil health management continues trending, its concepts are being applied to research in different regions. In the Midwest United States, a study reported that cover cropping improved soil productivity for no-till and tilled soils (Hammac et al., 2016). In Ontario, Canada, diverse cropping systems reportedly had better soil health than monoculture systems (Congreves et al., 2015). A study in Shanghai China reported that the most critical factors for evaluating soil health were dependent on local soil conditions (Bi et al., 2013). Research is contributing to a larger understanding of what different soil conditions imply about modern interpretations of soil management

The subtropical climate and diverse landscapes of North Carolina are host to soils with various textures and parent materials. Research in the area supports reduced tillage as a means to reduce soil erosion (Meijer et al., 2013), as well as cover cropping and organic residues to increase C pools (Larsen et al., 2014), but there are still many unanswered questions about the long-term effects of soil health management and whether current methods of evaluating soil health will produce more reliable recommendations for soil management. As the chemical, physical, and biological soil properties most closely associated with perceptions of soil health are more intently

evaluated, the variability in time and intensity that changes in land management will have on soil health should be carefully researched to avoid misinterpreting the effects these management practices actually have on soils.

To better understand how soil health may be interpreted in North Carolina, multiple agronomic trials comparing different management practices were included in a study of soil health indicators. The trials are located within the mountain, piedmont, and coastal plain physiographic regions that represent the diverse landscapes of North Carolina. First, different soil health tests were compared on their ability to use soil health indicators to differentiate agronomic management. Each test uses different soil health indicators but the extent to which the chosen indicators are able to differentiate the effects of agronomic management on soil health of North Carolina soils is uncertain. Soil organic matter content has a significant influence on other soil properties, which makes it essential for managing soil health. Because soil organic matter can be measured in different ways, results from different methods may change interpretations of the impact of agronomic management on soil organic matter. The second part of the study was to evaluate the relationships between different methods of measuring soil organic matter and the ability for those methods to differentiate agronomic management of North Carolina soils based on soil organic matter content. An agronomic trial in the piedmont included conservation tillage, but did not include cover cropping. A cover crop was added to this trial in order to observe the short-term effects of cover cropping on physical properties of piedmont soil. These objectives combined will provide insight on the implications of the current concepts and metrics of soil health management for North Carolina soils.

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Chapter 2: Soil Health Indicators Do Not Differentiate Agronomic Management of North Carolina Soils

ABSTRACT

Recent soil tests evaluating “soil health” on a broad scale may not properly consider the intrinsic limitations of soil properties, and have not been assessed in regionally unique soil conditions. To evaluate three soil tests in North Carolina, we used long-term agronomic management trials from three distinct physiographic regions: mountain (22 yr), piedmont (32 yr), and coastal plain (17 yr). Mountain and coastal plain trials included combinations of organic or chemical management with or without tillage; the piedmont trial included nine different tillage treatments. Soil samples were collected and submitted for analysis as recommended by the North Carolina Department of Agriculture and Consumer Services, Haney soil health test (HSHT), and Cornell comprehensive assessment of soil health (CASH). Plant nutrient concentrations varied but were still sufficient for crops. The CASH physical soil indicators, such as surface hardness and aggregate stability, were not statistically different, regardless of tillage intensity or management. Biological soil indicators (e.g., CO₂ respiration) responded differently to management, but this differentiation was inconsistent among locations and tests. Despite many years of conservation management, the CASH results described mountain soils as “low” or “very low” soil health for all but no-till organic management, which received a “medium” score. The HSHT results considered soil from all but moldboard plowing (piedmont) to be in good health. Finally, there was no correlation between soil health tests and crop yields from North Carolina soils. Soil health tests should be calibrated to better differentiate among soil management effects that vary depending on intrinsic soil limitations.

2.1 Introduction

Soil serves as a medium for many natural processes that rely on dynamic interactions among physical, chemical, and biological soil properties to regulate important ecological and environmental functions. The state of these processes in any given soil is called “soil health”, which is defined as the capacity of soil to function as a vital living ecosystem (Doran and Zeiss, 2000; Karlen et al., 2003; USDA-NRCS, 2015). Healthy soil is believed to be highly productive, optimally functional, and naturally able to recover from disturbances (Doran and Parkin, 1994; Kibblewhite et al., 2008). The physical, chemical, and biological soil properties that comprise soil health vary in quality because of regional differences in major soil forming factors, which include parent material, climate, biota, topography, and time (Buol et al., 2011). Soil-forming factors are well known and soil properties are measurable, but the current emphasis on soil health requires an integrative assessment of how intrinsic soil properties are affected by soil management.

The inherent complexity within soil systems complicates soil health evaluations because temperature, precipitation, and other climatic factors can interact with intrinsic soil properties in variable ways. These effects are not equal for all soils, which indicates that soil properties vary because of combinations of different physical, chemical, and biological processes. Several soil indicators have been suggested by researchers attempting to define soil health (Idowu et al., 2008; Morrow et al., 2016), but the effectiveness of combining analyses of different soil indicators into a comprehensive interpretation of soil health remains elusive.

Crop and soil management practices such as crop rotations and reduced tillage can significantly influence soil productivity (Arshad and Coen, 1992), and soil evaluations are used as guides for appropriate application of similar beneficial management practices. Because the soil health philosophy emphasizes interactions between soil properties in agronomic systems, it is increasingly important to make both qualitative and quantitative assessments of the soil properties

considered to be significant for soil evaluations and also to note how they are affected by agronomic management. It has been shown that no-till planting and cover cropping may add organic matter to soil, which improves physical structure (Tisdall and Oades, 1982), stimulates biological activity (Varvel et al., 2006), and enlarges the pool for C and nutrient cycling (Campbell et al., 1996), but there is limited research about the soil health indicators that are most representative of soil properties under different soil conditions.

A single soil health indicator is unlikely to provide a complete assessment of soil productivity (Liebig et al., 2001), so soil tests have been developed to analyze several soil properties simultaneously and interpret their use for a desired management objective. Some soil tests, like the Mehlich-3 soil test used by the North Carolina Department of Agriculture and Consumer Services (NCDA&CS), are regularly used to provide agronomic recommendations for plant nutrient applications (Hardy et al., 2014). There is a long history of agronomic recommendations from NCDA&CS soil testing for mineral nutrients, but their recommendations do not include biological or physical soil evaluations. Some soil tests integrated biological soil health indicators into their analyses as the importance of more comprehensive soil testing was recognized. The HSHT includes CO₂ respiration as a soil health indicator in addition to nutrient testing (Haney et al., 2006), whereas the CASH developed by the Cornell Soil Health Testing Laboratory includes multiple physical, chemical, and biological soil health indicators in its evaluation (Moebius et al., 2007). Although the HSHT and CASH analyze more soil properties than traditional soil tests, there is no confirmation that assessments of soil health indicators will guide soil management recommendations to improve soil health for different soils.

Because soil health tests are promoted as guides for soil management practices that can improve soil productivity, there is an increasing need to ensure that the soil health indicators used in testing are capable of differentiating among the effects of various soil management practices on

diverse soils. Soil tests that are not calibrated to quantify responses to management may provide misleading recommendations that do not improve environmental functions or agronomic productivity because of intrinsic soil limitations. We used three distinct long-term agronomic management trials to: (i) compare results from different soil tests, (ii) assess the ability of soil health indicators to differentiate the soil management effects on soil properties in North Carolina, and (iii) assess the relationships between soil health and crop yields.

2.2 Methods and materials

2.2.1 Experimental plots and design

There are three physiographic regions in North Carolina (coastal plain, piedmont, and mountain) that vary in soil characteristics (e.g., texture, organic matter, mineralogy), rainfall, temperature, and other climatic factors. To capture this variability, a research trial from each region was used in the analyses. These long-term agronomic trials are located in Goldsboro (17 yr), Reidsville (32 yr), and Mills River (22 yr), which represent the coastal plain, piedmont, and mountain physiographic regions of North Carolina, respectively (Figure 2.1). Each trial has a unique management history that broadened the scope of the soil health test evaluation (Table 2.1).

The Goldsboro (coastal plain) research trial was established in 1999 at the Center for Environmental Farming Systems research farm (35°22'59.9808"N, 78°2'19.6722"W) on soil mapped as Wickham sandy loam (fine-loamy, mixed semiactive, thermic Typic Hapludults) with interspersed sections of soil mapped as Tarboro loamy sand (mixed, thermic Typic Udipsamments). The trial began with four management treatments with asynchronous crop rotations, which meant that different crops were planted for different treatments depending on the year (Mueller et al., 2002). There were two treatments with chemical management using synthetic pesticides and fertilizer. One treatment included tillage (conventional tillage chemical, CTC) and the other was no-till (no-till chemical, NTC). They had 3-yr rotations of corn (*Zea mays* L.), peanut (*Arachis hypogaea* L.), and

cotton (*Gossypium hirsutum* L.) from 1999 to 2005 and changed to 3-yr rotations of corn, the hybrid variety of sorghum [*Sorghum bicolor* (L.) Moench] and sudangrass [*Sorghum × sudanense* (Piper) Stapf] lines combined into one species (sorghum-sudangrass), and double-crop soybean [*Glycine max* (L.) Merr.] with winter wheat (*Triticum aestivum* L.) in 2006. One organic treatment (conventional tillage organic 1, CTO1) included conventional tillage to produce soybean, sweet potato [*Ipomoea batatas* (L.) Lam.], and cabbage (*Brassica oleracea* L.) with winter wheat from 1999–2001. Corn and soybean were planted for CTO1 from 2002 to 2007, then sorghum-sudangrass in 2008 followed by two fallow years. Since 2011, CTO1 has had a 3-yr rotation with clean-tilled corn, soy bean, and a year of a stale seedbed with a sorghum-sudangrass cover crop. The other organic treatment (CTO2) included conventional tillage to produce continuous corn from 1999 to 2002. From 2003 to 2007, CTO2 had corn, soybean, and organic forage followed by a fallow period from 2008 to 2010. In 2011, CTO2 changed to a 3-yr rotation of corn, soybean, and sunflower (*Helianthus annuus* L.). For CTO2, a rye (*Secale cereal* L.) cover crop was also planted before soybean, and a rye and legume cover crop mixture was planted before corn and sunflower. Changes to the organic systems were intended to better manage pests and other obstacles detrimental to crop yields. The four treatments were replicated three times for a total of 12 plots ranging in size from 0.43 to 2.30 ha (1.08–5.68 ac). The plots were organized in a randomized complete block design. bean, and a year of a stale seedbed with a sorghum-sudangrass cover crop. The other organic treatment (CTO2) included conventional tillage to produce continuous corn from 1999 to 2002. From 2003 to 2007, CTO2 had corn, soybean, and organic forage followed by a fallow period from 2008 to 2010. In 2011, CTO2 changed to a 3-yr rotation of corn, soybean, and sunflower (*Helianthus annuus* L.). For CTO2, a rye (*Secale cereal* L.) cover crop was also planted before soybean, and a rye and legume cover crop mixture was planted before corn and sunflower. Changes to the organic systems were intended to better manage pests and other obstacles detrimental to

crop yields. The four treatments were replicated three times for a total of 12 plots ranging in size from 0.43 to 2.30 ha (1.08–5.68 ac). The plots were organized in a randomized complete block design.

Corn was planted in rows that were 76 cm (30 in.) apart and had varying row lengths depending on plot sizes. Approximate corn planting density was 67,500 seeds ha⁻¹ (27,000 seeds ac⁻¹) in conventional systems and 75,000 seeds ha⁻¹ (30,000 seeds ac⁻¹) in organic systems. After planting, N was added to corn crops at a rate of 170 kg N ha⁻¹ (150 lbs ac⁻¹) as suggested by the realistic yield database for soil series in North Carolina (North Carolina Interagency Nutrient Management Committee, 2014). Fertilizer N for chemically managed plots was in the form of urea-N; for organically managed plots, a combination of raw poultry litter and predicted N mineralization from cover crop residue was used to meet the 170 kg N ha⁻¹ rate. Salt fertilizers of P and K were applied in amounts recommended by the NCDA&CS soil test in chemically managed plots; organically managed plots received P and K based on the amount of raw poultry litter applied as N fertilizer. Before 2011, yield was estimated by harvesting 12 m (40 ft) of two rows with a combine. After 2011, yield was estimated from hand-harvesting 6 m (20 ft) of two rows and manually shelling the corn. All harvested corn was adjusted for uniform moisture content at 15.5% and yield was extrapolated to a per hectare harvest. Unharvested crop residue was left to decompose on the soil surface until the next cropping season. Monthly average temperatures for the April to October growing season ranged from 10 to 22°C (50–72°F) for lows and 24 to 33°C (75–91°F) for highs. Monthly average rainfall for the rain-fed plots ranged from 86 to 145 mm (3.4–5.7 in.) during the same period.

The Reidsville (piedmont) research trial at the Upper Piedmont Research Station (36°23'2.1372" N, 79°42'6.8436" W) has soil mapped as a Toast coarse sandy loam (fine, kaolinitic, mesic Typic Kanhapludults). In the first 5 yr from 1984 to 1989, the field was continuous corn and

from 1989 to 2015, there was a corn and soybean rotation (except from 2005 to 2007, when corn was planted for three consecutive years), all with a 96.5-cm (38 in.) row spacing (Cassel et al., 1995; Meijer et al., 2013). There were nine tillage treatments replicated four times for a total of 36 plots in a randomized complete block design. Each plot had six rows and an area of 5.8 by 15.2 m (19 by 50 ft). All piedmont plots were managed with chemical pesticides and fertilizer and included NTC, disking in spring (DS), in-row subsoiling in spring, chisel plowing in spring (CPS), chisel plowing in fall (CPF), chisel plowing and disking in spring (CPDS), chisel plowing and disking in fall, moldboard plowing and disking in spring (MPDS), and moldboard plowing and disking in fall (MPDF). Tillage treatments were selected to represent minimal soil disturbance (no-till) to severe disturbance (moldboard plow) and tillage traffic was restricted to the same rows every year.

Plot maintenance included yearly fertilizer applications in spring according to NCDA&CS soil test recommendations. Fertilizer N applications to corn were split throughout spring to a total of 179 kg ha⁻¹ (160 lbs ac⁻¹); no fertilizer N was applied to soybean crops. Planting density was approximately 75,000 seeds ha⁻¹ (30,000 seeds ac⁻¹) for corn and 343,894 seeds ha⁻¹ (137,500 seeds ac⁻¹) for soybean. Piedmont crop yields were calculated by harvesting 15.2 m (50 ft) of the middle two rows of a plot with a combine, adjusting the mass to a uniform moisture content of 15.5% for corn and 13% for soybean, and extrapolating yield to a per-hectare harvest. Crop stover remained in the field to decompose after harvest. For tilled treatments only, crop residues were tilled into the soil before planting the next crop. Average monthly temperatures for the piedmont area range from 12 to 19°C (54–66°F) for lows and from 21 to 31°C (70–88°F) for highs during the April to October growing season. Average monthly rainfall ranged from 100 to 120 mm (3.9–4.7 in.) during the same period.

The Mills River (mountain) research trial at the Mountains Horticultural Crops and Research Extension Center (35°25'39.126''N, 82°33'24.7068''W) began in 1994 on soil mapped as Delanco silt

loam (fine-loamy, mixed, semiactive, mesic Aquic Hapludults). The study included five treatments: NTO, NTC, chisel and disk tillage with organic management (CTO), chisel and disk tillage with chemical management (CTC), and chisel and disk tillage with no fertilizer or pesticide inputs (CTX) as a control treatment (Hoyt, 2005; 2007). The five treatments were arranged in a completely randomized design with four replications and a total of 20 plots, each sized 24.4 by 12.2 m (80 by 40 ft) with 16 crop rows. Organic plots were managed as such, but only received USDA organic status in 2010. From 1994 to 1999, there was a 3-yr rotation of corn with fall cabbage (*B. oleracea*), cucumber (*Cucumis sativus* L.) with fall cabbage, and tomatoes (*Solanum lycopersicum* L.). From 2000 to 2005, the cropping system was a 3-yr rotation of peppers (*Piper* sp.), yellow squash (*Curcubita pepo* L.) with fall broccoli (*B. oleracea*), and tomatoes. The cropping system was changed to continuous sweetcorn (*Z. mays* convar. *saccharata* var. *rugosa*) in 2006 and again changed in 2014 to an annual corn and soybean rotation with only the NTC, CTC, and CTX treatments (organic management treatments were removed). Each fall, winter wheat was planted as a cover crop in all chemically managed plots, and a cover crop mixture of winter wheat and crimson clover (*Trifolium incarnatum* L.) was planted in organic plots. Fertilizer N was applied at a rate of 202 kg N ha⁻¹ (180 lbs N ac⁻¹) in forms applicable to chemical or organic management, depending on the plot. The soil fertility of chemically managed plots was maintained by following NCDA&CS soil test recommendations (except for CTX), and organically managed plots received nutrients based on the composition of organic fertilizer applied as poultry litter pellets (Larsen et al., 2014; Edgell et al., 2015). Insect pests were managed with with Zeon Technology (Syngenta, Basel, Switzerland) [generic name, λ-cyhalothrin, (1a[S*],3a[Z])-cyano(3-phenoxyphenyl)methyl-3-(2-chloro-3,3,3-triñuoro-1-propenyl)-2,2-dimethylcyclopropanecarboxylate] in chemically managed plots and Entrust [a mixture of spinosyn A [(2R,3aS,5aR,5bS,9S,13S,14R,16aS,16bR)-2-([6-deoxy-2,3,4-triO-methyl-a-L-mannopyranosyl]oxy)-13-4 (dimethylamino)-2,3,4,6-tetradecoxy-b-D-

erythropranosyloxy-9-ethyl-2,3,3a,5a,5b,6,7,9,10,11,12,13,14,16a,16b-hexadecahydro-14-methyl-1H-8-oxacyclododeca(b)as-indacene-7,15-dione] and spinosyn D [(2R,3aS,5aR,5bS,9S,13S,14R,16aS,16bR)-2-([6-deoxy-2,3,4-tri-O-methyl- α -L-mannopyranosyl]oxy)-13-4-(dimethylamino)-2,3,4,6-tetradecoxy- β -Derythropranosyloxy-9-ethyl-2,3,3a,5a,5b,6,7,9,10,11,12,13,14,16a,16b-hexadecahydro-4,14-dimethyl-1H-8-oxacyclododeca(b)as-indacene-7,15-dione] (Dow Agrosiences, Indianapolis, IN) in organic plots. Weed pests were managed with synthetic herbicides in chemically managed plots but with mowing or rototilling between crop rows in organic plots. Sweetcorn was planted at approximately 65,000 seeds ha⁻¹ (26,400 seeds ac⁻¹). Yield was measured by hand-harvesting sweetcorn ears from 10-m (39-ft) lengths of the two center rows of each plot and extrapolating it to a per-hectare harvest.

2.2.2 Soil sampling

Soil samples for the HSHT were collected from piedmont and mountain trials as part of a USDA-NRCS research project in fall 2014. For NCDA&CS and CASH analyses, soil samples were collected in November 2015 from piedmont trials and in December 2015 from coastal plain and mountain trials. Soil sampling procedures for the HSHT and NCDA&CS were similar: 8 to 10 subsamples totaling 473 cm³ (2 cups) were collected from each plot using a soil probe with a 2-cm (0.78 in.) diameter to collect from the top 15 cm (6 in.) of soil. The soil was mixed, dried at 60°C for 24 h, and submitted to Cornell University according to recommendations from the NCDA&CS and the HSHT lab. Soil samples submitted to Cornell were collected according to recommendations in the CASH manual (Gugino et al., 2009). In brief, three to five subsamples were collected with an auger to a depth of 15 cm (6 in.) to obtain approximately 1400 cm³ (6 cups) of soil from each plot. After sampling, the soil was stored in sealed plastic bags and refrigerated at 4°C until being shipped to the Cornell Soil Health Testing Laboratory in January 2016. Three penetrometer (Field Scout SC-900, Spectrum, Aurora, IL) measurements to 45 cm (18 in.) depth were conducted in each plot while

soil moisture was approximately at field capacity. The Cornell Soil Health Testing Laboratory was contacted to verify that this approach was acceptable for measurement at the plot scale, since their recommended procedure is for 10 measurements. The greatest pressures from the 0 to 15 cm (0–6 in.) and 15 to 45 cm (6–18 in.) depths were included in penetrometer data submitted to Cornell.

2.2.3 Soil testing and soil health scores

The NCDA&CS uses the Mehlich 3 soil extractant (Mehlich, 1984a) to analyze soil for essential plant nutrients and the Mehlich buffer pH method (Mehlich, 1976) for lime recommendations. A separate procedure to measure humic matter (HM) includes a colorimeter and a 650 nm light filter to detect the percentage of HM in the soil solution after extracting humic and fulvic acids using an alkaline solution containing NaOH (Mehlich, 1984b). The nutrient results of the NCDA&CS soil test are based on soil volumetric concentrations and are converted into weight per area based on a 20-cm (7.9-in.) depth (Hardy et al., 2014). The NCDA&CS soil test's recommended nutrient applications are provided on a per-area basis and do not include guidelines for tillage, cropping, N fertilizer or other management needs beyond plant nutrients and lime.

The HSHT for essential plant nutrients relies on the Haney, Haney, Hossner, and Arnold (H3A) soil extractant that contains the organic plant root exudates typically associated with plant nutrient uptake from soil (Haney et al., 2006; 2010). The HSHT tool requires user inputs of expected crop yield and it provides fertilizer and cropping recommendations based on user inputs and soil analyses. Nutrient recommendations from the HSHT are based on concentrations of nutrients extracted from 4 g of soil, which is converted into units of weight per area. In addition to the nutrient extractions is a measurement of CO₂-C released (in mg L⁻¹) during a 24-h incubation of wetted soil (Franzluebbers et al., 2000). A HSHT score is calculated using the amount of CO₂-C release in 24 h along with a separate procedure from the H3A extract to measure soil concentrations

(in mg L⁻¹) of water-extractable organic C (WEOC) and water extractable organic N (WEON) as follows:

$$\text{Soil Health Score} = \frac{CO_2 - C}{10} + \frac{WEOC}{100} + \frac{WEON}{10}$$

The overall health score of the CASH is a combination of 12 quantifiable soil health indicators (Gugino et al., 2009; Moebius-Clune et al., 2016) of the most pertinent soil properties considered to affect soil health (Idowu et al., 2008). Several different analyses of physical, chemical, and biological soil properties are included in the CASH, as described in the manual. Soil health indicators for a soil sample are scored based on a database of results from processed soil samples that were used to create separate cumulative normal distribution curves normalized to values between 0 and 100 for each indicator. The soil health indicator curves are adjusted for fine, medium, and coarse soil textures, with the exceptions of surface hardness, subsurface hardness, soil respiration, and P, K and “minor elements” (plant micronutrients Fe, Mg, Mn, and Zn combined as one index), which are not adjusted for soil texture. Soil health indicator results from soil samples submitted for CASH analyses are compared with the cumulative normal distribution curve of the indicator and the reported health score is representative of where the measurement value of the soil indicator is located on the normalized cumulative distribution curve. For example, if a soil indicator measurement from a soil sample is better than 80% of all the samples used to create the curve, the soil sample would receive a score of 80 out of 100 for that indicator. This method is used to score all 12 indicators, and the overall CASH index score for a soil sample is reported as an unweighted mean of the 12 indicators, which is supplied along with soil management recommendations.

The CASH chemical assessment involves the modified Morgan extractant (McIntosh, 1969) to extract essential plant nutrients from the soil. The results are used to calculate four separate

indices for pH, P, K, and minor elements. The physical soil health indicators are available water capacity (AWC) measured by volume (Reynolds and Topp, 2008), surface and subsurface hardness measured with a penetrometer (Duiker, 2002), and aggregate stability measured with a Cornell sprinkle infiltrometer (Ogden et al., 1997). Biological soil health indicators measured for the CASH are organic matter (OM) by loss on ignition (LOI) (Broadbent, 1965), active C or permanganate oxidizable C (Weil et al., 2003), soil respiration of CO₂ after 96-h incubation (Zibilske, 1994), and soil protein content via spectrophotometry (Wright and Upadhyaya, 1996), which are all related to soil microbial activity.

2.2.4 Statistical analysis

A separate statistical analysis was conducted for each trial and soil testing method. Statistical analysis software (version 9.3, SAS Institute, Cary, NC) was used to perform an ANOVA for individual soil test parameters as they may have been affected by soil management treatments within the location. Differences in soil test parameters were analyzed using the proc glimmix procedure with block as a random effect. A similar ANOVA was conducted for each location to compare mean crop yields among treatments, for which the proc glimmix procedure was used with block and year as random effects in the model. Differences among treatments for each parameter were tested for significance at the 95% confidence level using the Scheffe means separation test. For statistical comparisons of fertilizer recommendations from NCDA&CS and HSHT, the analysis was between the two tests instead of the treatments. Correlations between the most recent crop yields and CASH index scores were calculated using the graphing features of Microsoft Excel 2013 software (Microsoft Corporation, Redmond, WA).

2.3 Results and discussion

2.3.1 The NCDA&CS soil Test

Soil test results from NCDA&CS included several soil properties and elemental analyses, but those pertinent for this study were HM, cation exchange capacity (CEC) estimated via summation, pH (1:1 soil/water by volume), and Mehlich-3 P and K. All soil test parameters considered in the analyses are summarized with *F*-test *p*-values organized by location (Table 2.2). Other plant nutrients were not considered for statistical analysis because they are not typically limiting factors in North Carolina soils.

Soil test P concentrations were different among the mountain treatments only, with NTO having greater Mehlich-3 P concentrations than CTC (Table 2.3) because it received poultry litter that provided excess P beyond critical soil test values. Although the critical P concentration suggested by NCDA&CS is 120 kg P ha^{-1} , previous research on mountain soils (Cahill et al., 2013) and soil test P research (Cox, 1992) have suggested that the critical levels are between 31 and 47 kg P ha^{-1} , and thus P was sufficient for all treatments at all locations. Soil P fertility has been linked to tillage because soil-adsorbed P tends to be depleted by tillage with greater erosion potential (Alberts and Moldenhauer, 1981; Gaynor and Findlay, 1995). The fact that tillage was not a significant factor controlling Mehlich-3-extractable soil P at any location was expected, because all plots had been continuously fertilized according to NCDA&CS recommendations for crop nutrients.

Both the coastal plain and mountain trials had different Mehlich-3-extractable K concentrations among treatments. The organic treatments (CTO1 and CTO2) of the coastal plain had greater K concentrations than the CTC and NTC treatments. Likewise, at the mountain location, NTO and CTO had greater K concentrations than CTC, but NTO had similar K concentrations to CTC and CTX. No differences in K availability were observed in the piedmont, whereas the average soil test P of NTC and CPF was slightly below the 195 kg K ha^{-1} recommended by NCDA&CS, but only slightly,

again demonstrating that the fertility of major plant nutrients was not a significantly limiting variable in the trials.

Treatments of the coastal plain and piedmont trials had similar CEC, but the CEC of mountain treatments was statistically greater in NTO than in NTC, CTC, and CTX. Because the CEC reported by NCDA&CS is a summation of the number of cations detected in the soil solution, it is not a direct measure of CEC. The addition of cations from organic fertilizers and liming agents can add cations that are subsequently detected and measured as CEC, but are not completely representative of the soil's capacity to hold cations. The mountain NTO treatment may have had a statistically greater CEC than the other treatments because it received additional cations in the form of organic fertilizer that was not incorporated into the soil, not because of the inherent ability of the soil to hold cations.

Soil HM, an organic C fraction sometimes correlated to soil organic matter (Kononova, 1966), was not different among the treatments (Table 2.3). The piedmont NT and MPDF treatments represented two tillage extremes that could influence OM content, but they did not differently affect soil HM as measured by the NCDA&CS soil test. Reduced tillage was expected to allow organic substances to accumulate in the soil, but the NCDA&CS analysis showed no consistent trend of increasing HM with reduced tillage. Neither tillage nor management produced statistical differences in HM among the treatments in the trials, which meant that HM was not a differentiating parameter in these management systems. Soil management effects on soil C content can vary, but one recurring theory is that tillage does not reduce soil organic C but instead redistributes it among tillage depths, which means that sampling depth could greatly affect soil C analyses (D'Haene et al., 2009). Although this is a possible explanation for why HM may not have differentiated among tillage treatments, some research indicates that the HM analysis itself may not be a reliable indicator for soil C content (Lamar and Talbot, 2009) and may not accurately represent soil OM.

2.3.2 Haney Soil Health Test

The HSHT, as mentioned previously, was only used in the mountain trial and three treatments at the piedmont. The piedmont treatments (NTC, CPDS, MPDS) were selected to represent varying degrees of soil tillage; fall tillage treatments were not included because no yield differences between spring and fall tillage systems occurred during the trial (Meijer et al., 2013). Soil nutrient availability detected by the HSHT was similar to NCDA&CS soil testing results, with the mountain treatments NTO and CTO having more P and K than the conventional treatments (Table 2.4). Organic treatments probably contained more nutrients because of the excess P and K typically found in organic fertilizers applied to satisfy N requirements (Smith et al., 1998; Larsen et al., 2014; Edgell et al., 2015).

Many soil extractants cannot simultaneously measure N and other plant nutrients (Holford, 1980), but the H3A extractant does not have this limitation (Haney et al., 2006). Measured values of $\text{NO}_3\text{-N}$ and $\text{NH}_4\text{-N}$ from the H3A extractant as well as predicted mineralized organic N represent the total N measurement of the HSHT. Both the CTC and CTO treatments of the mountain trial had greater total soil N than CTX, but not significantly more than the CTC and CTO tillage treatments (Table 2.4).

The amount of WEOC in soils was not statistically different among treatments at either location (Table 2.4). The amount of WEON measured in the mountain treatments was greater in soil from NTO than from CTO and CTX, but not CTC (Table 2.4). In the piedmont trial, more WEON was found in soil from NTC than in MPDS but not CPS. Plant residue contributes more organic C and N when left on the soil surface than when incorporated by tillage (Smith and Sharpley, 1993; Chen et al., 2014), and the treatments had increasing WEON with decreasing tillage intensity, but the differences among tillage effects were not great enough at either location to differentiate no-till management from all other tillage. The piedmont and mountain trials were conducted for over two

decades before this analyses, but research suggests that it may take centuries for crop residue deposition to significantly increase soil OM in some areas (Poeplau and Don, 2015; Strickland et al., 2015).

The amounts of CO₂-C released from soil of the mountain treatments ranged from 48 to 158 mg L⁻¹ on average, but large variability in CO₂-C release resulted in no statistical differences among treatments (Table 2.4). At piedmont locations, CO₂-C was statistically greater in NTC than in CPDS and MPDS. Microbial release of soil C can greatly vary depending on management, soil composition, soil collection methods (Rochette et al., 1991), and laboratory methodology (Parkin and Kaspar, 2004). Measurements of CO₂-C released from soil have been correlated to soil biological activity (Franzluebbers et al., 2000; Wang et al., 2003), and CO₂-C is likely to evolve more quickly in soil with more organic C and N being available to microbes (Touratier et al., 1999; Nguyen and Marschner, 2016). Although there was more CO₂-C from NTC in the piedmont trial, variability in the soil CO₂ released from treatments in the mountain trial meant that soil management was not consistently influencing soil biological activity as measured by microbial respiration.

The HSHT uses CO₂-C, WEOC, and WEON measurements to calculate soil health scores for soils. The average HSHT scores for mountain treatments ranged from 7 to 21, but these scores were statistically similar because of the large variability among replicate samples within treatments (Table 2.4). Piedmont HSHT scores ranged from 5 to 16 on average, and NTC scored higher than both CPDS and MPDS. Because a score of seven or greater from the HSHT is considered to be good soil health, all treatments except MPDS in the piedmont trial were considered to be “healthy” on average. Interpreting the practicality of HSHT scores is difficult, however, because CO₂-C release from soils as well as soil organic C and organic N pools can have significant temporal and spatial variability (Rochette et al., 1991). Soil texture also influences CO₂-C release (Franzluebbers et al., 2000), but it is not a factor considered in the HSHT scoring.

2.3.3 NCDA&CS and HSHT nutrient recommendations

The NCDA&CS test does not use its standard soil test to provide soil-specific N fertility recommendations. Instead, N recommendations in North Carolina are made by using the realistic yield expectation (RYE) tool, which is regularly updated to provide a realistic N rate based on soil mapping units and crop species (North Carolina Interagency Nutrient Management Committee, 2014). The HSHT samples were collected in the fall of 2014 and NCDA&CS samples were collected in the fall of 2015, but it was not expected that soil properties, including fertility, would significantly vary over the course of a single year. Expected yields from the RYE tool were entered into the HSHT tool, which recommends fertilizer based on expected yields and concentrations of soil nutrients extracted via H3A. The RYE tool recommended 129 kg N ha⁻¹ for the piedmont corn crop and 203 kg N ha⁻¹ for the mountain corn crop, and the HSHT recommended—for the same crop—a range between 121 and 139 kg N ha⁻¹ for piedmont corn and 205–246 kg N ha⁻¹ for mountain corn on average, which were statistically different for most of the treatments (Table 2.5). Fertilizer N recommendations from the HSHT are based on the amount of water-extractable NH₄-N, water-extractable NO₃-N, WEON, and microbial activity contributing to total N, which means that the HSHT fertilizer N recommendations are more dynamic than those in the RYE database. Microbial activity and soil organic N are considered by the HSHT to be significant factors contributing N to crops. The amount of N fertilizer recommended for corn production in the mountain trial was probably greater than the piedmont N fertilizer recommendation because the cooler mountain climate is favorable for greater corn yields, and the RYE database reflects this.

Soil test P was variable in both tests, which resulted in only MPDS having a greater P recommendation from the HSHT than from the NCDA&CS (Table 2.5). The amount of P fertilizer recommended by the HSHT is based on the amount of extracted inorganic P and organic P predicted by soil organic C to N ratios, which means that more P will generally be in soils receiving more

organic material. The recommendations for K fertilizer differed for several treatments, with NCDA&CS recommendations generally being greater than the HSHT recommendations. The differences in recommendations may be because the H3A extract lacks calibration for diverse soil conditions, whereas the Mehlich-3 extractant used by NCDA&CS has been tested and calibrated to predict plant uptake of P and K during a growing season (Indiati et al., 1997).

2.3.4 Comprehensive Assessment of Soil Health

Data from measurements of the soil health indicators are provided along with the CASH soil health index scores. There are 12 separately rated soil health indicators included with the CASH, and their index scores are averaged to calculate an overall soil health index score. Index scores range from 0–100, with 0 described as “very low” soil health by the assessment, 100 described as “very high” soil health, and intermediate descriptors of “low”, “medium”, and “high” between them (Moebius-Clune et al., 2016). The indicators are grouped into chemical, biological, and physical categories. Like the HSHT and NCDA&CS soil tests, plant nutrients were sufficient in soils at each location (Table 2.6). There were no statistical differences among treatments for any of the chemical soil health indicators (pH, P, K, or minor elements). The CASH index scores for pH ranged from 2 to 23 for coastal plain, 10 to 58 for piedmont, and 6 to 46 for mountain soil, but the results were too variable to differentiate among treatments. The NCDA&CS recommends a soil pH of 6.0 for corn and soybean crops grown in North Carolina, which many of the treatments averaged (Table 2.3), but the CASH index scores for treatments with soil pH < 6.0 indicated that soil pH was too low in these soils. Soil pH in these trials are not likely to limit crop growth, as they are considered appropriate for southern soils, which indicates that the scale at which the CASH is evaluating pH is not well adjusted for North Carolina soil conditions. Concentrations of nutrients extracted by the modified Morgan extractant have a good correlation to the concentrations of nutrients extracted via Mehlich 3 (Ketterings et al., 2002), but there is no reliable conversion of soil test results between the two

methods when different soil textures are considered. Soil extractants should be matched to the geographic area of the soils for which they have been developed and calibrated; otherwise, additional considerations will be needed for evaluating soil health across different regions (Ketterings et al., 2002; Herlihy et al., 2006).

Physical soil health indicators of the CASH were AWC, surface hardness, subsurface hardness, and aggregate stability. Tillage effects on soil penetration were inconsistent and there were no statistical differences among treatments at any location despite the large ranges in treatment means (Table 2.6). Subsurface hardness scores were also similar among treatments. Even no-till treatments failed to obtain better surface and subsurface hardness ratings from the CASH, which may indicate that the hardness test is not sensitive or consistent enough to differentiate management effects on the soils in the trials. Surface crusting typically develops in intensive tillage systems without residue cover (Pagliai et al., 2004), but crusting has also been observed in no-till systems to varying degrees (Rosa et al., 2013). In the piedmont trial, more soil crusting was observed in soil from tilled treatments without residue cover, but bulk density from 0 to 10 cm was lower when soil was tilled (Cassel et al., 1995), which complicates interactions between soil physical properties and their potential benefits to soil health. Subsurface hardening can result from compaction by machinery or plowing (Torbert and Reeves, 1995; Birkás et al., 2004), but with no statistical differences seen among physical soil health indicator scores for tillage treatments, there was no measured soil management effect on subsurface hardness even after many years of cropping on the mountain, piedmont, and coastal plain soils in North Carolina.

All treatments at each location scored “very low” for aggregate stability (Table 2.6), which is in the 0 to 40 range of the CASH index. No-till management is typically associated with improved soil structure (Carter, 2002; Bronick and Lal, 2005), so it was expected that no-till treatments would perform significantly better than tillage treatments, but management was not differentiated by the

aggregate stability test. Index scores for AWC were all within the medium (55–70) to high (70–85) range of the CASH index (Table 2.6), but were not statistically different among treatments within any of the three locations. Conservation tillage was implemented in each of the trials, but after many years of consistent management, this was still not differentiated from conventional tillage according to the physical soil health indicator measurements of the CASH.

The biological soil health indicator component of the CASH is comprised of OM, soil protein, soil respiration, and active C. No location had treatments with statistically different OM scores (Table 2.6) and, regardless of tillage or management, all treatments had “very low” (0–40) or “low” (40–55) soil health for OM according to the CASH index. Soil protein scores had no statistical differences among treatments within the coastal plain and mountain trials, but the soil protein scores of the piedmont soils were greater for NTC than for CPS, CPDS, MPDS, and MPDF (Table 2.6).

Soil respiration scores among treatments were statistically different only at the piedmont trials where soil respiration scores for DS and CPS were greater than that for MPDF (Table 2.6). Active C is different from soil respiration because it measures easily accessible C regardless of the current microbial activity in the sample. Unlike soil protein and soil respiration scores, there was no statistical difference among active C scores for piedmont treatments, which varied among replications of each treatment (Table 2.6). There were, however, statistical differences among active C scores for treatments in the coastal plain and mountain trials. Active C scores for coastal plain treatments were greater in NTC than in CTO1 and CTC, but not CTO2. For treatments in the mountain trial, NTO had more active C than CTC and CTX, but not NTC or CTO. The overall trend in biological soil health indicator scores showed that biological activity increased with reduced tillage and often with organic soil amendments. There was no consistent differentiation among management systems across the three trials in North Carolina, however, and biological soil

indicators for all treatments had average ratings of “very low” (0–40) or low (40–55) on the CASH index scale.

Overall soil health index scores from the CASH averaged between 38 and 46 for the coastal plain, 35–46 for piedmont, and 44–55 for mountain soil (Table 2.6). Very few of the individual soil health indicator component scores were statistically different among treatments, which is probably why overall CASH soil health index scores for the treatments were not statistically different for two of the three trials. All treatments in the coastal plain and piedmont trials were rated “very low” (0–40) or “low” (40–55) by the CASH index, but NTO in the mountain trial, which received a “medium” score of 55, was statistically greater than the score for NTC and CTX.

The only CASH index scores with statistical differences among management treatments were soil biological health indicator scores, but all of them received no better than “low” (0–40) scores on average, regardless of management. Because long-term trials were used in the research, the soils were assumed to be in a stable state that is representative of the long-term effects of soil management on the soil properties used to assess soil health, and the scores received by the CASH are likely to be representative of the equilibrium of soil management effects on soil health indicators.

Included with the CASH were soil management recommendations to implement practices that could ideally improve soil health scores. The recommended practices were already implemented in treatments that received less than ideal scores (NTC, NTO), so there was little more that could be done to improve the scores of individual treatments if the management followed only the soil health assessment guidelines. Soils in all the trials were managed to satisfy fertility requirements, so there was no concern about deficiencies in chemical soil health indicators. Physical soil health indicators did not differentiate among soil management systems, despite there being

varying degrees of tillage, and biological soil health indicators were inconsistently affected by management. As a consequence of the chemical, physical, and biological ratings, the overall soil health scores did not differentiate among the agronomic systems. The CASH did not provide more soil management information than that gained from soil tests designed for nutrient recommendations. If many years of conservation tillage did not result in discernable soil health scores among different types of soils and agronomic management, the current standards for soil health indicators may encompass too broad of a range of soils or lack the sensitivity to differentiate between the effects of recommended soil management practices.

Soils formed in different climates have intrinsic limitations in their ability to improve under favorable conditions (Sanchez et al., 2003), which implies that different standards may need to be considered to evaluate soil health. Researchers continue to evaluate soil indicators to determine how they can best be used to improve recommendations (Morrow et al., 2016), and some research is already being conducted to improve the CASH system to account for soil variation because of regional factors (Congreves et al., 2015; Moebius-Clune et al., 2016). Changes to soil health evaluations should consider that soil indicators vary in their significance for soils in different regions. For example, North Carolina soils are typical of Ultisols in the southeastern USA that have low organic matter content and an acidic pH, and have been irreversibly weathered to retain fewer nutrients than soils in the northeastern US climate (Buol et al., 2011), where the CASH was developed. By adjusting soil health assessments for regional soil properties, recommendations for improving soil management may better differentiate among agronomic management systems and also consider their practical implications for soil productivity.

2.3.5 Implications of soil C as a soil health indicator

A common parameter of each soil test is a surrogate measurement of soil C content. Soil C is a vital component of soil systems because C cycling is the driving force of many soil ecological

interactions (Goto et al., 1994). In many soil systems, C content is strongly correlated with microbial activity (Lavahun et al., 1996), which favors ecosystem processes. A soil C component included in a soil test can capture this important link between soil properties, but the measure of soil C availability that best differentiates among management systems has not been determined. Although soil C related to soil biological activity has been used to evaluate soil health, measurements of the same soil management systems tend to differ because of spatial and temporal variability of soil C content and microbial activity (Rochette et al., 1991).

In the case of soil C measurements used by soil tests considered in this study, HM (Table 2.3), CO₂ respiration (Table 2.4; Table 2.6), LOI (Table 2.6), and active C (Table 2.6) were the tests chosen to represent soil C. The value of the HM analysis used by NCDA&CS has been questioned in other research because of its inconsistent correlation with other soil C measurements (Lamar and Talbot, 2009). No statistical differences in HM content were observed for treatments in any of the trials, and this may be because HM is not a strong indicator of soil organic C. The OM content determined by the CASH also did not differ among treatments at any location. The LOI technique used by the CASH can be skewed because LOI generally lacks precision to quantify the organic C content of soils with less than 15% OM, which may cause soil organic C overestimations arising from heating temperatures and durations that can volatilize other soil constituents along with soil C (Szava-Kovats, 2009; Huang et al., 2012; Hoogsteen et al., 2015).

When other soil C measurements were conducted, however, some separation among soil management practices was observed. Measurements of WEOC by the HSHT did not differentiate among treatments of the piedmont and mountain trials; however, the HSHT analysis of CO₂-C release from soil managed by NTC in the piedmont trial was greater than that in soil managed via CPDS and MPDS (Table 2.4). Previous research from the piedmont trial revealed a greater abundance of fungal biomarkers in NTC (Muruganandam et al., 2009), and fungal activity is also

associated with decomposition of recalcitrant forms of C that may be converted to CO₂ (Strickland and Rousk, 2010; Malik et al., 2016). Although, the HSHT CO₂-C measurements did not show differences among the treatments in the mountain trial, previous research from there revealed differences in laboratory measurements of field-moist soil CO₂ respiration among treatments (Overstreet and Hoyt, 2008). Respiration previously measured from soil in the mountain trial was observed to be greater in the crop rows of organically managed plots than in crop rows of plots with synthetic chemical management, even though total C from the treatments was similar. It was suspected that additional C added from organic fertilizers stimulated biological activity. The CASH measurement of soil CO₂ respiration was not different among mountain treatments (Table 2.6) but did reveal that piedmont tillage treatments DS and CPS had greater CO₂ release than MPDF. Spatial and temporal variability in soil CO₂-C evolution may be the reason why soil tests are inconsistent in detecting differences between management systems.

By using an elemental analyzer and chloroform-fumigation-extraction, another soil organic C experiment conducted for the mountain trial detected more total soil organic C and microbial biomass C in NTO than in other treatments (Wang et al., 2011). A later experiment also revealed that NTO had more total C, high and light fraction particulate organic matter, and microbial biomass C than other treatments (Larsen et al., 2014). The analyses matched expectations that a combination of no-till and organic production would have more microbial activity and labile organic C than conventional tillage systems (Huggins et al., 1998; Zuber and Villamil, 2016). Active C results from the CASH analysis, however, showed that although active C was greater in NTO than in CTC and CTX (Table 2.6), it was not different from that in NTC and CTO.

The NCDA&CS soil test results showed no differences in HM among treatments (Table 2.3). The HSHT revealed more CO₂-C was released from NTC than from tillage treatments at the same location (Table 2.4), but the CASH CO₂-C analysis suggested otherwise (Table 2.6). Across all three

soil tests, the only C measurement that was different among treatments of the mountain trial was active C (Table 2.6), but when other soil C research was conducted on the mountain trial soils there were statistical differences among treatments for other C pools. Although most statistical differences among treatments were found in soil C analyses, patterns of differentiation using soil C analyses were inconsistent in their ability to differentiate among management systems. Inconsistency in the ability of soil C measurements to differentiate among soil management practices is problematic for soil health tests because of the importance of soil C to ecosystem functions of soil properties associated with C pools. If measurements of soil C availability are inconsistent for soil management, then management recommendations are also likely to be inconsistent. Other factors may therefore need to be considered to improve soil health in those systems.

2.3.6 Crop yields

The NCDA&CS, HSHT, and CASH soil tests only focus on soil properties, but crop yields can possibly be integrated with soil health indicators to provide a measure of sustainable agronomic management. Fields of the coastal plain and piedmont trials were typically used for producing corn and soybean, whereas sweetcorn was the predominant crop of the mountain trial.

Corn yields from the years 2002–2013 of the coastal plain trial were used in the analysis. Because corn was grown asynchronously across treatments, year-to-year variability may not be equally reflected in treatments, though overall yields during the period are still representative of management effects. Overall mean yields from treatments in the coastal plain trial were not statistically different during the period 2002–2013 (Table 2.7). Mean corn yields ranged from 5463 kg ha⁻¹ corn for CTC to 7105 kg ha⁻¹ corn for CTO2. The RYE for the Wickam sandy loam soil series is 9604 kg ha⁻¹ corn (North Carolina Interagency Nutrient Management Committee, 2014), which indicates that long-term soil management for corn yield from the coastal plain trial is not as

productive as it could be. This is partially because soil management in the coastal plain trial has been hindered by challenges such as pest pressure and flooding. Average rainfall did not significantly vary over the course of the trial, so it was not expected that rainfall provided an advantage for yields in any particular year.

Corn yields from the piedmont trial from 1987 to 2015 were considered because the first 3 yr of yield data were not available. Although the trial involved an annual rotation of corn and soybean, there were some years when corn was grown in successive crop years for various reasons. In total, there were 17 corn harvests in the piedmont trial, and mean corn yield was different among treatments (Table 2.7). The greatest yielding treatment was NTC, which yielded 6516 kg ha⁻¹ corn; the lowest yielding treatment was MPDF, which yielded 3374 kg ha⁻¹ corn. Corn yield from NTC was similar to that of in-row subsoiling in spring and CPF, but was greater than that for all other tillage treatments. The Toast coarse sandy loam soil series within the piedmont is predicted by RYE to yield 7846 kg ha⁻¹ (North Carolina Interagency Nutrient Management Committee, 2014). Although long-term yields have averaged less than RYE, yields in individual years of the trial have exceeded RYE (data not shown). Thus, it is possible for management conditions in the piedmont trial to produce the yields expected for the soil series, despite the soil receiving CASH index scores that imply otherwise (Table 2.6).

The NTC treatment at the piedmont trial averaged from the 10 soybean crop years from 1990 to 2014, yielded 2832 kg ha⁻¹ (Table 2.7). Other tillage treatments had soybean yields that were statistically similar to that of NTC, with the exception of MPDS and MPDF, which yielded 1991 and 1942 kg ha⁻¹ corn, respectively. The Toast coarse sandy loam soil series is predicted by RYE to yield 2556 kg ha⁻¹ soybean (North Carolina Interagency Nutrient Management Committee, 2014), which means that long-term soybean yields are exceeding expectations for the soil series despite the CASH index scores implying that the soils had poor soil health.

Sweetcorn was first planted in 1997 and 1998 for the mountain trial, but was not planted again until the continuous sweetcorn crop that lasted from 2007 to 2013. Mean sweetcorn yields for the trial ranged from the lowest at 9200 kg ha⁻¹ sweetcorn for NTO to the highest at 17,283 kg ha⁻¹ sweetcorn for NTC (Table 2.7). Long-term average yields from NTC and CTC were statistically greater than those from NTO and CTO. CTX plots were not included in the analysis because they produced no yield because of the lack of management. No RYE exists for sweetcorn grown in this soil series, so there is no direct yield comparison. Yield differences in the mountain trial were attributed to difficulties with preventing weed competition and pests, which decreased yields in organic treatments, which had with limited management options (Larsen et al., 2014; Edgell et al., 2015). Management was especially challenging in the NTO treatment, which was the least productive treatment of the mountain trial. Although NTO management had the highest CASH overall soil health index score on average (Table 2.6), because of management issues, the measured health of the soil was not reflected in crop yields.

The soil health philosophy promotes sustainable soil management practices that are believed to be beneficial for environmental health and agricultural productivity. Soil management in the trials included conservation practices such as long-term no-till agriculture and cover cropping, both of which are recommendations to improve soil health as defined by CASH (Gugino et al., 2009; Moebius-Clune et al., 2016). Based on the scores the soils received from the CASH, however, there was no correlation between the ability for long-term soil conservation to achieve acceptable soil health scores and high agricultural productivity (Figure 2.2). Recent corn yields from plots of the piedmont nine-tillage trial did not significantly correlate with CASH soil health scores ($r^2 = 0.01$) and neither did sweetcorn yields from plots of the mountain trial ($r^2 = 0.10$). The coastal plain data were not used to correlate recent crop yields and recent soil health scores because of the asynchronous cropping design of the trial. Even though soil health assessments did not clearly differentiate among

agronomic management systems, the yield results showed that conservation tillage frequently produces greater yields when used in combination with proper fertilizer and pesticide inputs. Given that these results are consistent with the recommended best management practices, it may be useful to consider potential crop yields in soil health assessments for agronomic management.

2.4 Conclusion

We submitted soil samples to NCDA&CS, the HSHT, and the CASH to assess how their analyses and soil management recommendations related to each other and whether soil testing could differentiate among soil health indicators in diverse agronomic systems and regions in North Carolina. The tests used different extractants and methodologies to evaluate soils, and none returned results with practical or quantifiable differences among management practices used on different soils. Only no-till organic management received an adequate soil health score from the CASH, and all but moldboard plowing had average scores of good soil health with the HSHT, which places these two methods at odds with each other in their soil health evaluations. There may be substantial variability in soil properties regardless of agronomic management or it could be that measurements of soil health indicators may not be sensitive enough to differentiate among soil management effects on properties of soils with different compositions. Because soil management effects on soil health indicators used in current soil health testing are limited by intrinsic soil properties, a weighted system of soil health indicators depending on soil and climate may make soil health assessments more practical for various soil uses. Most recommendations for improving the soil health focus on conservation tillage practices like no-till and cover cropping, and we did observe greater yields in some conservation tillage systems; instead, those yields were not consistently different from conventional tillage practices. Soil health management recommendations for agronomic systems need to be adjusted to account for differences in intrinsic soil properties that are currently incapable of supporting a broad standard for soil health.

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Table 2.1. Location, year established, number of treatments and replicates, experimental design, soil series, agronomic management, and cropping history of the research trials.

	Coastal plain	Piedmont	Mountain
Location	Goldsboro, NC	Reidsville, NC	Mills River, NC
Established	1999	1984	1994
Treatments (replicates)	4 (3)	9 (4)	5 (4)
Plot size	Various sizes	5.8 by 15.2 m (19 by 50 ft)	12.2 by 24.4 m (40 by 80 ft)
Experimental design	RCBD	RCBD	CRD
Soil series	Wickham sandy loam; Tarboro loamy sand	Toast coarse sandy loam	Delanco silt loam
Agronomic management	Chemical and organic, varied tillage	Chemical, varied tillage	Chemical and organic, varied tillage
Cropping history	Conventional systems 1999–2005: Corn, peanut, and cotton 2006: Corn, sorghum–sundangrass, and double-crop soybean Organic systems Organic 1 1999–2001: Soybean, sweet potato, cabbage 2002: Corn and soybean Organic 2 1999–2002: Continuous corn 2003–2007: Corn, soybean, organic forage 2011: Corn, soybean, and sunflower	1984–1989: Continuous corn 1990 to 2015: Corn–soybean rotation	1994–1999: Corn–cucumber–tomato rotation 2000–2005: Peppers, yellow squash, tomatoes 2006–2013: Continuous corn 2014–2015: Corn–soybean rotation

Table 2.2. The *p*-values for the *F*-test statistics of select soil parameters of soil analyses from the North Carolina Department of Agriculture and Consumer Services (NCDA&CS), Haney soil health test (HSHT), and Cornell comprehensive assessment of soil health (CASH).

Soil test	Parameter	Location		
		Coastal plain	Piedmont	Mountain
NCDA&CS	Humic matter	0.2919	0.1040	0.1844
	Cation exchange capacity	0.1775	0.9479	<0.0001
	pH	0.1492	0.0887	0.0652
	P	0.0701	0.5844	0.0219
HSHT	K	0.0020	0.0532	0.0003
	N	NA [‡]	0.0267	0.0005
	P	NA	0.2411	0.0018
	K	NA	0.1319	<0.0001
	CO ₂ respiration	NA	0.0111	0.1074
	WEOC [†]	NA	0.0363	0.0363
	WEON	NA	0.0346	0.0025
	Health score	NA	0.0088	0.0439
CASH	Available water capacity	0.4402	0.1140	0.8196
	Surface hardness	0.5488	0.0289	0.1132
	Subsurface hardness	0.7709	0.1379	0.1787
	Aggregate stability	0.5376	0.2640	0.2001
	Organic matter	0.4398	0.0137	0.3216
	Soil protein	0.0600	<0.001	0.0861
	CO ₂ respiration	0.3160	0.0006	0.1273
	Active C	0.0161	0.0061	0.0016
	pH	0.5439	0.1573	0.0519
	P	0.5013	0.0561	0.1090
	K	0.4547	0.6282	0.4380
	Minor elements	0.4547	NA	NA
	Overall soil health score	0.5281	0.0031	0.0090

† WEOC, water extractable organic C; WEON, water extractable organic N
‡ NA = Not assessed

Table 2.3. Abridged soil test results from the North Carolina Department of Agriculture and Consumer Services soil testing laboratory organized by location. Soil management treatments include no-till chemical (NTC), no-till organic (NTO), conventional tillage chemical (CTC), conventional tillage organic (CTO), conventional tillage without fertilizer or pesticides (CTX), in-row subsoiling (IRS), disking in spring (DS), chisel plowing in fall (CPF) or spring (CPS), chisel plowing and disking in fall (CPDF) or spring (CPDS), and moldboard plowing and disking in fall (MPDF) or spring (MPDS). Treatments are compared within each site using the Scheffe means comparison test at $p = 0.05$. Numbers in parentheses are SD of the means and letters indicate groupings for statistical differences.

Location	Treatment	Humic matter	CEC	pH	P	K
		%	cmol kg ⁻¹		kg ha ⁻¹	
Coastal plain	NTC	0.49 (0.10)	5.03 (1.0)	5.77(0.35)	265 (86)	239 (33) b
	CTC	0.58 (0.08)	4.90 (1.4)	5.57 (0.32)	355 (208)	248 (76) b
	CTO1	0.44 (0.08)	5.50 (1.4)	6.00 (0.10)	607 (246)	336 (42) a
	CTO2	0.48 (0.06)	7.17 (2.1)	6.10 (0.17)	455 (340)	384 (75) a
Piedmont	NTC	0.32 (0.12)	5.83(0.61)	5.75 (0.35)	157 (48)	181 (39)
	IRS	0.21 (0.08)	5.90 (1.05)	6.00 (0.22)	293 (207)	230 (35)
	DS	0.10 (0.04)	5.73(0.64)	5.98 (0.50)	128 (94)	236 (48)
	CPF	0.21 (0.18)	5.73 (0.84)	5.53 (0.35)	249 (60)	189 (17)
	CPS	0.20(0.02)	5.98 (0.68)	6.15 (0.13)	256 (68)	225 (25)
	CPDF	0.19 (0.12)	5.80 (0.56)	6.18 (0.34)	298 (246)	250 (23)
	CPDS	0.19 (0.09)	5.20 (1.02)	5.85 (0.13)	182 (63)	206 (21)
	MPDF	0.17 (0.17)	5.75 (0.66)	5.98 (0.33)	267 (290)	198 (34)
MPDS	0.25 (0.13)	5.98 (0.93)	6.15 (0.32)	368 (201)	200 (28)	
Mountain	NTC	0.29 (0.06)	5.80 (0.42) b	5.98 (0.47)	154 (25) ab	242 (43) c
	NTO	0.37(0.10)	8.20 (0.67) a	6.50 (0.14)	236 (68) a	434 (82) ab
	CTC	0.27 (0.10)	5.48 (0.32) b	6.30 (0.45)	102 (27) b	288 (21) bc
	CTO	0.23 (0.07)	6.57 (0.80) ab	6.68 (0.15)	144 (71) ab	474 (83) a
	CTX	0.45 (0.25)	5.98 (0.67) b	6.30 (0.14)	118 (53) ab	284 (64) bc

† CAC, cation exchange capacity

Table 2.4. Abridged soil test results of the Haney Soil Health Test (HSHT) performed on soil samples collected in 2014 by location. Soil management treatments include no-till chemical (NTC), no-till organic (NTO), conventional tillage chemical (CTC), conventional tillage organic (CTO), conventional tillage without fertilizer or pesticides (CTX), in-row subsoiling (IRS), disking in spring (DS), chisel plowing in fall (CPF) or spring (CPS), chisel plowing and disking in fall (CPDF) or spring (CPDS), and moldboard plowing and disking in fall (MPDF) or spring (MPDS). The HSHT uses organic C, organic N, and CO₂ respiration to calculate a health score for soils. Treatments are compared within each site using the Scheffe comparison at $p = 0.05$. Numbers in parentheses are SD of the means and letters indicate groupings for statistical differences.

Location	Treatment	kg ha ⁻¹			mg L ⁻¹			Health score
		N	P	K	CO ₂ respiration	WEOC [†]	WEON	
Piedmont	NTC	47 (7.0) a	119 (58)	119 (19)	124 (57) a	154 (26)	16 (2.2) a	16 (5.6) a
	CPDS	34 (9.4) ab	69 (46)	85 (12)	52 (13) b	101 (43)	10 (4.3) ab	8 (1.6) b
	MPDS	29 (3.4) b	67 (16)	124 (51)	34 (11) b	85 (11)	9 (0.97) b	5 (1.0) b
Mountain	NTC	62 (10) a	70 (15) b	107 (7.7) b	80 (25)	315 (56)	26 (3.6) ab	12 (2.7)
	NTO	75 (4.4) a	181 (86) a	215 (32) a	155 (53)	361 (28)	32 (1.7) a	21 (4.8)
	CTC	56 (7.0) ab	40 (19) b	113 (20) b	48 (37)	237 (103)	16 (8.2) b	7 (4.6)
	CTO	55 (10) ab	100 (25) ab	238 (36) a	158 (144)	269 (59)	23 (4.8) ab	19 (13)
	CTX	35 (12) b	49 (19) b	99 (13) b	51 (22)	216 (50)	16 (4.3) b	8 (2.7)

[†] WEOC, water-extractable organic C; WEON, water-extractable organic N

Table 2.5. Fertilizer recommendations for soil samples submitted to North Carolina Department of Agriculture and Consumer Services (NCDA&CS) soil testing and Haney Soil Health Testing (HSHT) laboratories by location. Soil management treatments include no-till chemical (NTC), no-till organic (NTO), conventional tillage chemical (CTC), conventional tillage organic (CTO), conventional tillage without fertilizer or pesticides (CTX), chisel plowing and disking in spring (CPDS), and moldboard plowing and disking in spring (MPDS).

Location	Treatment	N		P		K	
		RNR [†]	HSHT	NCDA&CS	HSHT	NCDA&CS	HSHT
kg ha ⁻¹							
Piedmont	NTC	129	121 NS	0	14 NS	31	0*
	CPDS	129	133 NS	31	63 NS	12	0 NS
	MPDS	129	138*	0	44*	21	0*
Mountain	NTC	203	218*	217	160 NS	58	7*
	NTO	203	205 NS	66	17 NS	0	0 NS
	CTC	203	225*	287	231 NS	53	9*
	CTO	203	225*	157	91 NS	0	0 NS
	CTX	203	246*	265	210 NS	67	16*

* Significantly different at the 0.05 probability level between NCDA&CS and HSHT fertilizer recommendations, tested using the Scheffe means comparison test

[†] Realistic nitrogen rate (RNR) from North Carolina Interagency Nutrient Management Committee

[‡] NS, not significant

Table 2.6. Cornell Assessment of Soil Health (CASH) soil indicator index scores for each parameter included in the standard analysis grouped by location. Soil management treatments include no-till chemical (NTC), no-till organic (NTO), conventional tillage chemical (CTC), conventional tillage organic (CTO), conventional tillage without fertilizer or pesticides (CTX), in-row subsoiling (IRS), disking in spring (DS), chisel plowing in fall (CPF) or spring (CPS), chisel plowing and disking in fall (CPDF) or spring (CPDS), and moldboard plowing and disking in fall (MPDF) or spring (MPDS). Treatments are compared within each site using Scheffe means comparison test at $p = 0.05$. Numbers in parentheses are SD of the means and letters indicate groupings for statistical differences.

Location & treatment	AWC	Surface hardness	Subsurface hardness	Aggregate stability	Organic matter	Soil protein	Soil respiration	Active carbon	pH	P	K	Minor elements [†]	Overall [‡]
————— CASH index score —————													
Coastal Plain													
NTC	76 (13)	18 (14)	33 (29)	4 (1.0)	21 (7.5)	20 (3.0)	24 (4.5)	35 (13) a	8 (14)	100 (0)	100 (0)	100 (0)	45 (5.2)
CTC	65 (24)	50 (17)	22 (31)	4 (0.6)	14 (5.1)	17 (2.1)	20 (4.6)	16 (5.9) b	2 (2.9)	99 (1.1)	92 (13)	85 (25)	38 (7.6)
CTO1	67 (7.4)	23 (35)	15 (20)	4 (0.6)	14(3.2)	17 (3.8)	29 (5.0)	20 (7.9) b	22 (31)	96 (6.4)	100 (0)	100 (0)	43 (8.7)
CTO2	60 (18)	38 (38)	31 (25)	3 (2.1)	20 (11)	18 (3.1)	27 (10)	14 (3.0) ab	23 (22)	100 (0)	100 (0)	100 (0)	46 (9.5)
Piedmont													
NTC	69 (7.0)	5 (7.2)	4 (2.2)	4 (0.6)	39 (8.5)	21 (6.8) a	29 (6.1) ab	31 (11)	16 (15)	100 (0)	100 (0)	100 (0)	43 (2.6)
IRS	57 (11)	40 (18)	8 (13)	8 (6.7)	39 (15)	16 (3.4) ab	28 (11) ab	22 (18)	25 (27)	100 (0)	100 (0)	100 (0)	46 (7.4)
DS	63 (7.3)	20 (16)	1 (0.8)	3 (1.2)	36 (10)	12 (2.8) ab	32 (8.1) a	28 (16)	57 (13)	100 (0)	97 (6)	100 (0)	46 (3.7)
CPF	57 (8.9)	44 (26)	11 (11)	5 (4.4)	27 (2.1)	12 (1.3) ab	27 (7.1) ab	18 (8.0)	49 (42)	100 (0)	100 (0)	100 (0)	46 (5.3)
CPS	58 (5.0)	28 (13)	1 (2.5)	5 (5.9)	36 (6.9)	11 (1.4) b	33 (7.8) a	24 (6.2)	56 (52)	100 (0)	100 (0)	100 (0)	46 (3.7)
CPDF	54 (2.6)	29 (6.8)	2 (1.7)	5 (3.8)	32 (13)	10 (1.3) b	25 (5.3) ab	20 (12)	58 (28)	100 (0)	100 (0)	100 (0)	45 (3.8)
CPDS	53 (11)	17 (19)	1 (1.0)	6 (4.1)	29 (9.5)	12 (2.6) ab	17 (6.1) ab	9 (4.4)	16 (32)	95 (10)	100 (0)	100 (0)	38 (3.1)
MPDF	54 (2.9)	20 (13)	1 (1.0)	7 (3.3)	23 (5.4)	7 (1.0) b	9 (4.0) b	3 (2.3)	10 (20)	83 (22)	99 (3)	100 (0)	35 (3.9)
MPDS	55 (7.1)	11 (18)	7 (9.5)	3 (2.5)	22 (7.0)	7 (0.5) b	18 (4.4) ab	6 (3.2)	35 (24)	100 (0)	98 (5)	100 (0)	39 (1.3)
Mountain													
NTC	87 (5.3)	14 (14)	15 (12)	8 (5.9)	36 (9.7)	19 (7.1)	23 (11)	17 (11) ab	7 (14)	100 (0)	98 (4.5)	100 (0)	44 (6.0) b
NTO	88 (0.8)	39 (11)	38 (4.4)	9 (5.3)	40 (11)	25 (9.9)	34 (7.0)	37 (14) a	46 (25)	100 (0)	100 (0)	100 (0)	55 (4.2) a
CTC	85 (5.6)	48 (29)	24 (10)	4 (2.4)	31 (7.9)	12 (2.2)	30 (10)	12 (4.4) b	44 (38)	93 (9.6)	100 (0)	100 (0)	48 (3.1) ab
CTO	87 (3.0)	32 (22)	27 (14)	5 (3.8)	33 (8.2)	14 (4.6)	26 (2.6)	21 (7.9) ab	39 (17)	100 (0)	100 (0)	100 (0)	49 (3.4) ab
CTX	85 (7.0)	51 (16)	24 (16)	11 (3.3)	25 (12)	17 (4.8)	20 (5.8)	4 (3.5) b	6 (11)	84 (18)	100 (0)	100 (0)	44 (3.0) b

[†] The score for minor elements is the mean of subscores for Fe, Mg, Mn, and Zn soil concentrations

[‡] The overall score is an unweighted average of the 12 individual indicator index scores.

[§] AWC, available water capacity

Table 2.7. Mean corn, sweetcorn, and soybean yields from the coastal plain, piedmont, and mountain trials, organized by location. Soil management treatments include no-till chemical (NTC), no-till organic (NTO), conventional tillage chemical (CTC), conventional tillage organic (CTO), in-row subsoiling (IRS), disking in spring (DS), chisel plowing in fall (CPF) or spring (CPS), chisel plowing and disking in fall (CPDF) or spring (CPDS), and moldboard plowing and disking in fall (MPDF) or spring (MPDS). Treatments are compared using the Scheffe means comparison test at $p = 0.05$. Numbers in parentheses are the SD of the means and letters indicate groupings for statistical differences.

Location	Treatment	Corn yield kg ha ⁻¹	Sweetcorn yield kg ha ⁻¹	Soybean yield kg ha ⁻¹
Coastal plain	NTC	6080 (3555)	NA	NA
	CTC	5463 (3741)	NA	NA
	CTO1	6195 (4013)	NA	NA
	CTO2	7105 (2940)	NA	NA
Mountain	NTO	NA [†]	9,200 (5,628) b	NA
	NTC	NA	17,283 (4,942) a	NA
	CTO	NA	9,362 (6,153) b	NA
	CTC	NA	14,957 (6,325) a	NA
Piedmont	NTC	6516 (3256) a	NA	2832 (1183) a
	IRS	5754 (3104) ab	NA	2669 (1016) a
	DS	4788 (2655) bcd	NA	2532 (1138) ab
	CPF	5462 (2919) ab	NA	2767 (1269) a
	CPS	5313 (2704) bc	NA	2780 (1121) a
	CPDF	4689 (2611) bcd	NA	2616 (1321) a
	CPDS	4327 (2641) cde	NA	2301 (1241) ab
	MPDF	3374 (2238) e	NA	1942 (1263) c
	MPDS	3764 (2393) de	NA	1991 (1347) bc

† NA = Not assessed

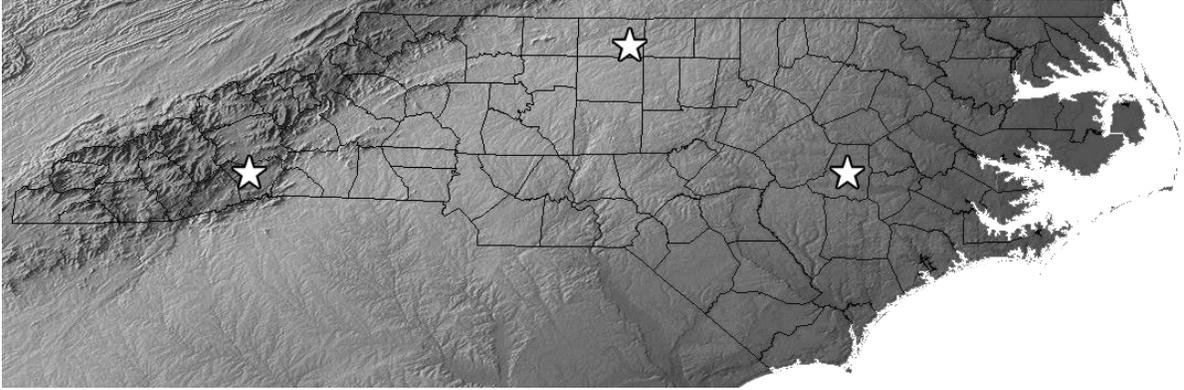


Figure 2.1. Map of tillage and cropping trials located across North Carolina. To the left is Mills River in the mountains, the center is Reidsville in the piedmont, and the right is Goldsboro in the coastal plain.

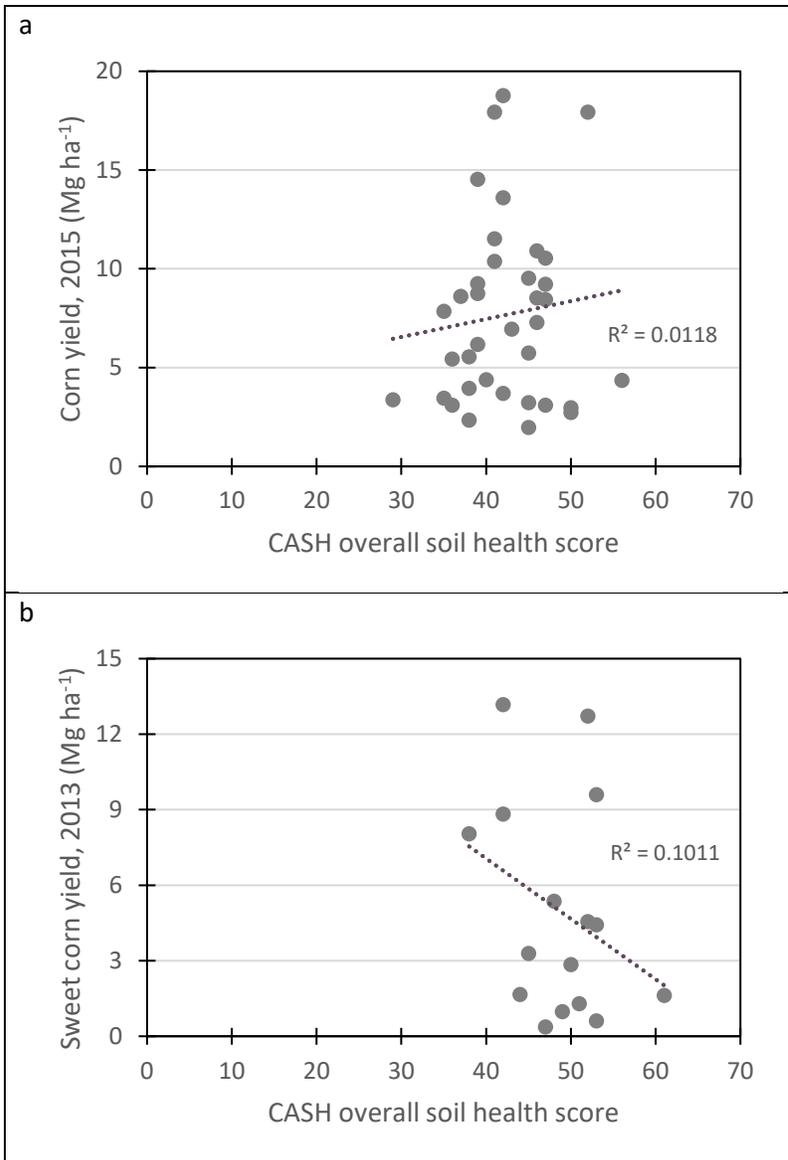


Figure 2.2. Correlation between the Cornell comprehensive assessment of soil health (CASH) overall soil health scores and recent crop yields (Mg ha⁻¹) for soils of the piedmont (a) and mountain (b) trials. Each solid circle on the graph represents an individual research plot.

Chapter 3: Comparing Four Methods for Measuring Soil Organic Matter in North Carolina Soils

Abstract

Soil organic matter (SOM) provides beneficial soil ecosystem services that are important for soil management, but it is unclear how results from different methods of measuring SOM should be compared when making soil management decisions. To compare different methods, we used 84 soil samples from long-term agronomic trials in the coastal plain, piedmont, and mountain regions of North Carolina. Coastal plain and mountain trials included combinations of tillage and management (conventional vs. organic), whereas piedmont trials were configured to evaluate tillage intensity. The methods used to measure SOM were Walkley-Black (WB), mass loss on ignition (LOI), automated dry combustion (ADC), and humic matter (HM) colorimetry. Correlations among LOI, WB, and ADC were significant ($p < 0.0001$) for the total population of soils, but variability due to location implied that HM had no correlation to other methods. For measures of soil organic carbon compared to SOM, the WB results were biased high compared to ADC, and ADC was more strongly correlated to LOI than WB. When using the methods to evaluate the effects of agronomic management on SOM, results varied for different methods and locations. Conservation management did not consistently accumulate more SOM than other soil management practices, and no method consistently differentiated soils based on management. Variation in SOM composition may be causing discrepancies in the way that SOM is interpreted using conventional methods. To avoid confusion about how agronomic management affects SOM, assessments should limit comparisons to those with similar measurement protocols.

3.1 Introduction

Organic matter has an important role in soil productivity. Studies have shown that increases in soil organic matter (SOM) can increase nutrient cycling (Delgado and Follett, 2002) and water availability for plants (Franzluebbers, 2002; Rawls et al., 2003), with limited effects in some situations (Minasny and Mcbratney, 2017). The structure and composition of SOM is variable and contains a complex mineral-organic matrix with compounds containing various C functional groups (Waksman and Stevens, 1930; Beyer, 1995; Shi et al., 2006). As such, C is closely related to SOM, which makes soil organic carbon (SOC) a likely indicator of SOM. Other elemental components of SOM are present in the mineral-organic matrix with less consistency and less abundance than C, and are not often used to estimate SOM content.

Measurements of SOM have commonly been used as a basis for evaluating the quality of soils (Karlen et al., 1997; Doran and Zeiss, 2000; Idowu et al., 2008). Because SOM is comprised of various C compounds, several methods have been developed to measure SOM as well as its constituent compounds. One common method is mass loss on ignition (LOI). This method involves thermal oxidation of SOM using a muffle furnace (Nelson and Sommers, 1996; Schulte and Hopkins, 1996). The amount of SOM oxidized using LOI is indicative of the organic mass of soil, but other methods can target fractions of SOM that are relevant to the reactivity of particular C compounds. Release of CO₂ from microbial respiration (Søe et al., 2004) and permanganate oxidation of SOC (Weil et al., 2003) are used to measure labile SOC. Recalcitrant SOC is more resistant to decomposition and representative of SOM that lasts longer in soils. Methods used to measure recalcitrant SOC are varied in their approach. Humic matter (HM) is a recalcitrant form of SOM defined by a specific chemical procedure for extracting humic acids from soils (Lunt, 1931; Mehlich, 1984). The Walkley-Black (WB) method was developed to measure recalcitrant SOC using a chemical oxidant (Walkley and Black, 1934; Walkley, 1947; Nelson and Sommers, 1996). Modern laboratory

procedures typically involve measuring SOC using dry combustion. Automated dry combustion (ADC) involves measurements of SOC based on CO₂ released from thermally-digested soil (Abella and Zimmer, 2007). Both LOI and ADC are dry combustion methods, but procedural differences between the two can lead to different interpretations of calculated amounts of total SOC. For ADC procedures, SOC is directly measured from the release of CO₂, and in the absence of inorganic carbonates, is considered the most reliable measure of SOC. Typical LOI procedures assume that the mass lost is entirely comprised of SOM, with estimates of SOC derived using adjustment factors (Chatterjee et al., 2009).

Studies of SOM have employed various measurements, but relationships between SOM measured using different analytical methods are not yet adequately understood for different soils. This may limit broader interpretations of SOM among researchers if it is found that results from one method of measuring SOM cannot be reliably compared to results from another. Although ADC is considered the standard for reporting SOC, there are also reports of SOC and SOM based solely on WB, LOI, and HM procedures. When comparing SOM measured using LOI, SOC measured using ADC or WB, and HM measured using colorimetry, variability in the composition and reactivity of SOM may cause discrepancies among the measurements for investigating changes in SOM. Further confounding interpretations of data is the estimate of SOC from SOM and vice versa. It is conventional to assume that SOC comprises 58% of the mass of SOM (Read and Ridgell, 1922; Bianchi et al., 2008), but SOC reportedly ranges from 50-66% of SOM (Lunt, 1931; Jain et al., 1997; Périé and Ouimet, 2008; Pribyl, 2010). This variability in the composition of SOM confounds estimates of SOM based on measurements of SOC. Strategies to manage SOM will only be as effective as the ability to measure and compare changes in SOM due to the effects of different soil management practices.

In a previous study of agronomic management across North Carolina, CO₂ release from microbial respiration, permanganate oxidation, HM colorimetry, and LOI were used to compare SOM in different soils (Roper et al., 2017). Measures of SOM using LOI did not differentiate soils in any of the trials. Differentiation of agronomic management effects on SOM using permanganate oxidation, CO₂ respiration, and HM colorimetry varied for soils in each physiographic region of North Carolina, with no consistency in management effects. After many years of consistent management, no difference in SOM was detected in soils when comparing conservation management practices to conventional tillage. It was not clear if these results were due to a lack of adequate sensitivity in the methods or if agronomic management had not differentially affected SOM accumulation. To further compare methods of measuring SOM in agronomic soils, SOM from the same soils was reevaluated using WB, LOI, and ADC, and compared to the previous colorimetric measurements of HM. Our objectives for this study were to (1) evaluate correlations between SOM measured using different methods and (2) assess the capacity for those methods to differentiate agronomic management of North Carolina soils.

3.2 Methods and materials

3.2.1 Agronomic trials

Soil samples were collected from four long-term agronomic trials established in the coastal plain, piedmont, and mountain physiographic regions of North Carolina (Figure 3.1). The main characteristics for each trial are provided in this text, and additional details are provided in Roper et al. (2017). For all trials, fertilizer was applied based on recommendations from the North Carolina Department of Agriculture and Consumer Services, except for organic plots that received P and K based on animal manure applied as fertilizer N. Plant residue remaining after harvest was not removed from plots.

The coastal plain agronomic trial is located at the Center for Environmental Farming Systems in Goldsboro, North Carolina (35°22'59.9808''N, 78°2'19.6722''W). This trial began in 1999 on soil mapped as Tarboro loamy sand (Mixed, thermic Typic Udipsamment) interspersed with Wickham sandy loam (Fine-loamy, mixed, semiactive, thermic Typic Hapludult). Four treatments with three replications are included in this study. One treatment involves no-till management with conventional pesticide and fertilizer applications (NTC) to produce various vegetable crops. The second treatment is conventional tillage with conventional chemical applications for managing soil fertility and pests (CTC) with the same crops as NTC. The last two treatments are organically managed with one including a three-year rotation of corn (*Zea mays* L.) and soybean (*Glycine max* L.) followed by a fallow year (CTO1) and the other including a three-year rotation of corn, soybean, and sunflower (*Helianthus annuus* L.) (CTO2).

The piedmont nine-tillage agronomic trial is located at the Upper Piedmont Research Station in Reidsville, North Carolina (36°23'2.1372'' N, 79°42'6.8436'' W). In 1984, the trial began with a continuous corn crop on soil mapped as Toast coarse sandy loam (Fine, kaolinitic, mesic Typic Kanhapludults), but has been managed as a corn and soybean rotation since 1990 (Waggar and Denton, 1989; Freese et al., 1993; Meijer et al., 2013). There are nine tillage treatments replicated four times. Treatments include NTC, in-row subsoiling (IRS), disking in spring (DS), chisel plowing in fall (CPF) and spring (CPS), chisel plowing plus disking in fall (CPDF) and spring (CPDS), and moldboard plowing plus disking in fall (MPDF) and spring (MPDS).

An additional agronomic trial at the Upper Piedmont Research Station is a four-tillage trial (36°22'44.4''N 79°41'36.1''W), on soil mapped as Pacolet sandy loam (Fine, kaolinitic, thermic Typic Kanhapludult). This trial began in 1984 with continuous corn and switched to a corn and soybean rotation in 1990 (Waggar and Denton, 1989; Denton and Waggar, 1992). The four tillage treatments

are replicated four times and include NTC, DS, alternating years of no-till and disk tillage (AYT), and cultivation tillage by disking in spring twice (DST).

The mountain agronomic trial is located at the Mountain Horticultural Crops Research and Extension Center in Mills River, North Carolina (35°25'39.126''N, 82°33'24.7068''W). This trial began in 1990 on soil mapped as Delanco silt loam (fine-loamy, mixed, semiactive, mesic Aquic Hapludults). The five management treatments were replicated four times and include NTC, no-till with organic management (NTO), CTC, conventional tillage with organic management (CTO), and conventional disk tillage in control plots managed without fertilizer or pesticides (CTX). Various vegetable crops were planted in the initial years of the trial, then the rotation changed to continuous sweet corn (*Zea mays* convar. *saccharata* var. *rugosa*) in 2007, and changed again to a corn and soybean rotation without organic management in 2014 (Larsen et al., 2014; Edgell et al., 2015). The legacy effects of previous management were expected to still be present at the time of sampling.

3.2.2 Soil sampling and analysis

In fall 2015, approximately eight subsamples of soil from the harvested rows of each plot were collected using a 2.2-cm diameter soil probe to a depth of 30 cm. Only soil from the 0-15 cm depth was used for this study. The soils were oven-dried at 60°C for 24 h before being processed and completely homogenized in a soil grinder in order to pass through a sieve with 2-mm openings. Sieved soil was mixed and stirred to reduce particle size segregation. Soils were subsequently stored at room temperature until analysis. Although the soils have previously been amended with CaCO₃ to reduce acidity, all soils were below pH 6.5 and did not respond to dilute HCl used to indicate the presence of carbonates.

Walkley-Black measurements of SOC followed a modified procedure from Nelson and Sommers (1996), which is itself a modified procedure from Walkley and Black (1947). The procedure involves using dichromate (Cr₂O₇²⁻) to oxidize SOC, and iron sulfate (FeSO₄) to reduce excess Cr₂O₇²⁻

in solution. The amount of $\text{Cr}_2\text{O}_7^{2-}$ consumed in the reaction is used to calculate the amount of SOC in the sample. For the analysis, SOC oxidation was conducted using a solution of 0.167 mol L^{-1} potassium dichromate ($\text{K}_2\text{Cr}_2\text{O}_7$). Reduction of excess $\text{Cr}_2\text{O}_7^{2-}$ was performed using 0.5 mol L^{-1} FeSO_4 .

Approximately 1 g of dried soil was placed in a 150 mL Erlenmeyer flask to which 10 mL of 0.167 mol L^{-1} $\text{K}_2\text{Cr}_2\text{O}_7$ was added. After mixing soil and $\text{K}_2\text{Cr}_2\text{O}_7$, 20 mL of concentrated sulfuric acid (H_2SO_4) was added to heat the solution and increase reactivity. The solution was then diluted to approximately 75 mL using deionized water and allowed to cool for at least 30 min. The remaining solution was filtered by gravity into a 250 mL Erlenmeyer flask after passing through 5-10 μm mesh filter paper in a glass funnel. After washing the soil and filter paper with deionized water multiple times, the solution was prepared for titration by adding 0.025 mol L^{-1} Ferroin [1,10-Phenanthroline iron(II) sulfate complex] color indicator. Titration of the solution with FeSO_4 proceeded with regular stirring until all $\text{Cr}_2\text{O}_7^{2-}$ was consumed. Blank samples (no soil added) were used to calibrate the concentration of FeSO_4 . Soil C concentrations were calculated with the assumption that all C was oxidized by $\text{Cr}_2\text{O}_7^{2-}$, no additional redox reactions with $\text{Cr}_2\text{O}_7^{2-}$ occurred while soil was suspended in the solution, and that 1 mol of C reacted for every 1 mol $\text{Cr}_2\text{O}_7^{2-}$ neutralized in solution before titrating with FeSO_4 . There was no adjustment of SOC to account for potentially incomplete oxidation as recommended by Walkley and Black (1934) because there is no standard adjustment for different soils (Meersmans et al., 2009).

Measurements of SOM using LOI were conducted with approximately 10 g of air-dry soil (Zhang and Wang, 2014). Soil was placed in a 35 mL porcelain crucible (6-cm diameter, 2.5-cm height), dried in a convection oven at 105°C , and placed in a sealed desiccator to cool. These steps were repeated until the soil and crucible had a constant mass. Next, a muffle furnace was preheated to 360°C and soil was heated in the furnace at that temperature for 2 h. After 2 h, the crucible with soil was removed from the furnace and placed in a desiccator to cool before being weighed. The

difference between the mass of the crucible and soil before heating and after heating was assumed to be the mass of SOM in the sample.

A Perkin-Elmer 2400 Series II CHNS/O Elemental Analyzer (Perkin-Elmer, Waltham, MA, USA) was used to measure SOC using ADC. Approximately 25 mg of soil was dried at 105°C and placed in a tin vial for analysis. Inside the instrument, each soil sample was heated to 925°C in the presence of a helium carrier stream with oxygen to promote oxidation. The resulting release of CO₂ was quantified by a thermal conductivity detector after being separated from other gases in a chromatography column. The detector output signal was converted to a concentration of C based on the mass of the original soil sample.

Colorimetric measurements of HM follow conventional procedures for extracting humic acids from soils (Mehlich, 1984; Hardy, 2014). A sub-sample of each soil was submitted to the North Carolina Department of Agriculture and Consumer Services for analysis. The procedure involves humic acid extraction from soil using 0.2 mol L⁻¹ NaOH, 0.002 mol L⁻¹ diethylenetriaminepentaacetic acid (DTPA), and 2% by volume ethyl alcohol (C₂H₅OH). After extraction, 5 mL of supernatant from the soil solution is diluted with 35 mL of deionized water in a polystyrene vial. Absorbance of the solution at 650 nm was measured in reference to a solution with only the extractant and water. Transmittance values were converted to a HM concentration based on previous HM calibrations conducted by the North Carolina Department of Agriculture and Consumer Services.

3.2.3 Statistical analysis

Statistical analyses were conducted using Statistical Analysis Software (SAS) version 9.4 (SAS Institute, Cary, NC). Correlations between measurements from each method were analyzed using the CORR procedure. Comparisons of SOM, SOC, and HM among treatments were conducted separately for each trial and for each methodology (ADC, LOI, WB, HM) using the MIXED procedure

with blocks as a random effect. A Tukey pairwise means comparison test with a 95% confidence interval was used for each analysis.

3.3 Results and discussion

3.3.1 Comparisons of conventional soil organic matter measurements

Among the four agronomic trials, SOM was measured from 84 soil samples collected from 84 different plots. The mean SOM content measured using LOI was consistent with SOM content for Ultisols like those used in this study (Oliveira et al., 2010; Marques et al., 2016), which is typically less than 30 g SOM kg⁻¹ or 3% of the total soil mass (Table 3.1). The lowest amounts were observed in coastal plain and piedmont soils, whereas the maximum amounts were in mountain soils.

Average SOC measured by WB was greater than SOC measured by ADC (Table 3.1). The two methods had similar ranges in measured values, but WB on average measured 1.75 times more SOC than ADC. Accepting the assumption that ADC provides a direct measure of SOC, the estimates of SOC using WB are biased high. The cause of discrepancies between WB and ADC is not clear from the results, but it does imply that the amount of SOC measured depends on the choice of methodology (Brye and Slaton, 2003).

Measurements of SOM using LOI were compared to SOC measured using ADC and WB. The average mass of SOC using ADC is approximately 33% of the average mass of SOM (Table 3.1). In comparison, average SOC measured using WB is 56% of the average mass of SOM. This is similar to the common assumption that approximately 58% of the mass of SOM is comprised of C (Read and Ridgell, 1922; Bianchi et al., 2008; Pribyl, 2010), whereas ADC results imply that the assumption is inaccurate for soils used in this study.

The average mass of HM was approximately 10% of the average mass of SOM (Table 3.1). As expected, HM colorimetry yielded the lowest masses among the 84 soil samples because HM is a different elemental component of SOM that comprises less of the total mass of SOM than SOC

(Beyer, 1993; Valladares et al., 2007; Khodorenko et al., 2012; Susic, 2016). It is also assumed that the HM extraction has the same relative efficiency for different soils, but recovery of HM is highly variable (Lamar and Talbot, 2009).

Although different methods of measuring SOC have variable results, understanding relationships among results from the methods could assist with interpretations of SOM measured in different ways. Correlations between the four methods are shown in Tables 3.2 and 3.3. Results for LOI, WB and ADC are grouped together in Table 3.2 because these methods are designed to measure total amounts of SOM or SOC, respectively.

The two measures of SOC, WB and ADC, were highly correlated ($p < 0.0001$) for the total population of soils, but the strength of the correlation varied with location (Table 3.2). Separating results for individual soils revealed a stronger correlation between ADC and WB for the mountain soil compared to the overall correlation, but other soils had weaker correlations for the same comparison. The correlation for the coastal plain soil was not significant, which implies that SOC measured by ADC and WB does not have a similar relationship for these four soils.

Relationships between SOC and SOM can vary based on the composition of SOM, but variability in the relationship may also be because of methodology. Both measures of SOC were highly correlated ($p < 0.0001$) to LOI, but differences between locations showed that ADC is more consistently correlated to LOI than WB (Table 3.2). Correlations between ADC and LOI for soils in individual locations were all significant (Table 3.2) and were greater than correlation coefficients between WB and LOI for the same soils. Also, soils in the coastal plain, piedmont four-tillage, and piedmont nine-tillage trials all had stronger correlations between ADC and LOI than the overall correlation, which implies that measurements of ADC and LOI are highly correlated for the soils despite potential variability in the composition of SOM.

As already noted, results from WB are biased high (Table 3.1) compared to ADC, which results in a larger proportion of measured SOC relative to SOM. Other research has also observed a systematic high bias when comparing WB to other methods (Brye and Slaton, 2003). Reliance on simple SOC to SOM comparisons should be approached with caution because of variation associated with soil depth, vegetative cover, and topographic position (Jain et al., 1997). Differences in the strength of correlations of ADC to LOI, and WB to LOI among locations (Table 3.2) may also reflect changes in the overall composition of SOM, with the WB protocol being less sensitive to these differences than ADC. Subtle differences due to variation in SOM composition can be masked when larger datasets are used (Christensen and Malmros, 1982).

The colorimetric measurement of HM was developed as an alternative to conventional measurements of SOM that use WB, LOI, and ADC protocols, and is used as a metric for soil testing in North Carolina (Hardy, 2014). The overall correlations for HM to WB and HM to ADC were significant (Table 3.3), but because correlations for individual soils varied in significance, HM had no consistent relationships with ADC or WB. Correlations between HM and LOI were not significant for any of the soils. Although HM has been considered as a surrogate for SOM for many years in soil testing (Mehlich, 1984), the method has mostly been used to assess potential herbicide activity (Strek et al., 1990). The amount of HM relative to other components of SOM has been evaluated (Sardessai, 1994; Valladares et al., 2007; Susic, 2016), but correlations between HM and those other components are unclear. Colorimetric measurements of HM appear to have little correlation to SOM due in part to the large degree of uncertainty in the measurement.

Discrepancies in the ratios of SOC to SOM measured using these methods reveal challenges for studying SOM when multiple methods are used. Comparing among different measurements of SOM will be difficult if relationships between the methods are not consistent or not representative of different soils. Both LOI and ADC are subject to positive bias due to the presence of inorganic

carbonates, which were tested for and determined not to be present in the 84 soil samples. Since LOI relies on changes in mass, loss of water from hydrated minerals is a possible source of error with a positive bias (Ball, 1964; Wright et al., 2008; Sun et al., 2009). Incomplete C oxidation is a source of negative bias for both WB and ADC (Walkley and Black, 1934; Meersmans et al., 2009). In addition to incomplete oxidation, WB measurements may have a positive bias due to oxidation of inorganic elements in the soil matrix, such as manganese or ferrous iron (Walkley, 1947). Correction factors for WB are typically recommended because of assumptions associated with incomplete oxidation of SOC, but the correction factors are also not consistent among soils (Jain et al., 1997; Meersmans et al., 2009).

Of the four protocols used, both LOI and ADC were more robust than the others, and were highly correlated for soils in different locations. Multiple studies utilize this strong relationship to derive calibration curves intended to help estimate SOC content based on SOM measured using LOI (Jain et al., 1997; Pérez et al., 2001; Konen et al., 2002; Konare et al., 2010). Others have argued against using conversions because of equations that are too imprecise (Szava-Kovats, 2009; Pribyl, 2010; Huang et al., 2012). For this study we focused on the ability for each method to detect differences in SOM content due to various tillage and soil conservation practices.

3.3.2 Differentiating agronomic management using measurements of soil organic matter

The LOI, ADC, WB, and HM methods have each been used as indicators of SOM, and can be evaluated separately for their utility in interpretations of the effects of agronomic management on SOM. There were 22 treatments among the four agronomic trials across North Carolina. The four methods were used on all 84 soil samples from the trials, and results were averaged per treatment. Because tillage is believed to be detrimental to SOM accumulation (Karlen et al., 2013; Liu et al., 2014), there is particular interest in comparing tillage treatments within the trials.

Treatments in the coastal plain were configured to compare inorganic and organic management as well as tillage. No-till and CTO1 had greater SOC than CTC based on WB measurements, but treatments were not differentiated by ADC, LOI, or HM (Table 3.4). Organic and no-till management in the coastal plain did not consistently have the most SOM based on measurements using these methods.

Differentiation of agronomic management effects on SOM in the piedmont nine-tillage trial was present for multiple methods. The LOI, ADC, and HM results exhibited a trend with IRS having more SOM than moldboard plowing (Table 3.4), but intermediate treatment effects differed between methods. Measured SOC from WB had no statistical difference among treatments. The NTC and IRS treatments disturb soil less than other tillage, but they did not consistently have more SOM than treatments with tillage. Conservation tillage in the piedmont nine-tillage trial did not substantially increase SOM relative to tilled soils.

The piedmont four-tillage trial included no-till management and different amounts of disk tillage. No method differentiated among treatments at this location (Table 3.4). Treatments in the four-tillage trial were arranged so that AY, DS, and DST had more soil disturbance than NTC, but no-till soils did not have more SOM than tilled soils. Because results from different methods of measuring SOM in the piedmont four-tillage trial show no difference in SOM due to management, it implies that either all the methods were incapable of detecting a difference or that there was no difference in SOM after 31 years of contrasting soil management.

In the mountain trial, management included organic and conventional practices as well as conservation and conventional tillage. Measurements of SOC using ADC indicated that NTO had greater SOC than soils of all other treatments (Table 3.4). For both LOI and WB, only the NTO and CTC treatments were different from each other. Although LOI and WB measurements resulted in similar differentiation of treatments in the mountain trial, results for the two methods were not

similar for any other location. There was no differentiation of soil management in this trial based on HM measurements. With the exception of HM, all methods imply that NTO had more SOM and SOC than CTC, but no-till with conventional management was similar in SOM and SOC to conventional tillage practices.

Both LOI and ADC differentiated management based on SOM in the piedmont nine-tillage and mountain trials (Table 3.4). Agronomic management effects on SOM were differentiated by WB in the coastal plain and mountain trials, but not in the piedmont trials. Management in only the piedmont nine-tillage trial was differentiated by HM. A summary of p-values for treatment effects separated by location and methodology is provided in table 5 in order to show the degree to which treatments were statistically different. Each location had non-tilled and tilled soils, with additional comparisons of organic and inorganic chemical management at the coastal plain and mountain locations. The ADC results indicate that no-till organic management accumulated more SOC than other soil management practices at the mountain location (Table 3.4), but for all other locations and methods, there was no consistent differentiation between conservation management and conventional management. Many years of contrasting soil management practices did not consistently result in different amounts of SOM for these soils.

Compared to methods used in Roper et al., results presented here are similar in that there is no clear differentiation of conservation and conventional agronomic management for many of the methods and locations involved. Our results indicate that conventional methods for measuring SOM are inconsistent in differentiating among agronomic management effects on SOM in North Carolina soils. There still remains significant uncertainty for correlations of SOM and SOC measured using these methods and their utility for comparisons of SOM in differently managed soils.

3.4 Conclusion

Even with our understanding of different methods to measure SOM and its elemental components, variation in the composition of SOM causes discrepancies in the way that SOM is interpreted when different methods are used to measure it. Correlations between methods can vary even for similar soils. Because ADC and LOI have the strongest correlations, these two methods are likely to be the most effective for evaluating SOM and SOC. As SOM becomes a more critical focus for soil management recommendations, it is important to note limitations among existing methods and their sensitivity to detect changes in SOM over time due to soil management. Until a reliable method of converting results between different analytical methods of measuring SOM is developed, comparisons of SOM and SOC among different analytical methods and different soils should be made with great caution.

3.5 References

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Table 3.1. Summary statistics[†] for mass loss on ignition (LOI), Walkley-Black (WB), automated dry combustion (ADC), and humic matter (HM) colorimetry. n = number of soil samples.

Method	n	Standard		Minimum	Maximum	Range
		Mean	deviation			
g SOM kg ⁻¹						
LOI	84	23.2	5.6	14.5	38.8	24.3
g C kg ⁻¹						
WB	84	12.9	a 2.1	9.7	18.8	9.1
ADC	84	7.7	b 2.1	3.7	13.3	9.6
g HM kg ⁻¹						
HM	84	2.37	1.29	0.3	6.6	6.3

[†] Letters after means indicate statistical groupings as determined by the Tukey pairwise means comparison test with a 95% confidence interval.

Table 3.2. Correlations between automated dry combustion (ADC), Walkley-Black (WB), and mass loss on ignition (LOI) for different soils. n = number of soil samples. r = Pearson's correlation coefficient.

Correlation	Trial	n	r	p-value
ADC to WB	Overall	84	0.55	<0.0001
	Coastal Plain	12	0.23	0.4694
	Piedmont nine-tillage	36	0.42	0.0102
	Piedmont four-tillage	16	0.53	0.0350
	Mountain	20	0.77	<0.0001
ADC to LOI	Overall	84	0.63	<0.0001
	Coastal Plain	12	0.79	0.0025
	Piedmont nine-tillage	36	0.57	0.0003
	Piedmont four-tillage	16	0.91	<0.0001
	Mountain	20	0.84	<0.0001
WB to LOI	Overall	84	0.56	<0.0001
	Coastal Plain	12	0.29	0.2936
	Piedmont nine-tillage	36	0.39	0.0198
	Piedmont four-tillage	16	0.64	0.0077
	Mountain	20	0.57	0.0100

Table 3.3. Correlations between humic matter (HM), Walkley-Black (WB), mass loss on ignition (LOI), and automated dry combustion (ADC) as a function of sampling location. n = number of soil samples. r = Pearson's correlation coefficient.

Correlation	Trial	n	r	p-value
HM to LOI	Overall	84	0.03	0.7850
	Coastal Plain	12	-0.15	0.6377
	Piedmont nine-tillage	36	-0.03	0.8558
	Piedmont four-tillage	16	0.09	0.7541
	Mountain	20	0.40	0.0781
HM to ADC	Overall	84	0.55	<0.0001
	Coastal Plain	12	0.22	0.4845
	Piedmont nine-tillage	36	0.40	0.0145
	Piedmont four-tillage	16	0.39	0.1356
	Mountain	20	0.51	0.0218
HM to WB	Overall	84	0.22	0.045
	Coastal Plain	12	-0.19	0.5638
	Piedmont nine-tillage	36	0.25	0.1425
	Piedmont four-tillage	16	-0.16	0.5438
	Mountain	20	0.53	0.0167

Table 3.4. The average concentrations of soil organic matter (SOM), soil organic carbon (SOC) and humic matter (HM) found in soils for treatments in the four agronomic trials across North Carolina. Results are presented as separate analyses[†] for each trial and method, which included mass loss on ignition (LOI), automated dry combustion (ADC), Walkley-Black (WB), and HM colorimetry.

Trial	Treatment	LOI	ADC	WB	HM
		g SOM kg ⁻¹	— g C kg ⁻¹ —		g HM kg ⁻¹
Coastal Plain	No-till chemical	17.1	8.3	13.2 a	3.98
	Conv. Tillage chemical	17.1	9.0	10.7 b	4.69
	Conv. Tillage organic 1	17.8	8.5	13.3 a	3.95
	Conv. Tillage organic 2	20.5	9.0	11.9 ab	3.68
Piedmont nine-tillage	No-till chemical	21.8 bc	7.5 ab	11.9	3.08 a
	In-row subsoiling	29.2 a	9.5 a	14.0	2.75 ab
	Disk, spring	23.9 abc	5.3 b	14.3	1.38 bc
	Chisel, fall	23.5 abc	7.9 ab	12.4	1.93 abc
	Chisel, spring	25.7 ab	8.1 ab	13.7	1.90 abc
	Chisel, disk, fall	21.2 bc	7.1 ab	12.2	1.32 bc
	Chisel, disk, spring	23.1 abc	6.5 ab	12.1	1.73 abc
	Moldboard, fall	22.0 bc	5.3 b	10.9	0.73 c
	Moldboard, spring	17.5 c	5.5 b	11.6	1.10 c
Piedmont four-tillage	No-till chemical	20.0	7.1	11.6	2.38
	Alternate year tillage	20.7	7.1	13.6	1.85
	Disk, spring	20.0	6.7	12.7	1.65
	Disk, spring, twice	17.6	5.9	12.4	1.67
Mountain	No-till organic	34.7 a	12.3 a	16.4 a	3.35
	No-till chemical	28.1 ab	8.8 b	14.6 ab	2.60
	Conv. Tillage organic	29.9 ab	9.3 b	13.9 ab	1.95
	Conv. Tillage chemical	26.3 b	7.3 b	12.7 b	2.30
	Conv. Tillage fallow	28.2 ab	8.8 b	15.0 ab	3.88

[†] Statistical analyses were conducted using the Tukey pairwise means comparison test with a 95% confidence interval. Letters after means indicate statistical groupings, and sections without letters indicate a lack of statistical differences among the treatments.

Table 3.5 Statistical p-values[†] for comparisons of agronomic management treatment differences in soil organic matter, soil organic carbon, and humic matter (HM) based measurements using mass loss on ignition (LOI), automated dry combustion (ADC), Walkley-Black (WB), and HM colorimetry.

	LOI	ADC	WB	HM
Coastal Plain	0.5849	0.9059	0.0269	0.4180
Piedmont nine-tillage	0.0013	0.0058	0.2652	0.0003
Piedmont four-tillage	0.3943	0.2743	0.7356	0.2496
Mountain	0.0524	0.0014	0.0331	0.2026

[†] Analyses were conducted using the Tukey means comparison with a 95% confidence interval.

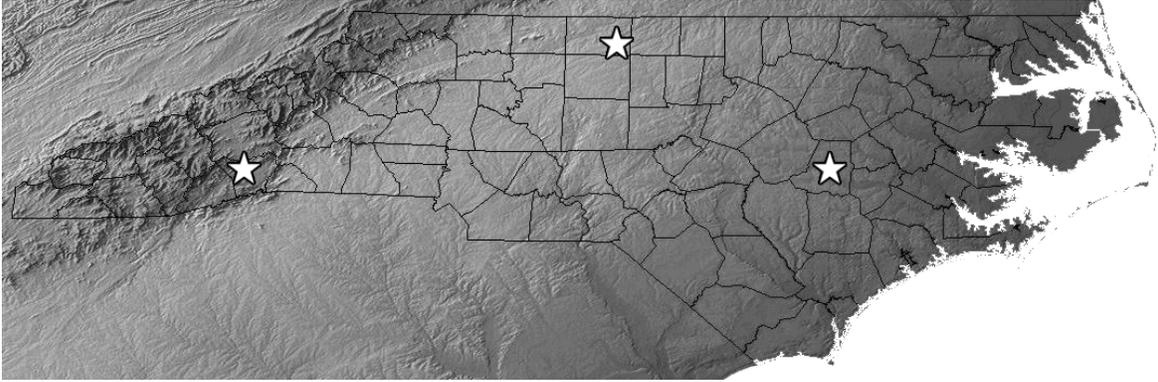


Figure 3.1. Map of agronomic trials located across North Carolina. Approximate locations of trials are indicated by stars. To the left is Mills River in the mountains, center is Reidsville in the piedmont, and right is Goldsboro in the coastal plain.

Chapter 4: Wheat Cover Crop Effects on Physical Properties of Piedmont Soil

Abstract

Conservation tillage is commonly recommended for improving physical resilience of soils, but because of differences in soil parent material and environmental conditions, the effects of conservation tillage may vary for soils in different areas. Cover crops are often proposed as a way to enhance the effects of conservation tillage, but this has not yet been proven for many soils. We added wheat (*Triticum aestivum*) as a cover crop to a corn (*Zea mays*) and soybean (*Glycine max*) rotation tillage trial after 31 years to learn how soil physical properties would change when cover crops are managed with conventional or conservation tillage. The trial included no-till management as well as tillage with chisel, disc, and moldboard plows. We measured cover crop biomass, aggregate stability, bulk density, penetration resistance, water content, water retention, and hydraulic conductivity of the soil during the first two years after adding the cover crop. Biomass from the cover crop was similar for all treatments. Megaaggregate stability (2.00-4.75 mm) and macroaggregate stability (0.25-2.00 mm) were greater in no-till soils compared to tilled soils with and without cover crops. Bulk density was less for no-till compared to only moldboard plowing in the second year. Penetration resistance varied in effect at different depths. There was no differentiation of tillage or cover cropping for any of the hydraulic properties. Results from the trial were mostly consistent with previous research showing that aggregate stability increased under conservation management but other physical properties were not as responsive even after adding a cover crop.

4.1 Introduction

Reducing tillage and adding cover crops are among the most common practices recommended for soil conservation and improving soil health in agronomic systems. These practices can potentially reduce soil erosion (Logan et al., 1991), reduce soil compaction (Tolon-Becerra et al., 2011), and improve hydraulic soil conditions for crop growth (Strudley et al., 2008). Cover crops may additionally improve nutrient cycling (Blanco-Canqui et al., 2015), soil carbon storage (Liu et al., 2005), and soil biological diversity (Chavarría et al., 2016). Despite these observed benefits, reduced tillage and cover cropping are not included in many soil management plans. Data from a survey conducted by the United States Department of Agriculture (USDA) in 2012 revealed that only 3% of land managed by the respondents to the survey was cover cropped (USDA, 2012). Low adoption may be due to multiple factors: low awareness of the potential benefits of conservation practices, lack of resources to implement these practices, or lack of realized benefits after implementing a cover crop. Cover crops are used to increase soil stability (Liu et al., 2005), increase soil carbon (Ruis and Blanco-Canqui, 2017; Beehler et al., 2017), and better manage soil nutrients (Utomo et al., 1990; Liebman et al., 2012; Chavarría et al., 2016), but these effects may take multiple years to be observed (Benoit et al., 1962) or in some cases the cover crop may exacerbate other management challenges (Unger and Vigil, 1998; Basche et al., 2014). Soil conservation and soil health management are not just focused on short-term changes in soil conditions and are instead focused on long-term sustainability of soil resources. If cover crops are not having the same effects on soils in different situations it may be due to variability in environments that may reduce or eliminate their expected long-term benefits (Grandy et al., 2006; Blanco-Canqui et al., 2015). Factors such as precipitation, temperature, soil moisture, sunlight, pest life cycles, and other phenomena that affect the success of agronomic systems should be considered before fully adopting any change in soil management.

In the piedmont region of North Carolina, agronomic systems are situated in a humid subtropical climate with moderately weathered Ultisols (Buol et al., 2011). The climate and diverse landscape of the piedmont is favorable for various crops and land management practices, which provides opportunities for different practices to be implemented in agronomic systems in order to adapt to local environmental conditions. Agronomic management without tillage (no-till) is a major aspect of soil conservation and soil health that may help improve soil resiliency in the piedmont. When land is managed with no-till, there is less disturbance and therefore less risk of erosion and soil degradation (Langdale et al., 1979; Logan et al., 1991; McGregor et al., 1992), but tillage continues to be an important tool for agronomic management. Tillage is used to reduce weed pressure (Kurstjens, 2007), incorporate nutrient and pesticide amendments (Verbree et al., 2010), and also to facilitate mineralization of previously deposited organic residues (Jones et al., 1998; Radicetti et al., 2017). Without tillage as a tool for crop and soil management, alternative strategies have to be developed to assist with land conservation, and the long-term effect of those strategies is not known for different areas of the piedmont.

To overcome the limited effect that conservation tillage has on soil physical properties, cover crops can be added to a soil management plan in order to increase plant biomass and the probability that organic residues will aggregate soil minerals to increase resiliency against environmental degradation (Tisdall and Oades, 1982; Lu et al., 1998; Liebbig et al., 2004). Cover crops are planted to protect soil during times when a cash crop is not covering the soil. Managing soil with no-till and cover crops is considered ideal for soil health and conservation, but tillage is still a desired control option for many people, especially during years when it is difficult to coordinate termination of the cover crop with planting the next crop.

Previous research in the piedmont region had evaluated soil physical properties as they are affected by different types of tillage. A study comparing no-till management and conventional tillage

using chisel plows, shanks, and moldboard plows, showed that residue remaining after tillage and planting had only 1% coverage for moldboard plowed soils compared to 75% for no-till (Cassel et al., 1995). The study also found that untrafficked inter-rows between crop rows had greater bulk density under no-till than when chisel plowed, showing a tradeoff between residue cover and compaction for untilled soils. A different study at the same location also observed greater bulk density in no-till soils compared to tilled soils, but also measured aggregate stability, which was greater in no-till soils compared to tilled soils (Freese et al., 1993). Years later, it was observed at the same location that the more intensively tilled areas were losing more soil than no-till areas (Meijer et al., 2013), which may have been because no-till soils had more stable aggregates. After many years of consistent management in this piedmont soil, there was direct evidence that no-till soil was better conserved and protected from erosion than tilled soils.

The overall goal of our experiment was to assess changes in physical soil properties due to the introduction of a winter wheat (*Triticum aestivum*) cover crop in a corn (*Zea mays*) and soybean (*Glycine max*) rotation in a long-term (>30 years) tillage study. Specific objectives were to measure potential changes to bulk density, cone index, aggregation, water content and retention, and saturated hydraulic conductivity. Monitoring these physical characteristics of the soil provides context to assess the effects of commonly recommended soil health and conservation practices in the NC piedmont.

4.2 Methods and materials

4.2.1 Experimental Design

The agronomic trial began in 1984 at the Upper Piedmont Research Station (36°23'2.1372'' N, 79°42'6.8436''W) to test the effects of different tillage practices on crop yields and physical soil properties in soil mapped as a Toast coarse sandy loam (Fine, kaolinitic, mesic Typic Kanhapludults). The trial initially had continuous corn but was changed in 1990 to a corn and soybean rotation with

crops planted in 96.5 cm rows (Freese et al., 1993; Cassel et al., 1995). Treatments at the start of the trial included no-till, planting with an in-row shank, and tillage with chisel, disk, or moldboard plows used in either fall or spring. No differences were observed between fall and spring tillage during the history of the trial and therefore the treatments were modified in fall 2015 to include a winter wheat cover crop as a factor instead of spring tillage. The treatments were converted to no-till (NNN), no-till with a cover crop (NNV), disking with a cover crop (NDV), chisel plowing without (CNN) and with a cover crop (CNV), chisel plowing and disking without (CDN) and with a cover crop (CDV), and moldboard plowing and disking without (MDN) and with a cover crop (MDV). The nine treatments were replicated four times in a randomized complete block design with a total of 36 plots each sized 5.8 x 15.2 m. All equipment traffic (tractors, planters, combines, etc.) across the plots was restricted to the same rows on every pass.

4.2.2 Agronomic Management

Winter wheat seed was acquired from animal feed stock with no specific variety considered. After harvesting the cash crop in fall 2015, the first winter wheat for the trial was planted using a seed-drill calibrated to release 45 kg of seed per acre. Seed was planted in all plots on the same day, and biomass from the cover crop was collected in spring after four to five months of growth. A quadrat with area dimensions of 0.91 x 0.91 m and 16 internal squares sized 0.23 x 0.23 m (25% of the length of the crop inter-row) was used to select areas from which biomass would be removed. The quadrat was randomly thrown into the plot area and wheat biomass within four randomly-selected squares of the quadrat was removed and stored in paper bags. This process was replicated twice to generate two biomass subsamples per plot. Biomass was dried in an oven at 60°C until a constant dry mass was achieved. The masses of the two dry mass subsamples were averaged and the total dry mass of the biomass was extrapolated to represent cover crop biomass for the area of

the entire plot. After collecting biomass, the cover crop was terminated using paraquat (1,1'-Dimethyl-4,4'-bipyridinium dichloride) herbicide.

Tillage was conducted each year approximately four to five days after terminating the cover crop. The chisel and disc plows were attached to a tractor and configured to plow two crop rows during each pass. The chisel was set to a depth of 25 cm and the disc was set to 12 cm. A moldboard plow was configured to plow one crop row with each pass and inverted soils from 25-30 cm deep. Tillage was always conducted on the same day for all plots.

4.2.3 Soil sampling and measurements

Soybean was planted May 31, 2016 and harvested November 18, 2016. Corn was planted May 18, 2017 and harvested November 16, 2017. Corn and soybean varieties were glyphosate tolerant and both glyphosate and atrazine were used to control weeds before and after planting. Planting density was approximately 75,000 seeds ha⁻¹ for corn and 343,894 seeds ha⁻¹ for soybean.

Bulk density and water content were measured in 2016 and 2017 for the 0-7.6 cm and 7.6-15.2 cm depths using a Uhland core sampler (7.6-cm diameter rings) in the crop row. Soil from the cores was weighed to obtain a wet mass, dried at 105°C until reaching a constant dry mass, and then re-weighed. Soil cone index (penetration resistance) was measured with a penetrometer (Field Scout SC-900, Spectrum, Aurora, IL) in the center untrafficked and trafficked rows of each plot in fall 2017 when soils seemed to be at or near field capacity moisture content (subjective observations based on weather and field conditions). Data were recorded digitally as the average pressure applied in 2.54 cm increments to a depth of 40 cm.

Soil samples for aggregate stability were collected from untrafficked and trafficked rows by collecting soil to a depth of 7.6 cm. The soil was shoveled from below and carefully broken apart to fit inside storage bags that were transferred to a lab to air-dry for several days. After the soil was dried, sieves were used to remove objects so that only air-dried aggregates between 1-4.75 mm in

diameter were used to measure aggregate stability. Aggregate stability was evaluated using a modified protocol from Beare and Bruce (1993), which includes the Yoder wet-sieving apparatus (Yoder, 1936) used for wet sieving. Sieves with mesh sizes 2 mm and 0.25 mm were stacked inside a large cylinder with enough water to submerge the 2-mm mesh on top. Approximately 30 g of air-dry soil, with rocks and other non-soil particles removed, was weighed and then placed on the 2-mm mesh above the cylinder reservoir. The sieves with soil were submerged for 10 min to soak and sieved in the water for another 10 min. The water reservoir beneath the sieves was drained through a 0.05 mm mesh to capture aggregates passed through the 0.25-mm mesh. Soil remaining on the sieves was washed into individual glass beakers for each mesh size and oven dried at 105°C until reaching a constant dry mass. Aggregate stability for the microaggregate (0.05-0.25 mm), macroaggregate (0.25-2.00 mm), and megaaggregate (2.00-4.75 mm) size fractions was calculated based on the mass of the soil recovered in each fraction divided by the mass of the original soil sample (Beare and Russell Bruce, 1993).

Additional soil samples were collected to measure saturated hydraulic conductivity (K_{sat}) and water retention for the 0-7.6 cm soil depth in the center untrafficked and trafficked inter-rows of each plot. These samples were collected using the same methodology used for bulk density and water content sampling but instead of being dried they were kept intact inside their rings, sealed with plastic, and preserved in cold storage at 4°C until analysis. Saturated hydraulic conductivity was measured using the constant pressure head method (Klute, 1986). After saturating the intact soil cores, they were sealed inside Buchner funnels connected to an air pressure device. Pressure was applied in increments of: 3.5, 50, 100, 333, and 500 cm H₂O and water outflow drained into graduated cylinders. After measuring outflow at 500 cm, soil cores were dried in an oven at 105°C until reaching constant dry mass. Water content at each pressure was used to create a water retention curve for the soils.

4.2.4 Statistical analyses

Statistical analyses were conducted using SAS 9.4 (SAS Institute, Cary, NC). Each parameter was compared among all treatments using the Tukey means comparison with a 95% confidence level. The significance of individual treatment, row, depth, and coverage factors was analyzed using the proc GLM procedure with a 95% confidence level. For measurements that were conducted in multiple years, the analysis considered each year as separate. For parameters with soil in untrafficked and trafficked row positions, the analyses were conducted separately for each row position. For parameters with multiple depths, the analyses were conducted separately for each depth. Soil hardness was analyzed in depth increments of 2.54 cm, and water retention analysis were conducted separately for each pressure.

4.3 Results and Discussion

4.3.1 Cover Crop biomass

A summary of significant factors for each measurement is included in table 4.1. Five of the nine treatments included a cover crop, and only treatments with a cover crop were included in the analysis of cover crop biomass. In 2016, the first spring after adding the cover crop to the rotation, biomass among the treatments was similar (Table 4.2). None of the treatments produced a statistically different amount of biomass than others, and this was mostly because of variability within treatments. The largest amounts of biomass for CDV and NDV were ten times greater than the smallest amounts of biomass collected (data not shown), and other treatments also had variability with orders of magnitude difference between minimum and maximum biomass collected. The effect was similar in 2017 with no treatment having more cover crop biomass than others. Biomass was also substantially less in 2017 for all treatments compared to 2016 ($p < 0.0001$), which was likely due to late planting and an abnormally cold winter. During the first two years of the cover

crop, none of the tillage treatments were more conducive to producing cover crop biomass than others and similar amounts of biomass were contributed to soils for each of the treatments.

4.3.2 Aggregate Stability

Aggregate size classes were separated into microaggregates (0.05-0.25 mm), macroaggregates (0.25-2.00 mm), and megaaggregates (2.00-4.75 mm) based on the mesh sizes used to evaluate aggregate stability. All aggregate stability measurements were conducted on soils collected from 0-7.6 cm depth. In untrafficked rows, megaaggregation was greater for both no-till treatments (with and without cover crops) compared to all other treatments (Table 4.3). The fraction of aggregates between 2.00-4.75 mm in no-till soils averaged 0.23-0.30 of the mass, which was more than twice as much as tilled soils. Soils with the least megaaggregate stability in untrafficked rows were MDN and MDV, which were moldboard plow treatments. This result indicates that soil management without tillage is more likely to produce large stable aggregates compared to tilled soils. Differences in the proportion of megaaggregates was less pronounced in the trafficked rows (Table 4.1). The NNN soils had a greater proportion of stable megaaggregates in trafficked rows than all other treatments except NNV (Table 4.3). In the trafficked rows, however, megaaggregates were 0.17 of the mass fraction compared to the average of 0.30 for untrafficked rows of the NNN soils. The controlled traffic between crop rows had decreased soil aggregation for this size fraction. Differences in aggregate stability due to the cover crop were not observed in the data (Table 4.1). There was no clear separation between the proportion of megaaggregates in cover cropped soils and winter fallow soils for any tillage treatment.

Macroaggregates are the intermediate soil aggregate size fraction, and the data show that macroaggregation in untrafficked rows is different among tillage treatments (Table 4.3). No-till soils had the largest amount of macroaggregate stability, but unlike the megaaggregate fraction, only NNN was different from tilled soils because NNV had similar macroaggregate stability to CNV, NDV,

and CDV. Additionally, moldboard plowing with and without cover had statistically less macroaggregate stability than NNN, NNV, NDV, CNV, and CDV, which was an additional degree of separation compared to the megaaggregate fraction. In trafficked rows, macroaggregate stability was still greater in NNN compared to other treatments except NNV and NDV (Table 4.3).

The microaggregate analysis showed that NNN had the smallest proportion of stable microaggregates in untrafficked rows compared to the same position for tilled treatments (Table 4.3). NNV was similar to NNN and CNV but had less stable microaggregates than all other tillage treatments. As expected, microaggregate stability was the inverse of megaaggregate stability, and tilled soils had greater microaggregate stability than no-till. Microaggregate stability in the trafficked rows was greater in MDN than NDV, NNV, and NNN, which was expected because NNN and NDV had more stable megaaggregates than MDN. Microaggregates in the trafficked rows of NNN and NNV comprised approximately twice the proportion of mass of soil compared to microaggregates in untrafficked rows of the same treatments. It is possible that equipment traffic was decreasing the likelihood of larger aggregates forming in these positions (Table 4.1). The effect of microaggregation is less pronounced than the resilience provided by larger aggregates as evidenced by the previous erosion study at the site (Meijer et al., 2013). The effect of tillage on soil resilience to erosion was previously observed at this location using LIDAR imagery to detect changes in elevation (Meijer et al., 2013), and the differences in macro and megaaggregate stability provides evidence for why no-till soils were less eroded than tilled soils. Results from the previous study conducted in 1992 (16 years ago) on the same soils also showed that more stable macroaggregates formed in no-till soils compared to tilled soils (Freese et al., 1993), and this occurred even without a cover crop. Additional biomass from the cover crop did not have a measurable effect on soil aggregation after two growing seasons (Table 4.1). This result is consistent with observations in previous research showing that it

takes multiple years for a cover crop to have a measurable increase in aggregate stability in tilled soils (Benoit et al., 1962), but the exact number of years may vary between locations.

4.3.3 Bulk density and hardness

A potential benefit of soil aggregation is the formation of inter-aggregate space that can increase porosity and decrease bulk density. Bulk density was measured from the 0-7.6 cm depth and 7.6-15.2 cm depth in untrafficked rows soon after planting in order to check soil bulk density in the planting zone after cover crop residue had been incorporated. In the first spring after terminating the cover crop, bulk density from 0-7.6 cm and from 7.6-15.2 cm was not different among treatments (Table 4.4) and was within ranges that are conducive to plant growth (Gerard et al., 1982; Vepraskas and Waggoner, 1989). In the second year after adding the cover crop to the rotation, bulk density in the 7.6-15.2 cm depth remained similar among treatments, but bulk density at the surface was greater in MDN compared to CDV, CNN, NNN, and NNV. This result is mostly consistent with macroaggregate stability in the untrafficked rows (Table 4.3), which was also measured in the second year. There was greater macroaggregate stability in NNN and NNV compared to MDN, and bulk density was less in NNN and NNV compared to MDN, which supports the idea that soils with more macroaggregates are less compact. Differences in surface bulk density for other treatments CDV and CNN did not follow any obvious trends, and there was no measured effect on bulk density due to the presence of the cover crop (Table 4.1).

Tillage at the start of the planting season loosens soil and incorporates cover crop residue, but after multiple passes of traffic from equipment, the loosened soil can reconsolidate resulting in high penetration resistance. Soil hardness was measured in trafficked and untrafficked rows to see how the interaction between cover cropping and tillage affected cone index after tillage and passes with heavy equipment. After measuring soil hardness to a depth of 40 cm, the only differences in pressure were that NNN required statistically more pressure at the depths 5 and 23-30 cm below

the surface of untrafficked rows compared to CNN (Figure 4.1). In trafficked rows, cone index increased closer to the surface than in untrafficked rows, but there were no statistical differences in cone index among treatments. Controlled traffic in these rows had consistently compacted soil over many years and likely removed differences in hardness due to tillage. A cone index greater than 3 Mpa is potentially restrictive to root growth (Gerard et al., 1982; Vepraskas and Wagger, 1989), and in untrafficked rows the average cone index was not greater than 3 MPa until reaching a depth of 25 cm (Figure 4.1a). For trafficked rows, however, restrictive root growth was observed at 15 cm (Figure 4.1b) for no-till soils. In previous research, hardness measured by cone index was statistically differentiated by the interaction between tillage and row position, with the most intensively tilled plots having a greater cone index at the surface of trafficked rows, but no-till plots having a greater cone index in untrafficked rows (Cassel et al., 1995). No-till soil in the previous study also had greater cone indices in untrafficked rows when compared to chisel and moldboard plowing. The data show that surface soil in no-till has consistently been tougher to penetrate, and that tillage decreases soil hardness.

4.3.4 Water content and retention

Water content was measured in untrafficked rows days after planting to measure potential water availability during the early planting season. Water content was measured from 0-7.6 cm and 7.6-15.2 cm increments for surface and subsurface comparisons, respectively. In the first year after adding the cover crop to the trial, water content was similar for all treatments in both the surface and subsurface increments (Table 4.4). Similar results were found the second year after adding the cover crop; none of the treatments had statistically different water content compared to others. Observations in the past had shown greater water content in no-till soil compared to chisel and moldboard plowed soils 25 days after planting (Cassel et al., 1995), but neither the cover crop nor

tillage intensity affected water content based on measurements taken around the time of planting for this study (Table 4.1).

Water content represents the amount of water in soil at a point in time, but water retention is a measure of the ability for soil to hold and release water at any time. Tillage is intended to loosen soils so that pore size is redistributed to favor more water retention, but as soils compact and settle after tillage, water retention by the soil may become unfavorable for management. Water retention in the top 7.6 cm was measured in trafficked and untrafficked rows to compare how tillage, traffic, and cover cropping may change water supply from the soil. In the untrafficked rows there was no statistical difference in water retention for pressures ranging from 0-500 cm H₂O (Figure 4.2a). The lack of differentiation is largely because of variation observed within tillage treatments that had soils with both a high capacity to retain water and soils with low capacity to retain water. The trend was similar for trafficked rows, which also did not differ in water retention for any pressure from 0-500 cm H₂O (Figure 4.2b). When soil water retention from this location was previously measured, no-till soils had less retention than tilled soils (Cassel et al., 1995). In this study, water retention was variable, and not consistent with previous observations. Soil water retention did not continue to be differentiated by tillage after many years of consistent management. Adding a cover crop to the trial also had not improved water retention relative to tilled soils.

4.3.5 Saturated hydraulic conductivity

Saturated flow through soil cores was measured to determine if tillage and cover cropping differently affected water movement in the surface 7.6 cm of untrafficked and trafficked rows. No statistical difference in K_{sat} was observed for untrafficked rows among treatments (Table 4.5). There was a large amount of variability in K_{sat} within and among all treatments. In trafficked rows, K_{sat} was also not different among treatments and had large variability (Table 4.5). Roots and other biomass from the cover crop did not increase K_{sat} compared to bare soil (Table 4.1). It may be difficult to

detect or measure differences in K_{sat} because of variability in field conditions that masks the effect of tillage and cover cropping.

4.4 Conclusion

After 33 years of the soils in the North Carolina piedmont agronomic trial being consistently managed with different tillage intensities, a winter wheat cover crop was added to evaluate interactions between cover crop biomass and common tillage practices. Past observations of the trial showed that no-till facilitated the formation of stable aggregates and reduced soil erosion compared to tilled soils (chisel plow, disc, and moldboard plow). These past observations did not include any noticeable improvements in soil bulk density or hydraulic properties. Conservation tillage is among the most commonly recommended practice for improving physical soil conditions for agronomic management, but few measurable differences in physical soil conditions were observed between no-till soil and tilled soils throughout this 33-year trial (Freese et al., 1993; Cassel et al., 1995; Meijer et al., 2013; Roper et al., 2017b). The winter-wheat cover crop added to the trial in fall 2015 represented another recommended soil conservation practice. After two years of maintaining tillage with and without a cover crop, aggregate stability was still the most indicative of differences in soil structure due to management, but no other measured physical characteristics of the soil seemed to respond to the cover crop or tillage intensity in the two-year time frame. The lack of differentiation among tillage and cover crop treatments for other physical properties was due to large variability, which means that these soil practices are unlikely to produce consistent differences in other soil physical properties in this piedmont soil. Although previous research showed that conservation tillage had improved soil resiliency to erosion and increased aggregate stability, the addition of the cover crop had not provided additional improvements in other measured soil physical properties after two years.

4.5 Acknowledgements

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Table 4.1 Table of p-values significance for different parameters measured in the study.

Parameter	Treatment	Position	Depth	Cover
Cover crop biomass	NS	-	-	-
Microaggregate stability	**	**	-	NS
Macroaggregate stability	**	*	-	NS
Megaaggregate stability	**	*	-	NS
Bulk density				
2016	NS	-	**	NS
2017	**	-	**	NS
Water content				
2016	NS	-	NS	NS
2017	**	-	NS	NS
K _{sat}	NS	NS	-	NS

* indicates significance with $p < 0.05$

** indicates significance with $p < 0.0001$

NS indicates no significance

- indicates that this factor was not considered for the measurement

Table 4.2. Biomass collected from the winter wheat cover crop in spring. Treatments included in the trial are no-till with a cover crop (NNV), disking with a cover crop (NDV), chisel plowing with a cover crop (CNV), chisel plowing and disking with a cover crop (CDV), and moldboard plowing and disking with a cover crop (MDV). Numbers in parenthesis are the SD of the means.

Treatment	Biomass	
	2016	2017
	————— kg ha ⁻¹ —————	
NNV	7854 (4382)	3608 (1331)
NDV	7030 (5116)	3055 (1291)
CNV	6744 (3376)	3627 (1507)
CDV	6559 (4503)	2952 (1693)
MDV	8308 (3473)	2952 (825)
Year	7297 (4057)	3238 (1327)

Table 4.3. Aggregate stability of soils collected from the untrafficked inter-rows and trafficked inter-rows of soils in the Upper Piedmont agronomic trial. Aggregates are separated into micro (0.05-0.25), macro (0.25-2.00 mm) and mega (2.00-2.75 mm) size fractions and numbers are the average fractional proportion of the mass of soil captured in each size fraction. Treatments included in the trial are no-till (NNN), no-till with a cover crop (NNV), disking with a cover crop (NDV), chisel plowing without (CNN) and with a cover crop (CNV), chisel plowing and disking without (CDN) and with a cover crop (CDV), and moldboard plowing and disking without (MDN) and with a cover crop (MDV). Letters indicate statistical groupings based on a Tukey means comparison test at the 95% confidence level. Columns without letters indicate no statistical difference between treatments.

Treatment	Untrafficked inter-row						Trafficked inter-row					
	Micro-aggregate stability		Macro-aggregate Stability		Mega-aggregate stability		Micro-aggregate stability		Macro-aggregate Stability		Mega-aggregate stability	
Fraction of stable aggregates												
NNN	0.23	c	0.40	a	0.30	a	0.41	c	0.36	a	0.17	a
NNV	0.29	bc	0.38	ab	0.23	a	0.52	bc	0.29	ab	0.11	ab
NDV	0.51	a	0.29	bc	0.09	b	0.56	b	0.29	ab	0.06	bc
CNN	0.56	a	0.25	cd	0.06	b	0.61	ab	0.24	bc	0.05	bc
CNV	0.47	ab	0.29	bc	0.11	b	0.60	ab	0.25	b	0.07	bc
CDN	0.56	a	0.22	cd	0.06	b	0.64	ab	0.21	bc	0.06	bc
CDV	0.50	a	0.28	bc	0.10	b	0.62	ab	0.23	bc	0.04	bc
MDN	0.66	a	0.16	d	0.03	b	0.70	a	0.16	c	0.03	c
MDV	0.60	a	0.17	d	0.03	b	0.64	ab	0.20	bc	0.03	bc

Table 4.4. Biomass collected from the winter-wheat cover crop in spring along with bulk density and water content in two depth increments for soils in the Upper Piedmont agronomic trial. Treatments included in the trial are no-till (NNN), no-till with a cover crop (NNV), disking with a cover crop (NDV), chisel plowing without (CNN) and with a cover crop (CNV), chisel plowing and disking without (CDN) and with a cover crop (CDV), and moldboard plowing and disking without (MDN) and with a cover crop (MDV). Letters indicate statistical groupings based on a Tukey means comparison test at the 95% confidence level. Columns without letters indicate no statistical difference between treatments.

Treatment	0-7.6 cm depth				7.6-15.2 cm depth				
	Bulk density		Water content		Bulk density		Water content		
	2016	2017	2016	2017	2016	2017	2016	2017	
	g cm ⁻³		cm ³ cm ⁻³		g cm ⁻³		cm ³ cm ⁻³		
NNN	1.39	1.33	b	25.0	0.29	1.52	1.63	0.20	0.24
NNV	1.33	1.32	b	21.5	0.33	1.53	1.59	0.23	0.28
NDV	1.36	1.42	ab	23.2	0.33	1.48	1.55	0.24	0.33
CNN	1.37	1.24	b	20.3	0.30	1.49	1.47	0.26	0.31
CNV	1.38	1.35	ab	21.3	0.30	1.55	1.51	0.24	0.31
CDN	1.41	1.37	ab	21.0	0.29	1.52	1.48	0.28	0.30
CDV	1.38	1.32	b	23.5	0.31	1.54	1.51	0.27	0.30
MDN	1.47	1.59	a	20.3	0.28	1.62	1.62	0.26	0.27
MDV	1.45	1.47	ab	24.0	0.29	1.63	1.55	0.32	0.32

Table 4.5: Saturated hydraulic conductivity measured in cm day^{-1} for the untrafficked and trafficked rows of soils in the Upper Piedmont agronomic trial. Treatments included in the trial are no-till (NNN), no-till with a cover crop (NNV), disking with a cover crop (NDV), chisel plowing without (CNN) and with a cover crop (CNV), chisel plowing and disking without (CDN) and with a cover crop (CDV), and moldboard plowing and disking without (MDN) and with a cover crop (MDV). Numbers in parenthesis are the standard deviations of the means.

Treatment	Saturated hydraulic conductivity	
	Untrafficked	Trafficked
	cm day ⁻¹	
NNN	3.5 (3.7)	1.2 (0.5)
NNV	31.2 (36.2)	0.8 (1.3)
NDV	7.9 (8.5)	4.6 (4.1)
CNN	46.1 (27.4)	3.5 (4.1)
CNV	4.4 (5.5)	1.1 (1.1)
CDN	17.2 (33.5)	1.5 (2.4)
CDV	7.7 (5.6)	1.6 (2.8)
MDN	13.2 (21.1)	0.04 (0.03)
MDV	8.3 (4.9)	1.3 (1.9)

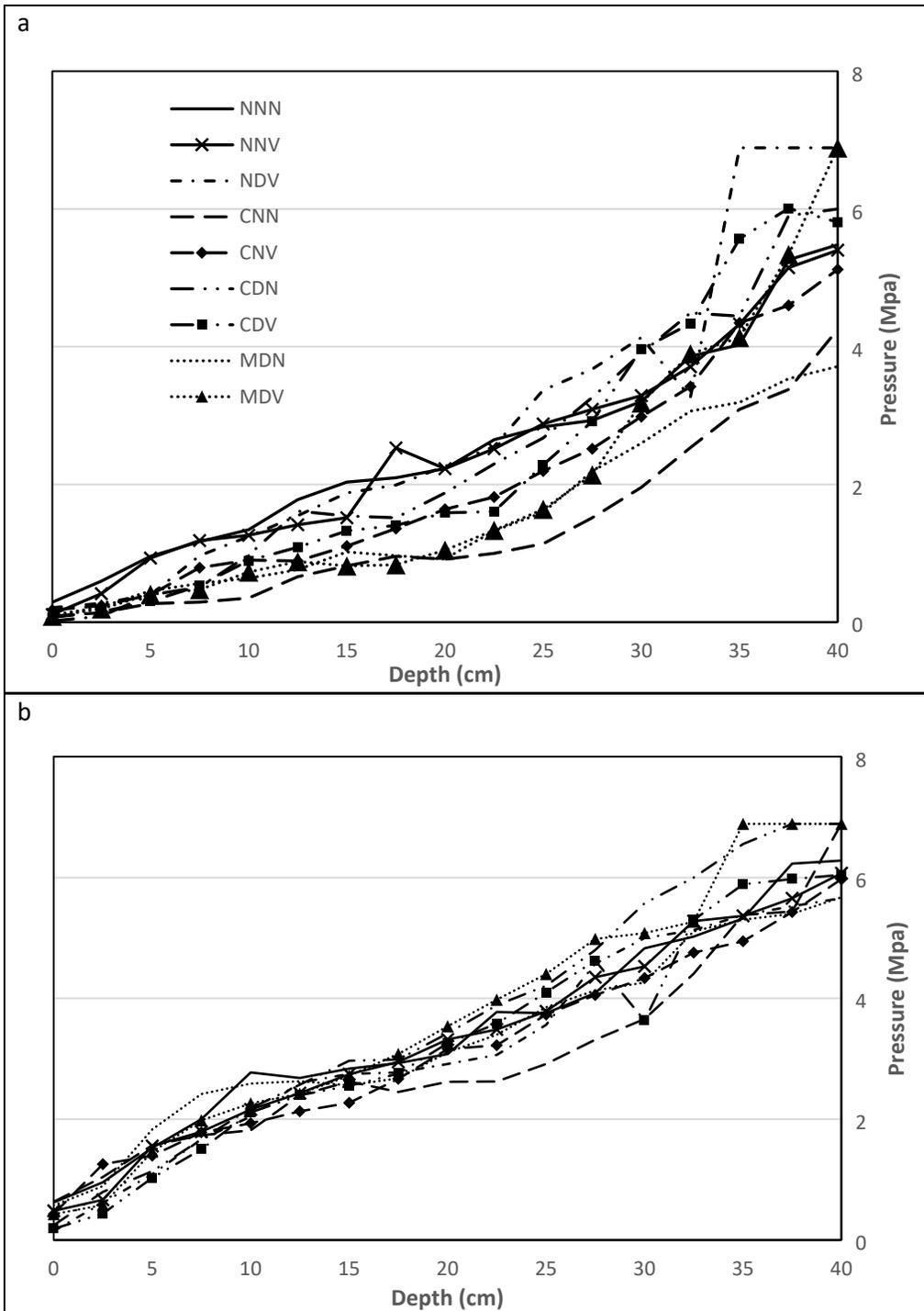


Figure 4.1. Soil hardness as measured by a cone penetrometer to 40-cm depth. Treatments included in the trial are no-till (NNN), no-till with a cover crop (NNV), disking with a cover crop (NDV), chisel plowing without (CNN) and with a cover crop (CNV), chisel plowing and disking without (CDN) and with a cover crop (CDV), and moldboard plowing and disking without (MDN) and with a cover crop (MDV). The graphs are for the (a) untrafficked inter-row position and (b) trafficked inter-row position.

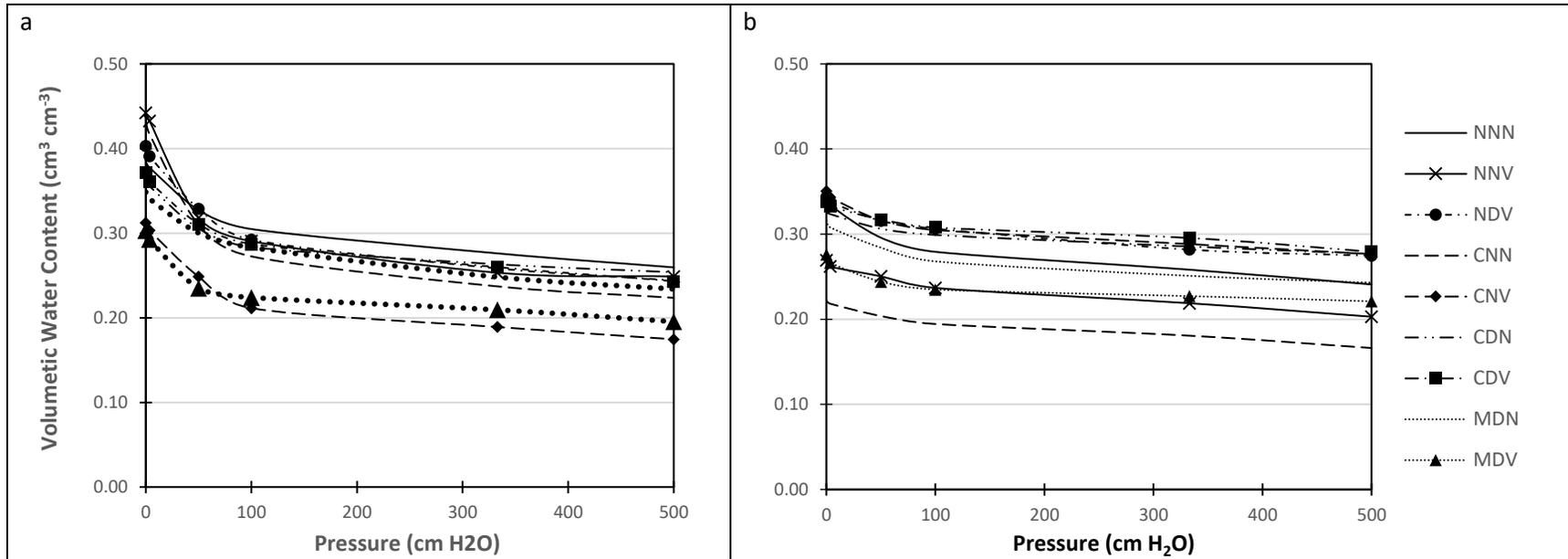


Figure 4.2. Water retention curves for soils in the untrafficked and trafficked inter-rows of soils included in the Upper Piedmont agronomic trial. Retention is reported as the volumetric water content at different pressures. Treatments included in the trial are no-till (NNN), no-till with a cover crop (NNV), disking with a cover crop (NDV), chisel plowing without (CNN) and with a cover crop (CNV), chisel plowing and disking without (CDN) and with a cover crop (CDV), and moldboard plowing and disking without (MDN) and with a cover crop (MDV). Graphs are for the (a) untrafficked inter-row position and (b) trafficked inter-row position.