OXIDATION-REDUCTION ENZYME ASSAYS AS SOIL QUALITY INDICATORS AND RELATIONSHIPS TO CROP PRODUCTIVITY

THESIS

Presented in Partial Fulfillment of the Requirements for the Degree Master of Science in the Graduate School of Ohio State University

By

YANG DING

Graduate Program in Environmental Science

Ohio State University

2018

Master's Examination Committee:

Professor Richard Dick, Advisor

Professor Warren Dick

Professor Brian Slater

ABSTRACT

There is growing recognition for the need to develop sensitive soil quality indicators that reflect soil management and that assist land managers in promoting long-term sustainability of terrestrial ecosystems. Potential analyses of soil quality indicator may be limited by sensitivity, temporal stability or practicality, enzyme activities have been tested as efficient soil quality indicators because they are simple and could be adopted by commercial soil testing laboratories. In order to be effective, soil quality indicators need to be integrated with other biophysical and socio-economic indicators, and crop yield is one of the most important economic indicators. Various soil quality indicators have been proposed, but few studies have investigated their relationship to crop yield. Landscape position and drainage is a major controller of crop yields, and therefore oxidation-reduction enzymes hold potential to be related to drainage.

Paired fields of no-tillage (NT) and conventional tillage (CT) management were sampled with most fields under a corn-soybean rotation. Soil samples were taken in September 2017 from fields in Ohio, Illinois and Iowa. Crop yields were measured in September 2015, 2016 and 2017. Soil drainage class information was determined by using the Web Soil Survey. Rhodanese (RA) and ammonium oxidation enzyme (AO) activities on field-moist or air-dried soil samples were determined in the 0-5 cm depth and 5-15 cm depth for NT and 0-15 cm depth for CT treatment. RA or AO activities were correlated with crop yields was analyzed. Drainage class was not correlated with RA, AO or crop yields, and there was no significant difference in crop yields between NT and CT. AO activities in 0-5 cm depth were significantly higher under NT compared to CT. RA activities in air-dried soil were significantly higher at NT 0-5 cm depth compared to CT.

The effect of air-drying was investigated as pre-treatment to enzyme assays because assays that can use air-dried soils are desirable for commercial applications as it stabilizes the sample (compared to field moist soil). It was found that air-dried soil samples provided the same rankings of treatments by RA or AO assays and would facilitate adoption of these assays for practical adoption by soil testing labs. Also air-drying increased the ability of RA assay to detect tillage difference. RA and AO activities were not significantly correlated with crop yield. However, enzyme activity/unit clay did increase the correlation r-values in relation to crop yields and increased the ability of enzyme assays to detect management effects. The lack of drainage class effects on yields or enzyme activities could be due to the study sites having only small changes in elevation between well drained and poorly drained sites. More studies are needed with samples taken from a stronger landscape position gradient, and other methods to test redox potential of soil should be adopted, such as measurement of reduction potential (E_h). In conclusion, enzyme activities were sensitive in detecting tillage effects and the air-drying pretreatment for RA and AO assay is feasible.

ACKKNOWLEGEMENT

First and foremost, I would like to thank my committee members Richard Dick, Warren Dick and Brian Slater for their assistance and patience throughout this entire process. A special thanks is also extended to Prof. Richard Dick, as my advisor, for supporting my program and sharing his valuable knowledge and ideas with me. I appreciated advice of Prof. Warren Dick and that he travelled a long distance to attend our every meeting. Prof. Brian Slater kindly helped me learn the Web Soil Survey and has been always available to answer my questions. This project would not be possible, and I would not be where I am today without their guidance and expertise.

I would also like to show my gratitude for Ohio State Laboratory staff for helping me around the lab and always offering me support. Specifically, Nicola Lorenz and Nathan Lee were integral in the completion of my research. Nicola trained me on lab technologies and assisted me with experiments. Nathan Lee is a great friend and colleague; he shares his experience in lab and assisted me in statistical analyses. Also, I want to thank the ESGP program. It gave me a chance to study in the U.S. and provided valuable support.

Last, but not least, I would like to thank my family. I would not have had the determination to continue my education without their encouragement and support. I am forever grateful for the support and love they have shown me throughout my endeavors.

VITA

June 2010 .....Xiangyang No.5 High School, Xiangyang, China

May 2015 .....B.S. Environmental Engineering, Wuhan Textile University

2017 to present .....Graduate Teaching Associate, School of Environment and Natural Resources, Ohio State University

PUBLICATIONS

Wei Shi, Qian Chen, Ping Zhang, Yang Ding, Huijie Dong, Lian Duan, Xiaoxia Li, Aihua Xu *"Catalytic Decolorization of Methyl Orange by "Rectorite–Sulfite System"* in *Catalysis Communications --Volume 56, 5 November 2014, Pages 32–35*

Wei Shi, Qian Chen, Lian Duan, Yang Ding, Zhongduo Xiong, Xiaoxiao Li and Aihua Xu *"Catalytic Generation of Hydroxyl Radicals by Dioxygen-mediated Oxidation of p-aminophenol by Simple Cobalt(II) Ions in Bicarbonate Aqueous Solution for use in Acid Orange 7 Decolorization* in *New J. Chem., --2014,38, 4938-4945 accepted 1st August 2014*

FIELDS OF STUDY

Major Field: Environmental Science

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

CHAPTER 1: ENZYME ACTIVITIES AS SOIL QUALITY INDICATORS: A REVIEW

ABSTRACT

With increasing public interest in sustainable agriculture, a tool for evaluating the quality of our soil resources is needed. Soil is an essential component of the Earth's biosphere, functioning in food production, and environmental quality maintenance. Unlike air and water, where standards have been established, standards for soil quality indicators have been difficult to establish due to the complexities of the interrelated biological, physical, and chemical properties of soils. To be practical and guide land management, a soil quality indicator needs to be able to detect changes rapidly (i.e. within a few years), be calibrated and interpreted, have seasonal stability, have high throughput capability, and be cost-effective. Some biological and chemical properties have been studied as indicators of soil quality, most notably microbial biomass, microbial diversity, nutrient mineralization, and soil organic matter are regarded as possible soil quality indicators. However, soil organic matter responds slowly to land management, while the biological properties may be too temporally responsive to short-term environmental factors (e.g. precipitation, tillage). Previous work in our laboratory and others' studies suggests certain soil enzyme assays have the potential to be suitable soil quality indicators because they are temporally sensitive to land management, seasonally stable, and analytically straight forward. This chapter reviewed the literature of soil quality measurement in general and enzyme activities in particular for their potential as soil quality indicators. Some enzyme activities have potential to detect soil management because of their sensitivity to land management, importance in nutrient cycling, and simplicity.

INTRODUCTION

Background

There are great concerns about the effects of human activities on the global environment, with soils being a critical resource for sustaining life on Earth (Sagan, 1992; Bhagat, 1990; Doran and Parkin, 1994). In June 1992, heads of states and delegates from 178 countries participated in the Union Nations Conference on Environment and Development. Since the 1980s, severe degradation of soil's productivity capacity occurred on more than 10% of the Earth's vegetated land as a result of soil erosion, atmospheric pollution, cultivation, over-grazing, land clearing, salinization, and desertification (World Resources, Inst., 1992; Sanders, 1992; Doran and Parkin, 1994).

Soil is a vital natural resource and is nonrenewable on a human time scale (Jenny, 1980). Protecting soils has become a national and world priority and an integral part of protecting the environment. A recent call for the development of a soil health index was stimulated by the perception that human health and welfare are associated with the quality and health of soils (Haberern, 1992; Doran and Parkin, 1994). Doran and Parkin (1994) defined soil quality as the "capacity of a soil to function within ecosystem boundaries to sustain biological productivity, maintain environmental quality and promote plant and animal health." However, quantifying soil quality remains an elusive goal because soil is a dynamic, living body that plays key roles in terrestrial ecosystems.

The components of soil include inorganic mineral matter (sand, silt, and clay), organic matter, water, gases, and living organisms, such as earthworms, insects, bacteria, fungi, algae, and nematodes (Doran and Parkin, 1994). Therefore, it is not plausible to

establish a single biological, physical or chemical measurement that could adequately reflect soil quality without taking into consideration the factors affecting the formation of a given soil (Doran and Parkin, 1994; Bandick and Dick, 1999). Since about 1993, national or regional programs have been established in several countries to monitor soil quality or the state of biodiversity (Stenberg, 1999; Nielsen and Winding, 2002). These countries include Canada, France, Germany, Switzerland, the Czech Republic, United Kingdom, Australia, United States, and New Zealand (Bloem, 2006). Although monitoring was initiated, little information was exchanged or published in the international literature (Bloem, 2006).

A suitable soil quality indicator should both a) identify a problem and b) monitor changes in soil health caused by management. Larson and Pierce (1991) stated that a system to assess soil quality can be considered analogous to a medical examination for humans. It can simultaneously indicate problems in soil health, track the origins of problems, and help monitor changes. Moreover, it is able to predict future changes. Granatsein and Bezdicek (1992) stated that a soil quality indicator should be able to identify problem production areas, make realistic estimates of food production, monitor changes in sustainability and environmental quality as related to agricultural management, and assist federal and state agencies in formulating and evaluating sustainable agricultural and land-use policies.

To be practical for use by scientists, farmers, conservationists, and policy makers, Doran and Parkin (1994) summarized a set of suitability criteria that basic soil quality indicators should meet: 1) encompass ecosystem processes and relate to process-oriented modeling; 2) integrate soil physical, chemical, and biological properties and processes; 3)

be accessible to many users, and applicable to field conditions; 4) be sensitive to detect changes and practices in management and climate; and 5) where possible, be components of existing soil data bases.

Soil Quality Indicators

Soil indicators are divided into physical, chemical, and biological categories, depending on how they affect soil functions. Any soil property to assess soil quality should be sensitive to management practices (Doran and Parkin, 1994) and can be changed easily in positive or negative ways. If changed, some properties and processes will recover at varying rates, while others are irreversible (Schoenholtz et al., 2001)

Physical properties relate to soil structure, gas exchange and water relations. Although soil texture is the most fundamental qualitative soil physical property controlling water, nutrients, and oxygen exchange, retention, and uptake (Schoenholtz et al., 2001), it changes little through time. Therefore, it is not very useful to detect management effects. Therefore, the physical properties are bulk density, available water capacity, and aggregate stability, that have been proposed by the USDA (2018) as indicators to help conservationists and soil scientists with soil health assessment. Corstanje et al. (2017) used a logical sieve approach based on key policy-related soil functions to determine the seven physical properties that were prioritized, including soil-packing density, soil water retention characteristics, aggregate stability, the rate of soil erosion, the depth of soil, soil structural, and soil sealing. Among these seven properties, packing density was derived by measuring the bulk density modified by clay content; soil water retention characteristics encapsulated plant-available water capacity, air capacity, macro porosity, and soil porosity.

Bulk density is an indicator of soil compaction and porosity. High bulk density causes restriction to root growth and poor water movement through the soil. Aggregate stability refers to the ability of soil aggregates to resist disruptive forces, such as tillage. Wet aggregate stability suggests how well soil can resist raindrop impact and water erosion, while the size distribution of dry aggregates can be used to predict resistance to abrasion and wind erosion (USDA, 2018).

Aggregate stability is highly related to organic matter and biological activity; it is an indicator of organic matter, biological activity, and nutrient cycling. Greater amounts of stable aggregates indicate better soil quality. Measurements of aggregate size distributions are the most relevant to the germination and early growth of plants on soils that are tilled, structurally stable, and are not compacted by traffic, while the measurements have less relevance to later growth or to early growth on untilled soils or tilled soils that are unstable or compacted by traffic (Kay and Grant, 1996).

Assessing water infiltration, availability, and retention are important for soil functions (Schoenholtz et al., 2001), especially water retention for plants and microorganisms. Available water capacity has been used for irrigation scheduling and is used in many crop-growth and hydrologic models (Kay and Grant, 1996). Except for the static soil properties discussed above, other physical indicators can be more complex constructs of several soil variables, such as soil-packing density (Corstanje et al., 2017).

Soil chemical properties, especially soil pH, nutrients, organic matter, and inorganic nutrients, were commonly used in traditional soil testing for crop nutrients indicators. Reganold and Palmer (1995) used chemical soil properties to evaluate the soil quality between different grass management systems in New Zealand. Soil testing for

nutrient status is well established and useful for crop and forage production. Adequately available nutrients are critical to crop production. Chemical analysis for soil nutrients has been foundational for maintaining agricultural productivity and has guided farmers since the 1900s. This critical component of soil health assessment has been mostly accepted and adopted by farmers and land managers. However, it does not reflect the ability of soils to provide services, such as phytopathogen suppression, resistance to erosion or as rooting media for optimal plant growth. Additionally, indicators need to reflect the degree of degradation or pollution in soils and identify end points for soil remediation.

Soil pH refers to the degree of soil acidity or alkalinity and affects soil's biological, chemical, and physical processes. Since the pH scale is in logarithmic units, a small change of pH units can induce significant changes in the chemical and biological processes in soil. In acidic soils, calcium, magnesium, nitrate-nitrogen, and phosphorus are deficient, whereas aluminum and manganese are abundant, which further exacerbates acidification. Very acid or alkaline pH levels slow organic mineralization due to poor microbial activity linked to bacteria. The yields of most crops decrease where pH is low and increase as pH rises to an optimum level. Many crops grow best if pH is close to neutral (pH 6–7.5), although a few crops prefer acidic or alkaline soils. Smith and Doran (1996) found that the highest yield of corn occurs at pH 6.8, whereas the highest yield of oats was at pH 7.5. Soil pH is easily measured in the field, can provide information on nutrient condition, and can be indicators of the effects on biological activity where certain microbial-mediated processes are affected by the shifts in pH (Smith and Doran, 1996). Soil pH is a good indicator because management practices can have a significant effect on pH over a short time. However, the buffer capacity of soil can confound the results

and make them difficult to interpret because soils with better buffer capacities are more resistant to a drop or rise in pH. Therefore, the quantity of limestone required to increase the pH of an acidic soil to the desired level must be specifically determined for each field before amending the soil.

Soil organic matter (SOM) or soil organic carbon (SOC) is commonly recognized as one of the key chemical parameters of soil quality. It is a critical pool in the carbon cycle and plays an important role in nutrient release and availability (Henderson, 1995). Bünemanna et al. (2018) reviewed sixty-two publications and found that total organic matter/carbon and pH were the most frequently proposed soil quality indicators. However, the quantitative assessment of its contribution to soil quality is lacking. Aune and Lal (1997) provided quantitative relationships between SOC and crop yield for tropical Oxisols, Ultisols, and Alfisols. They found a weak relationship between SOM and crop productivity ($r^2 = 0.37$). However, decreasing SOC had a strongly negative effect on crop productivity below a threshold value (SOC = 1%). Defining qualitative criteria for SOC is hampered by the fact that critical threshold values may be vastly different among soils orders; for example, the same percentage of organic C translates into different soil productivity capacity in Ultisols compared to Mollisols (Schoenholtz et al., 2001). Other environmental factors, such as climate and land use would also hamper this quantitative assessment. One example of a practical assessment of SOM in soil quality is the Wisconsin Soil Health Scorecard. Specific thresholds were used to indicate soil health: SOM = 4%–6% is healthy soil, SOM< 2% is unhealthy soil, and SOM = 2%–4% is impaired soil (Romig et al., 1996). However, this criteria would vary across soil types, climate regions, and landscapes. Although SOM is ubiquitous and changes in response to

managements, measurable changes in SOM generally are too slow in responding to perturbations (on the order of decades) to be useful to land managers.

SOM contains compounds with different levels of degradability, from very easily decomposable to extremely resistant to decomposition. Each C component has a different residence time in the soil and performs different functions (USDA, 2018). Particulate organic C (POC) and microbial biomass C (MBC) are important C fractions that reflect key processes, such as soil aggregation, and nutrient cycling (Wander, 2004). A number of studies have shown that POC and MBC are sensitive to management changes, such as reduced tillage, land use, and cover crop (Wander and Bidart, 2000; Grandy and Robertson, 2007). This sensitivity led to wide adoption of the methods as indicators of change in the soil ecosystem (Wander, 2004). However, POC and MBC are expensive measures due to the required labor and combustion analyzer to quantify the total C in the extracted fraction. Despite the cost, there is a large degree of variation in how researchers extract and define POC and MBC fractions. For example, POC can be fractionated by size or by density. MBC can be measured by chloroform fumigation-extraction or direct extraction after chloroform fumigation. These methodological variations can make comparisons of POC and MBC across studies difficult.

Potassium permanganate was first used to fractionate SOC via oxidation by Loginow et al. (1987). Weil et al. (2003) further developed and streamlined this method, using 0.02 mol L^{-1} $KMnO_4$ to measure the active carbon fraction of SOC. Therefore, this active carbon method was called permanganate oxidizable C (POXC). Reactive carbon is a fraction of the SOM pool that is oxidizable in the presence of potassium permanganate. Reactive carbon originates from the various fractions of SOM, including fresh organic

material, microbial biomass, particulate organic matter, other easily metabolized organic compounds, and C loosely bound to soil minerals. Reactive carbon is most readily degradable by microorganisms; however, it also includes the C bound to soil minerals. Because of this association to the mineral fraction, reactive carbon is considered a chemical indicator, not a biological indicator. Reactive carbon is more sensitive to management difference because of its relatively short turnover time compared to total organic carbon.

Culman et al. (2013) used a long-term trial to determine the temporal dynamics and long-term response of several simple measures of labile C and N to management. They found that measured labile SOM indicators (reactive carbon, C mineralization, and N mineralization) were able to reflect both short- and long-term dynamics in corn-based cropping systems in the upper Midwest. Reactive carbon was the most sensitive indicator of both management and crop rotational diversity. The history of crop rotation had a greater influence than a management regime on all soil measures, with the exception of reactive carbon, which made reactive carbon a better indicator of reflecting management difference in the short term. Weil et al (2003) showed that reactive carbon was related to most measures of soil microbial activity, including MBC, soluble carbohydrate C, and total SOC. Culman et al. (2010) found significantly positive relationships between reactive carbon and microbial biomass. Culman et al. (2011) found that POXC was significantly related to all soil C fractions, including POC, MBC, and SOC, and was more strongly related to heavier and smaller POC fractions, which suggests that POXC reflects a more processed and stabilized pool of labile soil C. POXC also demonstrated greater sensitivity to changes in management compared to POC, MBC, and SOC. However, the

relevance of reactive carbon in soil processes is not unequivocal due to the structural and functional heterogeneity.

Soil biological indicators provide information into the living component of soil. Biological indicators reflect the potential of soil to perform ecosystem functions and therefore be good candidate to assess soil quality. These indicators are dynamic and sensitive to land management. They respond rapidly to changes in the environment, such as drought and substrate stress, soil management and climate variability. Therefore, they have been suggested as soil quality indicators (Laishram et al., 2012), including earthworms, microbial diversity and activity, biomass, respiration and enzyme activities.

Among the soil fauna, earthworms are the most widely promoted as biological indicators. Earthworms are considered to be soil engineers, as they can modify soil structure and features with their etho-physiological action (Blouin et al., 2013). Low earthworm populations are an indicator of little or no organic residues inputs to soil and can affect drainage and aggregate stability, which in turn would cause low crop productivity. Earthworms are important not only because of their role in the soil but because of their implication in crop production. Van Groeningen et al. (2014) reviewed the literature and argued that the presence of earthworms can significantly increase crop yield by twenty-five percent. However, seasonal and climatic variations affect their abundance, distribution, and activity. For example, they are most active in the spring and autumn.

Soil respiration is one measure of biological activity and decomposition. It is defined as carbon dioxide being released from the soil surface through aerobic microbial decomposition of SOM. Because organic nutrients in organic matter are converted to

available inorganic forms for plant uptake, soil respiration is also known as carbon mineralization. Beneficial management that affects SOM, aggregation, and moisture can boost soil respiration. Higher soil respiration always indicates better soil quality. However, optimal conditions for soil respiration varies across soil types and climate regions. For example, clay particles would protect SOM from decomposition and reduce soil respiration, and microbial respiration more than doubles for every 10 °C rise, but the respiration decreases beyond a limiting temperature. Therefore, comparison of soil respiration across sites or seasons is not possible.

Phospholipid fatty acid and DNA are also gaining popularity in soil quality assessment. The molecular methods focusing on DNA and RNA hold great potential to perform faster, cheaper, and more informatively. Microbial biomass, composition, and activity are potentially useful soil quality indicators because they are linked to SOM dynamics and nutrient cycling (Gregorich et al. 1997; Bastida et al., 2006; Bastida et al., 2008), as well as sensitive to soil disturbance and changes due to tillage (Simard et al., 1994; Gregorich et al., 1997; Wander and Boller, 1999; Balota et al., 2004; Franchini et al., 2007). Acosta-Martinez et al. (2008) found differences in soil microbial community structures under pastures and trees compared to agricultural soils under vegetable plots. However, some biological measures may be too temporally responsive to short-term environmental factors (e.g. precipitation events, tillage, or inputs of organic matter), making it difficult to calibrate and interpret the assay. For example, Eric et al. (2001) found seasonal fluctuation in microbial diversity.

Although a lot of indicators have been proposed, a globally acceptable and applicable definition and methodology of assessment of soil quality is not in place

(Laishram et al., 2012). The Haney soil health test developed by Rick Haney and his USDA colleague Daren Harmel was designed to assess soil health. Traditional soil testing methods measured soil N, P, K, soil pH and SOM. However, the Haney soil heath test accounts for the contribution of soil microbes. Microbes can mineralize organic phosphorous and nitrogen and make them available to the crop. A few commercial laboratories already offer the Haney soil health test, but extensive field calibration research is required to confirm interpretations of the test. In recent years, the Cornell Soil Health Laboratory (CASH) developed the comprehensive assessment of soil health, which is able to identify physical, biological, and chemical measures to provide farmers an assessment of their fields' current soil health (Moebius-Clune et al., 2016). Roper et al. (2017) ran the CASH and Haney tests on the long-term replicated experiments in North Carolina. They had soil management (high tillage) that supposedly degraded soils indeed got high scores.

Velasquez et al. (2007) proposed that a general indicator of soil quality based on fifty soil properties including organic matter, soil morphology, physical condition, chemical fertility and biological properties. This indicator allows the evaluation of soil quality and monitoring of change but was only valid at a regional scale (Velasquez et al., 2007). Other soil quality assessment systems have also been developed, such as soil conditioning index (Abrahamson et al., 2007).

There is no globally acceptable calibrated measure or indexes of soil quality. Roper et al. (2017) showed that two soil quality indicators perform poorly.

Soil Quality in Relation to Crop Yields

Soil quality measurements need to be linked to important soil functions and used to predict sustainability or productivity (Herrick, 2000). High soil quality should maintain high productivity without significant soil or environmental degradation (Govaerts et al., 2006). Roper et al. (2017) submitted soil samples from different landscapes and under different management to the Haney soil health test and the Cornell comprehensive assessment of soil health for analysis. These two tests gave different scores for the same soil, and no correlation between soil health tests and crop yields was found. Roper et al. (2017) believed that current soil health indicators were limited by intrinsic soil properties. Hose et al. (2013) studied the effects of farm compost amendment on soil quality and four crops in a six-year field study. SOC, N content, and microbial biomass were remarkably increased when farm compost was applied. However, only potato yield was significantly higher after the application of farm compost.

Govaerts et al. (2006) established a minimum soil quality data set for a long-term tillage, residue management, and rotation trial for wheat and maize production systems. Several physical (e.g. aggregate stability) and chemical (e.g. soil C, N, K) indicators were chosen to assess soil quality. Zero tillage with crop residue retention improved the chemical and physical conditions of soil; however, a relationship of crop yield with these properties was not observed. A major challenge for any soil quality indicator to correlate with crop yield is that so many other factors such as seasonal weather, pests and diseases affect yields. Effects from these factors may confound the ability of soil quality indicators to predict crop yield.

Soil Quality Minimum Data Sets and Indexes

Although increasing the number of indicators can increase collinearity, complexity and costs of measurements would become prohibitive **(**Bünemanna et al., 2018). Therefore, the number of soil quality indicators needs to be reduced to a minimum data set. Larson and Pierce (1991) proposed that a minimum data set was needed for assessing soil health and standardizing methodologies and procedures. Burger (1997) and Powers et al. (1998) also proposed minimum data sets in forest soil quality. Their choices for a minimum data set were soil properties that are sensitive to management and can indicate these changes in a relatively short time and are related to soil health.

In summary, the criteria for potential soil quality indicator is that it can detect changes rapidly (a few years), can be calibrated, interpreted independent of soil type (a major obstacle for soil measures as soil quality indicators), has high throughout capability, is cost-effective, has seasonal stability, is accessible to farmers and land managers, and can be related to economic indicators (crop yield).

In the first, the selection was based on expert judgement, then statistical data reduction was done using multivariate techniques, discriminant analysis, and multiple regression. Using these techniques, the number of indicators finally selected typically ranges between 6 and 8 (Bünemanna et al., 2018). Ritz et al. (2009) presented a participatory approach for selecting soil biological indicators. Among 183 candidate biological indicators, a rank of twenty-one indicators was produced, scored by scientists and end users in a logical-sieve approach. The selection of a minimum data set from a larger set of soil quality indicators is necessary due to time and cost limitations.

After the minimum data set is determined, the data transformation step typically involves scoring the indicator variable on a zero to one scale. Various linear or non-linear mathematical functions are used to generate scores (Velasquez et al., 2007). However, there is little agreement on standards for data interpretation. Roper et al. (2017) tested the Haney soil health test and comprehensive assessment of soil health on various agricultural systems and found that scores between these two soil quality tests did not consistently give the same ranking. Therefore, consistent sampling and analytical methods are needed when adopting soil quality assessment.

Furthermore, some dynamic indicators are termed pedotransfer functions (Bouma, 1989) and are generally used to describe functions in which routinely measured properties are used to predict other properties that may be more difficult or practical to measure. Pedotransfer functions facilitate the adoption of a minimum data set to build models for soil quality assessment. Benjamin and Karlen (2014) evaluated five pedotransfer functions to determine the effectiveness of the least limiting water range in predicting soil water-holding capacity.

Enzyme Activities as Soil Quality Indicators

There is growing evidence that soil biological parameters, particularly soil enzyme activities, hold potential as early and sensitive indicators of soil ecological stress or restoration (Dick and Tabatabai, 1992; Dick, 1994; Dick et al., 1996; Dick et al., 1988a; Dick et al., 1988b; Bandick and Dick, 1999; Ndiaye et al., 2000; Ndour et al., 2001; Hinojosa et al., 2004; Acosta-Martinez et al., 2008; Vallejo et al., 2012).
As Doran and Parkin (1994) suggested, soil enzyme activity is an index that can integrate biological, chemical, and physical characteristics and that can be used to monitor long-

term effects of soil management. Taylor et al. (2002) mentioned two reasons for measuring soil enzymes. The first reason is that enzyme can provide information about the progress of remediation operations or the sustainability of particular land management. The second reason is that enzyme assays can reflect biological potential and possible resilience to environmental stress. Aside from these benefits, soil enzyme activities offer the potential to be sensitive to land management within 1–3 years, but seasonally stable to reflect the trajectory of a given system on the status of the soil (Bandick and Dick, 1999; Ndiaye et al., 2000).

Soil enzymes are found in three broad categories: 1) those associated with viable cells, either internally or on cellular surfaces; 2) excreted enzymes in soil solution; and 3) extracellular enzymes stabilized on soil colloids (Burns, 1982). The latter two groups have been referred to as "abiontic," a term coined by Skujinš (1976) to describe enzymes of biological origin but no longer associated with living cells. Stabilized enzymes on clays or humic colloids can remain catalytic, with typically 40%-60% of the activity associated with abiontic forms of many enzymes (Nannipieri et al., 1996; Knight and Dick, 2004). Abiontic enzyme activity provides a mechanism for the suitability of soil enzyme activities as a practical dynamic soil property. First, management systems that protect and improve soils through less disturbance and greater C inputs will stimulate microbial populations and enzyme production. Second, it seems plausible that practices that promote aggregation/organic matter accumulation would also promote the stabilization and protection of abiontic enzymes in the soil–humic matrix (Knight and Dick, 2004). Therefore, enzyme activity should provide useful information on whether soil management promotes SOM development long before measurable changes in organic

C can be detected because soil enzyme activity assays are much more sensitive to soil management than total C analysis is.

Another important characteristic for a dynamic soil property is the need for temporal sensitivity, which has seasonal stability yet detects the effects of land management trajectory on the status of a soil. Although biological properties would be expected to be temporally sensitive, measures associated with the viable population may be too sensitive (Ndiaye et al., 2000) to recent environmental conditions (e.g. moisture and temperature) or management practices (e.g. incorporating fresh plant material). In these cases, a temporary and large change in microbial responses may obscure the true status of a region's soil. This is another advantage of soil enzyme activity that has a significant amount of its activity associated with the abiontic fraction, because accumulation of the stabilized form is incremental and therefore moderates the more temporally variable enzyme activity of viable cells. Therefore, soil enzyme activities are temporally sensitive and seasonally stable. Knight and Dick (2004) used microwave irradiation to denature β-glucosidase associated with viable cells and found that soils under long-term agricultural practices were statistically different based on abiontic activity and not on viable cell activity. Moreover, Acosta-Martinez et al. (2008) found no significant difference in the released intracellular arylsulfatase activity, which represented 47% of the total arylsulfatase activity, under different managements. However, total arylsulfatase activity, determined in chloroform-fumigated soils, showed difference under different land use.

Abiontic enzyme activity also provides the mechanism for calibrating enzyme activities as a soil quality indicator, in dependent of soil type and by reducing seasonal

effects. Roper et al. (2017) stated that the soil quality indicator needs to be calibrated across soil types and climates. A major limitation of most measures of soil properties is that they vary considerably, simply as a function of the soil type and season. These differences can be much greater than the subtle changes due to land management, making the interpretation of dynamic soil measures difficult (Schutter et al., 2001). This is true for enzyme activities; however, research across diverse soils in Oregon (Knight, 2002; Knight and Dick, 2004) and soils in Columbus (Vallejo et al., 2012) showed that normalizing enzyme activity to either C or clay content can separate land management effects independent of soil type. Thus, enzyme activity seems to be a rare soil measure that could be calibrated and interpreted directly independent of soil type.

A soil indicator should have a relationship with soil structure, particularly aggregation. A major impact of cultivation and poor soil management is loss of aggregation (Gupta and Germida, 1988; Borchers and Perry, 1992). Loss of aggregation further exacerbates soil degradation by decreasing porosity, which in turn reduces water infiltration and storage and increases vulnerability to erosion. Although it is unlikely that soil enzymes directly participate in soil structure development, there is evidence that soil biology has an important role in developing soil structure. Indeed, studies on compaction from forestry and agricultural practices have shown significant negative correlations of soil bulk density and positive correlations of water infiltration rates (Dick et al. 1988a; Reganold, 1988; Martens et al. 1992) and aggregation (Miller and Dick, 1995ab) with enzyme activities. Thus, soil enzyme activities would be more a practical surrogate for reflecting changes in soil structure because the procedure is considerably simpler than the labor-intensive procedures of many standard soil structural methods.

For commercial labs and agencies to adopt an indicator, rapid soil handling and high throughput procedures are desirable. A major advantage of enzyme assays in this regard is that air-dried soils can be used. Bandick and Dick (1999) and Lee et al. (2007) compared field-moist and air-dried soils from different management systems and found that although activity went down in most cases, the ranking of treatments on air-dried samples was the same as that of field-moist samples. They hypothesized that air drying could further improve the ability of enzyme assays to give the true trajectory of management effects on soils because air drying would be expected to denature enzymes associated with viable populations (reducing the effects of recent management or environmental impacts on the highly sensitive and temporally variable living microbial enzyme pool). Thus, more of the activity is associated with enzymes stabilized in the soil matrix that are less susceptible to recent environmental conditions and better reflect the long-term trajectory on soil dynamics or health.

A further advantage for practical applications is that, unlike most other measures of soil microbial properties, enzyme assays are relatively simple to perform. Soils are typically incubated for one or just a few hours, followed by colorimetric or fluorescence determination of reaction products. Assays can be adapted for multi-enzyme, high throughput activity analyses using 96-well microplates (Dick at al., 2013; Dick et al., 2018). Thus, the ability to use air-dried samples combined with a simple methodology allows for large-scale processing and analysis of samples for practical applications, with commercial soil testing labs.

Another major challenge for any soil quality indicator is that it correlates with crop yield because abiotic and edaphic factors can control yield. One major factor, if not

the largest, is drainage (personal communication, Laura Lind said, 2018). A potential way to account for the confounding effects of drainage on yields might be to include an oxidation-reduction assay as covariant with the enzyme assays that can detect soil management effects. It could be expected that they will be highly correlated with landscape position and drainage and therefore with crop yield. For reduction enzyme activities, there should be a negative correlation with increasing drainage capability, whereas an oxidation assay would be expected to have a negative correlation. The goal would be to ultimately use an oxidation assay as a co-variant with other enzyme data to determine whether this improves the relationship of enzyme-based soil quality indicators with crop yield.

In sum, soil enzyme activity assays are advantageous as a potential indicator for soil quality because a) of operational practicality, b) they are sensitive integrative 'biological fingerprints' of past soil management, and c) they are accessible and cost-effective.

CONCLUSIONS

An integrative soil quality indicator is needed for the management of sustainable agriculture. To guide land managers and policy makers, the indicator needs to be sensitive to management changes, seasonally stable, cost-effective, and easily adopted. Enzyme activities have the potential to meet the above criteria. However, not all enzyme assays tested so far meet these conditions, and many more could be considered but need to be evaluated for their potential as soil quality indicators. The criteria of choosing enzyme assays are based on their sensitivity to land management, importance in nutrient

cycling, and simplicity. Deaminase was not a good soil quality indicator, while β-glucosidase was suggested as a soil quality indicator due to its importance in the C cycle, seasonal stability, and land management treatment (Bandick and Dick, 1999). Enzyme assays in the N and P cycles should be avoided, as activities of these enzymes are suppressed by repeated applications of inorganic fertilizers or strongly influenced by pH or liming (Chunderova and Zubets, 1969; Mathur and Rayment, 1977; Spiers and McGill, 1979; Dick et al. 1988b; McCarty et al. 1992; Clarholm, 1993; Dick, 1994). Furthermore, β-glucosidase and arylsulfatase are two possible soil quality indicators, as they have consistently detected land management changes (Bandick and Dick, 1999; Ndiaye et al., 2000; Acosta-Martinez et al., 2008). In this thesis, a novel idea that was investigated is oxidation-reduction enzyme assays as a soil quality indicator to be correlated with crop productivity. Rhodanese and ammonium oxidation enzymes (ammonia monooxygenase) were investigated as a preliminary screening tool for developing a suitable assay.

REFERENCES

Abrahamson, D. A., M. L. Norfeet, H. J. Causarano, J. R. Williams, J. N. Shaw and A. J. Franzluebbers. 2007. Effectiveness of the soil conditioning index as a carbon management tool in the southeastern USA based on comparison with EPIC. Journal of Soil and Water Conservation 62(2): 94–102.

Acosta-Martinez, V., D. Acosta-Mercado, D. Sotomayor-Ramirez, and L. Cruz-Rodriguez. 2008. Microbial communities and enzymatic activities under different management in semiarid soils. Applied Soil Ecology 38: 249- 260.

Aune, J.B., Lal, R., 1997. The tropical soil productivity calculator — a model for assessing effects of soil management on productivity. In: Lal, R., Stewart, B.A. (Eds.), Soil Management — Experimental Basis for Sustainability and Environmental Quality. Adv. Soil Sci., Lewis Publishers, London, UK, pp. 499–520.

Balesdent, J., C. Chenu, M. Balabane, 2000. Relationship of soil organic matter dynamics to physical protection and tillage. Soil and Tillage Research 53: 215-230.

Balota, E.L., A.C. Filho, D.S. Andrade, R.P. Dick. 2004. Long-term tillage and crop rotation effects on microbial biomass and C and N mineralization in a Brazilian Oxisol. Soil and Tillage Research 77: 137–145.

Bandick, AK. and RP. Dick. 1999. Field management effects on enzyme activities. Soil Biology and Biochemistry 31:1471-1479.

Bastida, F., Moreno, J.L., Hernández, T., García, C. 2006. Microbiological degradation index of soils in a semiarid climate. Soil Biology and Biochemistry 38: 3463–3473.

Benjamin, J. G., D. L. Karlen. 2014. LLWR techniques for qualifying potential soil compaction consequences of crop residue removal. Bioenergy Research 7: 468-480.

Bastida, F., Zsolnay, A., Hernández, T., García, C., 2008. Past, present and future of soil quality indices: a biological perspective. Geoderma 147: 159–171.

Bhagat, S.P. 1990. Creation in crisis. Brethren Press, Elgin, lL.

Bloem, J. Schouten, A. J. Sørensen, S. J. Rutgers, M. Werf, A. van der Breure, A. M. 2006. Microbiological methods for assessing soil quality 3: 23-49.

Blouin,M., M.E. Hodson, E.A. Delgado, G.Baker, L. Brussard, K.R. Butt, J. Dai, L.Dend ooven, G. Peres, J.E. Tondoh, D.Cluzeau, J.J. Brun. 2013. A review of earthworm

impact on soil function and ecosystem services. European Journal of Soil Science, 64: 161-182

Borchers, J. G., and D. A. Perry. 1992. The influence of soil texture and aggregation on carbon and nitrogen dynamics in southwest Oregon forests and clear cuts. Canadian Journal of Forest Research 22:298-305.

Bouma, J. 1989. Using soil survey data for quantitative land evaluation. Advances in Soil Science 9: 177-213

Bünemanna, E. K., G. Bongiornoa, Zhanguo Bai, R. E.Creamer, G. De Deyn, R. Goede, L. Fleskens, V. Geissen, T. W. Kuyper, P. Mäder, M. Pulleman, W. Sukkel, J. W. van Groenigen, L. Brussaard. 2018. Soil quality-a critical review. Soil Biology and Biochemistry 120: 105-125.

Burger, J.A. 1997. Conceptual framework for monitoring the impact of intensive forest management on sustainable forestry. In: Hakkila, P., Heino, M., Puranen, E. (Eds.), Proceedings of a Joint IEA Task XII Meeting on Forest Management for Bioenergy, Jyvaskyla, Finland. Finnish Forest Research Institute, Vaanta, Finland, pp. 147–156.

Burns, R.G. 1982. Enzyme activity in soil: Location and a possible role in microbial ecology. Soil Biology and Biochemistry 14:423-427.

Chunderova, A.I. and T. Zubets. 1969. Phosphatase activity in dernopodzolic soils. Pochvovedeniye 11:47-53.

Clarholm, M. 1993. Microbial biomass P, labile P, and acid phosphatase activity in the humus layer of a spruce forest, after repeated additions of fertilizers. Biology and Fertility of Soils 16:287-292.

Corstanje, R., T.G. Mercer, J. R. Rickson, L. K. Deeks, P. Newell-Price, I. Holman, C. Kechavarsi, and T. W. Waine. 2017. Physical soil quality indicators for monitoring British soils. Solid Earth 8: 1003-1016.

Culman S.W., M. A. Freemana, M. E. Schipanskib, J. Benistonc, R. Lalc, L. E. Drinkwaterd, A. J. Franzluebberse, J. D. Gloverf, A. Stuart Grandyg, J. Leeh, J. Sixh, J. E. Mauli, S. B. Mirksyi, J. T. Spargoi and M. M. Wanderj. 2011. Permanganate oxidizable carbon reflects a processed soil fraction that is sensitive to management. Soil Science Society of America Journal 76:494-504.

Culman, S. W., S. S Snapp, J. M Green, L. E Gentry. 2013. Short- and Long-term labile soil carbon and nitrogen dynamics reflect management and predict corn agronomic performance. Agronomy Journal 105: 493-502.

Culman, S.W., S.T. DuPont, J.D. Glover, D.H. Buckley, G.W. Fick, H. Ferris, and T.E. Crews. 2010. Long-term impacts of high-input annual cropping and unfertilized perennial grass production on soil properties and belowground food webs in Kansas, USA. Agriculture, Ecosystem and Environment 137: 13–24.

Dick, R. P. 1994. Soil enzyme activities as indicators of soil quality. *In* Doran et al. (eds.) Defining Soil Quality for a Sustainable Environment. Soil Science Society of America Special Publication, Madison, WI, pp. 107-124.

Dick, R. P., D. Breakwill, and R. Turco. 1996. Soil enzyme activities and biodiversity measurements as integrating biological indicators. p. 247-272. In J.W. Doran and A.J. Jones. (eds.). Methods for Assessing Soil Quality. Soil Science Society of America Special Publication 49. Madison WI. pp. 247-272.

Dick, R.P., D.D. Myrold, and E.A. Kerle. 1988a. Microbial biomass and soil enzyme activities in compacted and rehabilitated skid trail soils. Soil Science Society of America Journal 52:512-516.

Dick, R. P., P. E. Rasmussen and E. A. Kerle. 1988b. Influence of long-term residue management on soil enzyme activities in relation to soil chemical properties in a wheat-fallow system. Biology and Fertility Soils 6:159-164.

Dick, W.A., and M.A. Tabatabai. 1992. Potential uses of soil enzymes. *In* F.B. Metting, Jr. (ed.) Soil microbial ecology: Applications in agricultural and environmental management. Marcel Dekker, Inc., New York, pp. 95-127.

Dick, P. R., L. K. Dick, S. Deng, Xiufen Li, E, Kandeler, C. Poll, C. Freeman, T. G. Jones, M. N. Weintraub, K. A. Esseili, and J. Saxena. 2018. Cross-laboratory comparison of fluorometric microplate and colorimetric bench-scale soil enzyme assays. Soil Biology and Biochemistry 121: 240-248.

Doran, J.W., Parkin, T.B., 1994. Defining and Assessing Soil Quality. In: Doran, J.W., Coleman, D.C., Bezdicek, D.F., Stewart, B.A. (Eds.), Defining soil quality for a sustainable environment. Soil Science Society of America, Madison, pp. 3-21.

Franchini, J.C., C.C. Crispino, R.A. Souza, E. Torres, M. Hungria. 2007.Microbiological parameters as indicators of soil quality under various soil management and crop rotation systems in southern Brazil. Soil Tillage Research 92: 18–29.

Govaerts, B., K.D. Sayre, J. Deckers. 2006. A minimum data set for soil quality assessment of wheat and maize cropping in the highlands of Mexico. Soil and Tillage Research 87: 163-174.

Govaerts, B., K. D. Sayre, J. Deckers. 2005. A minimum data set for soil quality assessment of wheat and maize cropping in the highlands of Mexico. Soil and Tillage Research 87: 163-174.

Granatsein, D., and D.F. Bezdicek. 1992. The need for a soil quality index: Local and regional perspectives. American Journal of Alternative Agriculture 17:12-16.

Grandy, A.S., and G.P. Robertson. 2007. Land-use intensity effects on soil organic carbon accumulation rates and mechanisms. Ecosystems 10: 58– 73.

Gregorich, E.G., M.R. Carter, J.W. Doran, C.E. Pankhusrst, L.M. Dwyer. 1997. Biological attributes of soil quality. In: Gregorich, E.G., Carter, M.R. (Eds.), Soil quality for crop production and ecosystem health. Elsevier, Amsterdam, pp. 81-113.

Gupta, V.V.S.R., and J.J. Germida. 1988. Distribution of microbial biomass and its activity in different soil aggregate size classes as affected by cultivation. Soil Biology and Biochemistry 20:777-786.

Haberern, J. 1992. Viewpoint: A soil health index. J. Soil Water Conservation 47:6.

Henderson, G.S., 1995. Soil organic matter: a link between forest management and productivity. In: McFee, W.W., Kelly, J.M. (Eds.), Proceedings of the 8th North American Forest Soils Conference on Carbon Forms and Functions in Forest Soils. Soil Science Society of America., Madison, WI, pp. 419–435.

Herrick, J.E. 2000. Soil quality: an indicator of sustainable land management? Applied Soil Ecology 15: 75-83.

Hinojosa M.B, J.A. Carreira, R. García-Ruíz, and R.P. Dick. 2004. Soil moisture pre-treatment effects on enzyme activities as indicators of heavy metal-contaminated and reclaimed soils. Soil Biology and Biochemistry 36-1559-1568. American Journal of Alternative Agriculture 7:17-24.

Hose, T. D., M. Cougnon, A. De Vliegher, B. Vandecasteele, N. Viaene, W. Cornelis, E. V. Bockstaele, D. Reheul. 2013. The positive relationship between soil quality and crop production: A case study on the effect of farm compost application. Applied Soil Ecology 75: 189-198.

Kay, B.D., C.D. Grant. 1996. Structural aspects of soil quality. In: MacEwan, R.J., Carter, M.R. (Eds.), Soil Quality is in the Hands of the Land Manager. Centre for Environmental Management, University of Ballarat, Victoria, Australia, pp. 37–41.

Knight T, and R. P. Dick. 2004. Differentiating microbial and stabilized β-glucosidase activity relative to soil quality. Soil Biology and Biochemistry 36: 2089-2096.

Knight, T. 2002. Soil enzyme stability in disturbed ecosystems. MS thesis. Oregon State University, Corvallis OR.

Larson, W.E., and F.J. Pierce. 1991. Conservation and enhancement of soil quality. In Evaluation for sustainable land management in the developing world. Vol. 2. IBSRAM Proc. 12 (2). Bangkok, Thailand. Int. Board for Soil Research and Management.

Laishram, J., K. G. Saxena, R. K. Maikhuri and K. S. Rao. 2012. Soil quality and soil health: A review. International Journal of Ecology and Environmental Sciences 38(1): 19–37.

Loginow, W., W. Wisniewski, S. Gonet, and B. Ciescinska. 1987. Fractionation of organic C based on susceptibility to oxidation. Polish Journal of Soil Science 20: 47–52.

Martens, D.A., J.B. Johanson, and W.T. Frankenberger, Jr. 1992. Production and persistence of soil enzymes with repeated additions of organic residues. Soil Science 153:53-61.

Mathur, S.P. and A.F. Rayment. 1977. Influence of trace element fertilization on the decomposition rate and phosphatase activity of a mesic fibrisol. Canadian Journal of Soil Science 57: 397-408.

McCarty, G.W., D.R. Shogren, and J.M. Bremner. 1992. Regulation of urease production in soil by microbial assimilation of nitrogen. Biology and Fertility of Soils 12: 261-264.

Miller, M. and R.P. Dick. 1995a. Dynamics of soil C and microbial biomass on whole soil and aggregates in two cropping systems differing in C-input. Applied Soil Ecology 2:253-261.

Miller, M., and R. P. Dick. 1995b. Thermal stability and activities of soil enzyme activities as influenced by crop rotation Soil Biology and Biochemistry 27:1161-1166.

Moebius-Clune, B.N., D.J. Moebius-Clune, B.K. Gugino, O.J. Idowu, R.R. Schindelbeck, A.J. Ristow, H.M. van Es, J.E. Thies, H.A. Shayler, M.B. McBride, K.S.M Kurtz, D.W. Wolfe, and G.S. Abawi, 2016. Comprehensive Assessment of Soil Health – The Cornell Framework, Edition 3.2, Cornell University, Geneva, NY.

Nannipieri, P., P. Sequi and P. Fusi. 1996. Humus and enzyme activity. *In* A. Piccolo (ed.) Humic Substances in Terrestrial Ecosystems. Elsevier, New York, pp. 293-328.

Ndiaye, EL, J.M. Sandeno, D. McGrath, and R.P. Dick. 2000. Integrative biological indicators for detecting change in soil quality. American Journal of Alternative Agriculture 15:26-36.

Ndour Ndeye Yacine Badiane, J.L Chotte, E. Pate, D. Masse, and C. Rouland. 2001. Use of soil enzyme activities to monitor soil quality in natural and improved fallows in semi-arid tropical regions. Applied Soil Ecology, Vol. 18, Issue 3, pp. 229-238.

Powers, R.F., Tiarks, A.E., Boyle J.R., 1998. Assessing soil quality: practicable standards for sustainable forest productivity in the United States. In: Adams, M.B., Ramakrishna K., Davidson, E.A. (Eds.), The Contribution of Soil Science to the Development and Implementation of Criterial and Indicators of Sustainable Forest Management. Soil Science Society of America, Special Publication No. 53, pp. 53–80.

Reganold, J.P. 1988. Comparison of soil properties as influenced by organic and conventional farming systems. American Journal of Alternative Agriculture 3:144-155.

Reganold, J.P., A.S Palmer. 1995. Significance of gravimetric versus volumetric measurements of soil quality under biodynamic, conventional, and continuous grass management. Journal of Soil and Water Conservation 50: 298-305.

Ritz,

Ritz, K., H.I.J. Black, C.D. Campbell, J.A. Harris, C. Wood. 2009. Selecting biological indicators for monitoring soils: a framework for balancing scientific and technical opinion to assist policy development. Ecological Indicators 9: 1212-1221.

Romig, D.E., Garlynd, M.J., Harris, R.F., 1996. Farmer-based assessment of soil quality: a soil health scorecard. In: Doran, J.W., Jones A.J. (Eds.), Methods for Assessing Soil Quality. Soil Science Society of America, Special Publication No. 49, pp. 39–60.

Roper, W. R., D. L. Osmond, J. L. Heitman, M. G. Wagger and Schris Reberg-Horton. 2017. Soil health indicators do not differentiate among agronomic management systems in north Carolina soils. Soil Science Society, Vol. 81, Issue 4, pp. 828.

Sagan, C. 1992. To avert a common danger. Parade Magazine, March I, pp. 10-14.

Sanders, D.W. 1992. International activities in assessing and monitoring soil degradation. America.

Schoenholtz, S. H., H. Van Miegroet, J. A. Burger. 2001. A review of chemical and physical properties as indicators of forest soil quality: challenges and opportunities. Forest Ecology and Management 138: 335-356.

Schutter, M., J. Sandeno, and R.P. Dick. 2001. Seasonal, soil type, and alternative management influences on microbial communities of vegetable cropping systems. Biology and Fertility of Soils 34 : 397-410.

Simard, R.R., D.A. Angers, C. Lapierre. 1994. Soil organic matter quality as influenced by tillage, lime, and phosphorus. Biology and Fertility of Soils 18: 13–18.

Skujins, J. 1976. Extracellular Enzymes in Soil. CRC Critical Reviews in Microbiology 4:383-421.

Smit, E., P. Leeflang, S. Gommans, J.V.D. Broek, S.V. Mil and K. Wernars. 2001. Diversity and seasonal fluctuation of the dominant members of the bacterial soil community in a wheat field as determined by cultivation and molecular methods. Applied and Environmental Microbiology, vol. 67, no. 5, 2284-2291.

Smith JL and JW Doran. 1996. Measurement and use of pH and electrical conductivity for soil quality analysis. In Methods for assessing soil quality. Soil Science Society of America Special Publication 49: 169-182.

Spiers, G.A. and W.B. McGill. 1979. Effects of phosphorus addition and energy supply on acid phosphatase production and activity in soils. Soil Biology and Biochemistry 11: 3-8.

Taylor, J.P., M. Wilson, S. Mills, R.G. Burns. 2002. Comparison of microbial numbers and enzymatic activities in surface soils and subsoils using various techniques. Soil Biology and Biochemistry 34: 387–401.

Vallejo, V.E., Z. Arbelia, Wi. Terán, N. Lorenz, R. P. Dick, and F. Roldan. 2012. Effect of land management and Prosopis juliflora (Sw.) DC trees on soil microbial community and enzymatic activities in intensive silvopastoral systems of Colombia. Agriculture, Ecosystems and Environment 150 :139– 14.

Velasquez, E., P. Lavelle and M. Andrade. 2007. GISQ, a multifunctional indicator of soil quality. Soil Biology and Biochemistry 39(12): 3066–80.

Van Groeningen, J.W., I.M. Lubbers, H.M.J.Vos, G.G. Brown, G.B. De Deyn, K.J. Van Groenigen. 2014. Earthworms increase plant production: a meta-analysis. Nature, 4.

Wander, M. 2004. Soil organic matter fractions and their relevance to soil function. p. 67–102. In Magdoff, F., and R.R. Weil (ed.) Soil organic matter in sustainable agriculture. CRC Press, Boca Raton, FL.

Wander, M.M., and M.G. Bidart. 2000. Tillage practice influences on the physical protection, bioavailability and composition of particulate organic matter. Biol. Fertil. Soils 32: 360–367.

Wander, M.M., G.A. Bollero. 1999. Soil quality assessment of tillage impacts on Illinois. Soil Science Society of America Journal 63: 961–971.

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.

World Resources Institute. 1992. New York Times, March 3

CHAPTER 2: SOIL ENZYME ACTIVITIES UNDER VARIOUS DRAINAGE CLASSES AND THEIR CORRELATIONS WITH CROP PRODUCTIVITY

ABSTRACT

While most soil quality indicators can be limited by sensitivity, stability, and practical feasibility, enzyme activities, compared to other chemical, physical, and microbial properties of soil are operationally simple and economical feasible for adoption by commercial labs. Enzyme activities have been shown to be sensitive to land management. However, to be effective, soil quality indicators need to be calibrated to enable interpretation of causal relationships between soil quality and ecosystem functions. Ideally, feasible soil quality indicators for agriculture should have a relationship to crop yields. In agriculture, drainage is one of the most important factors that controls crop yields. Oxidation-reduction enzyme activities would be expected to be affected by drainage due to varying redox conditions; however, this is entirely uninvestigated. Therefore, the objective was to determine correlation of oxidation enzymes with crop yields and if oxidation assays are sensitive in detecting differences due to tillage management or drainage class. Soil samples were taken from paired fields of no tillage (NT) and conventional tillage (CT) with most fields under a corn-soybean rotation. Rhodanese (RA) and ammonium oxidation enzyme (AO) were chosen as oxidation enzymes because of short-time incubation (1 to 5 hours). Drainage class information was determined by using the Web Soil Survey. The results showed that crop yields were not correlated with RA or AO activities. Drainage class did not significantly affect RA, AO, or crop yields. Inorganic or organic inputs on soil may confound the results. AO assay detected significantly higher activity in NT at 0-5 cm depth compared to CT at 0-15 cm depth. AO activity/unit clay also indicated this tillage difference. Although correlation of

the enzyme activities with crop yield was not significant, enzyme activities/unit clay increased the correlation coefficients.

INTRODUCTION

Soil is a vital natural resource that, on a human time scale, is nonrenewable (Jenny, 1980). To guide land managers and policy makers, soil indicators are needed that are temporally sensitive and reflect the ability of ecosystems to deliver services. Protecting soils is a national priority and an integral part of protecting the environment but quantifying dynamic soil properties remains an elusive goal. Although soil testing for nutrient status is well established and useful, it does not reflect the ability of soils to provide services, such as resistance to erosion, phytopathogen suppression, or as rooting media for optimal plant growth. To be practical, soil indicators should have high throughput capability, detect changes rapidly, be able to be calibrated and interpreted, and have seasonal stability.

Biological properties of soils hold the potential to meet these criteria for practical applications as soil indicators. They are very responsive temporally and are considered as an integrative indicator, as plant roots and microorganisms (the source of most enzyme activity in soil) depend on optimal soil habitat conditions ,such as soil structure, aeration, water holding capacity, and availability of energy resources (e.g., soil organic matter). All of these properties are related to optimal plant growth and resistance to soil erosion. However, biological properties may be too temporally sensitive as soil dynamic properties. Short-term environmental factors, such as precipitation, tillage, or inputs of organic matter have an important influence on biological properties, making it difficult to calibrate and interpret the assay. Chemical and physical properties have even been used as crude measures of soil tilth. Most notably, determination of soil organic matter has been related to general soil tilth, but tilth does not account for important properties

contributing to soil quality, such as soil structure and microbial diversity. Measurable changes in soil organic matter generally are too slow in responding to perturbations to be useful to land manager. However, soil enzyme activities offer the potential to be sensitive to land management within 1 to 3 years and seasonally stable to reflect the trajectory of a given soil management system on the status of the soil (Bandick and Dick, 1999; Ndiaye et al., 2000). Also, soil enzyme activities are more responsive to environmental factors than soil organic matter.

Soil enzymes are found to be associated with viable cells and excreted in soil solution or stabilized on soil colloids (Burns, 1982). These abiontic enzymes that are no longer associated with living cells but remain catalytic are called "abiontic," a term coined by Skujins (1976). Typically, 40-60% of the activities are associated with abiontic forms (Nannipieri et al., 1996; Knight and Dick, 2004). Abiontic enzyme activity provides a mechanism for the suitability of soil enzyme activities as a practical dynamic soil property. Practices that protect and improve soils stimulate microbial populations and enzyme production. Moreover, it is possible that practices that promote organic matter accumulation would promote stabilization and protection of abiontic enzymes in the soil-humic matrix (Knight and Dick, 2004).

Therefore, enzyme activity can provide useful information on whether soil management is promoting soil organic matter. Another advantage of soil enzyme activity is that a significant amount is associated with the abiontic fraction, which moderates the more temporally variable enzyme activity of viable cells. Therefore, soil enzyme activities are temporally sensitive and seasonally stable. Indeed, Knight and Dick (2004) used microwave irradiation to denature β-glucosidase associated with viable cells and

found that soils under long-term agricultural practices were statistically different based on abiontic activity and not by viable cell activity.

An effective soil indicator should have a relationship to soil structure, particularly aggregation. A major impact of cultivation and poor soil management is loss of aggregation (Gupta and Germida, 1988; Borchers and Perry, 1992). Loss of aggregation will further exacerbate soil degradation. Studies on compaction from forestry and agricultural practices have shown significant negative correlations of soil bulk density, and positive correlations of water infiltration rates (Dick et al., 1988a; Reganold, 1988; Martens et al., 1992) and aggregation (Miller and Dick, 1995ab) with enzyme activities. Thus, soil enzyme activities may be a more practical means for reflecting change in soil structure because the procedure is simpler than the labor-intensive procedures of standard soil structural methods. Besides these benefits, for certain enzyme assays that have been tested, air-dried soil samples can be used (Bandick and Dick, 1999). This advantage could facilitate adoption by commercial labs.

One challenge for any soil quality indicator is that it should correlate with crop yield. However, crop growth and yield are controlled by complex abiotic and edaphic factors. There is very little information relating yields to soil quality. No relationship of the soil quality (e.g., soil respiration) and crop yield was found (Roper et al., 2017; Hose et al., 2013. Landscape position and drainage are major controllers of yields and could confound correlation between a soil quality indicator and yield. For example, Wright et al. (1990) found higher crop yields on summits. To address this challenge, oxidation-reduction enzyme assays were investigated, as they are expected to be highly correlated with landscape position and drainage and, therefore, with crop yield.

Rhodanese (RA) and ammonium oxidation enzyme (AO) were tested along drainage gradients because incubation time is short (1 to 5 hours) and reaction product is single and easily measurable. The initial idea was to take soil samples along landscape gradients, but, as fields where soil samples were collected were almost plains, the Web Soil Survey was used to determine drainage class of fields. Drainage class refers to the frequency and duration of wet periods under conditions similar to those under which the soil formed. Three classes of natural soil drainage are recognized on the fields: poorly drained, somewhat poorly drained, and well drained. These classes are defined in the Soil Survey Manual and were derived from Web Soil Survey.

A second challenge is to directly interpret soil quality tests independent of soil types. This is because most chemical, physical, and biological properties vary widely due to soil type (especially due to textural difference), and the variation is much greater than the variation due to soil management.

One solution that previous research supports is to apply an enzyme activity/unit clay ratio as a soil indicator that is interpretable independent of soil type (Knight, 2002; Vallejo et al., 2010). Previous results in our lab on the soil quality research from long-term plots across the United States showed enzyme activity/unit clay discriminated management systems that ranged from improving to degrading soils, in dependent of soil types. The objective of this research is to determine whether RA and AO could detect difference in soil tillage treatments or drainage class.

MATERIALS AND METHODS

Site Description

The study was done at eighteen sites overall: six sites were located in Iowa, six in Illinois, and six in Ohio. At each site, two side-by-side fields were selected; one field was under no-tillage (NT) and the other was under conventional tillage (CT) with disturbance and soil mixed to a 15 cm depth. Most fields had a corn-soybean rotation. Properties of sites are shown in Table 2.1.

Experimental Design, Yield Estimation, and Soil Sampling

Soil samples were collected from nine paired Ag Spectrum and conventional fields in Illinois (3 sites), Iowa (3 sites), and Ohio (3 sites) in September 2017. Crop yields were measured in September 2015, 2016, and 2017. Each paired site was adjacent. To measure crop yield for a site, five plants were harvested that had one ear/plant or ten soybeans plant were randomly harvested in 5 m (the 5 m section of a row equals $1/1000^{th}$ of an acre) of a row at the site, and the total number of plants in the row was recorded. Corn plants and soybean plants were dried in oven (65 ℃) and grains were removed and weighed. Corn or soybean yield based on a dry weight was calculated as follows (Nielsen, 2015):

Yield (Mg ha^{-1}) = weight of grains from 5 or 10 plants × number of plants in 5 m section × 1000.

To reduce year-to-year variability and allow yield comparisons regardless of crop, the relative yield was calculated as the actual treatment yield (NT or CT) divided by average yield at each site. All sites were sampled by taking 15 soil cores per sampling location. At NT sites, samples were taken in 0-5 cm depth and 5-15 cm depth. For CT

sites, samples were taken in 0-15 cm depth tillage which is the typical depth to which crop residues are incorporated by tillage. Soil samples were placed in bags and brought back to the laboratory and stored at -4°C until analysis. All samples were passed through a 2-mm sieve, and moisture was measured prior to enzyme analysis.

Soil Enzyme Activity Analyses

RA enzyme was determined according to the methods described by Tabatabai and Singh (1976). Four g of soil were incubated in Erlenmeyer flasks (50 ml) at 37 °C, with 8ml of THAM buffer, 1 ml 0.1 M $Na_2S_2O_3$, and 1 ml 0.1 M KCN. After 1 h, $CaSO_4$- formaldehyde solution was added, the suspension was filtered, and SCN^- was determined colorimetrically. For each soil sample, one replication were applied. The control samples were performed by the same procedures for RA activity, but the soil was autoclaved. Standard ferric thiocyanate solution was used to determine the calibration curve. Activity of RA was expressed as SCN^- nmol g^{-1} h^{-1}.

AO enzyme was determined by the method of International Organization of Standardization (ISO 15685, 2004), which was first described by Berg and Rosswall (1985) as an estimation of the potential and actual oxidation rates of ammonium oxidizers. Five g of soil were incubated at 25 °C with ammonium sulfate and sodium chlorate. After 5 h, potassium chloride solution was added, the suspension was filtered, then ammonium buffer and color reagent were added into the suspension, and nitrite released was determined colorimetrically. For each soil sample, one replication was applied. The control samples were performed by the same procedures, but the soil was incubated at -20°C for 5 h. Standard potassium nitrite solution was used to determine the calibration curve. Values of AO enzyme activity was expressed as µg NO_2-N $g^{-1}5$ h^{-1}.

Both, RA and AO were also calculated on a per unit clay basis. Weighted enzyme activity = (1/3) × enzyme activity in 0-5 cm depth + (2/3) × enzyme activity in 5-15 cm, which was used to represent enzyme activity in NT at a depth of 0-15 cm.

Soil Chemical Characteristics and Soil Texture

All soil samples were sent to Midwest Laboratories (13611 B Street, Omaha, Nebraska) for determination of chemical properties. Organic matter was determined by loss of weight on ignition (NCR, 2011); phosphorus was determined by extraction with dilute acid and ammonium fluoride (weak bray)/colorimetric (NCR, 2011); pH was measured with electrode in a 1:1 soil: water solution (NCR, 2011); soil texture was determined by hydrometer method (ASA, 1982); potassium, magnesium, and calcium were extracted with neutral ammonium acetate and measured by inductively coupled argon plasma detection (RMST, 1974; NCR, 2011). The results are presented in Table 2.1.

Statistical Analyses

All statistical tests were performed using SAS software, Version 9.1 for Windows (SAS Institute Inc., 2004). Two-way analysis of variance models was used to assess the main effects and interaction of spatiality and drainage. The Shapiro-Wilk test was used to determine normality ($p < 0.05$). Pair-wise comparisons were calculated using Tukey's Honestly Significant Difference comparisons based on a 95% or 90% confidence interval. However, nonparametric Kruskal-Wallis rank order analysis was done to determine the influence of different treatments if statistics did not meet the normal distribution criteria of general linear models. Linear regression was used to examine relations between enzyme activities and crop yield. Spearman's rank correlation was used as a

nonparametric alternative to linear regression, when normality could not be achieved through data transformations.

RESULTS

Enzyme Activity

Effects of drainage class on enzyme activities were analyzed separately under NT and CT in each state, as presented in Tables 2.2 to 2.7. For AO activity, no significant difference was found except for soils under CT (0-15 cm depth) in Ohio. AO activity of somewhat poorly drained soils had two times the activity of well-drained soils, with an even more significant difference between poor drainage and good drainage (Tables 2.2, 2.3, 2.4). RA responded similarly as AO to drainage class with no significant difference due to drainage (Tables 2.5, 2.6, 2.7).

Table 2.8 shows that AO activities of NT (0-5 cm depth depth) compared to CT (0-15 cm depth) was significantly higher, and AO activity/unit clay did also indicate this significant difference. There was no significant effect of tillage on AO activity and AO/unit clay, in the NT (5-15 cm depth) compared to the CT (0-15 cm depth; Table 2.8). However, calibrating AO activity as enzyme AO activity/unit clay did not change the fact that AO assay had a higher value in CT (0-15 cm depth) compared to NT (5-15 cm depth). Table 2.9 shows the effects of soil management on RA activity and RA/unit clay. Although no significant difference was observed, means of RA activity in NT at 0-5 cm depth was higher compared to CT (0-15 cm depth). Compared to NT (5-15 cm depth), RA activity was higher in CT (0-15 cm depth). RA activity/unit clay did not detect a tillage difference, but it remained the same ranking as enzyme activity within NT and CT.

Table 2.15 presents correlations of enzyme activities and crop yields (2017) with nutrients and organic matter. AO in NT (0-5 cm depth depth) is significantly correlated with organic matter, P, Mg, and Ca. In CT (0-15 cm depth), AO was significantly correlated with organic matter and Mg. Although RA in NT (0-5 cm depth) and CT (0-15 cm depth) was not significantly correlated with nutrients and organic matter. The correlation coefficient of RA at NT (0-5 cm depth depth) with Mg was -0.34 and for RA at CT (0-15 cm) with K, r = 0.33. Mg and K had important effects on RA.

Crop Yield

Average soybean yield on sites in Ohio was much higher than state average soybean yield (Table 2.10). Average soybean yield on sites of Illinois was much lower than state average yield (Table 2.10). The differences may exist between reported yield and measured yields, and this difference was higher in soybean yields compared to the difference in corn yields (Table 2.10).

Figure 2.1 shows organic matter under CT and NT. Organic matter in surface soil (0-5 cm depth) under NT is higher than organic matter in subsurface soil (5-15 cm depth) and organic matter under CT (0-15 cm depth). Figures 2.2, 2.3, 2.4 present organic matter in soils from different drainage classes. Organic matter in poorly drained soils is higher than organic matter in somewhat poorly and well-drained soils.

However, organic matter differences among drainage classes does not affect crop yields. Effects of drainage class on crop yields are shown in Table 2.11. There was no significant effect of drainage on crop yields in Iowa and Illinois. However, in Ohio, corn yield on well-drained soils was significantly higher than on somewhat-poorly drained soils, but no significant difference was found between corn yield on well-drained soils

and poorly drained soils. Results of effects of drainage on relative crop yields are shown in Table 2.12. No significant difference of relative yields due to drainage class was found, which is consistent with the result of Figure 2.5. Figure 2.5 presents drainage class did not significantly affect crop yield from 2015 to 2017.

Table 2.13 shows effects of soil management on measured crop yields in 2017 in Ohio, Illinois, and Iowa. Corn or soybean yields were not significantly affected by tillage. Table 13 shows the effects of soil management on relative crop yields from fields of three states, and relative crop yields did not show significant differences under different soil management, which is consistent with the result of Figure 2.6. Figure 2.6 presents tillage did not significantly affect crop yield from 2015 to 2017.

Crop Yield and Correlation

Table 2.19 shows correlation coefficients between crop yields and weighted means of enzyme activities or weighted means of enzyme activities/unit clay. Although both correlations were not significant at $\alpha = 0.05$, AO activities were slightly positively correlated with corn yields ($r = 0.02$, $p = 0.91$). However, AO activities were greater correlated with soybean yields ($r = 0.27$, $p = 0.27$). RA activities were negatively correlated with soybean yield ($r = -0.39$, $p = 0.1$) and slightly positively correlated with corn yields ($r = 0.057$, $p = 0.71$). Enzyme activity as ratios of activities to clay percentage had greater correlation coefficients than actual activities for the relationship with corn yields. Correlation coefficients between two enzymes and corn yields increased to 0.08 (AO) and 0.16 (RA), although these r values were not significant at $\alpha = 0.05$. For correlations with soybean yields, r values went from negative to positive with actual

activity and RA activity/unit clay ($r = 0.61$, $p = 0.0055$), respectively. Correlation between AO activity and soybean yields became significant when calibrated as AO/unit clay. Table 2.17 shows that enzyme activity/unit clay had a different ranking within NT (0-5 cm depth) or CT (0-15 cm) compared to enzyme activity. Table 16 shows that statistical significance was observed between AO (0-5 cm) and relative yield, between RA (0-5 cm) and relative yield.

Tables 2.19 and 2.20 present correlations between enzyme activities and actual crop yields in 2017 under different management. Under NT management, no significant correlation was found. However, significant correlation between RA activity and soybean yields was observed. A significant correlation was also observed between AO activity/unit clay and soybean yields. Tables 2.21 and 2.22 present correlation coefficients between crop yields and enzyme activities from soil samples at NT in 0-5 cm depth or 5-15 cm depth. There was no significant correlation at these depths of enzyme activities or activity/unit clay. However, RA activity is more significantly related with crop yields in 5-15 cm depth and AO activity is more significantly correlated with crop yields in 0-5 cm depth.

Table 2.15 shows crop yield was influenced by nutrients and organic matter across sites. Soybean yield was significantly correlated with Ca. Although no significantly correlation was observed in corn yield, r-value of corn with K was 0.25.

DISCUSSION

Effects of Drainage and Tillage on Enzymes

Both AO and RA were chosen because they are simple assays and perform oxidation reactions. Thus, it would be expected the microbial community would be stimulated to have higher levels and activity of oxidation enzymes under aerobic conditions in well-drained soils that would decrease down a drainage gradient of moderate to poorly drained soils.

The three drainage classes in this study were well drained (ready loss of water but not rapidly), somewhat poorly drained (water is removed slowly when soil is wet at a shallow depth for significant periods during the growing season), and poorly drained (water is removed so slowly that the soil is wet at shallow depths periodically during the growing season or remains wet for long periods) (Soil Survey Manual, USDA).

Generally, no significant difference was found in RA activities due to drainage class. The enzyme is distributed widely and has been detected in plants (Chew, 1973), and several bacteria (Brown et al., 1965; Smith and Lascelles, 1966; Stearns, 1953) and in flooded or non-flooded soil (Tabatabai and Singh, 1976). Little is known about effects of drainage on RA activity. Although flooded and non-flooded soils are markedly different in their physical, chemical, and biological properties (Ponnamperuma, 1972), Ramesh et al. (1984) found that flooding both increased or decreased RA activity depending on the soil type. Although this study applied to more extreme drainage conditions (non-flooded vs. flooded), it is consistent with our results that drainage condition did not significantly affect RA activity across soil types.

AO activity generally was not significantly affected by drainage except for one field in Ohio. On this field, AO activity in poorly and somewhat poorly drained soils was

higher than in well-drained soils. In this study, the first step in the AO process (formation of hydroxyl amine) was measured to estimate potential oxidation rates of ammonium oxidizers. The transformation of ammonia to nitrite is regarded as a limiting factor of nitrification, which is an essential step in the global nitrogen cycle. The AO reaction should be optimal under aerobic conditions and thus be elevated in well-drained soils over poorly-drained soils. Lutz Breuer et al. (2002) studied gross nitrification in tropical rain-forest soil and found it was influenced by temperature and moisture. They found gross nitrification was positively correlated with increasing soil temperature and negatively correlated with soil moisture. Sami and Tim (2008) also showed nitrification and N mineralization rates being higher in well-drained soils.

One factor for these effects of drainage on RA and AO activities is that the soil samples taken at sites in Ohio, Illinois, and Iowa were fairly flat and did not have major differences in elevation between well- and poorly-drained soils. Thus, differences between landscape position or drainage may not have been great enough to cause significant shifts in AO and RA activities. As soil samples were collected across sites, intrinsic properties of soil could confound the results. Further, the effect of drainage on AO was possibility confounded by influences of inorganic and organic inputs, such as organic matter, P, Mg, and Ca.

Considering accessibility and convenience, the drainage class map form the Web Soil Survey was applied. However, the drainage class map may not accurately represent the redox potential. Soil redox potential can be more accurately determined by measuring reduction potential (E_h) on site or in a laboratory. Although accurate assessment of redox status by electrodes is still a matter of controversy, redox equilibrium at the electrodes

may have not been reactive (Fiedler et al., 2007), it is able to be a reference for comparison of redox potential, and this method could provide more accurate assessment of soil redox condition than the indirect method of drainage class from the Soil Survey.

AO activity for NT in the 0-5 cm depth was higher than CT (0-15 cm depth). However, soil management did not have a significant effect on AO activities at the 5-15 cm depth of NT compared to CT (0-15 cm depth). A statistically significant difference was not observed, but RA activity was higher in NT at the 0-5 cm depth compared to CT (0-15 cm depth), and RA and AO activities were both higher in CT at the 5-15 cm depth. AO activity/unit clay indicated a management difference, while RA activity/unit clay did not. Enzyme activity/unit clay was possibly adopted in AO assay to indicate soil quality across sites.

Soil is an inhospitable environment for free extracellular enzymes, as they are denatured, degraded, and inactivated upon addition to soil (Burns, 1978). However, soil provides conditions where enzymes can become stabilized by clay and organic matter but remain catalytic (McLaren, 1975). This is because enzymes immobilized on humus or clay colloids are resistant to denaturing (Sarkar et al., 1980; Nannipieri et al., 1982). For some enzymes, the immobilized fraction in soil can be found in greater concentrations than those directly associated with viable microbial cells. Forty to sixty percent of the activities are associated with abiotic (Skujins, 1976) forms of enzymes (Nannipieri et al., 1996; Knight and Dick, 2004). As both RA and AO retained high levels of activity after air drying the soil (see Chapter 3), this would indicate that these enzymes are stabilized in the soil matrix and remain catalytic. Thus, enzyme activity/unit clay has potential to distinguish treatment effects independent of soil type for these assays.

Enzyme activity/unit clay detected soil tillage effects in some cases that did not occur on direct enzyme activity. This was the case for AO activity/unit clay. RA activity/unit clay was not significantly affected by tillage difference. However, some enzymes are completely inactivated when sorbed to clay surfaces (Jawed et al., 1989). Therefore, depending on the biochemical properties, these results show that there can be differential effects among enzymes for detecting soil management that may related to how enzymes are stabilized in the soil matrix.

Effects of Drainage and Tillage on Crop Yield

Tillage and drainage did not have a significant influence on crop yields in our research. Many soil properties, such as organic matter (Ciha, 1984; Stone et al., 1985; Wright et al., 1990), pH (Kreznor et al., 1989; Moore et al., 1993), available water, soil texture, and fertility have been found to affect crop yield. In our case, soybean and corn yields across sites were correlated with nutrients and organic matter. Soybean yield in our study was significantly correlated with Ca in soil, which could suggest this nutrient was limited in some cases. Thus, nutrient levels across the farm sites could have confounded the results for detecting yield effects due to tillage and drainage on crop yield.

Another factor that controls yield is topographic or landscape position. In certain years, 60% or more of the yield variability can be explained by a combination of soil properties and topographic features (Kravchenko and Bullock, 2000; Yang et al., 1998). Topographic features can have a direct effect on crop yield by influencing drainage and an indirect effect through its influence on distribution of physical and chemical soil properties (Franzmeier et al., 1969; Bennett et al., 1972). Therefore, other topographic features may confound the effects from drainage.

In general, tillage of poorly drained soils can increase crop yields (Nakajima et al., 2013), as drainage improves aeration and availability of nutrients (Lal and Taylor, 1970; Cannell et al., 1979). Thirty three percent of the cultivated area in the Midwestern region of the United States is tile-drained (Power et al., 2000) and has become a routine practice (Nangia et al., 2010). Tilling increases yields because poorly drained soils may have high organic matter content and may be of higher quality overall. However, crop yields are not always as consistent as expected along changes in landscape or drainage gradients. For example, Wright et al. (1990) found higher crop yields on the summit, whereas Afyuni et al. (1993) found higher crop yield on foot slopes.

NT retains residue on the soil surface and increases soil organic matter compared to intensive tillage systems (Martino and Shaykewich, 1994; Six et al., 2002; Kumar et al., 2012), which was the case for our study (Figure 1). Soil health and continuous no-till system are closely connected (Duiker, et al., 2017). But in our case, NT crop yields were found to be equal to CT crop yields from 2015-2017. The benefits from NT system on crop yields due to improved soil quality may take more time. Studies have shown that NT systems can increase crop yield over CT on well-drained soils (Griffith et al., 1973). Many investigations in the Corn Belt, as reported by Griffith et al. (1988), have found that NT has more efficient moisture use by crops and improved soil physical properties on well-drained soils.

However, lower NT yields on poorly-drained soil have been observed (Griffith et al., 1973), for which the mechanism is not well understood, although, in our study, poorly-drained soils (as classified by the Web Soil Survey) did not have NT crop yields significantly lower than CT. This could be because of inaccuracies of the soil survey,

meaning it may have misidentified some sites in terms of drainage class. This is supported by the fact that these same poorly-drained sites had a similar texture to well-drained soils, whereas poorly drained soils normally have greater clay content. Also, visual observations at the study sites concluded that the landscapes were relatively flat, with very small elevation difference between well-drained and poorly drained sites.

Correlation Between Enzyme Activities and Crop Productivity

RA as an oxidation enzyme was expected to decrease along a gradient from well- to poorly drained soils. Therefore, a positive correlation between RA and crop yields was expected. However, a slightly negative relation was found between soybean yields and RA. This negative correlation was more evident at the 5-15 cm depth. No significant negative correlation was observed between RA and crop yields. AO was expected to be positively correlated with crop yields, and, although a slightly positive correlation was observed between AO activity and crop yields, no significant correlation existed between AO activity and crop yields.

Enzyme activity, as a soil quality indicator, has potential to be an integrative indicator of soil microbial and physical properties and to be sensitive to disturbance applied on soils (Bandick and Dick, 1999; Ndiaye at el., 2000; Vallejo et al., 2012). To be effective and practical, soil quality indicators must be able to reflect crop yield. However, little information about relationship between soil quality and crop yields has been ocumented.

Good drainage does have a positive influence on crop yields; however, this is based on comparisons of extremely poorly drained soils with well-drained soils (Nakajima et al., 2013; Lal and Taylor, 1970; Cannell et al., 1979; Power et al., 2000). As

sites in the present study were relatively flat with limited elevation differences, drainage difference may not be important enough to cause significant effects on crop yields or oxidation enzymes. Under this condition, correlation between crop yields and enzyme activities cannot be dependent on landscape position or drainage.

Also, RA and AO can be controlled by a host of factors. For example, RA has been shown to influence forest type (Lettl, 1986), chemicals (Deng, 1990; Singh and Tabatabai, 1977), and trace elements (Singh and Tabatabai, 1977). Thus, other factors could be confounding the effects of landscape position on enzyme activities. In our case, organic matter, K, P, Mg, and Ca were correlated with RA, AO, and crop yields. AO activity was significantly correlated with organic matter, P, Mg, and Ca. Soybean yield was significantly correlated with Ca. Therefore, inputs on soils like fertilizer would also complicate the results.

The main conclusion is that AO and RA activities were not correlated with crop yields, and this was likely due to the similarity of the topography and confounding effects on yields due to variations of crop management (e.g. fertilizer management) across farm sites and tillage treatments. Enzyme activity/unit clay increased the correlation of RA or AO with crop yields, which supported the hypothesis that calibration of enzyme assay as enzyme activity/unit clay could reduce effects from soil types. This increase may be due to immobilized enzymes in clay, which are more stable and less sensitive to temporal disturbances, such as precipitation and temperature, and reduce the influence of soil type.

CONCLUSIONS

Correlation of the enzyme activities to crop yield was not significant. RA and AO activities were not significantly affected by drainage across sites either, but RA activity/unit clay and AO activity/unit clay did increase r values with crop yields. This study may be limited by number of soil samples that did not represent strong elevation and drainage gradients and because some poorly-drained sites may have been tilled, which would produce high yielding sites. Another issue is that the Web Soil Survey may not have accurately classified drainage for every site because its scaling resolution was too low. In this study, inputs on soil were not taken into consideration, but this may complicate the effects of drainage on crop yield. Intrinsic properties of soil may also complicate the results, as soil samples were collected across sites. RA and AO activity of soils on more distinct and steeper landscape gradients need to be investigated, and redox potential of soil can be identified by other more practical methods, such as measurement of Eh.

Based on this study, it can be concluded that RA and AO on relatively flat landscapes were not affected by natural drainage. This was reflected in crop yield, which also was not affected by drainage. This stands in contrast to the majority of the literature that has shown crop yields to be lower in poorly-drained soils than well-drained soils.

AO activity and AO activity/unit clay in NT (0-5 cm depth) did detect differences due to tillage; however, RA activity and RA activity/unit clay were not sensitive to tillage.

TABLES

Table 2.1 Soil properties of the soil sampling sites in Ohio, Illinois, and Iowa.

Site name	Coordinates	Series Name	Taxonomic name	Soil type	Drainage class‡	pH	Organic matter	Sand	Clay	P	K	Mg	Ca
							%			μg g^{-1}			
IA-MET	N41°50'18.14" W90°20'50.16"	Dinsdale	Fine-silty, mixed, superactive, mesic Typic Argiudolls	silt, loam	1,2	5.7	3.2	9	23	20	218	305	1699
IA-DIE	N41°50'45.12" W90°38'33.82"	Klinger	Fine-silty, mixed, superactive, mesic Aquic Hapludolls	silt, loam	1,2,3	6.5	4.7	26	22	30	195	533	2422
IA-VIC	N41°50'0.09" W90°34'37.59"	Tama	Fine-silty, mixed, superactive, mesic Typic Argiudolls	silt, loam	1	5.9	2.8	11	22	7	104	287	1733
IL-MCK	N40°37'25.04" W90°29'25.56"	Sable	Fine-silty, mixed, superactive, mesic Typic Endoaquolls	silt, clay, loam	1,2,3	6.1	4.3	10	28	20	168	369	2794
IL-EMO	N40°37'28.41" W90°30'43.21"	Ipava	Fine, smectitic, mesic Aquic Argiudolls	silt, clay, loam	2,3	6	3.8	7	29	23	168	291	2516

Table 2.2 (continued) Soil properties of the soil sampling sites in Ohio, Illinois, and Iowa.

Site name	Coordinates	Series Name	Taxonomic name	Soil type	Drainage class‡	pH	Organic matter	Sand	Clay	P	K	Mg	Ca
							%			μg g^{-1}			
IL-WEA	N40°37'19.67" W90°29'46.13"	Osco	Fine-silty, mixed, superactive, mesic Typic Argiudolls	silt, clay, loam	1,2	6.3	4.6	11	28	19	153	406	3018
OH-CIR1	N39°40'13.24" W82°55'44.48"	Crosby	Fine, mixed, active, mesic Aeric Epiaqualfs	loam	1,2	5.8	2.4	33	22	16	93	312	1495
OH-CIR2	N39°39'16.98" W82°56'2.04"	Miamian	Fine, mixed, mesic Typic Hapludalfs	loam	1,2	5.5	1.9	42	18	38	135	148	954
OH-CIR3	N39°39'12.58" W82°57'9.25"	Miamian	Fine, mixed, mesic Typic Hapludalfs	loam	1,2	5.9	2.8	38	16	16	90	270	1344

† pH, organic matter percent, P, K, Mg, Ca, sand, and clay percentages are averaged across drainage class with a regional location.

‡ 1 = well-drained soils; 2 = somewhat poorly drained soils; 3 = poorly drained soils

Table 2.3 Effect of drainage class on soil ammonium oxidation enzyme activity in Illinois.

		Poorly drained		Somewhat poorly drained		Well-drained	
Management	Depth (cm)	Mean	Standard deviation	Mean	Standard deviation	Mean	Standard deviation
		NO_2^- µmol g^{-1} 5 h^{-1}					
No tillage	0-5	3.47 a†	3.00	1.42 a	0.94	1.68 a	1.15
	5-15	1.23 a	0.78	0.43 a	0.30	0.43 a	0.21
Conventional tillage	0-15	0.88 a	0.69	0.51 a	0.26	1.05 a	1.14

† Values within a row followed by same letters are not significantly different ($p < 0.05$).

Table 2.4 Effect of drainage class on soil ammonium oxidation enzyme activity in Iowa.

		Poorly drained		Somewhat poorly drained		Well-drained	
Management	Depth (cm)	Mean	Standard deviation	Mean	Standard deviation	Mean	Standard deviation
		SCN^- nmol g^{-1} h^{-1}					
No tillage	0-5	3.29 a†	0.26	2.09 a	1.57	0.79 a	0.97
	5-15	0.82 a	0	0.61 a	0.40	0.44 a	0.39
Conventional tillage	0-15	5.62 a	0	1.07 a	0.27	0.96 a	0.36

† Values within a row followed by same letters are not significantly different ($p < 0.05$).

Table 2.5 Effect of drainage class on soil ammonium oxidation enzyme activity in Ohio.

		Poorly drained		Somewhat poorly drained		Well-drained	
Management	Depth (cm)	Mean	Standard deviation	Mean	Standard deviation	Mean	Standard deviation
		NO_2^- µmol g^{-1} 5 h^{-1}					
No tillage	0-5	0.67 a†	0.78	3.16 a	4.29	1.93 a	1.53
	5-15	0.73 a	0.77	1.70 a	2.25	0.83 a	0.50
Conventional tillage	0-15	2.46 a	0.23	1.82 a	0.14	0.88 b	0.50

† Values within a row followed by same letters are not significantly different ($p < 0.05$).

Table 2.6 Effect of drainage class on soil rhodanese enzyme activity in Illinois.

Management	Depth (cm)	Poorly drained		Somewhat poorly drained		Well-drained	
		Mean	Standard deviation	Mean	Standard deviation	Mean	Standard deviation
		SCN^- nmol g^{-1} h^{-1}					
No tillage	0-5	153.44 a[†]	60.99	165.64 a	17.03	223.96 a	64.36
	5-15	125.08 a	0.21	133.15 a	63.30	163.74 a	106.12
Conventional tillage	0-15	242.25 a	87.53	179.89 a	37.34	219.47 a	75.23

[†]Values within a row followed by same letters are not significantly different ($p < 0.05$).

Table 2.7 Effect of drainage class on soil rhodanese enzyme activity in Iowa.

Management	Depth (cm)	Poorly drained		Somewhat poorly drained		Well-drained	
		Mean	Standard deviation	Mean	Standard deviation	Mean	Standard deviation
		SCN^- nmol g^{-1} h^{-1}					
No tillage	0-5	164.37 a[†]	43.19	214.69 a	42.69	209.76 a	26.22
	5-15	119.45 a	29.13	143.10 a	92.32	230.32 a	172.72
Conventional tillage	0-15	54.75 a	N/A	209.60 a	136.19	181.67 a	77.87

[†] Values within a row followed by same letters are not significantly different ($p < 0.05$).

Table 2.8 Effect of drainage class on soil rhodanese enzyme activity in Ohio.

		Poorly drained		Somewhat poorly drained		Well-drained	
Management	Depth (cm)	Mean	Standard deviation	Mean	Standard deviation	Mean	Standard deviation
		SCN^- nmol g^{-1} h^{-1}					
No tillage	0-5	207.51 a[†]	108.46	209.82 a	122.83	181.80 a	146.86
	5-15	134.24 a	23.09	111.37 a	14.96	159.42 a	122.64
Conventional tillage	0-15	163.74 a	57.35	167.57 a	22.00	126.71 a	58.76

[†] Values within a row followed by same letters are not significantly different ($p < 0.05$).

Table 2.9 Effect of soil management on ammonium oxidation enzyme activities and ammonium oxidation enzyme activities/unit clay.

Enzyme	Depth in no tillage	No tillage		Conventional tillage (0-15 cm)	
		Mean	Standard deviation	Mean	Standard deviation
AO activity[§]	0-5 cm	1.85 a[†]	1.63	1.18 b	1.04
AO activity	5-15 cm	0.71 a	0.65	1.18 a	1.04
AO enzyme activity/unit clay[¶]	0-5 cm	8.21 a[‡]	7.35	5.56 b	5.10
AO enzyme activity/unit clay	5-15 cm	3.20 a	2.68	5.56 a	5.10

[†] Values within a row followed by the same lowercase letters are not significantly different ($p < 0.1$).
[§] Unit of AO activity is NO_2^- µmol g^{-1} 5 h^{-1}.
[¶] Unit of AO activity/unit clay is NO_2^- µmol g^{-1} 5 h^{-1} clay $percent^{-1} \times 10^4$.

Table 2.10 Effect of soil management on rhodanese activities and rhodanese enzyme activities/unit clay.

Enzyme	Depth in no tillage	No tillage		Conventional tillage (0-15 cm)	
		Mean	Standard deviation	Mean	Standard deviation
RA activity[‡]	0-5 cm	192 a[†]	79	174 a	74
RA activity	5-15 cm	156 a	101	174 a	74
RA activity/unit clay[§]	0-5 cm	914 a	495	787 a	359
RA activity/unit clay	5-15 cm	725 a	506	787 a	359

[†] Values within a column followed by same lowercase letters are not significantly different ($p < 0.1$).
[‡] Unit of RA activity is SCN^- nmol g^{-1} 1 h^{-1}.
[§] Unit of RA activity/unit clay is SCN^- nmol g^{-1} 1 h^{-1} clay $percent^{-1} \times 10^4$.

Table 2.11 2017 crop yield in Ohio, Iowa, and Illinois.

State	2017 Reported Statewide Crop Yield†		2017 Crop Yield at Sites‡	
	Corn	Soybean	Corn	Soybean
	Mg ha^{-1}			
Ohio	11.90	3.33	10.70	4.69
Illinois	13.52	3.90	11.85	2.27
Iowa	13.58	3.80	11.23	3.56

† 2017 state average crop yields (USDA, 2018).
‡ 2017 average crop yield for sites where crop samples were collected.

Table 2.12 Effects of drainage class on corn and soybean yield in Ohio, Illinois, and Iowa in 2017.

			Poorly drained		Somewhat poorly drained		Well-drained	
State	Crop	Management§	Mean	Standard deviation	Mean	Standard deviation	Mean	Standard deviation
			Mg ha^{-1}					
Iowa	Corn	NT	N/A	N/A	11.00 a	1.80	11.20 a	3.20
		CT	12.10 a†	N/A	12.10 a	0.01	11.10 a	1.30
	Soybean	NT	3.800 a	0.20	3.300 a	0.30	N/A	N/A
Illinois	Corn	NT	12.00 a	1.80	12.800 a	2.70	12.40 a	0.07
	Soybean	CT	2.700 a	N/A	2.500 a	0.20	2.300 a	0.40
Ohio	Corn	NT	8.300 a	N/A	7.700 ac	0.70	12.70 ab	1.80
		CT	9.000 a	4.10	13.40 a	N/A	10.90 a	1.60

† Values within a row followed by same letters are not significantly different ($p < 0.05$).
‡ Drainage class 1 in Ohio = very poorly drained.
§ NT is no tillage; CT is conventional tillage.
¶ Different capital letters in a column indicate significant difference ($p < 0.01$).

Table 2.13 Effect of drainage on relative yields ‡ from fields in Ohio, Illinois, and Iowa.

State	Management§	Poorly drained		Somewhat poorly drained		Well-drained	
		Mean	Standard deviation	Mean	Standard deviation	Mean	Standard deviation
Iowa	NT	108.0 a†	2.99	96.47 a	7.90	99.60 a	10.79
	CT	110.8 a	N/A	100.4 a	10.39	98.55 a	5.85
Illinois	NT	98.32 a	1.96	97.22 a	5.46	99.70 a	11.53
	CT	96.82 a	11.72	99.89 a	7.57	109.4 a	1.97
Ohio	NT	98.47 a	N/A	98.93 a	11.33	100.7 a	13.33
	CT	89.04 a	3.45	118.0 a	12.36	97.98 a	16.41

†Values within a row followed by same letters are not significantly different ($p < 0.05$).

‡The relative yield was calculated as a ratio of yield at each site to the site mean.

§NT is no tillage; CT is conventional tillage.

Table 2.14 Effect of soil management on crop yields from fields in Ohio, Illinois, and Iowa in 2017.

Management	Corn yield		Soybean yield	
	Mean	Standard deviation	Mean	Standard deviation
	Mg ha^{-1}			
No tillage	11.4 a[†]	2.50	3.40 a	1.00
Conventional tillage	11.2 a	1.40	3.50 a	1.60

[†] Values within a column followed by same letters are not significantly different ($p < 0.05$).

Table 2.15 Effect of soil management on relative crop yields‡ from fields in Ohio, Illinois, and Iowa.

	Relative crop yield	
Management	Mean	Standard deviation
No tillage	99.4 a†	9.15
Conventional tillage	101 a	11.2

† Values within a column followed by same letters are not significantly different ($p < 0.05$).
‡ The relative yield was calculated as a ratio of yield at each site to the site mean.

Table 2. 16 Correlation of enzyme activities with relative yield‡.

Enzyme activity†	Relative yield (divided by average yield of a site)	Relative yield (divided by average yield of a state)	Relative yield (divided by average yield of all sites)
RA (0-5 cm)	-0.462 *	-0.331	-0.286
RA (5-15 cm)	0.046	-0.043	-0.125
AO (0-5 cm)	0.350	0.600 *	0.472 *
AO (5-15 cm)	0.337	0.255	0.217

*Significant at the 0.05 probability levels.
†The unit of AO activity is NO_2^- µmol g^{-1} 5 h^{-1}, and the unit of RA activity is SCN^- nmol g^{-1} 1 h^{-1}.
‡Relative yield was calculated by dividing yield on a sampled place by average yield of a site or a state or all sites.

Table 2.17 Correlations coefficients (r) of enzyme activities† or crop yields‡ (2017) with organic matter and extractable nutrients.

Tillage / crop yield	Enzyme	Organic matter (%)	P µg g^{-1}	K µg g^{-1}	Mg µg g^{-1}	Ca µg g^{-1}
No tillage (0-5 cm)	RA	-0.21	-0.17	-0.14	-0.34	-0.21
No tillage (0-5 cm)	AO	0.42 *	0.37 *	0.18	0.63 ***	0.38 *
Conventional tillage (0-15 cm)	RA	0.22	-0.04	0.33	-0.04	0.21
Conventional tillage (0-15 cm)	AO	0.46 **	0.23	0.17	0.46 **	0.25
Corn		0.17	0.10	0.25	-0.13	0.08
Soybean		-0.33	0.06	-0.2	-0.25	-0.63 **

* ,*, **, *** Significant at the 0.05, 0.01, and 0.001 probability levels, respectively.

† The unit of AO activity is NO_2^- µmol g^{-1} 5 h^{-1}, and the unit of RA activity is SCN^- nmol g^{-1} 1 h^{-1}.

‡ The unit of crop yield is Mg ha^{-1}.

Table 2.18 Enzyme activities and enzyme activities/unit clay in NT[§] (0-5 cm depth) and CT[§] (0-15 cm depth).

		AO activity				RA activity			
		No tillage (0-5 cm)		Conventional tillage (0-15 cm)		No tillage (0-5 cm)		Conventional tillage (0-15 cm)	
Site name	Clay (%)	Actual[†]	Unit clay[‡]	Actual	Unit clay	Actual	Unit clay	Actual	Unit clay
IA-MET	23	0.757	3.241	0.980	4.376	230.4	985.1	240.7	1100
IA-DIE	22	3.335	15.63	2.270	10.26	173.7	813.0	133.7	574.7
IA-VIC	22	0.824	3.823	0.849	4.011	212.6	1014.6	167.7	773.1
IL-MCK	28	2.380	8.372	0.763	2.363	198.9	816.9	189.6	648.4
IL-EMO	29	1.872	5.917	0.498	1.989	168.9	533.5b	207.2	816.2
IL-WEA	28	1.030	4.240	1.400	4.470	167.8	650.1	258.8	859.2
OH-CIR1	22	2.800	13.56	1.090	4.340	146.1	836.2	129.7	526.2
OH-CIR2	18	0.640	3.280	1.360	7.480	287.0	1655 a	166.4	1071
OH-CIR3	16	2.460	14.10	1.650	11.33	125.0	777.3	121.8	840.0

[†]Actual is actual enzyme activity, unit of AO activity is NO_2^- μmol g^{-1} 5 h^{-1}, unit of RA activity is SCN^- nmol g^{-1} 1 h^{-1}.
[‡]Unit clay is enzyme activity/unit clay, unit is NO_2^- μmol g^{-1} 5 h^{-1} clay percent$^{-1}\times10^4$ and SCN^- nmol g^{-1} 1 h^{-1} clay percent$^{-1}\times10^4$.

Table 2.19 Correlation coefficients (r) of crop yields (Mg ha^{-1}) in terms of weighted enzyme activity or enzyme activity/unit clay across all sites.[†]

	Soybeans		Corn	
Enzyme activity	r	p[‡]	r	p
RA	-0.39	0.10	0.06	0.71
RA/clay	0.22	0.36	0.16	0.31
AO	0.27	0.27	0.012	0.91
AO/clay	0.61	0.006	0.08	0.61

[†] The crop yields are actual measured yields from fields in 2017 using either no tillage or conventional tillage and measured at 0-15 cm depth. The unit of RA activity is SCN^- nmol g^{-1} 1 h^{-1}, and the unit of AO activity is NO_2^- µmol g^{-1} 5 h^{-1}. Enzyme activity/unit clay = (enzyme activity)/(clay percentage) × 100; the units are NO_2^- µmol g^{-1} 5 h^{-1} clay percentage^{-1} × 10^4 and SCN^- nmol g^{-1} 1 h^{-1} clay percentage^{-1} × 10^4.
[‡] The correlations are significant ($p < 0.05$).

Table 2.20 Correlation coefficients (r) of crop yields (Mg ha^{-1}) in terms of enzyme activity or enzyme activity/unit clay under conventional tillage.[†]

	Soybeans		Corn	
Enzyme activity	r	p[‡]	r	p
RA	-0.63	0.08	0.04	0.85
RA/clay	0.15	0.71	0.29	0.18
AO	0.50	0.18	0.007	0.98
AO/clay	-0.72	0.04	0.05	0.83

[†] The crop yields are actual measured yields from fields in 2017 using conventional tillage and measured at 0-15 cm depth. The unit of RA activity is SCN^- nmol g^{-1} 1 h^{-1}, and the unit of AO activity is NO_2^- µmol g^{-1} 5 h^{-1}. Enzyme activity/unit clay = (enzyme activity)/(clay percentage) × 100; the units are NO_2^- µmol g^{-1} 5 h^{-1} clay $percentage^{-1} \times 10^4$ and SCN^- nmol g^{-1} 1 h^{-1} clay $percentage^{-1} \times 10^4$.
[‡] The correlations are significant ($p < 0.05$).

Table 2.21 Correlation coefficients (r) of crop yields (Mg ha^{-1}) in terms of weighted enzyme activity or enzyme activity/unit clay under no tillage. †

	Soybeans		Corn	
Enzyme activity	r	p‡	r	p
RA	-0.006	1.00	0.13	0.56
RA/clay	0.28	0.43	0.03	0.90
AO	0.02	0.96	0.06	0.80
AO/clay	0.09	0.80	0.06	0.81

† The crop yields are actual measured yields from fields in 2017 using no tillage and measured at 0-15 cm depth. The unit of RA activity is SCN^- nmol g^{-1} 1 h^{-1}, and the unit of AO activity is NO_2^- µmol g^{-1} 5 h^{-1}. Enzyme activity/unit clay = (enzyme activity)/(clay percentage) × 100; the units are NO_2^- µmol g^{-1} 5 h^{-1} clay $percentage^{-1} \times 10^4$ and SCN^- nmol g^{-1} 1 h^{-1} clay $percentage^{-1} \times 10^4$.

‡ The correlations are significant ($p < 0.05$).

Table 2.22 Correlation coefficients (r) of crop yields (Mg ha^{-1}) in terms of enzyme activity or enzyme activity/unit clay under no tillage (0-5 cm depth).[†]

	Soybeans		Corn	
Enzyme activity	r	p[‡]	r	p
RA	0.09	0.81	-0.005	0.98
RA/clay	0.47	0.18	0.03	0.89
AO	0.04	0.92	0.12	0.59
AO/clay	0.07	0.85	0.12	0.58

[†] The crop yields are actual measured yields from fields in 2017 using no tillage and measured at 0-5 cm depth. The unit of RA activity is SCN^- nmol g^{-1} 1 h^{-1}, and the unit of AO activity is NO_2^- µmol g^{-1} 5 h^{-1}. Enzyme activity/unit clay = (enzyme activity)/(clay percentage) × 100; the units are NO_2^- µmol g^{-1} 5 h^{-1} clay $percentage^{-1} \times 10^4$ and SCN^- nmol g^{-1} 1 h^{-1} clay $percentage^{-1} \times 10^4$.

[‡] The correlations are significant ($p < 0.05$).

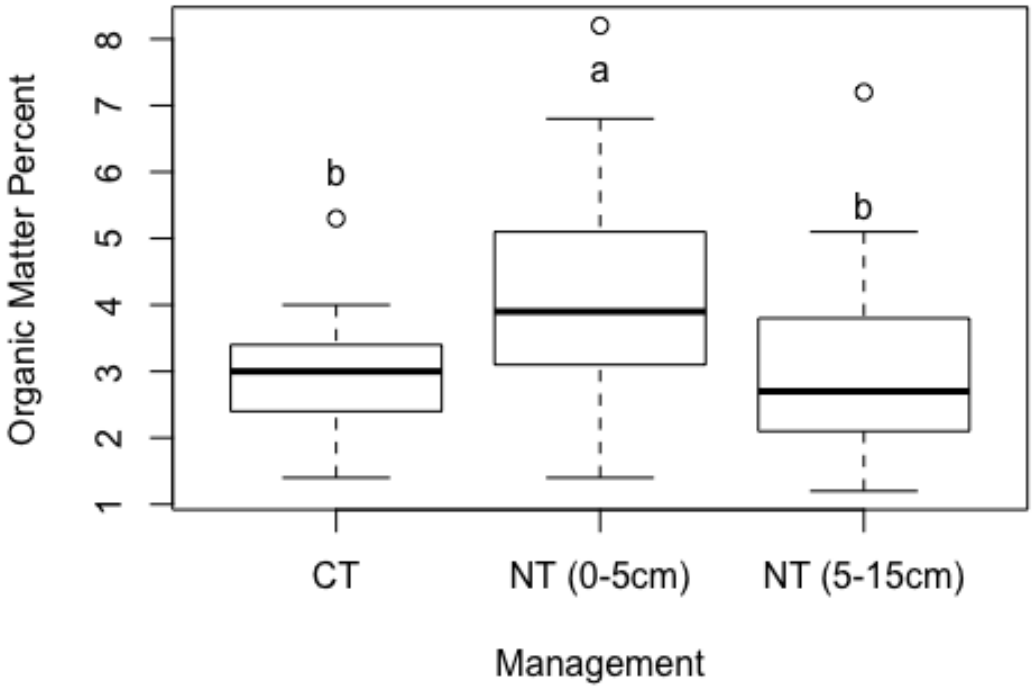
Table 2.23 Correlation coefficients (r) of crop yields (Mg ha^{-1}) in terms of enzyme activity or enzyme activity/unit clay under no tillage (5-15 cm depth).†

	Soybeans		Corn	
Enzyme activity	r	p‡	r	p
RA	-0.21	0.56	0.14	0.52
RA/clay	0.08	0.84	0.02	0.94
AO	-0.02	0.96	-0.04	0.87
AO/clay	0.10	0.78	-0.10	0.67

† The crop yields are actual measured yields from fields in 2017 using no tillage and measured at 5-15 cm depth. The unit of RA activity is SCN^- nmol g^{-1} 1 h^{-1}, and the unit of AO activity is NO_2^- µmol g^{-1} 5 h^{-1}. Enzyme activity/unit clay = (enzyme activity)/(clay percentage) × 100; the units are NO_2^- µmol g^{-1} 5 h^{-1} clay $percentage^{-1} \times 10^4$ and SCN^- nmol g^{-1} 1 h^{-1} clay $percentage^{-1} \times 10^4$.
‡ The correlations are significant ($p < 0.05$).

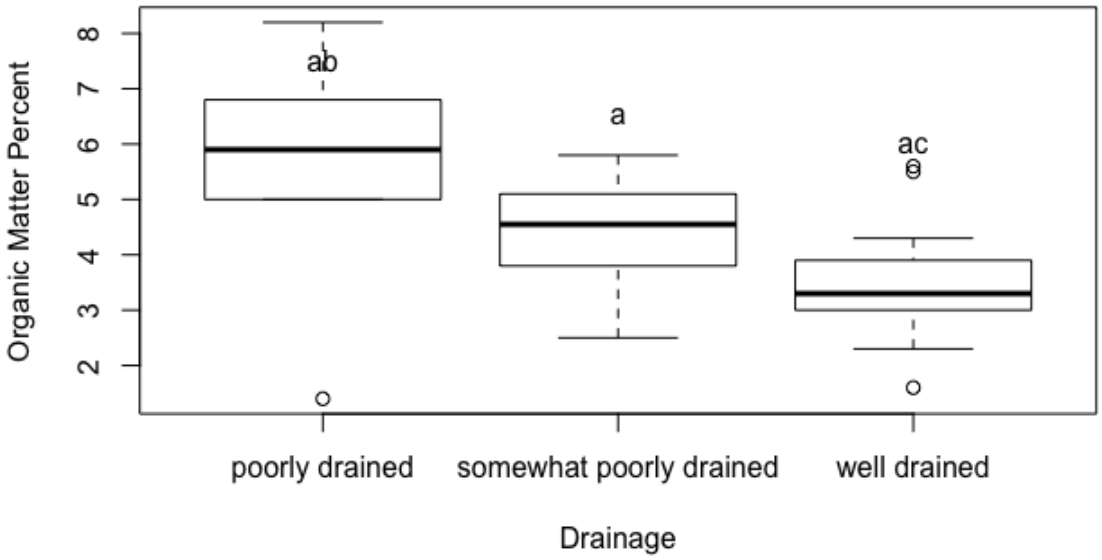
FIGURES

Figure 2. 1 Organic matter percentage under conventional tillage (CT) and no tillage (NT). For NT, the organic matter percentage was measured at 0-5 cm depth and at 5-15 cm depth. For CT, the organic matter percentage was measured at 0-15 cm depth.



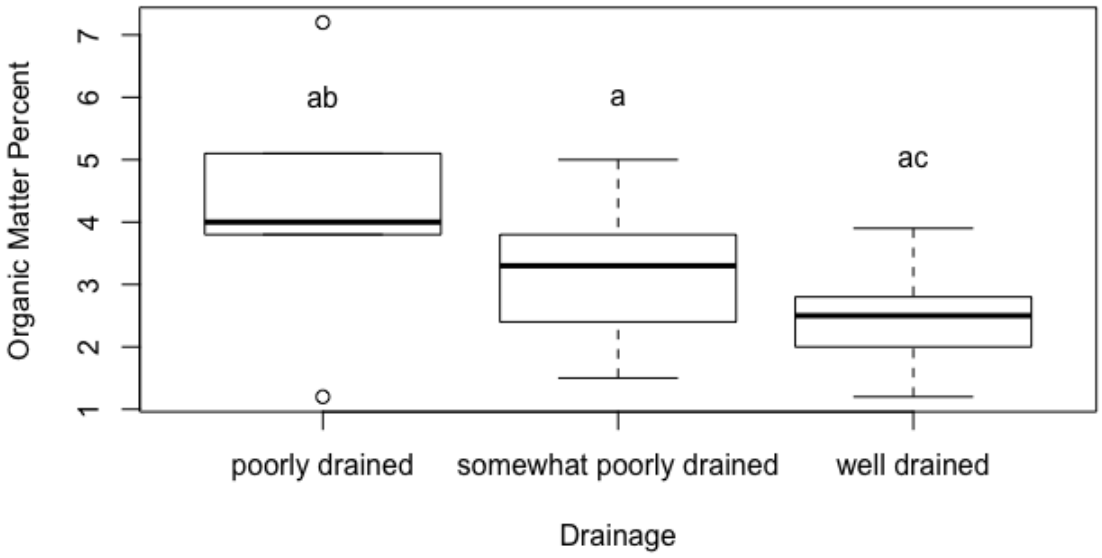
† Boxes with the same letters are not significantly different ($p < 0.05$).

Figure 2.2 Organic matter percentage under no tillage (0-5 cm depth) for drainage from fields in Ohio, Illinois, and Iowa.



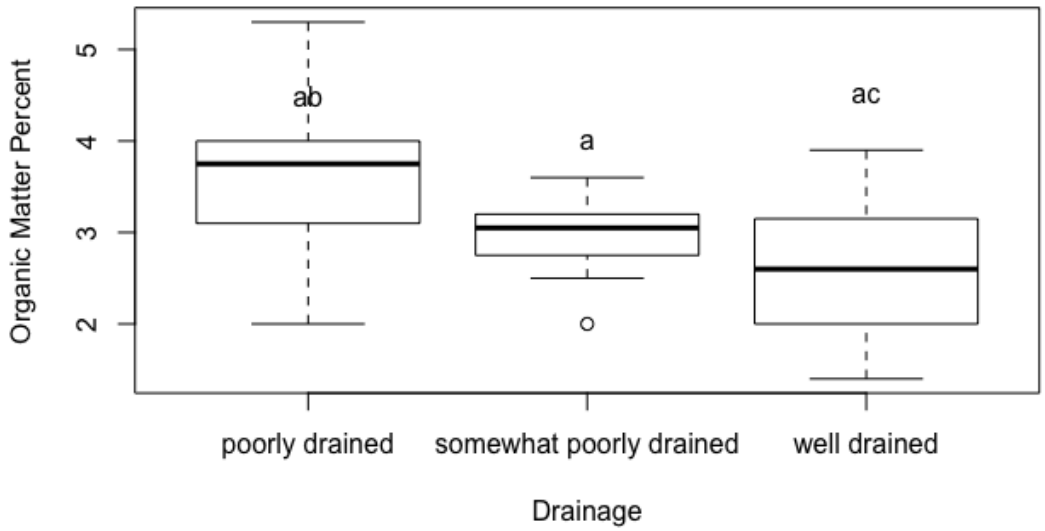
† Boxes with the same letters are not significantly different ($p < 0.05$).

Figure 2.3 Organic matter percentage under no tillage (5-15 cm) for drainage from fields in Ohio, Illinois, and Iowa.



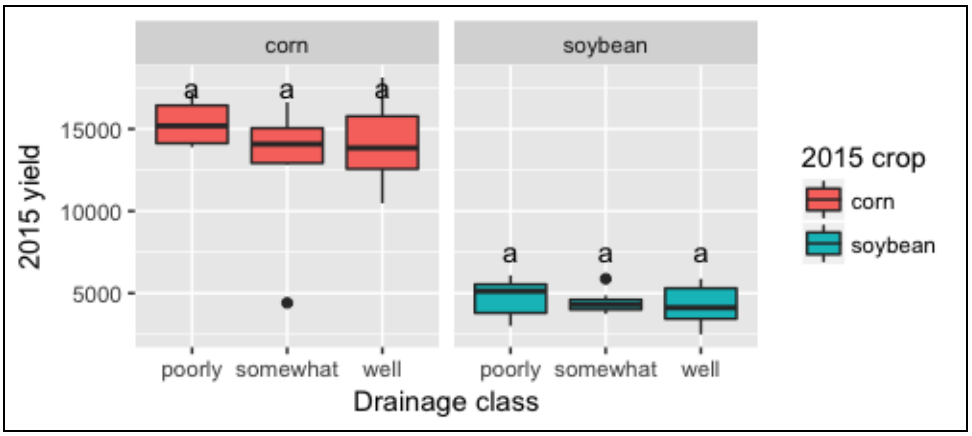
† Boxes with the same letters are not significantly different ($p < 0.05$).

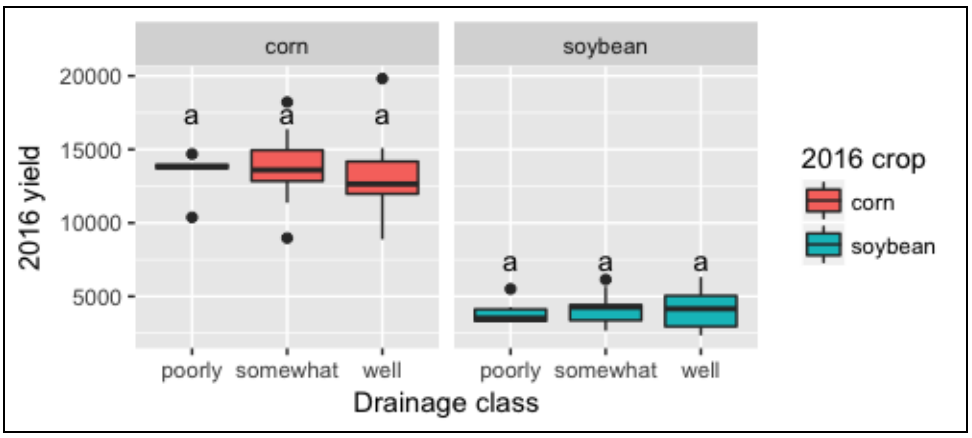
Figure 2.4 Organic matter percentage under conventional tillage (0-15 cm) for drainage from fields in Ohio, Illinois, and Iowa.

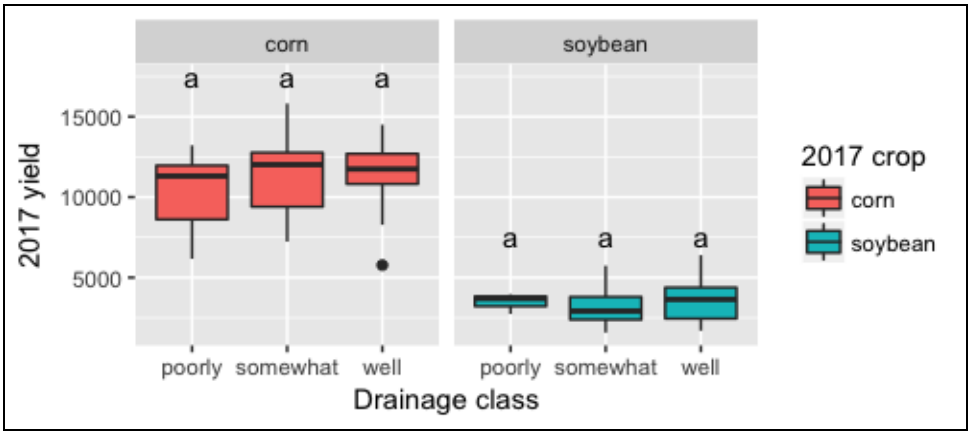


† Boxes with the same letters are not significantly different ($p < 0.05$).

Figure 2.5 Crop yields (kg ha^{-1}) in 2015, 2016, and 2017 for fields with various natural drainage types: poorly, somewhat poorly, and well drained.

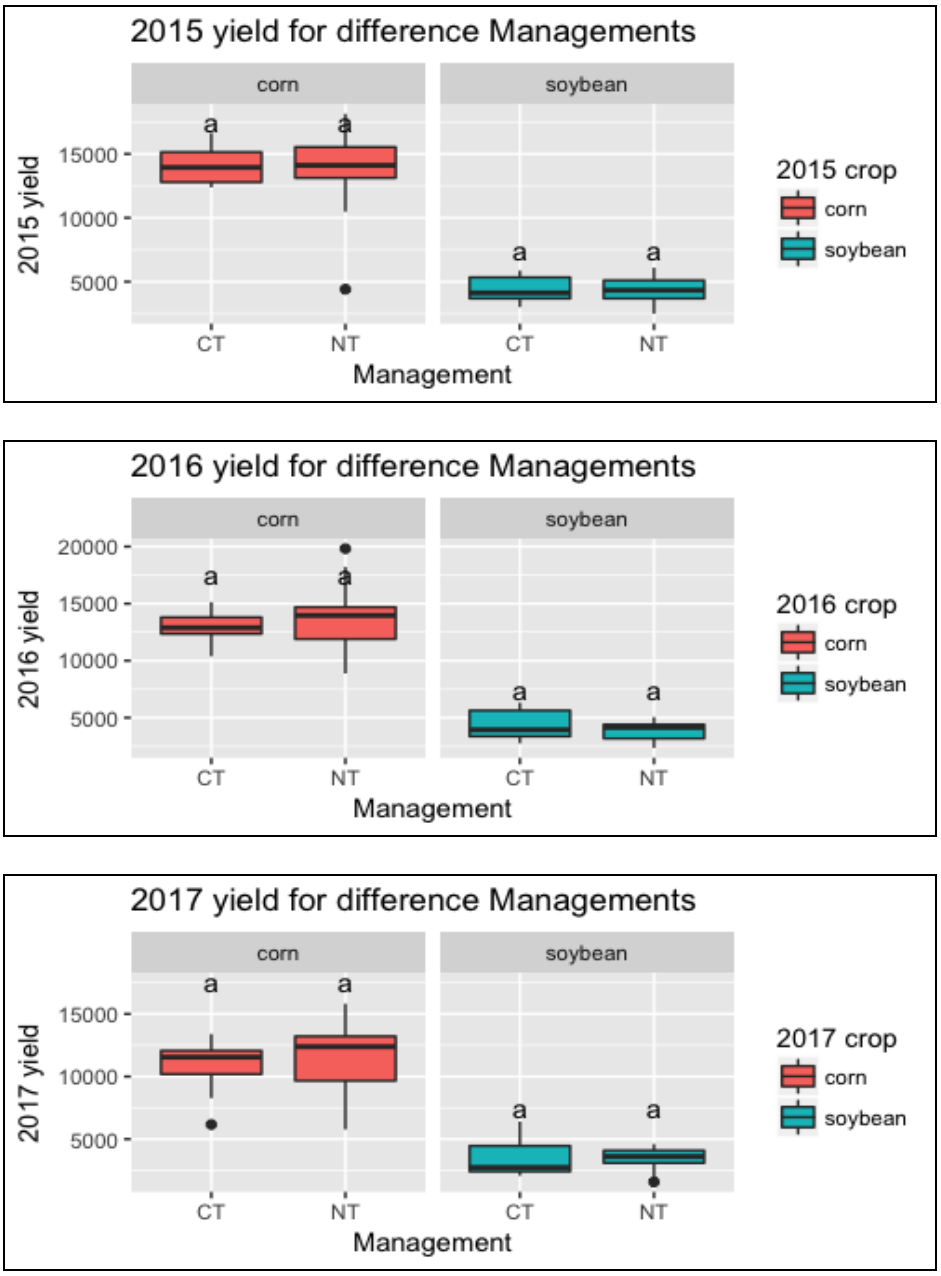






† Boxes with the same letters are not significantly different ($p < 0.05$).

Figure 2.6 Crop yields (kg ha^{-1}) in 2015, 2016, and 2017 under NT and CT.



† Boxes with the same letters are not significantly different ($p < 0.05$).

REFERENCES

Afyuni, M. M., D. K. Cassel and W. P. Robarge. 1993. Effect of landscape position on soil water and corn silage yield. Soil Science Society 57: 1573-1580.

ASA. 1982. Methods of Soil Analysis - Part 2: Chemical and Microbiological Properties, Second Edition. American Society of Agronomy. pp. 549-556.

Bandick, A., and R. P. Dick. 1999. Field management effects on soil enzyme activities. Soil Biology and Biochemistry 31:1471-1479.

Bennett, O.L. 1972. Effects of north and south facing slopes on yield of Kentucky Bluegrass with variable rate and time of nitrogen Application. Agronomy Journal 64: 630– 635.

Borchers, J. G., and D. A. Perry. 1992. The influence of soil texture and aggregation on carbon and nitrogen dynamics in southwest Oregon forests and clear cuts. Canadian Journal of Forest Research 22:298-305.

Brown, T. J., P. J. Butler and F. C. Happold. 1965. Some properties of the rhodanese system of Thiobacillus denitrificans. Biochemical Journal 97: 651-657.

Burns, R. G. 1978. Enzymes in soil: Some theoretical and practical considerations. In Soil Enzymes (R. G. Burns, Ed.), pp. 295-339. Academic Press, London.

Burns, R.G. 1982. Enzyme activity in soil: Location and a possible role in microbial ecology. Soil Biology and Biochemistry 14:423-427.

Cannell, R.Q., K. Gales, R. W. Snaydon, B.A. Suhail. 1979. Effects of short-term waterlogging on the growth and yield of peas (Isum-Sativum). Annals of Applied Biology 93: 327.

Chew, M. Y. 1973. Rhodanese in higher plants. Phytochemistry 12: 2365-2367.

Ciha, A.J. 1984. Slope position and grain yield of soft white winter wheat. Agronomical Journal. Vol.76, pp.193– 196.

Deng, S. 1990. Sulfur oxidation and rhodanese activity in soils. Soil Science, Vol. 150, pp. 552-560.

Dick, R.P., D.D. Myrold, and E.A. Kerle. 1988. Microbial biomass and soil enzyme activities in compacted and rehabilitated skid trail soils. Soil Science Society of America Journal 52:512-516.

Duiker, S., J. C. Myers and L. C. Blazure. 2016. Soil Health in Field and Forage Crop production. USDA Natural Resources Conservation Service, Penn State University Extension, Capital Resource Conservation & Development and Clinton County Conservation District, Harrisburg, PA, p. 27.

Fiedler, S., M. J. Vepraskas and J. L. Richardson. 2007. Soil redox potential: importance, field measurements, and observations. Advances in Agronomy 94: 1-54.

Franzmeier, D.P. 1969. Properties of some soils in the Cumberland plateau as related to slope aspect and position. Soil Science Society of America, Proceedings 33: 755– 761.

Hose, T. D., M. Cougnon, A. De Vliegher, B. Vandecasteele, N. Viaene, W. Cornelis, E. V. Bockstaele, D. Reheul. 2013. The positive relationship between soil quality and crop production: A case study on the effect of farm compost application. Applied Soil Ecology 75: 189-198.

Griffith, D. R., E. J. Kladivko, J. V. Mannering, T.D. West and S. D. Parsons. 1988. Long-term tillage and rotation effects on corn growth and yield on high and low organic matter, poorly drained soils. Agronomy Journal 80: 599-605.

Gupta, V.V.S.R., and J.J. Germida. 1988. Distribution of microbial biomass and its activity in different soil aggregate size classes as affected by cultivation. Soil Biology and Biochemistry 20:777-786.

Jenny, Hans. 1980. The soil resource: origin and behavior. Ecological studies 37. Springer-Verlag, New York.

Knight T, 2002. Soil enzyme stability in disturbed ecosystems. MS thesis. Oregon State University, Corvallis OR.

Knight T, and R. P. Dick. 2004. Differentiating microbial and stabilized β-glucosidase activity relative to soil quality. Soil Biology and Biochemistry 36: 2089-2096.

Kravchenko, A.N. 2000. Correlation of corn and soybean grain yield with topography and soil properties. Agronomical Journal 92: 75– 83.

Kreznor, W.R. 1989. Soil, landscape, and erosion relationships in a northwest Illinois watershed. Soil Science Society of America Journal 53: 1763– 1771.

Kumar, S., A. Kadono, R. Lal and W. Dick. 2012. Long-term no-till impacts on organic carbon and properties of two contrasting soils and corn yields in Ohio. Soil Science Society America Journal 76: 1798–1809.

Lal, R., and G.S. Taylor. 1970. Drainage and nutrient effects in a field lysimeter study. 2. Mineral uptake by corn. Soil Science Society America: Proceedings 34: 245–248.

Lettl, A. 1986. Thiosulfate sulfurtransferase (Rhodanese) in forest soils. Forestry and game management research institute.

Lutz Breuer, Ralf Kiese, and Klaus Butterbach Bahl, 2002. Temperature and moisture effects on nitrification rates in tropical rain-forest. Soil Science Society of America Journal 66:834-844.

Martens, D.A., J.B. Johanson, and W.T. Frankenberger, Jr. 1992. Production and persistence of soil enzymes with repeated additions of organic residues. Soil Science 153:53-61.

Martino, D.L., C.F. Shaykewich. 1994. Root penetration profiles of wheat and barley as affected by soil penetration resistance in-field conditions. Canadian Journal of Soil Science 74: 193–200.

McLaren, A. D. 1975. Soil as a system of humus and clay immobilized enzymes. Chemica Scripta 8: 97-99.

Miller, M. and R.P. Dick. 1995a. Dynamics of soil C and microbial biomass on whole soil and aggregates in two cropping systems differing in C-input. Applied Soil Ecology 2:253-261.

Miller, M., and R. P. Dick. 1995b. Thermal stability and activities of soil enzyme activities as influenced by crop rotation Soil Biology and Biochemistry 27:1161-1166.

Moore, I.D. 1993.Soil attribute prediction using terrain analysis. Soil Science Society of America Journal 57: 443– 452.

Nakajima, T., E. G. Mbonimpa, S. Gautam, U.R. Somireddy, A. Kadono, R. Lal, R. Chintala, R. Rafique and N. Fausey. 2013. Soil science and plant nutrition 60: 108-118.

Nangia, V., P. H. Gowda, D. J. Mulla. 2010. Effects of changes in N-fertilizer management on water quality trends at the watershed scale. Agricultural Water Management 97: 1855–1860.

Nannipieri, P., B. Ceccanti, C. Conti and D. Bianchi. 1982. Hydrolases extracted from soil, their properties and activities. Soil Biology and Biochemistry 14: 257-263.

Nannipieri, P., P. Sequi and P. Fusi. 1996. Humus and enzyme activity. *In* A. Piccolo (ed.) Humic Substances in Terrestrial Ecosystems. Elsevier, New York, pp. 293-328.

NCR. 2011. Recommended Chemical Soil Test Procedures for the North Central Region. No. 499 (revised). North Central Regional Research Publication. North Dakota State University. pp. 5-32.

Ndiaye, E.L., J. M. Sandeno, D. McGrath, and R.P. Dick. 2000. Integrative biological indicators for detecting change in soil quality. American Journal of Alternative Agriculture 15: 26-36.

Nielsen, R. L. 2015. Estimating grain yield prior to harvest. Purdue University. URL: http://www.kingcorn.org/news/timeless/YldEstMethod.html.

Ponnamperuma, F. N. 1972. The chemistry of submerged soils. Advances in Agronomy 24: 29-96.

Power, J.F., R. Wiese, D. Flowerday. 2000. Managing nitrogen for water quality-lessons from Management Systems Evaluation Area. Journal of Environmental Quality 29: 355–366.

Powers, R.F., Tiarks, A.E., Boyle J.R., 1998. Assessing soil quality: practicable standards for sustainable forest productivity in the United States. In: Adams, M.B., Ramakrishna K., Davidson, E.A. (Eds.), The Contribution of Soil Science to the Development and Implementation of Criterial and Indicators of Sustainable Forest Management. Soil Science Society of America, Special Publication No. 53, pp. 53–80.

Ramesh, C. R., N. Behera and N. Sethunathan. 1984. Rhodanese activity of flooded and nonflooded soils. Soil Biology and Biochemistry, Vol. 17, No.2, pp. 159-162.

Reganold, J.P. 1988. Comparison of soil properties as influenced by organic and conventional farming systems. American Journal of Alternative Agriculture 3:144-155.

Roper, W. R., D. L. Osmond, J. L. Heitman, M. G. Wagger and Schris Reberg-Horton. 2017. Soil health indicators do not differentiate among agronomic management systems in north Carolina soils. Soil Science Society, Vol. 81, Issue 4, pp. 828.

RMST. 1974. Handbook on Reference Methods for Soil Testing. Council on Soil Testing and Plant Analysis. pp. 60-65.

Sarkar, J. M., L. Batisticand and J. Mayaudon. 1980. Les hydrolases du sol et leur association avec les hydrates decarbone. Soil Biology and Biochemistry 12: 325-328.

Singh, B.B. and M.A. Tabatabai, 1976. Rhodanese activity of soils. Soil Science Society of America Journal 40: 381-385.

Singh, B. B. and M. A. Tabatabai. 1977. Factors affecting rhodanese activity in soils. Soil Science, Vol. 125, pp. 337-342.

Six, J., C. Feller, K. Denef, S. M. Ogle, J. C. D. Sa and A. Albrecht. 2002. Soil organic matter, biota and aggregation in temperate and tropical soils - Effects of no-tillage. Agronomy 22: 755–775.

Skujins, J. 1976. Extracellular Enzymes in Soil. CRC Critical Reviews in Microbiol. 4:383-421.

Smith, A. J. and J. Lascelles. 1966. Thiosulphate metabolism and rhodanese in Chromatium sp. strains. Journal of General Microbiology 42: 357-370.

Stearns, R. H. 1953. Respiration, rhodanese and growth in Escherichia coli. Journal of Cellular and Comparative Physiology 41: 163-l 70.

Stone, J.R. 1985. Effect of erosion and landscape position on the productivity of piedmont soils. Soil Science Society of America Journal, Vol. 49, pp. 987– 991.

USDA. 1993. Soil Survey Manual. http://websoilsurvey.nrcs.usda.gov/. Soil Survey Division Staff.

USDA.2017. National Agricultural Statistics Service. https://www.nass.usda.gov/Statistics_by_State/. United States Department of Agriculture.

Vallejo, V.E., Z. Arbelia, Wi. Terán, N. Lorenz, R. P. Dick, and F. Roldan. 2012. Effect of land management and Prosopis juliflora (Sw.) DC trees on soil microbial community and enzymatic activities in intensive silvopastoral systems of Colombia. Agriculture, Ecosystems and Environment 150:139-14.

Wright, R. J, D. G. Boyer, W. M. Winant, H. D. Perry. 1990. The influence of soil factors on yield differences among landscape positions in an Appalachian cornfield. Soil Science, Vol. 149, Issue 6.

Yang, C. 1998. Spatial variability of field topography and wheat yield in the Palouse region of the Pacific Northwest. Transactions of ASAE, Vol. 41, pp. 17– 27.

CHAPTER 3: AIR DRYING'S EFFECTS ON OXIDATION ENZYME ASSAYS AND THE RELATIONSHIP OF ENYZME ACTIVITIES TO CROP YIELD

ABSTRACT

There is growing recognition of the need to develop a sensitive soil-quality indicator that reflects land management so as to assist land managers in promoting the long-term sustainability of terrestrial ecosystems. Soil-enzyme activities are useful indicators of soil quality, as they are very sensitive to disturbances. Sample storage and pretreatment affect the results of enzyme assays, which are normally determined in field-moist samples. However, the use of air-dried soils is preferred because such samples are easier to store and handle. Many studies have been done on effects of air drying on specific enzymes activities, but little is known about effects of air drying on rhodanese (RA) and ammonium oxidation enzyme (AO). An important aspect of soil quality is that it should correlate with yield. RA and AO, as oxidation enzymes, are potentially sensitive to soil drainage and landscape position, which affect crop yields. Thus, RA and AO are of interest relative to crop yields, and air-drying pretreatment could facilitate these enzyme assays. The objective of this study was to evaluate the effects that air drying has on enzyme assays and to determine the correlations of particular enzymes with crop yields. The results were that RA and AO were not correlated with crop yields. However, this result was limited by the limitations of the landscape, the accuracy of the drainage-class map, and the actual on-site redox potential of the soil. More investigation is needed for other landscapes. We found that air-dried pretreatment decreased RA and AO activities but that the use of air-dried soil samples provided the same rankings for various tillage managements and increased the enzyme assays' ability to indicate management differences relative to the use of field-moist soil. Thus, enzyme assays that are run on air-dried soil and can maintain treatment effects and can even be more sensitive to soil

management. The results for the air-dried enzyme activity/unit clay did reveal a tillage difference, whereas those for the field-moist enzyme activity/unit clay did not. This is a desirable characteristic for a soil-quality indicator because it allows for a more temporally stable sample, facilitates pretesting, and increases the enzyme assay's ability to act as a quality index, thus making it a more practical method for commercial soil-testing labs.

INTRODUCTION

Soil properties are susceptible to change under various sample-storage and pretreatment regimes. In most cases, soil samples are refrigerated or frozen until the analysis to preserve the original characteristics of the samples. This practice is common in scientific work. Air drying gives the soil samples physical conditions that are ideal for handling and conservation without refrigeration (Zornoza et al., 2006; Lopes et al., 2015; Moreira et al., 2017). Commercial labs and agencies desire rapid soil handling and high-throughput procedures if they are to adopt an indicator. Moreover, the use of air-dried soil and short incubations facilitates routine soil-testing procedures; the use of air-dried soils can also reduce variability within a same soil sample and reduce storage space for refrigeration (Haney et al., 2004). Therefore, air-drying pretreatment can greatly facilitate enzyme assays and encourage a biochemical parameter's adoption as a soil-quality index (Bandick and Dick, 1999).

Li and Sarah (2003) studied the effect of air drying on some enzyme activities along a climatic transect in Israel from the Judean Mountains in the west to the Dead Sea in the east. They found no difference in the field-moist soil samples in terms of arylsulfatase, acid phosphatase, and alkali phosphatase. Chen (2003) used air-dried soils to determine soil phosphatase activity in Chinese fir plantation for rhizosphere and bulk soil, finding differences depending on the soil profile and on the sampling distance from the tree stem. Speir ad Ross (1981) examined the effects that air drying had on the activities of invertase, amylase, cellulase, xylanase, urease, protease, phosphatase, and sulfatase in nine New Zealand soils from pastures. They found that air drying the soils reduced these substances' activities, with losses ranging from slight (for sulfatase) to very

large (for protease). Urease activity increased with air drying in all soils except one, and most of those increases were significant. In addition, Sparling et al. (1986) compared phosphatase and phosphor-diesterase activities before and after air drying soil samples form grassland in New Zealand. They observed declines in both with air drying. Zornoza et al. (2006) evaluated the effects of air drying (both with and without rewetting) on the activities of β-glucosidase, acid phosphatase, and urease for various locations, degradation statuses, and seasons. They found that urease, β-glucosidase, and phosphatase activities were hardly affected by air drying. However, air drying and rewetting caused fluctuations in these enzymes' activities.

Bandick and Dick (1999) compared field-moist and air-dried soils from various management systems and found that, although enzyme activities went down in most cases, α-and β-glucosidase, amidase, arylsulfatase, and urease consistently had the same ranking for field-moist and air-dried samples in soil plots when comparing crops (including their agricultural practices) and a nearby pasture grass in the northwestern United States. They hypothesized that air drying could further improve enzyme assays' ability to provide the true trajectory of management effects on soils because air drying denatures the enzymes associated with viable populations, thus reducing the effects that recent management and environmental impacts have on the highly sensitive and temporally variable living microbial enzyme pool. Thus, more of the activity is associated with the enzymes that are stabilized in the soil matrix; these enzymes are less susceptible than other enzymes to recent environmental conditions and better reflect the long-term trajectory on soil dynamics or health.

Singh and Tabatabai (1977) investigated the effects that pretreatments had on RA and found that, for field-moist soils, air drying resulted in a marked decrease (average: 44%) in RA activity. However, they did not study whether air-drying pretreatments change the various soil-management treatments' rankings in terms of RA.

A further advantage for practical applications is that, unlike most other measures of soil's microbial properties, enzyme assays are relatively simple to perform. Soils are typically incubated for a few hours before colorimetric or fluorescence determinations of the reaction products are conducted. Assays can be adapted for multi-enzyme, high-throughput activity analyses using 96-well microplates (Dick at al., 2013). Thus, the ability to use air-dried samples and the simple methodology allows for large-scale processing and analysis of samples for practical applications.

For most measures of soil properties, a major limitation is that they vary considerably as a function of soil type and season. These differences can be much greater than the subtle changes due to land management, making interpretations of dynamic soil measures difficult (Schutter et al., 2001). This is true for enzyme activities; however, the research across diverse soils in Oregon (Knight, 2002; Knight and Dick, 2004) and (in this study) in Ohio have shown that normalizing enzyme activity to carbon content or clay content can separate out the land-management effects, independent of the soil type. Thus, enzyme activity seems to be a rare soil measure that can be calibrated and interpreted independently of the soil type.

Fungal biomass and bacterial abundance have been consistently found to be greater in organic systems than in conventional systems (Birkhofer et al., 2008; Diepeningen et al., 2006; Gunapala and Scow, 1998; Mulder et al., 2003; Shannon et al.,

2002; Yeates et al., 1997) Others have found that bacterial abundance is less sensitive to management than fungal abundance is (Yeates et al., 1997). However, many scholars have reported a difference in soil microbial diversity and biomass based on conventional versus organic soil management. These results cannot provide the basis for a calibrated soil-quality test, independent of soil type.

Furthermore, a good soil-quality indicator should be correlated with crop yield. However, few studies have tested the correlation between crop yield and enzyme assays. Tautges et al. (2016) found a slight negative correlation between β-glycosaminidase and protein content in spring wheat. In this research, the correlation between oxidation enzymes and crop yield were studied. For oxidation enzymes, there should be a positive correlation with drainage capability. RA and AO were chosen as the oxidation enzymes because they are simple assays and perform oxidation reactions.

The objective of this research is to study air drying's effects on oxidation enzyme assays, as well as the relationship between enzyme activities and crop productivity.

MATERIALS AND METHODS

Site Description

This study was conducted at 18 sites in Iowa, Illinois, and Ohio. Each site included two side-by-side fields: one under no-tillage (NT) and one under conventional tillage (CT), with disturbance and soil mixture of up to 15 cm in depth. Most of the fields had a corn-soybean rotation. The properties of the soil are shown in Table 3.1.

Experimental Design and Soil Sampling

Soil samples were collected from nine pairs of Ag Spectrum and conventional fields in Illinois (3 sites), Iowa (3 sites), and Ohio (3 sites) during September 2017. We measured the crop yields in September 2015, 2016, and 2017. The paired sites were adjacent. To measure a site's crop yield, five corn cobs or ten soybean plants were randomly harvested in a 5-m section of a row (equal to 1/1000th of an acre) at the site; the total number of plants in the 5 m section of the row was then recorded. Both the corn plants and the soybean plants were dried in an oven (65 ℃) and weighed the seeds. The yield of each crop (per acre) was equal to the weight of the seeds from the sampled plants × the number of plants in the 5 m section × 1000 (Nielsen, 2015). The yields of crops were reported using the dry weight. The crop yield on the site was expressed as the grain mass (in Mg) per hectare. All the sites were sampled by taking 15 soil cores in each sampling spot. At the NT sites, samples were took at depths of 0-5 cm and 5-15 cm. For the CT sites, samples were took at depths of 0-15 cm, which is the typical depths to which tillage incorporates crop residues. The soil samples were placed in bags and brought back to the laboratory, where they were stored at -4 ℃ until analysis. All the

samples were passed through a 2 mm sieve and moisture was measured prior to the enzyme analysis. After air drying the samples at room temperature for 48 h, their moisture was measured again; all the RA activities and randomly selected AO activities were also measured.

Soil Enzyme Activity Analyses

After air drying the samples, the rhodanese enzyme activity was determined according to the methods described by Tabatabai and Singh (1976). Each 4 g sample of soil was incubated in an Erlenmeyer flask (50 ml) at 37 °C, with 8 ml of THAM buffer, 1 ml 0.1 M $Na_2S_2O_3$, and 1 ml 0.1 M KCN. After 1 h, we added the $CaSO_4$-formalhyde solution, filtered the suspension, and colorimetrically determined the SCN^- levels. The control samples were performed using the same procedures as for rhodanese activity, except that the soil was autoclaved. Standard ferric thiocyanate solution was used to determine the calibration curve. The activity of rhodanese was expressed as SCN^- nmol g^{-1} h^{-1}.

After air drying, ammonium oxidation enzyme was determined by the method of International Organization of Standardization (ISO 15685, 2004), which Berg and Rosswall (1985) first described as estimation of both potential and actual oxidation rates for ammonium oxidizers. 5 g of soil was incubated at 25 °C with ammonium sulfate and sodium chlorate. After 5 h, potassium chloride solution was added, and the suspension was filtered, then the ammonium buffer and color reagent were added into the suspension, and the amount of nitrite released was determined colorimetrically. The same procedures were performed on the control samples, except that the soil was incubated at -20 °C for 5 h. The standard potassium nitrite solution was used to determine

the calibration curve. The values of ammonium oxidation enzyme activity were expressed as µg NO_2-N g^{-1} 5 h^{-1}.

Both rhodanese and ammonium oxidation were calculated on a per-unit-clay basis.

Soil Chemical Characteristics

Soil samples were sent to Midwest Laboratories (13611 B Street, Omaha, Nebraska) to determine their chemical properties. The organic matter was determined using the loss of weight on ignition (NCR, 2011); the phosphorus was determined by extraction with dilute acid and ammonium fluoride (weak Bray) or by a colorimetric assessment (NCR, 2011); the pH was measured with an electrode in a 1:1 soil: water solution (NCR, 2011); the soil texture was determined using the hydrometer method (ASA, 1982); potassium, magnesium, and calcium were extracted with neutral ammonium acetate and measured using inductively coupled argon plasma detection (RMST, 1974; NCR, 2011). The results are presented in Table 3.1.

Statistical Analyses

All the statistical tests were performed using SAS version 9.1 for Windows (SAS Institute Inc., 2004). Two-way analysis of variance models were used to assess the main effects and the interaction of spatiality and drainage. The Shapiro-Wilks test was used to determine normality ($p < 0.05$) and the pairwise comparisons was calculated using Tukey's honestly significant difference comparisons (based on a 95% or 90% confidential interval). However, a nonparametric Kruskal-Wallis rank-order analysis was used to determine the influence of the various treatments if the statistics did not meet the normal distribution criteria of general linear models. Linear regression was applied to examine the relationship between enzyme activities and crop yield. Spearman's rank

correlation was used as a nonparametric alternative to linear regression when we could not achieve normality through data transformation.

RESULTS

Effects of Drainage and Tillage

In field-moist soil samples, AO activities were higher for NT at the 0-5 cm depth than for CT (0-15 cm depth), but no significant difference was found in RA activities. After air drying, both enzymes decreased (Table 3.4), but the significant treatment effects and the rankings of the treatments was the same as in the assays run on field-moist soil (Table 3.4). Tables 3.8 and 3.9 present air-dried RA activity, AO activity, and enzyme activity/unit clay for NT and CT. The AO activity was higher in the NT at 0-5 cm depth than in the CT (0-15 cm depth). The AO activity/unit clay also had this difference. Although AO activity in NT (5-15 cm depth) did not have a tillage difference, AO activity/unit clay at this depth was significantly higher for CT (0-15 cm depth) than for NT (5-15 cm depth). The RA activity for NT at the 0-5 cm depth did indicate a tillage difference; it was higher compared to CT (0-15 cm depth). The RA activity/unit clay for NT (0-5 cm depth depth) was also higher than for CT (0-15 cm depth). We found no significant difference in RA activity or in RA activity/unit clay for NT at the 5-15 cm depth compared to CT (0-15 cm depth). The use of enzyme activity/unit clay increased the difference in enzyme activity between NT and CT. These results were also supported by Table 3.2, which shows that the use of enzyme activity/unit clay did change the

rankings of the enzyme assay. This change increased the enzyme assay's ability to detect management differences, especially when measuring air-dried soil.

Tables 3.5 through 3.8 show RA and AO activities in air-dried soil samples under various levels of drainage. Although we observed no significant difference in enzyme activities, for NT, AO activity was higher in well-drained soil than in poorly drained soil. RA activity was also higher in well-drained soil than that in poorly drained soil except for the Ohio samples; however, none of these differences were significant. RA in field-moist soil also had higher activity in well-drained soil than in poorly drained soil except for in the Ohio samples. Generally, air-drying pretreatment did not change the RA rankings within each drainage class.

Correlation of Air-Dried Enzyme Activities with Crop Yields

Table 3.3 indicates that the enzyme activities for air-dried and fresh soil were highly correlated. Table 11 shows the coefficients for the correlations between the crop yields and the weighted means of the air-dried enzyme activities or the weighted means of the air-dried enzyme activities/unit clay. However, neither correlation was significant at $\alpha = 0.05$. AO activities were slightly positively correlated with soybean yields ($r = 0.22$, $p = 0.48$) and slightly negatively correlated with corn yields ($r = -0.04$, $p = 0.88$). RA activities were negatively correlated with both soybean yields ($r = -0.26$, $p = 0.28$) and corn yields ($r = -0.13$, $p = 0.39$). The ratio of enzyme activity to clay percentage had greater correlation coefficients than did the actual activity in the relationship with corn yields. All r values for the enzyme activity/unit clay were higher than those for the enzyme activity alone. In addition, some of the r values went from negative for enzyme activity to positive for enzyme activity/unit clay.

Tables 3.12 and 3.13 present the correlations between the enzyme activities and the actual crop yields in 2017 under NT and CT. Under CT, we found higher correlation coefficients in the enzymes with soybean yields than in those with the corn yields. RA was negatively correlated with both soybean and corn yields; AO was positively correlated with soybean yields but negatively correlated with corn yields. Generally, enzyme activity/unit clay had higher r values than enzyme activity alone. However, we found no significant correlation in this comparison under CT or under NT.

Tables 3.14 and 3.15 present the coefficients for the correlations between crop yields and enzyme activities for soil samples at the 0-5 cm and 5-15 cm depths (NT). There were no significant correlations at these depths for the enzyme activity or for the enzyme activity/unit clay, with the exception of AO activity/unit clay. In the correlations of the enzyme activities with crop yields, only one positive correlation was observed: AO with soybean yields.

DISCUSSION

Singh and Tabatabai (1977) investigated the effects that pretreatments had on RA; they found that the air drying of field-moist soils resulted in a marked decrease in RA activity (average: 44%). However, they did not study whether air-drying pretreatments change the rank of RA activity for all the soil-management treatments. In the present study, the AO for both field-moist and air-dried soil was significantly higher in NT (0-5 cm depth depth) than in CT (0-15 cm depth). In the RA assay in air-dried soil, we also detected higher activity in NT at the 0-5 cm depth. The air-drying pretreatment decreased the RA and AO activities; however, this pretreatment did not change the rankings within the tillage management types, and it increased the RA assay's ability to serve as a soil-

quality indicator. Bandick and Dick (1999) compared 11 enzyme activities in field-moist and air-dried samples for two soil treatments and found that, although activity went down in most cases, the rankings of the treatments for the air-dried samples were the same as for the field-moist samples. Just as with these enzymes, RA and AO have the ability to be practical for use in commercial labs.

The use of the air-dried enzyme activity/unit clay increased the enzyme assays' sensitivity to tillage. The air-dried AO activity/unit clay in NT at the 5-15 cm depth was significantly lower than in CT at the 0-15 cm depth, although the AO activity/unit clay in field-moist soil samples did not detect this difference. Air drying could denature the part of the enzyme related to viable microbes and increase the enzyme's sensitivity to tillage in the soil matrix. In addition, calibrating the enzyme assay using enzyme activity/unit clay could reduce the influence due to the soil type, therefore increasing the enzyme assay's ability to detect tillage differences.

We had expected RA, as an oxidation enzyme, to decrease along a gradient from well-drained to poorly drained soils. Therefore, we had expected a positive correlation between RA and crop yields. However, we actually found a slightly negative relationship between soybean yields and RA. This negative correlation was most evident at the 5-15 cm depth. We observed significantly negative correlation between RA and crop yields. We had expected AO to be positively correlated with crop yields. Although we observed slightly positive or negative correlations between AO activity and crop yields, none of these were significant. Although air-drying pretreatment did change the r values of the enzyme activity for the crop yields, this difference was not large.

To be effective and practical, soil-quality indicators must reflect crop yields. However, there is very little documented information about the relationship between soil quality and crop yields.

RA is distributed widely and has been detected in plants (Chew, 1973), several bacteria (Brown et al., 1965; Smith and Lascelles, 1966; Stearns, 1953), and in the soil (Tabatabai and Singh, 1976). RA catalyzes the formation of thiocyanate and sulfite from thiosulfate and cyanide ($S_2O_3^{2-} + CN^- \longrightarrow SCN^- + SO_3^{2-}$). Because RA apparently does not play an important role in nutrient cycling, the correlation between RA and crop yields might not be significant.

Good drainage does have a positive influence on crop yields; however, this is based on comparisons between extremely poorly drained soils and well-drained soils (Takajima et al., 2013; Lal and Taylor, 1970; Cannell et al., 1979; Power et al., 2000). Because the sites in the present study were relatively flat and had limited elevation differences, their drainage differences may not be important enough to cause significant effects on crop yields or oxidation enzymes. The redox potential of soil can also be determined further using other methods, such as the measurement of reduction potential (E_h) instead of the drainage class. In addition, RA and AO can be controlled by a host of factors. For example, RA has been shown to be influenced by forest type (Lettl, 1986), chemicals (Deng, 1990; Singh and Tabatabai, 1977), and trace elements (Singh and Tabatabai, 1977). Thus, other factors could confound the effects that landscape position have on enzyme activities.

Our conclusion is that air-dried AO and RA activities are not correlated with crop yields, likely due to the similarity of the topography across the sampling sites or intrinsic

properties of soil. The use of enzyme activity/unit clay increased the correlations of RA and AO with crop yields. This increase may be due to the immobilized enzymes in clay, which are stable and relatively insensitive to temporal disturbances such as precipitation and temperature; this would reduce the influence of soil type.

CONCLUSIONS

Air drying did decrease RA and AO activities, but it did not change the ranking of enzyme activities between NT and CT treatments, relative to the ranking of those activities in field-moist soil samples. Furthermore, air-drying pretreatment increased the enzyme assays' ability to detect tillage differences, and calibrating the enzyme activity as the enzyme activity/unit clay further increased this ability. Air-drying pretreatment is desired because it facilitates the adoption of these assays for practical commercials applications; the calibration of enzyme activity is a possible method to indicate soil quality, independent of soil type.

In this study, we hypothesized that landscape position would significantly affect AO, RA, and crop yields, which would also be associated with drainage. However, we found no significant correlation between corn or soybean yields with air-dried activities (either absolutely or on a per-unit-clay basis). One possible reason is that the fields were relatively flat, meaning that there were no distinct differences in drainage that would change the yields or enzyme activities. The Intrinsic properties of soil and fertilizer input may also have confounded this correlation. More research is needed in areas with greater differences in landscape position and drainage. Air-drying pretreatment barely changed

the correlation between enzyme activities and crop yield. The use of RA activity/unit clay and AO activity/unit clay did increase the correlations with crop yields.

TABLES

Table 3.1 Properties of selected sites in Ohio, Illinois, and Iowa.

Site name	Coordinates	Series Name	Taxonomic name	Soil type	Drainage class‡	pH	Organic matter	Sand	Clay	P	K	Mg	Ca
							——————%———			——————— μg g^{-1}——			
IA-MET	N41°50'18.14" W90°20'50.16"	Dinsdale	Fine-silty, mixed, superactive, mesic Typic Argiudolls	silt, loam	1,2	5.7	3.2	9	23	20	218	305	1699
IA-DIE	N41°50'45.12" W90°38'33.82"	Klinger	Fine-silty, mixed, superactive, mesic Aquic Hapludolls	silt, loam	1,2,3	6.5	4.7	26	22	30	195	533	2422
IA-VIC	N41°50'0.09" W90°34'37.59"	Tama	Fine-silty, mixed, superactive, mesic Typic Argiudolls	silt, loam	1	5.9	2.8	11	22	7	104	287	1733
IL-MCK	N40°37'25.04" W90°29'25.56"	Sable	Fine-silty, mixed, superactive, mesic Typic Endoaquolls	silt, clay, loam	1,2,3	6.1	4.3	10	28	20	168	369	2794
IL-EMO	N40°37'28.41" W90°30'43.21"	Ipava	Fine, smectitic, mesic Aquic Argiudolls	silt, clay, loam	2,3	6	3.8	7	29	23	168	291	2516

Table 3.2 (continued) Properties of selected sites in Ohio, Illinois, and Iowa.

Site name	Coordinates	Series Name	Taxonomic name	Soil type	Drainage class‡	pH	Organic matter	Sand	Clay	P	K	Mg	Ca
							——————%———			——————— µg g^{-1}——			
IL-WEA	N40°37'19.67" W90°29'46.13"	Osco	Fine-silty, mixed, superactive, mesic Typic Argiudolls	silt, clay, loam	1,2	6.3	4.6	11	28	19	153	406	3018
OH-CIR1	N39°40'13.24" W82°55'44.48"	Crosby	Fine, mixed, active, mesic Aeric Epiaqualfs	loam	1,2	5.8	2.4	33	22	16	93	312	1495
OH-CIR2	N39°39'16.98" W82°56'2.04"	Miamian	Fine, mixed, mesic Typic Hapludalfs	loam	1,2	5.5	1.9	42	18	38	135	148	954
OH-CIR3	N39°39'12.58" W82°57'9.25"	Miamian	Fine, mixed, mesic Typic Hapludalfs	loam	1,2	5.9	2.8	38	16	16	90	270	1344

† pH, organic matter percent, P, K, Mg, Ca, sand, and clay percentages are averaged across drainage class with a regional location.
‡ 1 = well-drained soils; 2 = somewhat poorly drained soils; 3 = poorly drained soils

Table 3.3 Enzyme activities and enzyme activities/unit clay in no tillage and conventional tillage.

			AO activity				RA activity			
			No tillage (0-5 cm depth)		Conventional tillage (0-15 cm depth)		No tillage (0-5 cm depth)		Conventional tillage (0-15 cm depth)	
Site name	Coordinates	Clay (%)	Actual†	Unit clay‡	Actual	Unit clay	Actual	Unit clay	Actual	Unit Clay
IA-MET	N41°50’18.14” W90°20’50.16”	23	N/A	N/A	0.850	3.820	202.7	873.1	174.6	798.9
IA-DIE	N41°50’45.12” W90°38’33.82”	22	2.560	11.81	N/A	N/A	114.2	532.4	101.5	439.6
IA-VIC	N41°50’0.09” W90°34’37.59”	22	N/A	N/A	0.510	2.470	113.0	550.6	96.20	443.9
IL-MCK	N41°50’0.09” W90°34’37.59”	28	0.2100	0.9700	1.030	2.580	153.9	631.1	120.7	411.3
IL-EMO	N40°37’28.41” W90°30’43.21”	29	1.030	3.040	0.3200	1.350	112.7	360.6	112.3	445.0
IL-WEA	N40°37’19.67” W90°29’46.13”	28	2.210	8.380	1.740	5.500	148.1	571.5	161.0	533.0
OH-CIR1	N39°40’13.24” W82°55’44.48”	22	2.160	11.26	0.6400	2.540	99.44	525.7	94.10	379.8
OH-CIR2	N39°39’16.98” W82°56’2.04”	18	2.020	10.26	1.650	7.170	197.1	1143	94.89	579.1
OH-CIR3	N39°39’12.58” W82°57’9.25”	16	2.020	11.35	1.040	7.120	100.7	630.2	95.40	659.2

† “Actual”: actual enzyme activity. The unit of AO activity is NO_2^- μmol g^{-1} 5 h^{-1}; the unit of RA activity is SCN^- nmol g^{-1} 1 h^{-1}.
‡ “Unit clay”: enzyme activity/unit clay; the units are NO_2^- μmol g^{-1} 5 h^{-1} clay percentage$^{-1}\times10^4$ and SCN^- nmol g^{-1} 1 h^{-1} clay percentage$^{-1}\times10^4$.

Table 3.4 Correlation coefficients (r) of enzyme activities in air-dried and field-moist soil.

Field-moist soil	Air-dried soil	
RA	RA	0.834***
RA/clay	RA/clay	0.803***
AO	AO	0.874***
AO/clay	AO/clay	0.753***

*** indicates significance at the 0.001 levels.

Table 3.5 Comparison of enzyme activities in field-moist and air-dried samples in no tillage (0-5 cm and 5-15 cm depth) and conventional tillage (0-15 cm depth).

		No tillage		Conventional tillage (0-15 cm)	
Enzyme	Depth of no tillage	Field-moist	Air dried	Field-moist	Air dried
RA[‡]	0-5 cm	192 a (79.5)	136 b (56.5)	174 a (74.0)	113 b (42.8)
AO[§]	0-5 cm	2.06 a (1.78)[†]	1.97 b (1.37)	1.21 a (0.76)	0.97 b (0.88)
RA	5-15 cm	156 a (101)	103 b (87)	174 a (74.0)	113 b (43)
AO	5-15 cm	0.92 a (0.78)	0.54 b (0.56)	1.21 a (0.76)	0.97 b (0.88)

[†] Numbers in parentheses indicate sample standard errors.
[‡] The unit of RA activity is released SCN^- nmol g^{-1} 1 h^{-1}.
[§] The unit of AO activity is released NO_2^- µmol g^{-1} 5 h^{-1}.
[¶] Different letters indicate that the means in a row are significantly different ($p < 0.1$).

Table 3.6 Effect of drainage class on soil ammonium oxidation enzyme activity (air-dried soil samples) across all sites.

Management	Depth	Poorly drained		Somewhat poorly drained		Well drained	
		Mean	Standard deviation	Mean	Standard deviation	Mean	Standard deviation
		NO_2^- µmol g^{-1} 5 h^{-1}					
No tillage	0-5 cm	1.29 a[†]	1.36	2.48 a	1.32	1.97 a	1.47
	5-15 cm	0.29 a	0.21	0.90 a	0.89	0.45 a	0.26
Conventional tillage	0-15 cm	1.82 a	0.10	0.88 a	0.79	0.78 a	0.81

[†] Values within a row followed by same letters are not significantly different ($p < 0.05$).

Table 3.7 Effect of drainage class on rhodanese activity (air-dried soil samples) across Illinois.

		Poorly drained		Somewhat poorly drained		Well drained	
Management	Depth	Mean	Standard deviation	Mean	Standard deviation	Mean	Standard deviation
		SCN^- nmol g^{-1} h^{-1}					
No tillage	0-5 cm	103 a[†]	14.5	123 a	39.4	180 a	41.7
	5-15 cm	77.2 a	18.6	71.0 a	56.8	92.1 a	45.5
Conventional tillage	0-15 cm	115 a	26.6	122 a	12.0	140 a	46.2

[†] Values within a row followed by same letters are not significantly different ($p < 0.05$).

Table 3.8 Effect of drainage class on soil rhodanese activity (air-dried soil samples) across Iowa.

		Poorly drained		Somewhat poorly drained		Well drained	
Management	Depth	Mean	Standard deviation	Mean	Standard deviation	Mean	Standard deviation
		SCN^- nmol g^{-1} h^{-1}					
No tillage	0-5 cm	104 a[†]	29.0	158 a	44.4	136 a	58.7
	5-15 cm	72.0 a	32.5	101 a	87.3	173 a	178
Conventional tillage	0-15 cm	53.9 a	N/A	148 a	87.3	121 a	48.8

[†] Values within a row followed by same letters are not significantly different ($p < 0.05$).

Table 3.9 Effect of drainage class on soil rhodanese activity (air-dried soil samples) across Ohio.

Management	Depth	Poorly drained		Somewhat poorly drained		Well drained	
		Mean	Standard deviation	Mean	Standard deviation	Mean	Standard deviation
		SCN^- nmol g^{-1} h^{-1}					
No tillage	0-5 cm	135 a[†]	N/A	144 a	53.6	132 a	91.1
	5-15 cm	105 a	N/A	88.6 a	42.5	106 a	68.9
Conventional tillage	0-15 cm	123 a	41.2	112 a	14.3	81.9 a	38.1

[†] Values within a row followed by same letters are not significantly different ($p < 0.05$).

Table 3.10 Effect of soil management on ammonium oxidation enzyme activities (air-dried soil samples) and ammonium oxidation enzyme activities/unit clay (air-dried soil samples).

		No tillage		Conventional tillage (0-15 cm)	
Enzyme	Depth in no tillage	Mean	Standard deviation	Mean	Standard deviation
AO activity[§]	0-5 cm	1.97 A[‡]	1.37	0.97 B	0.88
AO activity	5-15 cm	0.54 a	0.52	0.97 a	0.88
AO activity/unit clay[¶]	0-5 cm	9.60 A	7.33	4.32 B	3.32
AO activity/unit clay	5-15 cm	2.56 b[†]	2.07	4.32 a	3.32

[†] Values within a row followed by the same lowercase letters are not significantly different ($p < 0.1$).
[‡] Values within a row followed by the same capital letters are not significantly different at ($p < 0.05$).
[§] The unit of AO activity is NO_2^- µmol g^{-1} 5 h^{-1}.
[¶] The unit of AO activity/unit clay is NO_2^- µmol g^{-1} 5 h^{-1} clay $percent^{-1} \times 10^4$.

Table 3.11 Effect of soil management on rhodanese activities (air-dried soil samples) and rhodanese enzyme activities/unit clay (air-dried soil samples).

		No tillage		Conventional tillage (0-15 cm)	
Enzyme	Depth in no tillage	Mean	Standard deviation	Mean	Standard deviation
RA activity[§]	0-5 cm	136 a[†]	56.5	113 b	42.8
RA activity	5-15 cm	103 a	86.8	113 a	42.8
RA activity/unit clay[¶]	0-5 cm	645 A[‡]	321	507 B	203
RA activity/unit clay	5-15 cm	488 a	417	507 a	203

[†] Values within a row followed by the same lowercase letters are not significantly different ($p < 0.1$).
[‡] Values within a row followed by the same capital letters are not significantly different ($p < 0.05$).
[§] The unit of RA activity is SCN^- nmol g^{-1} $1h^{-1}$.
[¶] The unit of RA activity/unit clay is SCN^- nmol g^{-1} 1 h^{-1} clay $percent^{-1} \times 10^4$.

Table 3.11 Correlation coefficients (r) of crop yields (Mg ha^{-1}) in terms of weighted air-dried enzyme activity or enzyme activity/unit clay across all sites.[†]

	Soybeans		Corn	
Enzyme activity	r	p[‡]	r	p
RA	-0.26	0.28	-0.13	0.39
RA/clay	0.35	0.14	0.05	0.76
AO	0.22	0.48	-0.04	0.88
AO/clay	0.48	0.11	0.07	0.76

[†] The crop yields are actual measured yields from fields in 2017 using conventional tillage and measured at 0-15 cm depth. The unit of RA activity is SCN^- nmol g^{-1} 1 h^{-1}, and the unit of AO activity is NO_2^- µmol g^{-1} 5 h^{-1}. Enzyme activity/unit clay = (enzyme activity)/(clay percentage) × 100; the units are NO_2^- µmol g^{-1} 5 h^{-1} clay $percentage^{-1} \times 10^4$ and SCN^- nmol g^{-1} 1 h^{-1} clay $percentage^{-1} \times 10^4$.
[‡] The correlations are significant ($p < 0.05$).

Table 3.12 Correlation coefficients (r) of crop yields (Mg ha^{-1}) in terms of air-dried enzyme activity or enzyme activity/unit clay under conventional tillage.[†]

	Soybeans		Corn	
Enzyme activity	r	p[‡]	r	p
RA	-0.58	0.11	-0.16	0.48
RA/clay	0.27	0.49	0.18	0.41
AO	0.49	0.36	-0.05	0.88
AO/clay	0.60	0.24	0.23	0.50

[†] The crop yields are actual measured yields from fields in 2017 using conventional tillage and measured at 0-15 cm depth. The unit of RA activity is SCN^- nmol g^{-1} 1 h^{-1}, and the unit of AO activity is NO_2^- µmol g^{-1} 5 h^{-1}. Enzyme activity/unit clay = (enzyme activity)/(clay percentage) × 100; the units are NO_2^- µmol g^{-1} 5 h^{-1} clay $percentage^{-1} \times 10^4$ and SCN^- nmol g^{-1} 1 h^{-1} clay $percentage^{-1} \times 10^4$.
[‡] The correlations are significant ($p < 0.05$).

Table 3.13 Correlation coefficients (r) of crop yields (Mg ha^{-1}) in terms of air-dried weighted enzyme activity or enzyme activity/unit clay under no tillage.[†]

	Soybeans		Corn	
Enzyme activity	r	p[‡]	r	p
RA	-0.02	0.97	-0.09	0.69
RA/clay	0.36	0.31	-0.06	0.79
AO	0.09	0.92	-0.02	0.98
AO/clay	0.54	0.30	-0.07	0.88

[†] The crop yields are actual measured yields from fields in 2017 using no tillage and measured at 0-15 cm depth. The unit of RA activity is SCN^- nmol g^{-1} 1 h^{-1}, and the unit of AO activity is NO_2^- µmol g^{-1} 5 h^{-1}. Enzyme activity/unit clay = (enzyme activity)/(clay percentage) × 100; the units are NO_2^- µmol g^{-1} 5 h^{-1} clay $percentage^{-1} \times 10^4$ and SCN^- nmol g^{-1} 1 h^{-1} clay $percentage^{-1} \times 10^4$.

[‡] The correlations are significant ($p < 0.05$).

Table 3.14 Correlation coefficients (r) of crop yields (Mg ha^{-1}) in terms of air-dried enzyme activity or enzyme activity/unit clay under no tillage (0-5 cm depth).†

	Soybeans		Corn	
Enzyme activity	r	p‡	r	p
RA	-0.03	0.94	-0.09	0.69
RA/clay	0.33	0.35	-0.23	0.30
AO	0.05	0.92	-0.05	0.89
AO/clay	0.37	0.48	0.05	0.89

† The crop yields are actual measured yields from fields in 2017 using no tillage and measured at 0-5 cm depth. The unit of RA activity is SCN^- nmol g^{-1} 1 h^{-1}, and the unit of AO activity is NO_2^- µmol g^{-1} 5 h^{-1}. Enzyme activity/unit clay = (enzyme activity)/(clay percentage) × 100; the units are NO_2^- µmol g^{-1} 5 h^{-1} clay $percentage^{-1} \times 10^4$ and SCN^- nmol g^{-1} 1 h^{-1} clay $percentage^{-1} \times 10^4$.

‡ The correlations are significant ($p < 0.05$).

Table 3.15 Correlation coefficients (r) of crop yields (Mg ha^{-1}) in terms of air-dried enzyme activity or enzyme activity/unit clay under no tillage (5-15 cm depth).†

	Soybeans		Corn	
Enzyme activity	r	p‡	r	p
RA	-0.19	0.59	-0.06	0.77
RA/clay	0.018	0.97	-0.04	0.86
AO	0.41	0.42	-0.23	0.55
AO/clay	0.86	0.03	-0.22	0.58

† The crop yields are actual measured yields from fields in 2017 using no tillage and measured at 5-15 cm depth. The unit of RA activity is SCN^- nmol g^{-1} 1 h^{-1}, and the unit of AO activity is NO_2^- µmol g^{-1} 5 h^{-1}. Enzyme activity/unit clay = (enzyme activity)/(clay percentage) × 100; the units are NO_2^- µmol g^{-1} 5 h^{-1} clay $percentage^{-1} \times 10^4$ and SCN^- nmol g^{-1} 1 h^{-1} clay $percentage^{-1} \times 10^4$.

‡ The correlations are significant at $p < 0.05$.

REFERENCES

ASA - Methods of Soil Analysis - Part 2: Chemical and Microbiological Properties, Second Edition, 1982. American Society of Agronomy. pp. 549-556.

Birkhofer, K., T.M. Bezemer, J. Bloem, M. Bonkowski, S. Christensen, D. Dubois, F. Ekelund, A. Fliessbach, L. Gunst, K. Hedlund, P. Mader, J. Mikola, C. Robin, H. Setala, F. Tatin-Froux, W.H. Van der Putten and S. Scheu. 2008. Long-term organic farming fosters below and aboveground biota: implications for soil quality, biological control and productivity. Soil Biology and Biochemistry 40: 2297–2308.

Brown, T. J., P. J. Butler and F. C. Happold. 1965. Some properties of the rhodanese system of Thiobacillus denitrificans. Biochemical Journal 97: 651-657.

Chen, H. 2003. Phosphatase activity and P fractions in soils of an 18-year-old Chinese fir (Cunninghamia lanceolata) plantation**.** Forest Ecology and Management 178: 301-310.

Chew, M. Y. 1973. Rhodanese in higher plants. Phytochemistry 12: 2365-2367.

Deng, S. 1990. Sulfur oxidation and rhodanese activity in soils. Soil Science, Vol. 150, pp. 552-560.

Dick, L.K., G. Jia, S. Deng, and R. P. Dick, 2013. Evaluation of microplate and bench-scale β-glucosidase assays for reproducibility, comparability, kinetics, and homogenization methods in two soils. Biol. Fert. Soils (in press).

Gunapala, N., K.M. Scow. 1998. Dynamics of soil microbial biomass and activity in conventional and organic farming systems. Soil Biology Biochemistry 30: 805–816.

Haney, R.L., A.J. Franzluebbers, E.B.Porter, F.M. Hons, D.A. Zuberer. 2004. Soil carbon and nitrogen mineralization: influence of drying temperature**.** Soil Science Society of America Journal 68: 489-492.

Knight, T, 2002. Soil enzyme stability in disturbed ecosystems. MS thesis. Oregon State University, Corvallis OR.

Knight, T, and R. P. Dick, 2004. Differentiating microbial and stabilized β-glucosidase activity relative to soil quality. Soil Biology and Biochemistry 36: 2089-2096.

Lettl, A. 1986. Thiosulfate sulfurtransferase (Rhodanese) in forest soils. Forestry and game management research institute.

Li, X., P. Sarah. 2003. Enzyme activities along a climatic transect in the Judean Desert. Catena 53: 349-363

Lopes, A.A.C., D.M.G. Sousa, F. B. Jr. Reis, I.C. Mendes. 2015. Air-drying and long-term storage effects on β-glucosidase, acid phosphatase and arylsulfatase activities in a tropical Savannah Oxisol. Applied Soil Ecology 93: 68–77.

Moreira, R. S., M. K. Chiba, S. B. Nunes and I. C. D. de Maria. 2017. Air-drying pretreatment effect on soil enzymatic activity. Plant Soil and Environment 63: 29-33.

Mulder, C., D. de Zwart, H.J. van Wijnen, A.J. Schouten, A.M. Breure. 2003. Observational and simulated evidence of ecological shifts within the soil nematode community of agroecosystems under conventional and organic farming. Funct. Ecol. 17, 516–525.

NCR. 2011. Recommended Chemical Soil Test Procedures for the North Central Region. No. 499 (revised). North Central Regional Research Publication. North Dakota State University. pp. 5-32.

Nielsen, R. L. 2015. Estimating grain yield prior to harvest. Purdue University. URL: http://www.kingcorn.org/news/timeless/YldEstMethod.html.

RMST. 1974. Handbook on Reference Methods for Soil Testing, Council on Soil Testing and Plant Analysis. pp. 60-65.

Schutter, M., J. Sandeno and R.P. Dick, 2001. Seasonal, soil type, and alternative management influences on microbial communities of vegetable cropping systems. Biology and Fertility of Soils 34: 397-410.

Shannon, D., A.M. Sen, D.B. Johnson. 2002. A comparative study of the microbiology of soils managed under organic and conventional regimes. Soil Use Management 18: 274–283.

Singh, B. B. and M. A. Tabatabai. 1977. Factors affecting rhodanese activity in soils. Soil Science, Vol. 125, pp. 337-342.

Singh, B.B., Tabatabai, M.A. 1976. Rhodanese activity of softs. Soil Science Society of America Journal 40: 381-385.

Smith, A. J. and J. Lascelles. 1966. Thiosulphate metabolism and rhodanese in Chromatium sp. strains. Journal of General Microbiology 42: 357-370.

Sparling, G.P., T.W. Speir, K.N. Whale**.** 1986. Changes in microbial biomass C, ATP content, soil phosphomonoesterase and phosphodiesterase activity following air-drying of soil. Soil Biology and Biochemistry 18: 363-370.

Speir, T. W., and D. J. Ross. 1981. A comparison of the effects of air-drying and acetone dehydration on soil enzyme activities. Soil Biology and Biochemistry 13: 225-229.

Stearns, R. H. 1953. Respiration, rhodanese and growth in Escherichia coli. Journal of Cellular and Comparative Physiology 41: 163-l 70.

Tautges, N. E., T. S. Sullivan, C. L. Reardon and I. C. Burke. 2016. Soil microbial diversity and activity linked to crop yield and quality in a dryland organic wheat production system. Applied Soil Ecology 108: 258-268

Van Diepeningen, A.D., de Vos, O.J., Korthals, G.W., van Bruggen, A.H., 2006. Effects of organic versus conventional management on chemical and biological parameters in agricultural soils. Applied Soil Ecology 31: 120–135.

Yeates, G.W., R.D. Bardgett, R. Cook, P.J. Hobbs, P.J. Bowling and J.F. Potter. 1997. Faunal and microbial diversity in three Welsh grassland soils under conventional and organic management regimes. Journal of Applied Ecology 34: 453–470.

Zornaza, R., C. Guerrero, J. Mataix-Solera, V. Arcenegui, F. García-Orenes, J. Mataix-Beneyto. 2006. Assessment air-drying and rewetting pretreatment effect on some soil enzyme activities under Mediterranean conditions. Soil Biology and Biochemistry 38: 2125-2134.

Zornoza, R., C. Guerrero, J. Mataix-Solera, V. Arcenegui, F. García- Orenes, J. Mataix-Beneyto. 2006: Assessing air-drying and rewetting pre-treatment effect on some enzyme activities under Mediterranean conditions. Soil Biology and Biochemistry 38: 2125–2134.

COMPLETE REFERENCES

Abrahamson, D. A., M. L. Norfeet, H. J. Causarano, J. R. Williams, J. N. Shaw and A. J. Franzluebbers. 2007. Effectiveness of the soil conditioning index as a carbon management tool in the southeastern USA based on comparison with EPIC. Journal of Soil and Water Conservation 62(2): 94–102.

Acosta-Martinez, V., D. Acosta-Mercado, D. Sotomayor-Ramirez, and L. Cruz-Rodriguez. 2008. Microbial communities and enzymatic activities under different management in semiarid soils. Applied Soil Ecology 38: 249- 260.

Afyuni, M. M., D. K. Cassel and W. P. Robarge. 1993. Effect of landscape position on soil water and corn silage yield. Soil Science Society 57: 1573-1580.

ASA. 1982. Methods of Soil Analysis - Part 2: Chemical and Microbiological Properties, Second Edition. American Society of Agronomy. pp. 549-556.

Aune, J.B., Lal, R., 1997. The tropical soil productivity calculator — a model for assessing effects of soil management on productivity. In: Lal, R., Stewart, B.A. (Eds.), Soil Management — Experimental Basis for Sustainability and Environmental Quality. Adv. Soil Sci., Lewis Publishers, London, UK, pp. 499–520.

Balesdent, J., C. Chenu, M. Balabane, 2000. Relationship of soil organic matter dynamics to physical protection and tillage. Soil and Tillage Research, vol. 53, pp. 215-230.

Balota, E.L., A.C. Filho, D.S. Andrade, R.P. Dick. 2004. Long-term tillage and crop rotation effects on microbial biomass and C and N mineralization in a Brazilian Oxisol. Soil and Tillage Research 77: 137–145.

Bandick, A., and R. P. Dick. 1999. Field management effects on soil enzyme activities. Soil Biology and Biochemistry 31:1471-1479.

Bastida, F., Moreno, J.L., Hernández, T., García, C. 2006. Microbiological degradation index of soils in a semiarid climate. Soil Biology and Biochemistry 38: 3463–3473.

Bastida, F., Zsolnay, A., Hernández, T., García, C., 2008. Past, present and future of soil quality indices: a biological perspective. Geoderma 147: 159–171.

Bennett, O.L. 1972. Effects of north and south facing slopes on yield of Kentucky Bluegrass with variable rate and time of nitrogen Application. Agronomy Journal 64: 630– 635.

Benjamin, J. G., D. L. Karlen. 2014. LLWR techniques for qualifying potential soil compaction consequences of crop residue removal. Bioenergy Research 7: 468-480.

Bhagat, S.P. 1990. Creation in crisis. Brethren Press, Elgin, lL.

Birkhofer, K., T.M. Bezemer, J. Bloem, M. Bonkowski, S. Christensen, D. Dubois, F. Ekelund, A. Fliessbach, L. Gunst, K. Hedlund, P. Mader, J. Mikola, C. Robin, H. Setala, F. Tatin-Froux, W.H. Van der Putten and S. Scheu. 2008. Long-term organic farming fosters below and aboveground biota: implications for soil quality, biological control and productivity. Soil Biology and Biochemistry 40: 2297–2308.

Bloem, J. Schouten, A. J. Sørensen, S. J. Rutgers, M. Werf, A. van der Breure, A. M. 2006. Microbiological methods for assessing soil quality. Chapter 3, pp. 23-49.

Blouin,M., M.E. Hodson, E.A. Delgado, G.Baker, L. Brussard, K.R. Butt, J. Dai, L.Dend ooven, G. Peres, J.E. Tondoh, D.Cluzeau, J.J. Brun. 2013. A review of earthworm impact on soil function and ecosystem services. European Journal of Soil Science, 64: 161-182.

Borchers, J. G., and D. A. Perry. 1992. The influence of soil texture and aggregation on carbon and nitrogen dynamics in southwest Oregon forests and clear cuts. Canadian Journal of Forest Research 22:298-305.

Borchers, J. G., and D. A. Perry. 1992. The influence of soil texture and aggregation on carbon and nitrogen dynamics in southwest Oregon forests and clear cuts. Canadian Journal of Forest Research 22:298-305.

Bowles, T., Acosta-Martinez, V., Calderon, F., and Jackson, L., 2014. Soil enzyme activities, microbial communities, and carbon and nitrogen availability in organic agroecosystems across an intensively-managed agricultural landscape. Soil Biology and Biochemistry 68: 252-262.

Bouma, J. 1989. Using soil survey data for quantitative land evaluation. Advances in Soil Science 9: 177-213

Brown, T. J., P. J. Butler and F. C. Happold. 1965. Some properties of the rhodanese system of Thiobacillus denitrificans. Biochemical Journal 97: 651-657.

Bünemanna, E. K., G. Bongiornoa, Zhanguo Bai, R. E.Creamer, G. De Deyn, R. Goede, L. Fleskens, V. Geissen, T. W. Kuyper, P. Mäder, M. Pulleman, W. Sukkel, J. W. van Groenigen, L. Brussaard. 2018. Soil quality-a critical review. Soil Biology and Biochemistry 120: 105-125.

Burger, J.A. 1997. Conceptual framework for monitoring the impact of intensive forest management on sustainable forestry. In: Hakkila, P., Heino, M., Puranen, E. (Eds.), Proceedings of a Joint IEA Task XII Meeting on Forest Management for Bioenergy, Jyvaskyla, Finland. Finnish Forest Research Institute, Vaanta, Finland, pp. 147–156.

Burns, R. G. 1978. Enzymes in soil: Some theoretical and practical considerations. In Soil Enzymes (R. G. Burns, Ed.), pp. 295-339. Academic Press, London.

Burns, R.G. 1982. Enzyme activity in soil: Location and a possible role in microbial ecology. Soil Biology and Biochemistry 14:423-427.

Cannell, R.Q., K. Gales, R. W. Snaydon, B.A. Suhail. 1979. Effects of short-term waterlogging on the growth and yield of peas (Isum-Sativum). Annals of Applied Biology 93: 327.

Chen, H. 2003. Phosphatase activity and P fractions in soils of an 18-year-old Chinese fir (Cunninghamia lanceolata) plantation. Forest Ecology and Management 178: 301-310

Chew, M. Y. 1973. Rhodanese in higher plants. Phytochemistry 12: 2365-2367.

Chunderova, A.I. and T. Zubets. 1969. Phosphatase activity in dernopodzolic soils. Pochvovedeniye 11:47-53.

Ciha, A.J. 1984. Slope position and grain yield of soft white winter wheat. Agronomical Journal. Vol.76, pp.193– 196.

Clarholm, M. 1993. Microbial biomass P, labile P, and acid phosphatase activity in the humus layer of a spruce forest, after repeated additions of fertilizers. Biology and Fertility of Soils 16:287-292.

Corstanje, R., T.G. Mercer, J. R. Rickson, L. K. Deeks, P. Newell-Price, I. Holman, C. Kechavarsi, and T. W. Waine. 2017. Physical soil quality indicators for monitoring British soils. Solid Earth 8: 1003-1016.

Culman S.W., and others. 2011. Permanganate oxidizable carbon reflects a processed soil fraction that is sensitive to management. Soil Science Society of America Journal 76: 494-504.

Culman, S. W., S. S Snapp, J. M Green, L. E Gentry. 2013. Short- and Long-term labile soil carbon and nitrogen dynamics reflect management and predict corn agronomic performance. Agronomy Journal 105: 493-502.

Culman, S.W., S.T. DuPont, J.D. Glover, D.H. Buckley, G.W. Fick, H. Ferris, and T.E. Crews. 2010. Long-term impacts of high-input annual cropping and unfertilized perennial grass production on soil properties and belowground food webs in Kansas, USA. Agriculture, Ecosystems and Environment 137: 13–24.

Deng, S. 1990. Sulfur oxidation and rhodanese activity in soils. Soil Science, Vol. 150, pp. 552-560.

Dick, L.K., G. Jia, S. Deng, and R. P. Dick, 2013. Evaluation of microplate and bench-scale β-glucosidase assays for reproducibility, comparability, kinetics, and homogenization methods in two soils. Biol. Fert. Soils (in press).

Dick, R. P. 1994. Soil enzyme activities as indicators of soil quality. *In* Doran et al. (eds.) Defining Soil Quality for a Sustainable Environment. Soil Science Society of America Special Publication, Madison, WI, pp. 107-124.

Dick, R. P., D. Breakwill, and R. Turco. 1996. Soil enzyme activities and biodiversity measurements as integrating biological indicators. p. 247-272. In J.W. Doran and A.J. Jones. (eds.). Methods for Assessing Soil Quality. Soil Science Society of America Special Publication 49. Madison WI. pp. 247-272.

Dick, R. P., P. E. Rasmussen and E. A. Kerle. 1988b. Influence of long-term residue management on soil enzyme activities in relation to soil chemical properties in a wheat-fallow system. Biology and Fertility Soils 6:159-164.

Dick, R.P., D.D. Myrold, and E.A. Kerle. 1988a. Microbial biomass and soil enzyme activities in compacted and rehabilitated skid trail soils. Soil Science Society of America Journal 52:512-516.

Dick, W.A., and M.A. Tabatabai. 1992. Potential uses of soil enzymes. *In* F.B. Metting, Jr. (ed.) Soil microbial ecology: Applications in agricultural and environmental management. Marcel Dekker, Inc., New York, pp. 95-127.

Dick, P. R., L. K. Dick, S. Deng, Xiufen Li, E, Kandeler, C. Poll, C. Freeman, T. G. Jones, M. N. Weintraub, K. A. Esseili, and J. Saxena. 2018. Cross-laboratory comparison of fluorometric microplate and colorimetric bench-scale soil enzyme assays. Soil Biology and Biochemistry 121: 240-248.

Duiker, S., J. C. Myers and L. C. Blazure. 2016. Soil Health in Field and Forage Crop production. USDA Natural Resources Conservation Service, Penn State University Extension, Capital Resource Conservation & Development and Clinton County Conservation District, Harrisburg, PA, p. 27.

Doran, J.W., Parkin, T.B., 1994. Defining and Assessing Soil Quality. In: Doran, J.W., Coleman, D.C., Bezdicek, D.F., Stewart, B.A. (Eds.), Defining soil quality for a sustainable environment. Soil Science Society of America, Madison, pp. 3-21.

Fiedler, S., M. J. Vepraskas and J. L. Richardson. 2007. Soil redox potential: importance, field measurements, and observations. Advances in Agronomy 94: 1-54.

Franchini, J.C., C.C. Crispino, R.A. Souza, E. Torres, M. Hungria. 2007.Microbiological parameters as indicators of soil quality under various soil management and crop rotation systems in southern Brazil. Soil Tillage Research 92: 18–29.

Franzmeier, D.P. 1969. Properties of some soils in the Cumberland plateau as related to slope aspect and position. Soil Science Society of America, Proceedings, Vol. 33, pp. 755– 761.

Furthermore, they may include a time or rate dimension which makes them dynamic, These indicators are termed pedotransfer functions (Bouma, 1989) and are generally used to describe functions in which routinely-measured properties are used to predict other properties that are not measured (Kay and Grant, 1996).

Govaerts, B., K.D. Sayre, J. Deckers. 2006. A minimum data set for soil quality assessment of wheat and maize cropping in the highlands of Mexico. Soil and Tillage Research 87: 163-174.

Govaerts, B., K. D. Sayre, J. Deckers. 2005. A minimum data set for soil quality assessment of wheat and maize cropping in the highlands of Mexico. Soil and Tillage Research 87: 163-174.

Granatsein, D., and D.F. Bezdicek. 1992. The need for a soil quality index: Local and regional perspectives. American Journal of Alternative Agriculture 17:12-16.

Grandy, A.S., and G.P. Robertson. 2007. Land-use intensity effects on soil organic carbon accumulation rates and mechanisms. Ecosystems 10: 58– 73.

Gregorich, E.G., M.R. Carter, J.W. Doran, C.E. Pankhusrst, L.M. Dwyer. 1997. Biological attributes of soil quality. In: Gregorich, E.G., Carter, M.R. (Eds.), Soil quality for crop production and ecosystem health. Elsevier, Amsterdam, pp. 81-113.

Griffith, D. R., E. J. Kladivko, J. V. Mannering, T.D. West and S. D. Parsons. 1988. Long-term tillage and rotation effects on corn growth and yield on high and low organic matter, poorly drained soils. Agronomy Journal 80: 599-605.

Gunapala, N., K.M. Scow. 1998. Dynamics of soil microbial biomass and activity in conventional and organic farming systems. Soil Biology Biochemistry 30: 805–816.

Gupta, V.V.S.R., and J.J. Germida. 1988. Distribution of microbial biomass and its activity in different soil aggregate size classes as affected by cultivation. Soil Biology and Biochemistry 20:777-786.

Haberern, J. 1992. Viewpoint: A soil health index. J. Soil Water Conservation 47:6.

Haney, R.L., A.J. Franzluebbers, E.B.Porter, F.M. Hons, D.A. Zuberer. 2004. Soil carbon and nitrogen mineralization: influence of drying temperature. Soil Science Society of America Journal 68: 489-492.

Henderson, G.S., 1995. Soil organic matter: a link between forest management and productivity. In: McFee, W.W., Kelly, J.M. (Eds.), Proceedings of the 8th North American Forest Soils Conference on Carbon Forms and Functions in Forest Soils. Soil Science Society of America., Madison, WI, pp. 419–435.

Herrick, J.E. 2000. Soil quality: an indicator of sustainable land management? Applied Soil Ecology 15: 75-83.

Hinojosa M.B, J.A. Carreira, R. García-Ruíz, and R.P. Dick. 2004. Soil moisture pre-treatment effects on enzyme activities as indicators of heavy metal-contaminated and reclaimed soils. Soil Biology and Biochemistry 36-1559-1568. American Journal of Alternative Agriculture 7:17-24.

Hose, T. D., M. Cougnon, A. De Vliegher, B. Vandecasteele, N. Viaene, W. Cornelis, E. V. Bockstaele, D. Reheul. 2013. The positive relationship between soil quality and crop production: A case study on the effect of farm compost application. Applied Soil Ecology 75: 189-198.

Jenny, Hans. 1980. The soil resource: origin and behavior. Ecological studies 37. Springer-Verlag, New York.

Kay, B.D., C.D. Grant. 1996. Structural aspects of soil quality. In: MacEwan, R.J., Carter, M.R. (Eds.), Soil Quality is in the Hands of the Land Manager. Centre for Environmental Management, University of Ballarat, Victoria, Australia, pp. 37–41.

Knight T, 2002. Soil enzyme stability in disturbed ecosystems. MS thesis. Oregon State University, Corvallis OR.

Knight T, and R. P. Dick. 2004. Differentiating microbial and stabilized β-glucosidase activity relative to soil quality. Soil Biology and Biochemistry 36: 2089-2096.

Kravchenko, A.N. 2000. Correlation of corn and soybean grain yield with topography and soil properties. Agronomical Journal 92: 75– 83.

Kreznor, W.R. 1989. Soil, landscape, and erosion relationships in a northwest Illinois watershed. Soil Science Society of America Journal 53: 1763– 1771.

Kumar, S., A. Kadono, R. Lal and W. Dick. 2012. Long-term no-till impacts on organic carbon and properties of two contrasting soils and corn yields in Ohio. Soil Science Society America Journal 76: 1798–1809.

Lal, R., and G.S. Taylor. 1970. Drainage and nutrient effects in a field lysimeter study. 2. Mineral uptake by corn. Soil Science Society America: Proceedings 34: 245–248.

Laishram, J., K. G. Saxena, R. K. Maikhuri and K. S. Rao. 2012. Soil quality and soil health: A review. International Journal of Ecology and Environmental Sciences 38(1): 19–37.

Larson, W.E., and F.J. Pierce. 1991. Conservation and enhancement of soil quality. In Evaluation for sustainable land management in the developing world. Vol. 2. IBSRAM Proc. 12 (2). Bangkok, Thailand. Int. Board for Soil Research and Management.

Lettl, A. 1986. Thiosulfate sulfurtransferase (Rhodanese) in forest soils. Forestry and game management research institute.

Li, X., P. Sarah. 2003. Enzyme activities along a climatic transect in the Judean Desert.

Loginow, W., W. Wisniewski, S. Gonet, and B. Ciescinska. 1987. Fractionation of organic C based on susceptibility to oxidation. Polish Journal of Soil Science 20: 47–52.

Lopes, A.A.C., D.M.G. Sousa, F.B.Jr. Reis, I.C. Mendes. 2015. Air-drying and long-term storage effects on β-glucosidase, acid phosphatase and arylsulfatase activities in a tropical Savannah Oxisol. Applied Soil Ecology 93: 68–77.

Lutz Breuer, Ralf Kiese, and Klaus Butterbach Bahl, 2002. Temperature and moisture effects on nitrification rates in tropical rain-forest. Soil Science Society of America Journal 66:834-844.

Martens, D.A., J.B. Johanson, and W.T. Frankenberger, Jr. 1992. Production and persistence of soil enzymes with repeated additions of organic residues. Soil Science 153:53-61.

Martino, D.L., C.F. Shaykewich. 1994. Root penetration profiles of wheat and barley as affected by soil penetration resistance in-field conditions. Canadian Journal of Soil Science 74: 193–200.

Mathur, S.P. and A.F. Rayment. 1977. Influence of trace element fertilization on the decomposition rate and phosphatase activity of a mesic fibrisol. Canadian Journal of Soil Science 57: 397-408.

McCarty, G.W., D.R. Shogren, and J.M. Bremner. 1992. Regulation of urease production in soil by microbial assimilation of nitrogen. Biology and Fertility of Soils 12: 261-264.

McLaren A. D. 1975. Soil as a system of humus and clay immobilized enzymes. Chemica Scripta 8: 97-99.

Miller, M. and R.P. Dick. 1995a. Dynamics of soil C and microbial biomass on whole soil and aggregates in two cropping systems differing in C-input. Applied Soil Ecology 2:253-261.

Miller, M., and R. P. Dick. 1995b. Thermal stability and activities of soil enzyme activities as influenced by crop rotation Soil Biology and Biochemistry 27:1161-1166.

Moebius-Clune, B.N., D.J. Moebius-Clune, B.K. Gugino, O.J. Idowu, R.R. Schindelbeck, A.J. Ristow, H.M. van Es, J.E. Thies, H.A. Shayler, M.B. McBride, K.S.M Kurtz, D.W. Wolfe, and G.S. Abawi, 2016. Comprehensive Assessment of Soil Health – The Cornell Framework, Edition 3.2, Cornell University, Geneva, NY.

Moore, I.D. 1993.Soil attribute prediction using terrain analysis. Soil Science Society of America Journal, Vol. 57, pp. 443– 452.

Moreira, R. S., M. K. Chiba, S. B. Nunes and I. C. D. de Maria. 2017. Air-drying pretreatment effect on soil enzymatic activity. Plant Soil and Environment 63: 29-33.

Mulder, C., de Zwart, D., van Wijnen, H.J., Schouten, A.J., Breure, A.M., 2003. Observational and simulated evidence of ecological shifts within the soil nematode community of agroecosystems under conventional and organic farming. Functional Ecology 17: 516–525.

Nakajima, T., E. G. Mbonimpa, S. Gautam, U.R. Somireddy, A. Kadono, R. Lal, R. Chintala, R. Rafique and N. Fausey. 2013. Soil science and plant nutrition, Volume 60, pp. 108-118.

Nangia, V., P. H. Gowda, D. J. Mulla. 2010. Effects of changes in N-fertilizer management on water quality trends at the watershed scale. Agricultural Water Management 97: 1855–1860.

Nannipieri, P., B. Ceccanti, C. Conti and D. Bianchi. 1982. Hydrolases extracted from soil, their properties and activities. Soil Biology and Biochemistry 14: 257-263.

Nannipieri, P., P. Sequi and P. Fusi. 1996. Humus and enzyme activity. *In* A. Piccolo (ed.) Humic Substances in Terrestrial Ecosystems. Elsevier, New York, pp. 293-328.

NCR. 2011. Recommended Chemical Soil Test Procedures for the North Central Region. No. 499 (revised). North Central Regional Research Publication. North Dakota State University. pp. 5-32.

Ndiaye, E. L., J.M. Sandeno, D. McGrath, and R.P. Dick. 2000. Integrative biological indicators for detecting change in soil quality. American Journal of Alternative Agriculture 15:26-36.

Ndour Ndeye Yacine Badiane, J.L Chotte, E. Pate, D. Masse, and C. Rouland. 2001. Use of soil enzyme activities to monitor soil quality in natural and improved fallows in semi-arid tropical regions. Applied Soil Ecology, Vol. 18, Issue 3, pp. 229-238.

Nielsen, R. L. 2015. Estimating grain yield prior to harvest. Purdue University. URL: http://www.kingcorn.org/news/timeless/YldEstMethod.html.

Ponnamperuma, F. N. 1972. The chemistry of submerged soils. Advances in Agronomy 24: 29-96.

Power, J.F., R. Wiese, D. Flowerday. 2000. Managing nitrogen for water quality-lessons from Management Systems Evaluation Area. Journal of Environmental Quality 29: 355–366.

Powers, R.F., Tiarks, A.E., Boyle J.R., 1998. Assessing soil quality: practicable standards for sustainable forest productivity in the United States. In: Adams, M.B., Ramakrishna K., Davidson, E.A. (Eds.), The Contribution of Soil Science to the Development and Implementation of Criterial and Indicators of Sustainable Forest Management. Soil Science Society of America, Special Publication No. 53, pp. 53–80.

Ramesh, C. R., N. Behera and N. Sethunathan. 1984. Rhodanese activity of flooded and nonflooded soils. Soil Biology and Biochemistry, Vol. 17, No.2, pp. 159-162.

Reganold, J.P. 1988. Comparison of soil properties as influenced by organic and conventional farming systems. American Journal of Alternative Agriculture 3:144-155.

Reganold, J.P., A.S Palmer. 1995. Significance of gravimetric versus volumetric measurements of soil quality under biodynamic, conventional, and continuous grass management. Journal of Soil and Water Conservation 50: 298-305.

Ritz, K., H.I.J. Black, C.D. Campbell, J.A. Harris, C. Wood. 2009. Selecting biological indicators for monitoring soils: a framework for balancing scientific and technical opinion to assist policy development. Ecological Indicators 9: 1212-1221.

Romig, D.E., Garlynd, M.J., Harris, R.F., 1996. Farmer-based assessment of soil quality: a soil health scorecard. In: Doran, J.W., Jones A.J. (Eds.), Methods for Assessing Soil Quality. Soil Science Society of America, Special Publication No. 49, pp. 39–60.

Roper, W. R., D. L. Osmond, J. L. Heitman, M. G. Wagger and Schris Reberg-Horton. 2017. Soil health indicators do not differentiate among agronomic management systems in north Carolina soils. Soil Science Society, Vol. 81, Issue 4, pp. 828.

RMST. 1974. Handbook on Reference Methods for Soil Testing. Council on Soil Testing and Plant Analysis. pp. 60-65.

Sagan, C. 1992. To avert a common danger. Parade Magazine, March I, pp. 10-14.

Sanders, D.W. 1992. International activities in assessing and monitoring soil degradation. America.

Sarkar, J. M., L. Batisticand and J. Mayaudon. 1980. Les hydrolases du sol et leur association avec les hydrates decarbone. Soil Biology and Biochemistry 12: 325-328.

Schoenholtz, S. H., H. Van Miegroet, J. A. Burger. 2001. A review of chemical and physical properties as indicators of forest soil quality: challenges and opportunities. Forest Ecology and Management 138: 335-356.

Schutter, M., J. Sandeno, and R.P. Dick. 2001. Seasonal, soil type, and alternative management influences on microbial communities of vegetable cropping systems. Biology and Fertility of Soils 34: 397-410.

Shannon, D., A.M. Sen, D.B. Johnson. 2002. A comparative study of the microbiology of soils managed under organic and conventional regimes. Soil Use Management 18 : 274–283.

Simard, R.R., D.A. Angers, C. Lapierre. 1994. Soil organic matter quality as influenced by tillage, lime, and phosphorus. Biology and Fertility of Soils 18: 13–18.

Singh, B. B. and M. A. Tabatabai. 1977. Factors affecting rhodanese activity in soils. Soil Science, Vol. 125, pp. 337-342.

Singh, B.B., Tabatabai, M.A. 1976. Rhodanese activity of softs. Soil Science Society of America Journal 40: 381-385.

Six, J., C. Feller, K. Denef, S. M. Ogle, J. C. D. Sa and A. Albrecht. 2002. Soil organic matter, biota and aggregation in temperate and tropical soils - Effects of no-tillage. Agronomy 22: 755–775.

Skujins, J. 1976. Extracellular Enzymes in Soil. CRC Critical Reviews in Microbiology 4:383-421.

Skujins, J. 1976. Extracellular Enzymes in Soil. CRC Critical Reviews in Microbiol. 4:383-421.

Smit, E., P. Leeflang, S. Gommans, J.V.D. Broek, S.V. Mil and K. Wernars. 2001. Diversity and seasonal fluctuation of the dominant members of the bacterial soil community in a wheat field as determined by cultivation and molecular methods. Applied and Environmental Microbiology, vol. 67, no. 5, 2284-2291.

Smith JL and JW Doran. 1996. Measurement and use of pH and electrical conductivity for soil quality analysis. In Methods for assessing soil quality. Soil Science Society of America Special Publication 49: 169-182.

Smith, A. J. and J. Lascelles. 1966. Thiosulphate metabolism and rhodanese in Chromatium sp. strains. Journal of General Microbiology 42: 357-370.

Sparling, G.P., T.W. Speir, K.N. Whale**.** 1986. Changes in microbial biomass C, ATP content, soil phosphomonoesterase and phosphodiesterase activity following air-drying of soil. Soil Biology and Biochemistry 18: 363-370.

Speir, T. W., and D. J. Ross. 1981. A comparison of the effects of air-drying and acetone dehydration on soil enzyme activities. Soil Biology and Biochemistry 13: 225-229.

Spiers, G.A. and W.B. McGill. 1979. Effects of phosphorus addition and energy supply on acid phosphatase production and activity in soils. Soil Biology and Biochemistry 11: 3-8.

Stearns, R. H. 1953. Respiration, rhodanese and growth in Escherichia coli. Journal of Cellular and Comparative Physiology 41: 163-l 70.

Stone, J.R. 1985. Effect of erosion and landscape position on the productivity of piedmont soils. Soil Science Society of America Journal, Vol. 49, pp. 987– 991.

Tabatabai, M. A. and B. B. Singh. 1976. Rhodanese activity of soils. Soil Science Society of America Journal 40: 381-385.

Tautges, N. E., T. S. Sullivan, C. L. Reardon and I. C. Burke. 2016. Soil microbial diversity and activity linked to crop yield and quality in a dryland organic wheat production system. Applied Soil Ecology 108: 258-268.

Taylor, J.P., M. Wilson, S. Mills, R.G. Burns. 2002. Comparison of microbial numbers and enzymatic activities in surface soils and subsoils using various techniques. Soil Biol. Biochemistry 34: 387–401.

USDA. 1993. Soil Survey Manual. http://websoilsurvey.nrcs.usda.gov/. Soil Survey Division Staff.

USDA.2017. National Agricultural Statistics Service. https://www.nass.usda.gov/Statistics_by_State/. United States Department of Agriculture.

Vallejo, V.E., Z. Arbelia, Wi. Terán, N. Lorenz, R. P. Dick, and F. Roldan. 2012. Effect of land management and Prosopis juliflora (Sw.) DC trees on soil microbial community and enzymatic activities in intensive silvopastoral systems of Colombia. Agriculture, Ecosystems and Environment 150 :139– 14.

Velasquez, E., P. Lavelle and M. Andrade. 2007. GISQ, a multifunctional indicator of soil quality. Soil Biology and Biochemistry 39(12): 3066–80.

Van Diepeningen, A.D., de Vos, O.J., Korthals, G.W., van Bruggen, A.H., 2006. Effects of organic versus conventional management on chemical and biological parameters in agricultural soils. Applied Soil Ecology 31: 120–135.

Van Groeningen, J.W., I.M. Lubbers, H.M.J.Vos, G.G. Brown, G.B. De Deyn, K.J. Van Groenigen. 2014. Earthworms increase plant production: a meta-analysis. Nature, 4.

Wander, M. 2004. Soil organic matter fractions and their relevance to soil function. p. 67–102. *In* Magdoff, F., and R.R. Weil (ed.) Soil organic matter in sustainable agriculture. CRC Press, Boca Raton, FL.

Wander, M.M., and M.G. Bidart. 2000. Tillage practice influences on the physical protection, bioavailability and composition of particulate organic matter. Biology and Fertility of Soils 32: 360–367.

Wander, M.M., G.A. Bollero. 1999. Soil quality assessment of tillage impacts on Illinois. Soil Science Society of America Journal 63: 961–971.

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.

World Resources Institute. 1992. New York Times, March 3.

Wright, R. J, D. G. Boyer, W. M. Winant, H. D. Perry. 1990. The influence of soil factors on yield differences among landscape positions in an Appalachian cornfield. Soil Science, Vol. 149, Issue 6.

Wright, R.J. 1990. The influence of soil factors on yield differences among landscape positions in an Appalachian cornfield. Soil Science 149: 375– 382.

Yang, C. 1998. Spatial variability of field topography and wheat yield in the Palouse region of the Pacific Northwest. Transactions of ASAE, Vol. 41, pp. 17– 27.

Yeates, G.W., R.D. Bardgett, R. Cook, P.J. Hobbs, P.J. Bowling and J.F. Potter. 1997. Faunal and microbial diversity in three Welsh grassland soils under conventional and organic management regimes. Journal of Applied Ecology 34: 453–470.

Zornaza, R., C. Guerrero, J. Mataix-Solera, V. Arcenegui, F. García-Orenes, J. Mataix-Beneyto. 2006. Assessment air-drying and rewetting pretreatment effect on some soil enzyme activities under Mediterranean conditions. Soil Biology and Biochemistry 38: 2125-2134.

Zornoza R., C. Guerrero, J. Mataix-Solera, V. Arcenegui, F. García- Orenes, J. Mataix-Beneyto. 2006: Assessing air-drying and rewetting pre-treatment effect on some enzyme activities under Mediterranean conditions. Soil Biology and Biochemistry 38: 2125–2134.