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Introducing green manures in an organic soybean-winter wheat-corn rotation: Effects on crop yields, soil nitrate, and weeds

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INTRODUCING GREEN MANURES IN AN ORGANIC SOYBEAN – WINTER WHEAT – CORN ROTATION: EFFECTS ON CROP YIELDS, SOIL NITRATE, AND WEEDS

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GREEN MANURES IN AN ORGANIC SOYBEAN – WINTER WHEAT – CORN ROTATION: EFFECTS ON CROP YIELDS, SOIL NITRATE, AND WEEDS

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In organic soybean– winter wheat – corn rotations, animal manure is a common choice to maintain high yields, but leguminous crops grown as green manures after wheat harvest and incorporated into the soil before corn planting, can be an alternative when animal manure is not accessible. Forage legumes with high dry matter (DM) production and high biological N fixation have been shown to meet corn N demand. However, in Eastern Nebraska, lack of precipitation can reduce green manure growth and N fixation, leading to an insufficient N supply for corn, but corn growth can also be impacted by green manure soil water use. Our objectives were 1) to determine the green manure potential of four forage legumes, and 2) to evaluate management methods that optimize green manure benefits.

We conducted an experiment at the ARDC near Mead, NE, from 2011 - 2014. Red clover, white clover, alfalfa, and sweet clover were undersown into winter wheat in early spring. After wheat harvest, they were either mowed or not mowed, and terminated in the fall or the next spring. We measured green manure DM, weed DM, soil nitrate concentrations, and crop yields throughout the rotation. We compared green manure effects to effects of cattle manure, post-wheat soybean green manure, and a control (no fertilizer).

Red clover produced the most DM, up to 5.5 Mg ha^{-1} and showed excellent weed control, especially when mowed. Green manures did not increase soil N compared to the control. Corn yields were always significantly higher after cattle manure (7.6 to 8.1 Mg ha^{-1}) than after undersown green manures, and were lowest after red clover in 2012 (2.8 Mg ha^{-1}) and after white clover in 2013 (4.6 Mg ha^{-1}), because of the clovers' high soil water use and insufficient N production.

In our study, green manures established well, but increased corn yields compared to a control in only one of three years. Cattle manure was the most reliable method to maintain high crop yields. Future research should investigate combinations of cattle and green manure to increase N availability to corn and decrease N leaching losses after corn harvest.

DEDICATION

To Amelie, Justina, Elise, and Natalya

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CHAPTER 1

INTRODUCTION

Organic agriculture is a farming system that does not allow the use of synthetic fertilizers or pesticides (USDA, 2014). Fertilizers permitted under the organic regulations include manure from organic and certain conventional livestock operations and green manures. Animal manure is a common choice to maintain soil fertility in organic crop rotations. Its nitrogen content and availability vary depending on water content, age, and source of the manure, but it is usually regarded as an excellent fertilizer and soil conditioner (Schröder, 2005). However, manure from organically certified farms may not be easily available and the cost may be prohibitive for organic farmers without livestock. Other drawbacks of manuring include the potential for over-application of phosphorus as well as labor and machinery costs associated with manure application and incorporation (Lory et al., 2006).

Green manure crops are thus often planted during fallow periods of organic rotations to supply N and organic matter. By definition, green manures are grown specifically to enrich the soil (Pieters, 1927), although in practice they often have secondary purposes, such as providing ground cover and weed suppression. The term cover crops is typically reserved for plants grown for erosion control, but the terms green manure and cover crops are used interchangeably. The ability to suppress or outcompete weeds is important for a green manure because other methods of weed control, such as tillage, are not possible or economical during the period of green manure growth. Green manures take time to establish and their benefits accumulate the longer they are allowed to grow. In a soybean [*Glycine max* (L.) Merr.]–winter wheat (*Triticum aestivum*

L.)−corn (*Zea mays* L.) rotation the most practical time to plant cover crops is during the window after wheat harvest in July and before corn planting the following spring. In much of the Great Plains region, hot, dry weather in the summer can make cover crop establishment following winter wheat difficult, usually limiting the choice of legumes to drought resistant, warm season species such as soybean or chickpea (*Cicer arietinum* L.).

To give green manure crops better growing conditions and extend their growing season, cool-season legume species can be sown in early spring into winter wheat stands, enabling the farmer to maximize green manure dry matter (DM) production and nitrogen fixation without sacrificing a cash crop (Snapp et al., 2005). A green manure species for this type of rotation and length of growing season should meet several requirements: fix sufficient amounts of nitrogen that can be used by the subsequent cash crop and have a dense canopy with high dry matter production to suppress weeds and add organic matter to the soil. Further, it should not be overly competitive with the wheat and should be short enough so as not to interfere with grain harvest. It should be biennial or perennial and able to cover the soil in winter and resume growth early in the season.

Red clover (*Trifolium pratense* L.) and white clover (*Trifolium repens* L.) are cool-season legume species which meet these requirements. They are native to temperate moist regions of Eurasia, but are now widespread. As true clovers, they have papilonaceous legume flowers with 10 stamens (Taylor, 1985), but red clover grows more erect and white clover is creeping. Red clover is a winter-hardy legume that provides growers with several desired traits: It fixes N, has high biomass yields and forms a dense canopy (Taylor and Quesenberry, 1996). The soil fertility enhancing properties of clovers have long been known. Red clover was probably domesticated in the south of Spain (Kjærgaard, 2003) and white clover in the Mediterranean region (Williams, 1987) and both were quickly imported by other countries, reaching the Netherlands by the middle of the 16^{th} century, France in 1583, England in 1620, Germany in 1645, and the Danish island of Fehmarn in 1710 (Kjærgaard, 2003).

To understand the rapid adoption of clover in European agriculture, one has to recognize the condition of European agriculture in the Middle Ages. Centuries of farming had reduced the supply of soil nutrients, especially nitrogen, leading to a cycle of low yields of food and forage crops which in turn resulted in reduced production of meat and milk by cattle as well as lower cattle reproduction (Kjærgaard, 2003). Cattle manure was the main fertilizer for cereal grains, and several agricultural researchers at the time bemoan insufficient numbers of cattle on farms which they saw as the reason for low cereal yields (Schubardt, 1783; Hatzel, 1795). The advent of clovers improved forage production in terms of quantity and quality, as both white and red clovers contain much more highly-digestible protein than the meadow grass used before, and huge improvements in the health and productivity of cattle followed. Researchers also soon recognized that red clover stands, when plowed under after two to four years, improved soil fertility, and cereal yields. Replacing fallow by clover fields was by some accounts the savior of European agriculture (Kjærgaard, 2003). In Flanders, clovers were so instrumental in the success of Flemish husbandry that the proverb was coined “Without clover no man in Flanders would presume to call himself a farmer” (Weir, 1926, in Taylor and Quesenberry, 1996).

Clover cultivation was not restricted to Europe, for example F.H. King, in his travels to China in the early 20^{th} century, observed clover phases after rice in the rotation

(King, 1911). Red clover was documented in the United States in 1663 (Taylor and Quesenberry, 1996); in 1747 Benjamin Franklin wrote about using red clover to improve meager pastures (Bigelow, 1904), and in 1917 Pieters stressed the value of red clover green manure for the regions of the Eastern United States, Eastern Canada, and the Great Plains. More recently, Gibson et al. (2006) recommend intercropping winter wheat with red clover in Iowa as a means to replace up to 40 kg N ha^{-1} for the following corn crop.

White clover was introduced into New Zealand and Australia with early settlers in the 18th century where it became the most important pasture legume (Williams, 1987). While not as productive as red clover, it can be grazed or cut more often due to its stoloniferous growth (Black et al., 2009). Its perennial features, low-growing habit and winter-hardiness have contributed to its introduction into cropping systems. Japan's permaculture advocate Masanobu Fukuoka promoted white clover as a ground cover to control weeds in grain fields and orchards (Korn, 1982). In the United States, Hartwig and Ammon (2002) discussed white clover as living mulch in sustainably-farmed orchards where its main function is to prevent weed growth and soil erosion. In Denmark, white clover is intercropped with cereals in organic production systems to improve N availability to the grain (Thorsted et al., 2002). In Germany, white clover-grain intercrops significantly raised yields of subsequently sown oats and rye (Neumann et al., 2005).

Alfalfa and sweetclover (*Melilotus officinalis* (L.) Lam.), which are in different genera than the true clovers, are small-seeded forage legumes that are more drought tolerant than red clover (Blackshaw et al., 2010a; Blackshaw et al., 2010b) or white clover (Neal et al., 2011) and thus might be better suited to drier regions of the Great Plains. When intercropped with flax (*Linum usitatissimum* L.), oriental mustard (*Brassica* *juncea* (L.) Coss) or field peas (*Pisum sativum* L.) sweetclover, a biennial, increased the yields of subsequent spring wheat in each year of a three year study in Alberta, Canada (Blackshaw et al., 2001). However, in about half of the site years in the Alberta study, sweetclover reduced yields of intercropped species, an effect that has also been observed when intercropping sweet clover with wheat (Moyer et al., 2007). Intercropped alfalfa showed a positive impact on subsequent corn yields (Liebman et al.; 2012, Hesterman et al., 1992) and can be grazed or hayed in the fall of the establishment year or in the spring before termination. In addition, both alfalfa and sweet clover are able to suppress weeds effectively (Blackshaw et al., 2010a; Anderson, 2010).

The Haber-Bosch process of synthetical fixation of atmospheric N made agriculture less reliant on biological fixation of atmospheric N. Annual grain crops grown for animal feed replaced much of the forages, and agriculture became specialized, with livestock operations separate from cash crop operations. Green manure became almost obsolete in conventional farming, and animal manures were more often regarded as a waste product (Lory et al., 2006). Planting soil improving green manures or leys that contain forage legumes is now prevalent primarily in organic farming systems that are prohibited from using synthetic sources of N fertilizer (Drangmeister, 2003) or in integrated farming systems that use green manures simultaneously as forages.

Timing of green manure termination is a critical management decision as it affects the amount of soil water used by the green manures, the amount of biomass produced and the time available for decomposition. Green manure crops may use considerable amounts of soil water reducing the amount available for the next cash crop and thus lowering yields (Unger and Vigil, 1998). In drier regions, it may be advisable to terminate green manures in the fall. However, green manures that overwinter have higher total DM production (Stopes et al., 1996), potentially depositing higher amounts of organic matter in the soil. Secondary goals, such as winter ground cover, are important to some producers and might be the deciding factor in when to terminate. Termination time also determines when nutrients from green manures become available. Nutrient release from green manure decomposition should coincide with the subsequent cash crop's nitrogen demand but when terminated in the spring, decomposition time may not be sufficient to meet corn N demand when it peaks, about 60 days after planting (Pang and Letey, 2000). When turned under in the fall, however, the potential for N leaching from decomposing plant residues is higher (Crews and Peoples, 2005).

Mowing can be a management tool to improve biomass production of forage legumes, as clovers for example respond favorably to mowing (Black et al., 2009) and overall DM yield increases (Stopes et al., 1996). Mowing a green manure is recommended to prevent weed seed formation and dispersal (Drangmeister, 2003). Ross et al. (2001) found that clover mowed in their establishment year grew back faster than weeds, and reduced weed biomass. Mowing or mulching has been shown to decrease weed growth in perennial forage species such as alfalfa (Norris and Ayres, 1991). Mulching, where the plant residue is left in place after it is mowed, can also affect N availability from green manures. Mowing white clover and leaving the residue on the soil surface increased soil N concentrations while the clover was still growing (Thorsted et al., 2006).

Research that focuses on green manure management in organic production systems is mostly located in the humid areas of the Eastern United States (Blackshaw et al., 2010b, Snapp et al., 2005, Unger and Vigil, 1998) but the results are not always directly applicable in areas with drier and more variable climates, such as the Great Plains. In these areas, research in the management of green manures, in particular choice of species, mulching of green manures and termination time, is needed to realize green manure benefits while avoiding negative impacts on cash crops in the rotation. Our study in Eastern Nebraska attempts to answer the following general research questions:

1. Do forage legume green manures, undersown into winter wheat, increase cash crop yields in an organic soybean-winter wheat-corn rotation compared to post-wheat cover crops or post-wheat manure applications?
2. Do undersown forage legume green manures decrease weed pressure?
3. Do undersown forage legume green manures increase soil nitrate levels after termination?

Dissertation outline and objectives

To answer the research questions, two trials were carried out, as described in the Materials and Methods section. All undersown green manures were forage legumes. The objectives for the first trial were to: 1) compare grain yields and grain protein content of wheat undersown with a green manure with sole cropped wheat; 2) determine dry matter production of the undersown green manures and the effects of mulching and time of termination on undersown green manure productivity; 3) investigate the weed growth within green manures. We hypothesized that 1) winter wheat yields will not be affected by undersown green manures; 2) wheat grain protein will be enhanced by undersown green manures; 3) dry matter yield will be highest for red clover and lowest for white clover; 4) weed dry matter yield will be lowest in the red clover and highest in the white clover.

The objectives for the second trial were to: 1) compare the effects of soil amendments (green manures undersown into winter wheat, post-wheat soybean cover crop, post-wheat manure application, and a control) during the winter wheat phase of the rotation on yields of the crops that follow winter wheat in the rotation (corn one year after winter wheat, soybean two years after winter wheat); and 2) compare the effects of these soil amendments during the wheat phase on the soil nitrate levels for following rotation. We hypothesized that subsequent corn yields would be highest for plots receiving manure, similar for plots undersown green manures or soybean cover crop, and lowest for plots receiving no soil amendment (controls). We further hypothesized that soil nitrate levels compared to the control, would be highest after manure applications, and lowest after the control. The specific hypotheses, methods, results and interpretation for the experiments were organized in six chapters.

Chapters:

1 Introduction

2 Dry matter production of forage legume green manures frost-seeded into organic winter wheat

3 Soil nitrate dynamics following green manures and cattle manure in an organic grain crop rotation

4 Organic corn yields following green manures or cattle manure

5 Weed suppression of leguminous green manures in an organic soybean-winter wheat-corn rotation

6 Summary, limitations and reflections

The first chapter presents a general literature review on the role of green manures in cropping systems. The location, crop rotation, crop management, as well as the experimental layout and treatment design are explained.

In the second chapter, forage legume emergence and productivity in terms of dry matter are analyzed. Productivity was measured as dry weight four times during the growing season and depended on forage legume species, mulching and termination time. Grain yields and grain protein content of winter wheat undersown with green manures was measured, but was not affected by the undersown green manures.

The third chapter investigates the effects of clover-wheat intercrops on soil nitrate over the course of the rotation. Soil testing was begun within three weeks after clover broadcasting and continued through the corn and soybean phase of the rotation. Soil nitrate changes for each soil amendment treatment are compared. Manure treatments had a significant and lasting effect on soil nitrate throughout the rotation, but the soil nitrate amounts in green manure plots were not significantly different from those in control plots.

The fourth chapter discusses the effects of the different soil amendments on cash crop yields in the rotation. Corn yields after cattle manure were significantly higher than corn yields after green manures in each year. The difference was largest in the drought year of 2012, indicating a possible soil moisture deficit after green manures.

Weed growth as affected by clover intercrops is analyzed in the fifth chapter. Weed growth as dry weight was measured at the same time clover growth was measured. It was only sampled in the red and white clover plots, i.e. no weedy control was available. Red clover suppressed weeds more than white clover. Clover mowing also reduced weed dry weight.

Finally, the dissertation contains a conclusion that summarizes what we've learned, what limitations there were, new research questions that arise from this project and the role green manure forage legumes can play in the design of future organic cropping systems.

MATERIAL AND METHODS

Experimental site

The study was carried out at the University of Nebraska's Shelterbelt Research Area located at the Agricultural Research and Development Center near Mead, Nebraska (41˚ 29' N; 96˚ 30' W; 354 m above mean sea level). Windbreaks surrounded all four fields used for this trial on at least three sides (figure 1.1, 1.2). The windbreaks in the center were planted in 1964 and consisted primarily of two rows of eastern redcedar (*Juniperus virginiana* L.) and scattered Scotch pine (*Pinus sylvestris* L.) as well as invading hackberry (*Celtis occidentalis* L.), honey suckle (*Lonicera sp.*), and mulberry (*Morus sp.*). Their average height was 12.3 m. The windbreaks on the outside were planted in 1982 and consisted of double rows of pyramidal eastern redcedar (triple row on west side). Their average height was 9.4 m in the north and south and 8.4 m in the west. All experimental plots were located at least 15 m away from the closest windbreak to avoid competition for soil water as well as shading. Previous experiments in conventional fields at this site observed 15% yield increases for winter wheat due to the windbreaks (Brandle et al., 1984). About half of the soils at the site were Yutan silty clay loam (fine-silty, mixed, superactive, mesic Mollic Hapludalf) with some Filbert silt loam (fine, smectic, mesic Vertic Argiallboll) and to a lesser extent Tomek silt loam (fine, smectic, mesic Pachic Argiudoll) (figure 1.3 and 1.3a). The slope was between 0 and 5%. The study was carried out in three cycles, each starting in the winter wheat phase of the rotation, with the first cycle starting in 2011, the second in 2012, and the third in 2013.

Crop rotation and general field management

All fields in this study have been organically certified, with field 789 transitioning in 2006, and the other fields in 2007. Since then, these fields were in a soybean-winter wheat-corn rotation with every phase of the rotation present in each year. Before the beginning of the experiments, soil fertility was maintained by applications of steer or dairy manure after wheat harvest. Approximately 56 Mg ha^{-1} (solid weight 25%) of manure were applied annually with a custom-made spreader mounted on a semi-truck and disked in within 24 hours. For this experiment, the same rotation was used, but manure was only applied to selected plots (see section on treatments and experimental design).

To prepare for soybean planting, in the spring fields were disked (Keewanee 1010 disk, Kewanee, IL) and field cultivated (Hesston 2210, Hesston, KS). Soybean was planted with a Case IH air planter 900 (Case IH, Racine WI) at a row spacing of 0.76 m. To control weeds, soybean fields were rotary hoed within one week after planting. This was followed by two more passes with the rotary hoe and two passes with a row crop cultivator (Sukup, Sheffield, IA). Soybean was harvested with a Case IH 1640 combine (Case IH, Racine, WI) with a 6.1 m head. Winter wheat was no-till drilled into soybean stubble with a Sunflower 9410 drill (Beloit, KS). No mechanical weed control was carried out in the winter wheat fields. In the first cycle winter wheat was harvested with a Gleaner N combine (Duluth, GA) with a 4.6 m wide head and in the second and third cycle with a Case IH 1640 combine (Racine, WI) with a 6.1 m wide head. For corn, the same soil preparation and weed control practices as for soybean were carried out. Corn was planted with a John Deere 7100 planter at 0.76 m row spacing. Corn was harvested at maturity with the Case IH 1640 combine with a 4.6 m wide head.

Treatment and experimental design

<u>Undersown green manures</u>

Forage legumes were undersown into winter wheat in 2011 (first cycle), 2012 (second cycle) and 2013 (third cycle), in different fields each year according to the rotational sequence (figure 1.1). In the first cycle, the experiment was arranged as a completely randomized design with split-plot treatments. No blocking was used, because the initial N tests revealed no differences in soil nitrate levels among plots, and the field was uniform and non-sloping. The main treatment factors were type of forage legume (red clover and white clover) and mulching regime (mulched once in late summer or not mulched). Split plot treatments were time of clover termination, which were allowed to grow until the fall of the establishment year or the following spring. Main treatments and split-plot treatments were randomly assigned. There were four replications for the forage legume x mulching regime combination, for a total of 16 main plots. Main plot size was 9.1 m by 137.2 m and split plots measured 9.1 m by 68.6 m.

To compare sole-cropped wheat grain yields and grain protein with undersown wheat grain yields and grain protein, control plots (plots without undersown green manures) were created. They were assigned after green manures had already been planted, and thus had to be placed either on the east, north or south side of the field (figure 1.4). Control plots were not split or mulched and measured 9.1 m by 103 m.

In the second and third cycle, the experimental design was changed to a randomized complete block design with 14 replications in the second and 20 replications in the third cycle. In the second cycle, treatments were undersown red clover, undersown white clover, and a control (figure 1.5 and 1.5a). Treatments were randomly assigned to each block. Clovers were not mulched due to insufficient clover stand development (drought year of 2012). The fields used in this cycle were smaller, so plot size was decreased to 9.1 m by 30.5 m for main plots and 9.1 m by 15.3 m for split plots (again, only clover plots were split). In the third cycle, treatments were undersown red clover, undersown white clover, undersown alfalfa, undersown sweet clover and a control. Plots measured 9.1 m by 18.3 m and were not split. Mulching treatments were assigned randomly to red and white clover plots only. Termination time was randomly assigned to whole blocks for ease of management (figure 1.6).

<u>All soil amendments</u>

To compare the effects of undersown green manures with soil amendments applied after wheat harvest, additional treatments were randomly applied after wheat harvest to control plots (plots not undersown with green manures). In the first cycle, four control plots received dairy manure at a rate of 56 Mg ha^{-1}, four plots were planted with a soybean cover crop at a rate of 100 kg ha^{-1} and four plots received no soil amendments (controls). In the second and third cycle, soil amendment treatments were the same except a chickpea post-wheat cover crop at a rate of 100 kg ha^{-1} was added in the third cycle. Since the chickpea failed to establish, it was not included in the analysis.

Plot management

The large plot sizes in this experiment allowed for cultivation and harvest with standard size farm equipment. Clover seed was broadcast into the winter wheat in early spring with a Vicon broadcast spreader (Merseyside, United Kingdom). Seed density was 13.5 kg ha^{-1} for white clover and 22.4 kg ha^{-1} for red clover, alfalfa, and sweet clover, respectively. After wheat harvest, soybean and chickpea plots were no-till drilled. The manure plots were manured and immediately disked to incorporate manure. They were disked again in the fall to kill weeds. The control plots were disked twice to control weeds after wheat harvest. Forty days after wheat harvest, half the red and white clover plots were mulched at a height of 0.1 m with the vegetation remaining on the surface. One week later, tall weeds in the unmulched plots were cut to prevent them from developing seeds. The mower was set at a height of 0.3 m to avoid injury to the green manure canopy. At the end of the growing season, half the undersown green manure plots were terminated by disking twice. The other undersown green manure plots were terminated in the spring by disking twice (see table 1.1 for dates of field operations).

Crop cultivar choice

Each cycle, the same varieties of clover and wheat were used. The red clover variety Marathon is a multi-cut or medium red clover released in 1987 by the USDA-ARS and Wisconsin Agricultural Experiment Station. It is a very winter hardy variety and is more productive in its second year than Arlington red clover which is one of its parents and a very wide-spread cultivar. Marathon is resistant to northern anthracnose and moderately resistant to powdery mildew (Smith, 1994). It yields up to 7.9 Mg ha^{-1} and persists for up to four years in the field (Cooke, 1996). Rivendel white clover originated in Denmark and is a small-leafed variety. It is very winter hardy, tolerates grazing well and has good resistance to nematodes and Sclerotina clover rot (DLF-Trifolium, year not given). 'Yellow blossom' sweet clover is an unstated variety. Alfalfa 'Viking 3200' is a well-adapted variety released by Albert Lea Seeds.

Blaser et al. (2006) researched optimum seeding rates of red clover frost-seeded into winter wheat and recommended winter wheat seeding rates of 300 to 400 seeds m^{-2} and red clover seeding rates of 900 to 1200 seeds m^{-2} for maximum winter wheat grain yields and red clover dry matter production. Our winter wheat seeding rate was 100 kg ha^{-1} equivalent to 400 seeds m^{-2} and red clover seeding rate was 22.4 kg ha^{-1} or 1,300 seeds m^{-2}. In Denmark, white clover was undersown into spring barley at a rate of 8 kg ha^{-1} (Thorsted et al., 2002) and sown as a pure stand at a rate of 25 kg ha^{-1} in Great Britain (Stopes et al., 1996). We selected a white clover seeding rate of 13.5 kg ha^{-1} (2,300 seeds m^{-2}). Alfalfa establishment guidelines for Nebraska recommend drilling at rates of 11 kg ha^{-1} for stands with a companion crop (Anderson and Nichols, 1983) but because we used frost-seeding methods, this rate was doubled to ensure good stand establishment. The same seeding rate as for alfalfa was used for sweet clover. All green manure seeding rates used in this experiment are in the high range. Clover seeds were inoculated with either Apex Green (seed coating containing Rhizobia), Nitragin Gold (*Rhizobium leguminosarum* biovar trifolii), N-Dure (*Sinorhizobium meliloti* and *Rhizobium leguminosarum* biovar trifolii) or Prevail (*Rhizobium leguminosarum* biovar trifolii) provided by the seed supplier and approved for use on organic farms.

The wheat variety Overland is a semi-dwarf cultivar released by the Nebraska Agriculture Experiment Station, the USDA-ARS and the South Dakota Experiment station and is well adapted to the rainfed areas of the Northern Great Plains. It has relatively high yields and medium grain protein content. In trials in Southeast Nebraska from 2004-2006, Overland yielded 4.8 Mg ha^{-1} and had 11.8% grain protein (Nebraska Agricultural Experiment Station, 2007). All seeds were organically certified, except for Marathon and Overland in the third cycle. Clover seed was purchased from Welter Seed (Onslow, IA) and Albert Lea Seed House (Albert Lea, MN).

Data collection

Soil sampling

Soils were sampled either with a JMC Backsaver soil sampler with a 0.02 m diameter stainless steel probe (Forestry suppliers, Jackson, MI) or by using a hydraulically operated stainless steel probe with a 0.03 m diameter. Soil samples were taken about three weeks after undersowing the forage legumes, at wheat harvest, in the fall at forage legume termination and in the spring at forage legume termination (see table

1.2 for measurement schedule). During the corn phase, soils were sampled at corn planting and at corn harvest. During the soybean phase, soil samples were collected in June and at soybean harvest. In the following wheat phase, soils were sampled after wheat harvest. Sampling was done by pushing the probe first to a depth of 0.2 m, retracting it and collecting the soil in a bucket. Then the probe was inserted in the same hole to a depth of 0.6 m, and the soil from that depth was collected in a separate bucket. The soil from the two different depths was analyzed separately. Each soil sample consisted of three to five cores per experimental unit, with the higher number of cores in the larger plots. All soil samples were analyzed by Ward Laboratories (Kearney, NE). Details on the soil analysis are contained in chapter 3.

Emergence counts

Undersown green manure and weed emergence counts were taken approximately seven weeks after undersowing (see table 1.2). In the first and second cycle, three or four samples were collected from each experimental unit. In the third cycle, due to the increased number of experimental units, ten blocks were chosen and within these blocks, three to four samples were collected from each experimental unit. In all cycles, the sampling square size was equivalent to 0.1 m^2 and all emerged clovers and weeds were counted in each square. To determine corn emergence, the number of corn plants in two 3.1 m long rows per plot was recorded. Wheat and soybean emergence was not measured.

Grain yield

Winter wheat yield was determined by harvesting the center 4.6 m (first cycle only) or 6.1 m of each plot with one combine pass along the length of the plot. The grain from this pass was weighed on a trailer scale accurate to 4.5 kg (Parker grain cart 450, Kalida, OH). In the second and third cycle, due to smaller plot areas, grain was emptied into a trash can and weighed on a truck scale accurate to 1 kg. Wheat yields were not adjusted for moisture. From each plot, 1 kg of grain was collected and analyzed for protein with near-infrared (NIR) transmittance technology, adjusted to 12% moisture, using an Infratec 1241 grain analyzer (Foss, Eden Prairie, MN) in the first and second cycle and a DA 7250 grain analyzer (Perten Instruments, Springfield, IL) in the third cycle. Soybean yields were taken in a similar matter to wheat yields. To determine corn yield, the corn grain from one pass of the combine along the length of the plot was weighed on the trailer scale. Grain moisture and protein content were not analyzed for corn or soybean.

Biomass yield

Biomass production of legumes, weeds, and winter wheat was determined by taking above-ground vegetation samples from establishment of the green manure in winter wheat until termination (see table 1.2). For sampling, three areas per experimental unit were randomly selected and all vegetation within a 0.1 m^2 square was cut at ground level. At wheat harvest, biomass production of legumes, wheat, and weeds was determined by sampling from the parts of the plot that were not harvested. At the later sampling times (five weeks after wheat harvest, before fall termination, and the following spring before termination in the overwintered plots), legumes and weeds were sampled by randomly selecting three areas throughout the plot.

In the first cycle, all experimental units were sampled at harvest for a total of 132 samples (44 EU x 3 samples/EU). Due to the large number of experimental units in the second and third cycle, only eight experimental units per treatment were sampled. The total number of samples was 72 (8 EU/treatment x 3 treatments x 3 samples/EU) in the second and 120 (8 EU x 5 treatments x 3 samples/EU) in the third cycle. Biomass was stored in paper bags in an unheated ventilated greenhouse until sorting. It was then sorted into wheat, clover and weeds, and dried in a custom-made drying oven at 65˚C to constant weight (less than 1.5% difference between weighing times).

Weather data

Year-round climate data including air temperature and precipitation were obtained from the Mead climate station located at 41˚ 15' N; 96˚ 48' W (Automated Weather Data Network, ID a255369) which is part of the High Plains Regional Climate Network. This station is located about 1 km distance from the experimental site in an unsheltered area whereas all plots of the experimental site were under the influence of windbreaks. Windbreaks decrease air circulation which can increase air temperature compared to unsheltered areas.

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Table 1.1. Timing of field operations for each phase of the rotation. The year is given for the first management operation per calendar year. Operations in bold font were carried out on all plots. Other operations pertain only to the treatments assigned to individual experimental units.

Field operation	First cycle	Second cycle	Third cycle
Winter wheat planting	October 13, 2010	October 13, 2011	October 10, 2012
Intercrop planting	March 24, 2011	March 14, 2012	March 19, 2013
Wheat harvest	July 18 and 19	June 27	July 16
Soybean cover crop planting*	July 26	July 11	July 31
Manure spreading	August 1	July 10	July 31
Disking manure and control plots	August 19 September 7	July 5 (controls only) July 30 September 12	July 31
Mowing for weed control	August 16	July 18	August 30
Intercrop mowing	September 1	-	August 30
Soybean cover crop disking	November 1	November 1	
Intercrop fall termination	November 1	November 1 & 7**	November 20
Intercrop spring termination	May 1, 2012***	April 30, 2013	April 18, 2014
Pre-planting disk	March 15, 2012	April 30 May 7	April 18 May 6
Field cultivation	April 25**** May 10 May 14	May 14	May 7
Corn planting	May 14	May 15	May 9
Rotary hoe	May 23 May 27	May 23 June 3	May 17 May 23
Cultivate	June 11	June 12 June 19	June 2 June 12
Corn harvest	September 24	October 21	November 6
Pre-planting disk	April 4, 2013	March 24, 2014 May 6	-
Field cultivation	May 16	May 7	-
Soybean planting	May 16	May 7	-
Rotary hoe	May 24	May 17 May 23	-
Cultivation	June 12 June 19	May 29 June 13	-
Soybean harvest	October 1	October 11	-
Winter wheat planting	October 2	-	-

*Soybean cover crop plots were disked and soybeans planted on the same day each year.

**disked again due to incomplete kill after first disking

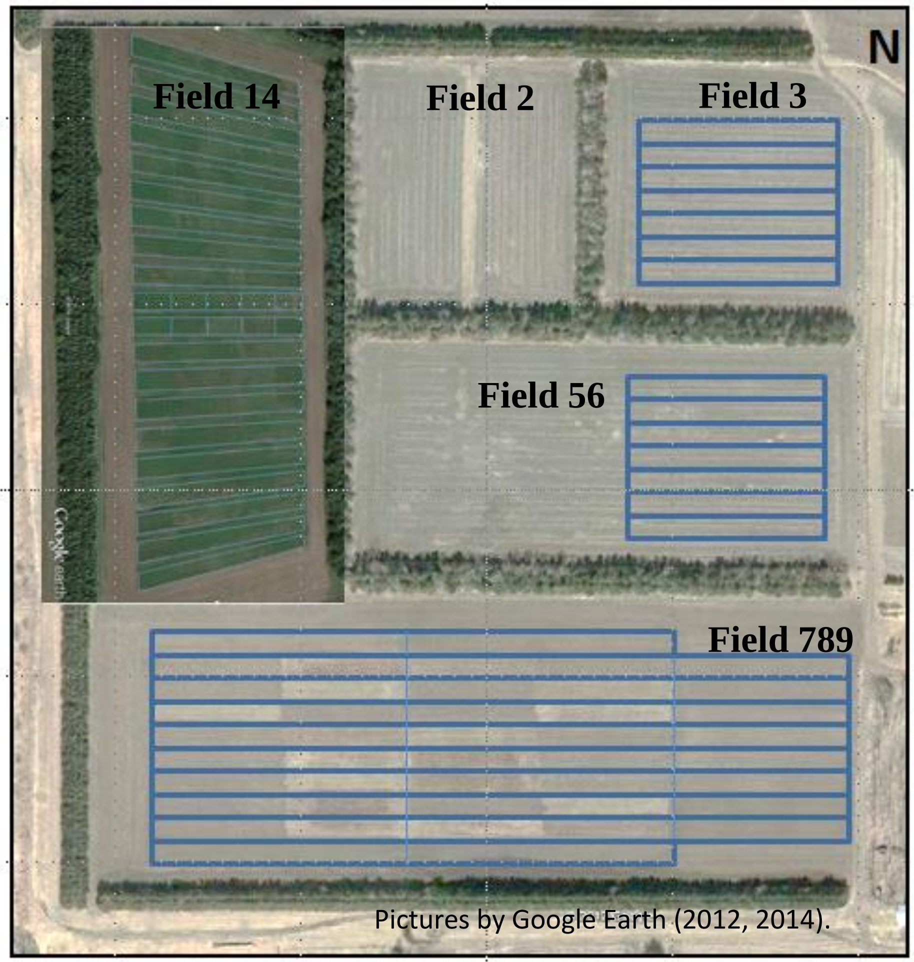
***Mowed clover plots first, then disked.

****Field cultivated all but clover plots

Table 1.2. Measurement schedule for each cycle. Measurements are shown in chronological order. Planting, mulching and termination dates are given for reference. Green manure and weed biomass sampling started at winter wheat harvest, and continued until green manure termination. Sampling could not always be completed in one day due to weather events and the large number of experimental units.

Type of measurement	First cycle	Second cycle	Third cycle
Winter wheat planting	October 13, 2010	October 13, 2011	October 10, 2012
Undersowing green manure	March 24	March 10	March 19
Initial soil sampling	April 6 - 13	March 29	April 2 – April 16
Green manure and weed emergence counts	May 6 – May 13	May 10	May 18
Biomass sampling (“Wheat harvest”)	July 18 & 19	June 28 & 29	July 17 & 18
Winter wheat harvest and yield test	July 18 and 19	June 27	July 16
Soil sampling	July 19 – August 3	June 28 & 29	July 22 & 23
Biomass sampling (“At mulching”)	August 23	July 30	August 23
Clover mulching	September 1	-	August 30
Biomass sampling (“Fall”)	October 11	October 11 & November 9	October 28
Soil sampling	November 1 – 7	November 16	November 20 & December 3
Green manure fall termination	November 1	November 1	November 20
Biomass sampling (“Spring”)	April 26, 2012	April 29, 2013	April 16, 2014
Soil sampling	April 26, 2012	April 30, 2013	April 22, 2014
Green manure spring termination	May 1, 2012	April 30, 2013	April 18, 2014
Soil sampling	May 14 & 15, 2012	May 15, 2013	May 9 & 10, 2014
Corn planting	May 14, 2012	May 15, 2013	May 9, 2014
Corn emergence counts	June 11 & 18	July 8, 2013	Not taken**
Corn harvest and yield test	September 24, 2012	October 21, 2013	November 6, 2014
Soil sampling	September 25 – 28, 2012	October 28, 2013	Not taken***
Soybean planting	May 16, 2013	May 7, 2014	-
Soil sampling	June 21, 2013	July 7, 2014	-
Soybean harvest and yield test	October 1, 2013	Not taken*	-
Soil sampling	October 10, 2013	-	-

*Soybean yields in 2014 were not taken because weeds had overgrown soybeans. High spring precipitation prevented timely weed control.
**Corn emergence counts were not taken due to wet soils.
***Soil samples after corn harvest in 2014 were not taken due to early hard freezes.



Rotation sequence (crops grown in each year) for each cycle during the study period

First cycle – field 789		
Year	Crops	Design
2011	Winter wheat-clover	Completely randomized design
2012	Corn	
2013	Soybeans	EU: 16 clover,
2014	Winter wheat	12 control

Second cycle – fields 3 and 56		Design
2012	Winter wheat-clover	Incomplete blocks
2013	Corn	14 blocks, 3
2014	Soybean	EU per plot

Third cycle – field 14		Design
2013	Winter wheat-clover	Incomplete blocks
2014	Corn	20 blocks, 5 EU per plot

Fig. 1.1. Experimental site with shelterbelts, experimental layout and rotation sequence. Images were taken March 2012 and September 2014 (field 14). Location of blocks (size not to scale) is drawn in field 14 (including plots in center blocks) and fields 3 and 56. Location of main plots is drawn in field 789. Different treatments during the winter wheat-clover phase are visible as lighter and darker shades. Total area is 16 ha.

Fig. 1.2. Layout of field 14, used in the third cycle. Blocks are shown as light blue rectangles and have an east-west orientation. There is a total of 20 blocks, each with five plots. For better illustration, plot borders are sketched in blocks 10 and 11 (center blocks). This picture was taken September 21, 2013, and shows the plots after wheat harvest. In block 10 (center left), treatments were (from top to bottom) white clover, post-wheat soybean cover crop, sweet clover, red clover, and alfalfa. In block 11, treatments were alfalfa, sweet clover, post-wheat chickpea cover crop, white clover and red clover.

Fig. 1.3. Soil survey map for the study site. The area of reference is the area within the blue rectangle. See figure 1.3a (below) for explanation of map unit symbols.

Note the treatment effects visible after wheat harvest in the first cycle.

Saunders County, Nebraska (NE155)			
Map Unit Symbol	**Map Unit Name**	**Acres in AOI**	**Percent of AOI**
7105	Yutan silty clay loam, terrace, 2 to 6 percent slopes, eroded	25.2	51.6%
7280	Tomek silt loam, 0 to 2 percent slopes	7.8	15.9%
7340	Filbert silt loam, 0 to 1 percent slopes	15.9	32.5%
Totals for Area of Interest		**48.9**	**100.0%**

Fig. 1.3a. Explanantion of map unit symbols used in figure 1.3. Obtained from http://websoilsurvey.nrcs.usda.gov/app/WebSoilSurvey.aspx (accessed December 18, 2014).

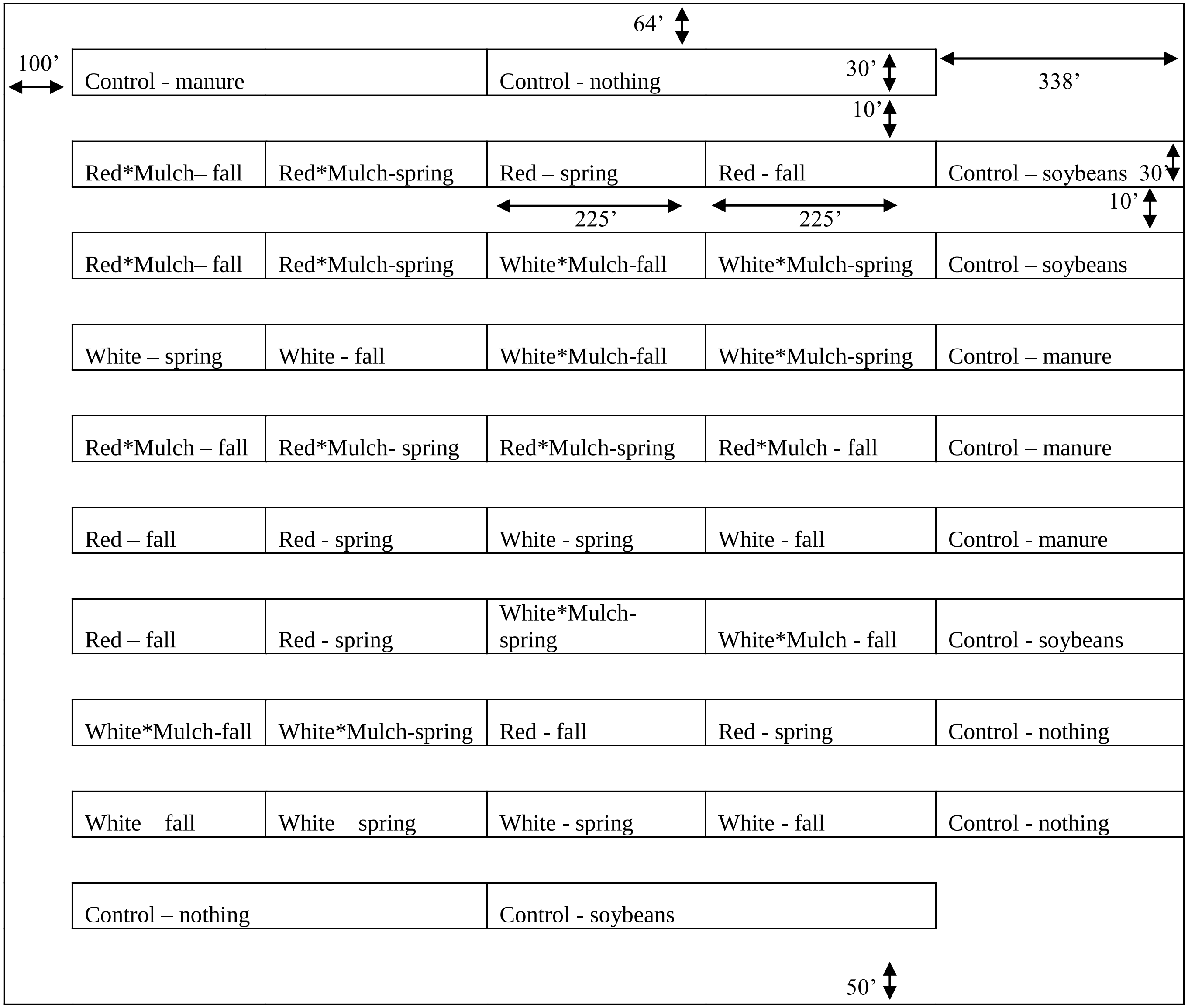


Figure 1.4. Plot map for first cycle, field 789

Explanation of terms:

Red*Mulch = red clover, mulched

White*Mulch = white clover, mulched

White = white clover, not mulched

Red = red clover, not mulched

fall/spring = time of clover termination (split-plot treatment)

Control = not undersown with either white or red clover
Term after “Control” indicates treatment that was applied after wheat harvest

Plot dimensions:
30’ x 225’ for clover
30’ x 338’ for east controls
30’ x 450’ for center controls

Not drawn to scale

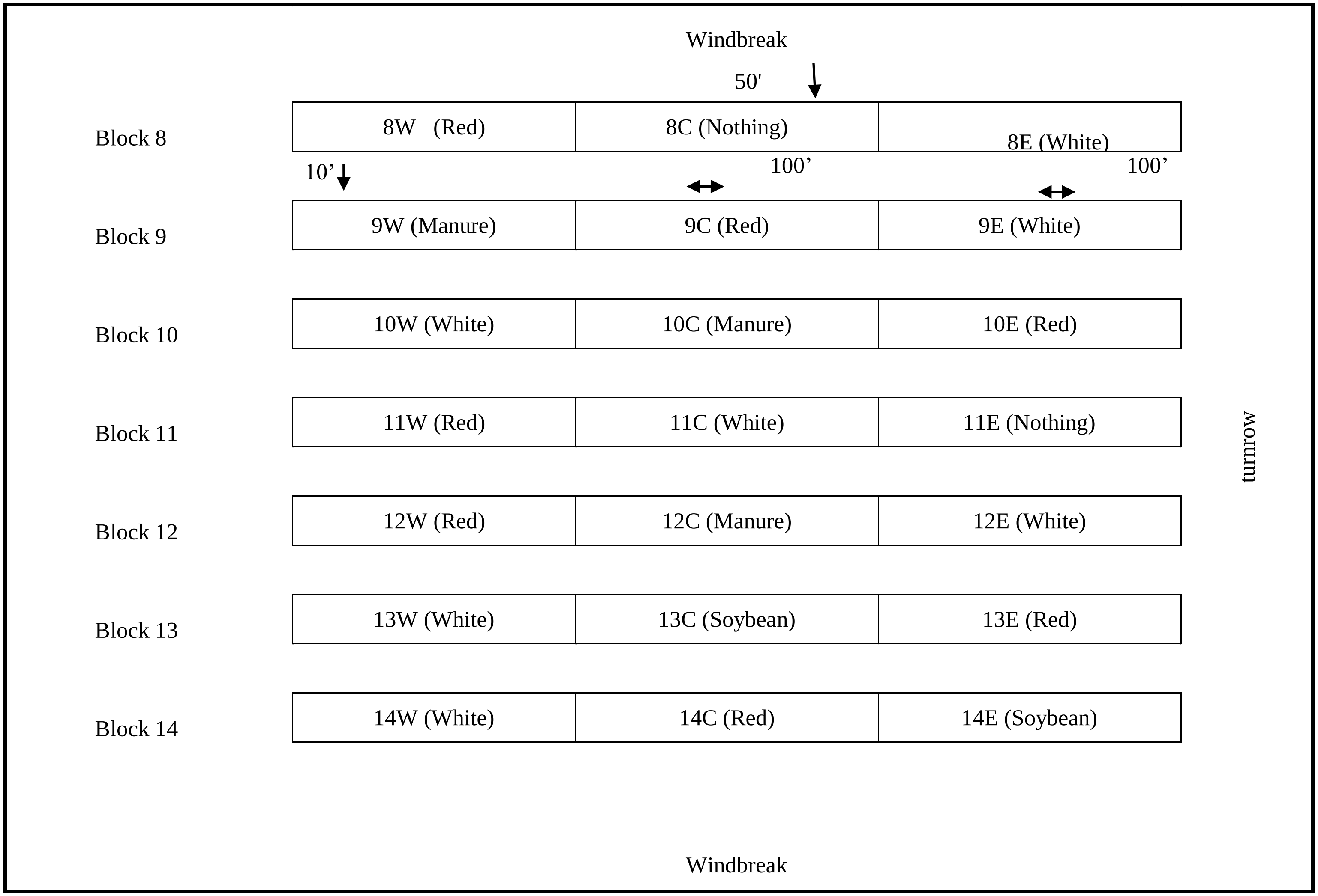


Figure 1.5. Plot map for second cycle, field 56. Explanation of terms: 8W = plot ID. White = undersown white clover. Red = undersown red clover. Manure = manure applied after wheat harvest. Nothing = control. Soybean = soybean cover crop. Mulching or termination time were not applied

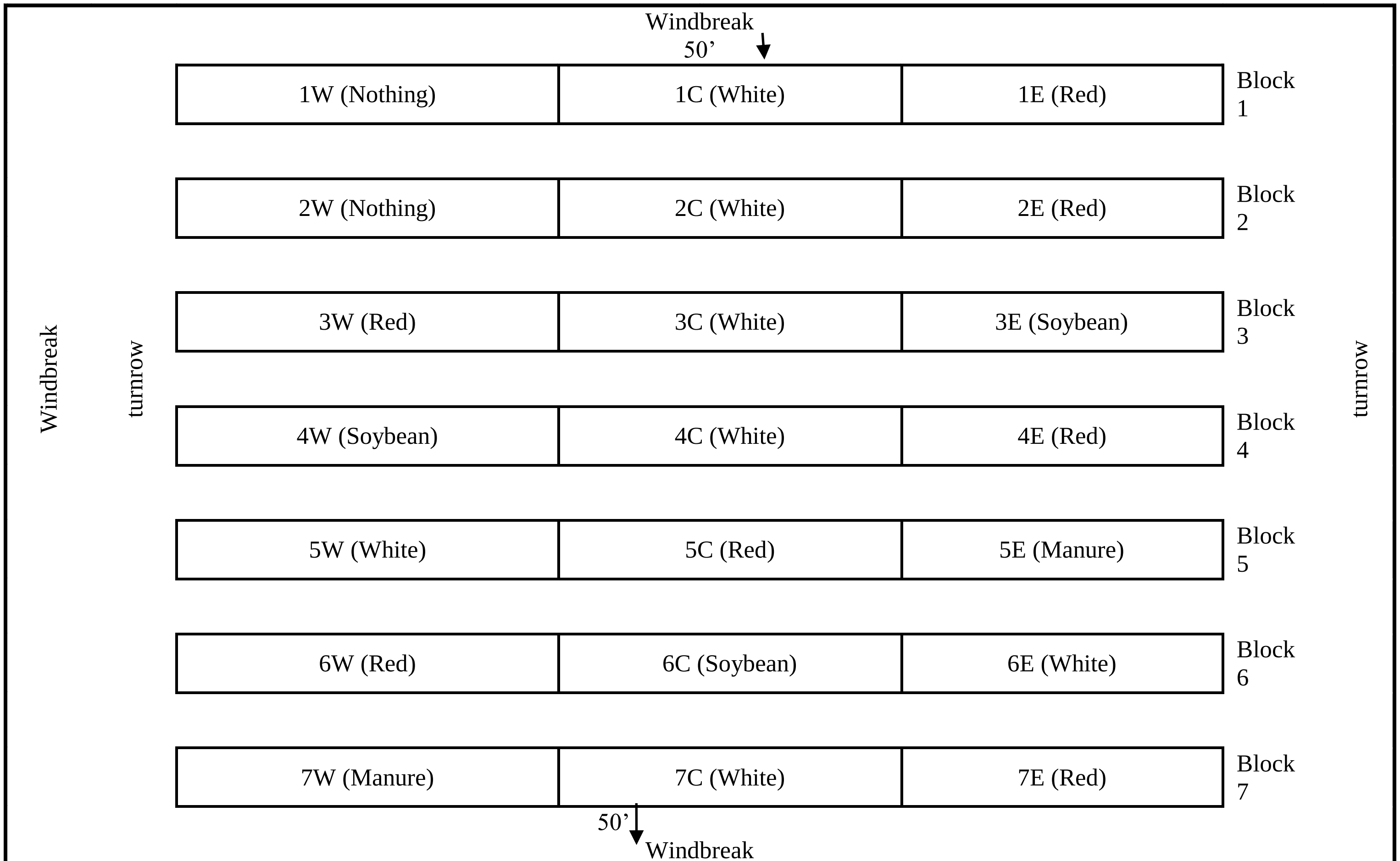


Figure 1.5a. Plot map for second cycle, field 3.

50’ Windbreak

Block					
1	RED	**MANURE**	ALFALFA	WHITE	SWEET
2	WHITE	**NOTHING**	SWEET	RED	ALFALFA
3	ALFALFA	SWEET	WHITE	**MANURE**	RED
4	RED	ALFALFA	**NOTHING**	WHITE	SWEET
5	**SOYBEANS**	ALFALFA	WHITE	RED	SWEET
6	ALFALFA	SWEET	**CHICKPEA**	WHITE	RED
7	RED	WHITE	**CHICKPEA**	SWEET	ALFALFA
8	WHITE	ALFALFA	SWEET	**NOTHING**	RED
9	WHITE	**CHICKPEA**	RED	SWEET	ALFALFA
10	ALFALFA	RED	SWEET	**SOYBEAN**	WHITE
11	RED	WHITE	**CHICKPEA**	SWEET	ALFALFA
12	WHITE	RED	ALFALFA	SWEET	**SOYBEAN**
13	RED	SWEET	WHITE	**NOTHING**	ALFALFA
14	SWEET	RED	WHITE	**NOTHING**	ALFALFA
15	WHITE	ALFALFA	SWEET	RED	**MANURE**
16	RED	ALFALFA	SWEET	WHITE	**MANURE**
17	**SOYBEAN**	SWEET	WHITE	ALFALFA	RED
18	SWEET	WHITE	**CHICKPEA**	RED	ALFALFA
19	SWEET	ALFALFA	RED	WHITE	**SOYBEAN**
20	RED	SWEET	**MANURE**	WHITE	ALFALFA

50’ 10’ 50’ 50’

Fig. 1.6. Plot map Third Cycle Field 14

Plots are: 60’ x 30’

Mow: dots

Fall disk: 1,3,6,9,10, 11,13,14, 18,19

Disk rest in spring

CHAPTER 2

DRY MATTER PRODUCTION OF FORAGE LEGUME GREEN MANURES FROST-SEEDED INTO ORGANIC WINTER WHEAT

Introducing leguminous green manures into grain-based rotations can benefit both conventional and organic farms. Legumes such as red clover (*Trifolium pratense* L.), white clover (*T. repens* L.), alfalfa (*Medicago sativa* L.) and sweet clover (*Melilotus officinalis* L.) add nitrogen to the soil and take up excess nutrients, thus preventing them from leaching. They cover otherwise bare soils before and after main crop harvest, reducing erosion (Pimentel, 1995). As green manures, their main purpose is to enrich the soil for subsequent crops with nitrogen and organic matter (Cherr et al., 2006), a function crucial in organic systems without livestock where legumes are the main source of N.

To realize the dry matter production and N fixing potential of slow-growing forage legumes, they can be planted during the winter wheat (*Triticum aestivum* L.) phase of a soybean [*Glycine max* (L.) Merr.]-winter wheat-corn (*Zea mays* L.) rotation to take advantage of the fallow period between winter wheat harvest and corn planting. In the central Great Plains, summer soil moisture can be low, so small-seeded forage legumes are often undersown into winter wheat in early spring by broadcasting seed on frozen soil (frost-seeding), allowing the freeze-thaw cycle to work the seeds into the soil. This practice is regarded as a practical and economical way of establishing red clover (Snapp et al., 2005), alfalfa (Hesterman et al., 1992) and sweet clover (Cicek et al., 2014) in the Central Great Plains. The green manure continues to grow in the field after winter wheat harvest and is terminated, usually by killing it mechanically, either in the fall of the establishment year or the following spring before corn planting.

The most important management decisions when introducing undersown green manures is the choice of species. High green manure biomass production is important, because it is highly correlated with N fixation (Peoples et al., 2001) and the productivity of the following crop (Parr et al., 2011; Amossé et al., 2013).Winter hardiness is required for winter ground cover and to resume growth in early spring. Low-growing, non-vining legume species are better suited because they rarely interfere with small grain growth and harvest. For example, Stute and Posner (1993) screened several forage legumes for their suitability to be intercropped with a small grain in Wisconsin conventional trials. Hairy vetch (*Vicia villosa* Roth) produced the most biomass but increased lodging of the small grain due to its vining growth habit. Red clover and white clover were better options because they did not interfere with the small grain while still producing up to 3.1 Mg ha^{-1} of dry matter (DM) for red clover and up to 1.8 Mg DM ha^{-1} for white clover. Sweet clover produced up to 3.6 Mg DM ha^{-1} but had very little regrowth if the small grain had to be cut low due to lodging. Other authors also reported on the high productivity of red clover as a green manure, for example Cicek et al. (2014) in organic trials in Manitoba found that undersown red clover yielded more biomass than undersown sweet clover, and more than pea (*Pisum sativum* L.), soybean or hairy vetch, which were grown as cover crops after wheat harvest.

Significant positive effects of a red clover green manure on corn yields have been observed in both organically and conventionally managed fields. In a two-year Iowa study by Liebman et al. (2012) corn yields were between 2.1 and 3.3 Mg ha^{-1} higher after an oat-red clover intercrop than after oats alone when no other fertilizer was applied under conventional management. Gentry et al. (2013) showed that red clover compared to winter fallow under organic or conventional management increased corn yields by 2.1 Mg ha^{-1} in one year of a two-year study, and there was no interaction between farming system (organic versus conventional) and the type of winter cover.

White clover is more commonly used as a green manure in Europe. In Great Britain, it yielded 12.2 Mg DM ha^{-1}, as much as red clover, over a growing period of 13 months with five mulchings under organic management (Stopes et al., 1996). In trials in organically managed fields in Germany, Neumann et al. (2005) found that yields of oat and winter rye increased by about 2 Mg ha^{-1} when grown after a winter wheat-white clover intercrop than when grown after sole cropped winter wheat. Alfalfa and sweet clover are often less productive than red clover (Cicek et al., 2014, Blaser et al., 2011), but because they are more drought-resistant they might be a better green manure choice for drier years (Neal et al., 2011, Blackshaw et al., 2010a).

Beside legume species selection, management tools that can optimize legume DM production and minimize risks associated with introducing perennial forage legumes include termination time and mowing/mulching regime. In the Central Great Plains, green manure crops can use scarce soil water and jeopardize growth and yields of subsequent cash crops (Unger and Vigil, 1998). It might be advisable to terminate the legume in the fall to avoid a soil water deficit. However, overwintering legumes produce more total biomass because they regrow in the spring. In addition, incorporating a green manure in the spring shortly before the planting of the cash crop reduces N loss from leaching of the decomposing plants and can improve the synchrony of N released by the legume and N demand by the cash crop (Crews and Peoples, 2005). Red clover DM production in the fall of the establishment year was higher than the following spring (3 Mg ha^{-1} versus 1.3 Mg ha^{-1}) in a study in New York state (Schipanski and Drinkwater, 2011) but few studies compare biomass yields at different incorporation times.

Organic farmers have few options to control weeds in a green manure crop, but mowing can significantly reduce weed pressure (Ross et al., 2001) and destroy volunteer wheat which is a host for mites and aphids that transmit several virus wheat diseases (Brakke, 1987). Further, red clover and white clover can be mowed to make high-protein, easily digestible hay for livestock (Black et al., 2009). While this was not an objective of this study and technically does not fit the definition of green manure, farmers might wish to market forage legume hay as an additional source of income. Typically, when mowing green manure, the plant residue would be left in place as mulch, so that nutrients released by the decomposing green manure are added back to the soil. Because it grows from stolons on the soil surface, white clover can be cut more often than red clover (Black et al., 2009). Defoliation, whether it is from mulching or haying, however, reduces the plant's photosynthesis ability and assimilation and can affect winter survival and dry matter production the following year (Taylor and Quesenberry, 1996). Information on the effects of mulching forage legumes in their first year is necessary to assess possible reductions in DM yield at green manure termination.

While green manures are intended to improve N supply to the following cash crop, undersown legumes may affect the companion small grain in several ways. For example, better nitrogen nutrition of wheat intercropped with pea (*Pisum sativum* L.) has been reported in low soil N environments (Bedoussac and Justes, 2010) and could be of interest for organic wheat producers wanting to increase wheat grain protein content and wheat yields. Indeed, studies in Denmark and Sweden have attempted to manipulate winter wheat-white clover intercrops to increase N transfer to the wheat (Bergkvist, 2003; Thorsted et al., 2006) but in these studies, wheat was planted into established white clover stands. In temperate regions of the United States, a legume frost-seeded into winter wheat would be small, with low nitrogen fixation at wheat jointing, when wheat grain yield responds the most to additional nitrogen (Hergert, 2014). This is likely why most studies in the temperate regions of the United States report little or no significant influences of undersown forage legumes on grain yield or grain protein content (Blaser et al., 2006; Hesterman et al., 1992).

Despite the potential benefits of using undersown green manures in organic grain-based rotations, few studies have been conducted under organic management conditions. Long-term organic-conventional farming system comparisons have found higher microbial biomass (Mäder et al., 2002) and soil organic matter (Pimentel et al., 2005) and higher (Pimentel et al., 2005) or lower soil mineral N (Drinkwater et al., 1995) in organically managed fields which could affect green manure DM yields as well as yields of intercropped winter wheat. Studies that measure undersown green manure DM production at more than one time during the season are scarce, but provide knowledge essential in understanding peak green manure productivity which can in turn inform timing of management decisions.

This study aims to better understand the influence of legume species, mulching and termination time on undersown green manure dry matter production and the intercropped winter wheat grain yield and grain protein content in an organic grain-based systems. Our hypotheses were (i) red clover DM production would be highest, (ii) mulching would impact red clover DM more than white clover DM yield, (iii) DM yield at fall termination is higher than at spring termination, and (iv) winter wheat grain yields or grain protein contents would not be affected by undersown species.

We use the term undersown green manures to describe the establishment process and intent of forage legumes planted into winter wheat. In the literature, the term relay intercropping or relay cropping has been used recently by Amossé et al. (2013, 2014) as well as Cicek et al. (2014), respectively, to define this system. However, the terms “cropping” or “intercropping” risk confusing this system with one that produces two marketable crops which by definition, is not the intent of a green manure.

MATERIALS AND METHODS

Site and Soils

The site is located in eastern Nebraska at the Shelterbelt Research Area of the Agricultural Research and Development Center near Mead (41˚ 29’ N; 96˚ 30’ W; 354 m above mean sea level). Soils were mostly Yutan silty clay loam (fine-silty, mixed, superactive, mesic Mollic Hapludalf) with some Filbert silt loam (fine, smectic, mesic Vertic Argiallboll) and to a lesser extent Tomek silt loam (fine, smectic, mesic Pachic Argiudoll) with a slope of less than 5%. Moderately dense windbreaks consisting of two or three rows of mostly eastern redcedar (*Juniperus virginiana* L.) at a height of 8.4 m to 12.3 m surrounded all fields used for this trial on at least three sides (figure 1.1).

Experimental and treatment design

The first cycle of this study was initiated in 2011, the second in 2012, and the third in 2013, respectively, with the undersowing of forage green manures into winter wheat. Fields and rotation sequences are available in figure 1.1. A completely randomized design was used in the first cycle, with type of forage legume (red or white clover) and mulching regime (mulched or not mulched) as main treatments with four reps for each clover by mulching combination. Clovers were mulched (mowed at a height of 0.1 m with plant residue left in place) 40 days after winter wheat harvest. In the fall, each clover plot was divided in half, with one half of the plot terminated in the fall of the establishment year and the other half terminated in the spring of the second year (figure 1.4). Control plots (n=12), i.e. plots not undersown with clovers, were established after clover planting and thus had to be placed on north, east, and south side of the field. In the second and third cycle, the experimental design was a randomized complete block design with 14 replications in the second and 20 replications in the third cycle, respectively. Treatments in the second cycle were red clover, white clover, and a control. The mulching treatment was not used, because clover DM production was very low due to drought conditions. Plots were again divided in the fall, with each half receiving either the fall or the spring termination treatment. Treatments in the third cycle were undersown red clover, white clover, alfalfa, sweet clover, and a control. Mulching was randomly assigned to red and white clover plots, but not the other treatments. Termination time was randomly assigned to whole blocks, to make disking with field-size equipment easier.

Crop Management

The semi-dwarf winter wheat ‘Overland” was no-till drilled into soybean stubble with a Sunflower 9410 drill (Beloit, KS) at a seeding rate of 100 kg ha^{-1} equivalent to 400 seeds m^{-2} in October (see table 1.1 for planting dates). Forage legumes were frost-seeded into winter wheat stands the following spring with a Vicon broadcast spreader (Merseyside, United Kingdom) at a rate of 22.4 kg ha^{-1} for red clover ‘Marathon’, alfalfa ‘Viking 3200’, and yellow sweet clover VNS, and at a rate of 13.5 kg ha^{-1} for white clover ‘Rivendel’. Number of seeds per kg for ‘Marathon’ and ‘Rivendel’ was 600,000 seeds kg^{-1} and 1,700,000 seeds kg^{-1}, respectively, as stated on seed tags. Number of seeds per kg were obtained from the USDA Plants Database for alfalfa and sweet clover, and were 500,000 and 570,000 seeds kg^{-1}, respectively. Table 2.1 shows purity, rate of germination, percentage of hard seed and inoculant for each legume species. The same cultivars were used each year and were chosen for high DM production capacity and winter hardiness. Forage legumes were terminated by disking twice with a Keewanee 1010 disk (Kewanee, IL).

Data Collection

Emergence counts of the forage legumes were taken approximately seven weeks after frost-seeding (table 1.2) in at least eight plots per treatment. In each of the randomly selected plots, three samples were taken by counting all forage legume seedlings within a 0.1 m^2 quadrat. The following formula was used to calculate the number of viable seed (actual seeding rate)

Target seeding rate x %purity x %germination = Actual seeding rate

Wheat plants were not counted. Wheat was harvested at maturity with a Gleaner N combine (Duluth, GA) with a 4.6 m wide head in the first cycle and with a Case IH 1640 combine (Racine, WI) with a 6.1 m wide head in the other cycles. Wheat grain yield was determined by weighing all grain from one pass along the center of each plot on a grain cart (Parker 450, Kalida, OH) with an accuracy of 4.5 kg. In the second and third cycle, plots were shorter and wheat grain from the center strip of each plot was emptied into a trash can and weighed on a truck scale with an accuracy of 1 kg. Wheat yields were not adjusted for moisture. Wheat grain protein from each plots was analyzed with near-infrared (NIR) transmittance technology with a Foss Infratec 1241 (Eden Prairie, MN) in the first and second cycle, and a Perten DA 7250 (Springfield, IL) in the third cycle. Dry matter (DM) production of undersown forage legumes was determined by taking above-ground biomass samples starting at winter wheat harvest (“Wheat harvest”), 35 days post-harvest to assess DM at mulching (“35 d post-harvest”), in the fall when DM accumulation had largely ceased (“October”) and in the overwintered plots in the spring shortly before spring termination (“April”) (table 1.2). Whole wheat plant biomass was taken at wheat harvest. For biomass sampling, three areas per plot were randomly selected and all vegetation growing within a 0.1 m^2 quadrat was cut at ground level, sorted into clover, weeds, and wheat (only at wheat harvest), dried at 65˚C to constant weight and then weighed. All dead plant material was discarded. Year-round climate data was available from the Mead climate station located in an area about 1 km away and not surrounded by windbreaks (Automated Weather Data Network, ID a255369, High Plains Regional Climate Network).

Water use of red and white clover was not measured, but was estimated using reported water use efficiency (WUE) values, measured DM values and observed precipitation (table 2.2). Total water use (soil water and precipitation, percolation was neglected) was calculated using estimated WUE and observed precipitation. Then, soil water use was estimated. Water use efficiency (according to Badaruddin and Meyer, 1989):

1. WUE (kg ha^{-1} mm^{-1}) = DM (kg ha^{-1})/ Total water used (mm)
2. Total water used (mm) = DM (kg ha^{-1})/ WUE (kg ha^{-1} mm^{-1})
3. Total water used (mm) = Precipitation (mm) + Soil water (mm)
4. Soil water (mm) = Total water used (mm) – Precipitation (mm)

Statistical Analysis

Emergence and DM data were analyzed with ANOVA implemented using PROC GLIMMIX in SAS 9.4 (SAS Institute, Cary, NC). For DM and emergence measurements, the means of the subsamples were calculated using PROC MEANS before conducting the ANOVA. To compare emergence, DM, and wheat protein across cycles, blocks were imposed on the completely randomized design in the first cycle after the data was collected (n=8). Cycle, forage legume species, mulching and their interactions were fixed effects and block was a random effect. For DM, sampling time was not used as a variable, i.e. a separate ANOVA was conducted for each sampling time. Wheat yield was not compared across cycles because it was not adjusted for moisture. Least-square means were compared with the relatively conservative Tukey or Tukey-Kramer (for unequal sample sizes) tests using a significance level of $\alpha = 0.05$.

RESULTS AND DISCUSSION

Climate conditions

Table 2.2 has monthly air temperature averages and monthly precipitation totals. In the first cycle, temperatures in March, April, May, and June were within

0.3˚C of the normal. Precipitation was 30 mm lower than the normal of 44 mm in March, 13 mm higher than the normal of 74 mm in April, 55 mm higher than the normal of 101 mm in May and 23 mm higher than normal in June. July temperature was 26˚C, 2˚C warmer than normal, and had 72 mm of rain, 15 mm less than normal. August had normal temperature and 19 mm more rainfall. September was 2.5˚C cooler than normal and dry, with only 19 mm, 51 mm less than normal. October was warmer than normal by about 1.5˚C and dry, 40 mm less than the normal of 56 mm. Dry conditions prevailed between November and March, with 30 mm less rainfall than the 133 mm normal for this period, while temperatures were on average 3 ˚C above the normal for this period (3.9˚C versus 0.9˚C) with March being 7.6 ˚C above the average.

In the second cycle, dry and warm conditions continued. April was 2.5˚C warmer than normal, May 3.1 ˚C, June 2.2, and July 1.4 ˚C warmer than the 30-year average. Rain fell until June, although there was a 36 mm deficit compared to the normal rainfall amount between March and June. July had 2 mm of rainfall, and temperatures were 27.8, a record high. Temperatures in August and September were close to normal, but precipitation was 14 mm in August, and 34 mm in September, with drought conditions in much of the area. Temperatures in October were 1 ˚C below normal, but 2 ˚C above normal in November. Hardly any precipitation fell until April, when 11 mm more than normal fell. April was much cooler than normal.

In the third cycle, drought conditions improved. May received 27 mm more rain than normal, and June rainfall was normal, with average temperatures.

July was dry again, with only 16 mm, but also 0.9 ˚C cooler than normal. August rainfall was 35 mm less, and September 13 mm more than normal, and September was 2.5˚C higher than normal. October and November temperatures were close to normal, and rainfall 18 mm above normal for these two months. There was no precipitation and no snow cover between December and March, and temperatures were 2.6 ˚C below normal in December, 0.9 ˚C below normal in January and 4 ˚C below normal in February, a record cold. April received about 84% of its normal precipitation and had average temperature.

Clover emergence

Forage legume frostseeding resulted in successful establishment in each cycle and for each species (table 2.3). Clover species and cycle were significant, as well as the interaction between species and cycle (table 2.4).

In the first cycle, the percentage of viable seed or actual seeding rate could not be calculated for red clover due to missing information (table 2.1) so the average actual seeding rate from the second and third cycle was used (992 seeds m^{-2}). Percent viable seeds emerged (emergence/actual seeding rate x 100) was 64%, 49%, and 93% in the first, second, and third cycle. For white clover, this percentage was lower in each cycle, with 32%, 13%, and 45% in the first, second, and third cycle. The lowest emergence of viable seeds was in the second cycle, probably because the seedbed in March was frost-free, lacking the freeze-thaw cycle necessary to incorporate clover seeds into the soil. Because of the high temperatures, wheat resumed growth earlier in the spring which could have lowered light transmission and thus clover emergence. In the first cycle, snowfall immediately after clover broadcasting provided cover and moisture and in the third cycle, seeds were broadcast onto snow. Cool spring temperatures in the third cycle did not hinder germination, as red clover germinates at 3 ˚C, white clover at 5 ˚C and alfalfa and sweet clover at 1˚C (Agriculture and Forestry Alberta, 2000). In March and April, precipitation did not differ much from normal in each cycle, but temperatures were much higher than normal in the second cycle, and lowest in the third. High evapotranspiration rates could have lowered soil water availability for seeds in the second cycle, and very low evapotranspiration probably improved germination in the third cycle.

Our actual seeding rates (viable seeds m^{-2}) differed from cycle to cycle due to differences in purity and germination rate of the seeds. However, the plant density was likely sufficient to establish dense stands. Blaser et al. (2006) frost-seeded red clover into winter wheat and triticale (X *Triticosecale* Wittmack) in a two year study under conventional management in Iowa and found red clover (‘Cherokee’, 94% germination, 100% purity) target seeding rates of 1,200 seeds m^{-2} resulted in 90 to 107 plants m^{-2} and seeding rates of 1,500 seeds m^{-2} resulted in 126 to 130 plants m^{-2} seven weeks after planting, much lower than the plant densities observed in our study. Cicek et al. (2014), in a study under organic growing conditions in Manitoba, used 400 red clover seeds m^{-2} and obtained less than 25% emergence after eight weeks. Red clover was intercropped with a fall rye cultivar that is 4 cm taller than 'Overland' and could have decreased light transmittance to the clover.

White clover is rarely sown alone, thus few studies have investigated white clover emergence in pure clover stands. In Alberta, 49% of white clover seeds broadcast as cover crops had emerged ten weeks after planting in a high-fertility site and 67% in a low-fertility site (Ross et al., 2001). Alfalfa and sweet clover plant populations at seven weeks after frost-seeding were intermediate between red clover and white clover (926 and 772 plants m^{-2}, respectively). In the study in Manitoba (Cicek et al., 2014) less than 20% of sweet clover planted at 400 seeds m^{-2} had emerged 8 weeks post-planting.

High seeding rates in our study produced high plant densities, but a much higher percentage of seeds emerged than what was reported in the literature. Clover population was not documented later in the season, but by observation, clover plant density was substantially lower at wheat harvest and later in the season. Plant density decrease over the growing season can be described with the self-thinning rule (Westoby, 1984). This simple population model predicts the mortality rate of plants in even-aged stands as a function of plant biomass accumulation. Depending on the growing conditions, biomass accumulates until the carrying capacity is reached and plants start to die off as a consequence of competition. For example, stand densities of alfalfa and red clover undersown into winter cereals in Iowa at rates of 900 seeds m^{-2} were between 5 and 22% of the seeding rate at cereal harvest. Yet dry matter production was not significantly influenced by intercrop plant density at harvest (Blaser et al., 2011).

White clover seed costs were \$356 ha^{-1}, twice as high as red clover seed costs. Sweetclover seed was cheapest at \$109 ha^{-1}. To save costs on forage legume seeds, all forage legumes used in this study can likely be frost-seeded at 50% of the rate used here or 75% if broadcast on frost-free ground.

Green Manure Dry Matter Production

Green manure DM production is shown in figure 2.1. In the first cycle, red clover DM was significantly higher than white clover DM at each sampling time (table 2.5). Clovers grew slowly during the time they were growing with winter wheat (figures 2.2 and 2.3). Red clover DM at “Wheat harvest” was 0.43 Mg ha^{-1}, and white clover DM was only 0.03 Mg ha^{-1} with a high standard error because white clover establishment was spotty and many subsamples did not contain any white clover biomass. Clover DM accumulation increased rapidly after winter wheat harvest, supported by timely rainfall in July and August. At “35 d post-harvest”, red clover had increased its DM by a factor of three and white clover by a factor of ten. At the “October” sampling, six weeks after mulching, the mulched clovers yielded significantly less DM than those that were not mulched, yet all treatments had at least 1.4 Mg DM ha^{-1}. Unmulched red clover had 5.45 Mg DM ha^{-1}, the highest DM yield obtained for any forage legume during this study. These high DM yields were obtained despite low rainfall in September and October, probably because soil water was sufficient. Clover biomass production was also high in the spring (“April”), with 5.2 Mg ha^{-1} for the mulched red clover and the interaction between clover species and mulching was significant.

Mulching increased red clover DM by 1.52 Mg ha^{-1} but decreased white clover DM by 1.46 Mg ha^{1}.

The second cycle began in the drought year of 2012, and the lack of water had a devastating impact on both red and white clover. Very little biomass was produced with less than 0.8 Mg ha^{-1} at any sampling time. Red clover DM at “Wheat harvest” was 0.21 Mg ha^{1}, less than half of the first cycle’s DM. White clover was 0.01 Mg ha^{1}. Red clover DM had decreased at “October”, indicating plants died. White clover DM increased slightly until “October” to 0.22 Mg ha^{-1}. In the spring, some clover regrew after rainfall.

In the third cycle, forage legume species was significant at “Wheat harvest” and at “35 d post-harvest”, with red clover DM significantly higher than white clover, and alfalfa and sweet clover were intermediate. Forage legume DM yields at wheat harvest were much higher than in the previous cycles, due to above-normal precipitation between April and June. Alfalfa and sweet clover plants were as tall as the winter wheat, obstructing wheat harvest. Thirty-five days later, before mulching, dry matter weight had doubled for red clover, sweet clover and alfalfa, and quadrupled for white clover. At the “October” sampling time, mulching and type of clover were not significant. However, mulched red clovers had 0.62 Mg DM ha^{-1} and mulched white clovers 1 Mg DM ha^{-1} less than the unmulched red and white, respectively. Red clover (averaged across mulching) was significantly higher than white ($P = 0.005$) and sweet clover ($P=0.005$). In “April”, DM yield was below 0.8 Mg ha^{-1} for all forage legumes. Very cold winter temperatures and the lack of snow cover likely caused winter-kill. Mulching did not significantly impact clover DM, and neither did the interaction. However, the mulched white clover had about 0.01 Mg DM ha^{-1}, less than a tenth of the other forage legume species. Species had a significant impact with red clover (averaged across mulching) significantly higher than white clover DM.

Red clover DM at winter wheat harvest in the third cycle was similar to values obtained by a conventional study in Iowa (Blaser et al., 2011). Red clover DM values thirty-five days after wheat harvest in the first and third cycle were also similar to those found by Blaser et al. (2006, 2011). In October, unmulched red clover was 5.45 Mg ha^{-1} in the first, and 3.5 Mg ha^{-1} in the third cycle, higher than reported from a study on forage legumes undersown into winter wheat under organic management in France (Amossé et al., 2014). An organic study in Manitoba, with much less rain during the growing season, but also cooler summer temperatures, had red clover DM yields above 3.5 Mg ha^{-1} in two out of five site years (Cicek et al., 2014).

While producing less DM, white clover was observed to densely cover the ground by fall despite spotty initial establishment in the first and third cycle. Its stoloniferous growth habit enables it to produce lateral stems at an early age that grow along the soil surface, eventually becoming individual plants (Black et al., 2009). In the fall, however, unmowed white clover DM was 2.2 Mg ha^{-1} and 2.45 Mg ha^{-1} in the first and third cycle, respectively. Amossé et al. (2014) also found that undersown white clover had the lowest DM yields at wheat harvest, but by the fall, had 3.58 Mg ha^{-1}, outyielding red clover and alfalfa. White clover phyllochron is shorter and its leaf expansion faster than red clover's (Black et al.,

2009) which helps explain the relatively high white clover DM in the fall in our study, despite very low initial biomass weights.

Undersown alfalfa was a treatment in the study in France, and was the lowest performing forage legume, with only 1.36 Mg DM ha^{-1} (Amossé et al., 2014). In a conventional study in with fall-and spring planted forages in Alberta, fall-planted alfalfa had higher yields than fall-planted red clover one year after planting, but the alfalfa yields were no more than 1.2 Mg DM ha^{-1}, likely due to their semi-arid climate (Blackshaw et al., 2010b).

Undersown sweet clover was used in the study in Manitoba, where it did not produce more than 1.45 Mg DM ha^{-1} in any year, but yielded more than most cover crops planted after wheat harvest (Cicek et al., 2014). On the other hand, in Alberta, Blackshaw et al. (2010a) planted sweet clover into spring wheat in May, and terminated approximately 13 months later, when sweet clover had produced 10 Mg DM ha^{-1} in each of two years.

The rate of DM production can impact the ability of weeds to grow in a green manure stand. Undersown red clover that was well established with DM yields of approximately 1.5 Mg ha^{-1} at winter wheat harvest had a competitive advantage over weeds, effectively suppressing weed growth after wheat harvest (Anderson, 2015). In the third cycle, high forage legume yields at wheat harvest corresponded to very low weed DM. Likewise, in the first cycle, low clover DM at wheat harvest resulted in much higher weed DM in the fall. White clover always had low DM yields at wheat and might not be able to suppress weeds in its establishment year due to its slower growth rate (chapter 5).

Mulching in late summer lowered fall DM yields of red and white clover significantly in one out of two years, however, mulched red clover in the fall produced at least 2.9 Mg ha^{-1} similar to a study in Iowa, where red clover that was mowed (biomass removed) in late summer yielded up to 3.1 Mg ha^{-1} in October (Blaser et al., 2006). Mulching in late summer could have contributed to the death of the mulched white clover in the spring of the third cycle, because it removes carbohydrates in the plant, and thus lowers the plants' ability of winter survival (Anderson, 2015). Farmers may mulch forage legumes in the first year to destroy weeds and volunteer wheat and still obtain considerable red and white clover biomass yields, but if the green manure is to overwinter, mulching before September 1 is advisable to lessen the risk of winter kill (Anderson, 2015). Mulching returns the nutrients contained in the green manure to the soil, where they become available for the current or subsequent crops. Mowing for hay, on the other hand, removes the nutrients contained in the legume biomass, and must be weighed against the economic gains from the sale of hay.

Water availability likely had the greatest influence on green manure DM production. Early in the season, winter wheat used most of the available water, but as wheat matured and senesced, the fraction of soil water taken up by clover increased, whereas the fraction taken up by winter wheat decreased. Precipitation is especially important after wheat harvest, so that forage legumes can utilize full sunlight and soil nutrients.

In a study in North Dakota, sole-cropped red clover planted in May and terminated in October with DM yields between 2.3 and 4.3 Mg DM ha^{-1} used between 222 and 388 mm of total water (soil water and precipitation) and had an average water use efficiency (WUE) of 12 kg ha^{-1} mm^{-1} (Badaruddin and Meyer, 1989). In a study in Australia with different irrigation schemes and year-round clover growth, red clover and white clover WUE was 17.5 and 15.5 kg ha^{-1} mm^{-1}, respectively, because DM production was much higher (Neal et al., 2011). Using the red clover WUE value from North Dakota and unmulched red clover DM values observed in our study (figures 2.1), red clover water use in the fall of the first cycle was 454 mm. Precipitation between April and October was 575 mm, the same as the 30-year mean. Overwintered red clover produces new biomass in the spring, and total water used is the sum of fall water use and spring water use. Unmulched red clover in the spring of the first cycle used an additional 308 mm of water, for a total of 762 mm of water. The total precipitation (April 2011 – April 2012) was 753 mm, 29 mm less than normal. It is likely that red clover had emptied the soil water profile by April of 2012, with too little soil water for corn growth.

Water use in the third cycle was less because DM yields were lower, with 292 mm in the fall and an additional 71 mm in the spring, for a total of 363 mm (table 2.2). Precipitation between April of 2013 and April of 2014 was 643 mm, allowing for recharge of soil water. White clover WUE is slightly less than red clover's, but it also yields less DM, so total water use for white clover is smaller. Red and white clover have relatively low WUE, meaning they require more water to produce a unit of DM than other crops such as alfalfa which had a WUE of 15 kg ha^{-1} mm^{-1} in North Dakota (Badaruddin and Meyer, 1989).

Air temperatures in the North Dakota study were not given, but typically temperatures and thus evapotranspiration are higher in Nebraska, and more water is needed to produce the same amount of green manure biomass (Robinson and Nielsen, 2015). Using WUE values from North Dakota and green manure DM from our study, it is likely that the amount of total water needed between May and October to produce 1 Mg red clover DM ha^{-1} is at least 83 mm. Red clover yields of 3 Mg DM ha^{-1} and more in the fall were only achieved in our study in the first and third cycle, when precipitation from April to October was at least 550 mm, but this assumption need to be supported by future research.

The benefits of overwintering forage legumes such as winter ground cover and extended biomass production in the spring can aggravate potential drawbacks such as the legume's use of soil water. If soil moisture for the following crop is a concern, legumes should be terminated in the fall. If farmers desire winter ground cover and living roots in their fields, but want to limit the legume's soil water use, termination in early spring, as soon as the ground is workable, could be an option. Termination with an undercutter has been shown to preserve soil moisture as compared to termination with a disk (Wortmann et al., 2012).

Winter Wheat Grain Yields and Grain Protein

Winter wheat grain yields were not adjusted for moisture, but trailer samples at the grain elevator and on-field trailer samples at harvesting showed moisture to be between 12 and 15% (all treatments combined). Thus, wheat grain yield was not compared across cycles. No significant impacts of undersown forage legumes on wheat grain yield were detected in any of the years (table 2.4). However, while not significant, the difference between controls (highest-yielding) and white clover treatments (lowest-yielding) in the third cycle, was relatively large (0.6 Mg ha^{-1}) (table 2.6). Other authors also report little or no influence on winter wheat yields when undersown with red clover (Blaser et al., 2011, Amossé et al., 2013, Blackshaw et al., 2010b), white clover (Amossé et al., 2013) or alfalfa (Amossé et al., 2013, Blackshaw et al., 2010b), because winter wheat has a competitive advantage over the later-planted forage legumes. Competition between legumes and cereals can also be masked when winter wheat is fertilized with N, as was the case in the study by Blaser et al. (2011).

Wheat grain protein was significantly influenced by cycle and clover species, but not by the interaction between the two (table 2.4). Wheat grain protein was significantly different in each year, 11.62%, 10.84%, and 11.86% in the first, second and third cycle, respectively (table 2.6). Wheat undersown with white clover had significantly higher grain protein (11.51%) than wheat undersown with red clover (11.33%), albeit a small difference, but these treatments were not different from the sole-cropped wheat (the control) which had 11.47% grain protein. Blaser et al. (2011) did not find an effect of undersown red clover or alfalfa on winter wheat grain protein, but Amossé et al. (2013) found that winter wheat grain protein was reduced by undersown red clover and black medic (*Medicago lupulina* L.) in some site years.

Winter wheat grain yields were not influenced by undersown green manures in this study. Wheat grain protein was 0.18% higher in the white clover treatment than in the red clover treatment, but not significantly higher than the control treatment. No treatment reached the 12% grain protein necessary to market winter wheat as bread wheat. It is likely that little competition for soil water and nutrients occurred between green manures and winter wheat, because green manures had less than 0.5 Mg ha^{-1} at winter wheat harvest in each cycle, with the exception of red clover, alfalfa, and sweet clover in the third cycle. For the same reason, it is not likely that N was transferred from legumes to the wheat. Peoples and Baldock (2001) give a general rate of about 25 kg N that is fixed for each ton of forage legume DM, so the clover N fixation at wheat harvest was negligible in our study. More importantly, wheat grain yield and grain protein content are determined much earlier, between wheat tillering and anthesis, at which time clover plants had only 3 to 5 leaves (figure 2.2). By visual observation, during the time winter wheat and forage legumes were intercropped, most legume growth occurred after wheat matured and leaves dropped, increasing light transmittance to the undersown legumes (figures 2.3 through 2.5).

CONCLUSION

Green manures must produce high amounts of biomass, to provide sufficient N to the following corn crop, as well as fulfill other functions, such as weed control and winter ground cover. We tested four species of forage legumes undersown into winter wheat for their potential as green manures in a soybean-winter wheat-corn rotation under organic management. We hypothesized that (i) red clover DM production would be highest, (ii) mulching would impact red clover DM more than white clover DM yield, (iii) DM yield at fall termination would be higher than at spring termination, and (iv) winter wheat grain yields or grain protein contents would not be affected by undersown species.

In the two cycles with successful green manure establishment, red clover had the highest and white clover had the lowest DM yields at each sampling time. Mulching lowered clover DM for both species only in October of the first cycle. In April of the first cycle, mulching increased red clover DM but decreased white clover DM. In the first cycle DM yields were high at both termination times, but in the third cycle they were low at spring termination. Red clover produced at least 2.9 Mg DM ha^{-1} and white clover at least 1.4 Mg DM ha^{-1} at three out of six termination times. Winter wheat yields were not affected by undersown green manures, but wheat grain protein was slightly lower in winter wheat undersown with red clover than in winter wheat undersown with white clover, although not significantly different from a control.

Both red and white clover are vulnerable to low precipitation, especially during early summer. Since the frequency of drought years is predicted to increase in the Central Great Plains, alfalfa and sweet clover might be better choices as undersown green manures. However, in our study, they were less productive than red clover in years with at least 550 mm of growing-season precipitation. Apart from DM yields, green manure seed costs are likely to be a factor in species selection, an advantage for red clover and sweetclover, due to their much lower seed costs.

Forage legumes in our study were established by broadcasting in early spring and had high emergence rates. Frost-seeding at a rate of 50 or 75% of the rates used in our study is likely sufficient for good stand establishment, while also reducing seed cost. All seeds are available as organically certified seeds and as improved varieties.

We quantified green manure DM production four times during the growing season, illustrating legume biomass production lows and highs. This information is useful for farmers that grow green manures for the first time, helping determine whether a stand will be productive. This information could also be used as a starting point for future research investigating optimum times for green manure mulching and termination. Our study indicates that forage legumes mulched in late summer can still produce high DM yields in the fall, but we did not investigate the effect of different mulching times on DM production. The highest DM yields were obtained in the fall in each year thus fall termination lets farmers take advantage of high green manure yields while lowering the risk of yield loss of the following crop due to soil moisture deficits. Undersowing with forage legumes is an efficient method of establishing a green manure with high dry matter productivity in Eastern Nebraska. Red clover can be recommended as a high-yielding, cost-efficient green manure species, but research should be conducted to find more drought-resistant forage legumes.

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Table 2.1. Green manure seed information (purity, germination rate, percentage of hard seed, inoculant) as well as seed cost per hectare for each year.

	Purity %	**Germination %**	**Hard seed %**	**Inoculant**	**Seed cost** $ ha^{-1}
Green manure species			**First cycle**		
Red 'Marathon'	na	na	Na		183
White 'Rivendel'	66	82	9	Apex Green	356
			Second cycle		
Red 'Marathon'	100	63	24	Nitragin Gold	188
White 'Rivendel'	99	80	11	Prevail	356
			Third cycle		
Red 'Marathon'	100	90	5	Nitragin Gold	188
White 'Rivendel'	66	77	13	N-Dure & Apex Green	356
Yellow Sweetclover VNS	100	67	30	Nitragin Gold	109
Alfalfa 'Viking 3200'	100	81	10	na	233

Table 2.2. Total monthly precipitation and average daily air temperatures (average of daily nighttime low and daytime high temperature) for each month in each cycle, starting with the month of undersowing (March) and ending with the month of spring termination (April of the next year).

	Sum of precipitation				**Average daily temperature**			
	30-year mean	First cycle (2011-2012)	Second cycle (2012-2013)	Third cycle (2013-2014)	30-year mean	First cycle (2011-2012)	Second cycle (2012-2013)	Third cycle (2013-2014)
Month	Mm				˚C			
March	44	11	21	22	4.9	5.2	12.5	0.8
April	74	81	93	85	10.2	10.3	12.8	7.4
May	103	158	80	130	16.4	16.2	19.3	16.0
June	101	126	92	105	21.9	22.1	23.3	21.4
July	87	72	2	16	24.2	26.4	27.8	23.5
August	84	103	14	49	22.9	23.1	23.1	23.6
September	70	19	34	83	18.2	15.6	17.7	20.7
October	56	16	35	84	10.9	12.5	9.4	10.5
November	39	34	2	29	3.0	3.9	5.0	2.1
December	21	0	0	0	-3.7	-2.5	-2.5	-6.3
January	13	0	8	0	-5.8	-1.8	-4.7	-6.7
February	16	38	3	0	-2.3	-1.7	-2.3	-6.3
March	44	21	22	0	3.9	12.5	0.8	1.5
April	74	85	85	62	10.2	12.9	7.4	10.1

Table 2.3. Emergence of undersown green manures approximately seven weeks after broadcasting into winter wheat stands. Standard error is given in parentheses. Means within a column that are followed by the same letter are not significantly different from each other. Actual seeding rate (viable seed m^{-2}) is target seeding rate x %purity x %germination.

Green manure species	**Target seeding rate**	**Actual seeding rate**	**Emergence**	**Percentage of viable seeds emerged**
	Seeds m^{-2}	Seeds m^{-2}	Plants m^{-2}	%
			First cycle	
Red clover	1,300	992*	632 (93)	64
White clover	2,300	1239	400 (93)	32
			Second cycle	
Red clover	1,300	817	399 (70)	49
White clover	2,300	1,825	243 (70)	13
			Third cycle	
Red clover	1,300	1,167	1088 (83) a	93
White clover	2,300	1,162	522 (83) b	45
Alfalfa	1,100	888	926 (86) a	104
Sweet clover	1,300	869	772 (86) ab	89

*Because red clover seed information in the first cycle was missing, the average of the red clover actual seeding rate of the second and third cycle was used for calculating the %viable seeds emerged.

Table 2.4. Source of variation for green manure emergence, wheat yields and wheat grain protein content. D.f. = degrees of freedom. Alfalfa and sweet clover were not included in the cycle*species ANOVA.

	Numerator d.f.	Emergence	Wheat grain yield	Wheat grain protein
Green manure species	1	<0.001	0.543	<0.001
Cycle	2	<0.001	-	0.044
Species*cycle	2	0.026	-	0.102
Denominator d.f.		42	94	92

Table 2.5. Source of variation for green manure DM yields at each sampling time. Because sample sizes varied with sampling times, denominator degrees of freedom are presented. P-values are significant at α = 0.05 (Tukey test).

		P-values at biomass sampling times			
Source of variation	**Numerator d.f.**	**"Wheat harvest"**	**"35 d post-harvest"**	**"October"**	**"April"**
			First cycle		
Clover species	1	<0.001	<0.001	0.001	0.001
Mulching	1	-	-	0.015	0.935
Species x Mulching	1	-	-	0.142	0.003
Denominator d.f.		14	14	12	12
			Second cycle		
Clover species	1	0.001	0.006	0.112	0.004
Denominator d.f.		13	13	5	11
			Third cycle		
Clover species	1	0.03	0.012	0.061	<0.001
Mulching	1	-	-	0.149	0.063
Species x Mulching	1	-	-	0.674	0.090
Denominator d.f.		7	7	3	5
			Third cycle		
All forage legumes	3	0.045	0.052	0.002	<0.001
Denominator d.f.		21	21	15	21

Table 2.6. Winter wheat grain yield and grain protein at wheat harvest for each treatment. Wheat grain yield was combine-harvested at maturity, not adjusted for moisture. Wheat grain protein is adjusted to 12% moisture. Standard error of the mean is given in parentheses. Means followed with the same letter are not significantly different at α=0.05. Alfalfa and sweet clover were only used in the third cycle.

Treatment	**First cycle**		**Second cycle**		**Third cycle**	
	Grain yield	**Grain protein**	**Grain yield**	**Grain protein**	**Grain yield**	**Grain protein**
	Mg ha^{-1}	%	Mg ha^{-1}	%	Mg ha^{-1}	%
Winter wheat only (control)	3.65 [a] (0.082)	11.75 [a] (0.097)	3.64 [a] (0.205)	10.71[a] (0.090)	4.13 [a] (0.226)	11.96[a] (0.077)
Winter wheat-red clover	3.75 [a] (0.101)	11.48 [a] (0.117)	3.82 [a] (0.205)	10.81 [a] (0.090)	3.73 [a] (0.229)	11.71[b] (0.077)
Winter wheat-white clover	3.79 [a] (0.108)	11.64 [a] (0.117)	3.85 [a] (0.205)	10.99 [a] (0.090)	3.52 [a] (0.226)	11.91[ab] (0.077)
Winter wheat-alfalfa	-	-	-	-	3.58 [a] (0.226)	11.88 [ab] (0.077)
Winter wheat-sweet clover	-	-	-	-	3.69 [a] (0.226)	11.79 [ab] (0.077)
P-value	0.562	0.103	0.602	0.142	0.265	0.05

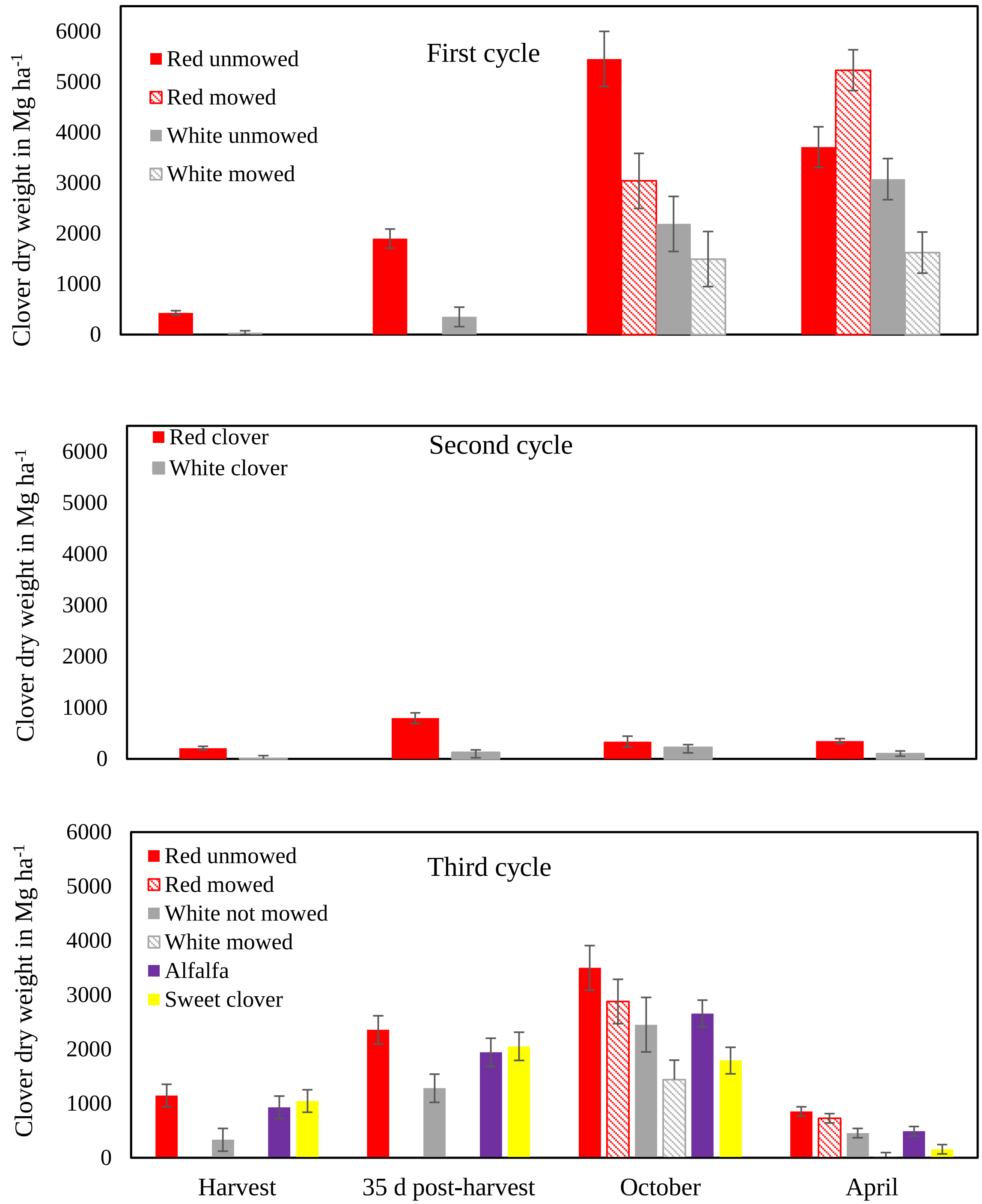

Fig. 2.1. Green manure DM production for each sampling time in each cycle. Mowed indicates clovers were mulched (mowed with plant matter left in place). “Harvest” – DM at winter wheat harvest, “35 d post-harvest” – DM at mulching time. “October” - DM before fall termination, “April”- DM before spring termination for overwintered green manures. Alfalfa and sweet clover were not mowed, and only used in the third cycle

Figure 2.2. Undersown clover and winter wheat on May 1, 2011 (5 weeks post-planting).

Figure 2.3. Clover and wheat at wheat flowering 11 weeks post-planting. White clover (top), red clover (below) with weeds (*Chenopodium album* L.).

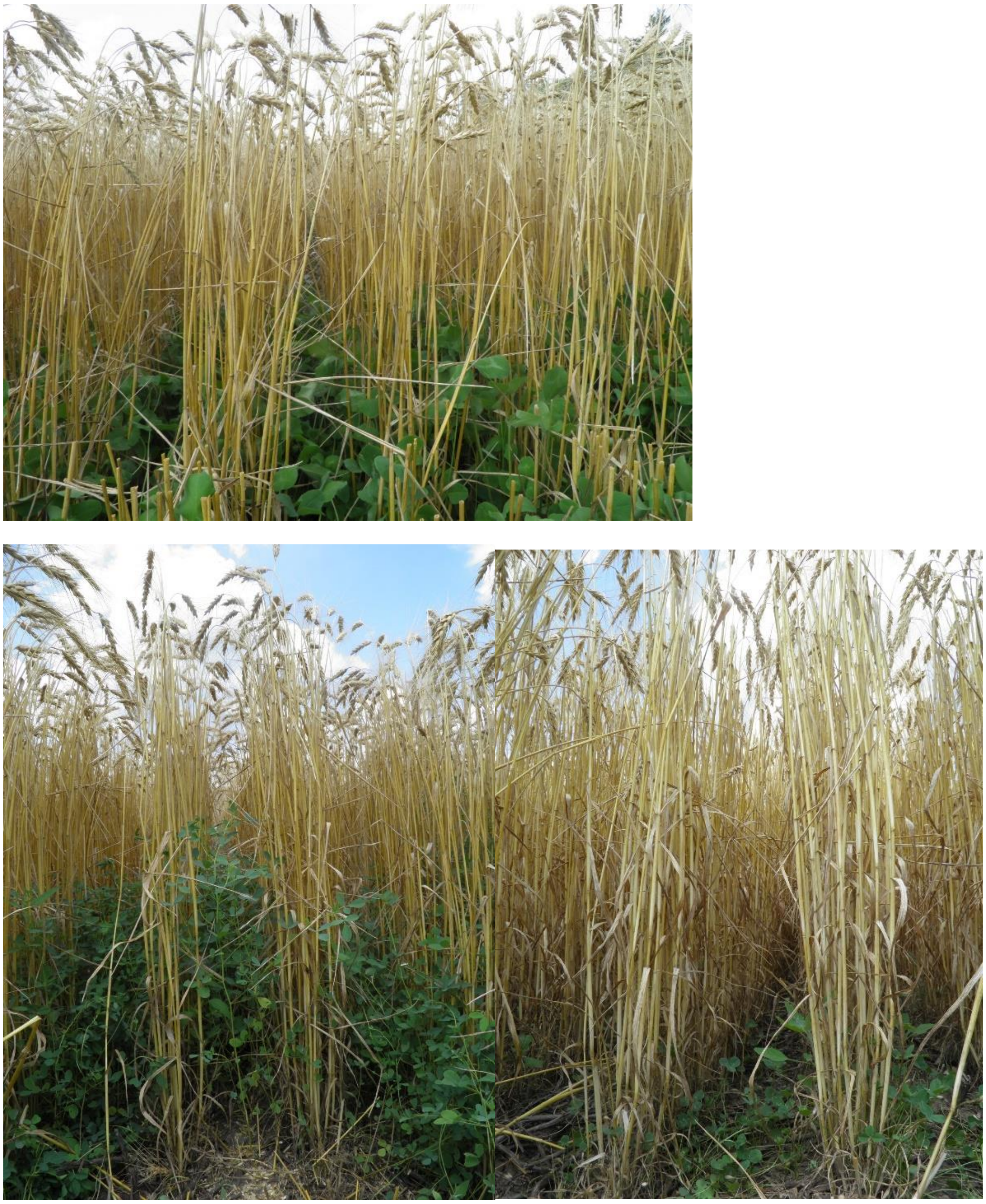

Figure 2.4. Green manures at wheat harvest on July 17, 2013 (18 weeks post-planting). Red clover (top), alfalfa (center), white clover (bottom).

Figure 2.5. Green manures after wheat harvest in August 2013. Alfalfa (top), red clover (below).

CHAPTER 3

SOIL NITRATE DYNAMICS FOLLOWING GREEN MANURES AND CATTLE MANURE IN AN ORGANIC GRAIN CROP ROTATION

An insufficient supply of plant available soil nitrogen is often cited as the main reason for lower yields in organic grain cropping systems (Berry et al., 2002; de Ponti et al., 2012). Nitrogen contained in the grain or straw is removed at harvest, and must be replaced to maintain or increase soil fertility as required by National Organic Program (NOP) standards. Organic farming systems principally rely on two types of soil amendments: animal manures and green manures. Green manures are usually leguminous crops because they can fix atmospheric nitrogen, and are a net nitrogen addition to organic systems. Animal manures, e.g. from cattle (*Bos Taurus*) are also used to provide N, however, this N originated from forage crops. On integrated farms, manuring constitutes a cycling, rather than net addition, of N contained in forage crops, but manuring can be viewed as a net N addition for farms that import manure.

Cattle manure is a commonly used animal manure in the Western Corn Belt and I will limit my discussion to this type of animal manure. Organic regulations mandate its application in composted form, with some exceptions (USDA, 2014). It is a reliable method to maintain or increase soil fertility as it can increase soil nitrogen, soil organic matter, and soil microbial biomass (Schröder, 2005). Nutrient concentration tables for dairy and beef manure are available from university extension offices, but a manure nutrient analysis before application is a more accurate assessment of mineral N (contained as ammonium [NH_4^+] in manure) and organic N, as well as other nutrients per unit. Typical nutrient contents of solid dairy manure at 46% dry matter (DM) are 0.15% NH_4^+-N and 0.7% organic N and total N application rates can be high with common application rates. Soil testing for nitrate is recommended before manure application to determine whether crops will respond to additional N (Koelsch and Shapiro, 2006).

Despite its benefits, manure is not used by all organic farmers because it can be expensive and difficult to obtain for farmers without livestock. Organic regulations still allow the use of manure from conventional sources on organic farms, but this can lead to unintended imports of antibiotic and pesticide residues contained in manure and/or animal bedding. Further, over-application of Na, K, Ca and Mg can occur with frequent manuring, increasing salinity of the soil. Organic farmers need alternatives to animal manures that build up or maintain soil nitrogen concentrations without the drawbacks of animal manures.

Green manures are crops grown solely for the purpose of improving soil fertility, usually by the addition of nitrogen and organic matter (Cherr et al., 2006). Two of the most important species of green manure plants for temperate climate zones are red clover (*Trifolium pratense* L.) and white clover (*Trifolium repens* L.) which were first domesticated in medieval Spain (Taylor and Quesenberry, 1996) and reached Northern Europe by the 18th century. In the severely nitrogen-limited farming systems of Europe, farmers soon realized the capacity of clovers to increase yields of subsequent crops and used them widely to replace fallows in the rotation as well as supply high-quality forage to livestock.

In the 20th century, with the advent of synthetically fixed nitrogen, their use began to decline (Kjærgaard, 2003). Yet in low-input and organic farming systems, clover green manures could be reintroduced to improve N nutrition of subsequent crops (de Ponti et al., 2012). Red and white clover can be established by undersowing into winter cereals in early spring. Most clover biomass production occurs after cereal harvest, during a period in which the field would otherwise be fallow (Gaudin et al., 2013). They are winter-hardy and can be plowed under either in the fall of the establishment year or spring before the next crop, making them a better choice than summer annual cover crops which can be hard to establish during hot and dry weather in the Western Corn Belt ecoregion.

Nitrogen fixation of legumes can vary widely depending on legume inoculation, nutrient status of the legume and soil mineral N concentration (Peoples et al., 2012). High soil N concentrations inhibit nodulation and increase the uptake of soil N relative to atmospheric N (Downie, 2014). Three factors are important to understand how much N was added to the soil by a green manure: The weight of the above-ground biomass (DM), the percentage of N in the legume above-ground biomass (%N in DM), and the fraction of above-ground biomass N that was derived from the atmosphere (%Ndfa). The total amount of N from fixation (Ndfa) in above-ground biomass is:

$$\text{DM (kg ha}^{-1}) \times \text{\%N in DM} \times \text{\%Ndfa} = \text{Ndfa (kg ha}^{-1})$$

Estimates of the amount of % Ndfa can be determined experimentally with ^{15}N natural abundance methods. Because the abundance of this stable isotope is slightly higher in the soil than in the atmosphere, legumes deriving N from the atmosphere will have a different tissue $^{15}N/^{14}N$ ratio than plants taking up N only from the soil solution (Carlsson and Huss-Danell, 2003). However, for farmers, it is difficult to determine the Ndfa of a legume green manure. It is highly correlated to green manure DM production (Carlsson and Huss-Danell, 2003) and as a general rule Peoples and Baldock (2001) estimate 25 kg N fixed for every Mg of forage legume DM. Most of the research on clover N fixation comes from studies where they are grown as forages or leys, sometimes in grass-clover swards, over several years. In these systems, clovers are mowed three to six times a year, and DM yield is the sum of the DM produced at each mowing, frequently resulting in amounts of Ndfa above 100 kg ha^{-1} $year^{-1}$ (Carlsson and Huss-Dannell, 2003). Mowing can be necessary to control weeds and improve plant vitality but if the mowed residues are allowed to remain on the surface (mulching), they will return N to the soil and can lower biological N fixation rates (Hatch et al., 2014).

The amount of Ndfa is likely different in a cereal-undersown green manure system, because growing periods are shorter and plants are usually not mowed. Due to the cereal's competitive advantage at the time of green manure planting, clover DM production and thus N accumulation is very slow until wheat harvest. Schipanski and Drinkwater (2011) found that red clover undersown into winter wheat had a greater %Ndfa than monocropped red clover and red clover undersown into taller cereals such as spelt (*Triticum aestivum var. spelta),* fixed less N. Table 3.1 lists recent research findings on %Ndfa and total Ndfa from red and white clover in organic systems.

A high amount of N at the time of green manure or cattle manure incorporation is important but the release of N from the organic fertilizers must be synchronized with crop N demand to avoid N deficits. Mineralization, the microbially driven process of converting organically bound N to mineral N, is dependent on the C and N content of the organic matter which the mineralizing bacteria and fungi use for energy and growth. The end product of mineralization is NH_4^+, but it will only be released once microbes meet their own N needs, and at high C/N ratios microbes take up mineral N from the soil solution to synthesize protein, thus immobilizing mineral N in their biomass. For this reason, materials with a C/N ratio below 25 will typically decompose quickly and release N, whereas those with a C/N ratio above 25 will lower soil mineral N concentrations (Robertson and Groffman, 2007). Nitrogen-rich legume tissue usually decomposes fast, for example Stute and Posner (1995) and Dou et al. (1995) found that red clover had released 50% of its N four weeks after incorporation in the spring. Corn has the highest N demand about 60 days after planting (Pang and Letey, 2000), and termination of green manures in the spring might improve the matching of N release and corn N uptake as compared to fall termination. On the other hand, fall termination of green manures is sometimes necessary to allow the recharge of soil moisture for the cash crop (Unger and Vigil, 1998). Cattle manure releases about 25 to 50% of its organic N in the year after application, and at lower rates in the following years (Koelsch and Shapiro, 2006), increasing mineralization rates in frequently manured fields (Schröder, 2005).

Synchronizing crop N demand with fertilizer N mineralization rates not only prevents N deficits in the crop, but also loss of N. Ammonium released during mineralization of green and animal manures is either taken up into microbial biomass (see above), adsorbed to clay minerals and soil organic matter, taken up by plants, converted to ammonia (NH_3) or converted to nitrate (NO_3^-) which is the dominant form of mineral N in temperate agricultural soils (Schachtschabel et al., 1998). Nitrate is water soluble and becomes part of the soil solution. It can be taken up by soil microbes or plants, but if crop demand is lower than N supply, nitrate can be lost either through leaching into lower soil layers or through denitrification into N gases. Leaching losses are highest after rainfall (Wick et al., 2012) and are public health and environmental concerns as nitrate becomes a pollutant, contributing to unsafe nitrate levels of groundwater (Exner et al., 2014) as well as eutrophication of surface waters including the Gulf of Mexico (Diaz and Rosenberg, 2008). Denitrification is an anaerobic process and occurs when water-filled pore space of soils exceeds 60% (Linn and Doran, 1984), for example after rainfall or flooding. Incorporation of green manures can lead to gaseous N losses due to denitrification and leaching (Gardner and Drinkwater, 2009). Animal manure is more prone to gaseous N losses in the form of ammonia, especially when it is surface applied, because of the high ammonium content of manures (Schröder, 2005). Leaching rates after spreading animal manure are typically higher than after green manure because of much higher rates of total applied N. For soybean (*Glycine max* L.)-winter wheat (*Triticum aestivum* L.)-corn (*Zea mays* L.) rotations in the Western Corn Belt, the period between corn harvest and soybean planting is most vulnerable to nitrate loss, followed by the period between wheat harvest and corn planting. Efforts to reduce nitrate loss include cover cropping and changes in tillage and/or fertilizer application rates and need to be targeted to these periods (Syswerda et al., 2011).

Optimum soil nitrate levels during crop growth are important in all farming systems, but under organic management, deficient N levels are harder to correct and excessive N levels lead to a greater relative loss because of the longer time it takes to either "grow" N by using legume green manures or cycle N by using animal manure. Attempts to quantify overall N additions of either green manures or animal manures are useful in selecting replacements for mineral fertilizers in conventional systems, but they are not easily transferable to studies under organic management seeking replacements for cattle manure. Nitrogen in both animal manure and green manure is subject to mineralization before it becomes plant available, but the length of time over which N becomes available varies depending on the amount and quality of C as well as the C/N ratio of the fertilizer. Further, as many researchers have pointed out, several factors influencing decomposition are different in organic soils, for example, soil organic matter under organic management is higher (Marriott and Wander, 2006), total N is higher (Liebig and Doran, 1999), and microbial activity is enhanced (Mäder et al., 2002). These differences likely stem from the continuous input of high amounts of carbon-rich organic fertilizers, including animal manures and plant residues and as a consequence, carry-over or residual effects of previous animal manure or green manure applications are high, potentially confounding effects of first-time usage of organic fertilizers such as green manures (Gentry et al., 2013). Research that follows organic fertilizers for more than one season is needed to distinguish between actual treatment effects and residual effects of previous fertilization. In addition, since the most practical time to apply either animal or green manure in soybean-winter wheat-corn rotations is after wheat harvest, once in three years, we need to understand the soil nitrate dynamics over the course of the entire rotation, until they are applied again, to be able to prevent soil fertility issues. Thus, investigating long-term effects of organic fertilizers on soil nitrate are essential in avoiding either loss of N (through leaching or denitrification) or crop deficits of N (through asynchrony of soil N supply and crop N demand).

With our research, we wanted to determine whether undersown green manures can be viable alternatives to cattle manure in their ability to increase soil nitrate levels for the subsequent corn crop as well as for other crops over the whole rotation. We further wanted to investigate whether soil nitrate levels at corn harvest and subsequent crop harvests differ for cattle or green manure, which can be useful to determine the potential for nitrate loss in this system. We hypothesized that (i) red clover increases soil nitrate more than white clover; (ii) clover mulching lowers soil nitrate levels versus not mulching; (iii) terminating in the fall versus in the spring increases soil nitrate levels at corn planting; (iv) at corn planting, soil nitrate is highest under cattle manure, intermediate under red and white clover green manures and lowest under post-wheat soybean cover crop and control treatments; (v) over the course of a whole rotation, cattle manure increases soil nitrate levels over a longer period than green manures.

MATERIALS AND METHODS

This site is located near Mead, NE, in the Western Corn Belt Plains ecoregion. All fields used in this study are organically certified, in a soybean-winter wheat-corn rotation with a history of cattle manure applications after winter wheat harvest. For a description of the site, soils and climate see Chapter 1. Experiments were carried out in three cycles, each cycle in different fields according to the crop rotation: field 789 in 2011 (first cycle), fields 3 and 56 in 2012 (second cycle), and field 14 in 2013 (third cycle) (figure 1.1). Soil samples were taken until 2014, so that soil nitrate data is available for one rotation for the first cycle, for two years for the second cycle and for one year for the third cycle. The experimental design was a CRD in the first cycle, and a RCBD for the other cycles. Treatments were types of organic soil fertility amendments: undersown green manure (red clover or white clover), post-wheat cover crop (soybean green manure), cattle manure, and no fertility amendment (control). All soil fertility amendments were applied in the wheat phase of the rotation: the clovers were undersown into winter wheat in March, soybean green manures were planted in July after winter wheat harvest, and cattle manure was applied between winter wheat harvest and November (table 1.1). Undersown red clover and white clover received two other randomly assigned treatments: mulching and time of termination. Half of the clover plots were mowed with the plant residues remaining on the surface (mulching) once 40 days after winter wheat harvest and half of the plots were not mulched. Clover plots were split and either terminated in the fall of the first year or the following spring about two to three weeks before corn planting (see table 1.2 for sampling times). Clover plots were terminated by disking twice (Keewanee 1010 disk, Kewanee, IL). Table 1.1 contains dates of all management operations carried out on the plots. It should be noted that tillage operations are not the same for each treatment in this experiment. Manure and nothing plots were tilled two to three times more than the green manure plots. Tillage has a well-established positive impact on soil N mineralization, and confounds the results of soil nitrate testing. However, our research results are intended to help organic farmers make management decisions, and thus our tillage operations were selected to reflect operations typical for organic farmers.

To assess the amount of N accumulated by the green manures (clovers and soybean), the percentage of C and N contained in the above-ground green manure DM were measured at termination in the fall of 2011 and spring of 2012 (first cycle). The full analysis can be found in Shi (2013). These values are multiplied with the above-ground DM weight at fall or spring termination (table 3.2). The first-year N values are also used in the other years to estimate N contained in the above-ground biomass. A dairy manure nutrient analysis from 2008 (Midwest Laboratories, Omaha) from the same dairy research farm that provided the manure in the first cycle was used to obtain N and C content of the dairy manure. Beef manure was used in the second and third cycle, and published values were used for C and N content. Estimated N available for corn in the first year were 112 kg N ha^{-1} (dairy manure) and 196 kg N ha^{-1} (beef manure) (table 3.2a).

Soil nitrate was measured at planting and harvest of each crop, including the green manures crop, to follow the seasonal dynamics of nitrogen (table 1.2).

Sampling times were “Cloverplanting” (within four weeks after undersowing clover into wheat), “Wheatharvest 1” (within two weeks after wheat harvest and before the other soil amendments were applied), “Fallkill” (at the time of clover termination in the fall), “Springkill” (at spring termination, data not shown), “Cornplanting” (same day corn was planted), “Cornharvest” (within one week after corn was harvested), “Soybeanplanting” (three weeks after soybean was planted in the first cycle, and two month after soybean was planted in the second cycle, because soils were too wet for sampling). “Soybeanharvest” and “Wheatharvest 2” (three years after the first wheat harvest) were only taken in the first cycle.

Soil was sampled by randomly taking five cores (in large plots) or three cores (in small plots) with a JMC Backsaver soil sampler with a 0.02 m diameter stainless steel probe (Forestry suppliers, Jackson, MI) or with a hydraulically operated stainless steel probe with a 0.03 m diameter. Soil was sampled by pushing the probe to a depth of 0.2 m and collecting this soil in a bucket. The probe was then inserted in the same hole to a depth of 0.6 m, and soil from this depth was collected in a separated bucket. Soil was air-dried and sent to Ward Laboratories (Kearney, NE) for analysis. Samples were extracted with calcium phosphate and analyzed for nitrate with a flow injection analyzer (Lachat Instruments, Milwaukee) (Ward Laboratories, Kearney).

Statistical analysis was carried out using SAS 9.4 (SAS Institute, Cary, NC). Each field was analyzed separately for the duration of the study. To compare only the effects of the two clovers, analysis of variance was first conducted with the red and white clover only. The effects of type of clover, mulching and time of termination were analyzed for each sampling time separately. Proc MIXED Method=type 3 was used for the ANOVA with split-plots and PROC GLIMMIX for all others. Then, analysis of variance was carried out for all soil amendments (clover [red and white combined], manure, soybean green manure and control) over the course of each cycle. Because the same plots were sampled repeatedly, sampling time was modeled as a repeated measure. Several covariance pattern models were tested for best fit with AICC and the first-order ante dependence model was selected as it was the most parsimonious. Multiple mean comparison with a Tukey test at a significance level of 0.05 was carried out with PROC GLIMMIX (see appendix for SAS code). Soil nitrate was analyzed separately for the upper soil layer (0 – 20 cm) and the lower layer (20 – 60 cm).

RESULTS AND DISCUSSION

Legume Dry Matter N and Estimates of N Derived from the Atmosphere

Red clover, white clover, and soybean green manure above-ground DM N and C/N can be found in table 3.2. At fall termination in the first cycle (2011), soybeans contained much more N in their above-ground DM than either red or white clover, however, some N transfer from the shoot to the roots had likely already occurred in the clover to prepare for winter dormancy. In the spring of 2012, both clovers contained more than 100 kg N ha^{-1} in their above-ground DM.

Only fresh clover biomass was sampled in the spring and fall, so any dead clover plant material was not included in the DM weights.

The DM portion of N that was derived from the atmosphere (%Ndfa) was not measured in our study. Values of %Ndfa from the literature on legume green manure N fixation in organic systems are in table 3.1. For red clover, these values range from 53% to 89%, and for white clover from 71 to 91%. In the fall of the first cycle, when the study's highest clover DM was measured, fall-terminated red clover can be expected to contain approximately 55 to 90 kg ha^{-1} and white clover 37 to 46 kg ha^{-1} of fixed N in its above-ground dry matter. With high biomass production and higher N content in the spring of 2012, red clover could have added between 50 and 111 kg N ha^{-1} from shoots alone. Assuming that only about 50% of the amount of N fixed by red clover is available for the subsequent crop (Stute and Posner, 1995, Dou et al., 1995) with the remainder of the N becoming available later or entering the stable pool of N in the soil, even the high range of N fixed by red clover would only provide about 55 kg N ha^{-1} for the following corn crop.

Soil Nitrate Changes in the First Cycle

Soil nitrate changes following red and white clover in the first cycle

In the first cycle soil nitrate levels were measured for one rotation (2011 – 2014). The effects of clover are discussed first, and then compared with the effects of the soybean green manure and cattle manure.

Table 3.3 shows the P-values for the test of the effects of type of clover, mulching and termination time on soil nitrate at each sampling time in the first cycle. Figure 3.1 shows the seasonal soil nitrate concentrations for the mulching x clover simple effects for the first cycle. Soil nitrate was similar at all sampling times, except corn planting and corn harvest. Type of clover was only significant at corn harvest, caused by the high value of the red mulched clover. Mulching clover decreased soil nitrate at fall termination by 2 ppm, possibly because clover residue released soil nitrate which in turn reduced biological N fixation which was also observed by Hatch et al. (2014). Mulching increased soil nitrate by 6 ppm in the red clover at corn harvest. In our study, higher soil nitrate at corn harvest in mulched red clover plots was probably caused by lower corn nitrate uptake in these plots. Drought conditions during 2012 affected corn more in plots that previously had highly productive clover stands such as mulched red clover plots (see chapter 2), probably leading to soil moisture deficits (Unger and Vigil, 1998), and corn growth was severely stunted in these plots.

Fall-terminated clover plots had 5 ppm more soil nitrate than spring terminated clover plots at the time of corn planting which was the largest effect of any treatment, and close to significance (table 3.3). Fall-termination increases the time for mineralization, but also for N losses. The spring of 2012 was very warm (figure 2.2.), which likely resulted in rapid mineralization for spring-terminated plots. At corn harvest, fall-terminated plots had 4 ppm less soil nitrate than spring-terminated plots (8 ppm versus 12 ppm), and spring-terminated mulched red clover had 22 ppm of soil nitrate, 10 ppm more than the next-highest treatment (table 3.3). As stated above, this is likely confounded by the water use of the highly productive red clover stands at spring-termination in 2012. Soil water deficits likely reduced corn growth and thus uptake of N, and possible also N mineralization rates, compounded by drought conditions during the summer months. At soybean planting, more than one year after the incorporation of the spring-terminated clovers, these plots still had significantly higher soil nitrate (12 ppm) than the fall-terminated plots (10 ppm).

Soil nitrate concentrations in the lower soil layer (20 – 60 cm) were not affected by type of clover or mulching at any sampling time. Termination time was significant at corn planting (table 3.4), with soil nitrate concentrations of 21 ppm in fall-terminated plots versus 18 ppm in spring-terminated plots. Data for red and white clover is shown combined (clover) in figure 3.2 (see below).

Soil Nitrate Changes Following Dairy and Green Manures in the First Cycle

At both the 0 – 20 cm and 20 – 60 cm depth, dairy manure had significantly higher soil nitrate concentrations than clover at three of five sampling times between corn planting and the second wheat harvest (table 3.5). During the same time, clover soil nitrate levels were never significantly different from the control. Variability in soil nitrate was higher between sampling times than between treatments, indicating the seasonal changes caused by weather, crop use, and field management of soil nitrate.

Soil nitrate values for all soil amendments (manure, clover, soybean green manure and control) over the course of one rotation are shown for the depth of 0 –

20 cm and 20 – 60 cm in figure 3.2. The type of soil amendment, sampling time and their interaction were highly significant (table 3.5). Red and white clover values are combined because they were not significantly different from each other (see above), but are shown separate from the soybean green manure, because of their differences in agronomic management and botanical characteristics.

At wheat harvest in July 2011, clover was already growing, but the other treatments had not yet been applied. At this initial soil test, there was a small (4 ppm) but significant difference between the future soybean cover crop plots and the future control plots. At fall termination manured plots had soil nitrate levels about one magnitude higher than either green manure type. Manure contained about 56 kg of ammonium (NH_4^+ -N) ha^{-1} which was quickly converted to ammonia (NO_3^-) in the soil, although loss of ammonium in the form of ammonia between spreading and disking (incorporation) is likely. For example, about 50% of the total ammonium was lost as ammonia emissions between application and incorporation of cattle manure over the course of 120 hours (Webb et al., 2012). Time between application and incorporation was not given, but Laboski et al. (2013) found that 75% of the total ammonia lost from surface-applied cattle manure was emitted within six to eight hours after application. Control plots also had at least 16 ppm more soil nitrate than the green manure plots. Manured and control plots were kept free of vegetation by disking which likely accelerated mineralization of N from wheat residue and/or manure and soil organic matter and nitrate accumulated. In contrast, living clover plants were taking up N from the soil solution in the clover plots. While red and white clover can meet up to 86 and

91%, respectively, of their N needs by biological N fixation via the symbiosis with rhizobia (table 3.1), this is an energy-consuming process for the plant and it will thus preferentially take up nitrate from the soil solution (Peoples and Baldock, 2001). Small amounts of N become available when nodules or plant roots die off, or release compounds such as exudates, lysates and ions (rhizodeposition) however, once this N is mineralized it is available for uptake by microbes or plants (Wichern et al., 2008). Compared to manured or control plots, green manure plots likely had lower nitrate leaching losses as they took up N into their biomass.

In May of 2012 at corn planting, soil nitrate was above 20 ppm for all treatments in the upper layer, the highest measured concentration during this rotation. The increase of soil nitrate from fall-termination to corn planting was highest for the green manures ($P < 0.001$) indicating rapid decomposition after incorporation. As expected, soil nitrate in the manure treatment was highest, reflecting the high amount of N applied with manure (see above) compared to modest amounts of N contained in the green manure biomass. Tillage was again different for the treatments, with all but the spring-terminated clover plots receiving one additional disking and field cultivation. Soybean green manure had a lower C/N ratio than clover at incorporation in the fall which accelerated mineralization and likely explains the higher soil nitrate in May in soybean green manures than clover (table 3.2). Low soil nitrate under clover is in contrast to other findings which report a soil nitrate peak four weeks after red clover spring incorporation that was several fold higher than a control (Dou et al., 1995).

Amossé et al. (2014) found that 12 weeks after spring incorporation, soil nitrate levels after red clover were 40% higher and after white clover 53% higher than after a control.

The summer of 2012 was warm and dry which affected corn yields and could have also decreased mineralization rates from green manures. May was 3 ˚C warmer, June 1 ˚C and July 3 ˚C warmer than normal, and precipitation during that same time was 174 mm, 117 mm below normal. Soil moisture was not measured, but it is likely that there was a soil moisture deficit after clovers, contributing to low corn yields in all but the control and manure plots (see chapter 4). Soil nitrate decrease between corn planting and corn harvest was highly significant for all treatments ($P < 0.001$), but the difference was highest for manured plots with 41 ppm. While uptake by the crop accounts for some of this, yields of manured corn were similar to those of the unfertilized corn, which only had a soil nitrate difference of 17 ppm between corn planting and harvest. Loss of N by denitrification is not likely, since soil moisture was probably too low for denitrification (Linn and Doran, 1984). However, reduced evapotranspiration of the drought-stressed crop could lead to more deep percolation of soil N after precipitation events (Pang and Letey, 2000).

During the remainder of the rotation, soil nitrate levels were lower and less variable. The time between corn harvest and soybean planting is vulnerable to nitrate leaching (Syswerda et al., 2011) and manured plots lost 12 ppm of soil nitrate during this period, but this was not significant ($P = 0.375$). Soil nitrate levels between soybean planting and soybean harvest were similar for all treatments, indicating biological N fixation as a source of N for this crop. Winter wheat is immediately no-till planted after soybean harvest, so N being released from decomposing soybean residue is likely taken up quickly. Soil nitrate levels at wheat harvest the following July are only 2 – 4 ppm lower than at soybean harvest. Between the first wheat harvest in July of 2011 and the second wheat harvest in July of 2014 soil nitrate decreased slightly, but significantly for clovers ($P < 0.001$), but not for the other treatments.

Soil nitrate in the 20 to 60 cm layer was lower but showed a similar trend as the upper layer with the highest soil nitrate concentrations in the manure treatment (figure 3.2). Treatment and sampling time were significant, as was their interaction (table 3.5). Soil nitrate is highly mobile and moves with precipitation from the upper to the lower layers, from where it is either taken up by plant roots or leached. The decrease in soil nitrate between corn planting and corn harvest was significant for each treatment ($P < 0.001$), ranging from 38 ppm in the manure plots to 18 ppm in the clover plots, reflecting the high nitrate uptake from this layer by corn. Between corn harvest and soybean planting, soil nitrate increased significantly under clover ($P < 0.001$). It is possible that N released from continuing green manure mineralization in the upper layer accumulated in the lower soil layer after corn growth ceased. After one rotation, soil nitrate concentrations at the second wheat harvest were lower for all treatments than at the first wheat harvest and this difference was significant for clover ($P < 0.001$) and manure ($P = 0.020$).

Soil Nitrate Changes in the Second Cycle

This cycle was impacted by drought conditions with above-normal temperatures and very little precipitation for much of the clover's growing season (2012) resulting in very little clover DM production and hence very little N accumulation in the clover treatments, but the soybean green manure still produced 118 kg N ha^{-1} in its above-ground DM (table 3.2). Since biomass production was very low, the effects of clover mulching and time of termination were not evaluated.

Soil nitrate levels under red clover were not significantly different from those under white clover in both layers (table 3.6), and they are shown combined as Clover in figure 3.3. The effects of treatments, sampling time and their interaction were significant for each layer (table 3.5). For the 0 – 20 cm soil depth, the highest soil nitrate concentrations were measured at fall kill, with manured plots having the greatest, and green manure plots the lowest soil nitrate levels. The difference in soil nitrate levels between the green manures and the control was probably mostly due to tillage in the control plots, rather than plant uptake, since little biomass was present in the green manure plots (see chapter 1). Nitrate levels decreased between fall kill and corn planting for the manured and control plots ($P < 0.001$), reflecting N lost to denitrification and/or leaching, either to the 20 – 60 cm layer, or deeper in the soil profile. Nitrate under the green manures increased significantly between fall termination and corn planting ($P < 0.010$), but at much lower concentrations than in the cycle started in 2011, likely reflecting the low biomass production and N accumulation in the green manures.

Nitrate levels in the 20 – 60 cm layer were highest at corn planting, with values similar to those in the layer above, from which much of the nitrate originated. For the remainder of the sampling season, soil nitrate levels under all treatments were low and similar among treatments and layers.

Soil Nitrate Changes in the Third Cycle

The effect of type of clover, mulching (if applicable) and termination time (if applicable) was tested separately for each sampling time (table 3.7). Since none of the treatments or interactions were significant, red and white clover were combined across mulching and termination time and entered as Clover in the comparison with the other organic soil amendments. Type of amendment, sampling time and their interaction were significant for both soil depths measured (table 3.5).

This cycle had sufficient moisture for high clover biomass production, but the soybean cover crop failed. Very cold winter temperatures and lack of snow cover in the winter of 2013/2014 reduced clover survival, with very little clover biomass at spring kill. Soil nitrate was measured only until corn planting, when it was low, but mineralization was likely delayed due to the cold winter and spring.

At the 0 – 20 cm soil depth at wheat harvest, clover plots were significantly lower in soil nitrate than the other plots (no other treatments had been applied yet, so all other plots were “controls”) which could be the result of high clover DM production (see chapter 1) and subsequently high clover soil nitrate uptake (Figure 3.4). In the 20 – 60 cm layer, soil nitrate levels were below 1 ppm for all treatments, probably due to uptake by winter wheat. In contrast to the previous years, soil nitrate in the upper layer between wheat harvest and fall termination did not increase under the manure treatments and decreased under the control treatments ($P = 0.033$). In the lower layer, it was below 1 ppm for all treatments.

All treatments increased soil nitrate significantly between fall termination and corn planting in the upper layer ($P < 0.02$ for each treatment) but at corn planting, soil nitrate levels overall were lower than in previous years. Very cold winter temperatures and a cool spring might have slowed mineralization rates compared to previous years (table 2.2). In the upper layer, soil nitrate after clover was significantly higher than after the other treatments which were not significantly different from each other. Values for soil nitrate were between 4 and 6 ppm in the lower layer, with clover treatments significantly higher than control treatments. Low soil nitrate after soybean green manures was likely caused by its very low N accumulation (table 3.2). The insignificant effect of the manure treatment was somewhat puzzling. It is possible that manure was applied at lower than assumed rates or that nitrogen concentrations in the manure were lower than in previous years.

N Availability for Corn

This site has a history of cattle manure applications every three years and residual effects (along with tillage) probably explain high soil nitrate concentrations even under control treatments. For example, 4% of beef manure is available three years after application (Koelsch and Shapiro, 2006) and with manure applications spanning several decades, residual effects are high (Schröder, 2005). Corn might not respond to the application of a fertilizer lower in N (such as green manure), if mineralization rates from previous manure applications are sufficient for high yields. It might take several rotations without cattle manure applications to separate the effects of the control treatment or green manure treatments (Schröder, 2005). Soil nitrate concentrations in the upper 0.3 m of the soil layer, taken when corn is about 0.3 m tall (in early June or time of pre-sidedress), are correlated to corn yields, and used to calculate N fertilizer needs of corn (Magdoff, 1991). If soil nitrate levels measured with the Magdoff test (or pre-sidedress nitrate test) are between 20 and 30 ppm no additional N is normally necessary. Thus, soil nitrate in the first cycle at corn planting was likely sufficient under all treatments, even before most of the green manure and dairy manure had mineralized (figure 3.2). In the second cycle, soil nitrate at corn planting was above 20 ppm only in the manure treatment (figure 3.3), and in the third cycle, it was below 20 ppm in all treatments (figure 3.4). But mineralization probably increased substantially by early June during warm and moist weather conditions observed in the spring of the second and late spring of the third cycle. Soil nitrate sampling during corn growth, along with corn tissue N sampling, would have allowed us to better understand the interactions between fertilizer N mineralization and corn N uptake.

While corn yields suffer if soil nitrate is too low, large-scale environmental damage occurs when soil N supply is greater than crop N demand, for example during periods of fallow or bare soil, leading to loss of mineral N through leaching or denitrification (Crews and Peoples, 2005). While all green manure biomass N has to be mineralized to be available to the plant, manure N is both in the organic and NH_4^+-N (ammonium) form.

In our rotation, the period between manure application and corn planting likely has potential for denitrification and leaching because of the high amount of N applied, coupled with a lack of N uptake by plants and frequent tillage over a period of 9 to 10 months. Manured plots, which received 56 kg NH_4^+-N ha^{-1} in the first and 134 kg NH_4^+-N ha^{-1} in the second and third cycle, showed a spike in soil nitrate levels after cattle manure application in the fall likely due to nitrification of ammonium. Leaching of nitrate in organic manure-based grain systems needs to be the focus of more research, as minimizing leaching not only reduces pollution, but also economic losses for farmers.

CONCLUSION

In this study, we tested four hypotheses:

(i) Red clover increases soil nitrate more than white clover; (ii) Clover mulching lowers soil nitrate levels versus not mulching; (iii) Terminating in the fall versus in the spring increases soil nitrate levels at corn planting; (iv) At corn planting, soil nitrate is highest under cattle manure and lowest under control treatments; (v) Over the course of one rotation, cattle manure increases soil nitrate levels over a longer period than green manures.

We found that (i) Red clover did not have a different effect on soil nitrate than white clover; (ii) At fall-kill in the first cycle, mulching lowered soil nitrate by 2 ppm, a small, but significant difference. At corn harvest in the first cycle, mulching significantly increased soil nitrate by 6 ppm (54%), and mulched red clover had at least 8 ppm more soil nitrate (at least 50%) than any other treatment. These effects were likely confounded with high soil water use of mulched clovers that led to reduced corn growth and subsequently reduced soil nitrate uptake; (iii) Termination time did not have a significant effect at corn planting, but spring-termination significantly increased soil nitrate by 4 ppm (36%) at corn harvest. This is also likely confounded with clover water use and subsequent reduction of corn growth ; (iv) At corn planting, in the first and second cycle, soil nitrate was highest after manure (66 ppm and 19 ppm) and lowest after clover (27 ppm and 8 ppm) but in the third cycle, highest after clover (15 ppm) and lowest after control (6 ppm); (v) for the subsequent sampling times, a significant positive effect of manure on soil nitrate was observed in the first cycle.

As expected, using cattle manure increased soil nitrate at more sampling times and in higher magnitude than any other treatment. Soil nitrate after the incorporation of a clover green manure was similar to a soybean green manure and reflected the much smaller amounts of N contained in green manure compared to cattle manure. Even though control treatments received no additional N, they were similar in soil nitrate to the green manures, likely due to the long history of high applications of composted cattle manure which had a residual or carry-over effect. However, cattle manured plots also showed the steepest decline in soil nitrate between corn planting and corn harvest and it is likely that more N was lost to denitrification or leaching from cattle manure than from the green manures. While green manuring can be beneficial for this type of system, for example in suppressing weeds (see chapter 5), soil nitrate levels are more likely maintained with regular applications of cattle manure. To address issues of N loss, research could investigate the growing of green manures or more specifically N catch crops after the application of cattle manure or after corn harvest, the times with the highest potential for N leaching or denitrification.

For organic farmers without access to livestock manure, including an undersown green manure in a soybean-winter wheat-corn rotation has been recommended to increase N availability to the following crop without losing a year of cash crop production (Snapp et al., 2005). In the first cycle of our study, one year of green manuring did not maintain soil nitrate levels as required by organic standards. Green manure N accumulation is highly correlated with DM production which greatly depends on weather. In this study, green manure DM showed high variability between years (chapter 2), carrying a greater risk of inadequate N accumulation for subsequent cash crops, especially in drought-prone areas such as the Western Corn Belt Plains ecoregion. Finally, to answer the question whether undersown green manures can maintain soil nitrate levels as well as animal manures, long-term studies investigating repeated undersowing of clovers into winter wheat in soybean-winter wheat-corn rotations are needed. The inclusion of multiyear green manure leys should be re-examined, as they accumulate higher amounts of N. However, in light of the variability in weather conditions, especially precipitation, in this ecoregion, research should be directed towards more drought-tolerant species of green manure crops, such as alfalfa or sweet clover.

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Table 3.1. Studies on organic undersown clover – winter wheat systems that determine the portion of N derived from atmosphere (%Ndfa). Amount of N from fixation was based on clover DM yield at termination time and %Ndfa. Clover was grown for one season, except when allowed to overwinter and terminated in the spring before planting of the next crop. The studies include %Ndfa values and the amount of Ndfa yr^{-1} derived from a two year organic grass-clover ley that contained both red and white clover.

Author and year	Location and management	Method of N determination	System	%Ndfa	Amount of N from fixation (kg N ha^{-1})	Other findings
Schipanski and Drinkwater, 2011	Central New York state, 15 fields from 7 farms, soil fertility gradient based on management, including organic	15 N natural abundance, reference plant orchardgrass	Winter wheat - red clover	74% in the fall 68% in the spring	65 34	%Ndfa was higher for undersown than monoculture red clover %Ndfa was not influenced by fertility gradient
Amossé et al., 2014	South-east France, 6 livestock-free organic farms	15 N natural abundance using B values from the literature, weeds as reference plants	Winter wheat - red clover Winter wheat – white clover	84% in the fall 71% in the fall	62 67	
Oberson et al., 2013	Switzerland, 21-year DOK trial, two year old ley in low-fertilizer input organic system, cut 5 times yr^{-1}	15 N abundance, reference plant perennial ryegrass	Grass – clover ley (red clover) Grass –clover ley (white clover)	83 – 86 % 91%	104 yr^{-1} 37 yr^{-1}	Total grass-clover ley Ndfa was 141 kg ha^{-1} yr^{-1} Ndfa not significantly affected by farming system

Table 3.2. N contained in above-ground DM at termination. The percentage of N and C, as well as C/N of green manures were taken in the fall of 2011 and spring of 2012 (Shi, 2013). For calculating the amount of dry matter N in the subsequent cycles, the same values as in 2011/2012 were used, and multiplied with the above-ground DM taken in that cycle (numbers in italics)(see chapter 2 for green manure DM). During the second cycle, the clover crop failed and during the third cycle, the soybean cover crop failed.

				Dry matter N content		
	Green manure	**N**	**C/N**	**First cycle**	**Second cycle**	**Third cycle**
		%			**Kg ha^{-1}**	
Fall termination	Red clover	1.92	22	105	*6*	*67*
	White clover	2.4	18	53	*3*	*59*
	Soybean	4.15	11	172	*118*	*3*
Spring termination	Red clover	3.86	11	143	*13*	*33*
	White clover	3.56	12	109	*4*	*16*

Table 3.2.a. N contained in cattle manure applied. Dairy manure was used in the first cycle and beef manure in the second and third cycle. Both were applied at 56 Mg ha^{-1}. Dairy manure parameters for ammonium, organic and total N, as well as the estimated first year availability, are from a nutrient analysis. Beef manure parameters are from Koelsch and Shapiro (2008) for a beef (paved feedlot), preplant applied and incorporated immediately.

Type of manure	**Cattle manure DM and N content**				**Total N available in the first year at 56 Mg ha^{-1}**	
	DM	NH_4^+-N	Organic N	Total N	NH_4^+-N	Total N
	%	Kg Mg^{-1}	Kg Mg^{-1}	Kg Mg^{-1}	Kg ha^{-1}	Kg Mg^{-1}
Dairy manure	25	2	3.3	5.3	56	112
Beef manure	29	2.5	4.5	7	134	196

Table 3.3. Source of variation, degrees of freedom (d.f.), and P-values for soil nitrate levels in the first cycle (2011 – 2014) under clover at 0 – 20 cm soil depth. Numerator d.f. = 1 for all treatments. A separate ANOVA was carried out at each sampling time because not all treatments were present at each sampling time. At Soybeanharvest, only half the plots were sampled and not all treatments were represented in equal numbers.

Sampling time	Main treatment	Denominator d.f.	P-Value
Clover planting	Clover	14	0.896
Wheat harvest 1	Clover	14	0.167
Fall kill	Clover	12	0.452
	Mulching	12	0.018
	Clover x Mulching	12	0.870
Corn planting	Clover	12	0.546
	Mulching	12	0.492
	Termination	12	0.086
	Clover x Mulching	12	0.272
	Mulching x Termination	12	0.614
	Clover x Termination	12	0.689
	Clover x Mulching x Termination	12	0.246
Corn harvest	Clover	12	0.010
	Mulching	12	0.001
	Termination	12	0.024
	Clover x Mulching	12	0.005
	Mulching x Termination	12	0.224
	Clover x Termination	12	0.535
	Clover x Mulching x Termination	12	0.227
Soybean planting	Clover	12	0.862
	Mulching	12	0.457
	Termination	12	0.015
	Clover x Mulching	12	0.339
	Mulching x Termination	12	0.893
	Clover x Termination	12	0.599
	Clover x Mulching x Termination	12	0.131
Soybean harvest	Clover	3	0.258
	Mulching	3	0.247
	Termination	5	0.288
	Clover x Mulching	3	0.548
	Mulching x Termination	5	0.810
	Clover x Termination	5	0.052
	Clover x Mulching x Termination	5	0.900
Wheat harvest 2	Clover	12	0.483
	Mulching	12	0.791
	Clover x Mulching	12	0.825

Table 3.4. Source of variation, degrees of freedom, and P-values for soil nitrate levels in the first cycle under clover at 20 – 60 cm soil depth. Numerator d.f. = 1 for all treatments. A separate ANOVA was carried out because not all treatments were sampled each time. No data is available for Fall kill.

Sampling time	**Main treatment**	**Denominator d.f.**	**P-value**
Wheat harvest 1	Clover	14	0.944
Corn planting	Mulching	12	0.512
	Clover	12	0.946
	Termination	12	0.026
	Mulching x Clover	12	0.848
	Mulching x Termination	12	0.606
	Clover x Termination	12	0.505
	Mulching x Clover x Termination	12	0.296
Corn harvest	Mulching	12	0.088
	Clover	12	0.708
	Termination	12	0.185
	Mulching x Clover	12	0.116
	Mulching x Termination	12	0.887
	Clover x Termination	12	0.444
	Mulching x Clover x Termination	12	0.670
Soybean planting	Mulching	12	0.181
	Clover	12	0.947
	Termination	12	0.120
	Mulching x Clover	12	0.650
	Mulching x Termination	12	0.965
	Clover x Termination	12	0.343
	Mulching x Clover x Termination	12	0.839
Soybean harvest	Mulching	3	0.211
	Clover	3	0.706
	Termination	5	0.926
	Mulching x Clover	3	0.772
	Mulching x Termination	5	0.155
	Clover x Termination	5	0.257
	Mulching x Clover x Termination	5	0.625
Wheat harvest 2	Mulching	12	0.818
	Clover	12	0.221
	Mulching x Clover	12	0.263

Table 3.5. Source of variation, degrees of freedom, and p-values for soil nitrate levels after all soil amendments (clover, cattle manure, soybean cover crop, or control). Soil nitrate values are available for one rotation (3 years) for the first cycle, for 2 years for the second, and for one year for the third cycle. ANOVA was carried out separately for each cycle and depth. Sample size was unequal for the various sampling times.

Source of variation	**Numerator d.f.**	**Denominator d.f.**	**F-value**	**P-value**
		First cycle		
		0-20 cm		
Soil amendment	3	37.9	50	0.001
Sampling time	6	40.7	108	0.001
Amendment x time	18	66.2	33	0.001
		20-60 cm		
Soil amendment	3	45.0	47	0.001
Sampling time	5	63.2	94	0.001
Amendment x time	15	85.7	7	0.001
		Second cycle		
		0-20 cm		
Soil amendment	3	45	29	0.001
Sampling time	5	26	52	0.001
Amendment x time	15	42	9	0.001
		20-60 cm		
Soil amendment	3	38	26	0.001
Sampling time	5	51	72	0.001
Amendment x time	15	61	13	0.001
		Third cycle		
		0-20 cm		
Soil amendment	3	39	3	0.037
Sampling time	2	44	64	0.001
Amendment x time	6	49	11	0.001
		20-60 cm		
Soil amendment	3	22	5	0.011
Sampling time	2	29	118	0.001
Amendment x time	6	38	3	0.011

Table 3.6. Source of variation, degrees of freedom, F-value and P-value for soil nitrate levels after red or white clover for the second cycle. Effect of mulching and termination was not evaluated in this rotation.

Source of variation	**Numerator d.f.**	**Denominator d.f.**	**F-value**	**P-value**
		0 – 20 cm		
Clover	1	49	1	0.349
Sampling time	5	36	136	0.001
Clover x time	5	36	0	0.921
		20 – 60 cm		
Clover	1	55	1	0.432
Sampling time	5	40	95	0.001
Clover x time	5	40	0	0.999

Table 3.7. Source of variation, degrees of freedom, and P-values for soil nitrate levels under red and white clover, at 0 – 20 cm and 20 – 60 cm soil depth for the third cycle. Numerator d.f. = 1 for all treatments. Separate ANOVA were carried out for each sampling time because of differences in the treatment design and sample sizes.

Sampling time	Source of variation	Denominator d.f.	P-Value
		0 – 20 cm	
Clover planting	Clover	19	0.066
Wheat harvest	Clover	7	0.245
Fall kill	Clover	17	0.157
	Mulching	17	0.591
	Clover x Mulching	17	0.269
Corn planting	Clover	2	0.259
	Mulching	2	0.660
	Termtime	2	0.618
	Clover x Mulching	2	0.726
	Clover x Termtime	2	0.701
	Mulching x Termtime	2	0.734
	Clover x Mulching x Termtime	2	0.909
		20 – 60 cm	
Clover planting	Clover	19	0.054
Wheat harvest	Clover	7	0.381
Fall kill	Clover	17	0.856
	Mulching	17	0.815
	Clover x Mulching	17	0.815
Corn planting	Clover	2	0.964
	Mulching	2	0.516
	Termtime	2	0.702
	Clover x Mulching	2	0.656
	Clover x Termtime	2	0.926
	Mulching x Termtime	2	0.936
	Clover x Mulching x Termtime	2	0.980

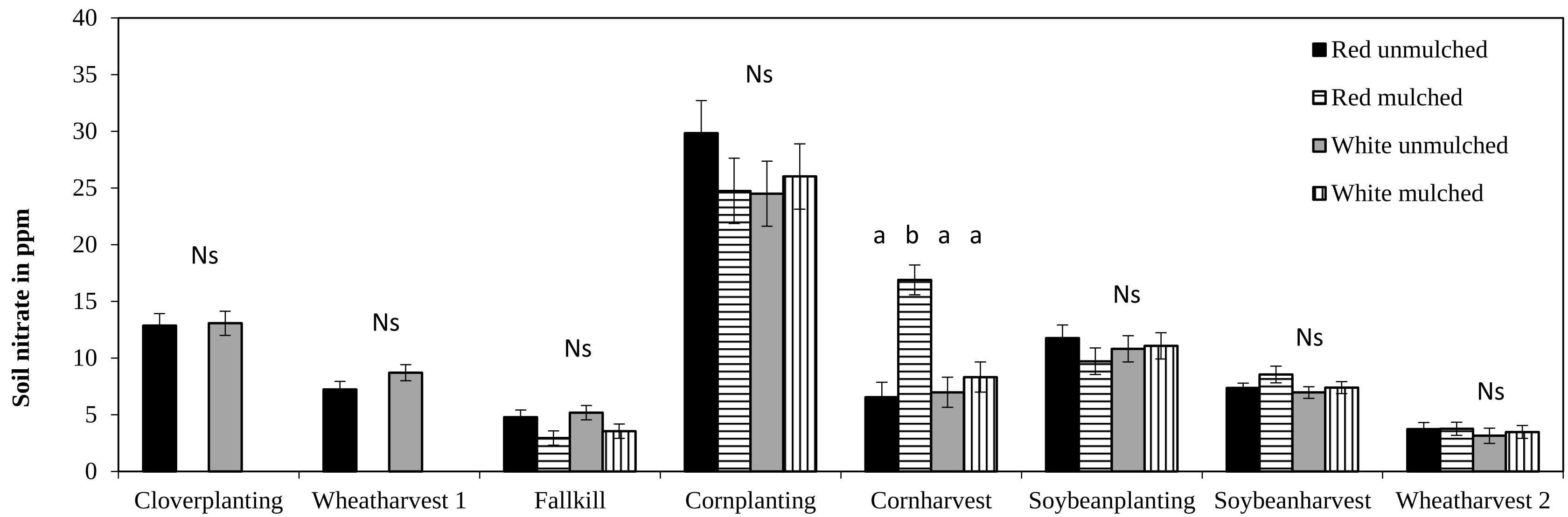

Figure 3.1. Soil nitrate dynamics under clover in first cycle in 0 – 20 cm soil depth. Red and white clover were undersown in winter wheat, mulched 6 weeks after wheat harvest, and terminated either at Fallkill or in the spring before Cornplanting. A separate ANOVA was carried out for each sampling time. Means that are significantly different at $\alpha = 0.05$ are indicated with a different letter. Error bars are standard errors of the mean.

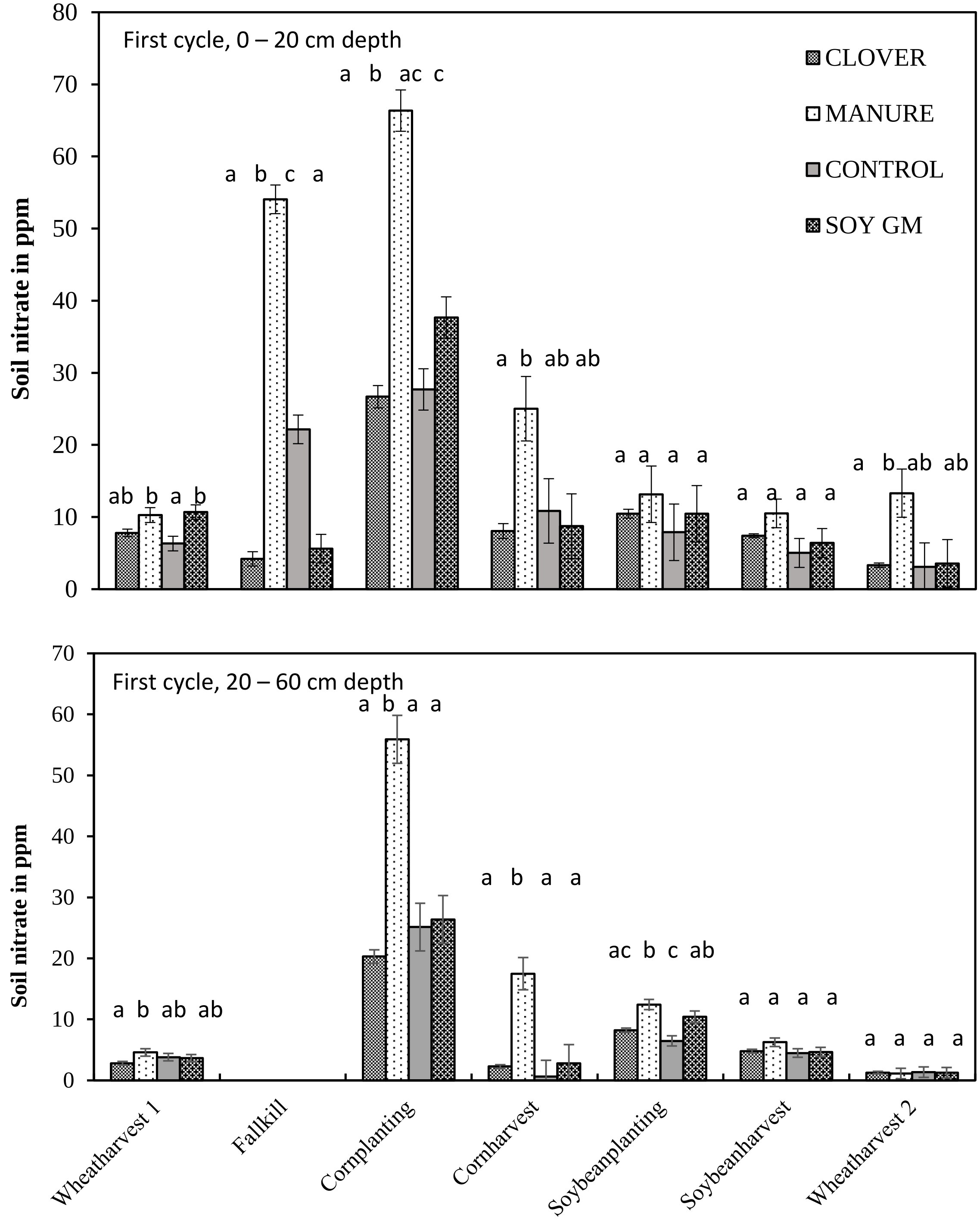

Figure 3.2. Soil nitrate levels during the first cycle for all soil amendments. Red and white clover are combined (Clover) and were present at Wheatharvest 1. All other treatments were applied after Wheatharvest 1. No samples in the 20 to 60 cm soil depth were taken at Fallkill. At each sampling time, treatments that are not significantly different at 0.05 are indicated with the same letter. Error bars are standard errors of the mean.

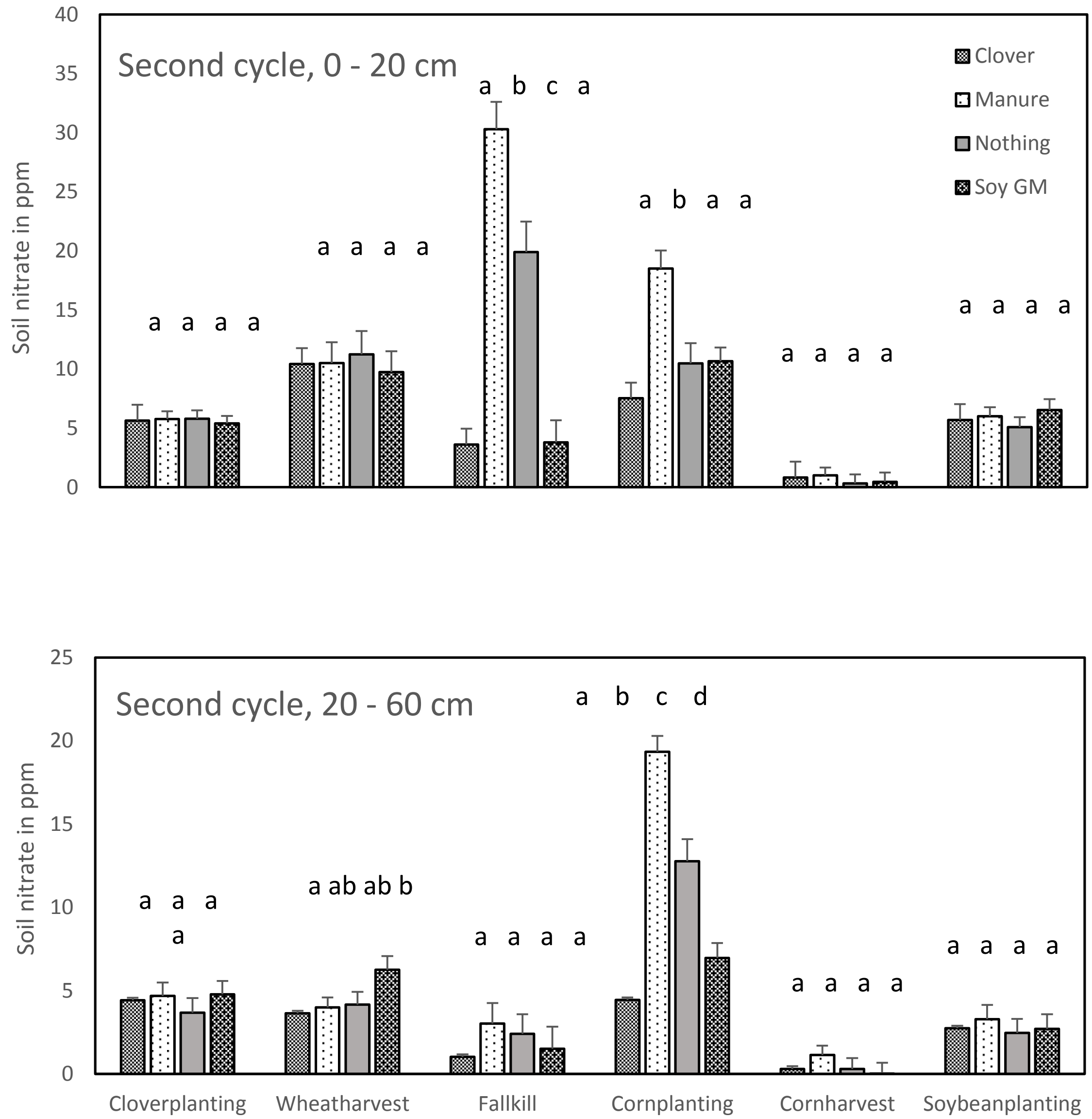

Figure 3.3. Soil nitrate level during the second cycle for all soil amendments. Red and white clover are combined (Clover) and were present at Wheatharvest. All other treatments were applied after Wheatharvest. At each sampling time, treatments that are not significantly different at 0.05 are indicated with the same letter. Error bars are standard errors of the mean.

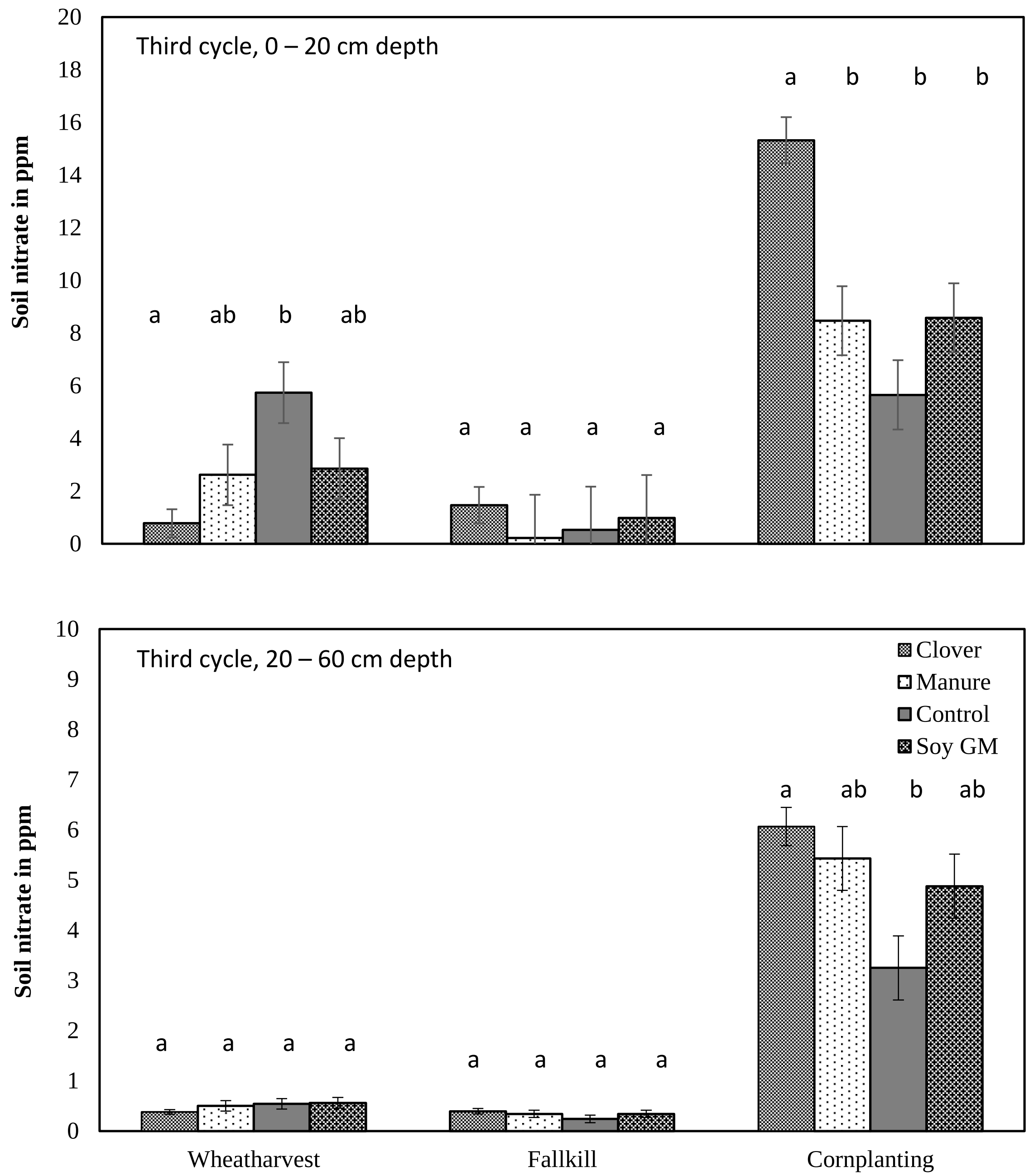

Figure 3.4. Soil nitrate levels during the third cycle for all soil amendments. Red and white clover are combined (Clover) and were present at Wheatharvest. All other treatments were applied after Wheatharvest. At each sampling time, treatments that are not significantly different at 0.05 are indicated with the same letter. Error bars are standard errors of the mean.

CHAPTER 4

ORGANIC CORN YIELDS FOLLOWING GREEN MANURES OR CATTLE MANURE

Organic farming systems often have lower cash crop yields than conventional farming systems (Seufert et al., 2012; Cavigelli et al., 2008; Mäder et al., 2002). Despite lower yields, organic corn (*Zea mays* L.) production in the United States currently has higher returns per hectare than conventional corn production, because of lower operating costs and price premiums for organic products (Foreman, 2014). However, this should not be a reason to become complacent of lower yields in organic farming systems. Demand for organic feed grains, especially corn and soybeans (*Glycine max* [L.] Merr.), far outstrips supply, and feed corn is now the tenth most imported organic food product (Organic Trade Association, 2015). Market theory dictates that increasing supply of organic products (either through imports or the conversion of domestic conventional farmland to organic farmland) will lower organic premiums in the future. Moreover, if organic farming is to play a substantial role in feeding the world population, it has to become much more productive (de Ponti et al., 2012). Organic farmers in the USA need to increase yields as an essential part of increasing efficiency and total supply of organically grown crops. Several researchers have identified nutrient limitations, especially nitrogen, as the factor most limiting crop yields in organic systems, because fertilizers permitted under organic regulations typically have low concentrations of readily available N (Berry et al., 2002; de Ponti et al., 2012).

For organic farms with livestock, the manure from livestock can be a plentiful and inexpensive source of N, some of which is readily available. Because the organic N mineralizes over weeks to years, frequent manuring can increase soil N and lead to higher crop yields over time (Schröder, 2005). If manure cannot be used, leguminous plants, especially forage plants cultivated as green manures, provide N, as well as other benefits. Legumes can fix considerable amounts of N depending on the species, length of growing period, and other factors, but all legume N must undergo mineralization before it becomes plant available (Crews and People, 2005). The amount of legume N fixed is closely correlated with legume DM production (Carlsson and Huss-Danell, 2003), and DM production can be manipulated by mowing and whether or not the green manure will be allowed to overwinter (for more detailed description of these factors, see the previous chapters).

Direct comparisons of yields of organic crops fertilized with either green manures or animal manures are complicated because the year-to-year variation in weather can greatly influence green manure DM production and thus the amount of N fixed (Carlsson and Huss-Danell, 2003). Data from long-term trials with green manures and animal manures in the rotation can be a better source of information on the comparative effects of these two soil amendments on yields, because long-term trials include a range of temperature regimes and precipitation levels observed for a particular rotation. Several long-term organic farming system trials have reported that crop yields from organic grain systems that include animal manure are higher than from organic grain systems that are based solely on N derived from legumes. For example, in the Wisconsin Integrated Cropping Systems Trial, corn in an oat (*Avena sativa* L.)/alfalfa (*Medicago sativa* L.)-alfalfa-corn rotation where corn was fertilized with cattle (*Bos taurus*) manure yielded more than corn in a soybean-winter wheat (*Triticum* *aestivum* L.)/red clover (*Trifolium pratense* L.)-corn rotation (8.95 versus 8.17 Mg ha^{-1}) (Posner et al., 2008).

Wortman et al. (2012a) reported crop yields between the years of 1996 to 2007 from the Long-Term Crop Rotation experiment conducted near Mead, NE about 1 km from our site. The organic animal manure system consisted of a soybean-corn/sorghum (*Sorghum bicolor* [L.] Moench)-soybean-winter wheat rotation with cattle manure applications at an average rate of 31 Mg ha^{-1} before corn/sorghum and winter wheat (actual application rate based on soil tests and crop N removal). The organic forage grain system consisted of alfalfa-alfalfa-corn/sorghum-winter wheat which received manure before alfalfa in half of the study years to improve P nutrition of the forage. Corn yields were higher in the organic animal manure system (6.56 Mg ha^{-1}) than in the organic forage grain system (5.05 Mg ha^{-1}). Soil nutrient concentrations were higher in the organic animal manure system than the organic forage grain system, especially for P, but also for K, Ca, Mg and Zn. Concentrations of N were not reported. Soil organic matter was above 3% for all farming systems, but highest for the organic animal manure system. For reference, corn yields were 7.65 and 7.35 Mg ha^{-1} in the conventional and diversified conventional farming system, respectively, and researchers speculated that the yield gap was caused by high weed pressure in the organic plots. Long-term corn yields from the Rodale Institute in Pennsylvania were only slightly higher for the organic manure-based system (6.43 Mg ha^{-1}) than for the organic legume-based system (6.37 Mg ha^{-1}) after the first five years of the trials (Pimentel et al., 2005).

Not all long-term organic system trials compare systems that receive animal manure with those that use green manures as their only N source. Rotation complexity and length of green manure period are also factors that influence corn yields. The USDA-ARS Beltsville

Farming Systems Project in Maryland compares corn grown in three organic rotations that differ in complexity but use green manure as the main N source, and in case of poor green manure stands, were fertilized with animal manure. The four-to-six year rotation that included two years of hay (either red clover and orchardgrass [Dactylis glomerata L.] or alfalfa) had higher corn yields (6.15 Mg ha^{-1}) than the three-year corn-soybean-winter wheat/hairy vetch (*Vicia villosa* Roth) rotation (5.55 Mg ha^{-1}). Overall organic yields were low due to N deficiencies and to a lesser extent weed pressure (Cavigelli et al., 2008). Corn grain yields in an organic long-term trial in Iowa which has similar growing season length to our site, but higher average precipitation, were 10.48 Mg ha^{-1} for a soybean-oat/alfalfa-corn rotation where alfalfa was undersown in oat and terminated before corn the following spring and 11.17 Mg ha^{-1} for a soybean-oat/alfalfa-alfalfa-corn rotation where alfalfa was undersown in oat and terminated two years later before corn planting. Both organic rotations received composted swine manure at a rate of 158 kg N ha^{-1} before corn. Yields from both organic rotations were not significantly lower than those obtained in the conventional system (11.3 Mg ha^{-1}) which received 158 kg N ha^{-1} in the form of urea (Delate et al., 2014).

While green manures can provide N sufficient for high corn yields, green manure crops use soil water, potentially leaving soil water deficits for a subsequent crop if rainfall is not adequate. Average annual precipitation at our site is 708 mm, higher than the 500 mm sometimes reported as the threshold for using cover crops (Robinson and Nielsen, 2015). While the USDA guidelines recommend killing a cover crop in this area at cash crop planting, earlier termination of the cover crop/green manure will lower the risk for subsequent crop failure in years with insufficient precipitation (Unger and Vigil, 1998).

In this study, we wanted to determine the effect of three different types of green manures in an organic soybean-winter wheat-corn rotation compared to the effects of animal manure or a control on corn yields. Further, we investigated the effect of green manure management (mulching and time of termination) on corn yields. We hypothesized that:

1. Manured plots will have the highest and control plots the lowest corn yields.
2. Green manures that are terminated in the fall will have higher corn yields in years with limited precipitation, because they will use less soil water.
3. Green manures that are terminated in the spring will have higher corn yields in years with non-limited precipitation, because they will produce more total N.
4. Mulching reduces green manure DM and will increase corn yields in years with limited precipitation.

MATERIALS AND METHODS

For the detailed description of the site, soils, and experimental design, see Chapter 1. Weather data were obtained from the High Plains Regional Climate Center (HPRCC) for the Mead Agrofarm Climate Station, located about 1 km from the study site and not surrounded by windbreaks (Automated Weather Data Network, ID a255369, High Plains Regional Climate Network). Because climate data for this station was not available before 1994, long-term climate data was obtained from the Mead South-Southeast station, and averaged for the years 1971 to 2000.

In this soybean-winter wheat-corn rotation, all soil amendments are applied during the wheat phase of the rotation. The soil amendments were two types of green manures: Forage legume green manure (red or white clover undersown in spring in winter wheat) and summer annual green manure (soybean green manure planted after winter wheat harvest); as well as cattle manure (applied after wheat harvest at 56 Mg ha^{-1}) and a control (no soil amendments). Nitrogen contents of green manures and cattle manures are shown in tables 3.2 and 3.2a, respectively. The forage legumes were either mulched (mowed with the plant residues left in place) once in the summer of the establishment year or not mulched and terminated in the fall of the establishment year or the spring of the second year, two weeks before corn planting (see chapter 2 for red and white clover varieties, planting densities, and treatments). For a list of management operations and dates during each phase of the rotation, see table 1.1.

Corn was planted at 75,000 kernels ha^{-1} in rows 0.76 m apart in each year. All corn seed was obtained from Blue River Hybrids and was organically certified. Tall varieties were selected, as they can compete better with weeds. Further variety selection criteria included good plant health and high yield potential, for example, the variety in 2014 yielded up to 10.71 Mg ha^{-1} in variety trials (Blue River Hybrids, 2013; 2014). Varieties were 63H30 in 2012 (111 days) and 67H19 (113 days) in 2013 and 2014.

Corn stand counts were carried out on June 11 and June 18 in 2012, and on July 8 in 2013, by counting all corn plants in two randomly selected, 3 m rows per experimental unit. Corn stands were not counted in the summer of 2014. Corn was harvested at maturity (full dry down) using a field-size combine (see Chapter 1) and weighed in the field. Corn grain moisture was not measured, so no adjustments for corn moisture were made. Plots were 9.1 m wide, and contained 12 or 13 rows of corn, allowing for two passes with the 6 row combine. Grain from one pass was emptied into a grain cart with a scale accurate to 4.5 kg and weighed, and this weight was used to determine yield:

Yield [Mg ha^{-1}] = Corn grain yield [kg] x 10 /(4.55 m x plot length [m])

In the first cycle, size of the undersown green manure plots was 624 m^2 and size of soybean green manure, cattle manure and control plots was 937 m^2. In 2013 all plots were 277 m^2 and in 2014 all plots were 166 m^2. Reduction in plot size reduced grain weights per plot, and increased the error due to scale inaccuracies. Error due to scale inaccuracies was also higher in large plots that had low grain weight due to treatment and/or blocking effects. Some of this error was reduced by changing from a completely randomized design in the first cycle to an incomplete randomized block design with 13 replications for forage legume green manure in the second and 20 replications in the third cycle, respectively. However, the incomplete treatments (soybean green manure, cattle manure and control) were only replicated four or five times in each cycle.

Statistical analysis was carried out using SAS 9.4 (SAS Institute, Cary, NC). ANOVA was carried out with the GLIMMIX procedure, using treatment as a fixed factor and block (in 2013 and 2014) as random factor. Treatments were not compared across years since yields were not adjusted for moisture. Because the undersowing of red and white clover into this type of rotation is a new method, we were especially interested in the effects of treatments applied to red and white clover: mulching and termination time (see chapter 2). Thus, a separate ANOVA was run including only type of undersown green manure (red or white), mulching (mulched once or never mulched) and termination time (fall or spring). Means were compared with Fisher's LSD at a significance level of $\alpha = 0.1$. The higher probability of a Type I error was chosen to reduce the risk of a Type II error, as Type II errors can be more harmful in agronomic research than Type I errors (Campbell et al., 2015). The Type I error in this study would be to infer that there was a yield difference when in reality there was none. The Type II error would be to not find the yield difference that actually exists. In case of a negative effect of green manuring, a Type II error could lead to economic losses from lower yields, whereas it could lead to lost economic opportunity in case of a positive effect of green manuring.

To determine if soil nitrate levels were adequate for certain yield goals, we used the University of Nebraska corn fertilizer recommendations based on the following algorithm (Shapiro et al., 2008)

$$\text{N need (lb/ac)} = [35 + (1.2 \times EY) - (8 \times NO_3\text{-N ppm}) - (0.14 \times EY \times OM) - \text{other N credits}] \times Price_{adj} \times Timing_{adj}$$

Where:

EY = Expected yield (bu/ac)

NO_3-N ppm = average nitrate concentrations in 0 – 60 cm depths, in parts per million

OM = percent organic matter

Other N credits = N from legumes, manure, other organic materials

$Price_{adj}$ and $Timing_{adj}$ = adjustment factors for corn and N prices, and application time

We also used the corn nitrogen calculator (Ferguson et al., 2008) which calculates fertilizer N for a desired corn yield goal using the formula above, with modifications for soil texture, number and thickness of soil layers sampled, and depth of rooting zone. Expected yield values with and without N credits, using soil nitrate levels at corn planting (chapter 3), are given in table 4.1.

In organic farming, it is difficult to feed a crop "on-demand" because most soil amendments mineralize slowly and somewhat unpredictably, and are difficult to apply to a standing crop. In the case of green manures, they may not fix enough N for the subsequent crop, depending on weather and management. In our study, any N deficits at corn planting could not be corrected by additional fertilizer applications. We thus adjusted the yield "goal" in the N calculator to the level where no additional N besides that from soil nitrate and N credits would be needed and used this value as the yield estimate. The algorithm that estimated the yields assumes that water is not limiting and did not take into account any other factors affecting corn growth, such as temperature and radiation. Despite the limitations to using the corn N calculator for a yield estimate, it is likely more predictive than a yield estimate based solely on soil nitrate levels at corn planting.

RESULTS AND DISCUSSION

Weather

Weather conditions varied widely, including Nebraska's hottest and driest year on record (2012), leading to large variations in green manure DM production which in turn influenced N and soil water available for the corn crop. In the first cycle, precipitation during the green manure establishment year (2011) was above the 30-year mean (normal) during most of the growing season, favoring high green manure DM production (table 2.2). September and October had less than 30% of their average precipitation, but green manure DM at fall termination was high (chapter 2). Temperatures in January (2012) and February were 5 and 2 ˚C higher than normal, and there was little snowcover, increasing evaporation from the soil. March average temperature was 12.5 ˚C which is 9˚C above normal, and April temperatures 2 ˚C above normal, initiating early regrowth and high evapotranspiration of the overwintered clover. Precipitation was 20 mm below normal in March, 12 mm above normal in April, and 20 mm below normal in May, not adequate to replenish soil moisture deficits left after the green manures. June rainfall and temperature were close to the normal of 101 mm and 22 ˚C, respectively, but in July, temperatures were 28˚C which are 4˚C above normal, and precipitation was only 2 mm. August received 14 mm of rain, 70 mm less than normal and September received 34 mm of rain, about half the amount of the normal rainfall.

In the second cycle, drought conditions that had prevailed from September of 2012 to March 2013 were abated when rainfall was 117 mm more than the 275 mm normal during April, May and June of 2013. July was dry, with only 15 mm of rain, and August had 36 mm less rain than normal. September and October precipitation was above normal. Temperatures were within 1 ˚C of normal between May and August; only September temperatures were 2 ˚C higher than normal.

In the third cycle, a very cold and dry winter was followed by a cool spring. Nighttime temperatures until mid-May of 2014 were often below 5 ˚C and the last nighttime frost occurred on May 16. During the last week of May, temperatures rose rapidly and measured 23 to 24 ˚C, about 4 to 5 ˚C higher than the average daily temperature during this period. Overall, the average temperature for May was 1 ˚C above normal and rainfall in May was close to normal. June temperatures were normal, but precipitation was twice as high as normal (200 mm). July temperatures were 2.4 ˚C below normal and precipitation was only 24 mm. Temperatures in August and September were normal, but precipitation for the two months was 70 mm above normal.

Corn Emergence and Growth

In the first cycle, type of clover and mulching did not significantly affect corn emergence, but termination time did (table 4.2). Fall-terminated clover plots had 52,200 plants ha^{-1}, 6,600 plants ha^{-1} more than spring-terminated clover plots (table 4.2). Manure, soybean cover crop and control plots had stand counts of 52,800. During planting, the planter malfunctioned and some rows were not planted, probably explaining most of the differences in corn counts (figure 4.1). In addition, plants were buried during cultivation. In the second cycle, type of soil amendment had no influence on corn emergence, and the overall corn count was 46,800 plants ha^{-1}. While red clover residue had allelopathic effects on corn emergence in laboratory experiments, effects subsided after several weeks (Sturz and Christie, 1996). Liebman and Sundberg (2006) found that allelopathic effects of red clover were higher for small-seeded species (such as many weed species) than large-seeded species. Discussions of allelopathic impacts of red or white clover green manures on following crop seedling emergence in the field were rarely found in the literature and likely had less impact than equipment problems on corn emergence. However, lower corn emergence after spring-terminated red clover in dry years was due to the uptake of soil water of the clover (Hesterman et al., 1992).

The preceding clover green manure had a profound negative effect on corn growth in the first cycle and to a lesser extent in the second cycle. Red clover terminated in the spring of the first cycle had used approximately 763 mm water, as much as the precipitation received during its time in the field. Reduced corn growth after different treatments was visible early. Corn after green manures was shorter than corn after manure or control in the first and second cycle (figure 4.2) and exhibited symptoms of N deficiency, such as yellowing of leaves (figure 4.3) in the second cycle. Temperatures that were continuously above 35˚C impacted corn pollination in corn fields across this area in 2012. In our study, clover treatments were most affected, having overall lower numbers of kernels per ear due to unpollinated rows and shorter ears, than the manure or control treatments (figure 4.4). At corn harvest, the control and manure plots were relatively free of weeds (figure 4.5), but the clover plots were very weedy, even though prior to corn planting, red clover plots were practically weed free (see chapter 5). It is likely that due to inhibited corn growth and lack of canopy closure weeds were able to re-infest the clover plots after the last weed management operations had been carried out. Clover can also become a weed in subsequent crops due to regrowth after incomplete kill, germination of hard seed, or from seeds produced during the green manure year but this was not observed in any of the cycles in this study.

Historical Corn Grain Yields at this Site

On this site, before the transition to organic management, corn was grown irregularly on each field, and mean grain yields were 9.22 Mg ha^{-1}. During the six years of organic management, mean yields were 8.7 Mg ha^{-1} and corn was grown once every three years on each field (table 4.4). Excluding the two years when the crop was damaged by late-season hail or storms, organic corn grain yields were 9.72 Mg ha^{-1} (combine-harvested at dry-down, not adjusted for moisture). Before the study began, fields 789 and 14 have reached corn yields > 10 Mg ha^{-1} under organic management, and it is likely that field 356 (field 2 was not included in the study) could attain corn yields > 10 Mg ha^{-1}, given its yield history under conventional management and similarities in soil type and slope. For reference, the long-term corn grain yield for rainfed conventional corn at Mead, NE, is 9.6 Mg ha^{-1} (Grassini, 2013).

Effects of Organic Soil Amendments on Corn Grain Yield

When comparing all soil amendments (red clover, white clover, soybean green manure, control and cattle manure) type of soil amendment had a significant impact on corn grain yields (table 4.5). In the first and second cycle, corn grain yields were highest after the control treatment, but not significantly different from the second highest yielding treatment, cattle manure (figure 4.6). In the third cycle, cattle manure was the highest and control the lowest yielding treatment (figure 4.6). Despite stark differences in precipitation and temperatures between years, manured plots were relatively consistent in yields (7.61 Mg ha^{-1}, 7.6 Mg ha^{-1} and 8.14 Mg ha^{-1} in 2012, 2013, and 2014, respectively), although the actual grain weight could be slightly different since corn grain was not adjusted for moisture (see above). Manure treatments in this study were not as high as previous corn grain yields obtained at this site under organic management with manure applications.

Corn yields after manure and control treatments

Field 789 used in the first cycle had corn yields > 10 Mg ha^{-1} in the previous years under organic management (table 4.4). High soil nitrate levels were measured at corn planting, probably due to warm and moist spring weather that favored mineralization, as well as high amounts of green manure DM that had been incorporated. Soil nitrate under manure was 59 ppm, sufficient for high yields. For example, the pre-sidedress nitrate test used in the Midwest considers soil nitrate concentrations of 20 to 30 ppm in the top 0.3 m as the limit above which yield gains from additional N fertilizer are not likely (Magdoff et al., 1990). Yield estimates based on the UNL N calculator are high for all treatments except the control, because of high soil nitrate levels and N credits from manure and green manures (table 4.1). No treatment reached the yield estimate because water was the limiting factor. Corn could not take up all available soil nitrate, resulting in high soil nitrate levels after corn harvest in all treatments (see chapter 3). For reference, conventional corn yields at the Shelterbelt farm were between 4.18 and 7.22 Mg ha^{-1}.

In the second cycle, water was not limiting during May and June, but little rain fell during July and August. Yields from fields 3 and 56 are available for only two years under organic management which happen to be years with storm damage to the crop and how much of the yield loss was due to storm damage is not known. During early corn growth, N probably was limiting, given that soil nitrate levels at corn planting were much lower than the ones observed in the previous cycle for manure. Reduced mineralization of manure during the drought year could have caused low soil nitrate levels in the manure plots. However, yields were higher than calculated, probably because more N became available between corn planting and the period of rapid corn N uptake. Yang et al. (2014) reported that conventional rainfed corn yields for Mead in 2013 were 10.36 Mg ha^{-1}, above average, despite the lack of rainfall during the critical period of July and August.

In the third cycle, moisture was not limiting during corn growth. Soil nitrate levels were very low for all treatments at corn planting (see chapter 3), probably because the very cold winter and cold spring had delayed mineralization. Nighttime temperatures were frequently below 5 ˚C (see above), and freezes still occurred until mid-May. Yield estimates based on soil nitrate were thus low, but were surpassed by actual yields for each treatment. With the rapid rise in temperatures in the last half of May, mineralization in the manured plots was sufficient to obtain corn yields close to the two year organic mean of 8.95 Mg ha^{-1} for this field. Weather conditions were favorable for high corn yields, with above average precipitation, below average evapotranspiration, minimum and maximum temperatures, although solar radiation was also below average (Grassini et al., 2014). It is likely that N limited higher yields in the manured plots. Control plots had the lowest yields of any treatment in this cycle, and lower yields than control treatments in the previous cycles. Soil nitrate was similar to that of manured plots at planting (table 4.1), but in contrast to the manured plots, soil organic matter was the only source of potentially mineralizable N and N availability severely restricted corn grain yields.

Corn yields after green manures

Compared to cattle manure, green manures (forage legumes and soybean) lowered corn yields in each cycle of the study. Corn yields were relatively consistent after soybean green manures (6.05, 6.22, and 6.1 Mg ha^{-1} in the first, second, and third cycle, respectively)(table 4.1). Treatment mean differences to manure were numerically similar in the first two cycles, but not significant in the first cycle ($P = 0.285$) and significant in the second cycle ($P = 0.091$), reflecting the differences in sample size and experimental design between both cycles. No clear trend between soybean green manure DM and N production was noticeable. In the first cycle, soybean green manures produced 4.15 Mg DM ha^{-1} and 172 kg N ha^{-1} in the fall before incorporation, while in the second cycle, they produced 2.84 Mg DM ha^{-1} and 118 kg N ha^{-1} (see chapter 2). In the third cycle, soybean green manure DM production was negligible, yet subsequent corn yields were similar to those of the first and second cycle, although they were the second-lowest yielding treatment in that cycle.

Corn yields after clover green manures were affected by clover type and termination time (table 4.2) but for comparison purposes, means for red and white clover are averaged across mulching and termination in Figure 4.6. Corn yields after red or white clover were always significantly lower than after manure. After red clover, they ranged from 37% of manured corn yields in the first cycle to 79% in the second to 87% in the third cycle. Correspondingly, after white clover, corn grain yields ranged from 59% to 60% to 83% of the manured corn grain yields for the first, second and third cycle, respectively.

The most likely causes for low corn grain yields after green manures are corn N deficits (either through low green manure N content or green manure N release that is not in synchrony with corn N demand) (Crews and Peoples, 2005), and soil water deficits incurred by green manures (Unger and Vigil, 1998). Measurements of corn tissue N or soil water content during the corn growing season were not carried out, but it is likely that both low soil water and lack of N from green manures caused low corn grain yields in different cycles. Soil water use and green manure N production are both positively correlated with green manure DM production and length of growing period (Carlsson and Huss-Danell, 2003; Badaruddin and Meyer, 1989). If soil water deficits by green manures were the reason for low corn yields, we would expect green manure treatments with the highest DM and/or longest growing period to result in the lowest corn grain yields. If green manures failed to produce sufficient N, we would expect green manure treatments with the lowest DM and/or shortest growing period to result in the lowest corn grain yields.

In the first cycle, type of clover significantly impacted corn grain yields when corn after red clover yielded 37% less than corn after white clover. Termination time also significantly impacted corn grain yields with spring termination lowering corn grain yields by 44% relative to fall termination. None of the interactions were significant and main treatment means are presented in table 4.3. At corn planting, soil nitrate levels after each green manure treatment were sufficient to produce corn yields of at least 11 Mg ha^{-1}, thus we assume that nitrogen was not the limiting factor for corn yields.

Biomass production in the first cycle was high, with red clover yielding significantly more DM than white clover. At each termination time, plots with the highest DM yields resulted in the lowest yields of subsequent corn. Mulching, although not a significant effect on corn grain yield, had a significant effect on clover DM (chapter 2), and for each clover type and termination time, mulched clover plots had slightly higher corn grain yields (fig. 4.7). We thus assume that in the first cycle, soil moisture deficits incurred by green manures limited corn production. Clover plots terminated in the fall had some soil water recharge until corn planting, although the combined precipitation from November 2011 to April 2012 was 170 mm, 30 mm less than the average. In addition, the winter and spring of 2012 were very mild, with early green manure regrowth and higher-than-normal evapotranspiration potential. Red clover plots terminated in the spring had used an estimated 762 mm of water, about the same amount of water as was received through precipitation. Rainfall after clover termination from May – June 2012 was about 85 and 90% of the average, and in July and August, the combined rainfall was about 15 mm, 160 mm less than normal. Water requirements for a 113-day corn variety in south-central Nebraska were approximately 650 mm (Kranz et al., 2008), and in the first cycle, water requirements for corn after clover green manures were not met. In return, corn growth was severely and irrevocably stunted. Hesterman et al. (1992) also reported that in years with precipitation deficits, corn following undersown red clover or alfalfa did not have a positive yield response, even though the precipitation deficit in their study was much less pronounced. Red clover did not reduced soil water compared to a control, but alfalfa did in one year of a study in Alberta where annual precipitation was less than 400 mm (Blackshaw et al., 2010).

While clover DM yield might explain most of the variation in corn grain yield, some variation is likely due to morphological and physiological differences between the clovers. For example, DM yield of the spring-terminated unmulched white clover was almost the same as that of spring-terminated unmulched red clover, but corn grain yield after white clover was twice as high as after red clover. It is possible that red clover which forms an extensive taproot, might have emptied the soil profile to a lower depth than white clover which has a shallow root system. In Winnipeg, relay-cropped red clover had significantly less soil water than a control in the fall of the establishment year, and these differences extended to a soil depth of 0.8 m (Thiessen Martens et al., 2001). Differences in water use efficiency could also play a role, though in a study from New South Wales, red and white clover had similar water use efficiency, which was lower than that of most other forage crops tested, including alfalfa (Neal et al., 2011).

The NRCA cover crop termination guidelines for this region recommend terminating a cover crop at the planting of the next crop (USDA-NRCS, 2014) but clearly, much earlier termination such as in the fall of the establishment year would minimize risk of cash crop failure. The method of green manure termination can also impact soil water conservation, for example Wortmann et al. (2012b) found that undercut cover crops preserved soil moisture in two years compared to disked cover crops and a control (no cover crops), although cover crops in that study were only grown for about two months before the cash crop.

In the second cycle, red clover treatments yielded 3.2 Mg ha^{-1} more than red clover treatments in the first cycle while white clover treatments yielded about the same as in the first cycle. Type of clover had a significant effect on the following corn grain yield (table 4.2), with corn after red clover yielding 24% more than corn after white clover (mulching and termination time were not analyzed) (table 4.3). Clovers were planted in the drought year of 2012 and had very low biomass yields (less than 0.4 Mg DM ha^{-1} for either type of clover and termination time) and hence very little N was accumulated by the clovers (chapter 2 and 3). Corn after all green manure types was yellowing and short (figure 4.3). We suspect that N deficiency was the cause for low corn grain yields after green manures in this cycle. While soybean green manure produced 2.84 Mg DM ha^{-1} it might not have fixed much N because drought conditions shift N accumulation from N fixation to soil nitrate uptake (Purcell et al., 2004). Corn yields after green manures were significantly lower than those after the control, which also did not receive N, probably due to tillage which increases mineralization from soil organic matter. Soil nitrate at corn planting was significantly higher under the control (13 ppm) than soybean (7 ppm) or clover (4 ppm) (chapter 3). Tilled soils also warm faster in the spring, giving corn a better start in these plots than in the plots that were tilled shortly before corn planting.

In the third cycle, corn grain yields were higher and growth was not limited by water. Type of clover, termination time, mulching or any of the interactions were not significant (table 4.2), but fall-terminated plots yielded 0.5 Mg ha^{-1} more than spring-terminated plots (table 4.3). Green manure DM yield was high in the fall (chapter 1), but very low in the spring probably because cold and lack of snow cover led to winterkill of the clovers. Temperatures in the spring were cool and possibly slowed mineralization, resulting in very low soil nitrate under all treatments in the spring (chapter 2). Symptoms of N deficiency such as yellowing leaves and stunted growth were not observed after green manure treatments, but corn grain yields after green manures were likely N limited.

In Wisconsin red clover terminated one day before corn planting released about 50% of its N within 4 weeks of spring killing, and increased corn yields significantly compared to a control. Corn yields were 10.5 Mg ha^{-1}, similar to 179 kg N ha^{-1} (Stute and Posner, 1995). Similar high yields after red clover were also observed in another conventional system in Iowa (Liebman et al., 2012). However, the requirements for high corn yields after red clover (high green manure DM in the spring and sufficient precipitation) were not met during our study.

Ultimately, the adoption of green manures in this area before corn will depend on the producer's yield goals. If the yield goal is the attainable corn yield, the yield possible at a certain site under organic management with optimum nutrient and rainfall conditions (Dobermann and Shapiro, 2004), cattle manure applications are advisable. However, producers may accept lower corn yields, if green manures have other economic benefits for their farm, such as for forage, weed control, or replacing the cost of purchasing cattle manure. In years with poor green manure growth, producers can supplement with other fertilizers, such as animal manure or compost, to avoid large yield losses.

Profitability of green manuring

Costs for corn production, including the preseason's fertilizer (cattle manure or green manure), seedbed preparations, seed costs and weed control, determine profitability along with corn sales. After wheat harvest, cattle manure and control plots were disked more frequently (table 1.1). When using green manures, seed costs are the single-largest expense, with white clover the most and sweetclover the least expensive (table 4.6). When using cattle manure, spreading (including labor and fuel cost for hauling and spreading) is the largest expense, however, in our example, the cost of spreading manure and additional disking is lower than the cost of buying, planting and killing green manures, except for sweetclover. Other expenses related to corn planting and harvest, such as corn seed costs, weed control and combining are not altered by treatments. Profitability was highest for the highest-yielding treatment, cattle manure, followed by alfalfa, sweetclover and red clover. Soybean green manure and the control, which were the lowest-yielding treatments had the lowest profits. This underlines the need for high yields to achieve high profits.

CONCLUSION

We found that green manures lowered yields of subsequent corn compared to cattle manure. This was caused predominantly by soil moisture deficits left by a highly productive clover green manure in the first cycle, and N deficits after low yielding green manures in the second year. In the third cycle, N deficits also lowered corn yields after green manures, but overall, yields were highest and the yield gap between manure and green manure treatments was smallest.

In this study, soil moisture deficits caused by high clover DM were more damaging to the corn grain yield than N deficits by low clover DM, as seen in the water-limited yields of the first cycle compared to the N-limited yields of second cycle. The drought of 2012 was extreme and illustrates the need for precautions to excessive green manure soil water use. If green manures are to be included in a rotation during the small grain phase, fall termination is advisable. In some forage legume species, mulching can limit DM yield and transpiration over the canopy. Soybeans are more drought-tolerant and might be a better choice than clovers as green manures.

The legacy of high manure applications in these fields has likely led to high amounts of total soil N which are often found in organically managed, manure-based systems (Marriott and Wander, 2006; Poudel et al., 2002) and explain relatively high yields obtained in control treatments in two years. It is possible that total soil N decreases when animal manure is replaced by green manures in a farming system, but this could take several years due to the slow mineralization rates of green manures and demonstrates the need for research that spans several rotations.

High yields in organic production systems can probably best be achieved by a combination of animal manure and green manure as demonstrated by the Iowa long-term trials. Longer periods of green manures improve soil N and subsequent corn yields more than shorter periods. Farmers need to be flexible with green manures, for example supplementing with animal manure, if green manure DM production is insufficient to meet corn N demand. Future research should be directed towards finding optimal application times and rates for manure, as well as optimum times and lengths of green manure periods in the rotation.

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Table 4.1. Estimated corn yield potential for each cycle, assuming water is not limiting. Estimates for yield w/out N fertilizer or N credits were taken from Shapiro et al. (2008) and are based on a yield goal of 150 bu/ac (9.42 Mg ha^{-1}), soil nitrate levels measured at corn planting in 0 – 20 cm and 20 – 60 cm soil depth (chapter 3), and 3% soil organic matter. Yield w/ credits was calculated with the Corn N Recommendations Calculator (Ferguson et al., 2008), based on medium/fine textured soils, previous crop of corn (for manure and control treatments) or clover 0-29 (red and white clover treatments) or soybeans (soybean green manure treatment), soil nitrate levels measured at corn planting in 0 – 20 cm and 20 – 60 cm soil depth (chapter 3), 3% soil organic matter, and credits for N from manure (1 year ago, Fall 1 day application method). Manure = cattle manure, red clover = undersown red clover, white clover = undersown white clover, control = no fertilizer at all, soybean GM = soybean green manure.

Treatments	**Soil nitrate**	**Estimated yield w/out N credits**	**Estimated yield w/ N credits**	**Actual yield**
	ppm	Mg ha^{-1}	Mg ha^{-1}	Mg ha^{-1}
		First cycle		
Manure	59.3	>11.3	17.58	7.61
Red clover	22.3	7.53	11.17	2.83
White clover	22.3	7.53	11.17	4.49
Control	26.0	11.3	6.72	8.37
Soybean GM	30.0	11.3	11.68	6.05
		Second cycle		
Manure	19.0	7.53	6.40	7.60
Red clover	5.3	<3.77	5.46	6.03
White clover	5.3	<3.77	5.46	4.60
Control	12.0	3.77	4.46	8.55
Soybean GM	8.3	<3.77	3.14	6.22
		Third cycle		
Manure	6.0	<3.77	3.14	8.14
Red clover	9.0	<3.77	6.72	7.05
White clover	9.0	<3.77	6.72	6.76
Control	4.0	<3.77	3.14	5.56
Soybean GM	6.3	<3.77	3.77	6.10

Table 4.2. Source of variation for corn emergence and grain yield as affected by type and management of preceding clover. Sample sizes varied from year to year due to changes in the treatment and experimental design.

		Corn emergence	**Corn grain yield**
	Denominator d.f.	**P-value**	**P-value**
Source of variation		First cycle	
Clover	24	0.278	0.012
Mulching	24	0.537	0.432
Termination time	24	<0.001	0.003
Clover x Mulching	24	0.967	0.902
Clover x Time	24	0.837	0.819
Mulching x Time	24	0.967	0.847
Clover x Mulching x Time	24	0.465	0.847
		Second cycle	
Clover	26 (emergence) 16 (yield)	0.938	0.005
		Third cycle	
Clover	18	-	0.497
Mulching	18	-	0.183
Termination time	18	-	0.536
Clover x Mulching	18	-	0.950
Clover x Time	18	-	0.664
Mulching x Time	18	-	0.901
Clover x Mulching x Time	18	-	0.756

Table 4.3. Corn emergence and corn grain yield main effects of treatment (type of clover, mulching and termination time) for each year. Means followed by the same letter are not significantly different at α = 0.1 (Fisher's LSD). Emergence was not counted in the third cycle.

Treatment	**First cycle**		**Second cycle**		**Third cycle**
	Corn emergence	**Corn grain yields**	**Corn emergence**	**Corn grain yields**	**Corn grain yields**
Type of clover	Plants ha^{-1}	Mg ha^{-1}	Plants ha^{-1}	Mg ha^{-1}	Mg ha^{-1}
Red clover	.	2.83[a]	45,200	6.03 [a]	7.06 [a]
White clover	.	4.49[b]	45,000	4.60 [b]	6.81 [a]
Mulching					
Mulched	.	3.91 [a]	-	-	6.83 [a]
Not mulched	.	3.42 [a]	-	-	7.05 [a]
Time of termination					
Fall termination	52,200[a]	4.69 [a]	-	-	7.19 [a]
Spring termination	45,600[b]	2.64 [b]	-	-	6.69 [a]

Table 4.4. Corn yield history of the organic site, in conventional and organic management. With the transition to organic management, this section has been in a soybean-winter wheat-corn rotation with each crop of the rotation present in each year and manure applications in the year before corn (only in manure treatments during study). Yields are combine-harvested, weighed on trailer, not adjusted for moisture. Discrepancies in whole field mean and study mean are due to measurement and fertility differences across each field (i.e. sites in each field that were very wet, weedy, or shaded by windbreaks) were not included in study mean, but are included in field mean. *NA, not available; **W.w., winter wheat; *** field borders around study plots were manured.

	Year	**Field Nr.**	**Preceding crop**	**Corn yield Mg ha^{-1}**	**Comments**
Conventional	1992	2	NA*	7.54	
	1993	2	Corn	7.92	Mean of two values
	1998	2	NA	10.44	
	1998	3	NA	13.78	
	1998	56	W.w.**	12.27	
	2001	3	NA	6.58	
	2003	789	W.w.	7.47	
	2005	14	NA	7.78	
	8-year conventional mean:			**9.22**	
Organic	2006	789	W.w.+ manure	10.17	
	2007	2356	W.w. + cover crop or manure	6.72	big windstorm in August
	2008	14	W.w.+ cover crop or manure	7.66	
	2009	789	W.w.+ manure	10.80	
	2010	2356	W.w. + manure	6.60	September hail
	2011	14	W.w.+ manure	10.24	
	6-year organic mean			**8.70**	
	Organic mean without storm/hail years			**9.72**	
	2012 (field mean)	789	W.w. + manure on borders + study	4.42	Study in 90% of field
	2013 (field mean)	2356	W.w. + manure on borders + study	5.32	Study not in 2, west 56 (wet)
	2014 (field mean)	14	W.w. + manure on borders + study	5.91	Study in 80% of field
	Field mean			**5.22**	
	2012 (study mean)	789	W.w. + study	5.87	Very low yields after clover
	2013 (study mean)	356	W.w. + study	6.60	
	2014 (study mean)	14	W.w. + study	6.01	
	Study mean:			**6.16**	

Table 4.5. Source of variation for corn grain yield and corn emergence as affected by organic soil amendment (undersown green manure, post-wheat green manure, manure or control).

	Numerator d.f.	**Denominator d.f.**	**Corn emergence**	**Corn grain yield**
			P-values	
		First cycle		
Soil amendment	4	35 (yield) 39 (emergence)	0.073	<0.001
		Second cycle		
Soil amendment	4	25 (yield) 37 (emergence)	0.789	<0.001
		Third cycle		
Soil amendment	6	51	-	<0.001

Table 4.6. Operational costs, income and profits for different soil amendments for corn. Numbers are based on the management operations and green manure seed costs in the third cycle. Costs for management operations are taken from University of Nebraska extension publications, corn seed, and sales price for organic corn are taken from extension publications of Iowa State University (see below). Green manure seed costs are the prices for green manures seeds in the third cycle (table 2.1).

Management operation	**Red clover**	**White clover**	**Alfalfa**	**Sweet clover**	**Soybean GM**	**Cattle manure**	**Control**
				$ $ ha^{-1}$			
Green manure seed 22 or 13.5 kg ha^{-1}	188	356	233	109	165	0	0
Spreading Seed or manure broadcast	13	13	13	13	16	84	0
Disking $28 ha^{-1}	56	56	56	56	112	112	112
Field cultivate $22 ha^{-1}	22	22	22	22	22	22	22
Corn seed 75,000 seeds ha^{-1}	191	191	191	191	191	191	191
Plant $33 ha^{-1}	33	33	33	33	33	33	33
Row cultivate $21 ha^{-1}	42	42	42	42	42	42	42
Combining $91 ha^{-1}	91	91	91	91	91	91	91
Total operational cost	636	804	681	557	672	575	491
				Mg ha^{-1}			
Corn yields	7.05	6.76	7.64	7.15	6.10	8.14	5.56
				$ ha^{-1}			
Income from corn sales, $512 Mg^{-1}	3610	3461	3912	3661	3123	4168	2847
Profits Income – Operational costs	3031	2715	3288	3161	2453	3594	2357

Source of operational costs, corn seed costs and corn sale prices:

Klein, R.N., R.K. Wilson, and J.Johnson. 2014. Crop budgets. Nebraska – 2015. EC872. http://www.ianrpubs.unl.edu/epublic/live/ec872/build/ec872.pdf

Organic crop production enterprise budgets. ISU Extension. https://www.extension.iastate.edu/agdm/crops/html/a1-18.html

Wilson, R.K. 2014. 2014 Nebraska farm custom rates - part 1. EC823. http://www.ianrpubs.unl.edu/live/ec823/build/ec823.pdf

Figure 4.1. Corn in first cycle (June 13, 2012). Planter problems caused gaps in the rows.

Figure 4.2. Corn height after clover (above) versus corn after dairy manure (below) on June 20, 2012. In the top picture, corn height is about 70 cm as measured on the pole and in the bottom picture, it is about 110 cm.

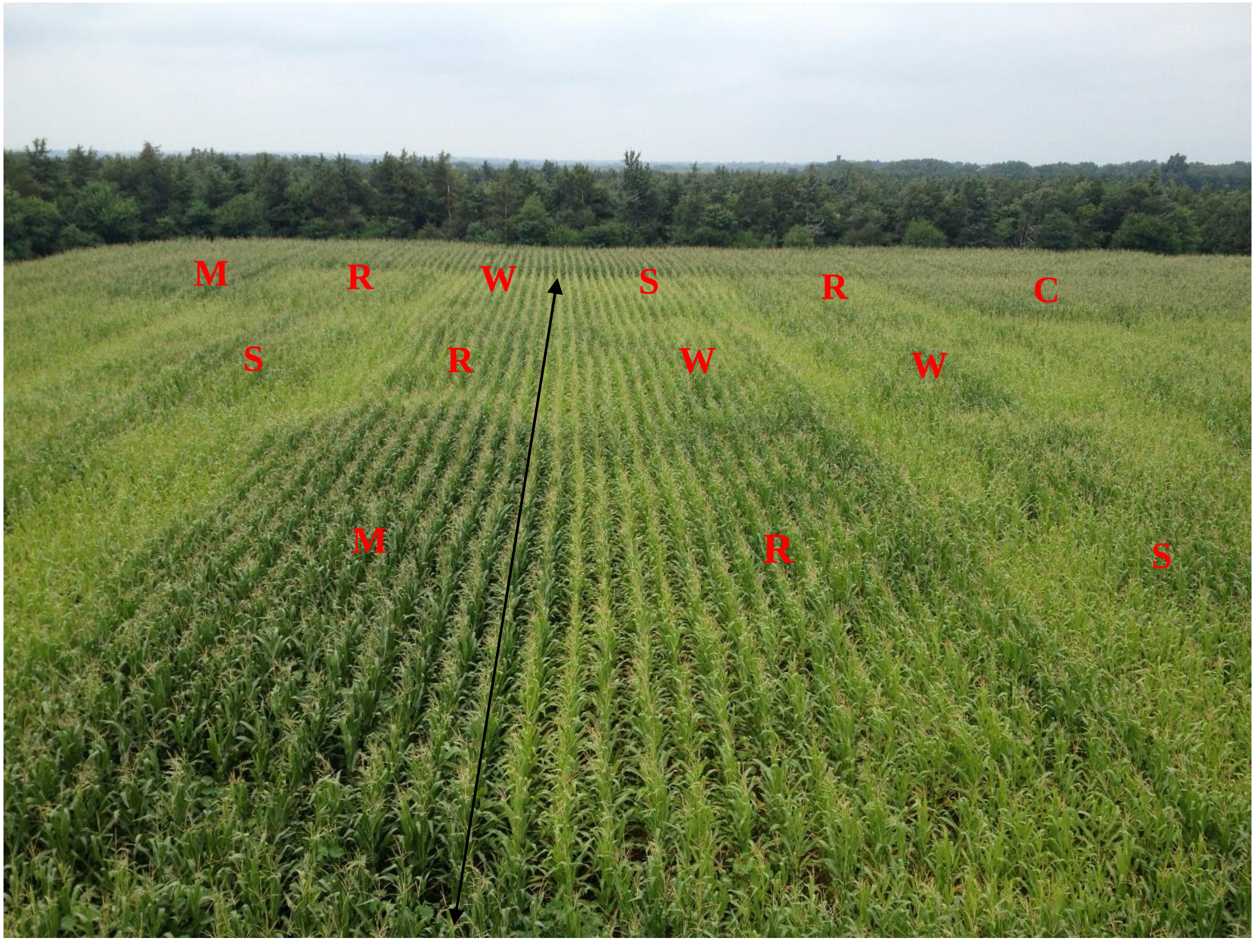
M
R
W
S
R
C
S
R
W
W
M
R
S

Figure 4.3. Variation in corn height and color after soil amendments. Field 3 (above) and field 56 (below) in 2013. The previous year's treatments are indicated with letters: M = manure, R = red clover, W = white clover, S = soybean green manure, C = control. Note the dark green color of both manure and control treatments. The black line indicates the length of a block (91.4 m) and letters indicate the treatments assigned to each of the three plots per block. Plots were 9.1 m wide and 30.4 m long and contained 12 rows of corn. Blocks were 3 m apart from each other. No fertilizer or tillage operations were carried out between the blocks and these gaps appear yellow as well. Manure was spread around the experimental area, thus the dark green color along the windbreaks and in the far back of field 56.

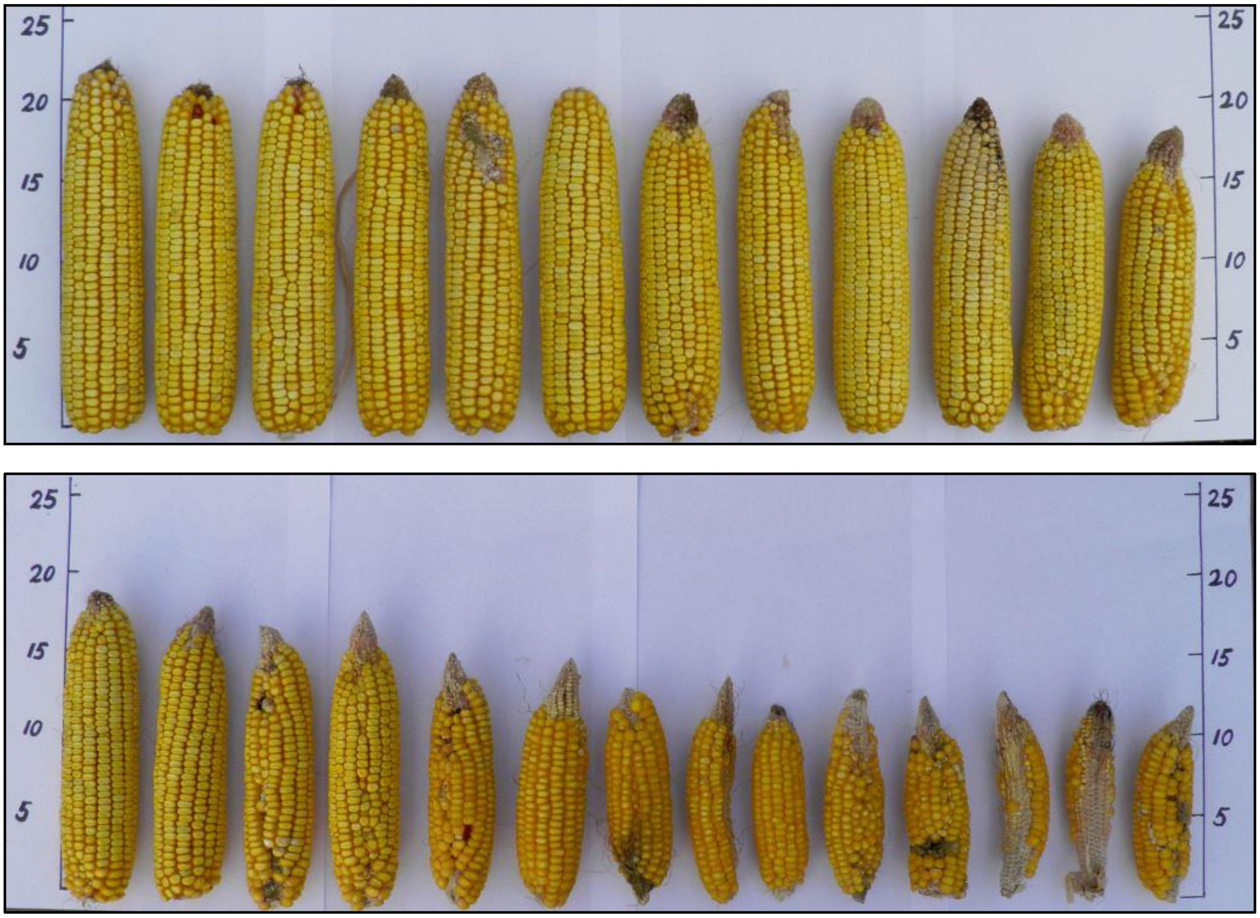

Figure 4.4. Differences in corn cob development in the first cycle (drought year). Cobs in control treatment (above) and after red clover (below). Scale on the ride and left side is in cm.

Figure 4.5. Weed infestation at corn harvest in manured plot (left) and clover plot (right) (first cycle). All plots had received the same weed control operations.

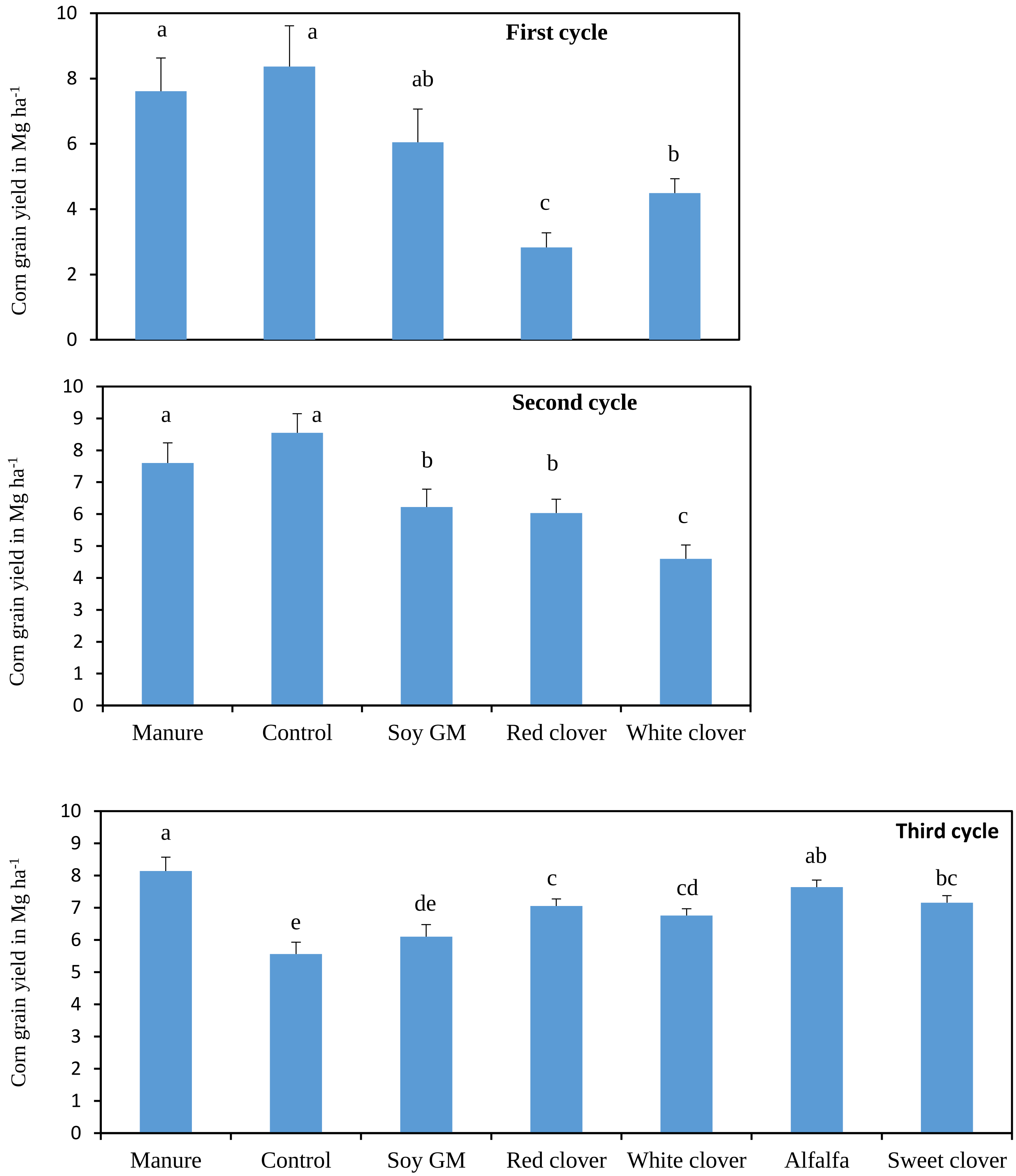

Figure 4.6. Corn grain yields after different soil amendments (soy GM = soybean green manure). Yields are presented for each cycle. Means that are not different at $\alpha = 0.1$ (Fisher's LSD) are indicated with the same letter.

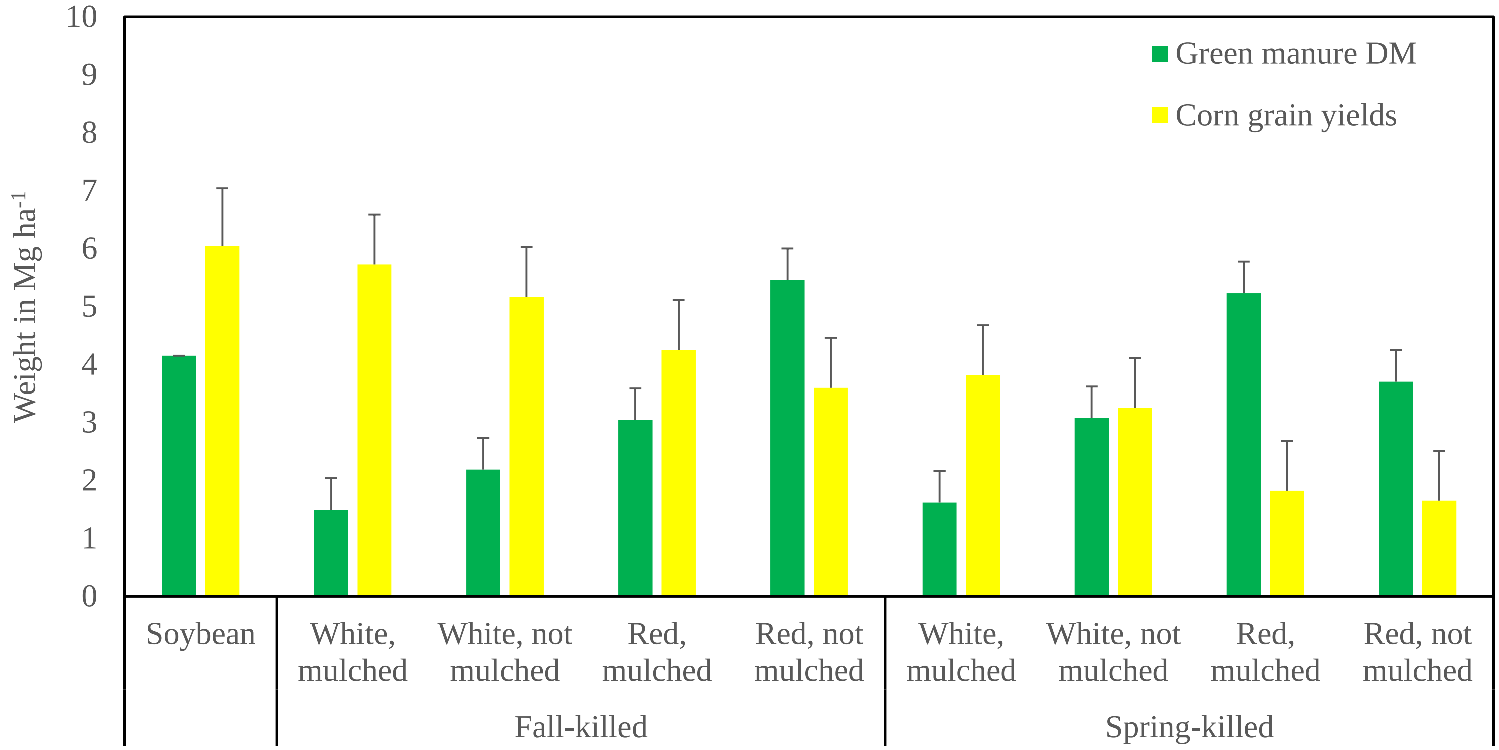

Fig. 4.7. Corn grain yields after green manures in the first cycle. Mulching clovers had a significant effect on clover DM yields, but not on corn yields.

CHAPTER 5

WEED SUPPRESSION OF LEGUMINOUS GREEN MANURES IN AN ORGANIC SOYBEAN-WINTER WHEAT-CORN ROTATION

Leguminous green manure crops that occupy the otherwise fallow period after winter wheat harvest in organic rotations with a small grain can increase soil nitrogen, soil organic matter, and subsequent crop yields (Snapp et al., 2005; Schipanski and Drinkwater, 2011). Replacing the fallow period with a green manure also eliminates tillage for weed control during fallow periods. Tillage is widely used for weed control in organically managed farms but is labor- and fuel intensive and can increase the risk of erosion (Carr et al., 2012). Even where the risk of erosion is small, the impacts of tillage on soil quality are stark: loss of soil organic matter, soil structure and aggregation, as well as the disruption of beneficial soil microorganisms such as fungi and earthworms (Triplett and Dick, 2008). However, to be able to replace tillage, the green manure species must be able to compete with weeds in order to carry out its purpose of biological N fixation and dry matter production. Further, if weeds are able to establish and proliferate during a green manure period, such as by depositing seeds or rhizomes, they can intensify weed problems for subsequent crops. Thus, it is important to assess green manures for their weed control potential.

Plants compete by consuming resources such as water and nutrients more efficiently, reducing light availability (shading), and releasing allelopathic compounds (Liebman and Dyck, 1993). Many of the characteristics of an ideal green manure, such as high DM productivity or quick growth, also make them efficient at competing with weeds (Brust et al., 2014).

Legume species such as red clover (*Trifolium pratense* L.), white clover (*Trifolium repens* L.) and soybean (*Glycine max* [L.] Merr.) can be excellent green manures, because of their high DM production, N fixation, and positive effects on subsequent corn yields (Cherr et al., 2006; Amossé et al., 2014; Yang et al., 2014). In soybean-winter wheat (*Triticum aestivum* L.)-corn (*Zea mays* L.) rotations in the Midwest, clover green manures are often established by undersowing into winter wheat in the spring, to take advantage of the higher soil moisture. Clovers are winter-hardy in the Western Corn Belt and can grow until their termination the following spring.

Undersown and overwintered red clover suppressed weeds by 99% compared with a control in trials in South Dakota (Anderson, 2015). White clover, a long-lived perennial, has demonstrated weed control when used as a perennial living mulch in orchards and vineyards (Hartwig and Ammon, 2002), but might be less effective when grown for shorter periods of time. In living mulch vegetable systems in the Netherlands, white clover reduced weeds less than red clover, but also impacted the crop less than red clover (Den Hollander et al., 2007). The suppressive ability of clover depends on several factors, including weed species. Red and white clover were not able to suppress brown mustard (*Brassica juncea* [L.] Czern.) in two-year study on a high-fertility site in Canada, because they were much smaller. Berseem clover (*Trifolium alexandrinum* L.) and Alsike clover (*Trifolium hybridum* L.) suppressed weeds better, probably because they grew taller (Ross et al., 2001). The same study tested the effect of mowing on clover and weed biomass production, and found that on the high-fertility site, red and white clovers regrew faster than the brown mustard after mowing, reducing mustard biomass by about 75% (red clover) and 25% (white clover). Mowing is recommended to prevent weed seed set in green manures, but can delay clover development (Drangmeister, 2003).

A cover crop planted after winter wheat harvest in the Western Corn Belt needs to be able to tolerate high temperatures and low soil moisture. Soybean is well adapted to this area, and might fare better than cover crops more typically used. It winterkills, eliminating the need for mechanical termination. Because of its shorter growing season (appr. July through October) it uses less soil water than undersown green manures, alleviating grower concerns over cover crop soil water use. In previous trials on this site, a soybean cover crop resulted in higher corn yields than berseem clover, Austrian winter peas (*Pisum sativum* L.), cow peas (*Vigna unguiculata* [L.] Walp.) or hairy vetch (*Vicia villosa* Roth) (Brandle, unpublished data). However, the effects of a soybean cover crop on weed growth were not investigated.

Our objectives were to compare the weed suppression potential of red clover, white clover and soybean grown as green manures in the wheat phase of an organic soybean-winter wheat-corn rotation. Red and white clover were undersown into the winter wheat, whereas soybean was planted after winter wheat harvest. In addition, the effect of mowing on the undersown green manures was investigated. The hypotheses were i) undersown green manure will suppress weeds more than soybean green manure, as they have a longer growing season; ii) among undersown species, red clover will suppress weeds more than white clover; iii) mowing clover green manures will improve weed suppression.

MATERIALS AND METHODS

For a detailed description of the site, soils, rotation, experimental design and management operations, see Chapter 1 and 2.

Red clover was frost-seeded (broadcast onto frozen soil) into winter wheat in March at a rate of 22 kg ha^{-1} and white clover was frost-seeded at a rate of 13 kg ha^{-1}. After winter wheat harvest, soybean was planted at a rate of 100 kg ha^{-1} as a cover crop in some of the plots that had no undersown green manure. In the third cycle, two additional, more drought tolerant undersown green manures were tested: alfalfa (*Medicago sativa* L.) and sweet clover (*Melilotus officinalis* L.). Chapter 2 illustrates clover growth and DM production. Half the clover plots were mulched (mowed with the plant residue left in place) 40 days after wheat harvest at a height of 0.1 m. To prevent weeds from going to seed, the other clover plots were mowed at a height of 0.3 m which cut the heads of tall weeds but did not defoliate the clover. Alfalfa, sweet clover and soybean green manure plots were not mowed.

Above-ground weed biomass in the red and white clover plots was sampled at the same time clover biomass was sampled (Chapter 2), at wheat harvest, 35 days post-harvest, at clover fall termination, and spring termination (table 1.2). Only results from the sampling at fall and spring termination are presented here. Above-ground weed biomass in the soybean cover crop was sampled once, at fall termination. Weed biomass was not sampled in the other treatments, as these were kept weed-free by disking. Thus, there is no green-manure free weedy control for comparison purposes. While it is instructive to have a control treatment to determine how much weed DM would have been produced without any weed control; in practical terms, producers will not (and should not) allow weeds to grow during a fallow period. In this farming system, the alternative to using green manures as weed control is clean cultivation of the fields.

Weed dry matter production was determined by placing a 0.1 m^2 quadrat in three randomly selected areas in each plot. All vegetation within the quadrat was cut to ground level, sorted into clover and weeds, dried at 65˚C to constant weight and then weighed. The most frequent weed species were noted, but weed DM was not determined for individual species, nor were all weed species identified.

Weed dry matter was analyzed with ANOVA implemented using the GLIMMIX procedure in SAS 9.4 (SAS Institute, Cary, NC). Species, mulching, and termination time and their interactions were fixed effects and block was the random effect. Values for alfalfa, sweet clover and soybean green manure were not included in the ANOVA. Least-square means were compared with the relatively conservative Tukey or Tukey-Kramer (for unequal sample sizes) tests using a significance level of $\alpha = 0.05$.

RESULTS AND DISCUSSION

Weed growth was higher in the years with high clover growth, probably because growing conditions that favor green manure growth also favor weed growth. However, year was not used as a factor in the ANOVA. Figure 5.1 shows the weed DM produced at each sampling time for each green manure. In the first cycle, at fall termination, type of clover was not significant, but mulching significantly lowered weed DM in both red and white clover treatments (table 5.1). The interaction between type of green manure and mulching regime was not significant. Weed DM at this sampling time was higher than at any other sampling time, almost 1.2 Mg ha^{-1} in the unmulched red clover, 1.4 Mg ha^{-1} in the unmulched white clover, and 1.13 Mg ha^{-1} in the soybean green manure (data for soybean green manure not shown). Surprisingly, clover DM was also high (table 2.4), almost 5.5 Mg ha^{-1} for the unmowed red clover, and 4.5 Mg ha^{-1} for the soybean green manure (data for soybean green manure not shown). Red clover stands with much lower DM yields have been effective at suppressing weeds, for example in South Dakota, undersown red clover DM in mid-September was about 1.5 Mg ha^{-1} and weed DM was less than 0.01 Mg ha^{-1} (Anderson, 2015). Clover DM in our study was low at wheat harvest, with little competitive advantage over weeds, and because of sufficient rainfall, water was not limiting plant growth (Liebman and Dyck, 1993).

The weed-suppressing effect of mulching is in line with results by Ross et al. (2001). The same study also observed that the weed growth stage at mowing influenced weed regrowth. When weeds (in this case, brown mustard) were mowed at late flowering, they did not regrow. In our study, the most common weed species after wheat harvest were pigweed (*Amaranthus* ssp.), lambsquarters (*Chenopodium album* L.) and volunteer wheat (self-sown kernels lost at wheat harvest). The summer annual weeds were probably more harmed by mulching than volunteer wheat, as they were in later stages of development (flowering) and did not regrow.

At spring termination, type of clover and mulching had a significant effect on weed DM (table 5.1). Red clover had suppressed virtually all new weed growth, with no weeds found in the unmowed plots, and 0.03 Mg ha^{-1} in the mowed plots. Red clover growth was initiated early in the spring due to warm temperatures. Red clover DM production was high, stands were uniform, with a dense canopy, preventing light from reaching the ground. As a result, weeds were not able grow. White clover stands were also productive in terms of DM yield, but weed DM, comprised mostly of volunteer wheat was almost as high as clover DM (figure 5.2). White clover is not competitive with grasses (Black et al., 2009), which is why it is most often grown with a companion grass in pastures or grass-clover leys (Oberson et al., 2013).

In the second cycle, weed DM was impacted by drought conditions. High temperatures in the spring had accelerated winter wheat development, and it was harvested about three weeks earlier than normal. After the removal of the wheat canopy, weeds did not grow as rapidly as in the first cycle, because of the lack of precipitation in July and August, combined with higher than normal temperatures (see chapter 4). Undersown green manures were not mowed, as they failed to develop more than 1 Mg DM ha^{-1} at any of the sampling times. At fall termination, weed DM was very low and similar in the red clover plots (0.23 Mg ha^{-1}) and white clover plots (0.38 Mg ha^{-1}), although some winter annuals such as Shepherd's purse (*Capsella bursa-pastoris* [L.] Medik.) were found that had probably emerged after rainfall in September and October. The DM yield of the soybean green manure was more than 2 Mg ha^{-1} but weed DM in the soybean treatment was 1.02 Mg ha^{-1} (data not shown), the highest among the treatments (figures 5.3, 5.4, 5.5). In the spring, weed DM was higher than in the fall, and not significantly impacted by type of clover. Weeds consisted of overwintered volunteer wheat and winter annual weeds such as field pennycress (*Thlaspi arvense* L.) that emerged due to normal amounts of precipitation in April (figures 5.6 and 5.7).

In the third cycle at fall termination, type of clover had a significant impact on weed DM, but mulching or their interaction did not (table 5.1). Red clover plots had 0.02 Mg weed DM ha^{-1} in the mulched and even less in the unmulched plots, and red clover green manure DM was highest (figure 5.8). Alfalfa green manure produced the second highest amount of DM, and reduced weed DM yields the second most (0.11 Mg ha^{-1}). Alfalfa controlled weeds better than red clover in semiarid regions in Canada (Blackshaw et al., 2010), but in Iowa, red clover reduced weed density more than alfalfa, although weed DM was similar between the two (Blaser et al., 2011). In our study, sweet clover and white clover green manures yielded similar amounts of DM and had the most weed growth, although weed biomass in each treatment was less than 0.7 Mg ha^{-1}.

Sweet clover reduced weed biomass by at least 75% compared to a weedy control in each of three years in a study in Canada. At sweet clover termination time in June of its second year, weed biomass was between 1 and 12% of total biomass (Blackshaw et al., 2001). This ratio of weed DM to sweet clover DM was much less favorable in our study in the fall, as weeds constituted about one fourth of the total plant biomass. The soybean cover crop failed to establish and weed DM was not sampled in these plots. In the spring, weed biomass was below 0.2 Mg ha^{-1} in all treatments and green manure biomass DM was also low (between 0.16 Mg ha^{-1} for sweet clover and 0.85 Mg ha^{-1} for red clover). Weed DM was not affected by type of undersown green manure, mulching or their interaction (table 5.1). Weed DM was highest in sweet clover (0.19 Mg ha^{-1}) and lowest in mulched red clover (0.06 Mg ha^{-1}). Very cold winter temperatures and the lack of snow cover probably delayed the emergence of annual weeds, and could have killed some volunteer winter wheat.

Of all green manures tested, red clover showed the best weed suppression. White clover did not suppress weeds as well, even in years with high white clover DM production. Because of its smaller size and slower growth, it is less competitive especially when it must compete with grasses such as volunteer wheat. Soybean was not an effective weed control because it did not develop a closed canopy although it yielded as much biomass as red clover in the first cycle, and much more than the clovers in the second cycle. Selecting soybean varieties or other cover crops suited that produce high amounts of biomass when planted after winter wheat harvest is important. In a Kansas study, a late maturing soybean variety yielded more than 5 Mg DM ha^{-1} in three of four years when planted as a cover crop after wheat. In the same study, sunn hemp (*Crotalaria juncea* L.) outyielded soybean in each year (Blanco-Canqui et al., 2012), however, weed suppression was not measured in this study. Tartary buckwheat (Fagopyrum tataricum [L.] Gaertn.) has been identified as a species for short-term, post-wheat harvest summer cover cropping because it grows fast even with limiting soil water and has high weed suppression potential (Brust et al., 2014).

Alfalfa and sweet clover were only tested in one year, but alfalfa was more competitive with weeds than sweet clover. In our study, it was not always clear whether high green manure DM production led to lower weed DM. Weather conditions, such as higher than normal precipitation, also increases weed growth and lessens competition for resources such as soil water. However, the ability to produce a dense, closed canopy that eliminates light transmittance to the soil surface was observed to result in much less weed biomass. Future research on the weed suppression potential of green manures should measure canopy light transmittance as this could help identify species with suitable canopy architecture.

In the context of finding weed-smothering cover crops for organic no-till systems research has focused on the weed suppressing ability of cover crops or green manures after they are killed (Carr et al., 2012). Whether the green manures in our study reduced weed growth in the following corn crop is not clear as we did not measure weed emergence and growth after green manure termination. Weeds in the corn were controlled by tillage. In the first cycle, soil moisture deficits after the clovers stunted corn growth. Because corn did not grow tall and did not close its canopy, it did not suppress weed emergence after mechanical weed control had ceased. High secondary weed infestations occurred in these plots.

One disadvantage of using undersown green manures in this rotation is the growth of volunteer wheat. This occurred in each year of our study, and comprised at least half of the weed DM. Volunteer wheat that emerges after winter wheat harvest can harbor a number of disease vectors, for example aphids which transmit Barley Yellow Dwarf virus and eriophyid mites which spread Wheat Streak Mosaic virus (Brakke, 1987). Winter wheat is usually planted in late September or October in this area, and can become infested with aphids and mites migrating in from volunteer wheat. To avoid disease infestations of newly planted winter wheat fields, producers must prevent volunteer wheat emergence. While a dense crop canopy after wheat harvest, as observed in the third cycle, can likely reduce the further growth of volunteer wheat, it is important to prevent the loss of wheat kernels at harvest by adjusting the combine. However, for organic producers, the surest method to destroy volunteer wheat is tillage.

CONCLUSION

Producers considering introducing green manures need to take into account how well the green manure can compete with weeds that will emerge if no other weed control operations are carried out. In our study, undersown red clover suppressed weeds better than any other green manure. Mulching or mowing did not always significantly decrease weeds, but it is an essential tool in preventing seed development or dispersal in taller weed species and should be part of the green manure management. However, when to mulch or mow is best determined by identifying the developmental stage of the weed species. Soybean, the only green manure species planted after wheat harvest, did not suppress weeds as effectively as red clover. Green manure DM production is important for weed DM reduction, but so is the ability of the green manure to exclude light transmittance to the soil surface. Research to find species that have this ability, while using less soil moisture than red clover, is needed, especially in the drier areas of the Midwest.

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Table 5.1. Weed dry matter in clover green manure at fall termination and spring termination. Mulching was carried out only in the first and third cycle, once at 40 days after wheat harvest, or not at all. Clover failed to grow in the second cycle due to the 2012 drought. Soybean green manure, alfalfa and sweet clover were not included in this analysis.

		Weed DM in Mg ha^{-1}					
		First cycle		Second cycle		Third cycle	
Clover type	Mowing	October	April	October	April	October	April
Red	unmulched	1.19	0.00	0.23	0.58	0.01	0.09
	mulched	0.31	0.03			0.02	0.06
White	unmulched	1.30	1.57	0.38	0.69	0.31	0.07
	mulched	0.78	3.08			0.61	0.16
P-value	Clover type	0.146	0.056	0.111	0.058	<0.001	0.323
	Mulching	0.002	0.047	-	-	0.150	0.627
	Interaction	0.56	0.027	-	-	0.186	0.220

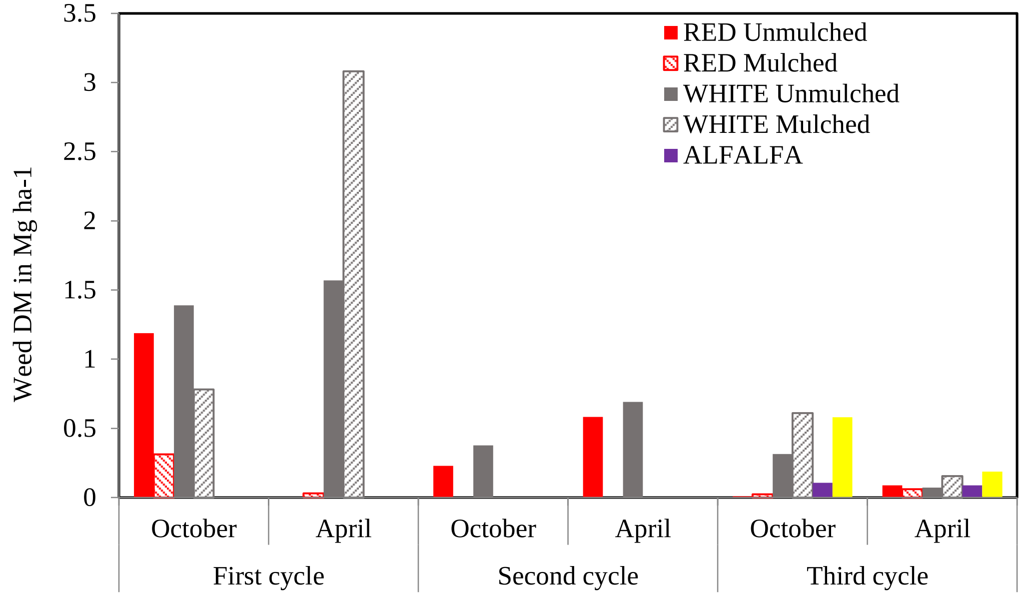

Figure 5.1. Weed dry matter of undersown green manures at termination time in each cycle. Alfalfa and sweet clover were only grown in the third cycle and were not mowed.

Figure 5.2. Volunteer wheat (yellowish) in unmowed white clover in the spring. Notice dead weed biomass.

Figure 5.3. Weed and clover growth at fall termination in the second cycle. Small white clover (probably emerged from hard seed after drought conditions eased), volunteer winter wheat, and Shepherd’s purse in clover plot on November 7, 2012.

Figure 5.4. Weed growth in soybean green manure plots at fall termination in the second cycle. The weed community was comprised mostly of volunteer winter wheat.

Figure 5.5. Weeds in red clover plots at fall termination in the second cycle.

Figure 5.6. Weeds, mostly field pennycress and volunteer winter wheat in white clover plots at spring termination in the second cycle.

Figure 5.7. Weeds and red clover in red clover plot at spring termination in the second cycle.

Figure 5.8. Weeds in red clover plots before mulching in the third cycle (August 20). Pigweed (*Amaranthus* ssp) with seed heads and velvetleaf (*A. theophrasti* Medik.) are growing in the back.

CHAPTER 6

SUMMARY

This study was conducted to find answers to the following research questions:

1. Do forage legumes green manures, undersown into winter wheat, increase cash crop yields in an organic soybean-winter wheat-corn rotation compared to post-wheat cover crops or post-wheat manure applications?
2. Do undersown forage legume green manures decrease weed pressure?
3. Do undersown forage legume green manures increase soil nitrate levels after termination?

Specific research questions and hypotheses are addressed in each of the previous chapters, but here I present overall conclusions drawn from this research. Very variable weather patterns, including a drought in the second cycle and a very cold, dry winter in the third cycle, characterized the four-year study period and influenced forage legume growth. In two seasons with above normal precipitation during the growing season, forage legumes grew well and produced DM yields comparable or higher than those regions with higher precipitation. However, in the drought year, red and white clover crops failed. Despite winter hardiness, all forage legumes suffered from winterkill in the third cycle, probably exacerbated by very dry conditions. Red clover was the most reliable DM producer, twice yielding more than 5.5 Mg ha^{-1}. White clover always had the lowest DM, although it produced 3 Mg ha^{-1} after a mild winter in the first cycle. Alfalfa and sweet clover, which were only grown in one year, were intermediate. The clovers did not impact winter wheat growth or yields, but alfalfa and sweet clover grew tall enough to obstruct wheat harvest, and for that reason, might not be a good choice for undersowing into winter wheat. Red clover reduced wheat grain protein in the last cycle.

High green manure DM yields are important, because they determine how much N is fixed and added to the soil for corn. However, high green manure DM production in our study also had negative effects, because of high water deficits incurred. Corn yields after forage legumes were limited by water, especially in the first cycle, when corn after highly productive red clover stands had stunted growth. Spring termination led to especially low red clover yields (1.7 Mg ha^{-1}). Corn yields after green manures were also N limited, especially in the third cycle, when DM yields were very low in the spring before incorporation. Corn yields reached 7.6 to 8.1 Mg ha^{-1} after cattle manure. They were always significantly lower for red clover (2.8 Mg ha^{-1}, 6 Mg ha^{-1} and 7 Mg ha^{-1} in the first, second and third cycle) and white clover (4.5 Mg ha^{-1}, 4.6 Mg ha^{-1}, and 6.8 Mg ha^{-1} in the first, second and third cycle). Alfalfa and sweet clover yields were 7.6 Mg ha^{-1} and 7.2 Mg ha^{-1}, respectively. It is difficult to obtain high corn yields using green manures alone, because they often do not produce enough N for the corn, or N is not released from decomposing green manures in synchrony with corn N demand. Further, green manure soil water use can be more damaging than insufficient N for the corn crop.

Green manures did not increase soil nitrate levels, but manure did. However, soil nitrate was not sampled during corn growth, so N release from the green manure DM was not known. Green manures had lower soil nitrate levels during their growth, and after corn growth and could possibly be used to take up excess N remaining after corn harvest.

Forage legumes, especially red clover, suppressed weeds very well. If mowed, farmers can expect almost 100% weed suppression in red clover stands. White clover stands were not competitive with volunteer wheat, which could lead to the transfer of virus diseases to newly planted wheat fields if disease vectors take refuge in volunteer winter wheat growing in green manure stands.

For a grower considering the introduction of green manures, two main concerns are the lack of soil N and/or the lack of soil water after the green manures are incorporated. Early termination, for example in the fall, can allow for soil water recharge. Lack of N, for example due to failed growth of the green manure, can probably be corrected by applying manure before corn growth.

Other studies have found that the continuing use of green manures can improve soil water holding capacity and soil organic matter, and help stabilize the system in drought years. However, farmers might not have the financial freedom to wait several years for this system to work. While green manures can have many benefits, such as weed control, as well as others not investigated in our study, high corn yields in an organic cash crop rotation were maintained with the application of cattle manure.

LIMITATIONS

Broad inferences from this study are limited because of errors made in the experimental design as well as in measurement. Using a randomized complete block design continuously would have made comparisons across years much easier. Yields were not adjusted for moisture, which also makes comparisons across years and with other studies difficult, because actual yields could be several percent higher or lower than those measured. However, in reality, the differences are likely minor, as grain was always harvested at maturity.

To explain with more certainty the reasons for corn yield losses after undersown green manures, we need information on the total water use and soil water use of green manures. This could have been carried out with measurements of soil water at several depths in the soil profile during the green manure as well as corn phase. It is also difficult from this data to calculate how much N actually entered and left the system. If manure, corn plant tissue, clover plant tissue and corn and wheat grain would have been analyzed for N and C each year, an N balance could be calculated. This would still not account for N leaching losses, or N volatization losses which were beyond the scope of this project. To make recommendations to farmers, it would also have been useful to test these green manures on farms in Eastern Nebraska, with different management systems, different soils and climates.

REFLECTION

This study has been an attempt to track the effects of several types of organic soil amendments throughout a rotation, to understand their interactions with the present and subsequent crops, soil nitrate concentrations and weed community. By measuring several distinct parameters, I have attempted to shed light on the connections between these variables of an agroecosystem, because in farming, like in the rest of the natural world, all things are connected. It is not to dismiss the merits of conventional agriculture to say that the understanding of some of these connections has been lost. It is not to undermine science to say that some agricultural research has focused on short-term gains and ignored long-term harm. It is not a call for a revolution in farming to say that we should change a few things. We should change a few things.

Organic agriculture relies on ecological and biological processes to maintain and improve soil fertility (Vogt, 2007). Some methods of organic farming, such as crop rotations, biological nitrogen fixation by legumes, recycling of nutrients, and mechanical weed control, are practiced by all organic (as well as many conventional farmers), but it really is the complexity and diversity of methods that contributes to the success of an organic farm. However, complexity and diversity are not usually a goal in conventional agriculture, nor are they easy to research for the scientific community, nor is it intuitive to solve problems by making things more complex. We have a penchant for simplicity.

The difficulty but also the fun (I prefer to call it a challenge) in this research project has been to follow and separate some of these connections, in order to say: A causes B. That did not happen very often, because most times A caused C and C together with some unknown variable caused B. Or no effect of A on B was observed, but maybe this is due to limitations in the statistical design that did not allow us to find the significant differences, because after all "Everything is different from everything else" (Casler, 2015). Or we were not able to look at something long enough to discover a difference. I am glad to have been able to extend the sampling season by one year, because the insight gained from one additional year of data changed the conclusions I had drawn until then.

This is the exciting thing in agronomic research: Every year is different. Every field is different. Even in a stand of genetically similar corn hybrids no two corn plants are the same. Yet we conduct our research to make inferences that generalize and summarize, that reduce the complexity, that categorize things as being the same or not the same, so that we can say: A causes B. Or: A does not cause B. So what inference can I make after four years of experiments?

For the purpose of being able to make recommendations based on my research, I learned it is important to have a goal, and then carefully select the methods to achieve this goal, keeping in mind the method's long-term effects, as well as side-effects. If the goal of a grower is to improve corn yields in an organic rotation in the Western Corn Belt, I would recommend cattle manure over leguminous green manures, as it increases yields both in the short-term and the long-term. If cattle manure is not an option, a soybean cover crop is preferable over undersown clovers, because their side-effect is high use of soil water. If the goal is to reduce tillage for weed control, then an undersown red clover is preferable over soybean cover crops.

I also learned that in farming, and organic farming especially, many things need to fulfill more than one purpose. For example, a clover stand grown as a green manure needs to be able to suppress weeds, otherwise it will create more problems than it solves. I did not have a good grasp on how important complexity and diversity are in the design of organic farming systems. Maybe our objects (see Chapter 1) could have been achieved with a combination of undersown clover and cattle manure, applied at a different rate (in the case of the manure) or time during the rotation. The clover could control weeds without tillage, preventing erosion, soil nitrate leaching and preserving organic matter. The manure would maintain high crop yields, as well as high organic matter and total soil nitrogen. More diversity in the selection of green manure, for example using a mix of species with varying degrees of drought tolerance, could improve green manure establishment in locations with variable weather (Wortmann et al., 2012).

Lengthening the period of clover growth could improve soil quality and subsequent crop yields further. A soybean-winter wheat-corn rotation, where the winter wheat was undersown with alfalfa, and the alfalfa remained for two years, had similar or higher profitability than shorter rotation without alfalfa leys but much higher inputs of synthetical fertilizers and pesticides (Davis et al., 2012). In South Dakota, fascinating research to reduce weeds in no-till organic farming systems has led to the design of nine-year rotations, where two years of summer annual crops are followed by two years of winter annual crops, another two years of summer annual crops and three years of a perennial forage such as alfalfa (Anderson, 2015). The old practice of clover leys, which helped medieval Europe increase its agricultural productivity (Kjærgaard, 2003) and is still the backbone of integrated farming systems in Europe (Drangmeister, 2003), should be reexamined in the United States as well. The clover or other types of perennial forages grown as leys not only increased soil fertility, but also supported livestock. It is my conviction that in order for organic farming to truly rely on and foster biological processes as the basis for the health of the soil and the health of the food grown from it, we must return livestock to the farm. Sir Albert Howard, one of the pioneers of organic farming, said: “The main characteristic of Nature’s farming can therefore be summed up in a few words. Mother earth never attempts to farm without livestock; she always raises mixed crops; great pains are taken to preserve the soil and to prevent erosion; the mixed vegetable and animal wastes are converted into humus; there is no waste; the processes of growth and the processes of decay balance one another; ample provision is made to maintain large reserves of fertility; both plants and animals are left to protect themselves against disease” (Howard, 1943, p. 4).

If organic farming is to follow the principles laid out by Sir Albert Howard, we need to make some changes. For me, the most important ones are to integrate animal husbandry with crop production. Perennial forage legumes, such as alfalfa and clover, should be reintroduced into rotations, grown both as a forage and for soil improvement. The improvements in soil quality, farm profitability, and environmental health could be immense. In my future career, I would like to conduct research in this area.

However, there are many critics that claim that organic farming principles such as using multi-year leys, or feeding cattle forages (for example, pasture-based), are reasons for low yields, higher land requirements, lower efficiency and higher prices of organic food production (Connor, 2013). Organic proponents argue that organic yields in fact are high or at least not as low as assumed. Seufert et al. (2012) in a large meta-analysis found that overall organic yields were 25% lower than conventional yields, but depended on the type of crops among other factors. The yield gap between organic and conventional agriculture is real, and growing (Posner et al., 2008). Just as worrisome for me, a consumer of organic products, is the price gap between organic and conventional foods. Is organic farming producing food for a wealthy few?

We must strive in organic farming research to continue to find ways to improve the productivity and yields of our systems, keeping in mind the long-term effects of our actions. Advances in breeding, technology, and equipment are available for organic agriculture as well. However, knowledge and appreciation of the complex and diverse interactions between plants, animals and the soil should be the framework for research. In agriculture, the soil is our greatest resource and our goal must be to sustain the health of the soil, as it is the basis for healthy food and healthy people. It is my hope that with my research on perennial forage legumes within an annual cropping system, I have made a small contribution towards this greater goal.

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APPENDIX

SAS input for repeated measures analysis with slicediff (soil nitrate over time in the first cycle, 0 – 20 cm, all soil amendments)

```
DATA REPEATEDMEASURES789;
input rep $ time $ treatment $ nitrate @;
cards;
1      WHARV1       CLOVER       7.7
2      WHARV1       CLOVER       7.5
3      WHARV1       CLOVER       6.9
.
.
.
14     WHARV2       CLOVER       4.2
15     WHARV2       CLOVER       2.7
16     WHARV2       CLOVER       3.4
;
run;
proc glimmix;
class rep time treatment;
model nitrate = treatment time treatment*time/ddfm=kr;
random  _residual_/subject=rep(treatment) type=ante(1);
lsmeans treatment time treatment*time/slicediff = (treatment
time) *slice diff gives means for each trt for a given time and
means for a given trt for each time*;
adjust=tukey
run;
```