SOIL HEALTH ON A SMALL-SCALE SUSTAINABLE VEGETABLE FARM IN SOUTH LOUISIANA

A Thesis

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Allison Hawkins Guidroz
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Table of Contents

List of Tables

List of Figures

Abstract

One of the biggest challenges facing the world today is the need to provide nutritious food to an ever-growing population in a way that does not compromise the ecosystem services of the soil that are necessary for life. The evidence for widespread degradation of the soil has been a major contributor in the increased interest in soil health. Most of the research in soil health has focused on field crops, which has led to a gap in the research of soil health on vegetable cropping systems. This objective of this study was to analyze the health of the soil on an intensive sustainable vegetable farm by measuring the chemical, physical and biological properties of the soil. Soil chemical assessments included macronutrients, micronutrients, pH, and soil organic matter (SOM). Soil physical assessments included particle size analysis and aggregate stability. Soil biological assessments included Fatty Acid Methyl Ester (FAME) and respiration. Key indicators of soil health were high in this system as compared to what other studies have reported in agricultural systems. These indicators included SOM, aggregate stability and a diverse microbial community as shown by FAME. An area to monitor in this system is the high nutrient levels, especially phosphorous, but the high aggregate stability, with an average of 93% in the areas in production at the end of the study, makes the possibility of runoff less of a concern. This study indicates that the management in this production system leads to positive impacts on the health of the soil.

CHAPTER 1. LITERATURE REVIEW

1.1. Introduction

Soil represents the difference between survival and extinction for most land-based life (Doran et al., 1996). It forms a thin layer that covers the earth's surface and performs many functions that are essential for life: providing a substrate to support plant growth, a reservoir for nutrients, influencing air and water quality, and providing a site for biological processes that are responsible for the decomposition and recycling of plant and animal products (Moebius-Clune et al., 2016). Soils influence air quality through interactions with the atmosphere and influences water quality by purifying water as it passes through the soil profile and acts as a site of water storage (Maikhuri and Rao, 2012). Soil is critically important to humankind as is documented by ancient civilizations that have collapsed or relocated because mismanagement destroyed the soils on which they depended (Montgomery, 2012; Wienhold et al., 2004).

One of the biggest challenges humanity faces today is the need to provide nutritious food to an ever-growing population. The challenge is twofold because not only must food production expand, but it must do so in a way that does not compromise the ecosystem services that are necessary for life on earth (Norris and Congreves, 2018). There is widespread degradation of soils around the globe, as shown through adverse changes in its physical, chemical, and biological properties, and contamination by inorganic and organic chemicals (Arshad and Martin, 2002; Oldeman et al., 2017). Degradation of soil contributes to erosion, loss of organic matter, soil contamination, compaction, increased salinity, and the release of organic carbon as carbon dioxide to the atmosphere (Houghton et al., 1983; Jones et al., 2012). In the last 40 years, 30 percent of the world's arable land has become unproductive and it has been estimated that the

rate of degradation caused by mismanagement is 10 million ha (about 25 million acres) per year (Pimentel, 2006). Given that it takes hundreds of years to form an inch of soil, this rate of erosion is a serious cause of concern (Wakatuski and Rasyidin, 1992).

Just as the degradation of the soil is well documented, the benefits of having a healthy soil has been widely studied and documented as well (Bennett et al., 2010; Wall et al., 2015). Healthy soil is linked with increased yields, decreased inputs, and the ability of the soil to withstand biotic and abiotic stresses (Brussaard et al., 2007). As greater production demands are placed on soils, farmers must learn to grow with sustainable soil management in mind. A major challenge within sustainable soil management is to conserve the function of the ecosystem, while optimizing agricultural yields (Kibblewhite et al., 2008). The quality and health of soils determine agricultural sustainability and environmental quality, which jointly determines plant, animal, and, ultimately, human health (Doran, 2002; Haberern, 1992).

Warkentin and Fletcher (1977) introduced the soil quality concept to complement soil science research, to make the understanding of soils more complete, and to guide the allocation of labor, energy, and other inputs as agriculture intensified and expanded to meet increasing world production demands. Concern about the decline in soil quality has increased through the years and to reverse the trend, there has been a call for developing definitions and indexes to measure the health of soil (Rodale Institute, 1991). The indexes provide an assessment tool for evaluating current management practices and a way to compare alternative management practices (Wienhold et al., 2004).

Soil Quality and Health Defined

Soil health and soil quality are terms which are widely used in sustainable agriculture to describe the general condition or quality of the soil resource (Kibblewhite et al., 2008). Terms that are often used interchangeably, soil health and quality are defined as "the capacity of soil to function as a vital living system, within ecosystem and land-use boundaries, to sustain plant and animal productivity, maintain or enhance water and air quality, and promote plant and animal health" (Doran and Zeiss, 2000). While often used synonymously, Moebius-Clune el al. (2016) suggest a distinction: soil quality referred to a soil's natural composition and properties (such as soil type), which is influenced by long-term process of soil formation and, generally, not affected by human management while soil health referred to soil properties that change as a result of soil use and management over the human time scale. Using the term soil health invokes the idea that the soil is an ecosystem that is full of life, which must be managed with care for optimal function (Moebius-Clune et al., 2016).

Managing soil in a way that enhances its health has many economic benefits to the producer as well. Healthy soils tend to have better plant yield and quality. There is reduced crop loss in times of environmental stresses like heavy rain, drought, and pest or disease outbreak. Fuel cost is reduced by less tillage and when needed, producers are able to get in the field sooner during wet periods. There are better profit margins with decreased losses and increased efficiency of fertilizer, pesticide, herbicide and irrigation applications (Moebius-Clune et al., 2016).

1.2. Integrated Concept of Soil Health

1.2.1 Soil as a System

Soil is a very complex system in which each component of the soil interacts and is affected by the other components as well as outside forces, both natural and imposed. Soil is a dynamic living system and, as such, is distinguished from weathered rock mainly by its biology which interacts with the abiotic physical and chemical components of the soil (Karlen et al., 1997; Kibblewhite et al., 2008). Soil health is enhanced by management decisions that consider the multiple functions of soil and is impaired by decisions that focus only on single functions, such as crop productivity. For optimal soil health, a balance between soil function for productivity, environmental quality, and plant and animal health is necessary (Doran, 2002).

1.2.2. Functions of the Soil

The concept of soil health is centered on the ability of the soil to perform specific functions like sustaining biological activity, regulating water flow, and buffering, storing, and cycling of nutrients (Karlen et al., 1997). Humankind relies on natural and managed ecosystems for many 'environmental good and services' (Costanza et al., 1997; Dally and Power, 1997; De Groot et al., 2002). Many of these functions are mediated by biological processes. These include processes necessary for food and fiber production like nutrient cycling, natural biological control of pests and diseases, and influencing water and air quality (Wall, 2004).

Soil scientists propose managing soils in a way that protects their ecosystem functions (Doran et al., 1996; Karlen et al., 1997; Kirchmann and Andersson, 2001). This view of protecting soil functions has been implemented at a policy level. As an example, in the EU the

Thematic Strategy for Soil Protection states, “Soil is essentially a non-renewable resource and a very dynamic system which performs many functions and delivers services vital to human activities and ecosystems survival” (European Commission, 2006).

Kibblewhite et al. (2008) propose four categories of ecosystem functions, which collectively provide the basis for all the major services provided by the soil: transformation of carbon, cycling of nutrients, soil structural stability, and biological regulations of soil populations.

- Transformation of carbon through the decomposition of plant residues and other organic materials with the activities of the soil biota, soil organic matter is produced, which has known benefits, namely, contributing to soil structure maintenance, increased cation exchange capacity, and increasing water storage (Moebius-Clune et al., 2016). Decomposition in itself is not only an essential ecosystem function and driver of nutrient cycles but also supports a detoxification and waste disposal service. Sequestration of carbon in soil also plays a role in regulating the emission of greenhouse gases such as methane and carbon dioxide (Kibblewhite et al., 2008).
- Cycling of nutrients in the soil for storage and supply to meet the requirement for plant growth and the living organisms in the soil (Bastida et al., 2006; Kelting et al., 1999).
- Soil structural stability by aggregation and formation of biostructures and pore networks across many spatial scales (Kibblewhite et al., 2008). The pore networks provide the habitat for microorganisms in the soil as well as spaces for air and water transport (Moebius-Clune et al., 2016). It also provides favorable rooting medium for plants and the aggregation helps resists erosion (Harris et al., 1996).
- Biological regulations of soil populations including organisms recognized as pests and diseases of agriculturally important plants and animals as well as humans (Kibblewhite et al.,

2008). This ability of soils to suppress plant diseases has been studied for many decades (Janvier et al., 2007).

1.3. Factors Controlling Soil Health

Soil is a dynamic ecosystem in which each part interacts and influences the others. Management that takes all components of this ecosystem into account has better results in maintaining and increasing the health of the soil. The main components of the soil are physical, chemical and biological in nature.

1.3.1. Soil Type (Physical)

Soil is formed through the interaction of five major factors: parent material, time, climate, topography and organisms present. This is what forms the mineral portion of the soil and affects texture and nutrient holding capacity of soils. While these factors largely determine the type of soil that develops, land management by humans can alter natural soils considerably. Mismanagement can cause a loss of surface horizons due to erosion, salinization due to poor irrigation practices, loss of natural soil organic matter caused by arable production, and compaction with frequent tillage (Doran, 2002; Pimentel et al., 1995). Thus, land-use and management are key factors impacting soil health. The fixed and variable abiotic factors interact with biotic ones to determine the overall condition of the soil system and its associated health (Kibblewhite et al., 2008).

1.3.2. Nutrients and pH (Chemical)

In natural ecosystems, plant requirements for nutrients such as nitrogen (N), sulfur (S), phosphorus (P), and trace elements are met wholly or in part by mineralization of organic matter (Allison, 1973). To maintain production where yields are N limited or where crop removal

occurs, any depletion of the N reserve is traditionally amended with fertilizer (both chemical and naturally occurring waste). Not only does the soil need an adequate supply of the nutrients, the pH of the soil must be at the appropriate level such that the nutrients are in a form available for plant uptake. Manipulation of nutrient supplies to increase productive outputs from the soil system by the addition of fertilizers has been one of the keystones of agriculture for centuries. Nonetheless, knowledge is limited about the impacts of nutrient additions on the condition of different groups of soil organisms and, in turn, on their function (Kibblewhite et al., 2008).

It is imperative in agriculture systems that nutrients are available in adequate proportions, but not in over abundance. Additions of nutrients beyond that which can be used by the soil-plant system lead to their damaging leakage from the soil system into other environmental compartments via leaching, runoff, and gaseous emissions (Doran, 2002). In this case, the soil system is polluted and considered unhealthy.

1.3.3. Organisms and functions (Biology)

While soil science is a mature field of study, especially when assessing the chemical and physical aspects of the soil, the importance of understanding and managing the soil's biological properties has moved beyond a few select scientists and innovative practitioners, to become a focus in broader circles in recent years (Moebius-Clune et al., 2016). As technologies have improved to measure microbial activity and abundance, the living nature of soil and the important role it plays in the soil has been reinforced. It has been estimated that the number of organisms in one teaspoon of soil can exceed 9 billion – almost 1.5 times that of the global population (Doran et al., 1999). Below ground biodiversity represents one of the largest reservoirs of biological diversity on Earth, with diverse life forms and functions that are involved

in many ecosystem processes, which provide key ecosystem services for humans (Decaëns, 2010). The microbial population in the soil is also of major importance as they play a key role in nutrient cycling, increasing nutrient availability, and water and nutrient use efficiency. Microbial activity is fundamentally governed by the availability of fixed carbon (the major 'currency' of the soil system), which is amenable to manipulation via agronomic factors such as vegetation type, and residue and other organic waste management (Kibblewhite et al., 2008).

1.4. Assessing Soil Health

1.4.1. Principles

Research indicates how critically important the soil is in the production of food and fiber, and in ecosystems function and the maintenance of local, regional, and global environments. As the evidence has increased, there has been an increased interest in evaluating the health of soil (Glanz, 1995). There is complex debate about the appropriate methods for soil assessment given the multicomponent nature of soil systems, the breadth of goods, services and functions that they are called upon to provide, and their spatial variability. No single indicator encompasses all aspects of soil health, nor would it be feasible to measure all possible indicators (Kibblewhite et al., 2008). Doran (2002) proposed the criteria for indicators of soil health relate mainly to their utility in defining ecosystem processes and integrating physical, chemical, and biological properties; their sensitivity to management and climatic variations; and their accessibility and utility to agricultural specialists, producers, conservationists, and policy makers.

There has been increasing interest internationally in developing soil health assessments for the protection of soil. For example, the European Commission implemented the Thematic Strategy for Soil Protection in Europe (European Commission, 2012), and identified erosion,

declining organic matter, contamination, compaction, salinization, loss in biodiversity, soil sealing, landslides, and flooding as the key threats to soil. The European Commission also has a report that sets out common principles for protecting soils across the European Union (European Commission, 2006). In the United States, in response to the degradation of soils, reduction of soil health, declines in crop productivity, and lower farm profits, growers, extension educators, researchers, and private consultants have been working to develop cost-effective protocols for assessing the health of soils. In 2006, the first version of Cornell Assessment of Soil Health was made publicly available (Moebius-Clune et al., 2016).

Most recently, United States Department of Agriculture (USDA) released a set of standard indicators and associated laboratory procedures to assess soil health. The National Resources Conservation Service (NRCS) and the Agricultural Research Service (ARS) led a diverse coalition of technical experts in selecting methods to assess six standard soil health indicators, which focus on key physical and biological processes in soils. Those indicators are organic matter recycling and carbon sequestration, soil structure stability, general microbial activity, carbon food source, bioavailable N, and microbial community diversity. The associated laboratory methods for assessing each indicator were chosen based on interpretability, ease of use, cost effectiveness, measurement repeatability, and ability to inform agricultural management decisions. Standardization of methods and protocols will allow for high quality data, which can be used to develop regional standards and algorithms that account for numerous soil and environmental factors (Stott & Moebius-Clune, 2018). While several indicators of soil health have been proposed, currently there is not a globally acceptable methodology of assessing soil quality (Laishram et al., 2012).

1.4.2. Measurement

Effectively measuring soil health has many challenges. Soil systems are open systems with great variability in soils spatially and temporally and their performance is variable and interactive with environmental factors, such as air temperature and precipitation, which are not easily controlled. Soil system performance does not respond immediately to altered conditions, and it is best if its assessment is made over significant time periods (Kibblewhite et al., 2008). While there are differences in options about what measurement methods are best to use, it is generally agreed that the measurements need to include physical, chemical and biological indicators with the goal of having as complete a picture of the soil as possible.

Physical measurements that are used in assessing soil health include aggregation, which measures soil structural stability, particle size analysis, which gives a general idea about some aspects of the soil (including permeability, cation exchange capacity, and pore space) and the habitat it provides. Bulk density and penetration resistance are physical measurements also used to assess the health of the soil. Chemical measurements include pH and levels of macro and micronutrients, which is what soil science has often focused on, as well as levels and pools of carbon, which gives an indication about how high microbial population the soil could support. The biological measurements have been made possible in recent years as technology has advanced. Biological measurements include microbial respiration, which measures general microbial activity, and fatty acid methyl ester (FAME) profiles, which gives information about the microbial community structure.

Soil organic matter is often chosen as the most important indicator of soil health because of its impact on other physical, chemical, and biological indicators of soil health (Reeves, 1997).

Stott and Moebius-Clune (2018) discussed some of the many beneficial properties of soil organic matter to the health of the soil:

- Improves physical structure by improved aggregation
- Slow release of plant nutrients, especially N
- Aids in trace element nutrition through chelation reactions
- Has high cation exchange capacity, increasing adsorption of cations
- Supports soil biological population, which promotes biological control of pests and pathogens
- Reduces and buffers toxins, both natural and anthropogenic
- Increase water holding capacity

Arshard and Martin (2002), note the interrelationship of soil indicators and how they are affected by other soil qualities:

Selected indicator	Other soil quality indicators affecting the selected indicator
Aggregation	Organic matter, microbial (especially fungal) activity, texture
Microbial Biomass	Organic matter, aggregation, bulk density, pH, texture, respiration
Available nutrients	Organic matter, pH, topsoil-depth, texture, microbial parameters (rates of mineralization and immobilization)

There is another question altogether about once the measurements are collected, 'How is one to know what levels are indicative of soil health?'. Kibblewhite et al. (2008) propose comparing levels to the 'agricultural equilibrium', which is what agroecosystems reach following the conversion from natural vegetation.

1.5. Impacts of Agriculture Practices on Soil Health

There are many anthropogenic factors that are meant to maximize yield, including artificial inputs such as chemical fertilizers, pesticides, and tillage to prepare a fine seed bed, that can negatively affect soil health (Edwards, 1993; Rhodes, 2012). Kibblewhite et al. (2008) noted that these practices and inputs supplement or even 'substitute' for biological functions that are seen as inadequate or insufficient for achieving required levels of production, which distorts the natural balance of the ecosystem and may compromise the output of other environmental services. Degradation of soils sometimes occurs rapidly and obviously, for example when poor soil management leads to gully erosion. Often degradation is slower and more subtle, however, and may only impact agricultural production and the wider environment over years. Examples include nutrient leakage, which can lead to degradation of the water supply, surface erosion, and loss of soil biodiversity (Kibblewhite et al., 2008).

Tillage also alters the microbial community present. Wardle (1995) reviewed and analyzed hundreds of papers and was able to draw several generalizations in respect to tillage and biological communities. The most striking effect was the relationship between tillage and the size of the organisms present. Increased tillage favors bacteria and less fungi present in the soil, as the physical tilling process injures the fungi. Studies showed biodiversity is lessened in agricultural systems as compared to natural systems (Bardgett and Van Der Putten, 2014).

Some production practices including tillage, application of chemical fertilizers, and changes in land use not only affect soil health, but also have been reported to be important contributors for elevating carbon dioxide content in the atmosphere (Lal, 2002; Swanepoel, 2016). Turner (2001) attributes 50 percent of carbon dioxide in the atmosphere to agricultural activities. Although, there are other sources that attribute 20 percent carbon dioxide to agricultural activities (Almaraz et al., 2009; Ball et al., 1999; Gesch et al., 2007)

Soil health in vegetable cropping systems is seriously threatened by intensive tillage and fertilization practices and by limited crop rotations. Inclusion of cover crops, compost application, and reduced tillage may help sustain soil quality (Edward et al., 1992; Willekens et al., 2014). Norris and Congreves (2018) concluded that alternative management practices often found in organic or conservation agriculture generally improve physical, chemical, and biological aspects of soil health without negatively affecting vegetable crop yields as compared to conventional management. Studies show that minimal tillage produced lower carbon dioxide emissions as compared to conventional tillage (Almaraz et al., 2009; Ball et al., 1999; Gesch et al., 2007). Willekens et al. (2014) found that combining reduced tillage and recurrent compost application resulted in a different soil microbial community structure and the total microbial biomass was 44% higher under reduced tillage as compared to conventional tillage and fungal biomass doubled in the surface layer by reduced tillage.

Osborne et al. (2010) stated that soil should be managed in such a way that it acts as a carbon sink and not a source of carbon emission. Reduced tillage and compost application has also been found to increase the amount of organic carbon content (Willekens et al., 2014). Recalcitrant carbon content is the main source of soil carbon sink (Campbell et al., 1967). It has been found that carbon sequestration could be an effective way to reduce atmospheric carbon dioxide, which

is the most important greenhouse gas (Khorramdel et al., 2013). Cheng et al. (2007) concluded from their study that soil carbon pools are the most promising targets for carbon sequestration, especially in terrestrial environments. Khorramdel et al. (2013) found that soil with low disturbances and absence of chemical fertilizers, especially N, resulted in improved levels of soil organic carbon and soil organic matter. Other long-term studies have similar findings. Edwards et al., (1992) found higher SOC rate with lower use of chemical fertilizers, especially N.

Intensive vegetable cropping systems

This study was conducted on a farm that was based on the agricultural models written about by Coleman (2018) and Fortier (2014). These model farms focused on small-scale, high-value, sustainable vegetable production using equipment scaled to size. To some, this size is not just considered small-scale, but a micro-scale, with 0.6 – 0.8 hectare of vegetables in production. Although this is a small area in production, there is a focus in high-value crops, intensively planted, with beds planted several times in a year. Farms with this model can gross $148,263 - $247,105 per hectare with a 40% profit margin (Martin, 2014). Farms using this model are able to gross and obtain these margins with a wide diversity of high value vegetable crops, sold to direct markets, with minimum production costs and a focus on quality.

This production style also focuses on sustainability. This is not a new technique and was studied extensively, especially in Europe in the 19th century. Often referred to as “biological farming”, there are three main areas early biological farmers focused on: long-lasting soil fertility through soil organic matter (Elliot, 1898; Hopkins, 1910; Poore, 1893), optimized nutritional value of food crops (Auchter, 1939), and minimized pest problems (Fessenden, 1828;

Howard, 1940). The goal of this type of farming is to maximum crop yields from minimum land area while seeking to preserve or improve the quality of the soil.

Norris and Congreves (2018) indicate in their review article, that previous agricultural research in soil health mainly focused on field cropping systems and largely ignored vegetable cropping systems. This has led to a conspicuous research gap, which must be addressed to progress toward sustainable food production. There is also a significate gap in soil health assessments in sustainable, high value intensive vegetable farming in the south and this is the only one, to our knowledge that has been conducted in Louisiana. This study was conducted to contribute to the data in assessments in the health of the soil in sustainable intensive vegetable production.

CHAPTER 2. MATERIALS AND METHODS

2.1. Site Description

2.1.1. Site Location

The site for this study was a small-scale, sustainable vegetable farm focused on growing high-value crops in Baton Rouge, Louisiana (30.340243, -91.133926). The soils were a Cancienne silt loam (Soil Survey Staff, 2019). The Taxonomic class was**:** Fine-silty, mixed, superactive, nonacid, hyperthermic Fluvaquentic Epiaquepts. The Cancienne series consists of very deep, level to gently undulating, somewhat poorly drained mineral soils that are moderately slowly permeable. These soils formed in loamy and clayey alluvium. They are on high and intermediate positions on natural levees and deltaic fans of the Mississippi River and its distributaries. Slopes range from zero to three percent.

Average annual precipitation for the area from 2017-2019 was 169 cm (Figure 2.1.). The highest rainfall during the testing period occurred in April 2019 (25.15 cm), and the least amount during the testing period occurred in March 2019 (2.97). Average high temperature was 26 °C and the average low was 14.3 °C. Weather data were reported by the Baton Rouge Ryan Airport (30.5372, -91.1469) weather station, which is 24 km from the study site.

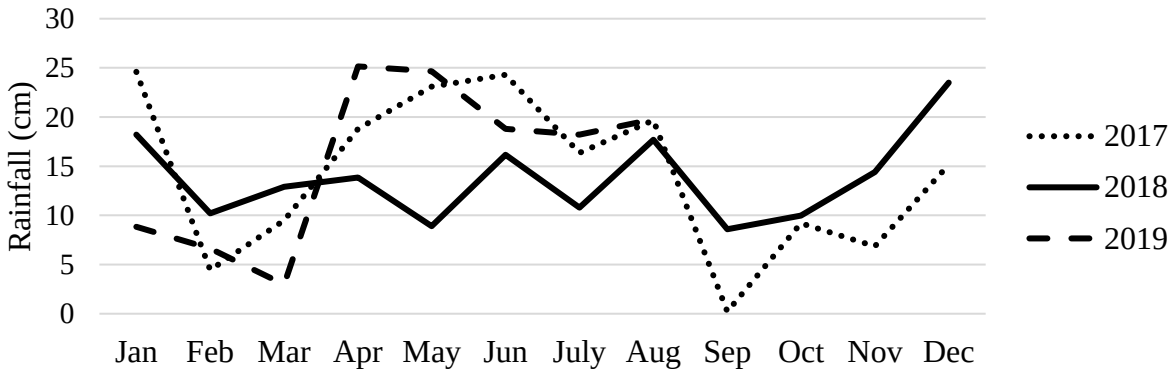

Figure 2.1. Monthly precipitation from 2017-2019 recorded at the Baton Rouge Metropolitan Airport.

2.1.2 Site Management

The site was a year-round small-scale, intensively managed, high-value vegetable farm selling to direct markets. Over 100 cultivars were grown throughout the year on the 0.6 ha farm. Crops grown included: Solanaceae (eggplants, peppers, tomatoes, and potatoes), Asteraceae (lettuce and dandelion), Brassicaceae (broccoli, cabbage, turnips, radishes, arugula, and kale), Chenopodiaceae (Swiss chard, spinach and beets), Cucurbitaceae (squash and cucumbers), Umbelliferae (dill, carrots, cilantro, fennel, and parsley), and Liliaceae (garlic, green onions and leeks). Crops were rotated frequently, and beds were often interplanted with more than one crop (radishes with greens, solanaceous crops with lettuce, and cole crops with radish).

2.1.2.1. Preparation of Site

To prepare the site for growing crops, native vegetation was cleared, the land was leveled and drainage was established with ditches and swales. A method called occultation was used to prepare soil with minimum tillage. In occultation, an opaque tarp was placed on the soil and weighed down with sandbags or rocks, such that light was not allowed to penetrate. This resulted in the death of the plants and the residues being mostly consumed by worms, fungi, and other organisms involved in decomposition. This not only reduced the labor necessary to prepare the soil, it decreased weed pressure and protected the soil from wind and water erosion. The tarps were left on the soil for two months and then beds were formed. The beds were 1.2 m apart center to center, 15.2 m long, with 0.76 m bed top and 0.45 m aisles. The beds were in a permanent location, which means the beds were not reformed seasonally or with crop changes, a technique used to reduce tillage. The growing areas of the beds were never walked on, to reduce compaction. The farm had standard size “zones”, which were made up of 25 beds for a total area of 15.2 m x 30.5 m in each zone.

2.1.2.2. Equipment

The equipment used in the formation and preparation of the garden beds was a two-wheeled walk-behind tractor (Grillo G108) with power take off (PTO) to allow operation of several implements (rotary plow, flail mower, and tiller). Before each crop planting, the previous crop was removed by physically pulling it up while leaving most roots behind. Fertilizer was then applied, the soil was lightly tilled (2.5 cm deep) to incorporate in the fertilizer, and compost was applied to the bed top and the next crop was planted. To maximize space and in effort to keep the soil covered, crops are often removed and replanted all in the same day. If deep tillage was necessary (for deep root penetration and increased water absorption) a tool called a broadfork was used. Typically, beds were broadforked annually in a manual operation. The broadfork was able to deeply aerate while preserving the soil structure and minimizing weed seeds coming to the surface. The broadfork has curved tines that were 45.6 cm long and 10 cm apart. For maintenance of weeds, light hand tools were used for shallow cultivation as well as extensive use of landscape fabric, made from 141 gram woven polypropylene fabric (DeWitt Company), which allowed air and water penetration while greatly reducing weed pressure.

2.1.2.3. Amendments

The site used only naturally derived amendments and organic pesticides and did not apply any herbicides in compliance with USDA National Organic Program (NOP) standards, although the farm is not certified by the USDA NOP. The site applied carbonaceous compost, sourced from a local company (Organic Products, LLC.), located 12.5 km from the farm. The feedstock was municipal yard-waste. The finished compost was passed through a 1.3 cm sieve. An average of 150 m^3 ha^{-1} of compost was applied annually on the site.

Initially, a pre-plant fertilizer (Scotts (11-2-2)) derived from hydrolyzed feather, meat, bone, blood meal and sulfate of potash was applied at a rate of 246 kg ha^{-1}. In addition, a supplemental fertilizer (Azomite (0-0-0.2)), which contained 70 metabolically active minerals and trace elements were applied at a rate of 23 kg ha^{-1}. Elemental sulfur (S) was applied to lower the pH at a rate of 246 kg ha^{-1}. Beds were planted an average of four times per year, so there was an annual application of 984 kg ha^{-1} (11-2-2), 92 kg ha^{-1} Azomite (0-0-0.2), and 984 kg ha^{-1} elemental S.

After soil was sampled and results were returned from January 2018, the amendment regimen was altered. For leafy greens and non-fruiting crops, only a preplant application of 246 kg ha^{-1} 13-0-0 (Nature Safe) was applied. The 13-0-0 was derived from feather meal, meat meal, and blood meal. For fruiting crops, a preplant application of 246 kg ha^{-1} 13-0-0 (Nature Safe) with an additional preplant fertilizer application of 123 kg ha^{-1} 8-5-5 (Nature Safe) was used. The 8-5-5 was derived from meat and bone meal, feather meal, blood meal and sulfate of potash. Another change to the amendment regimen after the first sampling was that applications of elemental S ceased.

2.2. Experimental Design

Soil samples were collected four times (January 2018, August 2018, January 2019, and August 2019) from four distinct management areas that represented different treatment durations (Figure 2.2): Old Fields (3 replications), New Fields A (2 replications), New Fields B (2 replications), and Control (2 replications). Each replication had a minimum of five rows (6.1 m x 15.3 m). The Old Fields had been in intensive high-value vegetable production since the farm started in the Summer of 2015. The New Fields A and New Fields B were unmanaged native vegetation for over 20 years and transitioned into production beginning in February 2016. A forestry mulcher was used to clear the site of the vegetation, which had the advantage of leaving the carbonaceous material behind on the site. The first samples for New Fields A and New Fields B were collected in January 2018, which was prior to the application of amendments and any vegetable production. The Control was an area that was under native vegetation the entire study and had not been under management for over 20 years; this served as a representation of a natural ecosystem in our study (Table 2.1).

Figure 2. 2. Map of sampling locations: Old (Replications 1, 2, & 3), New A (Replications 1 & 2), New B (Replications 1 & 2), Control (Replications 1 & 2)

Table 2. 1. Treatment, replication, production start date and crop history at sample date on intensive high-value vegetable farm during two-year study in South Louisiana.

			Crop History at Sample Date			
Treatment	Replication	Production Start Date	January 2018	August 2018	January 2019	August 2019
Old Field	1	Summer 2015	Brassicas	Brassicas	Lettuce/ Baby Greens	Eggplant/ tomato/ pepper
Old Field	2	Summer 2015	Flailed and tilled in	Baby Greens/ Lettuce	Brassicas	Eggplant/ tomato/ pepper
Old Field	3	Summer 2015	Lettuce	Flailed and tilled in	Lettuce/ Baby Greens	Eggplant
New Field - A	1	January 2018	Pre-amendment and planting	Tomatoes/ Peppers	Brassicas	Baby Greens
New Field - A	2	January 2018	Pre-amendment and planting	Tomatoes/ Peppers	Brassicas	Baby Greens
New Field - B	1	January 2018	Pre-amendment and planting	Peppers/ Squash/ Okra	Brassicas	Baby Greens/ Sunflower
New Field - B	2	January 2018	Pre-amendment and planting	Cucumber/ Squash/ Okra	Brassicas	Baby Greens/ Sunflower
Control	1	N/A	Native Vegetation	Native Vegetation	Native Vegetation	Native Vegetation
Control	2	N/A	Native Vegetation	Native Vegetation	Native Vegetation	Native Vegetation

2.3. Soil Sampling

Soil samples were collected in January and August both years. At each sampling, 32 soil cores were taken from the plots to a depth of 8 cm using a 1.9 cm soil probe. Samples were combined to get a representative sample and divided in two: (1) chemical analysis by the LSU AgCenter Soil Testing and Plant Analysis Laboratory (STPAL) and (2) biological and physical analysis in the LSU Soil Microbiology Lab.

Samples were transported to the lab in a cooler and soils for biological and physical analysis were stored at -20°C until analysis. Soil was sieved through a <4.75 mm mesh to remove plant matter and debris. Subsamples were saved for archive (20 grams) and field moist soil (10 grams) was saved for FAME analysis and gravimetric moisture content and stored at -20°C. The remainder of the sieved soil was air-dried.

2.4. Soil Variables Measured

2.4.1. Soil Nutrient Status

Macronutrients, micronutrients, soil pH, total C and total N were analyzed by the LSU AgCenter Soil Testing and Plant Analysis Laboratory (STPAL) (Table 2.2). Macronutrients and micronutrients (P, K, Ca, Mg, Na, S, Cu, and Zn) were analyzed based on Mehlich-3 extraction procedure (Mehlich, 1984) followed by inductively coupled plasma (ICP) analysis. Micronutrients (Mn, Fe, Cu and Zn) were determined by using diethylenetriaminepentaacetic acid (DTPA) extraction solution and ICP (Baker and Amacher, 1982). Soil pH was measured in 1:1 in deionized water:soil solution (McLean, 1982). Total C and N were determined using the Dumas dry combustion method by LECO C/N Analyzer (St. Joseph, MI).

Table 2. 2. Soil variables measured for samples of different treatments on an intensive high-value vegetable farm during two-year study in South Louisiana during 2018 and 2019.

Variable	Type of Analysis	Description	Analysis
Macronutrients/ Micronutrients	Chemical	P, K, Ca, Mg, Na, S, Cu, Zn[z]	Mehlich-3 Extraction
Micronutrients	Chemical	Mn, Fe, Cu, Zn	DTPA Extraction
Total C and N	Chemical	Dry combustion	LECO C/N Analyzer
pH	Chemical	1:1 Deionized water : soil solution	pH probe
Soil Organic Matter (SOM)	Chemical	Mass loss	Loss on Ignition (LOI)
Particle Size Analysis (PSA)	Physical	% Sand, Silt and Clay	Hydrometer
Soil Moisture Content	Physical	Amount of Soil Moisture	Oven Drying
Aggregate Stability	Physical	Ability to resist erosion	Wet Sieving Apparatus
Fatty Acid Methyl Ester (FAME)	Biological	Soil microbial community composition: Actinomycetes, Arbuscular Mycorrhizal Fungi (AMF), Fungi, Gram-Positive Bacteria, Gram-Negative Bacteria, Protozoa, Eukaryotes	Gas Chromatograph
Respiration – CO_2 – C Produced	Biological	Microbial activity	CO_2 Meter with Gaslab Software

[z]P = phosphorus, K = potassium, Ca = calcium, Mg = magnesium, Na = sodium, S = sulfur, Cu = copper, Zn = zinc, Mn = manganese, Fe = iron

2.4.2. Soil Organic Matter

The loss-on-ignition (LOI) method was used to determine soil organic matter (SOM) as developed by Nelson and Sommers (1996). Soil samples were oven-dried overnight (15-16 hours) at 105°C and weighed. The samples were then ignited at 400°C for 24 hours in a muffle furnace. After ignition, samples were weighed again. The loss on ignition was calculated with the following formula:

$$\%LOI = [(Weight_{105} - Weight_{400})/Weight_{105}] * 100$$

Soil organic matter was then estimated as recommended in the Cornell Soil Health Assessment Training Manual (Gugino et al., 2009) according to the following equation:

$$\%SOM = (\%LOI * 0.7) - 0.23$$

2.4.3. Gravimetric Moisture Content

Soil moisture was determined gravimetrically by oven drying. Five grams of field moist soil were placed in drying tins and dried in an oven at 105°C for 24 hours and weighed after drying. Percent soil moisture was calculated as:

$$\% \text{ Moisture} = [(\text{Field moist weight} - \text{Dry weight})/\text{Dry weight})] * 100$$

2.4.4. Particle Size Analysis

The hydrometer method described by Grossman and Reinsch (2002) was used to determine the percent of sand, silt and clay of the soil samples. Air dried soil (50 grams) was placed in 500 ml Nalgene bottles with 15 ml of 10% sodium hexametaphosphate to aid in the dispersion of soil particles. Bottles were filled with 250 ml deionized water and placed on a reciprocal shaker for two hours. The soil solution was then transferred to 1 L graduated cylinders. The cylinders were filled to the 1 L mark with deionized water and mixed with a plunger and hydrometer readings were collected at 40 seconds to measure sand content.

Hydrometer readings were taken again at 24 hours without mixing with the plunger for clay content. Silt content was calculated with the following formula:

$$\text{Silt} = [100 - (\text{Sand} + \text{Clay})]$$

Temperature was recorded at the time of the reading to correct for water viscosity if needed, but all readings fell below the threshold of >20° C, so no adjustments were needed. The texture of the soil samples was determined by plotting the sand, silt and clay content using the USDA soil textural triangle.

2.4.5. Water Aggregate Stability

Water aggregate stability was determined using the Eijkelkamp Wet Sieving Apparatus (Giesbeek, Netherlands). For each sample, four grams of air-dried soil were placed in a 0.25 µg sieve and pre-moistened to prevent slaking during the sieving process. Soils samples were immersed repeatedly in deionized water for three minutes. The sieved samples were collected and oven-dried at 110°C for 16 hours and weighed (W1). To correct for sand content, the remaining soil was immersed in a NaOH dispersing solution until remaining aggregates were disrupted and passed through the sieve. The dispersed soil samples were collected, oven-dried at 110°C for 16 hours and weighed (W2). The following formula was used to determine the aggregate stability:

$$\%\ \text{Water Stable Aggregates} = [W2/(W2 + W1)] * 100$$

2.4.6. Fatty Acid Methyl Esters (FAME) Analysis

Soil microbial community composition was determined using ester linked fatty acid methyl ester (EL-FAME) analysis according to Shutter and Dick (2000). Three grams of field moist soil samples were methylated by the addition of 0.2 *M* KOH in methanol and submerged in a 37°C water bath for one hour and vortexed every fifteen minutes. Samples were then cooled to

room temperature for five minutes and neutralized with 3 ml of 1.0 *M* acetic acid and vortexed. Samples were finally extracted with the addition of 3 ml of hexane inverted and centrifuged at 2200 rpm for five minutes. The organic phase was transferred to test tubes and concentrated using N_2 gas. Fatty acids were quantified using an Agilent 7890B gas chromatograph equipped with a fused silica capillary column and flame ionization detector using nitrogen as a carrier gas. Samples were analyzed using a temperature profile which ramped from 190 to 250°C per minute followed by a ramp to 300°C to clear the column. The concentration of FAME (nmol g^{-1} soil) was calculated using the internal standard 19:0 and relative abundance (mol%) was calculated by dividing each FAME by the total sum of all identified FAMEs in a sample. Fatty acids were identified using the library provided by MIDI (Microbial ID, Inc., Newark, DE). Selected FAMEs were used as microbial markers according to previous research (Table 2.3).

Table 2. 3. Fatty acid biomarkers used for identification of soil microbial groups using library provided by MIDI (Microbial ID, Inc.).

Indicator[z]	Marker[y]	References
Actinomycetes	17:0 10-methyl	Frostegard et al., 1993; Zelles, 1997
Actinomycetes	18:0 10-methyl, TBSA	Frostegard et al., 1993
Actinomycetes	16:0 10-methyl	Frostegard et al., 1993; Zelles, 1997
AMF	16:1 w5c	Paul and Clark, 1988; Pennanen et al., 1996
Fungi	18:3 w6c (6,9,12)	Paul and Clark, 1988
Fungi	18:1 w9c	Paul and Clark, 1988; Zelles, 1997
Fungi	18:2 w6c	Frostegard et al., 1993; Zelles, 1997
Fungi	20:1 w9c	Madan et al., 2002
GM -	16:1 w9c	Zelles and Bai, 1994
GM -	16:1 w7c	Ratledge and Wilkinson, 1988
GM -	17:0 cyclo	Frostegard et al., 1993
GM -	18:1 w7c	Frostegard et al., 1993
GM -	18:1 w5c	Ratledge and Wilkinson, 1988
GM -	19:1 w6c	Ratledge and Wilkinson, 1988
GM +	14:0 iso	Morgan and Winstanley, 1997
GM +	15:0 iso	White et al., 1998
GM +	15:0 anteiso	Laczko et al., 1997
GM +	16:0 iso	Frostegard et al., 1993
GM +	16:0 anteiso	Ratledge and Wilkinson, 1988
GM +	17:0 iso	Pennanen et al., 1996
GM +	17:0 anteiso	Laczko et al., 1997
GM +	18:0	Morgan and Winstanley, 1997
Protozoa	20:4 w6,9,12,15c	Pennanen et al., 1996; White et al., 1998
Eukaryotes	22:0	Zelles, 1999
Eukaryotes	23:0	Zelles, 1999
Eukaryotes	24:0	Zelles, 1999

[z] AMF = arbuscular mycorrhizal fungi, GM - = Gram-negative bacteria, GM + = Gram-positive bacteria

[y] FAMEs are described by the number of C atoms, a colon, the number of double bonds, then the position of the first double bond from the methyl (w) end of the molecule. Other notations are used for methyl, cis (c) isomers, iso and anteiso branched FAMEs.

2.4.7. Respiration – CO_2 – C Produced

Microbial activity was determined by measuring the CO_2 – C produced using a CO_2 Meter Sensor Device Kit (Ormond Beach, FL) and GasLab® v2-2.1.23 software. Pre-dried soil (40 g) was added to a beaker that allowed for natural capillary moistening upon the application of water. The beaker was placed in a 473 mL container with a 0-1% CO_2 Meter Sensor. The container was capped and incubated at room temperature for 24 hours. The GasLab® software logged the amount of CO_2 produced from the sample. To determine the amount of CO_2 – C produced, the following formula was used:

$$CO_2 - C = [(\text{Maximum } CO_2 - \text{Minimum } CO_2) * 12.01] / 44.01$$

2.5. Statistical Analyses

All data were analyzed in SAS (version 9.4) (SAS Institute, 2012) using a Mixed model (PROC MIXED) that utilized a completely randomized design (CRD) with repeated measures. The data was collected from nine locations unevenly distributed within four treatments (Old, New Fields A, New Fields B, Control). Each location was sampled four times, therefore the total number of observations was thirty-six. Treatment, time, and treatment by time effects were analyzed for significance ($\alpha \leq 0.05$). A common variance-covariance structure, compound symmetry (CS), was chosen to model the variance. Tukey's Honest Significant Difference post-hoc test was performed to separate means among treatment, time, and treatment by time. SAS's PDXMACRO was used to generate Tukey's letter groupings ($\alpha \leq 0.05$). Pearson's Correlation among the variables was found using SAS (version 9.4) (SAS Institute, 2012) using the PROC CORR function.

CHAPTER 3. RESULTS AND DISCUSSION

3.1. Soil Chemical Analyses

Extractable P and K, selected micronutrients (Mg, Fe, Mn, Zn), pH and SOM were chosen to discuss following the recommendations in the Comprehensive Assessment of Soil Health (Moebius-Clune et al., 2016) (Table 3.1). Complete chemical analyses by sample date can be found in Appendix A.

Phosphorus concentrations in the Old Fields were 108% greater than measured in the New and Control fields. The amount of P in all locations except Control was classified as "High" (>65 mg kg^{-1}) according to LSU AgCenter Soil Testing and Plant Analysis Laboratory (STPAL) and the Control was classified as "Medium" (>35 mg kg^{-1}). "High" indicated that a yield increase from adding additional P is not expected and "Medium" indicated that 75-95% of the crop yield is expected without the addition of P.

In response to the soil test results in January 2018, the fertilizer application was revised, with an application of just N (13-0-0) on many beds. The results indicate a response in the soil system, as the amount of P in the Old Fields (*P=0.014*) decreased from the first sample date (January 2018) to the last (August 2018). In January 2018, P averaged 312 mg kg^{-1} and in August 2019, P averaged 263 mg kg^{-1} in the Old Fields. In the other treatments, P did not change over the sample period (Figure 3.1).

The compost used may have contributed to the higher amount of P in the Old Fields. In this study, the SOM is highly correlated (0.947) with P. The compost that was applied was high in SOM (193 g kg^{-1}) and the Old Fields had the highest SOM (58 g kg^{-1}) of any treatment. The average nutrient analysis value for P in the Compost was 230 mg kg^{-1} (Table 3.2).

Table 3. 1. Soil properties from a study assessing soil health on an intensive high-value sustainable vegetable farm in South Louisiana during 2018 and 2019.

	Soil Properties[z]							
Treatment	P (mg kg^{-1})	K (mg kg^{-1})	Mg (mg kg^{-1})	Fe (mg kg^{-1})	Mn (mg kg^{-1})	Zn (mg kg^{-1})	pH (1:1 Water)	SOM (g kg^{-1})
Old Fields	275 (15.1) a[y]	286 (17.9) a	310 (8.5) ab	163 (35.9) a	33 (7.6) a	26.8 (4.0) a	5.6 (0.12) b	58 (1.7) a
New Fields - A	109 (6.2) b	284 (28.9) ab	281 (9.2) ab	41 (3.5) b	9.1 (1.8) b	22.8 (3.5) a	6.7 (0.10) a	39 (0.8) b
New Fields - B	101 (6.5) b	276 (22.7) ab	232 (8.4) b	18 (0.7) b	5.8 (1.1) b	16.8 (1.6) a	7.4 (0.07) a	42 (0.4) b
Control	35 (4.0) b	179 (14.5) b	337 (17.7) a	37 (1.3) b	9.1 (1.5) b	9.6 (0.60) a	6.8 (0.08) a	31 (1.1) b
P-value	*0.002*	*0.02*	*0.04*	*0.0002*	*0.0002*	*0.3*	*0.0006*	*0.002*

[z]Means for Mehlich-3 extractable phosphorus (P), potassium (K), magnesium (Mg); DTPA extractable iron (Fe), manganese (Mn), and zinc (Zn); soil pH, and soil organic matter (SOM).
[y]Means within a column not followed by the same letter are significantly different at $P < 0.05$ by Tukey's honest significant difference post-hoc test. Standard errors are in parentheses.

Table 3. 2. Soil properties of compost amendment added at an annual rate of 150 m^3 ha^{-1} on an intensive high-value sustainable vegetable farm in South Louisiana.

	Compost Soil Properites[z]							
	P (mg kg^{-1})	K (mg kg^{-1})	Mg (mg kg^{-1})	Fe (mg kg^{-1})	Mn (mg kg^{-1})	Zn (mg kg^{-1})	pH (1:1 Water)	SOM (g kg^{-1})
Compost	230 (19.8)[y]	1031 (76.1)	773 (69)	66 (3.7)	18 (4.3)	23 (1.6)	7.5 (0.5)	193 (0.5)

[z]Means for Mehlich-3 extractable phosphorus (P), potassium (K), magnesium (Mg); DTPA extractable iron (Fe), manganese (Mn), and zinc (Zn); soil pH, and soil organic matter (SOM).

[y]Standard errors in parentheses.

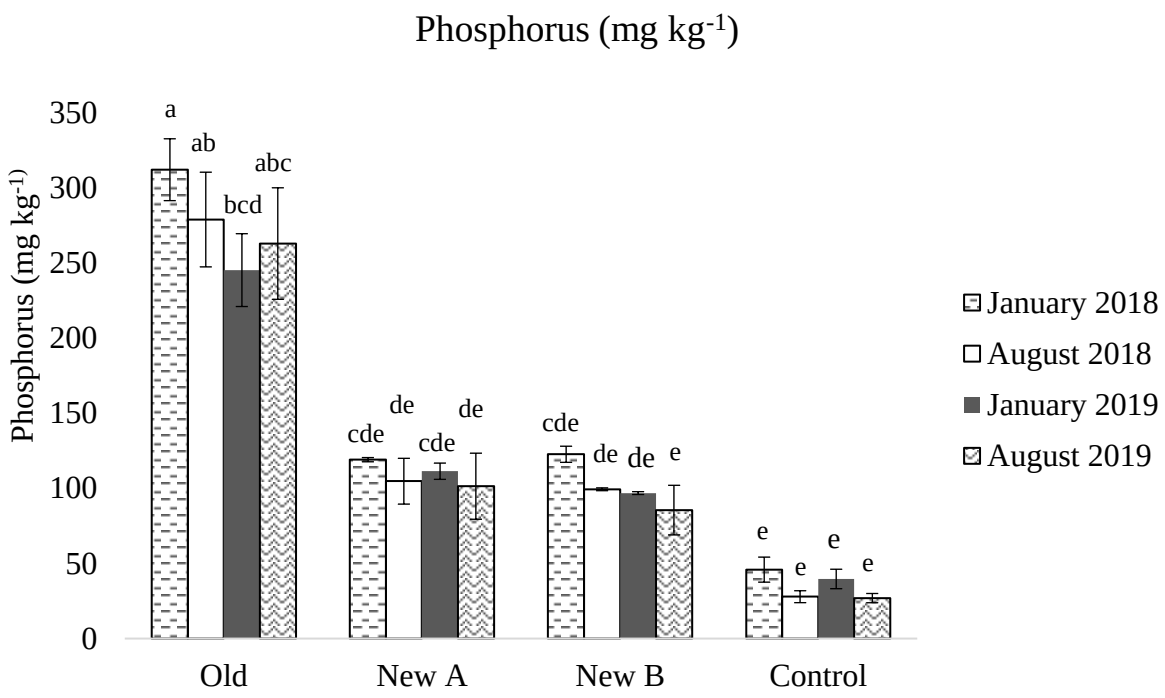

Figure 3. 1. Soil phosphorus (mg kg^{-1}) by sample date on an intensive high-value sustainable vegetable farm in South Louisiana during 2018 and 2019.

Iron (Fe), Manganese (Mn) and soil organic matter (SOM) were significantly higher in the Old Fields as compared to the other treatments. In the Old Fields, Fe was over five times higher than the value in the other treatments. All treatments were classified as "High" according to the LSU AgCenter STPAL (>13.5 mg kg^{-1}), which indicated that a yield increase from the addition of more Fe is not expected. The type of fertilizer and compost used may have contributed to the Fe levels. The fertilizer was derived from blood and meat meals and the compost averaged 66 mg kg^{-1} Fe. The Mn level in the Old Fields was four times higher than the values for the other treatments. According to the LSU AgCenter STPAL, the Old Fields were classified as "High" (>12.0 mg kg^{-1}) and the other treatments were classified between "Medium" (2.01-4.0 mg kg^{-1}) and "High" (>12 mg kg^{-1}).

The soil organic matter (SOM) level in the Old Fields was one and half times higher than that in the other treatments, while he lowest SOM level was in the Control (Table 3.1). There

was an additional 35% SOM in the Old Fields as compared to the Control. In a sugarcane (*Saccharum* spp.) production study conducted by Johnson et al. (2016) in Louisiana with the production study conducted by Johnson et al. (2016) in Louisiana with the same soil series as this study SOM averaged 17 g kg^{-1}, which was 70.4% less SOM than the soil from the Old Fields treatment in our study.

Soil organic matter is considered by some to be the most important baseline measurement of soil health (Doran and Parkin, 1994; Larson and Pierce, 1991). Haynes (2005) reported that SOM was a sensitive indicator of soil health. Stott and Moebius-Clune (2018) discussed how SOM influences many beneficial properties to the health of the soil: it acts as a long-term carbon sink, as a slow-release pool for nutrients, increases ion exchange capacity, influences nutrient cycling, soil aggregation, water holding capacity, and provides nutrients and energy to the plant and soil microbial communities. Previous literature reported that soils that are continually managed for high organic matter tend to require lower farm inputs and be more resilient to drought and extreme rainfall (Moebius-Clune et al., 2016). Reduced tillage, compost application, and lower use of chemical fertilizers are reported to increase SOM content in the soil, which stimulates both microbial community growth and the stabilization and sequestration of carbon (Edwards et al., 1992; Lal, 2002; Swanepoel et al., 2016; Willekens et al., 2014). The site for the current study had a production system that utilizes reduced tillage, relied on additions of compost and did not use any chemical fertilizers.

Although some nutrients were higher in the Old Fields as compared to the other treatments, pH (*P=0.0006*) was lowest in the Old Fields. The Old Fields had an average pH of 5.6, while the other treatments averaged 6.9. The pH of the Old Fields was 7.2 when the site was put into production in the Summer of 2015, considered “Very High”. To reduce the pH,

elemental S was applied at a rate of 244 kg ha^{-1} annually. The results indicate that the application of S was effective at bringing the pH down, but this level is lower than what is ideal for the crops. Based on the soil tests results of January 2018, the application of elemental sulfur was discontinued. In January 2018, the Old Fields had an average pH of 5.2 and in August 2019, the Old Fields had an average pH of 5.8, which indicated that the pH levels in the soil responded when the application of elemental S was stopped.

3.2. Soil Physical Analyses

3.2.1. Particle Size Analysis

All treatment soils were classified as loam according to the United States Department of Agriculture Natural Resource Conservation Service textural classification triangle (USDA NRCS, 2019) (Table 3.3). Soil texture is an inherent characteristic of the soil. Texture has an impact on many physical, biological and chemical processes in the soil, but is not changed easily by management. Loam is known as a medium textured soil with good water and nutrient holding capacity, while still allowing water to drain away.

Table 3. 3. Particle size analysis for treatments on an intensive high-value vegetable farm during two-year study in South Louisiana during 2018 and 2019.

Treatment	% Sand	% Clay	% Silt	USDA Texture Class[z]
Old Field	38 (1.0)[y]	15 (0.7)	47 (1.5)	Loam
New Fields - A	39 (2.0)	14 (0.0)	47 (2.0)	Loam
New Fields - B	40 (0.0)	17 (1.0)	43 (1.0)	Loam
Control	41 (0.5)	16 (0.0)	44 (0.5)	Loam

[z]Texture class calculated from the United States Department of Agriculture (USDA) soil textural triangle.

[y]Standard errors in parenthesis.

3.2.2. Water Aggregate Stability

The aggregate stability was the same in the Old Fields (90.1%) as the Control (93.4%) with both treatments higher than the New Fields (Table 3.4). There was a change in aggregate stability over time. Aggregate stability decreased (*P<0.0001*) from the January 2018 sampling to August 2018 (75%) sampling and then increased in January 2019 (*P<0.0001*) by 19%. Aggregate stability was similar for the January 2019 and August 2019 samplings.

Table 3. 4. Mean water aggregate stability (%) by treatment and date on an intensive high-value vegetable farm during a two-year study in South Louisiana.

Treatment	Aggregate Stability
Old	90 (1.9) a[z]
New A	83 (5.2) b
New B	85 (5.5) b
Control	93 (2.4) a
P-value	*0.003*
Sample Date	**Aggregate Stability**
January 2018	91 (2.4) a
August 2018	75 (4.3) b
January 2019	93 (2.2) a
August 2019	93 (1.4) a
P-value	<0.0001

[z]Means within a column not followed by the same letter are significantly different at $P < 0.05$ by Tukey's Honest Significant Difference post-hoc test. Standard errors are in parentheses.

There was an interaction with treatment and time for aggregate stability (*P=0.018*). In New Fields A, aggregate stability decreased (*P=0.005*) between the January 2018 (81%) and August 2018 (63%) samplings (Figure 3.2). Similarly, in New Fields B, aggregate stability

decreased (*P=0.004*) by 33% between the January 2018 (95%) and August 2018 (64%) samplings, while the Old Fields and Control stayed the same between the two samplings. Aggregate stability in New Fields A (*P=0.003*) and New Fields B (*P=0.003*) increased from the August 2018 and January 2019 samplings by 33% and 28% respectively. By the August 2019 sampling, there was no difference due to treatment.

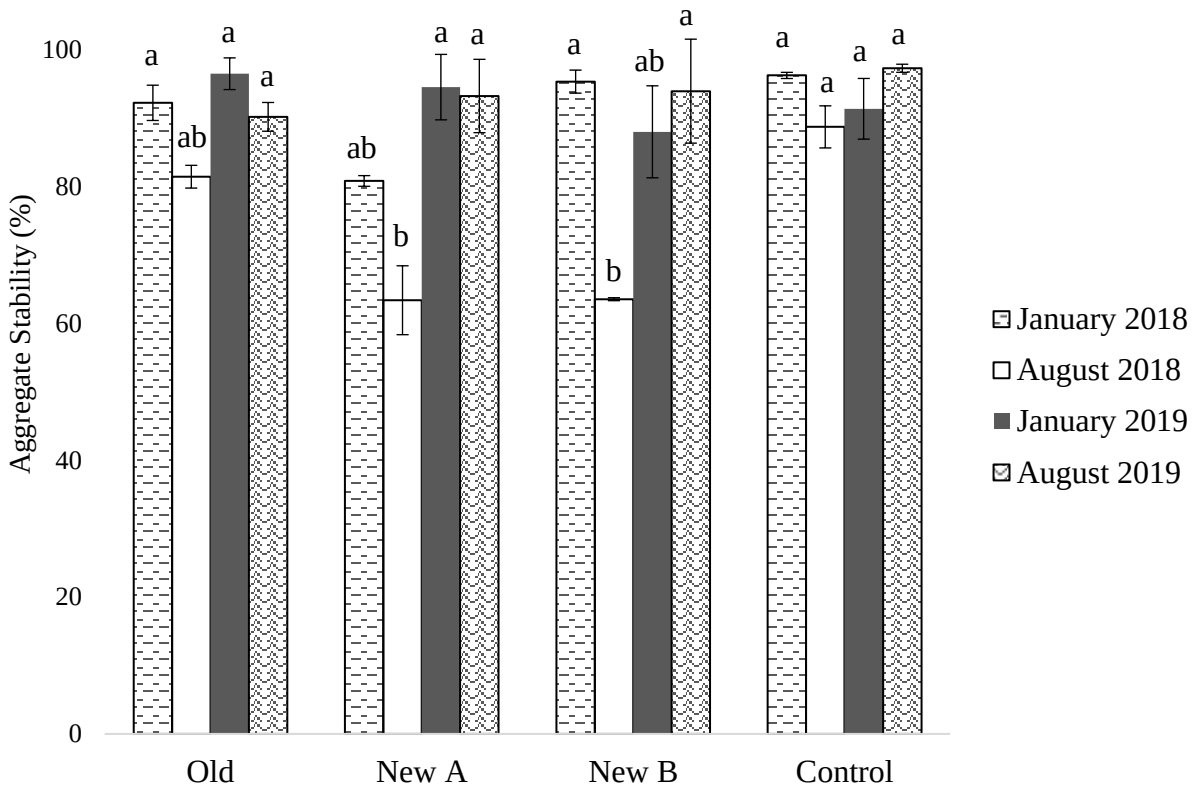

Figure 3.2. Water aggregate stability (%) by treatment and sampling dates on an intensive high-value vegetable farm during a two-year study in South Louisiana.

Aggregate stability is an important measurement as it influences the soil's ability to resist erosion. In general, there is consensus that the higher the aggregate stability, the more beneficial in terms of soil health. The results of our study suggest that when fields are put into production in this system, initially the aggregate stability decreases, but within a year, the aggregate stability rebounded to the level of pre-production aggregate stability and equivalent aggregate stability to

areas that had never been in production (Control). The high aggregate stability could be because in this system, it is a continuous cropping system, with actively growing roots nearly year-round. The root system may be what is causing the high aggregate stability as well as the high additions of organic matter, which has been shown to improve aggregation (Moebius-Clune, 2018).

3.3. Biological Analyses

3.3.1. Fatty Acid Methyl Ester (FAME)

The total FAME (nmol g^{-1}) was not affected by treatment (Table 3.4). Previous studies reported biodiversity was less in agricultural compared to natural systems (Bardgett and Van Der Putten, 2014; de Vries, 2013). In the current study, however, that is not the case as the Control represents a natural ecosystem and Old Fields, New Fields A and New Fields B are all intensively managed agricultural systems and there are no differences in total FAME due to treatment.

While there were no differences in the total FAME, there were differences in some of the individual microbial communities due to treatment. The highest amount of AMF were in the Control (*P=0.0003*) and the lowest AMF were in the Old Fields. Similarly, the control treatment had the highest Fungi:Bacteria ratio at 57% compared to an average of 41% in the other treatments. This trend observed in our study of lower AMF and lower Fungi:Bacteria ratio in agricultural systems compared to natural systems has been found in other studies. Wardle (1995) performed a meta-analysis and reported a strong correlation between tillage and the size of the organisms present. Increased tillage favored greater bacteria and less fungi present. It was also reported that standard tillage practices caused large reductions in fungal biomarkers in vegetable systems in California (Minoshima et al., 2007).

Total FAME , GMp, GMn, actinomycetes, and fungi were affected by sampling date (Table 3.4). The absolute abundance decreased from the first sampling (January 2018) to the second sampling (August 2018), increased in the third sampling (January 2019) and remained the same in the final sampling (August 2019). One possible explanation for the lower microbial abundance in the August 2018 sampling, could be the amount of rainfall received. July and August 2018 received the least amount of rain at 28.5 cm as compared to the other sampling dates averaging 34.6 cm in the sampling month and previous month. Compared to July and August 2019 with 38.0 cm, July and August 2018 with 28.46 cm the area received 25.1% less rain. Other studies have observed this pattern of decreased microbial abundance with lower precipitation (Angel et al., 2010; Cregger et al., 2012).

The relative abundance of fatty acids (mol%) was calculated by dividing each FAME by the total sum of all identified FAMEs in a sample. They followed a similar trend as the absolute abundance of fatty acids (nmol g^{-1}) in the treatment groups and by sample time. The Control had the highest AMF and saprophytic fungi, while the Old Fields had the lowest. These results were similar to other studies demonstrating a system with no tillage was more fungal dominated than a system that was under cultivation (Wardle, 1995). The Old Fields had the highest relative abundance of GMp bacteria as compared to the other treatments. Other studies have found a higher proportion of GMp bacteria in disturbed systems (Xue et al., 2018).

Table 3.5. Absolute abundance of fatty acid methyl ester (FAME, nmol g^{-1}) according to treatment and sample date on an intensive high-value vegetable farm during a two-year study in South Louisiana.

Treatment	Total FAME[z]	GMp	GMn	Actinomycetes	AMF	Fungi	F:B
Old Fields	139 (17.9) ab[y]	32.9 (2.7) a	11.9 (1.1) b	10.0 (0.8) a	3.3 (0.3) c	21.1 (2.1) b	0.38 (0.02) b
New Fields A	154 (22.5) a	31.1 (4.3) ab	17.0 (3.0) a	12.3 (1.9) a	5.6 (0.9) b	27.3 (4.3) a	0.41 (0.03) b
New Fields B	114 (12.7) b	23.4 (2.6) b	13.1 (1.4) ab	9.5 (1.3) a	4.6 (0.3) bc	20.603 (2.1) b	0.45 (0.02) b
Control	144 (17.1) ab	26.9 (3.2) ab	15.0 (1.8) ab	10.6 (1.2) a	9.7 (1.7) a	30.4 (4.2) a	0.57 (0.04) a
P-value	*0.05*	*0.04*	*0.01*	*0.09*	*0.0004*	*0.005*	*0.003*
Sampling Date	Total FAME[z]	GMp	GMn	Actinomycetes	AMF	Fungi	F:B
January 2018	161 (21) a[y]	16.1 (2.1) a	16.1 (2.1) a	10.8 (1.5) a	6.7 (1.5) a	30.8 (4.5) a	0.5 (0.04) a
August 2018	97 (4.1) b	8.9 (0.6) b	8.9 (0.6) b	6.7 (0.4) b	4.1 (0.7) a	18.0 (1.0) b	0.5 (0.03) a
January 2019	146 (12.9) ab	17.1 (1.8) a	17.1 (1.8) a	12.5 (1.2) a	4.6 (0.5) a	23.1 (2.3) ab	0.4 (0.02) b
August 2019	147 (11.4) ab	13.9 (1.3) a	13.9 (1.3) ab	12.2 (0.6) a	6.53 (1.60) a	25.82 (3.21) ab	0.43 (0.03) b
P-value	*0.02*	*0.01*	*0.01*	*0.001*	*0.02*	*0.04*	*<0.0001*

[z]Total FAME = Total Fatty Acid Methyl Esters GMp=Gram positive bacteria; GMn=Gram negative bacteria; AMF=arbuscular mycorrhizal fungi; fungi=saprophytic fungi; F:B=fungi to bacteria ratio

[y]Means within a column not followed by the same letter are significantly different at $P < 0.05$ by Tukey's honest significant difference post-hoc test. Standard errors are in parentheses.

Table 3.6. Relative abundance of fatty acid methyl ester (FAME, mol%) according to treatment and sample date on an intensive high-value vegetable farm during a two-year study in South Louisiana.

Treatment	Relative GMp[z]	Relative GMn	Relative Actinomycetes	Relative AMF	Relative Fungi
Old	23.7 (0.36) a	8.5 (0.37) b	7.2 (0.31) a	2.4 (0.14) c	15.1 (0.45) b
New A	20.3 (0.45) b	10.8 (0.34) a	7.9 (0.45) a	3.6 (0.18) bc	17.7 (0.65) ab
New B	20.7 (0.58) b	11.6 (0.49) a	8.1 (0.42) a	4.2 (0.32) b	18.4 (0.71) ab
Control	18.7 (0.39) b	10.4 (0.39) ab	7.4 (0.28) a	6.7 (0.61) a	21.1 (0.83) a
P-value	*0.001*	*0.001*	*0.10*	*0.0001*	*0.002*
Sample Date	Relative GMp	Relative GMn	Relative Actinomycetes	Relative AMF	Relative Fungi
January 2018	20.7 (0.83) a	10.3 (0.64) b	6.7 (0.26) b	4.4 (0.71) a	19.3 (1.08) a
August 2018	20.6 (0.81) a	9.2 (0.43) c	6.8 (0.23) b	4.3 (0.74) a	18.6 (0.89) a
January 2019	21.1 (0.46) a	11.6 (0.36) a	8.5 (0.24) a	3.2 (0.26) b	15.8 (0.69) b
August 2019	22.2 (0.91) a	9.4 (0.44) bc	8.4 (0.25) a	4.2 (0.68) a	17.2 (0.87) ab
P-value	*0.03*	*<0.0001*	*<0.0001*	*0.0001*	*0.001*

[z]GMp=Gram positive bacteria; GMn=Gram negative bacteria; AMF=arbuscular mycorrhizal fungi; fungi=saprophytic fungi

[y]Means within a column not followed by the same letter are significantly different at $P < 0.05$ by Tukey's Honest Significant Difference post-hoc test. Standard errors are in parentheses.

Although trends can be observed with the results from the current study, thresholds or ranges to define relative functioning have not been previously established, though with the standardization of procedures, more studies and increased available data will allow for thresholds and ranges to be established (Stott and Moebius-Clune, 2018). The standardization of analysis for this particular soil health measurement will be beneficial in order to better understand the impact of cropping systems and production practices on soil health.

3.3.2. Respiration – CO_2-C Produced

Our study showed a correlation between respiration and the ratio of Fungi: Bacteria (0.85) and a negative correlation to total nitrogen (-0.70). There was a difference in respiration among the treatments (*P=0.0002*) (Table 3.6). The Control fields had the highest measured respiration followed by New Fields A and New Fields B and the Old Fields had the lowest respiration. The respiration in the Old Fields was about half that of the Control. Other studies have shown that lower tillage results in increased soil microbial respiration, but that the differences in respiration were inconsistent among locations and tests (Roper et al., 2017).

The respiration and the amount of SOM and total C followed opposite trends in the treatment groups. The Old Fields had the highest SOM and total C with lowest respiration, while the Control had the lowest SOM and total C with highest respiration. This could be an indication that the microbial community in the Old Fields is more efficient at converting organic material into SOM. It also shows there is more carbon dioxide produced per unit of carbon in the Control as compared to the Old Fields.

There was a difference in respiration due to sample date. January 2018 had the highest recorded respiration and January 2019 had the lowest respiration (Table 3.6). Other studies have observed seasonal differences in the amount of CO_2 produced, typically with reduced CO_2 with lower temperatures (Ferreras et al., 2006; Morra et al., 2009). Our study did not follow this trend, with January having both the highest (2018) and lowest (2019) respiration rates. The weather data shows that the beginning of January 2018 (~0° C) was colder than January 2019 (~10° C), which, again, is in contrast to other studies that observed decreased CO_2 respiration with lower temperature.

Table 3. 7. Respiration – CO_2 – C produced (mg kg^{-1}) with 24 – hour incubation according to treatment and sample time on an intensive high-value vegetable farm in a two-year study in South Louisiana during 2018 and 2019.

Treatment	CO_2 - C (mg kg^{-1})
Old	1,276 (129.0) c[z]
New A	1,860 (182.9) b
New B	1,807 (155.6) b
Control	2,220 (96.9) a
P-value	*0.0002*
Sample Date	CO_2 - C (mg kg^{-1})
January 2018	2,221 (108.5) a
August 2018	1,931 (176.3) ab
January 2019	1,400 (146.1) c
August 2019	1,611 (187.1) bc
P-value	<0.0001

[z]Means within a column not followed by the same letter are significantly different at $P < 0.05$ by Tukey's honest significant difference post-hoc test. Standard errors are in parentheses.

Stott and Moebius-Clune (2018) noted that there continues to be a debate about interpretability of respiration results as well as laboratory standards to measure respiration. Cornell's Comprehensive Assessment of Soil Heath concluded that a 4-day incubation is required to obtain sufficient precision (Moebius-Clune et al., 2016). Others have promoted a 3-day incubation (Franzluebbers et al., 2000), still others have shown there to be no difference between a 24-hour incubation, like the Solvita®, and the longer incubations (Haney et al., 2017). Initially in this study, the Solvita® 24-hour incubation was used, but most of the samples

resulted in readings higher than the capacity of the Solvita® paddles. For this study, therefore, samples were incubated for 24 hours and a CO_2 Meter Sensor Device Kit and GasLab® v2-2.1.23 software was used to measure respiration. This method has not been used in other publications, which makes the comparison of the results to existing reports difficult.

3.4. Conclusions

The study results indicate that many of the measurements that are considered critical for soil health assessments were high in this system as compared to what other studies have reported in agricultural systems. These indicators included SOM, aggregate stability and a diverse microbial community as shown by FAME. They were at levels equivalent to the Control, which represents a natural ecosystem. There is a concern in this system about the higher nutrient accumulations, especially P. High aggregate stability, however, reduces the potential of P runoff, which can be an environmental concern. This study is similar to others that have shown that alternative management practices that resemble conservation agriculture have generally improved physical, chemical, and biological aspects of soil health.

More investigation is needed to determine how efficiently the microbial population is operating in an intensively managed production system. It is recommended that additional research include other biological assessments, such as enzyme activity to determine nutrient cycling potential. Additional research could also include yield and crop quality data along with soil health indicators and potential changes in the microbial community after various vegetable crops. As mentioned, there is a need to standardize laboratory procedures to develop reliable recommendations that can be used in determining indexes and thresholds. It is the hope that this research can contribute to the data available for analysis.

Appendix. Nutrients by sample date

Nutrients by sample date on an intensive high-value vegetable farm in a two-year study in South Louisiana during 2018 and 2019. Standard errors are in parentheses.

	Phosphorus (g kg^{-1})			
	January 2018	August 2018	January 2019	August 2019
Old	312 (20.6)	279 (31.5)	245 (24.3)	263 (37.2)
New A	119 (1.4)	105 (15.2)	111 (5.4)	102 (22.0)
New B	123 (5.4)	99 (0.9)	97 (0.9)	86 (16.5)
Control	46 (8.3)	28 (4.0)	40 (6.5)	27 (3.1)
	Potassium (g kg^{-1})			
	January 2018	August 2018	January 2019	August 2019
Old	355 (4.8)	410 (63.3)	416 (32.2)	301 (17.4)
New A	366 (63.2)	303 (51.0)	239 (36.9)	227 (8.0)
New B	361 (10.6)	291 (6.8)	233 (2.9)	220 (0.2)
Control	217 (20.0)	189 (0.6)	165 (39.9)	145 (15.3)
	Calcium (g kg^{-1})			
	January 2018	August 2018	January 2019	August 2019
Old	3172 (61)	3219 (252)	3213 (135)	3251 (224)
New A	4142 (446)	3944 (217)	3480 (343)	3803 (60)
New B	5771 (459)	4855 (120)	5076 (589)	4367 (191)
Control	3095 (202)	3005 (16)	2977 (25)	2669 (331)
	Copper (g kg^{-1})			
	January 2018	August 2018	January 2019	August 2019
Old	19 (2.1)	16 (1.3)	33 (5.4)	16 (1.3)
New A	5 (0.6)	6 (0.1)	8 (0.5)	6 (0.3)
New B	9 (0.2)	9 (0.8)	11 (0.9)	8 (0.5)
Control	5 (0.2)	5 (0.0)	6 (2.0)	5 (1.4)
	Magnesium (g kg^{-1})			
	January 2018	August 2018	January 2019	August 2019
Old	329 (9.9)	313 (17.5)	301 (12.6)	294 (23.0)
New A	268 (23.4)	305 (2.3)	257 (1.9)	295 (1.5)
New B	228 (18.1)	254 (0.9)	241 (0.5)	204 (3.0)
Control	344 (9.3)	367 (23.7)	342 (23.0)	294 (60.7)

(Table cont'd)

Sodium (g kg^{-1})				
	January 2018	August 2018	January 2019	August 2019
Old	82 (8.0)	162 (30.3)	58 (3.6)	77 (9.7)
New A	40 (0.6)	36 (2.8)	22 (5.3)	50 (3.2)
New B	54 (2.6)	52 (1.8)	90 (48.8)	58 (0.2)
Control	52 (1.2)	44 (7.9)	31 (18.1)	32 (5.9)
Sulfur (g kg^{-1})				
	January 2018	August 2018	January 2019	August 2019
Old	57 (3.9)	77 (24.6)	18 (2.7)	37 (8.5)
New A	33 (2.9)	188 (21.0)	55 (4.1)	41 (1.1)
New B	46 (12.1)	64 (16.7)	59 (27.1)	27 (2.0)
Control	28 (4.0)	49 (38.7)	12 (0.1)	14 (2.9)
Zinc (g kg^{-1})				
	January 2018	August 2018	January 2019	August 2019
Old	47 (13.5)	40 (11.7)	33 (9.9)	31 (7.0)
New A	54 (7.7)	38 (14.6)	41 (13.5)	31 (9.6)
New B	41 (4.4)	35 (3.6)	29 (3.8)	23 (1.8)
Control	16 (0.6)	16 (2.4)	16 (0.8)	11 (0.3)
Organic Matter (g kg^{-1})				
	January 2018	August 2018	January 2019	August 2019
Old	58 (4.1)	55 (2.6)	58 (2.4)	60 (4.9)
New A	38 (0.02)	39 (1.8)	41 (1.9)	37 (0.8)
New B	41 (0.77)	42 (0.4)	42 (1.5)	42 (0.1)
Control	29 (0.10)	27 (1.0)	32 (2.1)	34 (0.1)
Total Carbon (g kg^{-1})				
	January 2018	August 2018	January 2019	August 2019
Old	39 (5.0)	41 (1.6)	52 (8.4)	55 (7.7)
New A	24 (2.0)	21 (0.3)	29 (2.9)	26 (2.5)
New B	31 (1.5)	35 (4.6)	33 (3.9)	32 (4.7)
Control	23 (1.8)	18 (1.0)	25 (1.2)	26 (0.6)
Total Nitrogen (g kg^{-1})				
	January 2018	August 2018	January 2019	August 2019
Old	3.4 (0.06)	3.5 (0.11)	3.8 (0.15)	3.9 (0.07)
New A	2.3 (0.27)	2.5 (0.03)	2.7 (0.02)	2.2 (0.08)
New B	2.6 (0.06)	2.8 (0.05)	2.7 (0.07)	2.7 (0.21)
Control	2.1 (0.01)	2.2 (0.04)	2.4 (0.02)	2.5 (0.05)

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Vita

Allison Guidroz was born in Baton Rouge, Louisiana. She graduated from Louisiana State University, where she received the title of Bachelor of Science in Psychology in May 2011. In January 2018, she enrolled in the Graduate School at Louisiana State University under the direction of Dr. Carl E. Motsenbocker to pursue the degree of Master of Science in Plant, Environmental Management, and Soil Sciences, anticipating graduating at the May 2020 commencement.