8-2017

Effect of Different Cover Crop Species on Crop Production and Soil Health

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To the Graduate Council:

I am submitting herewith a thesis written by Mingwei Chu entitled "Effect of Different Cover Crop Species on Crop Production and Soil Health." I have examined the final electronic copy of this thesis for form and content and recommend that it be accepted in partial fulfillment of the requirements for the degree of Master of Science, with a major in Environmental and Soil Sciences.

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Effect of Different Cover Crop Species on Crop Production and Soil Health

A Thesis Presented for the

Master of Science

Degree

The University of Tennessee, Knoxville

Mingwei Chu

August 2017

ACKNOWLEDGEMENTS

First, I would like to thank my advisor, Dr. Forbes Walker for his support and guidance throughout my graduate study. His constant encouragement and valuable suggestions are highly appreciated.

I would also like to express my gratitude to my co-advisor, Dr. Sindhu Jagadamma for her unconditional support and patience during my laboratory research and thesis writing stages. Without her significant time investment and constant follow up, this study would not have been possible.

Thanks to Dr. Neal Eash for agreeing to serve on my committee member and for providing valuable suggestions and advice.

I would also like to thank my labmates Jaime Call, Surendra Singh, Shikha Singh, Xiuwen Li, Sarah Ottinger, Samantha Lee and Yan Sheng for their support and friendship. Thanks also to Melanie Stewart and Galina Melnichenko for the analytical support. I also wish to acknowledge Dr. Michael Buschermohle in providing valuable information about the field experiments.

Special thanks to my friends, Sarah Ouyang, Yang Yue, Locus Liang and Daihan Wang for their encouragement and support especially during the hard times.

Most importantly, I wish to express my indebtedness to my parents for their unconditional love, support and encouragement for making my graduate study a success.

ABSTRACT

Integration of cover crops (CCs) can provide several benefits to row crop production systems. Comprehensive studies to understand the effectiveness of a mixture of CCs versus single or double species CCs are limited. In this study, we evaluated the effect of single and double species CCs, and the soil health mix (SHM, a combination of five species recommended by the United States Department of Agriculture) on soil quality attributes and crop production in western Tennessee. The results showed an increase in soybean yield after 3-years of SHM treatment, which corresponded with significantly higher soil moisture content and soil inorganic nitrogen content compared to less diverse CC treatments and no-cover control. Overall the multi-species SHM showed potential for enhancing soil quality and crop yield.

The Haney's soil health test is a new approach to quantify the soil health status with heavy emphasis on soil biological properties. It introduced a new extractant for determining soil available nutrients, H3A; a new method of soil respiration measurement using Solvita gel system; and two new soil bioavailability parameters: water extractable organic carbon (WEOC) and water extractable organic nitrogen (WEON). The final Haney soil health score is calculated from the Solvita respiration, WEOC and WEON. In this study, components in Haney's soil health test were evaluated to test their effectiveness in Tennessee soils. The H3A extractant showed significant but weak correlation with the traditional extractants such as Mehlich-1 and Mehlich-3. The Solvita test did not provide a reliable estimation of potential mineralizable nitrogen, however, it correlated with many soil properties including soil carbon and nitrogen pools as well as the WEOC and WEON. Although the soil health score showed some extent of sensitivity to long-term cover crop treatments, it did not capture the variation in soil health status after 4 years of cover cropping with different species of cover crops. This study is a first step towards simultaneous suitability evaluation of a suite of CCs for

improving the sustainability of the agricultural belt of Tennessee. More similar studies are needed to help farmers make informed decisions of CC species selection for their cropping systems.

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CHAPTER I

INTRODUCTION AND GENERAL INFORMATION

Literature review

Conservation agriculture

The fast growing global population exerts enormous pressure on modern agriculture. It has been estimated that a 100 to 110 percent increase in global food demand will occur by 2050 compared to 2005 (Tilman et al., 2011) and to fulfill this huge demand, modern agriculture relies on multiple and practices. Modern plant breeding provides improved crop varieties to achieve higher food production. These varieties are also better adapted to the changing environments (Brummer et al., 2011). Agricultural mechanisation introduces machinery to improve farm labour efficiency, maximise marginal output and increase food production (Onwude et al., 2016). The application of fertilizers plays a key role in modern agriculture and the need to increase food production. The nitrogen (N) and phosphorus (P) fertilizer usage has increased by approximately 8 and 3 times, respectively, since 1961 (Lu and Tian, 2017). Other chemical products including pesticides, fungicides and herbicides increase plants' resistance to pests, diseases and weeds, which improve crop yield. Intensive crop production has led to a 15 to 20 fold increase in pesticides use worldwide (Oerke, 2006). It is projected that the total food production in the next 50 years will equal the cumulative production in the past 500 years (Hatfield and Walthall, 2015).

With the remarkable success in agricultural production, growing concerns about environmental pollution from agriculture also emerged. Increased chemical fertilizer application negatively impacted soil and water quality (Khan et al., 2013; Smith et al., 2016; Sobota et al., 2015; Sun et al., 2012). Pesticide residue accumulation in the environment is also a great concern (Vázquez-Boucard et al., 2014). Intensive tillage practices can accelerate soil erosion and nutrient loss (Beniston et al., 2015; Gao et al., 2016; Rhoton et al., 2002). These growing concerns call for a more environmental-friendly production system that fulfills the crop needs in a sustainable way. Conservation agriculture (CA) ensures both productivity and sustainability . The Food and Agriculture Organization of the United Nations (FAO) defines CA as an approach to manage agro-ecosystems for improved and sustained productivity and increased profits while enhancing or preserving the resource base and the environment. CA practices are developed based on three principles: minimum tillage, permanent soil cover and crop diversity (FAO, 2015). Cover cropping has emerged as one of the most versatile CA strategies because it offers multiple benefits to crops and soil.

Cover cropping

The Soil Science Society of America (SSSA) defines cover crops (CCs) as close-growing crops that provide soil protection, seeding protection and soil improvement between periods of normal crop production (SSSA, 2008). Current agricultural cropping systems generally do not cover the soil throughout the year. During the winter months, soil is typically unprotected which increases the risk of erosion and nutrient runoff, and could decrease the overall soil quality and crop productivity in the next growing season (Kaspar and Singer, 2011). Planting CCs is one conservation agricultural practice that is being promoted for controlling soil erosion, reducing weed growth, improving soil and water quality, and enhancing crop productivity (Hobbs et al., 2008). Integrating CCs into cropping systems can benefit soil's physical, chemical and biological properties.

Soil physical properties

CCs have the potential to alter soil physical properties directly through the formation of pores and aggregates by roots, and indirectly through the input and decomposition of plant residues (Blanco-Canqui et al., 2015). It has been observed that CCs can improve the soil aggregate stability by protecting the soil surface from raindrop impact, providing additional above and below ground biomass input, and increasing soil organic carbon (SOC) concentration and microbial activity (Blanco-Canqui et al., 2013). The improvement in soil aggregate formation and stability enhances storage of soil water and nutrients, improves root growth and in the long term can have a positive effect on soil hydraulic properties including water infiltration, water retention capacity, and saturated hydraulic conductivity (Keisling et al., 1994). With roots penetrating the compacted soil layer, CCs can also reduce soil compaction (Blanco-Canqui et al., 2012; Chen and Weil, 2010). Above and belowground root systems of the CCs also help in reducing soil erosion from wind and water (Blanco-Canqui et al., 2013; Clark et al., 1997; Truman et al., 2003).

Soil chemical properties

Numerous studies have been conducted on the effect of CCs in soil nutrient management. CCs scavenge nutrients from soil in winter and release them back to soil when the CC biomass decomposes. This process helps to reduce the nutrient loss through runoff, leaching and erosion during the non-growing season (Dabney et al., 2001; Eckert, 1991; Kaspar et al., 2001; Kleinman et al., 2005; Oliveira et al., 2017). Considering the mobile nature of nitrate (NO_3^-), N losses from NO_3^- leaching will reduce the fertilizer efficiency and increase non-point source pollution to nearby water bodies. During the winter months, CCs function as regular crops to reduce this massive loss of N (Kaspar and Singer, 2011; Malone et al., 2014). In addition to the enhanced N availbility by reducing N loss, leguminous CCs provide additional N input to soil by N-fixing from the atmosphere (Blanco-Canqui et al., 2015; Dabney et al., 2001; Kristensen and Thorup-Kristensen, 2004). Few studies have assessed the effect of CCs on soil potassium (K) content and soil pH. Some authors have reported that CCs did not change (Nyakatawa et al., 2001) or reduced soil pH (Jokela et al., 2009; Hargrove, 1986). Eckert (1991) reported an improvement of K content in the surface soil by growing cereal rye (*Secale cereale* L.) as a CC.

Soil biological properties

From a biological perspective, CCs improve plant coverage, organic matter input and biodiversity (Reddy et al., 2003; Reeleder et al., 2006; Tillman et al., 2004). In general, CCs can increase SOC concentration due to additional above and belowground biomass input (Poeplau and Don, 2015; Sainju et al., 2002). This effect varies mainly with CC species, soil type, tillage mangement and duration of cover cropping (Acuña and Villamil, 2014; Olson et al., 2014). Mbuthia et al. (2015) reported significantly higher soil microbial biomass N (SBN), total Fatty Acid Methyl Ester (FAME), b-glucosaminidase (N-cycling) activity and basal microbial respiration rates after 33-years of hairy vetch *(Vicia villosa*) cover cropping compared to wheat (*Triticum*) or no cover. CCs can also suppress weeds, plant pathogens and nematodes through chemical substances released as root exudates (Keating, 1999; Lawley et al., 2011). These benefits can improve overall soil quality and increase crop productivity.

Cover cropping and crop production

The impact of cover cropping on crop yield varies with several factors including CC species, growing season, annual precipitation and CC management strategies. A recent meta-analysis including data from 65 published articles from United States and Canada revealed that introducing CCs to row crop production systems did not reduce corn (*Zea mays*) productivity if properly managed (Marcillo and Miguez, 2017). In regions with higher precipitation (>800 mm), CCs often increase soil water storage capacity and benefit crop production. This indicates that CCs have potential to increase crop yield in areas receiving higher precipitation, while in semiarid regions CCs may reduce or have no effect on crop yield (Balkcom and Reeves, 2005; Blanco-Canqui et al., 2012). It is reported that CCs such as sunn hemp (*Crotalaria juncea* L.) and late-maturing soybean (*Glycine max*) increased crop yield in a no-till winter wheat–grain sorghum (*Sorghum bicolor*) rotation with a low rate of inorganic N application in the south-central Kansas with 878 mm mean annual precipitation (Blanco-Canqui et al., 2012). Strong N-fixing CC species show a faster and greater effect on crop yields than the species with low or no N–fixing capacity (Blanco-Canqui et al., 2015). In Tennessee (TN), a 33-year long-term study showed yield improvement in cotton (*Gossypium hirsutum* L.) by continuous hairy vetch planting (Mbuthia et al., 2015). This is also confirmed in a 12-year cotton-corn rotation field in the same location with hairy vetch as CC (Ashworth et al., 2016). Regardless, it may require many years of cover cropping to experience the yield increase (Andraski and Bundy, 2005; Decker et al., 1994).

Potential of multi-species cover crops

Different CC species perform distinct functions. For example, legume species can function as an additional N source, tap-rooted CCs such as brassicas can reduce soil compaction, and grasses such as rye can reduce erosion (Chen and Weil, 2010; Ebelhar et al., 1984; Kaspar et al., 2001). The growth of multi-species mixtures of CCs has the potential to provide multiple benefits simultaneously (Kramberger et al., 2014; Tosti et al., 2014). In recent years there has been an increasing interest in using multi-species CCs. A multi-species mixture called soil health mix (SHM) is being promoted by the United States Department of Agriculture-Natural Resources Conservation Serivce (USDA-NRCS) through the Environmental Quality Incentives Program (EQIP) (USDA, 2016a). A recent press release from the USDA-NRCS stated that a mixture of CCs works better than a single species (USDA, 2016b). This increasing interest in using CC mixtures warrants experimental data on their effect on soil quality and crop performance. After all, an assessment tool is needed to evaluate the overall effect of any CA practices including cover cropping on soil quality. The following sections describe the current state-of-the-art of the concept of soil quality and how we can improve soil quality by CA approaches such as cover cropping .

Soil quality

The terms soil health and soil quality are most often used synonymously. The concept of soil health dates back to ancient civilizations (Doran et al., 1996) and it is quite like that of human health. Essential biological, chemical and physical soil components must be present and function to permit the growth of healthy and high-yielding crops (Magdoff, 2001). For farmers, the term soil health is often favored over soil quality because it is easy to characterize based on descriptive and qualitative properties by using direct value judgments (unhealthy to healthy) (Romig et al., 1995). USDA-NRCS has been engaged in developing “soil health cards” that are appropriate for local conditions (USDA, 1997).

The concept of soil quality is changing with our increased understanding of soil. In the past, this concept was mainly summarized as its suitability for different uses (Olson, 1943). Interest in soil biology grew tremendously after the understanding of soil bacterial functions and N fixation by symbiotic and free-living bacteria, which leads to the addition of soil biological components to the term soil quality (Warkentin, 1995). Soil quality is defined as “the capacity of soil to function, within ecosystem and land use boundaries, to sustain productivity, maintain environmental quality, and promote plant and animal health” (Doran and Parkin, 1994). The USDA’s Guidelines for Soil Quality Assessment defines soil function as follows (Seybold et al., 1997):

- Sustaining biological diversity, activity, and productivity
- Regulating water and solute flow
- Filtering, buffering, and degrading organic and inorganic materials
- Storing and cycling nutrients and carbon
- Providing physical stability and support

Though soil health and soil quality are used interchangeably in the literature, soil health often focuses more on soil biological aspects while soil quality is a broader concept. According to Van Bruggen and Semenov (2000) and Franzluebbers (2016), health can only be associated with something that is living. For example, in the case of the newly emerging Haney's soil health test, heavy focus has been given to the biological soil properties (Haney, 2013).

Indicators of soil quality

To obtain an overall understanding of the quality or health of a specific soil, selection of specific indicators is needed as it is unrealistic and cumbersome to use all soil properties as indicators. One practical way is to identify a minimum set of meaningful soil properties as core attributes for soil quality assessment. These core attributes of soil quality are divided into three broad categories (chemical, physical and biological) depending on how they affect soil's functions (Larson and Pierce, 1991). The chemical indicators encompass many of the traditional soil properties including pH, electrical conductivity, soil nutrients and metal content. Physical indicators mainly represent the soil's functions to regulate water and solute flow, support physical stability and sustain nutrition (Karlen and Stott, 1994), which include soil structure, infiltration rate, hydraulic conductivity and compaction. Biological category covers indicators related to microbial activity and soil organic matter (Pankhurst et al., 1997). Besides, anthropogenic changes are also important because they represent the effects of land use and management on soil quality (Wienhold et al., 2004). These categories are not always clearly defined since several indicators often affect multiple soil functions. Choice of indicators various among soil quality assessment methods developed for different regions and they are chosen based on the convenience in measuring and significance in representing the problem of interest (Schloter et al., 2003). For example, the NRCS's indicator guide provides 17 indicators while the Cornell Soil Health Test covers 13 indicators and Haney’s Soil Health Score uses 3 indicators (Haney, 2013; Moebius-Clune et al., 2016).

Soil quality assessment methods

Although the indicators vary among different assessment methods, the general assessment approach is mostly similar. The first step is to set an assessment goal, collect information and select the proper indicators. The assessment goals typically fall in the areas of crop productivity, soil or environmental protection (Friedman et al., 2001). The next step is the data collection and analysis. Different indicators and data collection methods are needed for site-specific problems and agricultural systems (Shukla et al., 2006). A scoring system is often applied in various assessment methods, which provides convenience for evaluation, comparison of the test results to a reference value, and clear and quantitative understanding of the quality of the tested soil to the users. Reference values are often developed from the previous literature and research data, and knowledge about the pedogenesis of the specific soil (Stasch and Stahr, 1993). After obtaining the results, it is essential to interpret them and make recommendations for improvement, maintenance or remediation plan to achieve the set goal. Evaluation of the plan during and after the implementation is also essential to improve the whole process (Larson and Pierce, 1994; Romig et al., 1995; Seybold et al., 1997).

Soil management assessment framework (SMAF)

Andrews et al. (2004) developed the SMAF approach to calculate soil quality indices in response to agroecosystem management practices.. This framework outlines three basic steps such as indicator selection, indicator interpretation and index Integration and uses scoring functions to translate individual indicators to overall soil quality index(Andrews et al., 2002; Karlen and Stott, 1994)

Cornell Soil Health Test (CSHT)

By adapting the SMAF, Cornell's soil testing lab developed CSHT which is an integrative soil quality assessment tool covering physical, chemical and biological properties as soil health indicators. Indicators were selected from 42 potential indicator list and given weightage. Then, scoring functions were used to convert the indicators to soil health scores. Individual soil health scores were then integrated using an un-weighted average to give an overall soil health score with rankings (Moebius-Clune et al., 2016).

Alabama Soil Quality Index (SQI)

This was developed based on the CSHT by site-specific modifications to make it more suitable for Alabama soil. A fixed indicator list was given which mainly focused on soil chemical properties. One big difference was that a weight was assigned to each factor based on the judgment of the scientist panel instead of the un-weighed average in CSHT (Bosarge, 2015).

Haney's soil health test

Haney's soil health test is quite distinct compared to other soil quality/health assessment methods. Haney's test includes a unique set of parameters which are related to soil microbial activity and functions. It also offers modifications to fertilizer recommendations developed based on traditional soil testing. In addition to the N, P and K (which are part of many traditional soil testing), Haney's soil health test also provides water extractable organic C (WEOC) and water extractable organic N (WEON) contents (Haney, 2013). According to Haney et al. (2012), WEOC/WEON ratio was a more sensitive indicator of soil microbial activity than the traditional soil C:N ratio, and they suggest that a healthy soil should have a WEOC/WEON ratio below 20:1, which can be used as a practical threshold level to separate the healthy soils from soils that may have immobilized N with high microbial activity. The major outcome of Haney's soil health test is a "soil health score," which is calculated using the equation below:

$$\frac{\text{1-day } CO_2\text{-C}}{\text{Water extractable organic C:N}} + \frac{\text{WEOC}}{100} + \frac{\text{WEON}}{10}$$

The calculation combines three independent soil measurements. The health score varies from 0 to 50 (Haney, 2013).

Haney et al. (2006) developed a new multi-nutrient extractant called H3A for the simultaneous determination of N (ammonium and nitrate), P and K (Haney et al., 2010). This extractant is designed to mimic the chemical environment created around the actively growing roots mostly by the root exudates, so nutrients can be extracted near the field chemical conditions (Haney et al., 2006). Since soil pH and P solubility are highly interrelated (Golterman, 1988; Sharpley, 1993), it seems that the H3A extractant can provide more reliable P extraction efficiency. The original H3A extractant is composed of organic acids, lithium citrate, and two synthetic chelators that are diethylenetriaminepentaacetic acid (DTPA) and ethylenediaminetetraacetic acid (EDTA). After a series of modifications, lithium citrate, EDTA and DTPA were eliminated leaving only three organic acids such as citric acid, oxalic acid and acetic acid (Haney et al., 2016).

The core of this health test is the measurement of carbon dioxide (CO_2)-C evolved from a 24-hour long incubation of re-wetted air-dried soil. The Solvita gel system is used to measure the 1d-CO_2-C. The Solvita gel system is originally designed to quantify the relative difference in CO_2 evolution across varying types of composts in a short period (Brewer and Sullivan, 2003). This has been proved to be a reasonably accurate method to measure the soil CO_2 respiration due to its high correlation with traditional methods of CO_2 measurements including acid-base titration and Infra-Red Gas Analysis (IRGA) methods (Haney et al., 2008b). It is also recommended by the USDA Soil Quality Institute as an alternate soil respiration procedure (USDA, 1999). The flush of CO_2 following rewetting of dried soil is closely related to 28-day N and P mineralization as well as the initial WEOC and WEON concentrations (Haney et al., 2008a; Haney et al., 2012). This result also indicates the suitability of using the flush of CO_2 as a viable test for biological soil quality. Later a strong relationship between Solvita 1-day CO_2-C and potential N mineralization was developed using a suite of soils from across the US (r^2=0.82) (Haney et al., 2015). A Potential Mineralizable Nitrogen Calculator was developed to help users interpret the Solvita CO_2 result, and to provide N fertilizer recommendation by accounting for the potentially mineralizable N (Solvita, 2017).

Summary

Cover cropping is one of the conservation agricultural practices. Though CCs exhibit multiple benefits to agro-ecosystems, these benefits largely depend on several factors including soil type, cropping systems, weather and cover crop species. Moreover, cover cropping is considered a costy practice and it needs to be practised for a long period to observe notable impacts (Ryan et al., 2003). For many producers, it takes years to establish a profitable and convenient way to integrate CCs into their cropping systems (Dunn et al., 2016). Studies are needed to help farmers better understand the effects of CCs. There are growing interests in using multi-species CCs. However, studies on the effect of double or multi-species CCs in comparison with single species on soil quality and crop production are limited, especially in the south-east US. Owing to the fact that more and more farmers are interested in a diversity of CCs, it is essential to compare the performance of several species on CCs growing under similar soil type, environmental conditions and cropping systems. It will help farmers to make an informed decision on CC species selection rather than on assumptions or just availability of seeds.

Haney’s soil health test is a newly developed method which aims to evaluate the overall soil health status based on mainly soil biological characteristics (Haney, 2013). It is claimed to be convenient and fast compared to other methods. In addition to providing the soil health score, Haney's method is also designed to modify the nutrient recommendations developed through the traditional soil testing. Considering the fact that Haney's soil health test is originally developed for the state of Texas, more evaluation of the merit of this method on various soil types is needed to confirm its versatility in different regions of US.

Research objectives

In this study, we examined both short-term and long-term effects of different CC treatments (single species, double species, multi-species) on soil quality attributes and crop yield. We also evaluated the performance of Haney's soil health test in the row crop production belt of TN. The specific objectives are listed below:

Objective 1. Evaluation of the long-term and short-term effects of different CC species on soil quality attributes and crop yield.

Hypothesis: Multi-species CC mixtures will provide an improved effect on soil quality and crop yield, compared to single and double species CC species.

Objective 2. Evaluation of the three components of Haney's soil health test (extractant, CO_2 respiration and health score) on soil typical to TN.

Hypothesis: Haney's soil health test will not show a good result in representing the soil status in TN and it needs modification to make it better fit to TN.

Overall, the outcomes of this study are: 1) a comprehensive examination of the short-term and long-term effects of different CC species on soil quality attributes and crop yield; and 2) an evaluation of the effectiveness of Haney's soil health test for the row crop production systems of TN. These outcomes will provide information on how different CC species perform in TN soils and provide insights on modifying Haney's soil health test to adapt to other regions.

References

[1] Acuña, J. and M.B. Villamil. 2014. Short-term effects of cover crops and compaction on soil properties and soybean production in Illinois. Agronomy Journal 106: 860-870.

[2] Andraski, T.W. and L.G. Bundy. 2005. Cover crop effects on corn yield response to nitrogen on an irrigated sandy soil. Agronomy Journal 97: 1239-1244.

[3] Andrews, S., D. Karlen and J. Mitchell. 2002. A comparison of soil quality indexing methods for vegetable production systems in Northern California. Agriculture, ecosystems & environment 90: 25-45.

[4] Andrews, S.S., D.L. Karlen and C.A. Cambardella. 2004. The soil management assessment framework. Soil Science Society of America Journal 68: 1945-1962.

[5] Ashworth, A.J., F.L. Allen, A.M. Saxton and D.D. Tyler. 2016. Long-Term Corn Yield Impacted by Cropping Rotations and Bio-Covers under No-Tillage. Agronomy Journal 108: 1495-1502.

[6] Balkcom, K.S. and D.W. Reeves. 2005. Sunn-hemp utilized as a legume cover crop for corn production. Agronomy journal 97: 26-31.

[7] Beniston, J., M. Shipitalo, R. Lal, E. Dayton, D. Hopkins, F. Jones, A. Joynes and J. Dungait. 2015. Carbon and macronutrient losses during accelerated erosion under different tillage and residue management. European Journal of Soil Science 66: 218-225.

[8] Blanco-Canqui, H., M. Claassen and D. Presley. 2012. Summer cover crops fix nitrogen, increase crop yield, and improve soil–crop relationships. Agronomy journal 104: 137-147.

[9] Blanco-Canqui, H., C.A. Shapiro, C.S. Wortmann, R.A. Drijber, M. Mamo, T.M. Shaver and R.B. Ferguson. 2013. Soil organic carbon: The value to soil properties. Journal of Soil and Water Conservation 68: 129A-134A.

[10] Blanco-Canqui, H., T.M. Shaver, J.L. Lindquist, C.A. Shapiro, R.W. Elmore, C.A. Francis and G.W. Hergert. 2015. Cover crops and ecosystem services: Insights from studies in temperate soils. Agronomy Journal 107: 2449-2474.

[11] Bosarge, T. 2015. A Soil Quality Index for Alabama. M.S. Thesis, Auburn University, Auburn, AL.

[12] Brewer, L.J. and D.M. Sullivan. 2003. Maturity and stability evaluation of composted yard trimmings. Compost Science & Utilization 11: 96-112.

[13] Brummer, E.C., W.T. Barber, S.M. Collier, T.S. Cox, R. Johnson, S.C. Murray, R.T. Olsen, R.C. Pratt and A.M. Thro. 2011. Plant breeding for harmony between agriculture and the environment. Frontiers in Ecology and the Environment 9: 561-568.

[14] Chen, G. and R.R. Weil. 2010. Penetration of cover crop roots through compacted soils. Plant and Soil 331: 31-43.

[15] Clark, A.J., A.M. Decker, J.J. Meisinger and M.S. McIntosh. 1997. Kill date of vetch, rye, and a vetch-rye mixture: II. Soil moisture and corn yield. Agronomy Journal 89: 434-441.

[16] Dabney, S., J. Delgado and D. Reeves. 2001. Using winter cover crops to improve soil and water quality. Communications in Soil Science and Plant Analysis 32: 1221-1250.

[17] Decker, A.M., A.J. Clark, J.J. Meisinger, F.R. Mulford and M.S. McIntosh. 1994. Legume cover crop contributions to no-tillage corn production. Agronomy journal 86: 126-135.

[18] Doran, J.W. and T.B. Parkin. 1994. Defining and assessing soil quality. In: J.W. Doran, et al., editors, SSSA Special Publication, Defining Soil Quality for a Sustainable Environment. Soil Science Society of America and American Society of Agronomy, Madison, WI. p. 3-21.

[19] Doran, J.W., M. Sarrantonio and M.A. Liebig. 1996. Soil Health and Sustainability. Advances in Agronomy 56: 1-54.

[20] Dunn, M., J. Ulrich-Schad, L. Prokopy, R. Myers, C. Watts and K. Scanlon. 2016. Perceptions and use of cover crops among early adopters: Findings from a national survey. Journal of Soil and Water Conservation 71: 29-40.

[21] Ebelhar, S., W. Frye and R. Blevins. 1984. Nitrogen from legume cover crops for no-tillage corn. Agronomy journal 76: 51-55.

[22] Eckert, D. 1991. Chemical attributes of soils subjected to no-till cropping with rye cover crops. Soil Science Society of America Journal 55: 405-409.

[23] FAO, 2015, What is conservation agriculture?, http://www.fao.org/ag/ca/1a.html, Accessed [May 30, 2017]

[24] Franzluebbers, A.J. 2016. Should Soil Testing Services Measure Soil Biological Activity? Agricultural & Environmental Letters 10.2134/ael2015.11.0009. doi:10.2134/ael2015.11.0009.

[25] Friedman, D., M. Hubbs, A. Tugal, C. Seybold and M. Sucik, 2001, Guidelines for soil quality assessment in conservation planning, https://www.nrcs.usda.gov/Internet/FSE_DOCUMENTS/nrcs142p2_051259.pdf,

[26] Gao, Y., X. Dang, Y. Yu, Y. Li, Y. Liu and J. Wang. 2016. Effects of tillage methods on soil carbon and wind erosion. Land Degradation & Development 27: 583-591.

[27] Golterman, H. 1988. Reflections on fractionation and bioavailability of sediment bound phosphate. Arch. Hydrobiol. Beih. Ergebn. Limnol 30: 1-4.

[28] Haney, R.L., 2013, Soil Testing for Soil Health, https://www.usda.gov/oce/forum/past_speeches/2013_Speeches/Haney.pdf, Accessed [May 29, 2017]

[29] Haney, R.L., W. Brinton and E. Evans. 2008a. Estimating soil carbon, nitrogen, and phosphorus mineralization from short-term carbon dioxide

respiration. Communications in Soil Science and Plant Analysis 39: 2706-2720.

[30] Haney, R.L., W. Brinton and E. Evans. 2008b. Soil CO_2 respiration: Comparison of chemical titration, CO_2 IRGA analysis and the Solvita gel system. Renewable agriculture and food systems 23: 171-176.

[31] Haney, R.L., A.J. Franzluebbers, V.L. Jin, M.-V. Johnson, E.B. Haney, M.J. White and R.D. Harmel. 2012. Soil organic C: N vs. water-extractable organic C: N. Open Journal of Soil Science 2: 269-274.

[32] Haney, R.L., E.B. Haney, D. Harmel, D. Smith and M. White. 2016. Evaluation of H3A for Determination of Plant Available P vs. FeAlO Strips. Open Journal of Soil Science 6: 175-187.

[33] Haney, R.L., E.B. Haney, L. Hossner and J. Arnold. 2006. Development of a new soil extractant for simultaneous phosphorus, ammonium, and nitrate analysis. Communications in Soil Science and Plant Analysis 37: 1511-1523.

[34] Haney, R.L., E.B. Haney, L. Hossner and J. Arnold. 2010. Modifications to the new soil extractant H3A-1: A multinutrient extractant. Communications in soil science and plant analysis 41: 1513-1523.

[35] Haney, R.L., E.B. Haney, D.R. Smith and M.J. White. 2015. Estimating Potential Nitrogen Mineralisation Using the Solvita Soil Respiration System. Open Journal of Soil Science 5: 319-323.

[36] Hargrove, W. 1986. Winter legumes as a nitrogen source for no-till grain sorghum. Agronomy Journal 78: 70-74.

[37] Hatfield, J.L. and C.L. Walthall. 2015. Soil biological fertility: Foundation for the next revolution in agriculture? Communications in Soil Science and Plant Analysis 46: 753-762.

[38] Hobbs, P.R., K. Sayre and R. Gupta. 2008. The role of conservation agriculture in sustainable agriculture. Philosophical Transactions of the Royal Society B: Biological Sciences 363: 543-555.

[39] Jokela, W.E., J.H. Grabber, D.L. Karlen, T.C. Balser and D.E. Palmquist. 2009. Cover crop and liquid manure effects on soil quality indicators in a corn silage system. Agronomy Journal 101: 727-737.

[40] Karlen, D.L. and D.E. Stott. 1994. A framework for evaluating physical and chemical indicators of soil quality. In: J.W. Doran, et al., editors, SSSA Special Publication, Defining Soil Quality for a Sustainable Environment. Soil Science Society of America and American Society of Agronomy, Madison, WI. p. 53-72.

[41] Kaspar, T., J. Radke and J. Laflen. 2001. Small grain cover crops and wheel traffic effects on infiltration, runoff, and erosion. Journal of Soil and Water Conservation 56: 160-164.

[42] Kaspar, T. and J. Singer. 2011. The use of cover crops to manage soil. In: J.L. Hatfield and T.J. Sauer, editors, Soil Management: Building a Stable Base for Agriculture. American Society of Agronomy and Soil Science Society of America, Madison, WI. p. 321-337.

[43] Keating, K.I. 1999. Allelopathy: principles, procedures, processes, and promises for biological control. Advances in Agronomy 67: 141-231.

[44] Keisling, T., H. Scott, B. Waddle, W. Williams and R. Frans. 1994. Winter cover crops influence on cotton yield and selected soil properties. Communications in Soil Science & Plant Analysis 25: 3087-3100.

[45] Khan, S., M. Shahnaz, N. Jehan, S. Rehman, M.T. Shah and I. Din. 2013. Drinking water quality and human health risk in Charsadda district, Pakistan. Journal of cleaner production 60: 93-101.

[46] Kleinman, P., P. Salon, A. Sharpley and L. Saporito. 2005. Effect of cover crops established at time of corn planting on phosphorus runoff from soils before and after dairy manure application. Journal of soil and water conservation 60: 311-322.

[47] Kramberger, B., A. Gselman, J. Kristl, M. Lešnik, V. Šuštar, M. Muršec and M. Podvršnik. 2014. Winter cover crop: the effects of grass–clover

mixture proportion and biomass management on maize and the apparent residual N in the soil. European Journal of Agronomy 55: 63-71.

[48] Kristensen, H.L. and K. Thorup-Kristensen. 2004. Root growth and nitrate uptake of three different catch crops in deep soil layers. Soil Science Society of America Journal 68: 529-537.

[49] Larson, W. and F. Pierce. 1994. The dynamics of soil quality as a measure of sustainable management. Soil Science 35: 37-51.

[50] Larson, W. and F. Pierce. 1991. Conservation and enhancement of soil quality. In: C.R. Elliott, editor Evaluation for sustainable land management in the developing world: proceedings of the International Workshop on Evaluation for Sustainable Land Management in the Developing World. International Board for Soil Research and Management, Chiang Rai, Thailand.

[51] Lawley, Y.E., R.R. Weil and J.R. Teasdale. 2011. Forage radish cover crop suppresses winter annual weeds in fall and before corn planting. Agronomy journal 103: 137-144.

[52] Lu, C. and H. Tian. 2017. Global nitrogen and phosphorus fertilizer use for agriculture production in the past half century: shifted hot spots and nutrient imbalance. Earth System Science Data 9: 181-192.

[53] Magdoff, F. 2001. Concept, components, and strategies of soil health in agroecosystems. Journal of nematology 33: 169-172.

[54] Malone, R., D. Jaynes, T. Kaspar, K. Thorp, E. Kladivko, L. Ma, D. James, J. Singer, X. Morin and T. Searchinger. 2014. Cover crops in the upper midwestern United States: Simulated effect on nitrate leaching with artificial drainage. Journal of soil and water conservation 69: 292-305.

[55] Marcillo, G.S. and F.E. Miguez. 2017. Corn yield response to winter cover crops: An updated meta-analysis. Journal of Soil and Water Conservation 72: 226-239.

[56] Mbuthia, L.W., V. Acosta-Martinez, J. DeBruyn, S. Schaeffer, D. Tyler, E. Odoi, M. Mpheshea, F. Walker and N. Eash. 2015. Long term tillage, cover crop, and fertilization effects on microbial community structure, activity: Implications for soil quality. Soil Biology and Biochemistry 89: 24-34.

[57] Moebius-Clune, B.N., D.J. Moebius-Clune, B.K. Gugino, O.J. Idowu, R.R. Schindelbeck, A.J. Ristow, H.M.v. Es, J.E. Thies, H.A. Shayler, M.B. McBride, D.W. Wolfe and G.S. Abawi. 2016. Comprehensive Assessment of Soil Health – The Cornell Framework ManualCornell University, Geneva, NY.

[58] Nyakatawa, E., K. Reddy and K. Sistani. 2001. Tillage, cover cropping, and poultry litter effects on selected soil chemical properties. Soil and Tillage Research 58: 69-79.

[59] Oerke, E.-C. 2006. Crop losses to pests. The Journal of Agricultural Science 144: 31-43.

[60] Oliveira, R.A.D., J.J. Comin, T. Tiecher, R. Piccin, L.M. Somavilla, A. Loss, C.R. Lourenzi, C. Kürtz and G. Brunetto. 2017. Release of Phosphorus Forms from Cover Crop Residues in Agroecological No-Till Onion Production. Revista Brasileira de Ciência do Solo 10.1590/18069657rbcs20160272 doi:10.1590/18069657rbcs20160272

[61] Olson, K., S.A. Ebelhar and J.M. Lang. 2014. Long-term effects of cover crops on crop yields, soil organic carbon stocks and sequestration. Open Journal of Soil Science 4: 284-292.

[62] Olson, L. 1943. Columella and the beginning of soil science. Agricultural History 17: 65-72.

[63] Onwude, D.I., R. Abdulstter, C. Gomes and N. Hashim. 2016. Mechanisation of large-scale agricultural fields in developing countries–a review. Journal of the Science of Food and Agriculture 96: 3969-3976.

[64] Pankhurst, C.E., B.M. Doube and V.V.S.R. Gupta. 1997. Biological indicators of soil health: synthesis. In: C.E. Pankhurst, et al., editors, Biological Indicators of Soil Health. CAB International, Wallingford, UK.

[65] Poeplau, C. and A. Don. 2015. Carbon sequestration in agricultural soils via cultivation of cover crops–A meta-analysis. Agriculture, Ecosystems & Environment 200: 33-41.

[66] Reddy, K.N., R.M. Zablotowicz, M.A. Locke and C.H. Koger. 2003. Cover crop, tillage, and herbicide effects on weeds, soil properties, microbial populations, and soybean yield. Weed Science 51: 987-994.

[67] Reeleder, R., J. Miller, B.B. Coelho and R. Roy. 2006. Impacts of tillage, cover crop, and nitrogen on populations of earthworms, microarthropods, and soil fungi in a cultivated fragile soil. Applied Soil Ecology 33: 243-257.

[68] Rhoton, F., M. Shipitalo and D. Lindbo. 2002. Runoff and soil loss from midwestern and southeastern US silt loam soils as affected by tillage practice and soil organic matter content. Soil and Tillage Research 66: 1-11.

[69] Romig, D.E., M.J. Garlynd, R.F. Harris and K. McSweeney. 1995. How farmers assess soil health and quality. Journal of soil and water conservation 50: 229-236.

[70] Ryan, R.L., D.L. Erickson and R. De Young. 2003. Farmers' motivations for adopting conservation practices along riparian zones in a mid-western agricultural watershed. Journal of Environmental Planning and Management 46: 19-37.

[71] Sainju, U., B. Singh and W. Whitehead. 2002. Long-term effects of tillage, cover crops, and nitrogen fertilization on organic carbon and nitrogen concentrations in sandy loam soils in Georgia, USA. Soil and Tillage Research 63: 167-179.

[72] Schloter, M., O. Dilly and J. Munch. 2003. Indicators for evaluating soil quality. Agriculture, Ecosystems & Environment 98: 255-262.

[73] Seybold, C.A., M.J. Mausbach, D.L. Karlen and H.H. Rogers. 1997. Quantification of soil quality. In: R. Lal, et al., editors, Soil processes and the carbon cycle. CRC Press, Boca Raton, FL. p. 387-404.

[74] Sharpley, A.N. 1993. An innovative approach to estimate bioavailable phosphorus in agricultural runoff using iron oxide-impregnated paper. Journal of Environmental Quality 22: 597-601.

[75] Shukla, M., R. Lal and M. Ebinger. 2006. Determining soil quality indicators by factor analysis. Soil and Tillage Research 87: 194-204.

[76] Smith, D.R., R.D. Harmel, M. Williams, R.L. Haney and K.W. King. 2016. Managing Acute Phosphorus Loss with Fertilizer Source and Placement: Proof of Concept. Agricultural & Environmental Letters 10.2134/ael2015.12.0015. doi:10.2134/ael2015.12.0015.

[77] Sobota, D.J., J.E. Compton, M.L. McCrackin and S. Singh. 2015. Cost of reactive nitrogen release from human activities to the environment in the United States. Environmental Research Letters 10.1088/1748-9326/10/2/025006. doi:10.1088/1748-9326/10/2/025006.

[78] Solvita, 2017, Guide for Interpreting CO_2 Burst Results, https://solvita.com/soil/potential-min-n-calc/, Accessed [June 15, 2017]

[79] SSSA. 2008. Glossary of soil science terms 2008Soil Science Society of America Madison WI.

[80] Stasch, D. and K. Stahr. 1993. The soil potential concept-A method for the fundamental evaluation of soils and their protection. In: H.J.P. Eijsackers and T. Hamers, editors, Integrated Soil and Sediment Research: A Basis for Proper Protection. Kluwer Academic Publishers, Dordrecht, Netherlands p. 107-109.

[81] Sun, B., L. Zhang, L. Yang, F. Zhang, D. Norse and Z. Zhu. 2012. Agricultural non-point source pollution in China: causes and mitigation measures. Ambio 41: 370-379.

[82] Tillman, G., H. Schomberg, S. Phatak, B. Mullinix, S. Lachnicht, P. Timper and D. Olson. 2004. Influence of cover crops on insect pests and predators in conservation tillage cotton. Journal of economic entomology 97: 1217-1232.

[83] Tilman, D., C. Balzer, J. Hill and B.L. Befort. 2011. Global food demand and the sustainable intensification of agriculture. Proceedings of the National Academy of Sciences 108: 20260-20264.

[84] Tosti, G., P. Benincasa, M. Farneselli, F. Tei and M. Guiducci. 2014. Barley–hairy vetch mixture as cover crop for green manuring and the mitigation of N leaching risk. European Journal of Agronomy 54: 34-39.

[85] Truman, C.C., D. Reeves, J.N. Shaw, A.C. Motta, C.H. Burmester, R.L. Raper and E.B. Schwab. 2003. Tillage impacts on soil property, runoff, and soil loss variations from a Rhodic Paleudult under simulated rainfall. Journal of Soil and Water Conservation 58: 258-267.

[86] USDA, 1997, Maryland Soil Quality Assessment Book, https://www.nrcs.usda.gov/wps/PA_NRCSConsumption/download?cid=nrcs142p2_050955&ext=pdf., Accessed [April 30, 2017]

[87] USDA, 1999, Soil Quality Test Kit Guide, https://www.nrcs.usda.gov/Internet/FSE_DOCUMENTS/stelprdb1044790.pdf, Accessed [April 30,2017]

[88] USDA, 2016a, COVER CROP (340) REQUIREMENTS IN TENNESSEE, https://www.nrcs.usda.gov/wps/PA_NRCSConsumption/download?cid=nrcseprd414433&ext=pdf, Accessed [May 30, 2017]

[89] USDA, 2016b, Cover Crop Mixes? They Just Work Better, https://www.nrcs.usda.gov/wps/portal/nrcs/detail/il/newsroom/releases/?cid=STELPRDB1117185, Accessed [April 30, 2017]

[90] Van Bruggen, A. and A. Semenov. 2000. In search of biological indicators for soil health and disease suppression. Applied Soil Ecology 15: 13-24.

[91] Vázquez-Boucard, C., G. Anguiano-Vega, L. Mercier and E. Rojas del Castillo. 2014. Pesticide residues, heavy metals, and DNA damage in sentinel oysters Crassostrea gigas from Sinaloa and Sonora, Mexico. Journal of Toxicology and Environmental Health, Part A 77: 169-176.

[92] Warkentin, B.P. 1995. The changing concept of soil quality. Journal of soil and water conservation 50: 226-228.

[93] Wienhold, B.J., S. Andrews and D. Karlen. 2004. Soil quality: a review of the science and experiences in the USA. Environmental geochemistry and health 26: 89-95.

CHAPTER II

EFFECT OF COVER CROP SPECIES ON SOIL QUALITY AND CROP YIELD IN A CORN-SOYBEAN ROTATION IN WESTERN TENNESSEE

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Abstract

Cover crops (CCs) provide several benefits in row crop production systems. Long-term studies show that the effect of CCs vary with CC species, soil types and cropping systems, and it often takes a long time for CCs to deliver a significant benefit to a particular production system. Comprehensive studies to understand the effectiveness of a mixture of CCs versus single or double CCs are limited. In this study, we evaluated and compared the effect of single CC, double CCs and the soil health mix (SHM, a combination of five species recommended by the United States Department of Agriculture (USDA)) on soil quality and crop production. The study was conducted at the Research and Education Center in Milan, TN. Soil samples were collected from a 3-year and a 15-year long corn-soybean rotation system integrated with several treatments of CCs. The results showed an increase in soybean yield after 3-years of SHM treatment. SHM treatment also showed a significant increase in the soil moisture content. Soil organic carbon, total nitrogen and water extractable nitrate content did not change across the cover crop treatments. However, the cereal rye/hairy vetch double species treatment and the SHM increased the inorganic nitrogen content. These differences were observed only from the short-term experiment. Soil properties from the long-term experiment were statistically similar across the single species CC treatments in this experiment. This indicates that SHM, a combination of five CCs, has a good potential for enhancing soil quality and crop yield in the western TN.

Introduction

Cover cropping is a common conservation agricultural practice that reduces soil erosion, improves soil and water quality, and suppresses weed growth (Hobbs et al., 2008). The Soil Science Society of America (SSSA) defines cover crops (CCs) as close-growing crops that provide soil protection, seeding protection and soil improvement between periods of normal growing season (SSSA, 2008).

Cover crops are grown to reduce the fallow period in the cropping systems. The unprotected soil between the growing seasons increases the risk of erosion and nutrient runoff, which may decrease the overall soil quality with a subsequent decrease in crop productivity in the following growing seasons (Kaspar and Singer, 2011). When grown in winter seasons, CCs behave as a regular crop and alter soil physical properties through the formation of pores and aggregates (Blanco-Canqui et al., 2015; Drury et al., 2003; Papadopoulos et al., 2006). With roots penetrating the compacted soil layer, CCs can also alleviate or reduce soil compaction (Blanco-Canqui et al., 2012; Chen and Weil, 2010). They can provide additional biomass to increase soil organic carbon (SOC) concentration and microbial activity (Blanco-Canqui et al., 2013; Dabney et al., 2001). The aboveground biomass and belowground root system of CCs can reduce soil from wind and water erosion (Blanco-Canqui et al., 2013). The increased soil aggregation improves soil hydraulic properties such as infiltration rate and water retention (Blanco-Canqui et al., 2011; Keisling et al., 1994).

Another important function of CC is to scavenge nutrients from the soil during the off-season thus reducing the loss of nutrients by leaching and runoff (Kaspar and Singer, 2011). Leguminous CC species can also contribute to additional nitrogen (N) to the crop by fixing atmospheric N (Hauggaard-Nielsen et al., 2010). From a biological perspective, CCs improve organic matter input through above and belowground residues, which favors macroand microfauna activities (Reddy et al., 2003). Studies have shown higher earthworm and microarthropods population after integrating CCs (Reeleder et al., 2006). CCs also have been shown to suppress weeds, plant pathogens and nematodes through different mechanisms including allelopathy (Keating, 1999). These benefits can enhance the overall soil quality and system sustainability.

Soil health, previously called soil quality, is defined by the Natural Resources Conservation Service (NRCS) of the United States Department of Agriculture (USDA) as "the capacity of a soil to function." Doran and Zeiss (2000) expanded this definition as "the capacity of a soil to function, within ecosystem and land use boundaries, to sustain productivity, maintain environmental quality, and promote plant and animal health." Dabney et al. (2001) concluded that cover crops increase soil quality through improvement in soil physical, chemical and biological properties, including soil hydraulic conductivity, aggregation, cation exchange capacity and organic matter content. Recently, with more emphasis on soil biological properties, the term soil health is becoming more and more popular.

The enhanced soil quality attributes including additional N supply, improved aggregation and reduced nutrient runoff, and other benefits (e.g. weed suppression) from cover cropping, as discussed above, may contribute to increase in crop yield. Enhanced soil water storage by cover cropping may also impact positively to yield (Welch et al., 2016). In regions with higher precipitation (>800 mm), CCs often increase soil water storage capacity and benefit crop production (Balkcom and Reeves, 2005). N-fixing CC species show a stronger effect in increasing crop yields than CC species with no N–fixing capacity (Blanco-Canqui et al., 2015). In Tennessee (TN), a 33-year long-term study showed yield improvement in cotton (*Gossypium hirsutum* L.) by continuous hairy vetch *(Vicia villosa* Roth) planting (Mbuthia et al., 2015). The same result was found in a 12-year study in TN on a cotton-corn (*Zea* *mays*) rotation experiment with hairy vetch as CC (Ashworth et al., 2016). On the other hand, a few studies showed decreased yield or no change in yield by cover cropping relative to no-cover control because of reduced N availability, reduced water storage and allelopathic effect (Ewing et al., 1991; Johnson et al., 1998; Nielsen and Vigil, 2005; Salmerón et al., 2010).

In general, the benefits from CCs are strongly influenced by several factors including CC species, soil types and management strategies, and the fact that it may take several years to notice the benefits (Andraski and Bundy, 2005; Decker et al., 1994). Different CC species have different functions in soil. Leguminous CCs [hairy vetch and crimson clover (*Trifolium incarnatum* L.)] improve nutrient availability to crops and non-leguminous CCs [(radish (*Raphanus sativus*) and cereal rye (*Secale cereale* L.)) scavenge nutrients from deeper soil layers, control loss of soil and nutrients from landscapes and improve soil organic matter content (Blanco-Canqui et al., 2015). Multi-species mixtures of CCs have the potential to offer multiple benefits to the system (Tosti et al., 2014). In recent years, there has been an increasing interest in using multi-species CC mixtures. The USDA-NRCS is promoting a multi-species mixture called soil health mix (SHM) through its Environmental Quality Incentives Program (EQIP) (USDA, 2016a). A recent press release by USDA-NRCS explicitly stated that CC mixtures work better than single or double species CCs (USDA, 2016b). However, there is a lack of scientific data on the performance of this multi-species mix in relation to common single and double species CCs in different regions of US.

This study was conducted to evaluate and compare the effect of different CC species (single-, double- and multi-species) on soil properties and crop production. We hypothesized that the cropping systems integrated with multi-species mixture exhibit greater crop yield and favorable soil properties as compared to systems integrated with single and double-species CCs.

Materials and methods

Field design and soil sampling

This study was conducted at the University of Tennessee's Research & Education Center (REC) in Milan, TN. The soil is classified as a Lexington silt loam (fine-silty, mixed, thermic, Ultic Hapludalf). The mean annual rainfall of the region is 1361 mm.

This study used two existing field experiments on CCs at REC, Milan. The first experiment was established in 2002 and the second experiment was established in 2013. Both experiments were under no-till corn-soybean rotation with the same management.

For both experiments, the experimental design was a randomized complete block (RCB) design. In the short-term experiment, there are six CC treatments including single, double and multi-species (Table B-1). Each treatment had four replications. In the long-term experiment, there are four CC treatments each with three replications (Table B-2).

Table B-1 Cover crop treatments in the short-term experiment

Treatments	***Species***	
A (CR/CC)	Cereal Rye Crimson Clover	
B (CR/HV)	Cereal Rye Hairy Vetch	
C (W)	Wheat (*Triticum aestivum* L.)	
D (CR)	Cereal Rye	
E (SHM)	Soil Health Mix	Cereal Rye Whole Oats (*Avena sativa* L.) Purple Top Turnips (*Brassica napus* L var) Daikon Radish Crimson Clover
F (Control)	Control—no cover crop	

Table B-2 Cover crop treatments in the long-term experiment

Treatments	***Species***
A (AP)	Austrian Winter Pea (*Pisum sativum*)
B (HV)	Hairy Vetch
C (W)	Wheat
D (Control)	Control—no cover crop

Soil samples were collected on October 18th, 2016 after the harvest of the main crop (soybean) and just before the seeding of the winter CC. The sampling depth was 15 cm. Samples were collected using stainless steel probes (2.5 cm in diameter) from the two field trials, along with samples collected from nearby undisturbed woods and grassland to represent a relative ‘natural’ soil with no recent cropping history. From each plot, approximately 10-15 subsamples were randomly collected and then mixed into one composite sample. Multiple samples were also collected from each of the 7 pristine locations. Composite samples were stored in plastic Ziploc bags and placed in a cooler with blue ice while transporting to the laboratory. A subsection of each sample was air-dried and passed through a 2-mm sieve for analyzing physical and chemical properties, and the rest was stored in 4°C for analyzing biological properties.

Measurement of crop yield

Harvest was conducted using a plot harvester. Yield was measured using a yield monitor. Soybean yields were converted to Mg/ha using the following equation 1 (Johanns, 2013):

Yield (Mg/ha) = [(Bushels soybean/acre) * (27.22kg/bushels soybean) ÷ (0.404hectare/acre)] ÷ (1000kg/Mg) **Equation 1**

Measurement of soil properties

All chemicals and materials used were purchased from Fisher Scientific. All reagents were prepared using type 1 ultrapure water.

Soil pH

Soil pH was measured on a 1:2 soil/water suspension (Thomas, 1996). 10 g of air-dried soil was weighed and mixed with 20 ml of deionized (DI) water. Soil pH was measured using a Denver Instrument Model 250 pH meter after shaking and setting.

Soil moisture content

Soil moisture content was measured by gravimetric method (Gardner, 1986). Approximately 10 g soil sample was weighed into tared tin cups and dried in an oven at 105℃ for 24 hours. Samples were removed, cooled in a desiccator, and dry weight recorded. Soil moisture content was calculated using equation 2:

$$\frac{Mass_{Wet\ soil} - Mass_{Dry\ soil}}{Mass_{Dry\ soil}} \qquad \textbf{Equation 2}$$

Extractable nutrients

Phosphorous (P), potassium (K), calcium (Ca) and magnesium (Mg) were extracted using Mehlich-1 extractant (Savoy, 2009). Approximately 5 g air-dry soil was weighed into 50 ml centrifuge tubes. 20 ml of Mehlich-1 extractant was added to each tube. Tubes were capped and shaken at 180 oscillations/min for 5 minutes. The suspensions were filtered, filtrates collected and stored in the refrigerator for the analysis of P, K, Ca and Mg using a Perkin-Elmer 5300 & 7300 DV Inductively Coupled Plasma (ICP) unit.

Soil organic carbon and total nitrogen

Soil organic carbon and total nitrogen (T-N) were measured by combustion method using a Thermo Flash EA 1112 NC combustion analyzer. Air-dried samples were powdered using a mortar and pestle before analysis (Nelson and Sommers, 1996).

Soil labile carbon

Soil labile carbon, also called permanganate oxidizable carbon (POXC), was determined using the method developed by Weil et al. (2003). Soil samples were first oxidized by potassium permanganate ($KMnO_4$). The remaining $KMnO_4$ was immediately measured by a spectrophotometer. A standard curve was developed to correlate the solution concentration and the absorbance. The POXC content (mg/kg) was calculated using equation 3:

$$[0.02 - (\mathrm{a} + \mathrm{b} \times \mathrm{Abs})] * 9000 * \frac{0.02}{Wt} \qquad \textbf{Equation 3}$$

Where: 0.02 (mol/L) = initial solution concentration;

a = intercept of the standard curve

b = slope of the standard curve

Abs = absorbance of unknown

9000 (mg C/mol) = milligrams of carbon oxidized by 1 mole of $KMnO_4$

Wt = weight of air-dried soil sample in kg

Soil microbial biomass carbon

Soil microbial biomass carbon was analyzed using the chloroform fumigation-extraction method (Vance et al., 1987). 10g of fresh soil samples were fumigated with chloroform in the dark for 48 hours. Both fumigated and non-fumigated (control) samples were then extracted with 0.5M potassium sulfate (K_2SO_4). Total dissolved carbon was measured using a Shimadzu Total Organic Carbon Analyzer (TOC-5000). The difference between C in fumigated and non-fumigated samples is the chloroform labile C (EC). Microbial biomass C was calculated as EC divided by *k*, a constant, estimated at 0.45 (Beck et al., 1997).

Soil inorganic nitrogen

Soil inorganic nitrogen was analyzed using the extraction method modified based on the protocol described by Mulvaney (1996). Four grams of air-dry soil was extracted with 40 ml of 2M potassium chloride (KCl). NH_4-N and NO_3-N were measured from the extract using a Skalar San++ Continuous Flow Analyzer (CFA).

Soil water extractable nitrate nitrogen

Soil water extractable nitrate (WE-NO_3) was analyzed using the extraction method described by Haney et al. (2012). Four grams of air-dry soil was extracted with 40 ml of DI water. NO_3-N were measured using the Skalar CFA.

Statistical analysis

The effects of CC treatments on soil physical, chemical and biological properties and crop yield were evaluated by analysis of variance (ANOVA) based on Generalized Linear Model (GLM) in SAS 9.4 (SAS Institute, Cary, NC, 2013). Fisher's protected least significant difference (LSD) was used to determine significant differences among treatment means at $P \leq 0.05$.

Results and discussion

Soil pH

Soil pH ranged from 5.52 to 5.84 with an average of 5.64 in the short-term experiment, and 5.48 to 5.74 with an average of 5.61 in the long-term experiment (Figure B-1). Overall, cover cropping decreased soil pH in the short-term experiment in comparison with no cover control with the CR/HV treatment showed significantly lower pH than that of the control. No other statistically significant differences were found among treatments in either experiment. Previous studies also reported that leguminous CC might reduce soil pH (Hargrove, 1986; Jokela et al., 2009). At the grassland site, samples from the upslope location showed a slightly higher soil pH (5.54) than that from the downslope location (5.41). At the woodland site, samples from the three floodplain locations showed a relative lower soil pH (5.05) than from the two non-floodplain locations (5.66) (Table B-3).

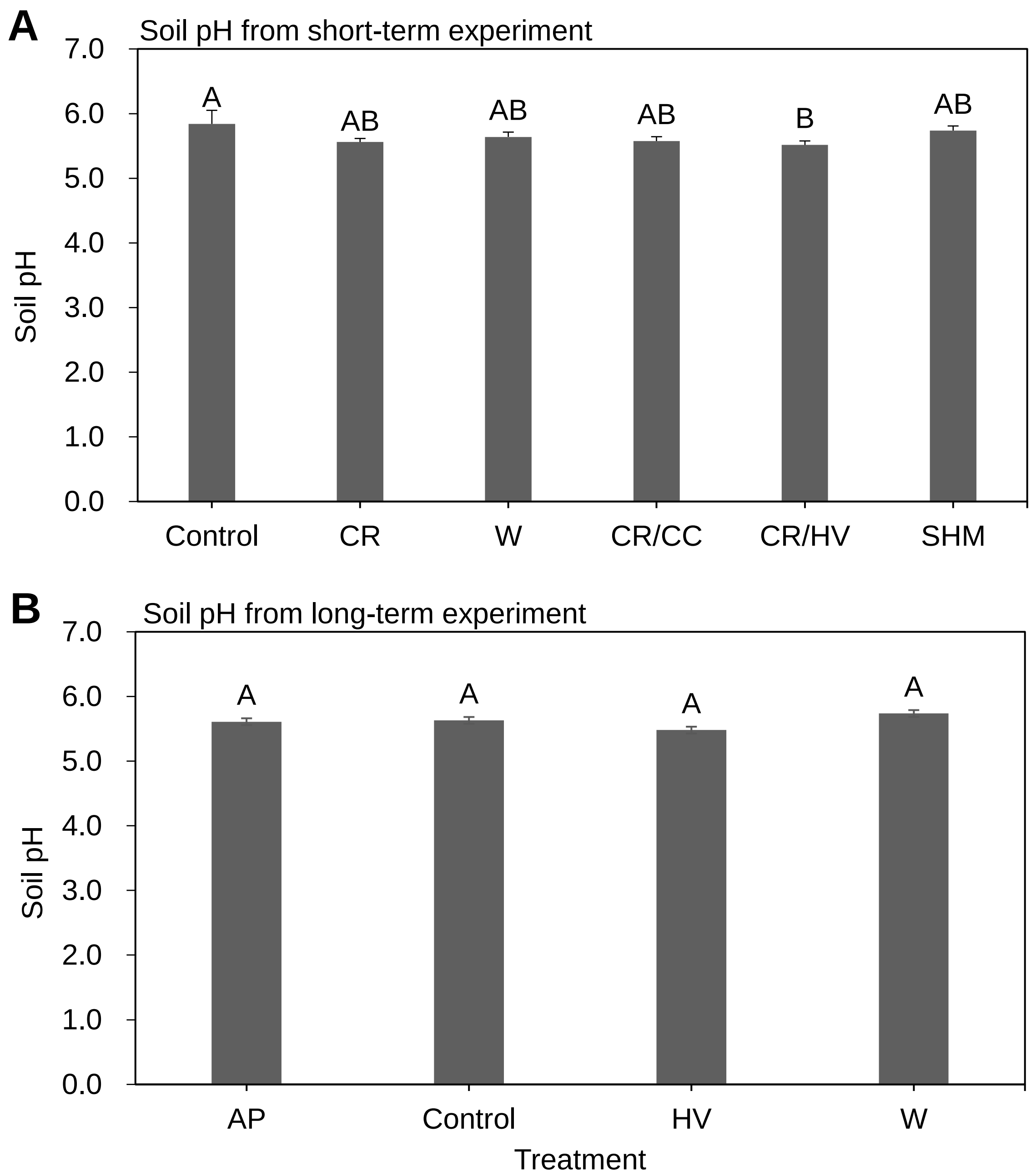

Figure B-1 Soil pH in response to cover crop treatments from the short- and long-term experiments. (A) Soil pH from the short-term experiment, (B) Soil pH from the long-term experiment. Different letters denote statistically different means at $P < 0.05$ and error bars represent standard error of the mean.

Soil gravimetric moisture content

Gravimetric soil moisture content ranged from 17.0 to 20.8% across the CC treatments in the short-term experiment and 21.2 to 23.0% across the CC treatments in the long-term experiment (Figure B-2). In the short-term experiment, the average soil moisture contents of the control, single-species, double-species and multi-species treatments were 17.0, 19.0, 19.2 and 20.4%, respectively. The SHM treatment showed a significantly higher soil moisture content than the control. Past studies demonstrated increased soil water retention as a result of cover cropping (Blanco-Canqui et al., 2012; Kaspar et al., 2001; Nielsen et al., 2015; Unger and Vigil, 1998). In regions receiving higher precipitation, growing CC could be a promising water storage strategy that could benefit crop production if a subsequent drought occurs (Blanco-Canqui et al., 2015).

In the long-term experiment, soil moisture content was found to be higher than that in the short-term experiment. This could be due to the different crops grown on these plots in 2016, which is corn in the long-term plot and soybean in the short-term plots. More crop residues remained on the soil surface from the corn may have caused higher retention of moisture in the long-term plots. However, we did not find significant differences in soil moisture content across the CC treatments in the long-term experiment. The results suggest that the increased diversity of CC species (double and multi-species), as in the short-term experiment, has a positive effect on soil water retention. The grassland site had an average moisture of 15.8% with higher moisture at the downslope (16.7%) than at the upslope (14.9%). Among the five woodland sites, three around the floodplain showed lower moisture content of 10.21% compared to the other two (13.3%) (Table B-4).

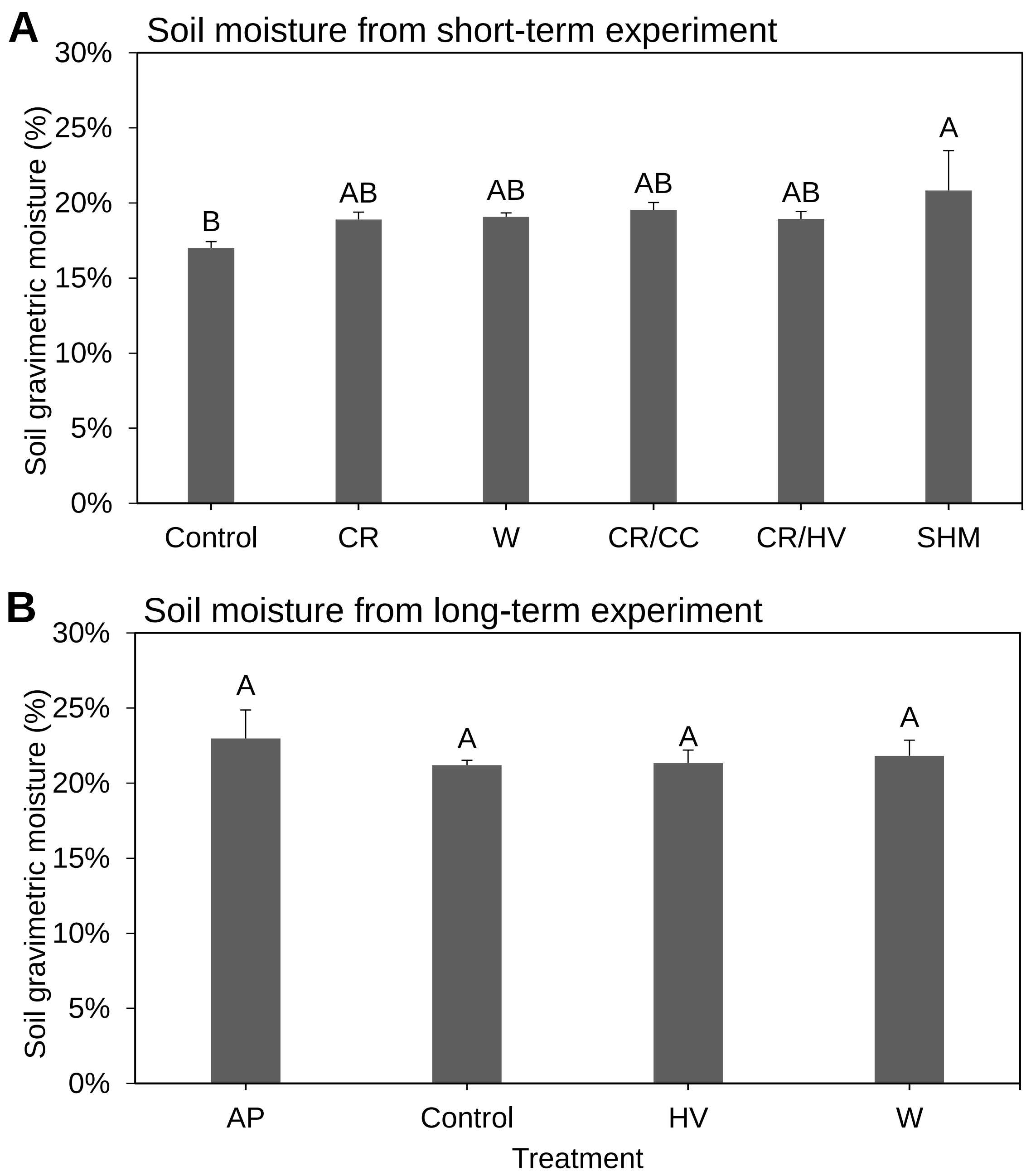

Figure B-2 Soil gravimetric moisture content in response to cover crop treatments in short- and long-term experiments. (A) Soil moisture content from the short-term experiment, (B) Soil moisture content from the long-term experiment. Different letters denote statistically different means at P < 0.05 and error bars represent standard error of the mean.

Soil extractable nutrients

Soil available nutrients were extracted using Mehlich-1 reagent. CR/HV and HV treatments showed numerically lower P levels than the control in the short and long-term experiments, respectively (Table B-3). Some authors have reported CC may reduce the available P because of their uptake and transformation of available P into organic form (Villamil et al., 2006), but this was not observed in either of these experiments, as the differences among CC treatments were not statistically significant. The woodland soils had an average P level of 15.3 mg kg^{-1}. The grassland soils had an average P level of 3.45 mg kg^{-1} with the grassland downslope had a slightly higher P content (3.67 mg kg^{-1}) than the grassland upslope (3.23 mg kg^{-1}) (Table B-4).

Soil K concentration ranged from 52.0 to 69.0 mg kg^{-1} across the CC treatments in the short-term experiment with the CR/CC and SHM treatments showed the highest and CR showed the lowest concentration (Table B-3). In the long-term experiment, K level ranged from 54.8 to 69.8 mg kg^{-1} with no significant differences across the CC treatments. The extractable K of natural soil is shown in Table B-4. Soils from the three woodland floodplain locations showed a lower K level (52.4 mg kg^{-1}) than from the two non-floodplain location (82.1 mg kg^{-1}), and the grassland downslope showed relatively higher K level than the grassland upslope (82.1 and 89.5 mg kg^{-1}).

The CR treatment showed the highest extractable calcium (Ca) content (703 mg kg^{-1}) which was significantly different from the W treatment (691 mg kg^{-1}) in the case of short-term experiment (Table B-3). The extractable Ca from the long-term experiments was not statistically different across the CC treatments. The Ca concentration ranged from 667 to 680 mg kg^{-1} for the samples from the grassland location and from 565 to 694 mg kg^{-1} for the samples from the woodland location (Table B-4).

Soil extractable magnesium (Mg) concentration ranged from 71.6 to 87.8 mg kg^{-1} and 84.3 to 119 mg kg^{-1} in the short and long-term experiments,

Table B-3 Soil extractable nutrients in response to CC treatments in the short and the long-term experiments. (A) Soil extractable nutrients from the short-term experiment. (B) Soil extractable nutrients from the long-term experiment. Numbers in parentheses are (±1) standard error of the mean, and lower case alphabets denote statistical significance at P < 0.05. Bolded ANOVA tables show statistical significance at P < 0.05.

A	**Soil extractable nutrient in short-term experiment (mg kg^{-1})**			
Treatment	P	K	Ca	Mg
Control	12.5(1.40)a	65.5(5.94)ab	694(4.56)ab	79.5(11.44)a
CR	10.8(1.15)a	52.0(1.00)b	703(2.35)a	71.6(17.56)a
W	12.3(2.50)a	61.5(2.40)ab	691(3.60)b	75.4(8.74)a
CR/CC	15.7(1.54)a	69.7(1.88)a	700(3.71)ab	87.8(8.87)a
CR/HV	9.9(2.51)a	57.4(2.80)ab	696(4.29)ab	85.1(10.14)a
SHM	11.8(2.74)a	68.9(0.60)a	699(4.87)ab	87.0(16.04)a
ANOVA Table (LSD Protected p<0.05)				
Cover crop	0.4961	**0.0036**	0.3420	0.9194

B	**Soil extractable nutrient in long-term experiment (mg kg^{-1})**			
Treatment	P	K	Ca	Mg
AP	11.4(6.18)a	58.2(2.10)a	688.9(15.00)a	103.2(37.50)a
Control	10.1(1.78)a	69.1(9.74)a	690.3(0.38)a	119.4(6.31)a
HV	8.1(0.82)a	54.8(5.60)a	681.2(9.79)a	84.3(31.10)a
W	13.0(4.08)a	69.8(4.78)a	684.1(8.39)a	105.2(26.90)a
ANOVA Table (LSD Protected p<0.05)				
Cover crop	0.8334	0.2859	0.9043	0.8480

Table B-4 Soil properties of the natural sites

	location		pH	Moisture (%)	P (mg kg-1)	K (mg kg-1)	Ca (mg kg-1)	Mg (mg kg-1)	SOC (g kg-1)	POXC (mg kg-1)	MBC (mg kg-1)	T-N (mg kg-1)	Inorganic N (mg kg-1)	WE-NO3 (mg kg-1)
Grass land	upslope		5.54	14.9	3.23	77.8	667	100	16.3	417	327	1734	6.19	3.88
	downslope		5.41	16.7	3.67	101.0	680	197	21.2	413	572	2095	7.46	1.93
Wood land	Flood plain	Near road	5.16	9.6	4.76	49.2	575	136	17.7	351	397	1632	5.99	0.12
		Near river	5.04	9.2	19.30	57.9	680	177	10.5	517	269	1021	20.5	3.34
		Away from river	4.94	11.9	12.00	50.2	565	143	14.0	491	307	1119	8.00	1.61
	near army area-1		5.68	14.3	14.50	93.8	694	82	22.5	560	108	1949	12.1	4.14
	near army area-2		5.63	12.2	22.50	70.3	687	107	17.9	535	330	1627	8.50	5.22

respectively, with no statistical differences across the CC treatments in both experiments (Table B-3). Average Mg content of grassland and woodland soils were 148 and 129 mg kg^{-1}, respectively (Table B-4). At the grassland site, the downslope showed about two times more soil Mg content than the upslope. The woodland samples from the floodplain had higher average Mg level than from the non-floodplain locations.

Overall, statistically significant differences were only found for K and Ca in short-term experiment under CC treatments compared to the control. The relatively deep sampling depth (0-15cm) could be the reason for the lack of statistically significant differences among treatments because CCs return nutrients to the top few centimeters of the soil in these no-tillage systems.

Soil organic carbon

Soil organic carbon (SOC) is important for sustainable crop production. Cover cropping has the potential to increase SOC by inputting more above and belowground biomass to soil (Blanco-Canqui et al., 2013). In this study, SOC was not significantly different among CC treatments in both the experiments (Figure B-3 A, B). SOC content in the short-term experiment ranged from 10.1 to 11.4 g kg^{-1} (Figure B-3A) and that in the long-term experiment ranged from 9.8 to 10.4 g kg^{-1} (Figure B-3B).

We measured relatively labile fractions of SOC such as permanganate oxidizable carbon (POXC) and microbial biomass carbon (MBC) (Vance et al., 1987; Weil et al., 2003). Jokela et al. (2009) and Steele et al. (2012) reported significantly greater soil POXC level in 0-5 and 0-7cm soil depth, respectively, using winter rye in comparison to no-cover, while no difference occurred in total SOC. Our study, however, found no significant difference in POXC across the treatments in both the short-term (values ranged from 311 to 346 mg kg^{-1}, Figure B-3C) and the long-term (values ranged from 285 to 359 mg kg^{-1}, Figure B-3D) experiments. Soil MBC also showed no significantly different results across the CC treatments in both short- and long-term

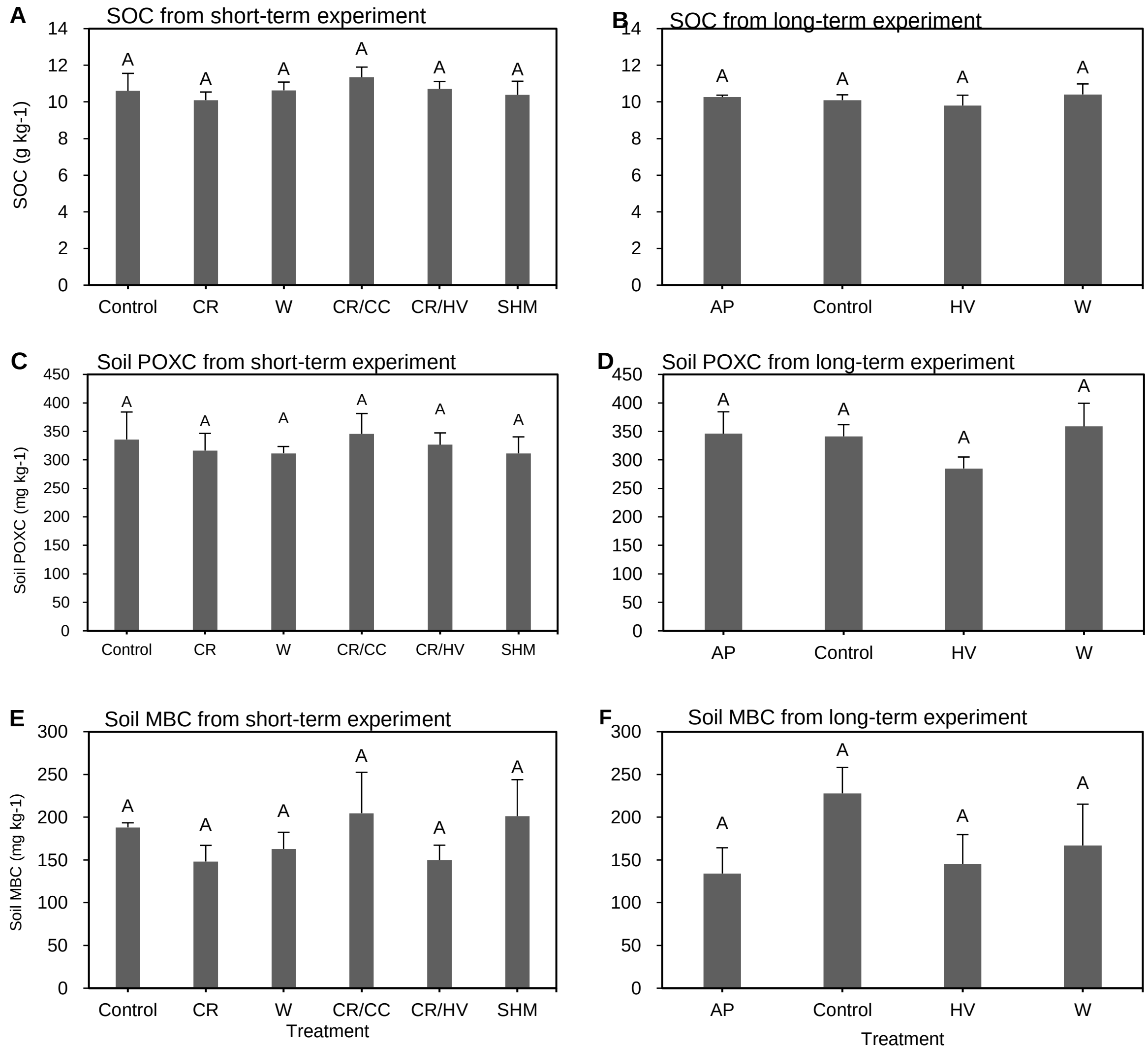

Figure B-3 SOC, POXC and MBC in response to CC treatments from the short and the long-term experiment. A and B - SOC from the short and the long-term experiments. C and D - POXC from the short and the long-term experiments. E and F- MBC from the short and the long-term experiments. Similar lowercase letters over the bars denote statistically similar means at P < 0.05 and error bars represent standard error of the mean.

experiments with values ranged from 150 to 205 mg kg^{-1} in short-term experiment (Figure B-3E) and 134 to 228 mg kg^{-1} in the long-term experiment (Figure B-3F).

It was surprising to find that no differences were found even in the labile C fractions possibly due to sampling time. We collected samples in October 2016, which was after the completion of the main cropping season. A spring sampling, coinciding with the termination of the CC, may have responded better to the labile C pools. Lack of response of SOC to CC treatments could also be attributed to the unique hot and humid climatic condition in TN, which favors accelerated C mineralization (Davidson and Janssens, 2006; Fang et al., 2005). Additionally, the CC treatments return biomass to the surface soil of these no-tillage systems, and the relatively deep sampling depth (0-15cm) that we chose could have diluted the effect on the first few centimeters of the soil layer.

For the double and multi-species treatments, the relatively short experimental time (3 years) may be the main reason that no difference was discovered. It may take a longer time for carbon content enhancement considering the effect of CC may not be detectable in the first several years after establishment (Acuña and Villamil, 2014; Blanco-Canqui et al., 2014). Natural soils showed higher values of SOC, POXC and MBC than that of cropland soils (Table B-4) which are expected due to the long-term undisturbed nature of these soils. At the grassland site, the downslope soil had higher SOC and MBC level (21.2 g kg^{-1} and 572 mg kg^{-1}, respectively) than the upslope soil, while POXC remained same. The average SOC, POXC and MBC contents in woodland site were 16.5 g kg^{-1}, 491 and 282 mg kg^{-1}, respectively.

Soil nitrogen

Nitrogen is one of the most important nutrients for plant growth, and cover cropping is a demonstrated strategy to influence soil N balance by fixing atmospheric N, and reducing leaching and erosion loss of N (Dabney et al., 2001; Dabney et al., 2010). The soil total nitrogen (T-N) level in the short-term experiment ranged from 1026 to 1158 mg kg^{-1} (Figure B-4A) and that in the long-term experiment ranged from 1030 to 1073 mg kg^{-1} (Figure B-4B) with no statistical differences across the treatments. Although T-N did not show significant differences among CC treatments, soil inorganic N (NH_4 + NO_3) varied significantly in the short-term experiment. The average inorganic N level of control, single species, double species and multi-species treatments was 15.5, 16.3, 19.4 and 19.4 mg kg^{-1}, respectively (Figure B-4C) in the short-term experiment with significantly higher inorganic N content observed for double and multi-species treatments compared to the control. The CR/HV treatment had the highest inorganic N level, followed by the SHM treatment, which was 32 and 25% higher than that of control. These results indicate that increasing the number of CC species can enhance the soil inorganic N pool. In the long-term experiment in which only single species CC was included, no significant difference in inorganic N was observed although all the three single species CCs such as HV, W and AP showed numerically higher inorganic N compared to the control (Figure B-4D). Though inorganic N level varied across the CC treatment, total and labile C level were not significantly affected by the treatments (Figure B-3). We speculate that the increased availability of inorganic N might have stimulated the microbial population that resulted in enhanced microbial utilization of organic C from the soil. The potential risk of N loss through leaching was measured as water extractable nitrate nitrogen (WE-NO_3). No statistically significant differences were found among treatments in the short-term experiments (Figure B-4E). This shows that though the CC mixtures significantly improved total inorganic N content of soil, they did not increase the potential risk of N loss through leaching. In the long-term experiment, W treatment showed significantly higher WE-NO_3 level than the other treatments (Figure B-4F).

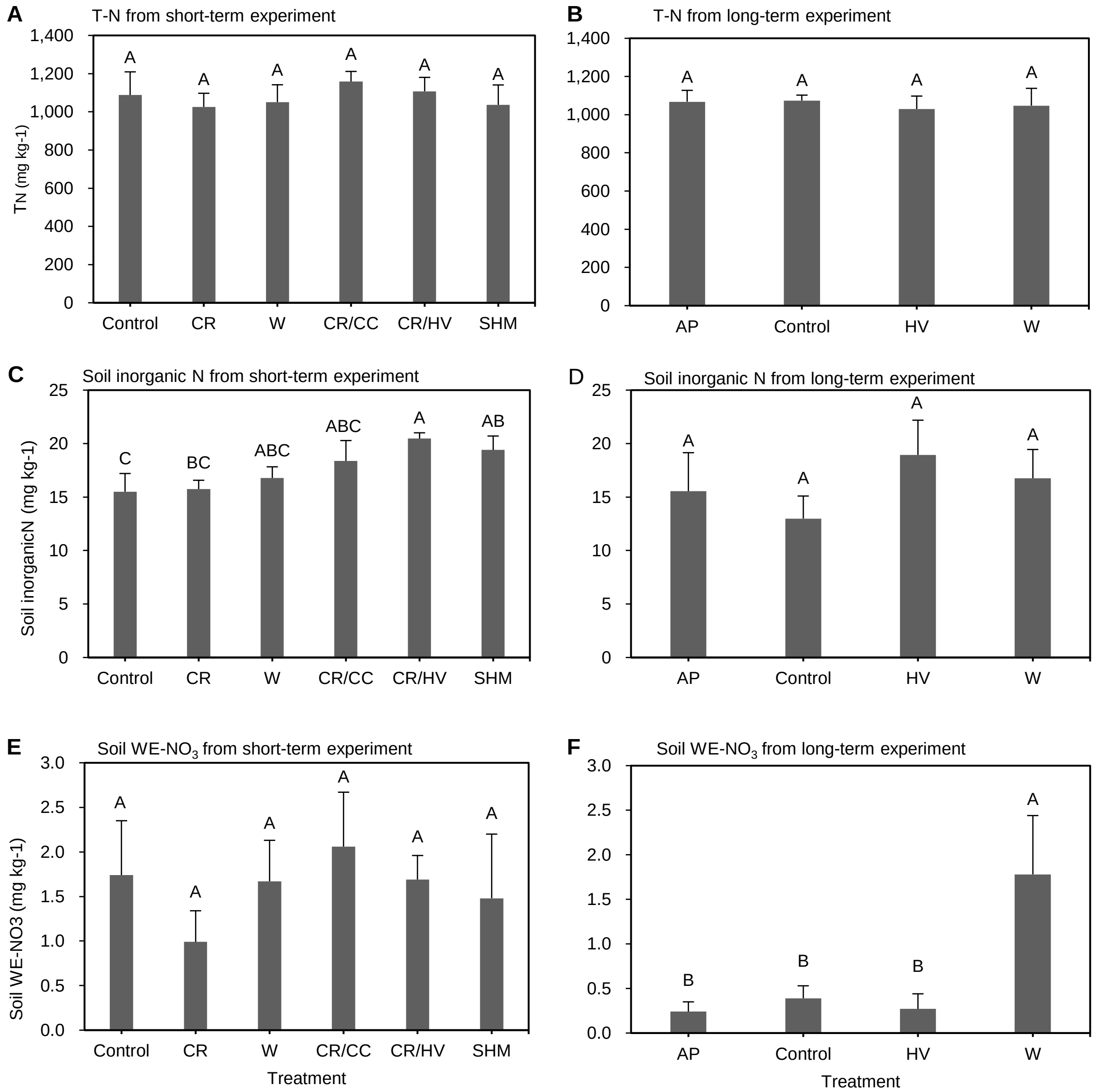

Figure B-4 TN, inorganic N and WE-NO3 in response to CC treatments from the short and the long-term experiments. A and B - TN from the short and the long-term experiments. C and D – Inorganic N from the short and the long-term experiments. E and F - WE-NO3 from the short and the long-term experiments. Different lowercase alphabets over the bars denote statistically different means at P < 0.05 and error bars represent standard error of the mean.

Crop yield

The 2016 soybean yield from the short-term experiment is shown in Figure B-5. The overall average yield across all cover crop treatments was 4.10 Mg ha^{-1}, which was 35% higher than the 2016 state average yield (USDA, 2017). The average yield of control, single, double and multi-species was 3.96, 4.02, 4.12 and 4.55 Mg ha^{-1}, respectively. Although the yield from single and double species treatments was numerically higher than that of the control, the differences were not statistically significant. The multi-species treatment (SHM) showed a significantly higher yield response, which was 15% higher than the control. We also observed increased gravimetric soil moisture content and soil inorganic N for the SHM treatment. Therefore, the yield increase could be due to the favorable effect of soil moisture (Figure B-2) and inorganic N status (Figure B-4C). It should be noted that the experimental site experienced a drought during the growing season especially in June (UNL, 2017) as shown in Table B-5 (NCEI, 2017). Under such a water limited condition, the increased soil water content under the SHM treatment might have favorably influenced the crop yield.

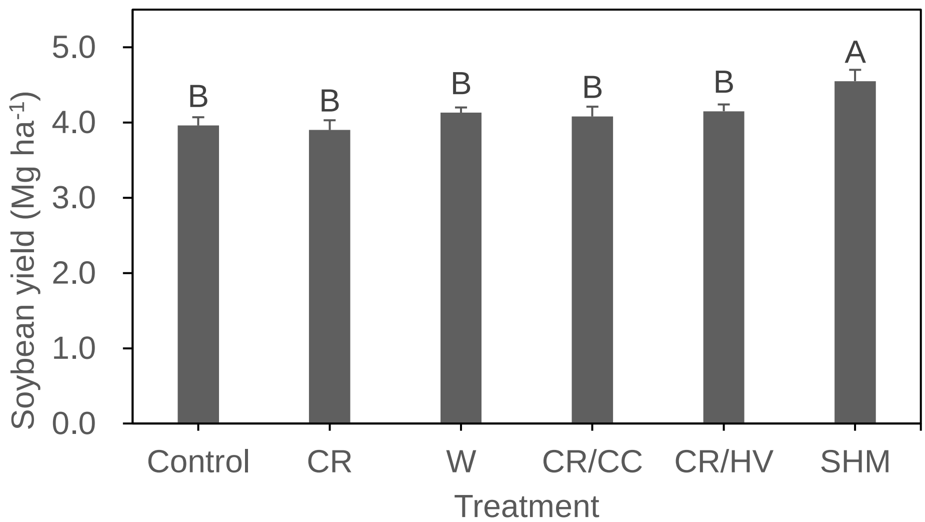

Figure B-5 2016 Soybean yield in response to the CC treatments in the short-term experiment. Different letters denote statistically different means at P < 0.05 and error bars represent standard error of the mean.

Table B-5 Climate data of Milan from 2013 to 2016

	Mean Temperature (°F)				Total precipitation (Inches)			
	2013	2014	2015	2016	2013	2014	2015	2016
Jan	40.1	30.6	36.0	35.2	6.93	2.63	1.18	1.79
Feb	40.7	36.1	30.4	42.7	4.04	5.41	5.69	5.07
Mar	43.0	43.3	47.1	54.0	5.98	5.51	4.43	12.19
Apr	57.6	58.8	61.3	60.5	10.86	5.93	4.43	2.86
May	67.0	69.9	68.8	66.4	9.75	2.70	10.47	4.35
Jun	76.8	76.7	78.0	79.1	5.43	9.06	6.13	1.68
Jul	76.0	74.2	81.4	82.4	6.95	2.07	3.33	3.44
Aug	76.6	78.9	75.7	80.5	2.90	5.18	4.79	6.15
Sep	72.4	70.5	72.6	75.1	5.89	1.85	2.56	1.04
Oct	59.9	61.4	60.7	66.2	4.35	3.50	3.47	0.58
Nov	45.3	42.3	54.1	53.2	3.41	2.27	9.35	3.30
Dec	39.5	42.9	51.0	No data	4.20	4.40	5.48	7.91

Conclusion

Growing multi-species CCs can potentially improve sustainable crop production by providing multiple benefits to the soil and the environment. However, studies on the effect of multiple species CCs on soil properties and crop production is scarce. In this study, we compared the effect of CCs on crop and soil responses by using two CC field trials on corn-soybean rotational systems. The first trial is three years long and it includes several single, double and multi-species CCs, and the second trial is fifteen years long with only single CC species. Although most soil properties measured did not show significant differences across the CC treatments, crop yield and gravimetric soil moisture content were significantly higher for the multi-species soil health mix (SHM). This indicates that the enhanced soil moisture content could be the driver for the yield increase. Double and multi-species CCs improved inorganic N level of soil, but not water extractable nitrate-N (WE-NO_3), which indicated that using double and multi-species CCs can increase soil inorganic N level to benefit crop growth without increasing the potential risk of N loss through leaching. Overall SHM exhibited promise for enhancing soil quality and crop production.

References

[1] Acuña, J. and M.B. Villamil. 2014. Short-term effects of cover crops and compaction on soil properties and soybean production in Illinois. Agronomy Journal 106: 860-870.

[2] Andraski, T.W. and L.G. Bundy. 2005. Cover crop effects on corn yield response to nitrogen on an irrigated sandy soil. Agronomy Journal 97: 1239-1244.

[3] Ashworth, A.J., F.L. Allen, A.M. Saxton and D.D. Tyler. 2016. Long-Term Corn Yield Impacted by Cropping Rotations and Bio-Covers under No-Tillage. Agronomy Journal 108: 1495-1502.

[4] Balkcom, K.S. and D.W. Reeves. 2005. Sunn-hemp utilized as a legume cover crop for corn production. Agronomy journal 97: 26-31.

[5] Beck, T., R. Joergensen, E. Kandeler, F. Makeschin, E. Nuss, H. Oberholzer and S. Scheu. 1997. An inter-laboratory comparison of ten different ways of measuring soil microbial biomass C. Soil Biology and Biochemistry 29: 1023-1032.

[6] Blanco-Canqui, H., M. Claassen and D. Presley. 2012. Summer cover crops fix nitrogen, increase crop yield, and improve soil–crop relationships. Agronomy journal 104: 137-147.

[7] Blanco-Canqui, H., R.B. Ferguson, V.L. Jin, M.R. Schmer, B.J. Wienhold and J. Tatarko. 2014. Can Cover Crop and Manure Maintain Soil Properties After Stover Removal from Irrigated No-Till Corn? Soil Science Society of America Journal 78: 1368-1377.

[8] Blanco-Canqui, H., M.M. Mikha, D.R. Presley and M.M. Claassen. 2011. Addition of cover crops enhances no-till potential for improving soil physical properties. Soil Science Society of America Journal 75: 1471-1482.

[9] Blanco-Canqui, H., C.A. Shapiro, C.S. Wortmann, R.A. Drijber, M. Mamo, T.M. Shaver and R.B. Ferguson. 2013. Soil organic carbon: The value to soil properties. Journal of Soil and Water Conservation 68: 129A-134A.

[10] Blanco-Canqui, H., T.M. Shaver, J.L. Lindquist, C.A. Shapiro, R.W. Elmore, C.A. Francis and G.W. Hergert. 2015. Cover crops and ecosystem services: Insights from studies in temperate soils. Agronomy Journal 107: 2449-2474.

[11] Chen, G. and R.R. Weil. 2010. Penetration of cover crop roots through compacted soils. Plant and Soil 331: 31-43.

[12] Dabney, S., J. Delgado and D. Reeves. 2001. Using winter cover crops to improve soil and water quality. Communications in Soil Science and Plant Analysis 32: 1221-1250.

[13] Dabney, S.M., J.A. Delgado, J.J. Meisinger, H.H. Schomberg, M.A. Liebig, T. Kaspar, J. Mitchell and W. Reeves. 2010. Using cover crops and cropping systems for nitrogen management. Advances in nitrogen management for water quality: 231-282.

[14] Davidson, E.A. and I.A. Janssens. 2006. Temperature sensitivity of soil carbon decomposition and feedbacks to climate change. Nature 440: 165-173.

[15] Decker, A.M., A.J. Clark, J.J. Meisinger, F.R. Mulford and M.S. McIntosh. 1994. Legume cover crop contributions to no-tillage corn production. Agronomy journal 86: 126-135.

[16] Doran, J.W. and M.R. Zeiss. 2000. Soil health and sustainability: managing the biotic component of soil quality. Applied soil ecology 15: 3-11.

[17] Drury, C., C. Tan, W. Reynolds, T. Welacky, S. Weaver, A. Hamill and T. Vyn. 2003. Impacts of zone tillage and red clover on corn performance and soil physical quality. Soil Science Society of America Journal 67: 867-877.

[18] Ewing, R., M. Wagger and H. Denton. 1991. Tillage and cover crop management effects on soil water and corn yield. Soil Science Society of America Journal 55: 1081-1085.

[19] Fang, C., P. Smith, J.B. Moncrieff and J.U. Smith. 2005. Similar response of labile and resistant soil organic matter pools to changes in temperature. Nature 433: 57-59.

[20] Gardner, W.H. 1986. Water content. In: L.D. Whittig, et al., editors, Methods of Soil Analysis: Part 1—Physical and Mineralogical Methods. Soil Science Society of America and American Society of Agronomy, Madison, WI. p. 493-544.

[21] Haney, R.L., A.J. Franzluebbers, V.L. Jin, M.-V. Johnson, E.B. Haney, M.J. White and R.D. Harmel. 2012. Soil organic C: N vs. water-extractable organic C: N. Open Journal of Soil Science 2: 269-274.

[22] Hargrove, W. 1986. Winter legumes as a nitrogen source for no-till grain sorghum. Agronomy Journal 78: 70-74.

[23] Hauggaard-Nielsen, H., L. Holdensen, D. Wulfsohn and E.S. Jensen. 2010. Spatial variation of N2-fixation in field pea (Pisum sativum L.) at the field scale determined by the 15N natural abundance method. Plant and soil 327: 167-184.

[24] Hobbs, P.R., K. Sayre and R. Gupta. 2008. The role of conservation agriculture in sustainable agriculture. Philosophical Transactions of the Royal Society B: Biological Sciences 363: 543-555.

[25] Johanns, A.M., 2013, Metric Conversions, https://www.extension.iastate.edu/agdm/wholefarm/html/c6-80.html, [July 10, 2017]

[26] Johnson, T., T. Kaspar, K.A. Kohler, S. Corak and S. Logsdon. 1998. Oat and rye overseeded into soybean as fall cover crops in the upper Midwest. Journal of Soil and Water Conservation 53: 276-279.

[27] Jokela, W.E., J.H. Grabber, D.L. Karlen, T.C. Balser and D.E. Palmquist. 2009. Cover crop and liquid manure effects on soil quality indicators in a corn silage system. Agronomy Journal 101: 727-737.

[28] Kaspar, T., J. Radke and J. Laflen. 2001. Small grain cover crops and wheel traffic effects on infiltration, runoff, and erosion. Journal of Soil and Water Conservation 56: 160-164.

[29] Kaspar, T. and J. Singer. 2011. The use of cover crops to manage soil. In: J.L. Hatfield and T.J. Sauer, editors, Soil Management: Building a Stable Base for Agriculture. American Society of Agronomy and Soil Science Society of America, Madison, WI. p. 321-337.

[30] Keating, K.I. 1999. Allelopathy: principles, procedures, processes, and promises for biological control. Advances in Agronomy 67: 141-231.

[31] Keisling, T., H. Scott, B. Waddle, W. Williams and R. Frans. 1994. Winter cover crops influence on cotton yield and selected soil properties. Communications in Soil Science & Plant Analysis 25: 3087-3100.

[32] Mbuthia, L.W., V. Acosta-Martinez, J. DeBruyn, S. Schaeffer, D. Tyler, E. Odoi, M. Mpheshea, F. Walker and N. Eash. 2015. Long term tillage, cover crop, and fertilization effects on microbial community structure, activity: Implications for soil quality. Soil Biology and Biochemistry 89: 24-34.

[33] Mulvaney, R.L. 1996. Nitrogen—inorganic forms. In: D.L. Sparks, et al., editors, Methods of soil analysis part 3—Chemical methods. Soil Science Society of America and American Society of Agronomy, Madison, WI. p. 1123-1184.

[34] NCEI, 2017, Global Summary of the Month Station Details, https://www.ncdc.noaa.gov/cdo-web/datasets/GSOM/stations/GHCND:USC00406012/detail, Accessed [July 5, 2017]

[35] Nelson, D.W. and L.E. Sommers. 1996. Total carbon, organic carbon, and organic matter. In: D.L. Sparks, et al., editors, Methods of soil analysis part 3—chemical methods. Soil Science Society of America and American Society of Agronomy, Madison, WI. p. 961-1010.

[36] Nielsen, D.C., D.J. Lyon, G.W. Hergert, R.K. Higgins, F.J. Calderón and M.F. Vigil. 2015. Cover crop mixtures do not use water differently than single-species plantings. Agronomy Journal 107: 1025-1038.

[37] Nielsen, D.C. and M.F. Vigil. 2005. Legume green fallow effect on soil water content at wheat planting and wheat yield. Agronomy journal 97: 684-689.

[38] Papadopoulos, A., S. Mooney and N. Bird. 2006. Quantification of the effects of contrasting crops in the development of soil structure: an organic conversion. Soil Use and Management 22: 172-179.

[39] Reddy, K.N., R.M. Zablotowicz, M.A. Locke and C.H. Koger. 2003. Cover crop, tillage, and herbicide effects on weeds, soil properties, microbial populations, and soybean yield. Weed Science 51: 987-994.

[40] Reeleder, R., J. Miller, B.B. Coelho and R. Roy. 2006. Impacts of tillage, cover crop, and nitrogen on populations of earthworms, microarthropods, and soil fungi in a cultivated fragile soil. Applied Soil Ecology 33: 243-257.

[41] Salmerón, M., J. Cavero, D. Quílez and R. Isla. 2010. Winter cover crops affect monoculture maize yield and nitrogen leaching under irrigated Mediterranean conditions. Agronomy journal 102: 1700-1709.

[42] Savoy, H. 2009. Interpreting Mehlich 1 and 3 soil test extractant results for P and K in Tennessee. Publication W229, University of Tennessee Agricultural Extension Service, Knoxville, TN.

[43] SSSA. 2008. Glossary of soil science terms 2008Soil Science Society of America Madison WI.

[44] Steele, M., F. Coale and R. Hill. 2012. Winter annual cover crop impacts on no-till soil physical properties and organic matter. Soil Science Society of America Journal 76: 2164-2173.

[45] Thomas, G.W. 1996. Soil pH and soil acidity. In: D.L. Sparks, et al., editors, Methods of Soil Analysis Part 3—Chemical Methods. Soil Science Society of America and American Society of Agronomy, Madison, WI. p. 475-490.

[46] Tosti, G., P. Benincasa, M. Farneselli, F. Tei and M. Guiducci. 2014. Barley–hairy vetch mixture as cover crop for green manuring and the mitigation of N leaching risk. European Journal of Agronomy 54: 34-39.

[47] Unger, P.W. and M.F. Vigil. 1998. Cover crop effects on soil water relationships. Journal of Soil and Water Conservation 53: 200-207.

[48] UNL, 2017, United States Drought Monitor, http://droughtmonitor.unl.edu/MapsAndData/MapArchive.aspx, Accessed [June 20, 2017]

[49] USDA, 2016a, COVER CROP (340) REQUIREMENTS IN TENNESSEE, https://www.nrcs.usda.gov/wps/PA_NRCSConsumption/download?cid=nrcseprd414433&ext=pdf, Accessed [May 30, 2017]

[50] USDA, 2016b, Cover Crop Mixes? They Just Work Better, https://www.nrcs.usda.gov/wps/portal/nrcs/detail/il/newsroom/releases/?cid=STELPRDB1117185, Accessed [April 30, 2017]

[51] USDA, 2017, 2016 STATE AGRICULTURE OVERVIEW, https://www.nass.usda.gov/Quick_Stats/Ag_Overview/stateOverview.php?state=TENNESSEE, Accessed [June 10, 2017]

[52] Vance, E., P. Brookes and D. Jenkinson. 1987. An extraction method for measuring soil microbial biomass C. Soil biology and Biochemistry 19: 703-707.

[53] Villamil, M., G. Bollero, R. Darmody, F. Simmons and D. Bullock. 2006. No-till corn/soybean systems including winter cover crops. Soil Science Society of America Journal 70: 1936-1944.

[54] Weil, R.R., K.R. Islam, M.A. Stine, J.B. Gruver and S.E. Samson-Liebig. 2003. Estimating active carbon for soil quality assessment: A simplified method for laboratory and field use. American Journal of Alternative Agriculture 18: 3-17.

[55] Welch, R.Y., G.D. Behnke, A.S. Davis, J. Masiunas and M.B. Villamil. 2016. Using cover crops in headlands of organic grain farms: Effects on soil properties, weeds and crop yields. Agriculture, Ecosystems & Environment 216: 322-332.

CHAPTER III

HANEY SOIL HEALTH TEST: EVALUATION ON A WEST TENNESSEE SOIL

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Abstract

The Haney’s soil health test is a new approach to quantify soil health status by focusing mostly on soil biological properties. It uses a new extractant H3A in place of the traditional Mehlich-1 or Mehlich-3 extractant for soil nutrient extraction; a new method of soil respiration measurement using Solvita gel system, and a new way of determining bioavailable carbon (C) and nitrogen (N) in soil by measuring water extractable organic C (WEOC) and water extractable organic N (WEON). The soil health score is calculated by combining the Solvita respiration, WEOC and WEON. In this study, we collected soil samples from the University of Tennessee (TN) Research & Education Center (REC) at Milan, Tennessee. Components in the Haney’s soil health test were evaluated to test their effectiveness in TN soil. Among the three extractants tested, the H3A extracted the least soil extractable phosphorus, potassium and calcium, showing significant but weak correlation with the traditional extractants. The Solvita test did not provide a reliable estimation of potential mineralizable nitrogen (PMN). However, it correlated with many soil properties including soil C and N pools as well as the WEOC and WEON. Although the soil health score showed some extent of sensitivity to long-term cover crop treatments, it did not capture the variation in soil health status after four years of cover cropping with different species of cover crops. Overall, the Haney’s soil health score needed more evaluations and modifications to fit better to TN soils and cropping systems.

Introduction

Global agricultural production has increased greatly over the past 50 years to meet the growing demand of the rapidly increasing population. Data indicate that total food production in the next 50 years will equal the cumulative production in the past 500 years (Hatfield and Walthall, 2015). With the remarkable improvement in agricultural production, growing concerns regarding shrinking croplands and increasing environmental pollution call for sustainable intensification of agricultural production. There is increased interest in the role of soil biology in crop production systems, in addition to the importance of improved soil chemical and physical properties. The term “soil health,” often considered similar and used interchangeably with “soil quality,” is becoming more popular with a focus on soil biological characteristics. Haney’s soil health test is a recently emerged approach, which primarily focuses on measuring soil biological activity.

Haney’s soil health test uses a unique set of parameters to provide an overall status of soil biological status (Haney, 2013). It provides a recommendation based on the bioavailability of soil nutrients in contrast to the current fertilizer recommendations which are based on traditional soil tests. Haney et al. (2006) developed a new multi-element extractant called H3A for simultaneous measurement of soil phosphorus (P), potassium (K) and inorganic N (ammonium, and nitrate). Modified H3A includes three organic acids (malic acid, citric acid and oxalic acid), which are commonly found in root exudates (Haney et al., 2016). This extractant was designed to mimic the soil pH of the rhizosphere with actively growing roots and root exudates, so nutrients can be extracted near the soil pH of rhizosphere (Haney et al., 2006). Since soil pH and P solubility are highly interrelated (Golterman, 1988; Sharpley, 1993), the H3A extractant was intended to provide a more reliable estimate of plant available P.

In addition to the soil available nutrient contents which are often provided by the traditional soil tests, Haney's soil health test measures water extractable organic C and organic N (WEOC and WEON) contents. Since WEOC and WEON are closely connected to the soil microbial activity, the WEOC/WEON ratio could be a more sensitive indicator of soil microbial activity than the traditional total C:N ratio (Haney et al., 2012). According to this approach, a healthy soil should have a WEOC/WEON ratio below 20:1, which can be used as a practical threshold to separate the healthy soils from soils that may have immobilized N with high microbial activity.

The core of this soil health test is the measurement of carbon dioxide (CO_2)-C evolved from a 24-hr long incubation of re-wetted air-dried soil using Solvita gel system. It was originally designed to quantify the relative differences in CO_2 evolution across various types of compost in a short period (Brewer and Sullivan, 2003). Later, this was suggested to be a reasonably accurate method to measure the soil CO_2 respiration due to its high correlation with traditional methods of CO_2 measurements including acid-base titration and Infra-Red Gas Analysis (IRGA) analysis (Haney et al., 2008b). Haney's approach also provides relationships between soil fertility and soil microbial respiration by demonstrating that the flush of CO_2 following rewetting of dried soil is closely related to 28-day N and P mineralization as well as the WEOC and WEON concentrations (Haney et al., 2008a; Haney et al., 2012). The flush of CO_2 measurement is also suggested to be a viable test for biological soil quality. Subsequently, a strong positive relation between Solvita 1-day CO_2-C and 7-day anaerobic N mineralization of various soils across the US was reported with $r^2 = 0.82$ (Haney et al., 2015). A "Potential Mineralizable Nitrogen Calculator" was developed to help users interpret the Solvita results and to provide the N fertilizer recommendation by accounting for the potentially mineralizable N (PMN) (Solvita, 2017).

The final output of Haney's soil health test is a "soil health score," which is calculated from the following equation:

$$\frac{\text{1-day } CO_2\text{-C}}{\text{Water extractable organic C:N}} + \frac{\text{WEOC}}{100} + \frac{\text{WEON}}{10} \qquad \textbf{Equation 1}$$

Depending on the soil types and management, results can vary from 0 to 50. A score above four is considered acceptable for many soils (Haney, 2013).

Considering that Haney's test was developed for the soils of Texas and has not been calibrated for other soils, there is a paucity of information to show the effectiveness of this approach in regions other than Texas. A recent study showed a low correlation (r^2 = <0.05) between 1-d CO_2-C and PMN in different soil types (Horwath, 2015), which indicates that Solvita 1-day CO_2 respiration is not a universal predictor of N mineralization. In this study, we conducted a detailed evaluation of all components of Haney's soil health test including the H3A extractant, the Solvita test and the Haney's soil health score for the soils of west TN. Soil available nutrients extracted with the H3A reagent was compared with that of Mehlich-1 and Mehlich-3 extractants. The Solvita CO_2 result was correlated with the result of traditional acid-base titration method of CO_2 determination and the PMN. The Solvita CO_2 result and the soil health score under different cover crop (CC) treatments were compared to evaluate their sensitivity to differentiate management-specific conditions in TN soils. Pearson correlation and principal component analysis were conducted to evaluate the effectiveness of the Haney's score to represent the health status of TN soils.

Materials and methods

Field design and soil sampling

This study was conducted at the University of TN Research & Education Center (REC) at Milan, TN. The soil is classified as a Lexington silt loam (fine-silty, mixed, thermic, Ultic Hapludalf). The mean annual rainfall of the region is 1361 mm.

This study used two existing field experiments on CC at REC, Milan. The first experiment was established in 2002 while the second experiment was established in 2013. Both experiments were under a no-till corn (*Zea mays* L)-soybean (*Glycine max*) rotation with same management, and the experimental design was a randomized complete block (RCB) design.

In the short-term experiment, there are six CC treatments including single-, double- and multiple species (Table C-1), each with four replications. In the long-term experiment, there are four CC treatments, each with three replications (Table C-2).

Table C-1 Cover crop treatments in the short-term experiment

Treatments	***Species***	
A (CR/CC)	Cereal Rye (*Secale cereale* L.) Crimson Clover (*Trifolium incarnatum* L.)	
B (CR/HV)	Cereal Rye Hairy Vetch	
C (W)	Wheat (*Triticum aestivum* L.)	
D (CR)	Cereal Rye	
E (SHM)	Soil Health Mix	Cereal Rye Whole Oats (*Avena sativa* L.) Purple Top Turnips (*Brassica napus* L var) Daikon Radish (*Raphanus sativus*) Crimson Clover
F (Control)	Control—no cover crop	

Table C-2 Cover crop treatments in the long-term experiment

Treatments	***Species***
A (AP)	Austrian Winter Pea (*Pisum sativum*)
B (HV)	Hairy Vetch
C (W)	Wheat
D (Control)	Control—no cover crop

The soil samples were collected on October 18th, 2016 after the harvest of the main crop (soybean) and just before the seeding of the winter CC. The sampling depth was 15 cm. Samples were collected using stainless steel probes (2.5 cm in diameter) from the two field trials, along with samples collected from nearby undisturbed woods and grassland to represent a relative 'natural' soil with no recent cropping history. From each plot, approximately 10-15 subsamples were randomly collected and then mixed to get one composite sample. Multiple samples were also collected from each of the 7 pristine locations. Composite samples were stored in plastic Ziploc bags and placed them in a cooler with blue ice while transporting to the laboratory. A subsection of each sample was air-dried and passed through a 2 mm sieve for analyzing physical and chemical properties, and the rest was stored in 4℃ for analyzing biological properties.

Measurement of crop yield

Harvest was conducted using a plot harvester. Yield was measured using a yield monitor. Soybean yields were converted to Mg/ha using the following equation 2 (Johanns, 2013):

Yield (Mg/ha) = [(Bushels soybean/acre) * (27.22kg/bushels soybean) ÷ (0.404hectare/acre)] ÷ (1000kg/Mg) **Equation 2**

Measurement of soil properties

All chemicals and materials used were purchased from Fisher Scientific. All reagents were prepared using type 1 ultrapure water.

Soil pH

Soil pH was measured from a 1:2 soil:water suspension (Thomas, 1996). 10 g of air-dried soil was weighed and mixed with 20 ml of ultrapure water. Soil pH was measured from the supernatant using a Denver Instrument Model 250 pH meter after shaking and setting.

Gravimetric soil moisture content

Soil moisture content was measured by gravimetric method (Gardner, 1986). Approximately 10 g soil sample was weighed into tared tin cups and dried in an oven at 105℃ for 24 hours. Samples were removed, cooled, and recorded dry weight. Soil moisture content was calculated using equation 3:

$$\frac{Mass_{Wet\ soil} - Mass_{Dry\ soil}}{Mass_{Dry\ soil}}$$ **Equation 3**

Extractable nutrients

Phosphorous (P), potassium (K), and calcium (Ca) were extracted using Mehlich-1, Mehlich-3 (Savoy, 2009) and H3A extractants (Haney et al., 2006). For Mehlich-1, approximately 5 g of air-dry soil was extracted with 20 mL of the extractant. Samples were shaken and filtered. For Mehlich-3, approximately 2.5 g of dry soil was extracted with 25 mL of the extractant. Samples were also shaken and filtered. For H3A, approximately 4 g of dry soil was extracted with 40 mL of the extractant. Samples were shaken, centrifuged and filtered.

All filtrates were collected and stored in the refrigerator for elemental analysis using a Perkin-Elmer 5300 & 7300 DV Inductively Coupled Plasma (ICP) unit.

Soil organic carbon and total nitrogen

Soil organic carbon and total nitrogen (T-N) were measured by combustion method using a Thermo Flash EA 1112 NC combustion analyzer. Air-dried samples were powdered using a mortar and pestle before analysis (Nelson and Sommers, 1996).

Soil labile carbon

Soil labile carbon, also called permanganate oxidizable carbon (POXC), was determined using the method developed by Weil et al. (2003). Soil samples were first oxidized by potassium permanganate ($KMnO_4$). The remaining $KMnO_4$ was immediately measured by a spectrophotometer at a wavelength of 550 nm. A standard curve was developed to correlate the solution concentration and the absorbance. The POXC content (mg kg^{-1}) was calculated using equation 4:

$$[0.02 - (\mathrm{a} + \mathrm{b} \times \mathrm{Abs})] * 9000 * \frac{0.02}{Wt}$$ **Equation 4**

Where: 0.02 (mol/L) = initial solution concentration a = intercept of the standard curve

b = slope of the standard curve

Abs = absorbance of unknown

9000 (mg C/mol) = milligrams of carbon oxidized by 1 mole of $KMnO_4$

Wt = weight of air-dried soil sample in kg

Soil microbial biomass carbon

Soil microbial biomass carbon was analyzed using the chloroform fumigation-extraction method (Vance et al., 1987). Briefly, 10g of fresh soil samples were fumigated with chloroform in the dark for 48 hours. Both fumigated and non-fumigated (control) samples were then extracted with 0.5M potassium sulfate (K_2SO_4). Total dissolved carbon was measured using a Shimadzu Total Organic Carbon Analyzer (TOC-5000). The difference between C in fumigated and non-fumigated samples is the chloroform labile C (EC). Microbial biomass C was calculated as EC divided by *k*, a constant, estimated at 0.45 (Beck et al., 1997).

Soil inorganic nitrogen

Soil inorganic nitrogen was analyzed using the extraction method modified based on the protocol described by Mulvaney (1996). 4 g of air-dry soil was extracted with 40ml of 2M potassium chloride (KCl). NH_4-N and NO_3-N were measured from the extract using a Skalar San++ Continuous Flow Analyzer (CFA).

Water extractable organic carbon, inorganic nitrogen and organic nitrogen in soil

Water extractable organic carbon (WEOC), water extractable inorganic nitrogen (WEIN) and water extractable organic nitrogen (WEON) were analyzed using the extraction method described by Haney et al. (2012). WEOC and water extractable total N (WEN) were determined by extracting 4 g of air-dry soil with 40 mL of ultrapure water and shaking for 10 minutes. Samples were then centrifuged, filtered and analyzed for WEOC and WEN using the SHIMADZU TOC-V (CPH) carbon analyzer and TNM-1 nitrogen measuring unit. WEIN concentrations were determined from the same extractant using the Skalar Continuous Flow Analyzer (CFA). WEON was calculated by subtracting inorganic N content from WEN.

Soil potential mineralizable nitrogen (PMN)

Soil PMN was measured using the 7-day anaerobic incubation method (Waring and Bremner, 1964). Approximately 5 g of dry soil was saturated with water and incubated at 40℃ for 7 days. Soil was extracted using 25 ml 2M KCl solution before and after the incubation, and soil NH_4-N was measured from the extract by the Skalar CFA. The difference of NH_4-N before and after the incubation is the PMN.

Soil Respiration

For the Solvita test, undisturbed fresh soil samples were sieved through a 2-mm sieve and air-dried. Approximately 40 g of sample was rewetted with 12 mL of DI water in the 50-mL plastic beaker and incubated in the 250-mL gas-tight jars with the Solvita gel paddle at around 22℃ for 24 hours. The paddles were taken out after the incubation and read by the Solvita digital color reader to measure the 1-day CO_2 flux.

The acid-base titration method was modified based on the method described by Anderson (1982). Approximately 10 g of fresh soil was weighed into a sealed glass jar and incubated at 22℃ for 24 hours. A vial containing 0.1M sodium hydroxide (NaOH) was included inside the jar. The NaOH solution which trapped the CO_2 was removed from the jar after 24 hours. The remaining alkali unreacted with CO_2 was back titrated by 0.1M hydrochloric acid (HCl) and the amount of CO_2-C was calculated.

Statistical analysis

The effect of CCs on soil health score was analyzed by analysis of variance (ANOVA) based on Generalized Linear Model (GLM) in SAS 9.4.

Fisher's protected least significant difference (LSD) was used to determine significant differences among treatment means at $P < 0.05$ (SAS Institute, Cary, NC, 2013). PROC CORR procedure of SAS was applied to determine relationships among different extractants and among soil properties. The principal component analysis (PCA) was conducted using PROC FACTOR procedure of SAS to determine the variables that show the strongest relationships to the overall soil status. The factors derived from PCA consist of contributions from 19 parameters including the general soil properties, the Haney's soil health score and the parameters used to calculate Haney's score. These factors derived from PCA were considered mutually orthogonal, uncorrelated, and successively explain the maximum residual variation (Sena et al., 2002). Total variance of each factor was defined as eigenvalue (Swan and Sandilands, 1995). Factors with high eigenvalue (>1) and could explain more than 5% of the total variance in the data was retained (Brejda et al., 2000; Wander and Bollero, 1999). The selected factors/parameters were evaluated for their effectiveness in representing the overall soil status using their factor loading.

Results and discussion

Evaluation of H3A extractant

Soil extractable P, K, and Ca were determined after extracting the soil samples with Haney's H3A, Mehlich-1 and Mehlich-3 extractants. For the short-and long-term experiments, Mehlich-3 extracted the highest P followed byMehlich-1 and H3A across the CC treatments (Figure C-1A, B). Across all study sites, Mehlich-3 extracted 45% higher and H3A extracted 86% lower soil P compared to Mehlich-1 (Table C-3). This result is in accordance with Haney et al. (2010) which reported that H3A extracted less P than Mehlich-3 and with Wolf and Baker (1985) which reported that Mehlich-3 extracted more P than Mehlich-1. Haney et al. (2006) reported that extractable P increased

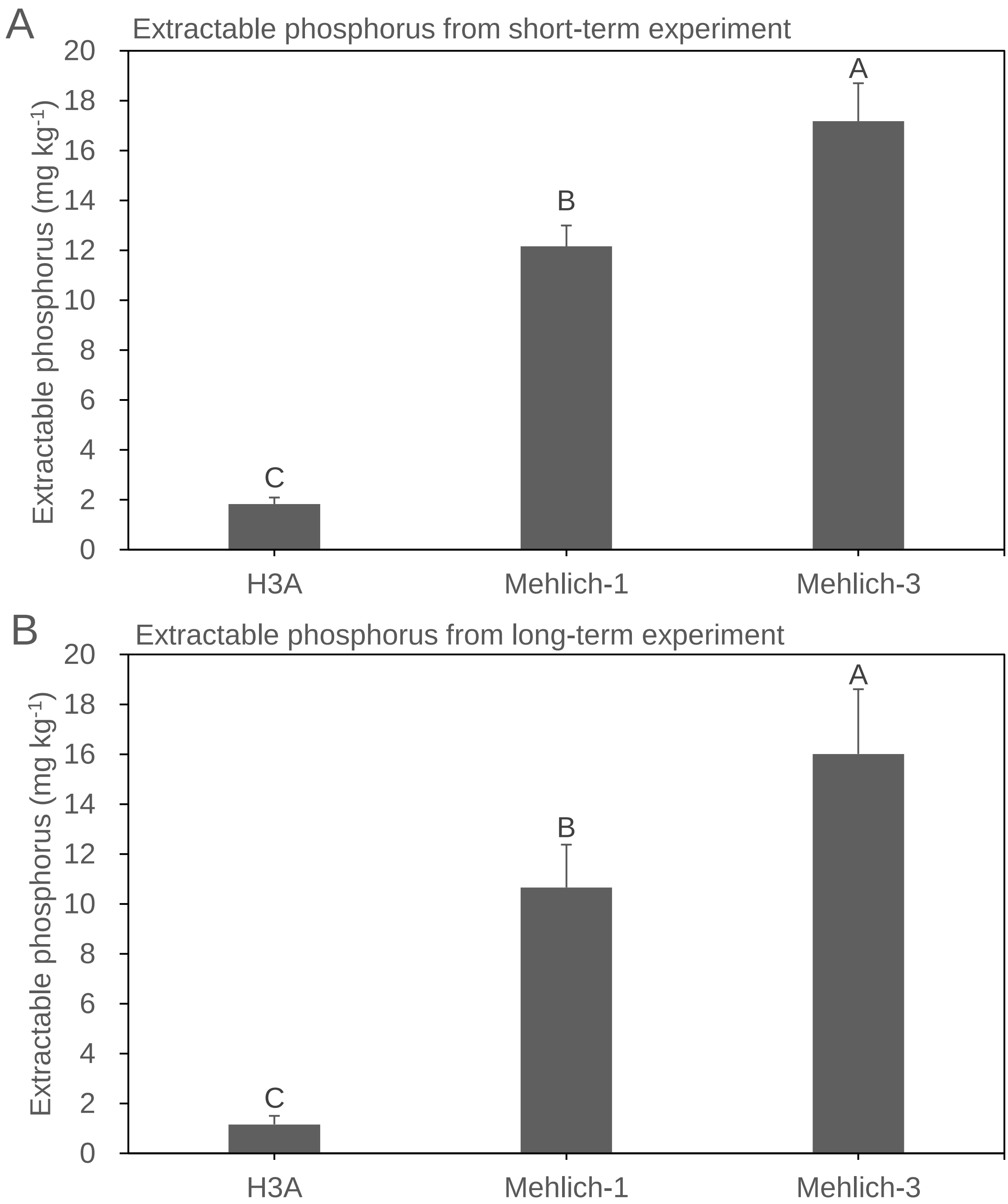

Figure C-1 Soil extractable phosphorus content from short- and long-term experiments using three extractants. (A) Extractable phosphorus from the short-term experiment, (B) Extractable phosphorus from the long-term experiment. Different letters denote statistically different means at P < 0.05 and error bars represent standard error of the mean.

Table C-3 Correlation matrix for soil-extractable P, K and Ca extracted with Mehlich-1, Mehlich-3, and H3A extractants. Correlation coefficient, P Value, and number of samples are listed under each pair of correlation.

	P H3A		K H3A		Ca H3A
P M1	0.66	K M1	0.56	Ca M1	0.44
	<.0001		<.0001		0.0033
	43		43		43
P M3	0.79	K M3	0.30	Ca M3	0.19
	<.0001		0.0489		0.2338
	43		43		43

Mean of 43 samples:

P: Mehlich-1: 11.62 mg kg-1, Mehlich-3: 16.84 mg kg-1, H3A: 1.67 mg kg-1

K: Mehlich-1: 64.08 mg kg-1, Mehlich-3: 90.97 mg kg-1, H3A: 50.83 mg kg-1

Ca: Mehlich-1: 686.50 mg kg-1, Mehlich-3: 1017.43 mg kg-1, H3A: 401.72 mg kg^{-1}

with decrease in extractant pH. This relationship between extracted P and extractant pH probably explain why H3A extracted less P than Mehlich-1 and Mehlich-3 since H3A consists of three weak organic acids that have higher pH than Mehlich-1 and Mehlich-3 (Golterman, 1988; Haney et al., 2010). H3A extractable P correlated well with Mehlich-1 and Mehlich-3 extractable P (Table C-3). But the correlation coefficients were lower than that reported by Haney et al. (2010). This may be due to the lower number of total samples in the present study.

Effect of extractant type on soil extractable K content followed a pattern similar to that of extractable P in both the short- and long-term experiments (Figure C-2A, B). Across all the sites, Mehlich-3 extracted the highest amount of K, followed by Mehlich-1 (29% lower than Mehlich-3) and H3A (44% lower than Mehlich-3) (Table-C3). However, compared to the H3A extracted P content, H3A extracted K content was much closer to Mehlich-1 and Mehlich-3 extracted K (Figure C-2A, B). H3A extractable K showed significant correlation with the Mehlich-1 and Mehlich-3 extractable K, but the correlation coefficients were lower than that of extractable P (Table C-3). Significant correlation of soil K extracted with H3A with that extracted with ammonium

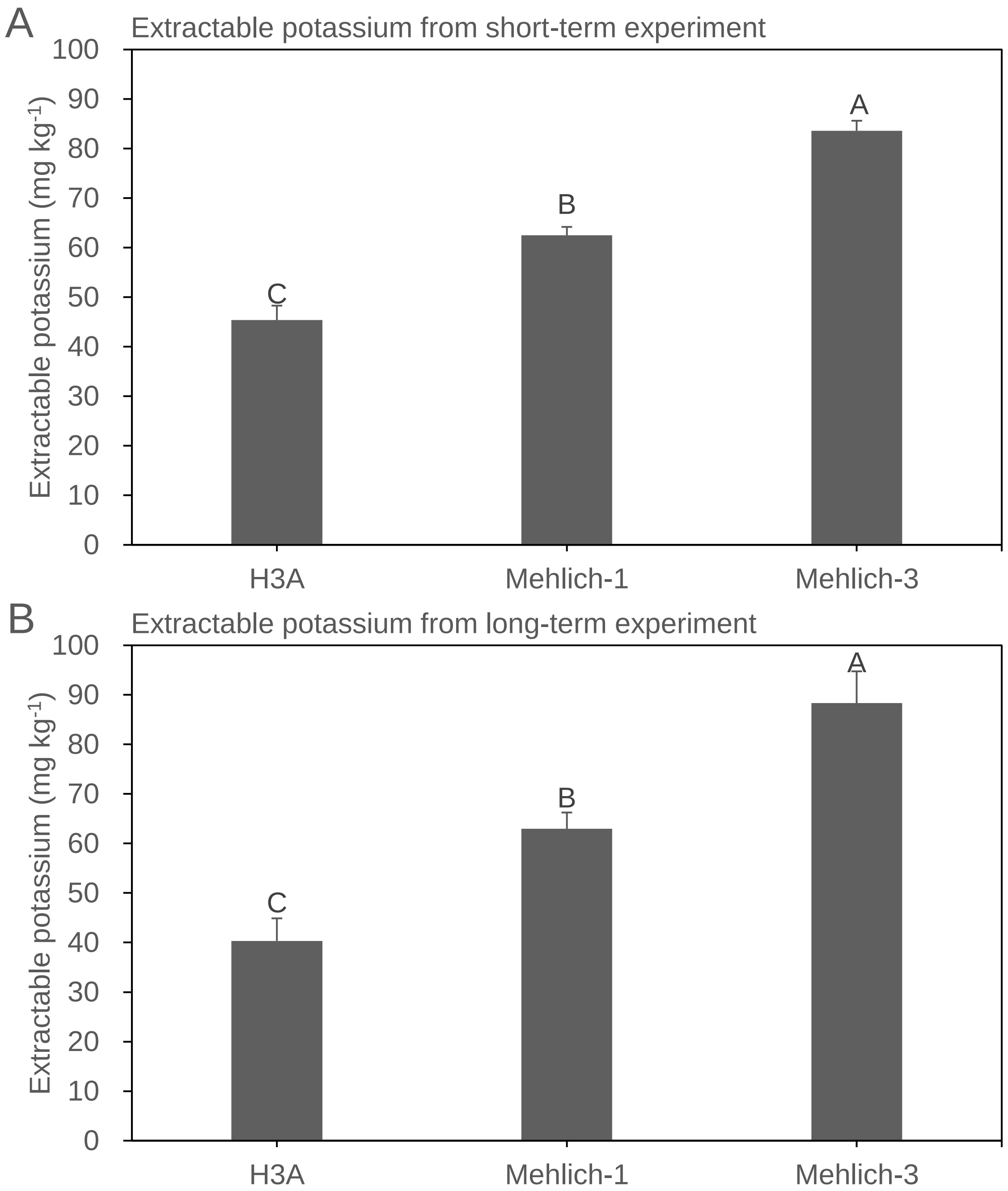

Figure C-2 Soil extractable potassium content from short- and long-term experiments using three extractants. (A) Extractable potassium from the short-term experiment, (B) Extractable potassium from the long-term experiment. Different letters denote statistically different means at P < 0.05 and error bars represent standard error of the mean.

acetate (NH_4-OAc) was reported by Haney et al. (2010). University of TN recommends Mehlich-1 as the extractant for determining available nutrients (Savoy, 2009a) and past studies comparing the extraction efficiency of H3A with Mehlich-1 are scanty (Haney et al., 2010; Hanlon and Savoy, 2009).

The pattern of soil extractable calcium (Ca) from the three extractants was similar to that of extractable P (Figure C-3A, B). Across the sites, Mehlich-3 extracted the highest amount of Ca, followed by Mehlich-1 (32% lower than Mehlich-3) and H3A (61% lower than Mehlich-3) (Table C-3). The lowest soil Ca extracted with H3A may have been due to the lower dissolution of Ca-associated phosphates by the H3A reagent, which is composed of weak organic acids. It could also be the reason for the lower extraction of soil P by the H3A. To confirm this, extracted iron and aluminum data by the three extractants are needed, which was not the focus of this study. H3A extractable Ca correlated with Mehlich-1 extractable Ca but showed no significant correlation with the Mehlich-3 extractable Ca (Table C-3) and the correlation coefficients were lower than that reported in Haney et al. (2010).

Evaluation of Solvita test

The Solvita 1-day CO_2-C (hereafter called as Solvita) and a series of soil properties were subjected to the correlation analysis. The Solvita showed significant positive correlation with soil pH, extractable K, Ca, SOC, POXC and T-N (Table C-4). Tu (2016) also reported a good correlation between Solvita, soil organic matter, inorganic N, permanganate oxidizable carbon (POXC) and PMN in Minnesota soilOur results did not show significant correlation of Solvita with the CO_2 determined by the traditional acid-base titration method (data not shown). Also, Solvita did not differ significantly across the CC treatments in the short-term experiment. But in the long-term experiment winter wheat produced significantly higher Solvita than hairy vetch (Figure C-4). Chemical and biological methods were developed to measure the PMN pool to help better estimate the plant available N pool so that over

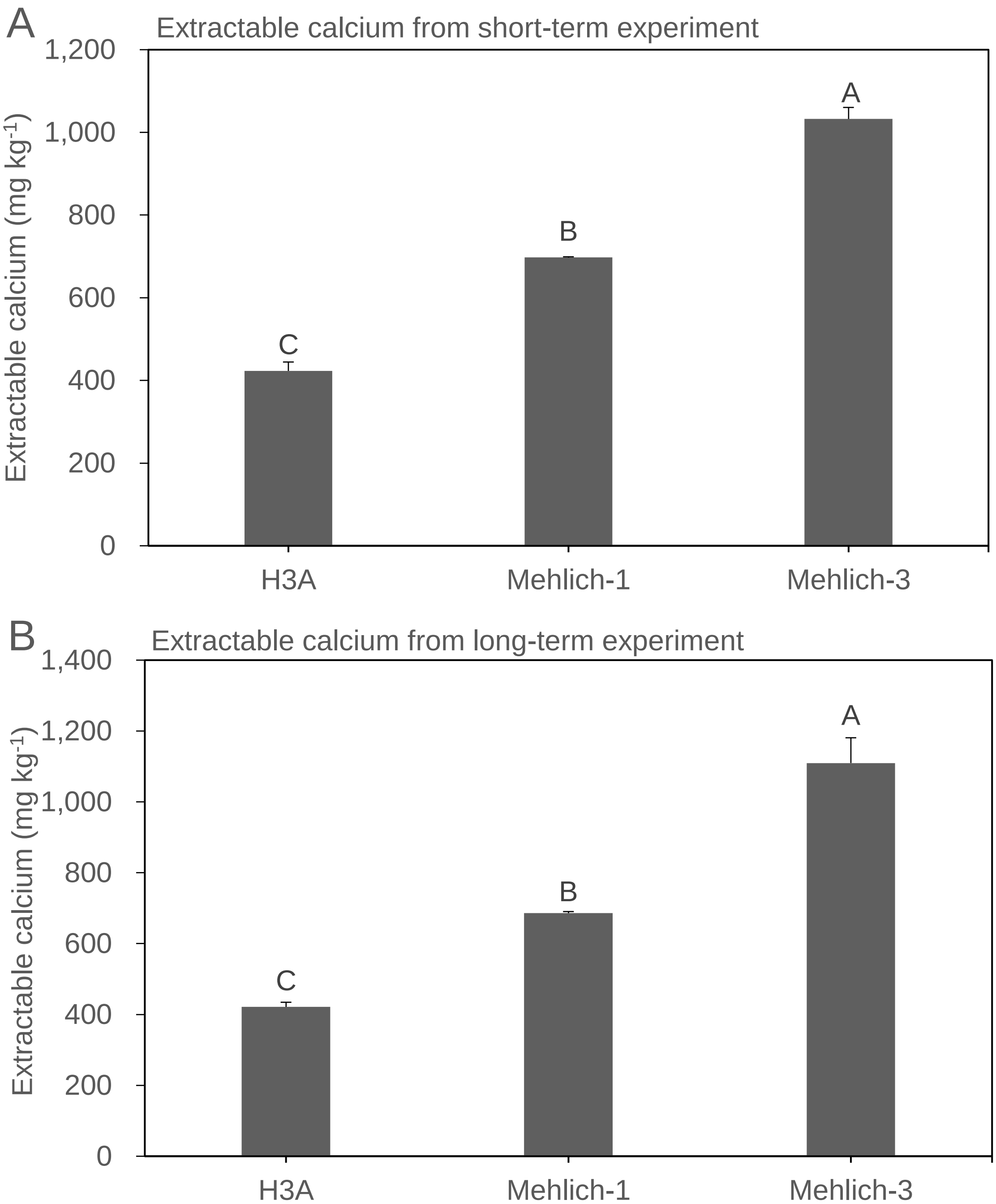

Figure C-3 Soil extractable calcium content from short- and long-term experiments using three extractants. (A) Extractable calcium from the short-term experiment, (B) Extractable calcium from the long-term experiment. Different letters denote statistically different means at $P < 0.05$ and error bars represent standard error of the mean.

Table C-4 Pearson's correlation result for soil properties and Haney's soil health score

Properties	pH	ω	Solvita	Titration	P	K	Ca	Mg	SOC	POXC	MBC	WEOC	T-N	NO_3	InN	WEON	PMN	C: N	WEC:N
ω	**																		
Solvita	**																		
Titration		-*																	
P																			
K			***	*															
Ca	***	**	**	-*															
Mg					-**														
SOC		-**	**	***		**	-*	*											
POXC		-**	**	**				**	***										
MBC		-**		*			-*	***	**	*									
WEOC	**	-***		**			-***	**	***	***	**								
T-N		-**	**	**	-*	**		**	***	***	**	**							
NO_3		**		-***			**	-**	-***	-**	-***	-**	-**						
InN		*		-***	*		*	-**	-**	-*	-**	-*	-**	***					
WEON					**									**	***				
PMN					-**			**											
C:N	-*	-*		*	**		-**					**							
WEC:N	-*	-***		**			-***	**	***	***	**	***	***	-***	-***			**	
Soil Health score	**	*	*				***		-*		-*	-***		**	*				-***

ω, gravimetric moisture content; Solvita, Solvita 1-day CO_2-C; Titration, acid-base titrated CO_2-C; P, extractable phosphorus; K, extractable potassium; Ca, extractable calcium; Mg, extractable magnesium; SOC, soil organic carbon; POXC, permanganate oxidizable carbon (labile carbon); MBC, microbial biomass carbon; WEOC, water extractable organic carbon; T-N, total nitrogen; NO_3, nitrate nitrogen; InN, inorganic nitrogen; WEON, water extractable organic nitrogen; PMN, potential mineralizable nitrogen; WEC:N, WEOC:WEON ratio. " * " means significant correlation at $P<0.05$, " ** " means significant at $P<0.01$ and " * " means significant at $P<0.0001$. Hyphen (-) means negative correlation.**

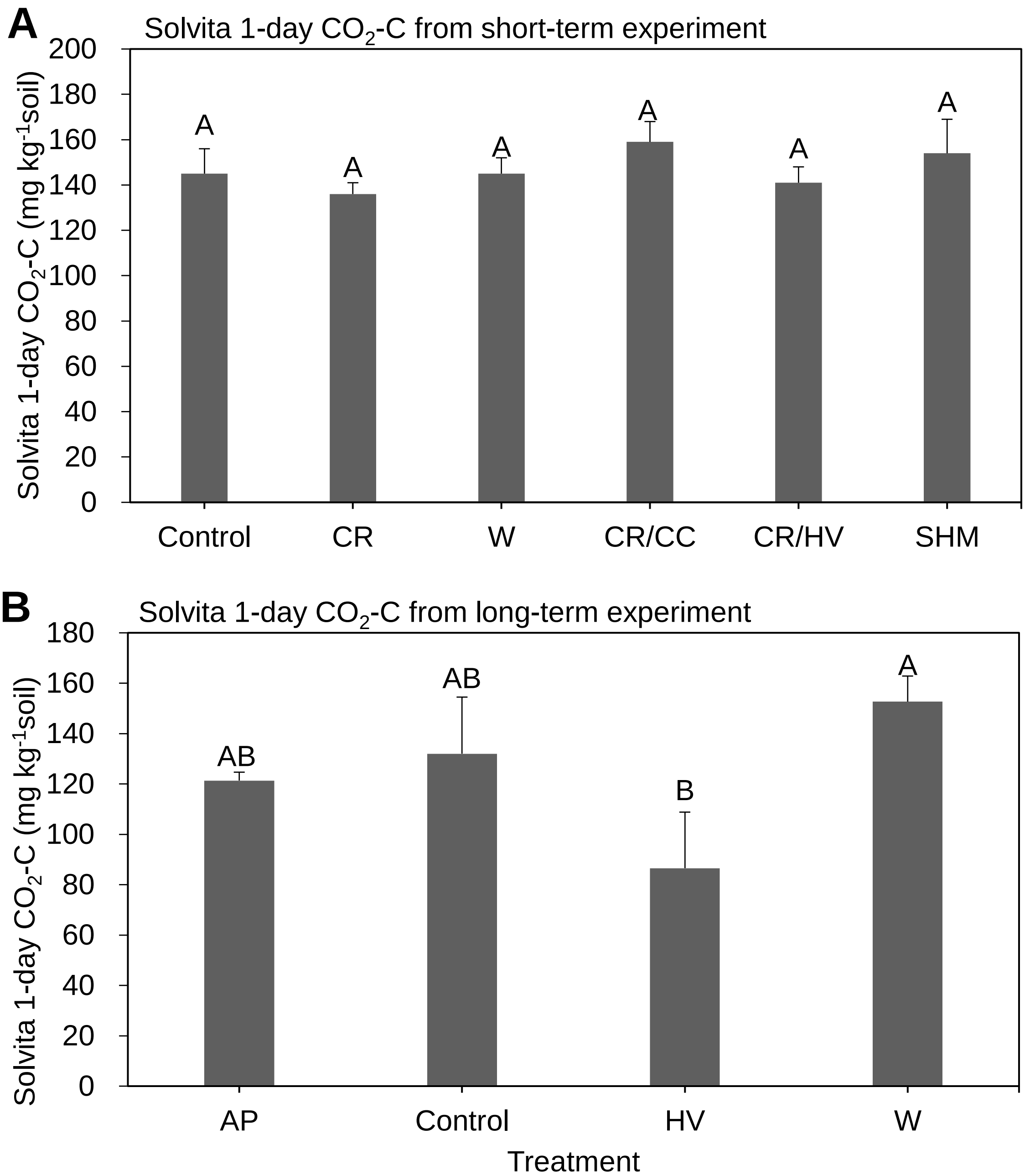

Figure C-4 Solvita 1-day CO_2-C in response to cover crop treatments in short- and long-term experiments. (A) Solvita 1-day CO_2-C from the short-term experiment. CR-Cereal Rye, W-Wheat, CC-Crimson Clover, HV-Hairy Vetch, SHM-Soil Health Mix. (B) Solvita 1-day CO_2-C from the long-term experiment. AP-Austrian Winter Pea. Different letters denote statistically different means at $P < 0.05$ and error bars represent standard error of the mean.

fertilization can be avoided (Horwath, 2015).Most of these methods either measured relative production of inorganic N by the mineralization process over a specific incubation period or extracted NH_4–N that correlated with the standard incubation measurement (Fox and Piekielek, 1978; McDonald et al., 2014; Øien and Selmer-Olsen, 1980; Stanford and Smith, 1972; Waring and Bremner, 1964). The Solvita method of PMN estimation was developed based on the observations of Franzluebbers et al. (1996) that the flush of CO_2, which indicates the size and activity of the soil microbial biomass, was a good biochemical indicator of mineralizable N in soils Haney et al. (2015). Consequently, a strong linear correlation between Solvita and 7-day PMN was developed (r^2=0.82). But in our study, Solvita showed lower correlations with the 7-day PMN from both short- and long-term experiments (Figure C-5A, B). There was one outlier data point in the long-term experiment, removing which increased the R^2 to 0.18 (data not shown). The relationship observed in our study was much weaker than that reported by Haney et al. (2015) (R^2=0.82). Additionally, compared to the previous study, the Solvita values were much higher and the PMN values were much lower in our study which indicates that the soils we tested had a higher microbial activity but lower nitrogen mineralization potential. N bioavailability is a process controlled by the exoenzyme-driven depolymerization reactions (Schimel and Bennett, 2004). Without thorough understanding of the enzymology involved in this process, it would be hard to predict the bioavailability of N. The low correlation between Solvita and PMN in our study indicates that Haney's approach for predicting bioavailable N to correct fertilization recommendation is not robust enough for the agroecosystems of TN.

Evaluation of Haney's soil health score

Haney's soil health score is calculated from Solvita 1-day CO_2-C, WEOC and WEON, reflecting its focus on soil biological properties.

Haney's soil health score under different CC treatments in short-term and

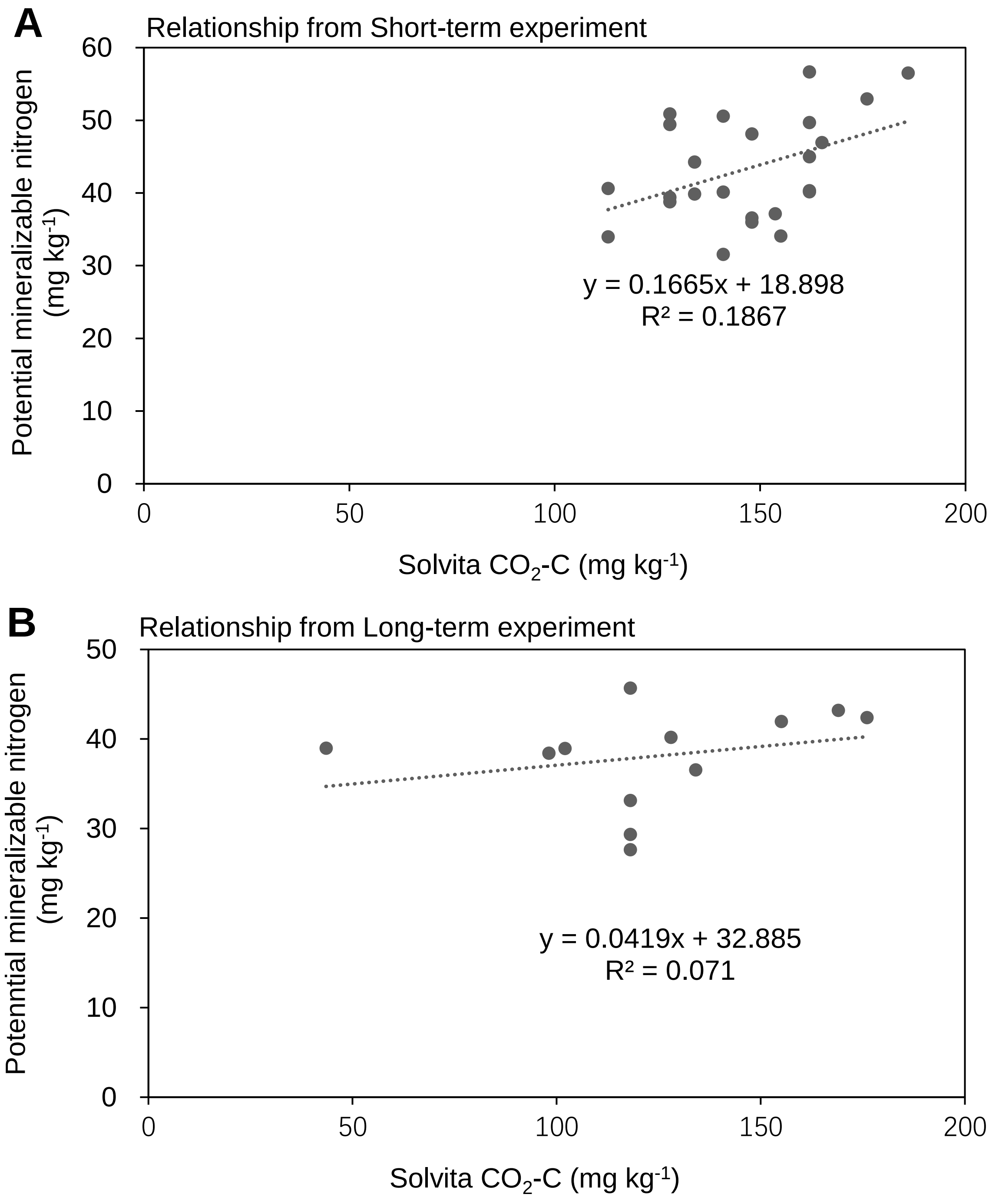

Figure C-5 Relationship between Solvita 1-day CO_2-C and 7-day potential mineralizable nitrogen. (A) Relationship from the short-term experiment. (B) Relationship from the long-term experiment.

long-term experiments are shown in Figure C-6. In the short-term experiment, the health score ranged from 30.80 to 33.98. Although the CR/CC showed relatively higher score, no significant differences were found among treatments. But in the long-term experiment, significantly higher soil health score was found for AP and W treatment than the HV treatment, which was corresponded to the result of Solvita. This indicates that the Haney's score was not sensitive to differentiate the soil health from short-term management changes, but it could detect the soil health differences from the long-term management changes. In addition, the similar trend in Solvita and the soil health score indicates that out of the three variables used to calculate the soil health score (Solvita, WEOC, WEON), the score is skewed by the Solvita results.

The Pearson's correlation coefficients for soil properties, soil health score and the parameters used to calculate the soil health score are shown in Table C-4. The WEOC showed strongly significant and positive correlation to titrated CO_2-C, SOC, POXC, MBC, T-N, C:N and WEC:N ($P<0.01$). Strong negatively significant correlations between WEOC and pH, moisture level, Ca, NO_3 and soil health score were also found. The WEON only showed strongly significant and positive correlation to soil P, NO_3 and inorganic nitrogen (InN). The health score showed significant relationships with several soil carbon fractions and inorganic N. The WEC:N ratio introduced by Haney et al. (2012) correlated with more carbon and nitrogen parameters than the traditional C:N ratio and the soil health score, indicating that it may be a better predictor of soil functions. No significant correlation was found between soil health score and soybean yield (data not shown).

In order to determine the variables with strongest relations to overall soil health, PCA was conducted using the soil properties, Haney's soil health score and Haney's soil health variables to form the smallest possible subsets of variables representing the majority of variance. Each of the first four groups

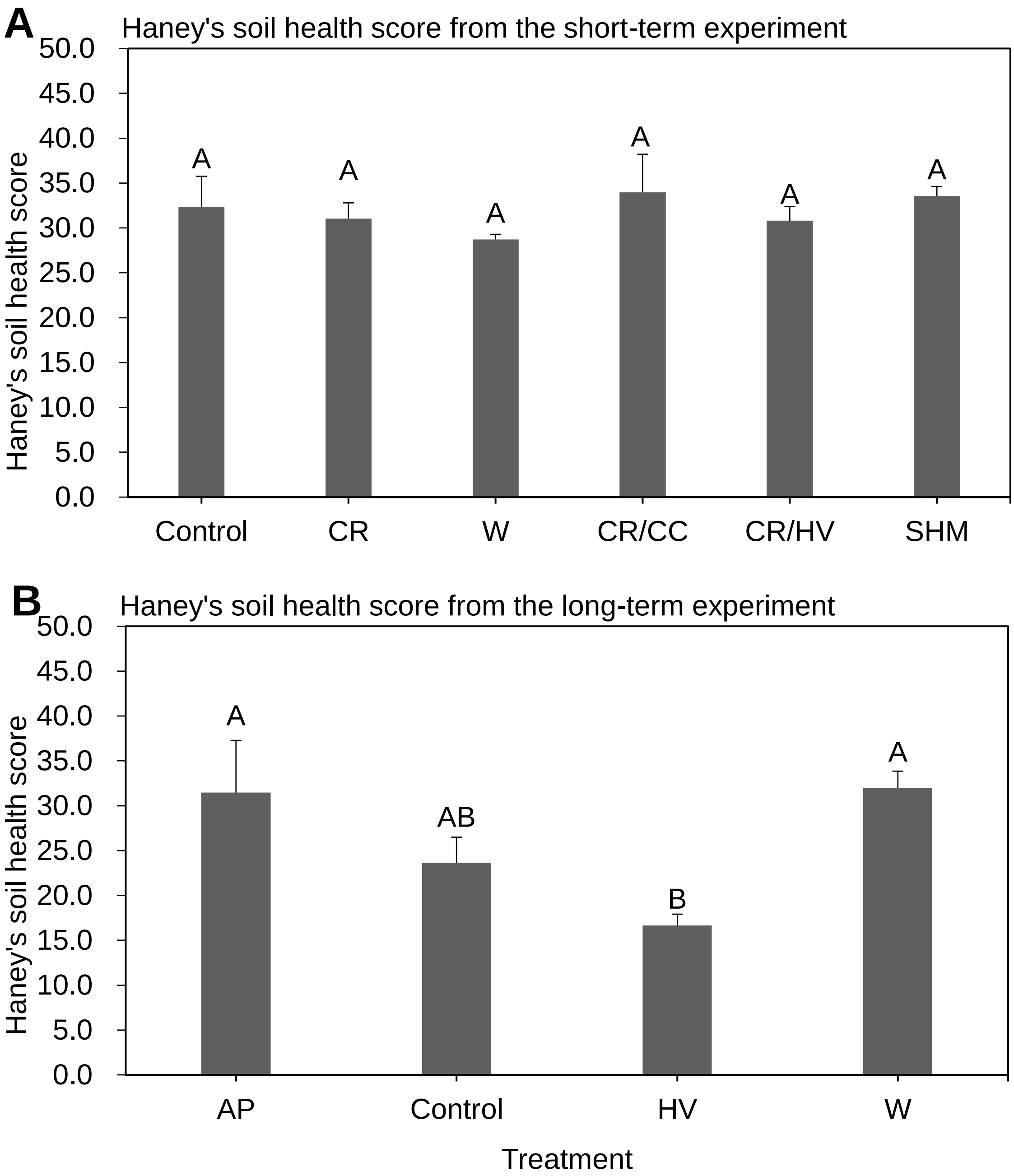

Figure C-6 Haney's soil health score under different CC treatment in the short-term and long-term experiments. (A) Haney's soil health score from short-term experiment. CR-Cereal Rye, W-Wheat, CC-Crimson Clover, HV-Hairy Vetch, SHM-Soil Health Mix. (B) Haney's soil health score from long-term experiment. AP-Austrian Winter Pea. Different letters denote statistically different means at $P < 0.05$ and error bars represent standard error of the mean.

or factors had eigenvalues >1 and were retained for interpretation (Table C-5). These four factors explained cumulative sample variance of 79%. In Factor 1, which explained about 40.4% of variance, WEOC:WEON ratio showed the highest factor loading (0.33) among variables, followed by NO_3 (-0.30), WEOC (0.29) and SOC (0.29). Solvita (0.42) and pH (0.40) showed higher factor loadings in factor 2, and explained 17% of the variance. Factor 3 explained 12.4% of the variance, in which P (0.50) and WEON (0.41) were highly weighted. PMN from Factor 4 was selected as highly weighted variable. Overall the factor loadings of variables in the PCA were low as compared to previous studies (Jagadamma et al., 2008). Variables used to calculate Haney's soil health test showed higher factor loadings among all the variables, but the soil health score itself didn't, which indicates that the calculation of Haney's soil health score may need further modification to better fit for the soils of TN.

Conclusion

In this study, we evaluated three components of Haney's soil health test: the extractant H3A, Solvita test and the soil health score. The H3A extractant showed significant correlation with Mehlich-1 for soil P, K and Ca, and with Mehlich-3 for soil P and K. But the correlations were relatively weaker than that reported previously. We didn't find correlation between Solvita, PMN and CO_2 determined by the traditional acid-base titration method, but Sovita correlated well with some other soil properties including pH, SOC, POXC and T-N. Haney's soil health score showed significant difference among different CC treatments only in the long-term study. Haney's soil health variables and soil health score correlated to some extent with soil properties such as pH, moisture, soil carbon and nitrogen pools such as SOC, POXC, MBC, T-N, InN and NO3. But the PCA analysis showed that the Haney's soil health variables (Solvita, WEOC, WEON), but not the health score, better explained the overall variance of tested soil properties. In conclusion, more samples need to be

Table C-5 Factor analysis results based on soil properties

Factors	Factor1	Factor2	Factor3	Factor4
Eigenvalue	8.1	3.4	2.5	1.9
Percent variance	40.4	17.0	12.4	9.37
Cumulative variance	40.4	57.4	69.8	79.2
Eigen vectors				
ω	-0.25	0.18	-0.17	-0.07
pH	-0.12	**0.40**	0.00	-0.06
POXC	0.26	0.11	0.26	0.09
NO_3	**-0.30**	-0.03	0.21	0.17
InN	-0.27	-0.06	0.26	0.22
SOC	**0.29**	0.19	0.16	-0.03
T-N	0.26	0.29	0.08	0.04
CO_2-C	0.25	0.12	0.00	-0.08
MBC	0.26	0.07	-0.09	0.25
P	-0.07	-0.18	**0.50**	-0.22
K	0.12	0.34	0.20	0.02
Ca	-0.22	0.34	0.11	0.08
Mg	0.20	0.05	-0.21	0.41
C:N	0.17	-0.29	0.28	-0.16
PMN	0.00	-0.05	-0.11	**0.63**
Solvita	0.06	**0.42**	0.32	-0.01
WEOC	**0.29**	-0.16	0.19	0.17
WEON	-0.09	-0.19	**0.41**	0.38
WEC:N	**0.33**	-0.07	0.01	-0.06
Soil health score	-0.26	0.23	0.13	0.10

ω, gravimetric moisture content; Solvita, Solvita 1-day CO2-C; Titration, acid-base titrated CO2-C; P, extractable phosphorus; K, extractable potassium; Ca, extractable calcium; Mg, extractable magnesium; SOC, soil organic carbon; POXC, permanganate oxidizable carbon (labile carbon); MBC, microbial biomass carbon; WEOC, water extractable organic carbon; T-N, total nitrogen; NO3, nitrate nitrogen; InN, inorganic nitrogen; WEON, water extractable organic nitrogen; PMN, potential mineralizable nitrogen; WEC:N, WEOC:WEON; factor loadings that are bold and underlined are considered highly weighted.

analyzed in order to compare the extraction efficiency of H3A extractant with Mehlich-1, which is the recommended extractant for the TN soil. The Solvita cannot provide reliable estimation of PMN of TN soils. Haney's soil health variables showed potential to represent the overall health status of TN soils, but the formula of soil health score needed modifications. According to the result of this study, parameters like SOC, NO_3, soil pH, P and PMN might help to improve the responsiveness of Haney's soil health score to the agricultural soils of TN. In addition, alternate sampling dates and sampling depths should be tested.

Reference

[1] Anderson, J.P. 1982. Soil respiration. In: A.L. Page, et al., editors, Methods of soil analysis. Part 2—Chemical and microbiological properties. Soil Science Society of America and American Society of Agronomy, Madison, WI. p. 831-871.

[2] Beck, T., R. Joergensen, E. Kandeler, F. Makeschin, E. Nuss, H. Oberholzer and S. Scheu. 1997. An inter-laboratory comparison of ten different ways of measuring soil microbial biomass C. Soil Biology and Biochemistry 29: 1023-1032.

[3] Brejda, J.J., T.B. Moorman, D.L. Karlen and T.H. Dao. 2000. Identification of regional soil quality factors and indicators I. Central and Southern High Plains. Soil Science Society of America Journal 64: 2115-2124.

[4] Brewer, L.J. and D.M. Sullivan. 2003. Maturity and stability evaluation of composted yard trimmings. Compost Science & Utilization 11: 96-112.

[5] Fox, R. and W. Piekielek. 1978. A rapid method for estimating the nitrogen-supplying capability of a soil. Soil Science Society of America Journal 42: 751-753.

[6] Franzluebbers, A.J., R.L. Haney, F. Hons and D. Zuberer. 1996. Determination of microbial biomass and nitrogen mineralization following rewetting of dried soil. Soil Science Society of America Journal 60: 1133-1139.

[7] Gardner, W.H. 1986. Water content. In: L.D. Whittig, et al., editors, Methods of Soil Analysis: Part 1—Physical and Mineralogical Methods. Soil Science Society of America and American Society of Agronomy, Madison, WI. p. 493-544.

[8] Golterman, H. 1988. Reflections on fractionation and bioavailability of sediment bound phosphate. Arch. Hydrobiol. Beih. Ergebn. Limnol 30: 1-4.

[9] Haney, R.L., 2013, Soil Testing for Soil Health, https://www.usda.gov/oce/forum/past_speeches/2013_Speeches/Haney.pdf, Accessed [May 29, 2017]

[10] Haney, R.L., W. Brinton and E. Evans. 2008a. Estimating soil carbon, nitrogen, and phosphorus mineralization from short-term carbon dioxide respiration. Communications in Soil Science and Plant Analysis 39: 2706-2720.

[11] Haney, R.L., W. Brinton and E. Evans. 2008b. Soil CO_2 respiration: Comparison of chemical titration, CO_2 IRGA analysis and the Solvita gel system. Renewable agriculture and food systems 23: 171-176.

[12] Haney, R.L., A.J. Franzluebbers, V.L. Jin, M.-V. Johnson, E.B. Haney, M.J. White and R.D. Harmel. 2012. Soil organic C: N vs. water-extractable organic C: N. Open Journal of Soil Science 2: 269-274.

[13] Haney, R.L., E.B. Haney, D. Harmel, D. Smith and M. White. 2016. Evaluation of H3A for Determination of Plant Available P vs. FeAlO Strips. Open Journal of Soil Science 6: 175-187.

[14] Haney, R.L., E.B. Haney, L. Hossner and J. Arnold. 2006. Development of a new soil extractant for simultaneous phosphorus, ammonium, and nitrate analysis. Communications in Soil Science and Plant Analysis 37: 1511-1523.

[15] Haney, R.L., E.B. Haney, L. Hossner and J. Arnold. 2010. Modifications to the new soil extractant H3A-1: A multinutrient extractant. Communications in soil science and plant analysis 41: 1513-1523.

[16] Haney, R.L., E.B. Haney, D.R. Smith and M.J. White. 2015. Estimating Potential Nitrogen Mineralisation Using the Solvita Soil Respiration System. Open Journal of Soil Science 5: 319-323.

[17] Hanlon, E.A. and H.J. Savoy. 2009. Producedures Used by State Soil Testing Laboratories in The Southern Region of The United

StatesSouthern Extension Research Activity Information Exchange Group-6 Clemson, SC.

[18] Hatfield, J.L. and C.L. Walthall. 2015. Soil biological fertility: Foundation for the next revolution in agriculture? Communications in Soil Science and Plant Analysis 46: 753-762.

[19] Horwath, W.R. 2015. Soil respiration tests as predictors of nitrogen mineralization potential. In: G. Mooso, et al., editors, Proceedings of The Western Nutrient management Conference. Western Region Nutrient Management Coordinating Committee, Reno, NV.

[20] Jagadamma, S., R. Lal, R.G. Hoeft, E.D. Nafziger and E.A. Adee. 2008. Nitrogen fertilization and cropping system impacts on soil properties and their relationship to crop yield in the central Corn Belt, USA. Soil and Tillage Research 98: 120-129.

[21] Johanns, A.M., 2013, Metric Conversions, https://www.extension.iastate.edu/agdm/wholefarm/html/c6-80.html, [July 10, 2017]

[22] McDonald, N.T., C.J. Watson, S.T. Lalor, R.J. Laughlin and D.P. Wall. 2014. Evaluation of soil tests for predicting nitrogen mineralization in temperate grassland soils. Soil Science Society of America Journal 78: 1051-1064.

[23] Mulvaney, R.L. 1996. Nitrogen—inorganic forms. In: D.L. Sparks, et al., editors, Methods of soil analysis part 3—Chemical methods. Soil Science Society of America and American Society of Agronomy, Madison, WI. p. 1123-1184.

[24] Nelson, D.W. and L.E. Sommers. 1996. Total carbon, organic carbon, and organic matter. In: D.L. Sparks, et al., editors, Methods of soil analysis part 3—chemical methods. Soil Science Society of America and American Society of Agronomy, Madison, WI. p. 961-1010.

[25] Øien, A. and A. Selmer-Olsen. 1980. A laboratory method for evaluation of available nitrogen in soil. Acta Agriculturae Scandinavica 30: 149-156.

[26] Savoy, H. 2009. Interpreting Mehlich 1 and 3 soil test extractant results for P and K in Tennessee. Publication W229, University of Tennessee Agricultural Extension Service, Knoxville, TN.

[27] Schimel, J.P. and J. Bennett. 2004. Nitrogen mineralization: challenges of a changing paradigm. Ecology 85: 591-602.

[28] Sena, M., R. Frighetto, P. Valarini, H. Tokeshi and R. Poppi. 2002. Discrimination of management effects on soil parameters by using principal component analysis: a multivariate analysis case study. Soil and Tillage Research 67: 171-181.

[29] Sharpley, A.N. 1993. An innovative approach to estimate bioavailable phosphorus in agricultural runoff using iron oxide-impregnated paper. Journal of Environmental Quality 22: 597-601.

[30] Solvita, 2017, Guide for Interpreting CO_2 Burst Results, https://solvita.com/soil/potential-min-n-calc/, Accessed [June 15, 2017]

[31] Stanford, G. and S. Smith. 1972. Nitrogen mineralization potentials of soils. Soil Science Society of America Journal 36: 465-472.

[32] Swan, A.R. and M. Sandilands. 1995. Introduction to geological data analysis. International Journal of Rock Mechanics and Mining Sciences and Geomechanics Abstracts 8: 387A.

[33] Thomas, G.W. 1996. Soil pH and soil acidity. In: D.L. Sparks, et al., editors, Methods of Soil Analysis Part 3—Chemical Methods. Soil Science Society of America and American Society of Agronomy, Madison, WI. p. 475-490.

[34] Tu, X. 2016. Solvita and Other Measures as Indicators of Soil N Availability in Minnesota Soils. M.S. Thesis, University of Minnesota, Retrieved from the University of Minnesota Digital Conservancy.

[35] Vance, E., P. Brookes and D. Jenkinson. 1987. An extraction method for measuring soil microbial biomass C. Soil biology and Biochemistry 19: 703-707.

[36] Wander, M. and G. Bollero. 1999. Soil quality assessment of tillage impacts in Illinois. Soil Science Society of America Journal 63: 961-971.

[37] Waring, S. and J. Bremner. 1964. Ammonium production in soil under waterlogged conditions as an index of nitrogen availability. Nature 201: 951-952.

[38] Weil, R.R., K.R. Islam, M.A. Stine, J.B. Gruver and S.E. Samson-Liebig. 2003. Estimating active carbon for soil quality assessment: A simplified method for laboratory and field use. American Journal of Alternative Agriculture 18: 3-17.

[39] Wolf, A.M. and D.E. Baker. 1985. Comparisons of soil test phosphorus by Olsen, Bray P1, Mehlich I and Mehlich III methods. Communications in Soil Science and Plant Analysis 16: 467-484.

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CHAPTER IV

SUMMARY

Cover cropping is one of the conservation agricultural practices used to reduce soil erosion and nutrient loss, typically during the non-growing cropping seasons. Recent studies show that cover crops (CCs) can bring multiple ecosystem benefits to soil, crop and the environment. The results of these studies, however, vary by a suite of factors including soil types, cover crop species, cropping systems, and regional climate. The additional cost and labor associated with establishing a CC integrated cropping system as well as the time it takes to see the benefits can be the major barriers to their adoption. Comprehensive studies are needed to help farmers better understand the benefits of the CCs they are dealing with. Growing interest emerged in recent years regarding the use of multi-species CCs (three or more species). But studies on the effect of multi-species CCs in comparison with single and double species on soil quality attributes and crop production are limited, especially in the southeastern USA. To evaluate the effectiveness of the USDA recommended soil health mix (SHM, a mixture of five cover crop species) as compared to the common single or double-species CCs, soil samples from a short-term (3-year) and a long-term (14-year) CC field experiments were collected during October 2016. Soil properties and crop yield data were also collected. The SHM showed an increase in soybean yield from the short-term experiment, which could be attributed to increased soil inorganic N and gravimetric moisture content from SHM treatments compared to other less diverse CC treatments and no-cover control. Overall, the multi-species CC showed potential to improve the soil quality and crop production.

Haney's soil health test was developed with a heavy focus on soil biological properties, which was not emphasized in previous soil quality assessment methods. With the faster soil respiration test (Solvita), the modified soil carbon and nitrogen measurements (water extractable organic carbon (WEOC) and water extractable organic nitrogen (WEON)), and an overall soil health score derived from Solvita, WEOC and WEOC, this test is claimed to be convenient and faster than the other methods. In addition to providing the soil health score, Haney's method also introduced a new soil nutrient extractant (H3A-combination of malic acid, citric acid and oxalic acid) for more reliable extraction of soil available nutrients for plants. Since the Haney soil health test was originally developed for soils of Texas, more evaluation is needed to confirm its applicability for agricultural soils of TN. In this study, the three components of the Haney's soil health test were evaluated. The H3A extractant showed a significant correlation to Mehlich-1 and Mehlich-3, but the relationship was weaker than had been reported previously. The use of the Solvita method to measure microbial activity did not give a reliable estimation of the potential mineralizable nitrogen (PMN) in TN soil, but it correlated well with some other soil properties including pH, SOC, POXC and T-N. Haney's soil health score showed significant difference among different CC treatments only in the long-term study. The correlation analysis showed that the soil health score and variables used to calculate the score (Solvita, WEOC, WEON) correlated with soil properties such as pH, soil moisture, soil carbon and nitrogen pools, but in the PCA the soil health score cannot explain much variance comparing to variables used to calculated it. This suggests that the algorithm used to assign a soil health score will need modification for soils in west TN. More soil samples from diverse cropping systems and locations need to be analyzed in order to compare the efficiency of extraction of H3A with Mehlich-1 and Mehlich-3and to test the robustness of Haney's soil health test in different regions.

VITA

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