THE USE OF FALL-PLANTED BRASSICACEAE COVER CROP MONO- AND BICULTURES FOR NUTRIENT CYCLING AND WEED SUPPRESSION

By

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ABSTRACT

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Cover crops have the potential to increase the sustainability of agronomic cropping systems. Farmers are increasingly interested in using oilseed radish (Raphanus sativus L. var. oleiformis Pers.), both alone and in mixtures, to suppress weeds, reduce fertilizer inputs, and improve crop yields. However, there is limited information to guide cover crop species selection. To evaluate differences between and within species, we evaluated biomass accumulation of six oilseed radishes, two brown mustards (Brassica juncea [L.] Czern.), two white mustards (Sinapis alba L.), one rapeseed (Brassica napus L.), and one hybrid turnip (Brassica rapa L. x B. napus L.) in field trials. Cover crop biomass accumulation within and between species was similar. The accessions provided rapid ground cover and accumulated biomass at similar rates. In Minnesota, dry aboveground biomass ranged between 3410-5542 kg ha⁻¹, while in Michigan biomass ranged between 2545-3572 kg ha⁻¹. There were no differences in N uptake for any of the accessions in either trial. Brassicaceae cover crops accumulated 100-131 kg N ha⁻¹ and 81-109 kg N ha⁻¹ in aboveground tissues in Minnesota and Michigan, respectively. Experiments were then conducted to investigate the growth and weed suppression of oilseed radish, annual ryegrass (Lolium *multiflorum* Lam.), cereal rye (Secale cereale L.), oats (Avena sativa L.), crimson clover (Trifolium incarnatum L.), hairy vetch (Vicia villosa Roth.), and winter pea [Pisum sativum var. arvense (L.) Poir.] both in monocultures and in biculture mixtures of oilseed radish plus each species. Cover crop and weed biomass varied across years. Oilseed radish comprised the majority of biculture fall biomass, and was more competitive in biculture with legumes than

grasses. Grass monoculture and grass biculture treatments were more effective at weed suppression in fall 2012, fall 2013, and spring 2014 than legume monoculture treatments. Crimson clover failed to establish in two out of the three years, and winter pea failed to survive the winter in two out of three years. This study also evaluated the impact of the cover crop monocultures and bicultures on a following corn crop in the absence of applied fertilizer. Overall, the cover crops did not reduce corn grain yield, with the exception of annual ryegrass and cereal rye treatments each in one of three years. Annual ryegrass and cereal rye reduced corn yield by 51% and 24%, respectively, compared with the weedy control. An additional experiment was conducted in Lansing and Hickory Corners, MI to determine the impact of fallplanted oilseed radish, annual ryegrass, and radish + ryegrass cover crops on nitrous oxide (N_2O) emissions. There were no differences between the cover crop treatments and the bare ground control for fall and spring-summer cumulative N₂O -N emissions. It appears nitrous oxide emissions did not represent a major pathway for N loss in this study. This work adds to the cover crop body of knowledge and provides information which will be of use when making recommendations to farmers.

This work is for my grandparents, Don and Joy Ackroyd. They don't understand why I've been ten years a college student, but they've supported me nonetheless. And for Aunt Mary "Buttons" Watts, who maybe does understand.

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KEY TO ABBREVIATIONS

DAP: days after planting GDD: growing degree days NIRS: near infrared spectroscopy PSNT: pre-side dress nitrogen-N test RY: relative yield VNS: variety not specified WAP: weeks after planting

CHAPTER 1

LITERATURE REVIEW

Cropping System Overview

Michigan's food and agricultural sector, which is the second most diverse in the country, generates over \$90 billion each year (USDA-NASS 2013). In 2013, Michigan's largest commodity group in terms of cash receipts was field crops. Corn (*Zea mays* L.), soybeans [*Glycine max* (L.) Merr.], and winter wheat (*Triticum aestivum* L.) were respectively ranked the 1st, 2nd, and 5th largest commodities in that group (USDA-NASS 2015). Michigan farmers planted 2.6 million acres of corn, 1.9 million acres of soybeans, and 620,000 acres of winter wheat in 2013 (USDA-NASS 2015). As the value of these commodities has increased over the last five years, so has the cost of inputs.

Farmers have minimal control over some expenses such as cropland rent. However, the use of other inputs such as fertilizer can be creatively managed to decrease costs and increase profitability. One large and increasingly expensive input for farmers is nitrogen (N) fertilizer. It is also particularly well-suited to creative management. The goal of fertility management strategies is to increase the nitrogen use efficiency (NUE) of the system by synchronizing fertilizer application with peak crop need, while minimizing N losses to the environment (Ribaudo et al., 2011). In addition to careful manipulation of N fertilization, Robertson and Vitousek (2009) advocated the incorporation of cover crops into crop rotations to improve the uptake of N fertilizer into the system and increase the sustainability of cropping systems. Cover crops are generally defined as species which are grown between cash crops, when the ground would otherwise lie fallow. In Michigan, one of the best windows to plant cover crops in a winter wheat-corn-soybean rotation is after wheat harvest. Winter wheat is typically harvested in

July, which allows adequate time to establish a cover crop before the onset of freezing temperatures. Research into the use of cover crops after wheat harvest would be of interest to a wide audience.

Benefits of Cover Crops Use

Cover crop use has grown steadily among farmers over the last five years (CTIC, 2015). This is a result of both more farmers using cover crops and current users increasing their cover crop acreage. The top f benefits that cover crop users seek from cover crops were increased overall soil health, increased soil organic matter, reduced soil erosion, weed suppression, and reduced soil compaction (CTIC, 2015). These results echo those of a survey by Singer et al. (2007), who found that the top two benefits ascribed to cover crops by farmers were the reduction of soil erosion (96% of respondents) and increases in soil organic matter (74% of respondents).

Improved soil physical properties

Cover crops have been documented to decrease soil erosion and improve soil physical properties (Meisinger et al., 1991). Oats (*Avena sativa* L.) and cereal rye (*Secale cereale* L.) each decreased rill and inter-rill erosion in at least one year of a three year study (Kaspar et al., 2001). In this study, cereal rye decreased erosion by 48-62% and oats by 51% during simulated rainfall events on ground with an average slope of 4.4%. The authors hypothesized that the decrease in erosion was due to decreased sediment detachment and an increase in ponding and sediment deposition. Hively and Cox (2001) found annual ryegrass (*Lolium multiflorum* Lam.) to be an acceptable choice for erosion control after soybean harvest as it reliably provided over 75% ground cover. Villamil et al. (2006) found that after two years, crop rotations including both hairy vetch (*Vicia villosa* Roth) and cereal rye had a higher soil organic matter (SOM) content

within the top 30 cm of soil than other rotations. They postulated that this was due to the addition of C from the cereal rye, plus N from the hairy vetch that fueled microbial activity which lead to the increased SOM levels. The same study found that rotations which included cereal rye or hairy vetch had 9-17% greater wet aggregate soil stability, depending on the rotation (Villamil et al., 2006). Rotations which include a cover crop can benefit from decreased soil bulk density, which means improved aeration and water infiltration as well as better cash crop establishment and root growth (Villamil et al., 2006). Williams and Weil (2004) observed soybean roots growing down into channels created by the roots of canola (*Brassica rapa* L.), oilseed radish (*Raphanus sativus* L.), cereal rye, and cereal rye plus oilseed radish cover crops.

Nutrient retention

Cover crops can supply N to the following cash crop and help retain nutrients in the system (Meisinger et al., 1991). Nitrogen is of particular concern due to its mobility and activity in the soil. Aside from incorporation into soil organic matter, N can leach, run off, or be lost to volatilization and other soil gaseous emissions (greenhouse gas emissions). Roughly 50% of the N applied to agricultural systems is lost through these pathways (Tonitto et al., 2006). In the best-case scenario, cover crops take up residual soil N and then that N is released from the biomass in synchrony with cash crop N demands. Wagger (1989) found that while cereal rye, crimson clover (*Trifolium incarnatum* L.), and hairy vetch accumulated more tissue N when terminated later in the spring, early termination resulted in faster N release. The author suggested that this faster N release offset the lower amount of N accumulated by the cover crops.

In the Midwest, N leaching is of most concern from November – May. Cover crops can decrease leaching by decreasing soil moisture levels via evapotranspiration and by taking up residual soil nitrate (NO₃) (Meisinger et al., 1991). Cereal rye is particularly effective at

decreasing nitrate leaching (Kaspar et al., 2007; Ruffo et al., 2004). Kaspar et al. (2007) found cereal rye decreased nitrate leaching by at least 50%, compared with a no-cover control, in all four years of a study. Tonitto et al. (2006) calculated that legumes reduced nitrate leaching by 40% while non-legume cover crops reduced leaching by an average of 70%. Collins et al. (2007) found mustard (*Brassica hirta* 'Martigena') took up 92-142 kg N ha⁻¹ and decreased nitrate leaching.

Aside from leaching and runoff, N could be lost from the system in the form of greenhouse gas emissions. Not much literature exists on the topic of cover crops and their impact on greenhouse gas emissions. However, Parkin and Kaspar (2006) found that while corn plots emitted significantly more N₂O than soybean plots, the inclusion of a cereal rye winter cover crop after both corn and soybean harvest each year did not affect N₂O emissions from each cash crop. Robertson et al. (2000) stated that the primary driver of N₂O fluxes in agricultural systems is the amount of N available in the soil. The more N available in the soil, the higher the flux. This assertion is supported by research done by Gomes et al. (2009), in which leguminous cover crops including vetch (Vigna sativa L.), cowpea (Vigna unguiculata L. Walp), pigeon pea (Cajanus cajan L. Millsp.), and lablab (Dolichos lablab L.) had larger cumulative N₂O emissions during the 45 days after cover crop residue management than a black oat (Avena strigosa Schreb) cover crop in no-till maize in a subtropical climate. In spite of this, the authors calculated that less than one percent of the N added to the soil by the legumes was subsequently lost as greenhouse gas emissions. Gomes et al. (2009) also determined that N_2O emissions in the 45 days after cover crop residue management correlated with the N added to the soil by the cover crop biomass, while after that period total soil N content drove N2O emissions. The N2O emissions took longer to peak in the black oat + vetch cover crop mixture treatment than in the

legume treatments, which supports the idea that creating a cover crop mixture with a "balanced" C:N ratio can improve N synchrony by delaying N loss from the system.

Weed suppression

Weeds can significantly reduce crop yield and thus their control is of considerable concern to farmers (Walsh et al., 2013). Farmers currently control weeds primarily through the use of herbicides and cultivation; both tactics have detrimental aspects. The intensive use of herbicides can lead to the development of herbicide-resistant weeds, while cultivation increases soil erosion. Cover crops are thus of interest as a weed-suppression tool. They can make environmental conditions unfavorable for weed germination and growth, increase the activity of weed seed predators and weed seedling pathogens, act as a mulch that smothers weeds, outcompete weeds for nutrients or light, and some are allelopathic (Conklin et al., 2002; Creamer et al., 1996; Pullaro et al., 2006). Stivers-Young (1998) found that oilseed radish and mustards suppressed weeds in the fall and their residues shaded out weeds in the spring. Cereal rye, crimson clover, and red clover planted during the wheat phase of a rotation were believed to also decrease weed biomass by shading (Smith et al., 2008). Weed species richness was likewise decreased, an effect the authors attributed specifically to the cover crops and not to rotational diversity. Several common cover crops are believed to be allelopathic including cereal rye, hairy vetch, and oilseed radish. For example, laboratory studies indicate that substances produced by Brassicaceae cover crops such as oilseed radish inhibit weed seed germination (Norsworthy and Meehan IV, 2005; Norsworthy and Meehan IV, 2009; Norsworthy et al., 2006).

Promising laboratory results do not always translate to the field. In one study, cereal rye and hairy vetch had no impact on weed density (Davis, 2010). Wang et al. (2008) found that sorghum-sudangrass [*Sorghum bicolor* (L.) Moench X *S. sudanese* (P.) Stapf] and Brassicaceae

cover crops reduced weed density and altered the composition of weed species, but did not eliminate the need for other methods of weed control. De Bruin et al. (2005) determined that when weed pressure was high, a late-season herbicide application in addition to the cereal rye cover crop was necessary for adequate weed control. Fall-planted cereal rye and crimson clover were more effective at suppressing weed growth than hairy vetch in a North Carolina study, though herbicides were still necessary for optimum corn yields (Yenish et al., 1996).

Increased cash crop yield

Cover crops can increase the yield of a following cash crop (Table 1.1). In a metaanalysis by Miguez and Bollero (2005), corn grown after legume cover crops yielded 24% more than that grown without a cover crop when no inorganic fertilizer was applied. Another metaanalysis found that as long as a legume cover crop provided at least 110 kg ha⁻¹ of N, cash crop yields did not differ between cover crop plots and those in which an inorganic fertilizer had been applied (Tonitto et al., 2006). The use of Dutch white clover (Trifolium repens L.) and medium red clover (Trifolium pratense L.) has also been shown to increase corn yield, as compared with a no-cover crop control (Hively and Cox, 2001). In a study by Williams and Weil (2004), soybean yields were greater at one site following an oilseed radish plus cereal rye mixture than after cereal rye and no-cover crop treatments. At another site they were greater following cereal rye than after a cereal rye plus oilseed radish mixture, oilseed radish, and no-cover crop treatments. The authors postulated that the cover crops provided the most benefit at the site with lower precipitation levels and more compacted soil. Smith et al. (2008) found a linear relationship between crop rotation diversity and corn yield. The more cash and cover crop species included in the rotation, the higher the yield. They attributed this effect to the N provided by legume species. Soybean and wheat yields also benefited from increased rotational diversity,

though not to the same extent as corn (Smith et al., 2008). Other studies have shown cover crops to generally have no impact on corn yield (Delate et al., 2003; Yenish et al., 1996) or soybean yield (De Bruin et al., 2005; Delate et al., 2003; Kaspar et al., 2007; Reddy, 2001; Ruffo et al., 2004) (Table 1.1). A meta-analysis by Tonnito et al. (2006) determined that when inorganic fertilizer was applied, cash crop yields did not differ between bare fallow plots and those in which a non-legume cover crop had been grown. The impact of the cover crop on cash crop yield is dependent on the interaction among a variety of variables including cover crop management, weed pressure, soil fertility, and the weather (De Bruin et al., 2005). Kaspar et al. (2007) found cereal rye to have no impact on corn yield so long as sufficient time was allowed between cover crop termination and cash crop planting.

Difficulties Associated with Cover Crop Use

In spite of the benefits provided by cover crops, cover crop adoption remains relatively low. Singer et al. (2007) found that in the last five years, only 11% of surveyed Corn Belt farmers had used cover crops. Several factors limit cover crop use, and they all relate to economics (Snapp et al., 2005). These factors can be loosely grouped into physical costs (e.g., of seed), time costs (e.g., cover crop planting), management concerns (e.g., cover crops may need to be planted at a busy point in the season like during cash crop harvest), interference with the cash crop (e.g., causing delayed planting), and difficulty choosing the right cover crop. In the CTIC survey (2015), the top five most common "barriers to adoption" that cover crop non-users cited were the time and labor associated with planting and management, the cost to plant/manage the cover crops, seed cost, difficulty getting the cover crop to establish, and concerns about delayed planting in the spring. Among cover crop users, the top five challenges associated with cover crops were establishment, seed cost, the time and labor associated with planting and management, cover crop species selection, and "no measurable economic return".

Interference with cash crop

Cover crops may negatively impact cash crops in a number of ways. In years or locations where moisture is limited, cover crops may deplete soil water reserves needed by the cash crop (Meisinger et al., 1991; Ruffo et al., 2004). Cover crops with high C:N ratios and large biomass production such as cereal rye can immobilize soil N (De Bruin et al., 2005) to the detriment of a cash crop. When weather or soil conditions are unfavorable for field work, the need to terminate cover crops prior to cash crop planting may lead to delayed planting (Long et al., 2013). Some cover crops can become weeds which compete with the cash crop for resources. Williams and Weil (2004) observed this interaction between canola and soybeans. They attributed poor soybean yield to a canola cover crop which was not adequately controlled prior to soybean planting. Living mulches such as alfalfa can also compete with the cash crop to decrease yield (Schmidt et al., 2007). When intercropped, cover crops such as cereal rye and winter pea can interfere with cash crop harvest by physically impeding harvest (Hively and Cox, 2001). Cover crops can also act as "green bridges", harboring insects or pathogens whose populations would otherwise subside during a fallow period (Odhiambo et al., 2012).

Decreased cash crop yield

While some studies have shown cover crops to be of benefit to the yield of a following cash crop, other studies have shown the opposite (Table 1.1). In a review by Miguez and Bollero (2005), corn following cereal rye yielded 1% less than that grown following no cover crop. The meta-analysis by Tonitto et al. (2006) determined that a legume cover crop had to provide at least 110 kg ha⁻¹ of N for a cash crop to yield the same as plots fertilized with inorganic fertilizer.

Otherwise, there was a yield reduction of 10%. Corn following cereal rye suffered a yield depression when insufficient time (eight days) was allowed between cover crop termination and corn planting (Kaspar et al., 2007). The authors suggested that the large amount of cereal rye biomass incorporated into the soil may have interfered with the corn. Terminated cover crops can also attract pests such as seed corn maggot [Delia platura (Meigen)] that can damage cash crop stand (Cullen and Holm, 2013). Westgate et al. (2005) observed a decrease in soybean dry matter accumulation and a delay in soybean maturity when soybeans were planted after cereal rye. In one of two years of a study, corn following annual ryegrass yielded less than the no-cover crop control (Hively and Cox, 2001). Work done in Ontario, Canada, found corn following oats or cereal rye to yield less than a no-cover control (Vyn et al., 2000). Because of these reductions in yield, a number of researchers contend that cover crop use is not economically feasible, particularly in the case of non-legume cover crops (Reddy, 2001; Reddy, 2009; Tonitto et al., 2006). These studies generally examined the economics of cover crop use in the context of a rotation with minimal diversity, such as continuous corn. In contrast, Smith et al. (2008) assert that the higher cash crop yields seen in the most diverse crop rotations (those including three cash crops and three cover crops) could offset the opportunity cost of planting high-return cash crops less frequently. Rotational diversity could act as a buffer against sudden changes in the commodity-market values of cash crops.

Types of Cover Crops

In the Midwest, three of the most commonly grown groups of cover crops are fall-planted grasses, legumes, and Brassicaceae species. Each type of cover crop has its advantages and its drawbacks.

Grasses

Grasses are a good choice for fall planting because their large biomass production allows them to take up residual soil NO_3 that might otherwise leach away over the winter. Their deep roots and large root biomass make them able N scavengers (Meisinger et al., 1991). In the Midwest, three of the most common grass cover crops are cereal rye, annual ryegrass, and oats. Cereal rye is the hardiest of the three, reliably survives the winter, and is one of the easiest cover crops to establish. Cereal rye is also one of the cover crops that is recognized as being the most likely to interfere with a following cash crop. This observation is due to its large potential dry aboveground biomass production, which ranges from 610 - 4640 kg ha⁻¹ in the Midwest in the fall (Kaspar et al., 2001; Vyn et al., 2000) and up to 6095 kg ha⁻¹ by the following spring (Ruffo et al., 2004) (Table 1.2). Cereal rye can cause delayed cash crop planting due to the difficulty of terminating it in the spring under adverse weather or soil conditions. It is also known to be allelopathic to following cash crops (Burket et al., 1997). Further, its relatively high C:N ratio and large biomass production increases the likelihood of N immobilization in the soil after cover crop termination, to the potential detriment of the following cash crop (Crandall et al., 2005). However, cereal rye remains a commonly used cover crop in spite of its potential risks because that same cover crop biomass which can make termination difficult also adds C to the soil and acts as a mulch. Its seed is relatively inexpensive and it is one of the hardiest cover crop species, making it a likely choice for late fall planting when weather delays the harvest of a preceding crop such as corn or soybean.

As with cereal rye, annual ryegrass may not be the best choice for a novice cover crop user due to concerns over termination in the spring (Creamer et al., 1997; Madden et al., 2004). When planted in the fall, it survives the winter and can produce 1240 - 6241 kg ha⁻¹ of dry aboveground biomass by the following spring (Francis et al., 1998; Kuo and Jellum, 2000)

(Table 1.2). One issue related to annual ryegrass is that of herbicide resistance. The resistance of annual ryegrass to several herbicide classes has been documented in the U.S. (Martins et al., 2012; Perez-Jones et al., 2005). This is no small concern given that 59% of respondents in the CTIC survey who grew row crops (2015) relied on herbicides for cover crop termination. Nonetheless, annual ryegrass is one of the few cover crop choices for planting in wet areas, and farmers have been experimenting with its use. There are several ryegrass species (*Lolium* spp.) grown as cover crops. Throughout this dissertation, "annual ryegrass" refers to *Lolium multiflorum* Lam.

Like cereal rye, oats has the potential for large biomass production in the fall. It has been reported to produce 3670 kg ha⁻¹ of dry aboveground biomass (Kaspar et al., 2001) and 12,500 kg ha⁻¹ of biomass in regions where it overwinters (Brennan and Smith, 2009) (Table 1.2). Unlike cereal rye, oats does not overwinter in the upper Midwest, and may be a good choice for farmers concerned with spring cover crop termination. Because oats winterkills several weeks to months prior to cash crop planting, there is little risk of N immobilization in the soil negatively impacting a following cash crop. However, oats has been documented to negatively impact corn growth in regions where it does overwinter, possibly partially due to allelopathy (Norsworthy, 2004). When purchased as bin-run seed, oats has one of the lowest seed costs of all the commonly grown cover crops.

Legumes

Legume cover crops can form symbiotic relationships with *Rhizobacteria* spp., which fix atmospheric N in nodules on the legume roots. Farmers make use of this symbiosis to add N to cropping systems. The shallower root systems of legumes means they are less likely to deplete soil moisture (Nielsen, 2001), which is of benefit in dry years or locations. In the Midwest, some

of the legume cover crops that can be grown include hairy vetch, winter pea (*Pisum sativum* var. *arvense* (L.) Poir.), and crimson clover. Hairy vetch is the hardiest of the three, and the easiest to establish. It has been documented to produce up to 8900 kg ha⁻¹ of dry aboveground biomass by May when planted the previous September (Singogo et al., 1996) (Table 1.2). In an Ohio study examining hairy vetch as part of a cover crop mixture, it produced 5500-7810 kg ha⁻¹ of biomass (Creamer et al., 1996). It is also has some of the highest N-fixation rates: 160-265 kg N ha⁻¹ compared with 10-30 kg N ha⁻¹ accumulated by crimson clover in a study by Creamer et al. (1996).

There is a need for research into ways to successfully establish legume cover crops (Hively and Cox, 2001). Crimson clover and winter pea are more difficult to establish than hairy vetch. Fall-planted crimson clover has been documented to produce 968 - 8000 kg ha⁻¹ of dry aboveground biomass by the following spring (Daniel et al., 1999; Decker et al., 1994) (Table 1.2). As with the other cover crops listed in Table 1.2, there is a large variation in biomass production due to differences in cover crop genetics, planting date, and site-specific factors such as the weather and soil fertility. In spite of its name, winter pea does not reliably survive the winter in Michigan. Under favorable weather conditions, winter pea can produce similar amounts of biomass as the other cover crops discussed in this chapter. In a study in North Carolina, winter pea produced 2400-6300 kg ha⁻¹ of biomass and accumulated 67-208 kg N ha⁻¹ (Parr et al., 2011). While hairy vetch is the most reliable of the three legumes, its large biomass production can make it difficult to terminate in the spring. Hairy vetch which was not successfully terminated has been documented to negatively impact corn yield through competition (Parr et al., 2011). Further, it is not suitable for some crop rotations. Farmers who grow small grains are advised to avoid hairy vetch as a cover crop, as it readily volunteers and can be a contaminant of

harvested seed. Given these issues and concerns over increasing weather variability, it is sensible to investigate the use of other legume options such as winter pea and crimson clover.

Brassicaceae species

Cover crops with rapid fall growth and large biomass production are particularly useful as N scavengers (Meisinger et al., 1991), and Brassicaceae cover crop species neatly fit these criteria. In a study conducted by Weil and Dean (2009), rapeseed and oilseed radish shoots scavenged more N in the fall than cereal rye. Brassicaceae cover crop species were another commonly used cover crop type in a recent survey, with 61% of respondents saying they had planted these cover crops (CTIC, 2015). Among the Brassicaceae species, oilseed radish has become a particularly popular choice in the Midwest (Ngouajio and Mutch, 2004; Sundermeier, 2008). With regard to nomenclature, the radishes grown as cover crops are closely related and are usually either oilseed radish (Raphanus sativus L. var. oleiformis) or forage oilseed radish (R. sativus L. var. longipinnatus) (Weil et al., 2009). The subspecies readily interbreed and management recommendations for both are the same (Weil et al., 2009). For the purpose of this dissertation, cover crop radishes will all be referred to as oilseed radish. Other Brassicaceae species of interest to farmers include rapeseed or canola (both *Brassica napus* L.), turnips (B. rapa L.), white mustards (Sinapis alba L.), and brown mustards (B. juncea [L.] Czern.). As with oilseed radish, there is confusion with regard to the common names associated with B. rapa L. A search of the Integrated Taxonomic Information System (itis.gov) reveals that common names for this species include field mustard, rape, mustard rape, turnip rape, and wild turnip. For the purposes of this dissertation, *B. rapa* L. will be referred to as turnip.

Fall-planted Brassicaceae cover crops grow quickly. Rapid and large biomass production is one of several attributes that farmers desire from oilseed radish. It has been shown to produce

close to 5000 kg ha⁻¹ of dry aboveground biomass in the fall when planted in August (Table 1.2) (Dean and Weil, 2009; Vyn et al., 1999). Other reported values range from 890 – 3947 kg ha⁻¹ (Stivers-Young, 1998; Vyn et al., 2000). White mustard has been documented to produce 1626 – 6397 kg ha⁻¹ of dry aboveground biomass in the fall (Collins et al., 2006; Stivers-Young, 1998). Table 1.2 provides an overview of biomass production values found in the literature of yellow mustard, rapeseed, and turnip.

In spite of farmer interest in, and marketing efforts involving, Brassicaceae cover crops there has been relatively little research in the Midwest to compare their growth both within and between species. In New York, Stivers-Young (1998) evaluated 'Adagio' oilseed radish, two white mustards (referred to as *B. hirta* L. but now called *S. alba* L.), forage kale (*B. oleracea* L.), turnip, and canola and found that the accessions produced the same amount of dry aboveground biomass in the fall, though oilseed radish plots had less surface cover crop residue in the spring than the other Brassicaceae treatments and oats. All of the cover crop treatments had lower fall soil NO₃ levels than the no-cover crop control plots. Some studies have examined a few accessions of one species such as oilseed radish, or a few accessions from the plant family such as oilseed radish and mustard. Dean and Weil (2009) found no difference in fall dry aboveground biomass production and N uptake between 'Adagio' oilseed radish, 'Daikon' oilseed radish, and 'Dwarf Essex' rapeseed. In two out of four site-years, oilseed radish produced more dry aboveground biomass and accumulated more shoot tissue N than canola in a study conducted in Quebec, Canada (Isse et al., 1999).

Biomass production is not the only Brassicaceae cover crop attribute of interest. Oilseed radish is reported to have the ability to alleviate soil compaction while suppressing weeds and scavenging nutrients (Snapp et al., 2005). The latter benefit has caught the attention of farmers,

who have also expressed interest in the use of Brassicaceae cover crops to help retain N in the system. Thorup-Kristensen (2001) determined that oilseed radish and canola roots grew faster and deeper than those of annual ryegrass, cereal rye, and oats and took up more soil NO_3 than the grass cover crops. Meisinger et al. (1991) noted that Brassicaceae cover crops are not generally winter hardy, and that they decompose more easily than grass cover crops, allowing rapid remineralization of N the following spring. Fall-planted oilseed radish accessions have been shown to accumulate 100-170 kg N ha⁻¹ (Allison et al., 1998; Axelsen and Kristensen, 2000; Isse et al., 1999; Thorup-Kristensen, 1994; Thorup-Kristensen, 2001; Thorup-Kristensen, 2006). Other Brassicaceae species likewise rapidly accumulate soil N. Stivers-Young (1998) found that five weeks after planting, turnip had accumulated 69 kg N ha⁻¹ while two accessions of white mustards accumulated 51-62 kg N ha⁻¹. This accumulated N could be of benefit to a following cash crop. There are concerns, however, as to the fate of the N accumulated by fall-planted oilseed radish. Because oilseed radish winter-kills, and because it has a relatively low C:N ratio, there is a window between decomposition in late winter/early spring and cash crop planting during which N released by the cover crop biomass could be lost from the system via surface runoff, leaching, or denitrification and subsequent diffusion into the atmosphere.

Cover crop mixtures

Farmers have long been known as innovators (Carlson and Stockwell, 2013) and have recently begun to experiment with the use of cover crop mixtures (multiple species grown at the same time). In spite of farmer interest in cover crop mixtures, there has been relatively little research on the topic (Carlson and Stockwell, 2013). Research that has been conducted has focused typically on a grass such as cereal rye mixed with a legume such as hairy vetch (Table 1.3). The trend towards the use of cover crop mixtures is driven by the desire to take advantage

of the complementary or synergistic benefits of cover crop species. A slow-to-establish N fixing legume such as hairy vetch pairs well with a rapid-growing Brassicaceae species such as oilseed radish. Furthermore, the combination of high and low C:N ratio species may improve N synchrony via reduced N immobilization (Creamer et al., 1997). Mixing a cover crop with a high C:N ratio such as cereal rye with one with a lower C:N ratio such as oilseed radish may "balance" the C:N ratio, avoiding the problems inherent in too high of a ratio such as N immobilization and too low of a ratio such as too rapid/early N release and subsequent leaching (Odhiambo and Bomke, 2001; Rosecrance et al., 2000). There is evidence in the literature to support this idea. Gomes et al. (2009) observed that in a vetch (Vigna sativa L.) plus black oat (Avena strigosa Schreb) treatment, N₂O emissions took longer to peak after cover crop termination than in pure legume treatments. Several cover crops such as hairy vetch, oilseed radish, and cereal rye, are believed to be allelopathic; therefore, using a mixture of cover crops offers the possibility of a wider spectrum of weed suppression (Creamer et al., 1997). Mixtures have been found to be more productive than cover crop monocultures, and are resilient in the face of extreme weather events (Wortman et al., 2012). This resiliency may contribute to higher and more consistent cash crop yields.

While cover crop mixtures can convey many benefits, there are potential disadvantages to their use. Some cover crops may be better candidates for inclusion in mixtures than others. Furthermore, there is the question of planting rates and proportions. Wortman et al. (2012) found mustard species to be more competitive and productive than legumes when grown in mixtures in Nebraska. Creamer et al. (1997) surveyed a range of cover crop species in 13 multi-species mixtures in Ohio and observed that some species were not suitable due to failure to overwinter or low biomass production. When a component species dominates in a mixture, it can also lessen

the benefits provided by the other cover crops. In both years of a study by Brainard et al. (2011), soybeans grown as a cover crop in a mixture with sorghum-sudangrass produced less biomass, nodulated less, and fixed less N than soybeans grown in monoculture. Furthermore, the cost of seed was greater for the mixture than for the sorghum-sudangrass monoculture treatment. When sown later in the fall (mid- to late September), hairy vetch planted in mixture with cereal rye produced less biomass than when planted in late August in Michigan (Hayden et al., 2015). It may be more difficult for a farmer to predict the effect of a cover crop mixture on a cropping system. Hayden et al. (2014) did not find evidence of synergy between cereal rye and hairy vetch when grown in mixture. The authors noted that altering the seeding proportions of the two species in mixture resulted in tradeoffs among the agroecosystem benefits provided by the cover crops. For example, higher proportions of hairy vetch lead to greater seed costs and less weed suppression but greater amounts of N fixed. More research is needed to determine which species are the best candidates for inclusion in mixtures in Michigan, and which mixtures provide the best combination of benefits and the least likelihood of risk to a following cash crop.

Dissertation Objectives

In spite of the popularity of cover crops, many questions remain about their growth characteristics and use. Research has not kept pace with farmer innovations on the use of cover crop mixtures. Farmers have requested more data on the benefits of cover crops, and how to successfully implement their use (Carlson and Stockwell, 2013). Key research priorities of interest to farmers include environmental impacts of cover crops, cover crop effects on cash crop yield, the performance of cover crop mixtures, and the synchronization of nutrient release in cover crop systems (Carlson and Stockwell, 2013). Kaspar et al. (2007) likewise identified a need for more research into the management of cover crops to avoid the risk of cash crop yield

reduction. Many questions remain to be answered about cover crops. Given the rising popularity of both oilseed radish and cover crop mixtures, these questions should be addressed with mixtures and oilseed radish in mind. Research topics of interest include: a) cover crop growth potential and cover crop interactions, especially with regard to the difficulty of establishing a mixed cover crop stand, b) cover crop impact on weeds, c) cover crop impact on cash crop yield as informed by the risk of N immobilization by grass cover crops, and d) the synchronization of N release by cover crops to match the N uptake demands of the following cash crop. Much has been said about oilseed radish, but there is relatively little research to support or disprove the claims. In particular, there are questions as to the benefits provided by oilseed radish in mixtures with other cover crops. Also of concern is the fate of N released by oilseed radish after decomposition.

APPENDIX

Common name	Scientific name	Overall impact on corn yield compared to no-cover crop control [†]	Reference
Annual ryegrass	Lolium multiflorum Lam.	Negative to neutral	Dapaah and Vyn, 1998
		Neutral to negative	Hively and Cox, 2001
		Neutral	Isse et al., 1999
		Neutral	Kuo and Jellum, 2002
		Negative to neutral	Vyn et al., 1999
Cereal rye	Secale cereale L.	Neutral	Crandall et al., 2005
		Neutral to negative	Kaspar et al., 2007
		Neutral	Kuo and Jellum, 2002
		Neutral	Vyn et al., 2000
		Neutral	Yenish et al., 1996
Oats	Avena sativa L.	Neutral	Vyn et al., 2000
Crimson clover	<i>Trifolium incarnatum</i> L.	Neutral to positive	Decker et al., 1994
	-	Neutral	Isse et al., 1999
		Neutral to negative	Parr et al., 2011
		Neutral	Yenish et al., 1996
Hairy vetch	Vicia villosa Roth.	Positive to neutral	Decker et al., 1994
		Positive	Kuo and Jellum, 2002
		Neutral to positive	Parr et al., 2011
		Neutral	Yenish et al., 1996
Winter pea	Pisum sativum var.	Positive to neutral	Decker et al., 1994
		Neutral to negative	Parr et al., 2011

Table 1.1. Overview of fall-planted cover crop impact on corn yield in the literature (assuming appropriate management of cover crop such as allowing adequate time between cover crop termination and corn planting).

Common name	Scientific name	Overall impact on corn yield compared to no-cover crop control [†]	Reference
Oilseed radish	Raphanus sativus L. var.	Positive to neutral Neutral Neutral to positive Neutral	Dapaah and Vyn, 1998 Isse et al., 1999 Vyn et al., 1999 Vyn et al., 2000
Annual ryegrass + hairy vetch	Lolium multiflorum Lam. +	Neutral to positive	Kuo and Jellum, 2002
Cereal rye + hairy vetch	Secale cereale L. + Vicia	Positive to neutral neutral	Kuo and Jellum, 2002 Parr et al., 2011
Cereal rye + winter pea	Secale cereale L. + Pisum	neutral	Parr et al., 2011

† When the study examined multiple fertilizer rates, the results from the no-added fertilizer treatment or the treatments averaged across rates (when presented) are summarized here.

Common name	Scientific name	Planted	Harvested	Total dry aboveground biomass (kg ha ⁻¹)	Reference
Annual ryegrass	Lolium multiflorum	Jul.	Nov.	1280 - 2530	Dapaah and Vyn, 1998
		Mar.†	Jun.	1685	Francis et al., 1998
		Mar.†	Oct.	3558 - 6241	Francis et al., 1998
		Sep.	Nov.	302 - 1691	Isse et al., 1999
		SepOct.	AprMay	1240 - 4650	Kuo and Jellum, 2000
		Oct.	AprMay	4420 - 4650	Kuo et al., 1997
		Aug.	Apr.	4310	Odhiambo and Bomke, 2001
		JulAug.	Nov.	3500	Thorup-Kristensen, 2001
		AprAug.	Nov.	660 - 2620	Vyn et al., 1999
Cereal rye	Secale cereale L.	Jul.	Nov.	1680	Axelsen and Kristensen, 2000
-		Jul.	Mar.	2660	Axelsen and Kristensen, 2000
		Oct.	Apr.	680 - 2660	Crandall et al., 2005
		Oct.	MarApr.	528 - 4600	Daniel et al., 1999
		Aug.	OctNov.	1910 - 4640	Kaspar et al., 2001
		SepOct.	AprMay	250 - 2740	Kaspar et al., 2007
		SepOct.	AprMay	1420 - 4190	Kuo and Jellum, 200
		Oct.	AprMay	4050 - 4190	Kuo et al., 1997
		Nov.	Mar.	4970	Norsworthy, 2004
		Aug.	Apr.	4240 - 5670	Odhiambo and Bomke, 2001
		SepOct.	AprMay	1800 - 12600	Parr et al., 2011
		Oct.	Apr.	1500 - 5730	Ranells and Wagger, 1996
		Oct.	Dec.	600 - 1000	Ranells and Wagger, 1997
		Oct.	Apr.	3360 - 4630	Ranells and Wagger, 1997
		Oct.	May	2236 - 6095	Ruffo et al., 2004
		OctNov.	Apr.	2280 - 6070	Sainju et al., 2005
		JulAug.	Nov.	2100	Thorup-Kristensen, 2001
		Aug.	Oct.	610 - 1480	Vyn et al., 2000

Table 1.2. Overview of cover crop dry aboveground biomass in the literature.
Common name	Scientific name	Planted	Harvested	Total dry aboveground biomass (kg ha ⁻¹)	Reference
Cereal rye	Secale cereale L.	Aug.	Apr.	650 - 2120	Vyn et al., 2000
		Oct.	MarApr.	4540 - 5140	Yenish et al., 1996
Oats	Avena sativa L.	Oct.	Dec.	≈700 - 2000	Brennan and Smith, 2009
		Oct.	Feb.	≈7500 - 12,500	Brennan and Smith, 2009
		Mar.*	Jun.	3108	Francis et al., 1998
		Mar.*	Oct.	9908	Francis et al., 1998
		Aug.	OctNov.	2170 - 3670	Kaspar et al., 2001
		Nov.	Mar.	3690	Norsworthy, 2004
		AugSep.	OctNov.	957 - 3922	Stivers-Young, 1998
		JulAug.	Nov.	3100	Thorup-Kristensen, 2001
		Aug.	Oct.	970 - 1630	Vyn et al., 2000
Crimson clover	<i>Trifolium incarnatum</i> L.	OctNov.	Apr.	1914 - 5824	Bauer et al., 1993
		Oct.	MarApr.	968 - 4270	Daniel et al., 1999
		SepOct.	AprMay	2100 - 8000	Decker et al., 1994
		Sep.	Nov.	141 - 1041	Isse et al., 1999
		Aug.	Apr.	1460 - 4930	Odhiambo and Bomke, 2001
		SepOct.	AprMay	2000 - 7800	Parr et al., 2011
		Oct.	Apr.	1420 - 4980	Ranells and Wagger, 1996
		Oct.	Dec.	90 - 140	Ranells and Wagger, 1997
		Oct.	Apr.	1120 - 1170	Ranells and Wagger, 1997
		Oct.	MarApr.	3500 - 3690	Yenish et al., 1996
Hairy vetch	Vicia villosa Roth.	Jul.	Nov.	1910	Axelsen and Kristensen, 2000
-		Jul.	Mar.	2390	Axelsen and Kristensen, 2000
		Oct.	MarApr.	738 - 2890	Daniel et al., 1999
		SepOct.	AprMay	2700 - 7200	Decker et al., 1994
		SepOct.	AprMay	2800 - 6600	Parr et al., 2011

Table 1.2 (cont'd)

()				Total dry	
Common name	Scientific name	Planted	Harvested	aboveground biomass (kg ha ⁻¹)	Reference
Hairy vetch	Vicia villosa Roth.	SepOct.	AprMay	910 - 3480	Kuo and Jellum, 200
5		Oct.	AprMay	2700 - 3480	Kuo et al., 1997
		Oct.	May	4150 - 4380	Norsworthy et al., 2010
		Oct.	Apr.	2920 - 4760	Ranells and Wagger, 1996
		OctNov.	Apr.	2440-5100	Sainju et al., 2005
		Sep.	May	5600 - 8900	Singogo et al., 1996
		Oct.	MarApr.	2190 - 2380	Yenish et al., 1996
Winter pea	Pisum sativum var.	OctNov.	Apr.	2673 - 4587	Bauer et al., 1993
		SepOct.	AprMay	1800 - 6000	Decker et al., 1994
		Oct.	May	4350 - 4910	Norsworthy et al., 2010
		SepOct.	AprMay	2400 - 6300	Parr et al., 2011
		Sep.	May	3200 - 7600	Singogo et al., 1996
Oilseed radish	Raphanus sativus L. var.	Jul.	Nov.	4750	Axelsen and Kristensen, 2000
		Aug.	Nov.	2400 - 3640	Dapaah and Vyn, 1998
		Aug.	OctNov.	1993 - 4912	Dean and Weil, 2009
		Sep.	Nov.	176 - 3120	Isse et al., 1999
		AugSep.	OctNov.	1560 - 3947	Stivers-Young, 1998
		JulAug.	Nov.	4700	Thorup-Kristensen, 2001
		Aug.	Nov.	1250 - 4840	Vyn et al., 1999
		Aug.	Oct.	890 - 1270	Vyn et al., 2000
		Mar.	May	≈2600 - 3000	Wortman et al., 2012b
Rapeseed/canola	Brassica napus L.	Aug.	OctNov.	2987 - 5053	Dean and Weil, 2009
		Nov.	Mar.	6800	Hartz et al., 2005
		Sep.	Oct.	181 - 1960	Isse et al., 1999
		AugSep.	OctNov.	1462 - 4010	Stivers-Young, 1998
		Mar.	May	≈2000 - 2400	Wortman et al., 2012

Table 1.2 (cont'd)

Common name	Scientific name	Planted	Harvested	Total dry aboveground biomass (kg ha ⁻¹)	Reference
Turnip	<i>Brassica rapa</i> L.	AugSep.	OctNov.	1974 - 3140	Stivers-Young, 1998
1	1	Jul - Aug	Nov	4000	Thorup-Kristensen 2001
		van Trag.	1.001.	1000	
Yellow mustard	<i>Brassica juncea</i> [L.]	Nov.	Mar.	6700 - 9200	Hartz et al., 2005
		Mar.	May	≈2200 - 3000	Wortman et al., 2012
White mustard	<i>Sinapis alba</i> L.	Aug.	Oct.	2360 - 6397	Collins et al., 2006
		Nov.	Mar.	8300 - 9200	Hartz et al., 2005
		AugSep.	OctNov.	1626 - 4358	Stivers-Young, 1998
		Mar.	May	≈1250 - 3100	Wortman et al., 2012

† Study was conducted in New Zealand, where March is the start of autumn.

Common name	Scientific name	Planted	Harvested	Total dry aboveground biomass (kg ha ⁻¹)	Reference
Annual ryegrass + hairy vetch	Lolium multiflorum Lam. Vicia villosa Roth.	Fall	Apr.	1880 - 2920	Kuo and Jellum, 2002
Annual ryegrass + crimson clover	<i>Lolium multiflorum</i> Lam. + <i>Trifolium incarnatum</i> L.	Aug.	Apr.	3400	Odhiambo and Bomke, 2001
Cereal rye + crimson clover	Secale cereale L. + Trifolium incarnatum L.	Aug.	Apr.	5060 - 6220	Odhiambo and Bomke, 2001
		Oct.	Apr.	2300 - 5180	Ranells and Wagger, 1996
		Oct.	Dec.	200 - 700	Ranells and Wagger, 1997
		Oct.	Apr.	3120 - 3340	Ranells and Wagger, 1997
Cereal rye + hairy vetch	<i>Secale cereale</i> L. + <i>Vicia</i> <i>villosa</i> Roth	Oct.	MarApr.	605 - 3650	Daniel et al., 1999
		SepOct.	AprMay	2700 - 9700	Parr et al., 2011
		Oct.	Apr.	3010 - 5420	Ranells and Wagger, 1996
		OctNov.	Apr.	5720-8180	Sainju et al., 2005
Cereal rye + winter pea	Secale cereale L + Pisum sativum var. arvense (L.) Poir.	SepOct.	AprMay	4000 - 9600	Parr et al., 2011
Oilseed radish + cereal rve	Raphanus sativus L. var. oleiformis + Secale cereale	Oct.	Nov.	667 - 4406	Cavadini, 2013
		AugSep.	Nov.	1640 - 2584	White and Weil, 2010

Table 1.3 Cover crop mixture dry aboveground biomass, as reported in the literature.

Table 1.3 (cont'd)

Common name	Scientific name	Planted	Harvested	Total dry aboveground biomass (kg ha ⁻¹)	Reference
Oilseed radish + oats	Raphanus sativus L. var. oleiformis + Avena sativa L.	Oct.	Nov.	592 - 5201	Cavadini, 2013
Oilseed radish + pea	Raphanus sativus L. var. oleiformis + Pisum sativum L.	Aug.	Oct.	2860 - 3180	Möller and Reents, 2009
Oilseed radish + common vetch	Raphanus sativus L. var. oleiformis + Vicia sativa L.			3300	Möller and Reents, 2009

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CHAPTER 2

GROWTH CHARACTERISTICS OF BRASSICACEOUS SPECIES USED AS FALL-PLANTED COVER CROPS IN MINNESOTA AND MICHIGAN

ABSTRACT

Research was conducted to evaluate the growth characteristics of two brown mustards (Brassica juncea [L.] Czern. 'Pacific Gold' and one variety not stated (VNS) accession), one rapeseed (Brassica napus L. 'Dwarf Essex'), one hybrid turnip (Brassica rapa L. x B. napus L. 'Pasja'), six oilseed radishes [Raphanus sativus L. var. oleiformis and R. sativus L. var. longipinnatus Daikon VNS #1, Daikon VNS #2, 'Defender', 'Driller', Groundhog™ (Ampac Seed Co., Tangent, OR), and 'Tillage'], and two white mustards (Sinapis alba L. 'Ida Gold' and 'Accent') as potential cover crops. Trials were conducted in St. Paul, MN and Bath, MI for two years. Cover crop performance among Brassicaceae species was generally similar. The only differences detected in final aboveground biomass were in Minnesota, where 'Ida Gold' mustard produced 57 and 67% more biomass than 'Driller' and Groundhog[™] oilseed radishes, respectively, and 'Pacific Gold' mustard produced 65% more biomass than Groundhog[™] oilseed radish. Differences in final oilseed radish dry root biomass also occurred in the Minnesota trial, where 'Driller' oilseed radish produced an average of 74% more dry root biomass than Daikon VNS #2, 'Defender', and GroundhogTM oilseed radishes. Aboveground biomass N uptake was similar among Brassicaceae species, ranging from 100-131 kg N ha⁻¹ in Minnesota and 81-109 kg N ha⁻¹ in Michigan. Ground cover and growth curves were generated for Groundhog[™] and Daikon VNS #1 oilseed radishes, 'Pasja' hybrid turnip, and 'Pacific Gold' mustard. In both Minnesota and Michigan, ground cover accumulation was similar for the four accessions, which can be expected to provide 50% ground cover within 315 accumulated growing degree days

(GDD) in Minnesota and 335 GDD in Michigan. In an average year, 315-335 GDD accumulate during the first three weeks of August in both states, indicating how rapidly these accessions can provide ground cover. In both Minnesota and Michigan, Daikon VNS #1 and Groundhog[™] oilseed radishes accumulated biomass at distinctly different rates than 'Pacific Gold' mustard and 'Pasja' turnip. Growth rates plateaued more quickly for the oilseed radishes than for the mustard and hybrid turnip in both trials. Flowering varied considerably among accessions in both trials. While the white and brown mustards generally flowered, 'Dwarf Essex' rapeseed, 'Pasja' hybrid turnip, and the named oilseed radishes did not. There were notable differences in winter survival, as well. The white mustard accessions did not survive the winter. Zero to ten % of the oilseed radish and brown mustard accession plants survived the winter. 'Dwarf Essex' rapeseed and 'Pasja' hybrid turnip respectively had survival rates of 7-88 and 1-59 %. The results from this research suggest that in the Midwestern U.S., Brassicaceae cover crops are similar in terms of time to canopy closure, aboveground biomass production, and aboveground N accumulation. If these characteristics are the only ones of concern to a producer, the producer would do well to choose the cheapest or most readily-available seed.

INTRODUCTION

Cover crops can contribute to the profitability and sustainability of crop production systems (Cherr et al., 2006; O'Reilly et al., 2011) as the costs of inputs such as fertilizer rise. The use of cover crops in the Brassicaceae (mustard family) has become increasingly common (Ngouajio and Mutch, 2004). Brassicaceae cover crops include mustards (e.g., *Brassica juncea* [L.] Czern. and *Sinapis alba* L.), radishes (*Raphanus sativus* L. var. *oleiformis* and *R. sativus* L. var. *longipinnatus*), rapeseed (*Brassica napus* L.), and turnip (*Brassica rapa* L. and *B. rapa* L. x *B. napus* L.). Researchers and university extension educators view these species as valuable additions to the spectrum of available cover crops (Ngouajio and Mutch, 2004; Snapp et al., 2006; Sundermeier, 2008). With regard to nomenclature, the radishes grown as cover crops are closely related and are usually either oilseed radish (*Raphanus sativus* L. var. *oleiformis*) or forage radish (*R. sativus* L. var. *longipinnatus*) (Weil et al., 2009). The subspecies readily interbreed and management recommendations for both are the same (Weil et al., 2009). Both subspecies will henceforth be referred to as 'oilseed radish'. Brassicaceae cover crops generally have small seeds, broad leaves, and deep roots (Snapp et al., 2007). Oilseed radish roots can reach a depth of 2.4 m in 11 weeks (Thorup-Kristensen and Kristensen, 2004). Since they are cool-season annual plants, Brassicaceae cover crops are useful in the often cool and unpredictable climate of the upper U.S. Midwest. These species can germinate at soil temperatures as low as 4°C soil (Snapp et al., 2006). When planted in the fall Brassicaceae cover crops are killed by cold winter temperatures in climates like Minnesota and Michigan, which allows for easy spring tillage operations (Stivers-Young, 1998).

Producers have expressed interest in the use of Brassicaceae cover crops to help manage N in production systems. Meisinger et al. (1991) stated in a research review that Brassicaceae cover crops on average reduced the amount of N leached by 60-75% as compared with non-cover crop controls. The researchers also noted that Brassicaceae cover crops are not winter hardy, and that they decompose more easily than grass cover crops, allowing for rapid remineralization of N. Fall-planted oilseed radishes have been found to assimilate 80-170 kg N ha⁻¹ (Allison et al., 1998; Axelsen and Kristensen, 2000; Dean and Weil, 2009; Isse et al., 1999; Thorup-Kristensen, 1994; Thorup-Kristensen, 2001; Thorup-Kristensen, 2006). Fall-planted mustards, turnips, and rapeseed have been shown to accumulate 50-170 kg N ha⁻¹ (Brennan and

Boyd, 2012; Dean and Weil, 2009; Muir and Bow, 2009; Stivers-Young, 1998). Even over short growing periods, Brassicaceae species are efficient N scavengers. Stivers-Young (1998) found that five weeks after planting, turnip (*B. rapa*) had taken up 69 kg N ha⁻¹ while two accessions of mustard (*S. alba,* formerly *B. hirta*) had accumulated 51-62 kg N ha⁻¹. In a study by Brennan and Boyd (2012), mustards (a mixture of *S. alba* and *B. juncea*) accumulated N at a faster rate than cereal rye and a legume/cereal rye mixture.

Brassicaceae cover crops can improve soil physical properties. Oilseed radish, in particular, can alleviate soil compaction and is sometimes referred to as a "biological tillage tool" (Chen and Weil, 2010). In a study using a mini rhizotron camera, Williams and Weil (2004) observed soybean (*Glycine max* (L.) Merr.) roots growing through channels in compacted soil created by oilseed radish and canola roots. Lehrsch and Gallian (2010) found that oilseed radish improved soil characteristics related to water dynamics (e.g., hydraulic conductivity and water infiltration through pores). Oilseed radish can increase soil aggregate stability, though this effect may be short-lived in conventional tillage systems (Dapaah and Vyn, 1998).

Brassicaceae cover crops are also of interest as a weed-suppression tool. Cover crops can create environmental conditions unfavorable for weed germination and growth. They can encourage the activity of weed seed predators and weed seedling pathogens (Conklin et al., 2002; Pullaro et al., 2006); act as a mulch that smothers or shades out weeds (Smith et al., 2008; Stivers-Young, 1998); compete with weeds for space, nutrients, or light (Linares et al., 2008); and some cover crops are allelopathic (Weston, 1996). Among Brassicaceae cover crops, oilseed radish is a good choice for the suppression of winter annual weeds in the fall due to quick growth and large biomass accumulation (Ngouajio and Mutch, 2004; Stivers-Young, 1998; Sundermeier, 2008). Forage radish 'Daikon' and oilseed radish 'Adagio' produced up to 4,912 kg ha⁻¹ dry

above-ground biomass in a study by Dean and Weil (2009). Other studies have found oilseed radish can produce 3,000-5,600 kg ha⁻¹ of biomass when planted in the summer or fall (Allison et al., 1998; Axelsen and Kristensen, 2000; Isse et al., 1999; Thorup-Kristensen, 2001; Thorup-Kristensen, 2006).

Brassicaceae cover crops also have the potential to reduce nematode and soil-borne pathogen populations. Certain cultivars of oilseed radish, such as 'Adagio' and 'Colonel', can be used as a trap-crop for sugar beet cyst nematodes (Smith et al., 2004). Brassicaceae cover crops produce glucosinolates, which upon breakdown form compounds [including isothiocyanates (ITCs)] that have been shown to be biocidal against soil-borne pathogens (Angus et al., 1994; Brown and Morra, 1997; Dunne et al., 2003; Kirkegaard et al., 1996; Sarwar et al., 1998). Greenhouse and field studies have shown that Brassicaceae residues can also reduce disease incidence in some cases (Blok et al., 2000; Larkin and Griffin, 2007; Snapp et al., 2007), but not others (Bensen et al., 2009; Hartz et al., 2005; Wiggins and Kinkel, 2005).

Research into Brassicaceae cover crops ranges across the U.S. and Canada (Collins et al., 2007; Dean and Weil, 2009; Vyn et al., 2000). Kaspar et al. (2007) identified several priorities for cover crop research, including the need for more information on specific cover crop cultivars. Furthermore, Cherr et al. (2006) argued that studies in which cover crop growth parameters are sampled multiple times over the course of the growing season may provide more meaningful information to producers than those which only collect data once. Finally, the U.S. Department of Agriculture Natural Resources Conservation Service (USDA-NRCS) has indicated a need for data on the growth of Brassicaceae cover crop species for inclusion into planning tools such as RUSLE2 (Revised Universal Soil Loss Equation, Version 2) and WEPS (Wind Erosion Prediction System), which assist producers in the selection of cover crops (J. Douglas, pers.

comm., 2010). The objectives of this research were to: a) compare the performance of Brassicaceae cover crop accessions within and between species, b) evaluate Brassicaceae cover crop performance in two Midwestern states (Minnesota and Michigan), and c) collect Brassicaceae cover crop growth data for use in USDA-NRCS models.

MATERIALS AND METHODS

Four mustards (two accessions of brown mustard and two accessions of white mustard), one rapeseed, one hybrid turnip, and six accessions of oilseed radish were evaluated in Minnesota and Michigan during the falls of 2010 and 2011. Individual accession names can be found in Table 2.1.

Site description, experimental design, and field management

The Minnesota trial was located on the University of Minnesota St. Paul campus (44.99° N; 93.19° W), where the soil was a well-drained Waukegan silt-loam (fine-silty over sandy mixed super active mesic Typic Hapludolls). The Michigan trial was located at the Natural Resources Conservation Service (USDA-NRCS) Rose Lake Plant Materials Center (PMC) (42.81° N; 84.44° W) in Bath, MI. The soil was a poorly drained Colwood loam (fine-loamy mixed active mesic Typic Endoaquolls). Field preparation and planting dates and methods are listed in Table 2.2. Both trials were conducted using a randomized complete block design with four replicates. Experimental units were a minimum of 3 x 10 m. Planting rates were: mustards 9.0 kg ha⁻¹, oilseed radishes 11.2 kg ha⁻¹, rapeseed 5.6 kg ha⁻¹, and turnip 2.2 kg ha⁻¹. In Minnesota in 2011 planting rates were adjusted so that each planting rate was of pure live seed (PLS). Adjustments were based on germination tests. Cover crops were drilled in August –

September of each year (Table 2.2). In Minnesota, plots were hand-weeded 7 d after planting (DAP) in 2010. Volunteer oat and annual grasses were controlled in the Michigan trial with quizalofop (quizalofop p-ethyl ethyl(r)-2-[4-(6-chloroquinoxalin-2-yloxy) - phenoxypropionate) at 0.07 kg a.i. $ha^{-1} + 1\%$ v v⁻¹ crop oil concentrate.

Data collection

Four accessions ('Pacific Gold' brown mustard, 'Pasja' turnip, and Daikon VNS #1 and GroundhogTM oilseed radishes) were chosen for more detailed measurements. In the Minnesota trial, % ground cover (amount of canopy closure) was estimated every 10-14 d by counting the number of centimeter marks visible on a meter stick placed horizontally on the ground under the canopy. In the Michigan trial % cover was estimated every 10-14 d using the grazing stick method (Smith et al., 2010). The grazing stick had a grid on which ten dots were printed. % cover was estimated by placing the grazing stick below the cover crop canopy, counting the number of visible dots on the grid, subtracting the result from ten, and then multiplying by 100. Above- and belowground biomass data were also collected every 10-14 d. In the Minnesota trial, one subsample per plot was collected using a 0.2 m² quadrat. Quadrat locations were flagged to avoid sampling in the same area multiple times.

Biomass samples were collected from all twelve Brassicaceae accessions once each year in the fall, before the onset of a hard freeze. In the Minnesota trial, two 0.25 m² subsamples were collected per plot. In Michigan, one 0.2-m² subsample was collected per plot. In both trials, plants in each quadrat were pulled by hand and separated into root and shoot fractions. Samples were dried at 60° C for 5-21 d, and weighed. These samples were then analyzed for tissue N

concentrations. In the Minnesota trial, tissue N was determined by combustion using a ThermoFinnigan FlashEA organic elemental analyzer (Thermo Fisher Scientific Inc., Waltham, MA) for 'Pacific Gold' mustard, 'Pasja' turnip, Groundhog[™] oilseed radish, and 'Dwarf Essex' rapeseed in 2010. For all other accessions, tissue N was determined with near infrared spectroscopy (NIRS) using a Perten DA 7250 NIRS analyzer (Perten Instruments, Inc., Springfield, IL). Different analysis methods were employed because of cost-effectiveness concerns. In the Michigan trial in 2010, samples were analyzed for total Kjeldahl nitrogen (TKN) content using the Hach method (Watkins et al., 1987). In 2011 samples were tested for TKN by A&L Great Lakes Laboratories (Fort Wayne, IN) using plant TKN AOAC method 978.04 (AOAC, 2012). Root and shoot N were analyzed separately in both Minnesota and Michigan.

Flowering status of the Brassicaceae species was noted every 10-14 d throughout the fall. Winter survival was determined in March-April by counting the number of green cover crop plants and calculating the % survival from cover crop stand counts the previous fall. The exception was in spring 2012 in Minnesota where % survival was estimated visually.

Weather data and growing degree day calculations

Daily air temperature and precipitation data for 2010-2012 in St. Paul, MN were obtained from the Minnesota Department of Natural Resources interactive online tool (MDNR, 2015) (Figure 2.1). Weather data for Bath, MI were obtained from the Michigan State University Enviro-weather website (MSUEW, 2015), with the exception of December-April precipitation data which were collected from the Haslett, MI weather station via the NOAA website (NOAA, 2015). The 30-yr monthly averages (1981-2010) for both locations were obtained from the National Oceanic and Atmospheric Administration (NOAA) website (NOAA, 2015).

Temperature data were used to calculate growing degree days (GDD) from the time of planting using the standard formula

$$GDD = (T_{max} + T_{min})/2 - T_{base}$$

where T_{max} is the maximum daily temperature and T_{min} is the minimum daily temperature. T_{base} was set at 5°C (Morrison et al., 1989). T_{max} and T_{min} values less than 5°C were set to 5°C before calculating GDD (McMaster and Wilhelm, 1997).

Data analysis

Data were checked for normality and homogeneity of variance. Aboveground final biomass, N accumulation, and oilseed radish root N accumulation were log base 10 transformed to improve normality. Data were analyzed using PROC MIXED in SAS Enterprise Guide v. 6.1 (SAS Institute Inc., Cary, NC). Replication was treated as a random factor. The Tukey-Kramer adjustment to ANOVA was used to test for significant differences in the means ($\alpha \le 0.05$). Means were separated using the PDMIX 800 macro (Saxton, 1998). Nitrogen accumulation was determined by multiplying tissue N content by biomass.

Percent ground cover and biomass accumulation data were collected season-long for Groundhog[™] and Daikon VNS#1 oilseed radishes, 'Pacific Gold' mustard, and 'Pasja' hybrid turnip. PROC GLM in SAS Enterprise Guide v. 6.1 (SAS Institute Inc., Cary, NC) was used to fit these data to second order polynomial (quadratic) equations in the form of

$$y = ax^2 + ax + b$$

where y is biomass accumulated or percent ground cover for a given accumulation of GDD, x is GDD accumulated, and b is the y-intercept of the equation. Data for each accession were combined across years to investigate the effect of GDD on biomass accumulation and percent ground cover

(Hayden, 2014). GraphPad Prism v.6.02 (GraphPad Software, La Jolla, CA) was used to conduct an extra sum-of squares F-test to determine whether one line could be fitted to the ground cover and biomass accumulation data for all accessions in each trial ($\alpha \le 0.05$). When the F-test was significant, the null hypothesis ("one equation fits the pooled accession data") was rejected and only equations for the individual accessions are presented. When the F-test was not significant, the general equation for the pooled data is presented. In all cases, the dependent variable was either percent cover or dry aboveground biomass, while the independent variable was GDD.

RESULTS AND DISCUSSION

Weather

The Brassicaceae cover crop growing season generally encompasses August – November, depending on planting date and the weather. September 2010 was 19% cooler than average in Minnesota (284 accumulated GDD compared with 338 GDD), but only 12% cooler than average in Michigan (319 vs. 358 GDD) (Figure 2.1). Growing degree days are calculated from temperature data and thus can serve as a rough proxy for these data. Winter and early spring temperatures are of interest because they can affect the overwintering of cover crops. March of 2012 was warmer than average in both Minnesota and Michigan (178 and 211 GDD, respectively, vs. an average of 0 GDD for both locations).

There were some differences in precipitation (Figure 2.1). It should be noted that the first planting date in each trial-year did not occur before mid-August, and the last data collection date in each trial-year occurred between 1 November and 14 November. In Minnesota, August - November 2010 was 25% wetter than average with a total precipitation of 399.3 mm as compared with the norm of 297.3 mm. In contrast, in Michigan August – November 2010 was

20% drier than average with 205.2 mm of precipitation as compared with 246.6 mm. The trends reversed in 2011, when Minnesota experienced a dry period from August – November in which total precipitation was 62% of average. Precipitation in Michigan during August – November 2011 was within norms and totaled 253.8 mm, compared with the average of 246.6 mm.

Ground cover and biomass accumulation of four Brassicaceae accessions

Ground cover accumulation. An extra-sum of squares F-test (H₀: one equation fits pooled accession data) was not significant for both the Minnesota and Michigan data (p = 0.18 and 0.71, respectively). A single second-order polynomial equation was thus fitted to the pooled GroundhogTM oilseed radish, Daikon VNS #1 oilseed radish, 'Pacific Gold' brown mustard, and 'Pasja' hybrid turnip ground cover accumulation data in each trial. In the Minnesota trial the fitted equation is

$$y = -0.0006x^2 + 0.80x - 142$$

where y is percent ground cover at a given number of GDD and x is GDD (Figure 2.2). The r^2 for this equation is 0.85, P = <0.0001. According to this equation, all four Brassicaceae accessions would be expected to reach 50% ground cover at 315 accumulated GDD. Based on the 30-yr average monthly GDDs (Figure 2.1), if these cover crops were planted on 1 August they would achieve 50% ground cover within three weeks of planting. While a single line fits all four accessions, based on the means and standard errors 'Pacific Gold' mustard percent cover declined earlier than the other Brassicaceae accessions (Figure 2.2). Mustards are more coldsensitive than oilseed radish and hybrid turnip (data not shown). Freezing weather at the end of the data collection period likely caused 'Pacific Gold' to wilt, thus the decreased ground cover. In the Michigan trial, the equation fitted to the percent ground cover data for all four accessions across two years is

$$y = -0.0005x^2 + 0.65x - 112$$

where y is percent ground cover and x is GDD (Figure 2.3). The r² for this equation is 0.89, P = <0.0001. Based on this equation, the four accessions could be expected to reach 50 % ground cover after 335 GDD. If GroundhogTM oilseed radish, Daikon VNS #1 oilseed radish, 'Pacific Gold' brown mustard, or 'Pasja' hybrid turnip was planted on 1 August in central Michigan, as in Minnesota it could reach 50% ground cover within three weeks of planting, based on 30-yr average monthly GDD accumulation (Figure 2.1). Averaged across the two years, all four accessions reached peak ground cover (90% or greater) by 550-555 GDD in both Minnesota and Michigan.

Rapid cover crop canopy closure is one mechanism through which cover crops confer benefits to a crop production system. Good canopy cover suppresses weeds by shading the ground and preventing emergence of winter annual weeds (Lawley et al., 2012; O'Reilly et al., 2011) and also helps decrease soil erosion by protecting the soil from the impact of raindrops (Dabney et al., 2001). While the equations in our study were generated from data collected from four accessions, rapid ground cover was also observed in the full set of twelve Brassicaceae accessions studied (data not shown). Progression of % ground cover by Brassicaceae cover crops is not commonly reported in the literature. Stivers-Young (1998) found that in the northeastern USA, *R. sativus* and *S. alba* (formerly *B. hirta*) reached 23-33% ground cover at 20 DAP and 100% ground cover by seven WAP; in another year of that study, ground cover for six different Brassicaceae accessions (including *R. sativus*, *B. napus*, *B. rapa*, and *S. alba* [formerly *B. hirta*]) ranged from 40-68% at 18 DAP.

Aboveground biomass accumulation. An extra-sum of squares F-test (H₀: one equation fits pooled accession data) was significant for both the Minnesota and Michigan data (p = 0.006 and 0.02, respectively). A single second-order polynomial equation thus could not be fitted to the pooled biomass accumulation data in each trial. However, in Minnesota a single line was fitted to 'Pacific Gold' and 'Pasja' data (p = 0.83), while another line fitted the Daikon VNS # 1 and GroundhogTM oilseed radish data (p = 0.99). The equations and r^2 and P values for the Minnesota trial are provided in Figure 2.4. Based on these equations, it would take roughly 485 GDD for the oilseed radishes to accumulate 2,000 kg ha⁻¹ of dry aboveground biomass. 'Pacific Gold' brown mustard and 'Pasja' hybrid turnip would require 475 GDD to reach 2,000 kg ha⁻¹ biomass. In spite of the similar numbers, 'Pacific Gold' mustard and 'Pasja' turnip produced more dry aboveground biomass than Daikon VNS #1 and GroundhogTM oilseed radishes and thus had steeper sloping lines. Growth rates for both groups were similar until near 500 GDD, at which point 'Pacific Gold' and 'Pasja' biomass accumulation rate increased compared to that of the radish accessions.

As in Minnesota, in Michigan a single line fitted 'Pacific Gold' and 'Pasja' data (p = 0.54), while another line fitted the Daikon VNS # 1 and GroundhogTM oilseed radish data (p = 0.88). The equations and r^2 and P values for the Michigan trial are provided in Figure 2.5. Based on these equations, it would require roughly 465 GDD for GroundhogTM and Daikon VNS #1 oilseed radishes to accumulate 2,000 kg ha⁻¹ of dry aboveground biomass, vs. 540 GDD for 'Pacific Gold' brown mustard and 'Pasja' hybrid turnip. Unlike in Minnesota, in Michigan the oilseed radish accessions had a larger initial growth rate than 'Pacific Gold' mustard and 'Pasja'

turnip, up until near 500 accumulated GDD when oilseed radish growth rate slowed as the mustard and turnip growth rate accelerated.

Rapid and large cover crop biomass production is of interest for several reasons. It allows cover crops to outcompete weeds (O'Reilly et al., 2011). Fast root growth makes Brassicaceae cover crops good N scavengers (Meisinger et al., 1991). Since Brassicaceae cover crops can also be used as forage (Barry, 2013), biomass production potential could be of interest to producers whose systems include livestock.

Characteristics of all twelve Brassicaceae accessions

Final biomass. Fall aboveground biomass data were combined over years for each trial since the accession*year interactions were not significant (P = 0.41 and 0.32 in the Minnesota and Michigan trials, respectively). In Minnesota, 'Ida Gold' mustard produced 57 and 67% more biomass than 'Driller' and GroundhogTM oilseed radishes, respectively (Table 2.3). 'Pacific Gold' mustard produced 65% more biomass than GroundhogTM oilseed radish. No other differences were detected. Nor were there any differences among the Brassicaceae accessions in final dry aboveground biomass produced in Michigan (Table 2.3). The failure to detect differences in the Michigan data may have been the result of insufficient subsample size and number. In Minnesota, biomass ranged from 3311 kg ha⁻¹ (GroundhogTM radish) to 5542 kg ha⁻¹ ('Ida Gold' mustard) (Table 2.3). In Michigan biomass ranged from 2476 kg ha⁻¹ ('Dwarf Essex' rapeseed) to 3747 kg ha⁻¹ ('Driller' oilseed radish). Differences in biomass between the trials were likely the result of varying weather and soil fertility.

Mustard, rapeseed, and hybrid turnip final root biomass data are not presented because the sampling protocol was insufficient to capture small, fibrous roots. Fall final oilseed radish

dry root biomass data were combined over years for each trial because the accession*year interaction was almost not significant (P = 0.04) in Minnesota and not significant (P = 0.78) in Michigan. In the Minnesota trial, 'Driller' oilseed radish produced an average of 74% more dry root biomass than Daikon VNS #2, 'Defender', and GroundhogTM oilseed radishes (Table 2.4). Oilseed radish root biomass ranged from 1226-2483 kg ha⁻¹ in Minnesota and 846-1294 kg ha⁻¹ in Michigan. No differences were detected in root biomass production in the Michigan trial. The failure to detect differences may have been the result of insufficient subsample size and number in Michigan, or because roots were hand-pulled instead of excavated with a shovel. Variability in the data may also have impacted the ability to detect differences.

Aboveground biomass production was in general accord with the literature. As in our Michigan trial, Stivers-Young (1998) detected no differences in biomass in northeastern U.S. early-fall plantings of oilseed radish (*R. sativus*), two accessions of mustards (*S. alba*, formerly *B. hirta*), turnip (*B. rapa*), and rapeseed (*B. napus*). However, for a later planting of Brassicaceae cover crops, by late October turnip had more biomass than the other Brassicaceae species, a lead it maintained at the mid November biomass sampling (Stivers-Young, 1998). Had biomass sampling in our study continued later into the fall, it is possible that differences in Michigan biomass might have been detected. Dry matter production in the Stivers-Young (1998) study ranged from 2525 kg ha⁻¹ to 2826 kg ha⁻¹ by late October, less than many accessions produced in our Minnesota trial in the same time frame, but close to the range produced in our Michigan trial (Table 2.6). In our Minnesota trial, biomass production by mid-October ranged from 3311 to 5542 kg ha⁻¹, while that found by Stivers-Young (1998) in mid-November ranged from 3947 kg ha⁻¹ to 4358 kg ha⁻¹ (Stivers-Young, 1998).

Nitrogen content. Tissue N concentration data allow for a partial estimation of N accumulated ha^{-1} when combined with biomass production data (Table 2.3). There was no treatment*year interaction for either location (P = 0.17 and 0.33 in Minnesota and Michigan, respectively), so aboveground N accumulation data were combined over years. There were no differences detected in aboveground N accumulation for the different Brassicaceae accessions in both Minnesota and Michigan. In Minnesota, N accumulation ranged from 100-131 kg N ha⁻¹ and in Michigan, N accumulation ranged from 81-109 kg N ha⁻¹. Nitrogen uptake was lower in the Michigan trial than the Minnesota trial, possibly due biomass production differences caused by varying soil fertility and weather conditions.

The treatment*year interaction was barely significant in the Minnesota trial (P = 0.04) and not significant in Michigan (0.40), so oilseed radish root N accumulation data were combined across years in each trial (Table 2.4). 'Driller' oilseed radish accumulated 39-59 % more N than the other oilseed radish accessions. This is not a surprise, given the large root biomass production of 'Driller' oilseed radish (Table 2.4). No other differences were detected in the Minnesota trial. Nor were differences detected in the Michigan trial.

Aboveground tissue N accumulation in our trials were similar to those found in other studies. Fall-planted oilseed radishes have been shown to accumulate 100-170 kg N ha⁻¹ (Allison et al., 1998; Axelsen and Kristensen, 2000; Isse et al., 1999; Thorup-Kristensen, 1994; Thorup-Kristensen, 2001; Thorup-Kristensen, 2006). In our Minnesota trial, all accessions had accumulated at least 100 kg N ha⁻¹ by roughly eight weeks after planting, while in Michigan the smallest amount of N accumulated by that point was 81 kg ha⁻¹ (Tables 2.1 and 2.3). Five weeks after planting, Stivers-Young (1998) found turnip to have accumulated 69 kg N ha⁻¹ while two white mustard accessions had accumulated 51 kg N ha⁻¹.

Flowering and seed production

Flowering varied among cover crop accessions in both trials (Table 2.7). The brown mustard accessions flowered within 45 days after planting (DAP), except in the Michigan trial in 2011 when brown VNS mustard did not flower and 'Pacific Gold' flowered within 75 DAP (Table 2.5). The white mustard accessions flowered within 37-65 DAP, with the exception of 'Ida Gold' which in Michigan flowered within 89 DAP in 2010 and did not flower in 2011. The VNS oilseed radish accessions flowered within 37-78 DAP. Daikon VNS #1 did not flower in the Michigan trial in 2011. Rapeseed, hybrid turnip, and the named oilseed radish accessions did not flower in either location in either year. Differences in flowering were likely due to the interaction between accession genotype and the environment. Factors affecting time to flowering include temperature and photoperiod (Nanda et al., 1996; Robertson et al., 2002). In a study of four Brassicaceae species including B. juncea (brown mustard), cooler temperatures were calculated to shorten the time to flowering by 22-41 growing degree days (base temperature 0 °C) for every 1 °C decline in average temperature (Nanda et al., 1996). Within a Brassicaceae species, different genotypes respond differently to temperature and photoperiod (Robertson et al., 2002).

Accessions that flower may be of interest to producers who want to provide a nectar source for beneficial insects. Insects were observed visiting mustard accession flowers as late as November in the Michigan trial (data not shown). Time to flowering is also of interest because plants which have reached the reproductive phase cease allocation of resources to vegetative structures (Rossato et al., 2001). Early flowering Brassicaceae accessions could provide reduced ecosystem services such as weed suppression due to potentially lower biomass production and

less canopy closure. Time to flowering is also of concern because Brassicaceae cover crop plants may produce seed to add to the weed seed bank. Although none of the accessions in this study set mature seed (data not shown), producers who grow Brassicaceae cover crops should monitor their crop and be prepared to control them with mowing or herbicides before seed set occurs. Brassicaceae cover crops could also become established as weeds via seeds which are planted but fail to germinate immediately, instead persisting as part of the soil seed bank. In Michigan, newly-germinated oilseed radish seedlings were observed in the spring both years, seven months after the cover crops were planted (data not shown).

Winter hardiness

The accessions varied in terms of winter hardiness (Table 2.5). 'Pasja' hybrid turnip and 'Dwarf Essex' rapeseed winter survival ranged from 1-59% and 7-88%, respectively, across years in Minnesota and Michigan. In the Minnesota trial only 'Pasja' hybrid turnip and 'Dwarf Essex' rapeseed survived the winter. However, in Michigan, 1-10% of several oilseed radish accessions and both brown mustards also survived winter 2011-2012 (Table 2.5). Increased winter survival in 2011-2012 was likely due to the weather. From December 2011-March 2012, there were 188 and 248 GDD accumulated in Minnesota and Michigan, respectively, compared to 15 and 50 GDD over the same period in 2010-2011 (Figure 2.1). Cover crops that overwinter may provide benefits such as continuous ground cover in the spring that decreases soil erosion. However, the need to terminate surviving cover crops may delay cash crop planting in wet years. If spring logistics are of concern to producers, cold-sensitive cover crop species such as mustards are a better choice.

CONCLUSIONS

The Brassicaceae accessions in these trials had similar growth characteristics and were suitable for use as fall-planted cover crops. With regard to percent ground cover and biomass accumulation, final aboveground biomass, and N uptake, little differentiation was detected. If these characteristics are the only ones of concern to a producer, the producer would do well to choose the cheapest or most readily-available seed. However, Brassicaceae cover crop do differ with regard to other traits, which could affect cover crop selection. In our trials, white and brown mustards flowered before winter-kill, while rapeseed, hybrid turnip, and the named oilseed radish accessions did not. In terms of winter survival, rapeseed and hybrid turnip had the highest incidence of over-wintering while mustards and oilseed radishes had the lowest incidence. Though not quantified in our trials, there are also known differences in Brassicaceae cover crop pest suppression. For example, 'Defender' oilseed radish was specifically bred to be a trap-crop for sugar beet cyst nematode.

Plant breeders continue to develop and refine Brassicaceae germplasm, so future work could continue to evaluate oilseed radish, turnip, and mustard cover crop accessions. Root biomass could be better quantified by excavating roots with a shovel, rather than pulling the plants by hand. It would be interesting to trial accessions under varying fertility management regimes, to better estimate how fall-planted Brassicaceae cover crops perform under limited vs. optimal nutrient conditions.

APPENDICES

APPENDIX A

CHAPTER 2 TABLES AND FIGURES

Table 2.1. Brassicaceae accessions evaluated in trials in St. Paul, MN and Bath, MI in 2010 and 2011.

Species	Common name Accessio	
Brassica juncea [L.] Czern.	Brown mustard	'Pacific Gold'
		Brown VNS
Brassica napus L.	Rapeseed	'Dwarf Essex'
Brassica rapa L. x B. napus L.	Hybrid turnip	'Pasja'
<i>Raphanus sativus</i> L.	Oilseed radish	Daikon VNS #1
		Daikon VNS #2
		'Defender'
		'Driller'
		Groundhog™
		'Tillage'
Sinapis alba L.	White mustard	'Ida Gold'
		'Accent'

	Minnesota	Michigan
Preceding crop	Soybean green manure;	Oats; harvested
	chopped and disked	
Field preparation prior to planting	Rototilled (15-20 cm deep);	Plowed and tilled (15-20 cm deep); also
	packed	rototilled and dragged in 2011
Cover crop planting dates	17 Aug. 2010 and 22 Aug. 2011;	13 Aug. 2010 and 1 Sep. 2011;
Cover crop planting methods	Wintersteiger cone-drill seeder †	Great Plains drill
Row spacing	15 cm	19 cm
Supplemental N	none	34 kg ha ⁻¹ N in the form of broadcast urea on
		2 Aug. 2010 and High NRG-N [™] liquid N
		fertilizer (Agro-Culture Liquid Fertilizers, St.
		Johns, MI) on 22 Aug. 2011

Table 2.2. Brassicaceae variety trial site characteristics at St. Paul, MN and Bath, MI in 2010 and 2011.

[†] 'Defender' oilseed radish was broadcast seeded and raked in on 29 Aug. 2011.

		Biomass		N uptake	
Common	Accession	Minnesota	Michigan	Minnesota	Michigan
name					
		——— kg h	a ⁻¹	——kg h	a ⁻¹ ———
Brown	Brown VNS	4652 abc‡¶	2555	100	86
	'Pacific Gold'	5453 ab	2998	112	96
Rapeseed	'Dwarf Essex'	4836 abc	2476	122	82
Hybrid turnip	'Pasja'	4245 abc	3541	106	109
Oilseed	Daikon VNS #1	3534 abc	3031	106	88
	Daikon VNS #2	3500 abc	2889	107	88
	'Defender'	4316 abc	3747	131	96
	'Driller'	3541 bc	2744	109	81
	Groundhog TM	3311 c	3109	101	87
	Tillage	3759 abc	2787	105	88
White	'Accent'	4808 abc	2544	120	81
	'Ida Gold'	5542 a	2545	124	84
P value		0.0006	0.12	0.37	0.92

Table 2.3. Final aboveground biomass production and N uptake of twelve Brassicaceae accessions evaluated in St. Paul, MN and Bath, MI⁺.

[†] Biomass sampling dates were 20-22 Oct. 2010 and 17 Oct. 2011 in Minnesota and 12 Oct. 2010 and 1 Nov. 2011 in Michigan.

‡ Within a column, means followed by the same letter do not differ ($\alpha < 0.05$).

¶ Minnesota biomass and N uptake means were back-transformed from log base 10.
		Biomass		N uptake		
Common	Accession	Minnesota	Michigan	Minnesota	Michigan	
name						
		—— kg l	na ⁻¹	——kg ha	a ⁻¹	
Oilseed	Daikon VNS #1	1855 ab‡¶	1253	33 b	22	
	Daikon VNS #2	1552b	1094	29b	21	
	'Defender'	1226b	846	30b	15	
	'Driller'	2483 a	1294	46 a	23	
	Groundhog™	1515b	1140	29b	21	
	Tillage	1851 ab	907	33 b	18	
P value		0.0001	0.27	0.001	0.22	
* D:	1	0.00 -+ .001	0 - 1 + 17 + 20	11 M	11204	

Table 2.4. Final dry root biomass production and N uptake of six oilseed radish accessions evaluated in St. Paul, MN and Bath, MI⁺.

[†] Biomass sampling dates were 20-22 Oct. 2010 and 17 Oct. 2011 in Minnesota and 12 Oct. 2010 and 1 Nov. 2011 in Michigan.

‡ Within a column, means followed by the same letter do not differ ($\alpha < 0.05$).

¶ Minnesota N uptake means were back-transformed from log base 10.

			Flower initiation date			Winter survival			
		Mini	Minnesota Michigan		Minnesota		Mich	Michigan	
Brassicaceae	Accession	2010	2011	2010	2011	2010	2011	2010	2011
							%		
Brown mustard	Brown VNS	27 Sep.	28 Sep.	27 Sep.	-	0	0	0	1
	'Pacific Gold'	27 Sep.	28 Sep.	27 Sep.	14 Nov.	0	0	0	6
Rapeseed	'Dwarf Essex'	_§	-	-	-	7	88	8	59
Hybrid turnip	'Pasja'	-	-	-	-	12	1	1	59
Oilseed radish	Daikon VNS #1	8 Nov.	19 Nov.	27 Sep.	-	0	0	0	2
	Daikon VNS #2	27 Sep.	28 Sep.	27 Sep.	1 Nov.	0	0	0	0
	'Defender'	-	-	-	-	0	0	0	10
	'Driller'	-	-	-	-	0	0	0	4
	'Groundhog'	-	-	-	-	0	0	0	0
	Tillage	-	-	-	-	0	0	0	2
White mustard	'Accent'	27 Sep.	28 Sep.	27 Sep.	1 Nov.	0	0	0	0
	'Ida Gold'	26 Oct.	9 Nov.	10 Nov.	-	0	0	0	0

Table 2.5. Flowering initiation date[†] and winter survival[‡] of Brassicaceae cover crops in MN and MI 2010-2011 and 2011-2012.

[†] Note that flower initiation dates are approximate as data were collected every 10 - 14 d.

‡ Winter survival data were collected on 6 Apr. 2011 and 29 Mar. 2012 in Minnesota and 4 Apr. 2011 and 19 Mar. 2012 in

§ Did not flower.



Figure 2.1. Monthly and 30-yr average growing degree days (base 5°C) and precipitation for St. Paul, MN (top) and Bath, MI (bottom) in 2010 and 2011. Growing degree days are represented on the left y-axis and by lines, while precipitation is represented on the right y-axis and by bars.



Figure 2.2. 'Pacific Gold' mustard (gray square), 'Pasja' turnip (black circle), Daikon VNS #1 oilseed radish (white diamond), and GroundhogTM oilseed radish (gray triangle) ground cover accumulation in Minnesota in 2011 and 2012. Polynomial regression was conducted with growing degree days (GDD) (base 5 °C) and percent ground cover as covariates. Means \pm SE.



Figure 2.3. 'Pacific Gold' mustard (gray square), 'Pasja' turnip (black circle), Daikon VNS #1 oilseed radish (white diamond), and GroundhogTM oilseed radish (gray triangle) ground cover accumulation in Michigan in 2011 and 2012. Polynomial regression was conducted with growing degree days (GDD) (base 5 °C) and percent ground cover as covariates. Means \pm SE.



Figure 2.4. 'Pacific Gold' mustard (gray square), 'Pasja' turnip (black circle), Daikon VNS #1 oilseed radish (white diamond), and GroundhogTM oilseed radish (gray triangle) dry aboveground accumulation in Minnesota in 2011 and 2012. Polynomial regression was conducted with growing degree days (GDD) (base 5 °C) and biomass as covariates. Means ± SE. Response of 'Pacific Gold' and 'Pasja' biomass production to GDD: $y = 0.0099x^2 + 1.84x - 1102$, $r^2 = 0.91$, P = <0.0001. Response of Daikon VNS #1 and GroundhogTM oilseed radishes biomass production to GDD: $y = -0.0034x^2 + 11.45x - 2754$, $r^2 = 0.94$, P = <0.0001.



Figure 2.5. 'Pacific Gold' mustard (gray square), 'Pasja' turnip (black circle), Daikon VNS #1 oilseed radish (white diamond), and GroundhogTM oilseed radish (gray triangle) dry aboveground accumulation in Michigan in 2011 and 2012. Polynomial regression was conducted with growing degree days (GDD) (base 5 °C) and biomass as covariates. Means \pm SE. R Response of 'Pacific Gold' and 'Pasja' biomass production to GDD: $y = 0.0059x^2 + 1.61x - 587$, $r^2 = 0.91$, P = <0.0001. Response of Daikon VNS #1 and GroundhogTM oilseed radishes biomass production to GDD: $y = -0.0058x^2 + 11.88x - 2254$, $r^2 = 0.97$, P = <0.0001.

APPENDIX B

NON-OILSEED RADISH BRASSICACEAE ROOT N CONTENT ANALYSIS OF VARIANCE

Table 2.6. Root tissue N concentrations of non-oilseed radish Brassicaceae cover crop accessions in trials in St. Paul, MN and Bath, MI⁺.

		Root N concentration			
Brassicaceae species	Accession	Minnesota	Michigan		
Brown mustard	Brown VNS	0.9b¶	1.7 ab		
	'Pacific Gold'	0.9b	1.6 ab		
Rapeseed	'Dwarf Essex'	1.6a	1.8 ab		
Hybrid turnip	'Pasja'	1.5 a	2.0 a		
White mustard	'Accent'	1.0b	1.2 b		
	'Ida Gold'	0.8b	1.3 b		
P-value		<0.0001	0.01		

[†] The tissue analyzed was collected 20-22 Oct. 2010 and 17 Oct. 2011 in Minnesota and 12 Oct. 2010 and 1 Nov. 2011 in Michigan.

 \ddagger Data were combined across years in both trials because there was no treatment*year interaction (P = 0.28 and 0.06 in Minnesota and Michigan, respectively).

¶Within a column, means followed by the same letter do not differ ($\alpha < 0.05$).

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CHAPTER 3

PERFORMANCE OF COVER CROP MONOCULTURES VS. BICULTURES: COVER CROP GROWTH AND IMPACT ON WEED BIOMASS AND CORN (Zea mays L.) YIELD

ABSTRACT

A three-year experiment was conducted in Lansing, MI to quantify biomass production of cover crop monocultures and biculture mixtures and investigate the impact of the cover crop treatments on fall and spring weed biomass production and corn (Zea mays L.) grain yield in a conventionally tilled system with no additional applied fertilizer. Cover crops investigated consisted of annual ryegrass (Lolium multiflorum Lam.), cereal rye (Secale cereale L.), oats (Avena sativa L.), crimson clover (Trifolium incarnatum L.), hairy vetch (Vicia villosa Roth.), and winter pea [Pisum sativum var. arvense (L.) Poir.] grown in monoculture and in biculture with oilseed radish (Raphanus sativus L. var. oleiformis Pers.). Oats and oilseed radish produced 2-11 times more fall aboveground biomass than crimson clover, the cover crop treatment with the smallest biomass production in all three years. Crimson clover failed to establish in two of the three years, while winter pea failed to survive the winter in two of the three years. Within each year, biomass production was similar across cover crop biculture treatments. Oilseed radish biomass exceeded that of the second species in each biculture, accounting for 54-99% of total biomass. Monoculture treatments produced more spring biomass than bicultures because the seeding rate of the complementary species in biculture was half that in monoculture. In addition, biomass relative yield values indicate that the oilseed radish competed with each complementary species in biculture. Oilseed radish competition with complementary species was greatest in the oilseed radish + crimson clover and oilseed radish + winter pea treatments and lowest in the

oilseed radish + cereal rye and oilseed radish + oats treatments. Overall, the cover crops did not reduce corn grain yield, with the exceptions of annual ryegrass and cereal rye each in one of three years. Annual ryegrass and cereal rye reduced corn grain yield by 51 and 24%, respectively, compared with the weedy control.

INTRODUCTION

The environmental services provided by cover crops are well documented. Cover crops can prevent soil erosion and improve soil physical properties (Chen and Weil, 2010; Hively and Cox, 2001; Kaspar et al., 2001; Meisinger et al., 1991; Villamil et al., 2006; Williams and Weil, 2004). Cover crops provide food and habitat for micro- and macro-organisms ranging from collembola to birds (Axelsen and Kristensen, 2000; Blanchart et al., 2006; Decourtye et al., 2010; Henderson et al., 2004). Cover crops can decrease the amount of N leached from agricultural systems (Kaspar et al., 2007; Ruffo et al., 2004; Tonitto et al., 2006). The use of cover crops has been suggested as one way to help decrease the nutrient load in the Mississippi River drainage basin that contributes to the hypoxic zone in the Gulf of Mexico (Strock et al., 2004). In spite of the benefits listed above, cover crop adoption remains low. In a 2006 survey of U.S. Corn Belt farmers, only 11% had used cover crops in the previous five years (Singer et al., 2007). Farmers have cited economic factors including time and monetary costs of planting and managing cover crops as key barriers to cover crop use (CTIC, 2015). There is a need for more data regarding cover crop use and ways to minimize the risk of cash crop yield reduction (Carlson and Stockwell, 2013; Kaspar et al., 2007).

The most commonly-grown cover crop species in the U.S. belong to three plant families: the Poaceae (grasses), Fabaceae (legumes), or Brassicaceae (mustards and oilseed radishes).

With regard to nomenclature, the oilseed radishes grown as cover crops are closely related and are usually either oilseed radish (*Raphanus sativus* L. var. *oleiformis* Pers.) or forage radish (*R. sativus* L. var. *longipinnatus* L.H.Bailey) (Weil et al., 2009). Said cover crop will be referred to as oilseed radish throughout this dissertation.

Grass cover crops include cereal rye, annual ryegrass, and oats. Compared with a weedy control, cereal rye and annual ryegrass have been found to increase soil organic carbon 200-400% more than Austrian winter pea and hairy vetch (Kuo et al., 1997). Kaspar et al. (2001) determined that cereal rye decreased inter-rill erosion up to 62%, while Ruffo et al. (2004) found cereal rye treatments to have 11% more ground cover residue than treatments that did not include cereal rye. Cereal rye has been found to take up residual soil N, decreasing the amount of N lost from corn/soybean [Glycine max (L.) Merr.] cropping systems (Kaspar et al., 2007; Ruffo et al., 2004). Due to ease of establishment and smaller seed costs (Table 3.1), grass species are commonly grown as cover crops. However, there are potential risks involved with grass cover crops. Annual ryegrass and cereal rye can cause delayed cash crop planting due to the difficulty of spring cover crop termination under adverse weather or soil conditions (Creamer et al., 1997; Madden et al. 2004). Annual ryegrass in the U.S. has been documented to be resistant to several herbicide classes (Martins et al., 2012; Perez-Jones et al., 2005), which could make cover crop termination difficult in no-till systems. The relatively large C:N ratio and biomass production of cereal rye increases the likelihood of N immobilization in the soil after cover crop termination (Crandall et al., 2005).

In the CTIC survey (2015), 57% of respondents reported the use of legume cover crops. Nitrogen fixed in the root nodules of legumes such as hairy vetch can be of benefit to cash crops directly as a fertilizer supplement (Ruffo et al., 2004; Sainju et al., 2001) and indirectly by

contributing to the formation of soil organic matter when in the presence of a C source (Villamil et al., 2006). Similar to grasses, legumes also entail costs and risks. Hairy vetch and winter pea are more costly to plant, per-hectare, than annual ryegrass, cereal rye, oats, and oilseed radish (Table 3.1). The small size of crimson clover seed can make planting and stand establishment difficult. Winter pea does not always survive the winter in cold climates such as that of Michigan. Legume cover crops may serve as hosts to plant-parasitic nematodes and pathogens that infect cash crops (Dabney et al., 2001). As with cereal rye, the large biomass production of hairy vetch can make it difficult to terminate in the spring. Parr et al. (2011) documented the negative impact of unsuccessfully terminated hairy vetch on corn yield, as a result of interspecific competition. Hairy vetch seed is a contaminant in harvested small grains and thus should not be grown in rotations that include wheat (*Triticum aestivum* L.) or barley (*Hordeum vulgare* L.) (Clark, 2008).

Due to the potential benefits (e.g., decreased erosion), Brassicaceae species such as oilseed radish (*Raphanus sativus* L. var. *oleiformis* Pers.) are also commonly grown as cover crops. Oilseed radish accumulates large quantities of biomass and provides dense ground cover (Lawley et al., 2011), scavenges residual soil N (Weil and Dean, 2009), may improve soil physical properties (Lehrsch and Gallian, 2010; Williams and Weil, 2004), and can be used to help manage weeds and other pests (Lawley et al., 2011; Melakeberhan et al., 2008; Stivers-Young, 1998; Wang et al., 2008). Brassicaceae cover crops involve many of the same costs and risks as grass and legume species. Mustards and oilseed radish are susceptible to the same pests and pathogens as Brassicaceae cash crops and should not be grown in close temporal proximity in a rotation (Snapp et al., 2006). The benefits of Brassicaceae cover crops, such as suppression of soil-borne pathogens, are sometimes overstated or oversimplified. For example, mustards and

oilseed radish are frequently noted for their bio-suppressive characteristics. Bensen et al. (2009), however, found no decrease in lettuce (*Lactuca sativa* L.) disease with long-term mustard (*B. juncea* and *S. alba*) use, in spite of evidence of decreased lettuce drop incidence in the short-term. Brassicaceae species, along with other cover crops, have been documented to negatively impact cash crop stand (Haramoto and Gallandt, 2005). While Brassicaceae species such as oilseed radish are frequently planted in the fall to scavenge residual soil N, there are concerns as to the fate of the accumulated N. Oilseed radish winter-kills and has a relatively small C:N ratio. After oilseed radish termination or winter kill, there is a window prior to cash crop planting during which N released by the cover crop biomass could be lost from the system leaching or denitrification (Radersma and Smit, 2011).

Cover crop impact on cash crop yield depends on a variety of factors including the specific cover crop and cash crop grown, cover and cash crop management, soil fertility, and the weather. Interestingly, the literature reveals that when more than one type of impact of a given cover crop on cash crop yield is observed, the impacts range either from positive to neutral or neutral to negative. Cover crop impact can vary across years.

Even for a single specified cash crop such as corn, the literature is mixed as to the effect of a cover crop on grain yield. Some studies have shown that a preceding annual ryegrass, cereal rye, oats, oilseed radish, crimson clover, hairy vetch, or winter pea cover crop generally had no impact on corn grain yield (Crandall et al., 2005; Isse et al., 1999; Kuo and Jellum, 2002; Parr et al., 2011; Vyn et al., 2000; Yenish et al., 1996). However, some studies have found cover crops to positively impact corn yield in at least some site-years (Dapaah and Vyn, 1998; Decker et al., 1994; Kuo and Jellum, 2002; Parr et al., 2011; Vyn et al., 1999). Legume cover crops such as hairy vetch can contribute N to cropping systems for use by the following cash crops (Miguez

and Bollero, 2005; Tonitto et al., 2006). Cover crops with extensive root systems such as oilseed radish and cereal rye can create macropores in the soil which help cash crop roots access water (Williams and Weil, 2004). Cover crops that have been terminated can conserve soil moisture by acting as a mulch (Unger and Vigil, 1998).

Cover crops also have the potential to decrease cash crop yield. Grass species such as annual ryegrass and cereal rye most often are observed to have negative or neutral impacts on corn yield (Dapaah and Vyn, 1998; Kaspar et al., 2007; Parr et al., 2011; Vyn et al., 1999). Farmers have cited the risk of delayed cash crop planting as one barrier to cover crop adoption (CTIC, 2015). Delayed planting often correlates to decreased yields in agronomic systems. Cover crops can deplete limited soil moisture resources in dryland regions or drought years (Meisinger et al., 1991; Ruffo et al., 2004). Cereal rye and other cover crops with large C:N ratios can immobilize soil N after cover crop termination (De Bruin et al., 2005). Cover crops with hard seed, that produce viable seed before termination, or are not completely terminated prior to cash crop planting may act as weeds that compete with the cash crop for resources such as sunlight and N. Williams and Weil (2004) attributed decreased soybean yield to competition between soybeans and a poorly controlled canola (*Brassica napus* L.) cover crop. Cover crops may also act as "green bridges", harboring insects or pathogens whose populations would otherwise subside during a fallow period (Odhiambo et al., 2012).

From 2014 to 2015, the percentage of respondents who planted cover crop mixtures increased from 60% to 67% (CTIC, 2015). Research has been done on a variety of cover crop bicultures, often consisting of a legume combined with a grass. The results of different studies involving mixtures are difficult to compare since planting rates and proportions of component species are a confounding factor (Miyazawa et al., 2014). Cover crops grown in mixtures can

benefit each other by occupying complementary niches (Hauggaard-Nielsen et al., 2001; Teasdale and Abdul-Baki, 1998). Kuo and Jellum (2002) suggested that upright grass cover crops provided vining hairy vetch with a support structure on which to grow and better access light. Cover crop mixtures can be more resilient and stable than pure stands when subjected to extreme weather events such as drought (Rusinamhodzi et al., 2012; Wortman et al., 2012). Mixtures may allow producers to take advantage of the complementary benefits provided by cover crops while mitigating potential risks (e.g., N immobilization during periods of high cash crop N demand) (Miyazawa et al., 2014; Smith et al., 2014; Teasdale and Abdul-Baki, 1998). For example, cereal rye provides fast ground cover and produces large amounts of biomass, protecting against erosion and building soil organic matter through the addition of C. The large C:N ratio of cereal rye, however, can lead to N immobilization after cover crop termination (Clark et al., 1994). Conversely, nodulating cover crops such as hairy vetch fix N that can offset the C produced by large C:N ratio species and contribute to cash crop N needs, but are slow to establish and provide ground cover. Combining a cover crop with a large C:N ratio such as cereal rye with one with a smaller C:N ratio such as oilseed radish may "balance" the C:N ratio, avoiding the difficulties inherent in too high a ratio (i.e. N immobilization) and too small a ratio (i.e. early N release from decomposing cover crop tissue and subsequent leaching) (Odhiambo and Bomke, 2001; Rosecrance et al., 2000). Clark et al. (1994) found that in cereal rye + hairy vetch bicultures, cereal rye scavenged residual soil N while hairy vetch fixed N, which increased corn grain yield an average of 14% compared with a no-cover crop control.

The theoretical benefits provided by mixtures are not always realized in field studies. Species selection, and planting rates and proportions dictate to what extent potential benefits are realized. In Nebraska, Wortman et al. (2012) found mustard species to be more competitive and

productive than legumes when grown in mixtures. In Ohio, Creamer et al. (1997) evaluated 13 multi-species mixtures and observed that some species were not suitable due to lack of winter hardiness or small biomass production. A dominant component species in a mixture can also lessen the benefits provided by the other cover crops. Brainard et al. (2011) found that soybeans grown in a mixture with sorghum-sudangrass produced less biomass, nodulated less, and fixed less N than soybeans grown in monoculture in both years of a study. Furthermore, the sorghumsudangrass monoculture treatment had a smaller seed cost than the mixture. Depending on the species and planting rates, seed mixtures can be more costly than monocultures (Table 3.1). Mixtures with a range of seed sizes may be difficult to plant, establish, and manage (Creamer et al., 1997). When sowed later in the fall (mid- to late September), hairy vetch planted in mixture with cereal rye produced less biomass than when planted in late August in Michigan (Hayden et al., 2015). Farmers may also have difficulty predicting the effect of a cover crop mixture on a cropping system. Hayden et al. (2014) did not find evidence of synergy between hairy vetch and cereal rye in mixture. The authors noted that altering the seeding proportions of the two species in mixture resulted in tradeoffs among the agroecosystem benefits provided by the cover crops. For example, larger proportions of hairy vetch lead to greater seed costs and less weed suppression but greater amounts of fixed N.

Despite farmer interest, there has been relatively little research on cover crop mixtures (Carlson and Stockwell, 2013). More research is needed to determine and which mixtures provide the best combination of benefits and the least likelihood of risk to a following cash crop. In this study, we chose to evaluate cover crop bicultures rather than polycultures because the planting and management of polycultures can be impractical (Creamer et al., 1997). Research also suggests that polycultures are not more beneficial to a cropping system than bicultures

(Miyazawa et al., 2014; Smith et al., 2014; Teasdale and Abdul-Baki, 1998). Due to a lack of information on bicultures in literature, the current study focuses upon bicultures of a Brassicaceae cover crop species (specifically, oilseed radish) combined with either a grass or legume. Annual ryegrass, cereal rye, oats, crimson clover, hairy vetch, and winter pea were chosen as complementary species to provide a range of characteristics such as large vs. small C:N ratio, winter-hardy vs. frost sensitive, and N-fixers vs. non-N fixers.

The objectives of this study were to: a) quantify biomass production of annual ryegrass, cereal rye, oats, crimson clover, hairy vetch, winter pea, and oilseed radish both individually and in bicultures, b) determine the effect of the cover crops on fall and spring weed biomass, and c) evaluate cover crop impact on corn grain yield in a conventionally tilled system with no applied fertilizer. We hypothesized that: a) bicultures will produce more biomass than monocultures, b) legume species will be less competitive in biculture with oilseed radish than grass species, c) cover crop treatments with the largest biomass production will be more effective at weed suppression than those with smaller biomass, d) the inclusion of oilseed radish in biculture with each legume will reduce the positive impact of the legume on corn yield when compared with the legume monocultures. To our knowledge this study is unique in that it investigated the performance of a variety of legumes and grasses included in biculture with oilseed radish.

MATERIALS AND METHODS

Site characteristics and field operations

A field experiment was conducted at the Michigan State University Agronomy Farm in Lansing, MI (42° 46' N, 84° 34' W) for three years. In 2011-2012 the soil was a Riddles-Hillsdale sandy loam (fine-loamy, mixed, mesic Typic Hapludalfs). In 2012-2013 the soils were a Riddles-Hillsdale sandy loam and a Metea loamy sand (loamy, mixed, mesic Arenic Hapludalfs). In 2013-2014 the soil was a Capac loam (fine-loamy, mixed, mesic Aeric Ochraqualfs). Field characteristics are listed in Table 3.2. The experiment had randomized complete block design with three (2012-2013) or four (2011-2012 and 2013-2014) replications. All plots were 3 x 12 m in size.

Cover crop treatments were planted using a Great Plains no-till drill or a John Deere planter in 19-cm wide rows into wheat stubble between mid-August and early-September (Table 3.2). Prior to planting, weeds were controlled with applications of glyphosate (N-(phosphonomethyl)glycine) at 1.22 kg ae ha⁻¹ tank-mixed with 2% w w⁻¹ ammonium sulfate (AMS). Cover crop treatments consisted of monocultures of: Groundhog[™] oilseed radish (Ampac Seed Co., Tangent, OR) (11 kg ha⁻¹), variety not stated (VNS) crimson clover (11 kg ha⁻¹) ¹), VNS hairy vetch (34 kg ha⁻¹), VNS winter pea (67 kg ha⁻¹), 'Bounty' annual ryegrass (44 kg ha⁻¹), 'Wheeler' cereal rye (126 kg ha⁻¹), and VNS oats (72 kg ha⁻¹). Biculture mixtures consisted of GroundhogTM oilseed radish (11 kg ha⁻¹) + VNS crimson clover (6 kg ha⁻¹), GroundhogTM oilseed radish (11 kg ha⁻¹) + VNS hairy vetch (17 kg ha⁻¹), GroundhogTM oilseed radish (11 kg ha⁻¹) + VNS winter pea (34 kg ha⁻¹), GroundhogTM oilseed radish (11 kg ha⁻¹) + 'Bounty' annual ryegrass (22 kg ha⁻¹), Groundhog[™] oilseed radish (11 kg ha⁻¹) + 'Wheeler' cereal rye (63 kg ha⁻¹) ¹), and GroundhogTM oilseed radish (11 kg ha⁻¹) + VNS oats (36 kg ha⁻¹). Additionally, a nocover crop (weedy) control and a bare ground control that was hand-weeded once in fall 2011 and sprayed with glyphosate (N-(phosphonomethyl)glycine) once per fall in 2012 and 2013 were included. Seeding rates were based on Michigan State University Extension and seed distributor recommendations as outlined in the Midwest Cover Crops Council online decision tool species fact sheets (MCCC, 2015). To ensure nodulation for N fixation, *Rhizobium leguminosarum* bv. *trifolii* inoculant was mixed by hand with the crimson clover seed prior to planting, while *Rhizobium leguminosarum* bv. *viciae* inoculant was applied to the hairy vetch and winter pea seed.

Cover crops which survived the winter were terminated the following spring with glyphosate (N-(phosphonomethyl)glycine) + 2,4-D (2,4-dichlorophenoxyacetic acid) 8-10 d prior to corn planting. The fields were prepared for planting using a chisel plow and a minimum of two passes of a soil finisher. The 102-day field corn cultivar 'DeKalb 52-59' (Monsanto, St. Louis, MO) was planted at a rate of 74,100 seeds per ha⁻¹ using a four-row planter with a row width of 76 cm. Weeds were controlled as necessary with glyphosate at corn stages V6 and V10. In order to avoid concealing the effects of cover crops, no irrigation or fertilizer was applied to either the cover crops or corn. Yearly weather data for Lansing, MI were obtained from the Michigan State University Enviro-weather website for the MSU-Hort station (MSUEW, 2015), with the exception of winter precipitation data which were collected from the Lansing, MI weather station via the NOAA website (NOAA, 2015). Temperature and precipitation 30-yr monthly means (1981-2010) for Lansing, MI were obtained from the National Oceanic and Atmospheric Administration (NOAA) website (NOAA, 2015). Weather conditions over the course of the experiment are presented in Figure 3.1.

Cover crop, weed, and corn data collection

Cover crop, weed, and volunteer wheat aboveground biomass were harvested from two randomly placed 0.25 m² quadrats per plot, with the exception of fall 2011 when one subsample

per plot was collected. Biomass was harvested in the fall prior to winter kill and in the spring prior to cover crop termination (Table 3.2). Oilseed radish roots were also pulled by hand from each quadrat in the fall. Tissue samples were separated into cover crop and weed material, dried at 70° C for 5-10 d, and weighed. Corn stand was assessed 2-4 wk after planting by counting the number of plants in 5.3 meter-lengths of each of two rows. Corn stand was again assessed prior to corn harvest. A plot combine was used to harvest the middle two rows of each plot. Corn yield was adjusted to 15.5% moisture.

Biomass relative yield calculation

Biomass relative yield (RY) was calculated using fall cover crop biomass. The RY index buffers the influence of population density on biomass and thus helps account for different planting rates between treatments (Bedoussac and Justes, 2011; Williams and McCarthy, 2001), allowing for a more distinct evaluation. Relative yield is calculated as

$$RY_A = Y_{AB}/(P_A * Y_A)$$

where RY_A was the relative biomass yield of species A, Y_{AB} was the dry biomass yield of species A when grown with species B, P_A was the proportional seeding rate of species A that is seeded in the biculture, and Y_A was the dry biomass yield of species A when grown in monoculture [adapted from Bedoussac and Justes (2011), who used the notation of Fowler (1982)]. In this study, the oilseed radish seeding rate was held constant across both monoculture and biculture treatments; P_A was thus 1. The complementary species to oilseed radish in each biculture treatment was drilled at half the monoculture planting rate; P_B for these species was thus 0.5.

Statistical analysis

All data sets were analyzed with SAS Enterprise Guide v. 6.1 (SAS Institute Inc., Cary, NC). Analysis of variance was completed using PROC MIXED, the LSMEANS and PDIFF statements, and the Tukey-Kramer adjustment for all pairwise comparisons ($\alpha \leq 0.05$). Data were tested for normality, equal variances, and treatment*year interactions prior to ANOVA and mean separation. Replication was deemed a random effect. Mean separation was conducted with the PDMIX 800 macro (Saxton, 1998). Because some species failed to survive the winter in some years, some species did not establish well, and some species did not overwinter well, maximum annual aboveground biomass was calculated using a combination of fall and spring data. For example, in the annual ryegrass treatment fall biomass values were deemed the maximum annual biomass if spring biomass production was smaller than fall due to poor winter survival. If spring values were larger than fall, then the spring biomass was used. For bicultures, oilseed radish biomass was added to the greater of the fall and spring biomass of the complementary species in each mixture. Corn yield data were square-root transformed to improve normality.

RESULTS AND DISCUSSION

Cover crop biomass production

Fall cover crop biomass. There was a significant treatment*year interaction (P < 0.0001), so fall cover crop total aboveground dry biomass is presented by year (Figure 3.2). The oilseed radish root: shoot ratio treatment*year interaction was not significant (P = 0.21), so data were combined across years (Table F.2). Oats produced the most biomass in the fall of 2011 and 2012, while oilseed radish produced the most biomass in 2013 (Figure 3.2). Crimson clover produced the least amount of biomass in all three years. Oats produced approximately 2.1 and 9.1 times more biomass than crimson clover in 2011 and 2012, respectively. Oilseed radish produced 11 times more biomass than crimson clover in 2013. Treatments that included oats consistently produced

some of the largest biomass across years. The large fall aboveground biomass production may have been the result of oats partitioning less biomass to the roots than annual ryegrass and cereal rye, which are more winter-hardy. The small fall biomass production of crimson clover was indicative of poor stand as a result of establishment issues. Crimson clover had the smallest-sized seed among the cover crops tested, which caused difficulties despite the use of a planter set up to handle small seeds. In addition, the crimson clover monoculture was planted at 11 kg ha⁻¹ in this study, which is a small seeding rate even when using a drill (MCCC, 2015). November 2011 was 2.8 and 3.9 °C warmer than November 2012 and 2013, respectively (Figure 3.1). Biomass was collected on 14 November 2011 (Table 3.2). The larger amount of crimson clover biomass collected in 2011 may have been due to the warmer temperatures. In September 2013 precipitation totaled 23% of the 30-yr average for the month (Figure 3.1). Dry conditions immediately following the 2 September cover crop planting (Table 3.2) may have delayed germination slowing cover crop growth and contributing to the decreased cover crop biomass observed in 2013 (Figure 3.2). In 2013, biomass production was smaller than in the other two years at least partly because biomass was collected 10-11 weeks after planting (WAP) in 2011 and 2012, but only eight WAP in 2013. While differences were observed in cover crop aboveground biomass, no differences were detected in oilseed radish root: shoot ratios (Appendix Table 3.7). Differences may not have been detected because they did not exist (i.e. oilseed radish was highly competitive in biculture), or because root sampling protocols were inadequate.

Variability in cover crop biomass across years and studies is common in the literature. The fall biomass production found in this study is similar to that found in other studies for annual ryegrass (Dapaah and Vyn, 1998; Francis et al., 1998; Isse et al., 1999; Thorup-Kristensen, 2001;

Vyn et al., 1999), cereal rye (Axelsen and Kristensen, 2000; Kaspar et al., 2001; Ranells and Wagger, 1997; Thorup-Kristensen, 2001; Vyn et al., 2000), and oats (Brennan and Smith, 2009; Francis et al., 1998; Kaspar et al., 2001; Stivers-Young, 1998; Thorup-Kristensen, 2001; Vyn et al., 2000). Fall legume biomass production values are less commonly reported in the literature, but crimson clover has been noted to produce 90-1040 kg ha⁻¹ (Axelsen and Kristensen, 2000; Isse et al., 1999; Ranells and Wagger, 1996), while Axelsen and Kristensen (2000) found hairy vetch to produce 1910 kg ha⁻¹ of dry aboveground biomass. Oilseed radish biomass ranges from 176–4840 kg ha⁻¹ (Axelsen and Kristensen, 2000; Dapaah and Vyn, 1998; Dean and Weil, 2009; Isse et al., 1999; Stivers-Young, 1998; Thorup-Kristensen, 2001; Vyn et al., 2000; Wortman et al., 2012).

Within each year, the cover crop biculture treatments generally produced similar amounts of fall biomass to each other (Figure 3.2). In all mixtures, oilseed radish biomass exceeded that of the second species in the biculture, accounting for 54-99% of total above ground biomass. Across the three years, oilseed radish dominated the oilseed radish + legume bicultures to a greater extent than the oilseed radish + grass bicultures (Figure 3.2).

While there has been no comprehensive evaluation of oilseed radish in biculture with grass and legume species, there are some reports of oilseed radish biculture performance in the literature. Cavadini (2013) found that fall-planted oilseed radish, oilseed radish + cereal rye, and oilseed radish + oats generally produced the same amount of dry aboveground biomass in the fall in each site-year. Oilseed radish was planted at a smaller rate in biculture than monoculture in that study. In one site-year, the oilseed radish in the oilseed radish + cereal rye biculture composed 70% of the dry aboveground production of that mixture even though both crops were planted at half the rate in biculture that they were in monoculture (White and Weil, 2010). In a

study in which seeding rates were the same in biculture for each species as in monoculture, Moller and Reents (2009) found that at one site there were no significant differences in cover crop dry aboveground biomass production of oilseed radish + pea (*Pisum sativum* L.), oilseed radish + common vetch (*Vicia sativa* L.), and oilseed radish, pea, and common vetch monocultures while at the other site, oilseed radish + pea yielded more biomass than the oilseed radish monoculture.

Spring cover crop biomass. Total spring cover crop biomass could not be combined across years due to a treatment*year interaction (P < 0.0001) caused by varying weather conditions across the three years of this study that impacted cover crop winter survival. February and March 2013 were 1.3 and 2.6 °C colder than the 30-yr averages for those months, respectively (Figure 3.1). January, February, and March 2014 were 5.4, 3.0, and 5.6 °C colder than the corresponding 30vr monthly averages. The winter killed cover crops of oilseed radish and oats did not contribute to cover crop biomass in the spring. In spring 2012, crimson clover, hairy vetch, and annual ryegrass produced 1.5-3.9 times more biomass than the bicultures of oilseed radish + crimson clover, + hairy vetch, + winter pea, + cereal rye, + annual ryegrass, and + oats (Figure 3.3). With the exception of cereal rye and oilseed radish + cereal rye, all of the other monoculture treatments produced 1.9-3.9 times more biomass than the corresponding biculture treatments. Monoculture treatments produced more biomass than the bicultures partly because the seeding rate in biculture was half that in monoculture and partly because the oilseed radish did not overwinter. In spring 2013, cereal rye and oilseed radish + cereal rye respectively produced a minimum of 855 and 703 kg ha⁻¹ more biomass than all other non-oat, non-oilseed radish + oat, and non-radish monoculture treatments, except annual ryegrass. Unlike in 2012, in 2013 biomass production was the same within each monoculture/biculture treatment pair. In 2014, cereal rye

produced at least 1.3 times more biomass than all of the other cover crops that survived the winter. Hairy vetch, annual ryegrass, and cereal rye produced more biomass than the corresponding biculture treatments.

The weather impacted spring biomass production. Winter pea was not difficult to establish (data not shown) but did not survive the winter in two of the three years. March 2013 was 2.6°C colder than the 30-yr average (Figure 3.1). November 2013 – March of 2014 was an average of 4.3 °C colder than the 30-yr average. The cold winter conditions in the second and third year of this study may help to explain why winter pea overwintered the first year of the study but not the other two years (Figure 3.3). In addition, in 2012-2013 and 2013-2014 winter pea was observed to have reached the reproductive stage prior to frost (data not shown), which would have further decreased the likelihood of winter survival. Weather clearly affected the spring biomass of the other cover crops, as well. For example, hairy vetch and annual ryegrass produced up to an order of magnitude more biomass in spring 2012 compared with 2013 and 2014 (Figure 3.3), likely because November-March was an average of 5.5 °C warmer in 2011-2012 than winter of other two years.

Our results show several cases where cover crops appear to have been suppressed by oilseed radish, such as crimson clover vs. oilseed radish + crimson clover in 2012, and crimson clover and hairy vetch vs. oilseed radish + crimson clover and oilseed radish + hairy vetch in 2014. Moreover, it cannot be determined from our study whether mixtures per se or differences in seeding rates account for differences in the biomass of the complementary species in mixture. For example, had cereal rye been sown in monoculture at half of the rate used in this study, reductions in intraspecific competition may have resulted in similar biomass as those observed at

the full monoculture rate, as has been documented in the literature (Clark et al., 1994; Hayden et al., 2014).

As with fall cover crop biomass, there is a wide range of values reported for spring dry aboveground biomass production of fall-planted cover crops. Spring biomass values found in this study were in agreement with those found in the literature, except in the cases of crimson clover and winter pea. Values reported in the literature for crimson clover and winter pea range from 968-8000 kg ha⁻¹ and 1800-7600 kg ha⁻¹, respectively, and are greater than those found in this study (Bauer et al., 1993; Daniel et al., 1999; Decker et al., 1994; Isse et al., 1999; Norsworthy et al., 2010; Odhiambo and Bomke, 2001; Parr et al., 2011; Ranells and Wagger, 1996; Ranells and Wagger, 1997; Singogo et al., 1996; Yenish et al., 1996). As previously discussed, crimson clover did not establish well in two of the three years of the study, while winter pea failed to survive the winter in two of the three years of the study. To improve the probability of winter pea survival over the winter, it has been suggested that the seed be planted relatively deep (5 cm) and not too early in the fall.

Maximum annual biomass. Fall and spring aboveground dry biomass data were used to calculate the total maximum likely amount of biomass contributed by each cover crop treatment from fall through spring, removing the confounding effects caused by some species that failed to establish or overwinter well. Due to a treatment*year interaction (p < 0.0001), data were not combined across year. In 2011-2012, no differences between treatments were detected (Table 3.3). In 2012-2013, oilseed radish + crimson clover produced 3.2 times more biomass than crimson clover and oilseed radish + hairy vetch yielded 5.7 times more biomass than hairy vetch. In 2013-2014, oilseed radish + crimson clover, oilseed radish + winter pea, and oilseed radish + annual ryegrass

respectively produced 4.6, 1.9, and 1.8 times more biomass than crimson clover, winter pea, and annual ryegrass.

The biomass results in this study are of interest because the presence and quantity of cover crop biomass is one of the mechanisms through which cover crops influence production systems. In some cases, this influence comes in the form of harm, such as when cereal rye biomass with a large C:N ratio immobilizes soil N (Crandall et al., 2005; Odhiambko and Bomke, 2001). In other cases, cover crop biomass can convey benefits to productions systems (Kuo et al., 1997). Cover crops decrease erosion by buffering the ground from the impact of raindrops and slowing the flow of water over the ground (Dabney et al., 2001). The rapid production of large amounts of biomass helps cover crops suppress weeds (O'Reilly et al., 2011). Non-legume cover crops scavenge residual soil nutrients, while legume cover crops fix atmospheric N (Dabney et al., 2001). In order to estimate the impact of cover crops on nutrient cycling, knowledge of cover crop potential biomass production is necessary.

Cover crop impact on weeds

Fall weed biomass. Due to a treatment*year interaction (P < 0.0001), total fall weed dry aboveground biomass data could not be combined. For the purposes of this discussion, "total weeds" denotes weed plus volunteer wheat biomass. Total weed densities varied between sites. Total weed densities were so small in fall 2011 that no weed data were collected. Total weed biomass was over an order of magnitude larger in fall 2012 than in 2013 (Figure 3.2 and Table 3.4). In both 2012 and 2013, the predominant weed species were common chickweed (*Stellaria media* (L.) Vill.) and *Lamium* spp. (data not shown). In fall 2012, there was 155-571 kg ha⁻¹ weed biomass in treatments with the least amount of weed biomass, less than the 1268-1586 kg ha⁻¹ biomass in the weedy control, crimson clover, hairy vetch, and winter pea treatments. In

2013, there was an order of magnitude less total weed biomass in the bicultures of oilseed radish + crimson clover, + hairy vetch, + winter pea, + annual ryegrass, and + cereal rye, and the annual ryegrass and cereal rye monocultures than in the weedy control. When bicultures were compared with monocultures, total weed biomass was smaller in the oilseed radish + crimson clover, oilseed radish + hairy vetch, and oilseed radish + winter pea treatments than in the corresponding legume monocultures and weedy control in fall 2012. The oilseed radish in these biculture treatments appeared to contribute to weed suppression. No differences in total weed biomass were detected among the annual ryegrass, cereal rye, oats, oilseed radish + annual ryegrass, oilseed radish + cereal rye, and oilseed radish + oats treatments. All were equally effective at suppressing weeds when compared with the weedy control. Irrespective of oilseed radish presence, annual ryegrass and cereal rye suppressed weed biomass. In fall 2013 the biculture treatments were no more effective at suppressing weeds than the corresponding monocultures, with the exception of oilseed radish + hairy vetch and hairy vetch.

Cover crops can suppress weeds (Conklin et al., 2002; Creamer et al., 1996; Stivers-Young, 1998; Hayden et al. 2012). This ability is of interest because winter annual weeds can harbor pests and diseases of cash crops (Groves et al., 2001; Venkatesh et al., 2009), they can grow too large to be easily controlled, and if they are left uncontrolled they may add to the weed seedbank and become problematic in future years of the rotation. In particular, the ability of cereal rye (Akemo et al., 2000; O'Reilly et al., 2011) and oilseed radish (Lawley et al., 2011; O'Reilly et al., 2011; Stivers-Young, 1998) to suppress weeds has been documented in the literature. Since large cover crop biomass production has been linked to effective weed suppression (Teasdale, 1996), it is not surprising that the treatments in this study which provided the most fall weed suppression also had some of the largest cover crop biomass (Figure 3.2).

There an average of at least 453 kg ha⁻¹ more total weed biomass across years in the crimson clover, hairy vetch, and winter pea treatments than in the annual ryegrass, cereal rye, oats, and oilseed radish treatments. The inclusion of oilseed radish in biculture with the legumes greatly increased fall weed suppression in 2012, probably because of the large biomass contributed by the oilseed radish in the biculture treatments (Figure 3.2). Other studies have also found that cover crop mixtures controlled weeds better than legume monocultures. Bicultures of cereal rye + crimson clover and cereal rye + hairy vetch were more effective at suppressing weed emergence and biomass production than crimson clover and hairy vetch monocultures (Teasdale and Abdul-Baki, 1998). Field pea (*Pisum sativum* L.) was less effective at weed suppression than a barley + pea biculture evaluated by Hauggard-Nielsen et al. (2001). Akemo et al. (2000) likewise found cereal rye and cereal rye + field pea to more effective at weed suppression than field pea grown in monoculture.

Spring weed biomass. Due to a treatment*year interaction (P < 0.0001), total spring dry aboveground total weed biomass data could not be combined across years. Spring total weed biomass did not vary in magnitude across years to the extent that it did in the fall (Figure 3.3 and Table 3.4). In spring 2012 the dominant weed species were wheat, common chickweed and common speedwell (*Veronica arvensis* L.) (data not shown). Total weed biomass was composed of 95% wheat (Figure 3.3). All of the cover crop treatments had at least 643 kg ha⁻¹ less total weed biomass than the weedy control; all treatments had significantly less total weed biomass than the weedy control (Table 3.4). In spring 2013, wild violet (*Viola* spp.), shepherd's-purse [*Capsella bursa-pastoris* (L.) Medik.], dandelion (*Taraxacum officinale* F.H. Wigg.), and *Lamium* spp. were the major weed species. No differences in total weed biomass were detected. In spring 2014 common chickweed, *Lamium* spp., and common lambsquarters (*Chenopodium*)

album L.) dominated in the field. In spring 2014, there was 596 kg ha⁻¹ biomass in the weedy control, more than the 0-179 kg ha⁻¹ biomass in all other treatments except oilseed radish (214 kg ha⁻¹), crimson clover (565 kg ha⁻¹), and winter pea (663 kg ha⁻¹). Differences in weed control across season and years were partly the result of varying cover crop biomass. Since different weeds react differently to cover crop residue (Teasdale, 1996), differences between years could also have been due to the varying weed composition in each field-year. The presence of volunteer wheat may also have played a role. Wheat itself is frequently used as a fall cover crop and is known to be a vigorous and hardy species.

Biomass relative yields (RY)

Fall biomass RYs were calculated for oilseed radish, each complementary species ("species B"), and the total biomass RY for each biculture treatment. There was no treatment*year interaction for any permutation of the biomass RYs (P = 0.75, 0.32, and 0.95, respectively). Since there were no treatment*year interactions, we averaged the oilseed radish and species B biomass RYs across the three years of this study and plotted them on the graph created by Williams and McCarthy (2001) (Figure 3.4). Oilseed radish biomass RY is on the y-axis and species B biomass RY is on the x-axis. All of the treatment points fell into the area bounded by the vertical line x = 0, the horizontal line y = 1, and the diagonal line biomass RY_{oilseed radish} = biomass RY_{species B}. According to Williams and McCarthy (2001), this positioning on the graph indicates that oilseed radish likely competed/interfered with the complementary species in each mixture. While the coordinates for the treatments all fell into the same space on the graph, those of oilseed radish + cereal rye and oilseed radish + oats fell closest to the line biomass RY_{oilseed radish} = biomass RY_{species B}. In these two treatments, the effects of the cover crops on each other were thus closest to "neutral". Notably, the one legume cover crop which
overwintered in all three years (hairy vetch) had coordinates nearest the horizontal line above which oilseed radish would be said to suppress the complementary species (Figure 3.4). It is harder to draw conclusions about crimson clover and winter pea as those legumes were clearly heavily influenced by weather conditions and establishment issues as previously discussed.

Another potentially confounding factor in this study was weeds, which were not controlled in treatment plots after cover crop planting. The weeds biomass data in Figure 3.2, however, indicate that oilseed radish was likely competing/interfering with weeds as well as the complementary species. Fall weed biomass production was smaller in the legume and oats bicultures than in the corresponding monoculture treatments. Since weed biomass was greater in monoculture treatments, the weeds were likely competing with each complementary cover crop and thus potentially decreasing monoculture treatment biomass. Since the RY equation calls for this biomass number to be multiplied by the biculture planting rate (0.5) and then used as the denominator to determine biculture species RY, our biomass RY calculations were likely underestimates.

Cover crop impact on corn

Corn grain yield. Corn grain yield data were not combined. In 2012, corn in the annual ryegrass treatment produced 27-48% less grain than that in all other treatments except cereal rye and oilseed radish + cereal rye (Table 3.5). In 2013 there were no differences in corn yield. In 2014, corn in the cereal rye treatment yielded less than that in all other treatments except oilseed radish + cereal rye and winter pea. A number of factors may have contributed to our inability to detect differences including variability in the data, soil type and fertility differences between field sites, and varying weather each year. The most differences were detected in 2012, which was a drought year (Figure 3.1). Cash crop yield possibly benefits most from cover crop influences in

years with adverse weather conditions or sites with adverse soil conditions. In 2014, the field tested at 3.8% soil organic matter. Soil N reserves may have obscured differences between controls and cover crop treatments. Weeds in the weedy control could have acted similarly to cover crops, also obscuring differences.

The results of our study with regard to the neutral to positive impact of legume cover crop monocultures and bicultures on corn yield are generally in agreement with the literature. Our study as not designed to separate cover crop effects on N from the cover crop rotation effect, both of which could help explain our results. There have been other studies involving cover crops in which there was at least one no-fertilizer treatment. Most involved the use of legume cover crops. Legume cover crops have been found to increase crop yield. This effect is often attributed primarily to the N contributed to the system by the legumes (Kuo and Jellum, 2002; Torbert et al., 1996). In a meta-analysis, Miguez and Bollero (2005) found winter legumes increased corn yield 37% in the absence of applied N fertilizer as compared to a bare ground control, while cover crop bicultures increased corn yield 21%. Smith et al. (2008) attributed the beneficial effect of a diverse crop rotation on corn yield in to the N contributed by legume cover crops; corn yield in the most diverse rotation was the same as the county average, despite the lack of conventional fertilizer. Contrary to our results, some studies have found grass cover crops to have no impact, or a positive impact, on corn yield. Andraski and Bundy (2005) found that corn yield benefited from the use of an oat or cereal rye cover crop as compared to fallow, but attributed the impact to rotation effects rather than N contribution from the cover crops. In the Miguez and Bollero (2005) meta-analysis, grass winter cover crops had no impact on corn yield regardless of whether N fertilizer was applied. Kuo and Jellum (2002) and Isse (1999)

determined that annual ryegrass had no impact on corn yield. Kuo and Jellum (2002), Crandall et al. (2005), Yenish et al. (1996), and Vyn et al. (2000) found the same of cereal rye.

As in our study, other studies have also found annual ryegrass or cereal rye to negatively impact corn yield in at least some years (Hively and Cox, 2001; Johnson et al., 1998). The negative effect of the grass cover crop treatments on corn in some years of our study could be due to a number of factors, including corn stands up to 15% smaller in the annual ryegrass and cereal rye treatments than in the bare ground control (Table F.4). Kaspar et al. (2007) postulated that a combination of large cereal rye biomass production and insufficient time between cover crop termination and corn planting lead to the decreased yield seen in one year of that study. In this study in 2014, there likewise may not have been sufficient time between cover crop termination and cash crop planting (Table 3.2) given the large amount of biomass produced by the cereal rye treatment (Figure 3.2-3.3). Other authors have noted the difficulty of terminating annual ryegrass (Creamer et al., 1996; Madden et al., 2004). We had difficulty creating a uniform seedbed during tillage in annual ryegrass plots, which could have interfered with corn planting or germination and thus affected stand (Table 3.5). In some years annual ryegrass or cereal rye may have immobilized soil N to the detriment of the corn. In the unfertilized treatment of a study by Kuo and Jellum (2000), corn N uptake was significantly smaller in annual ryegrass and cereal rye treatments than in a hairy vetch treatment, though not generally different than the weedy control. Other researchers have also suggested N immobilization as a mechanism by which cereal rye (Miguez and Bollero, 2006; Vaughan and Evanylo, 1999) and annual ryegrass (Vyn et al., 1999) decreased corn yields. Finally, in the two years of this study in which there were corn yield differences, there were also differences in corn tasseling in late July – early August (Table F.6). Tasseling in the annual ryegrass and cereal rye treatments was delayed. This may have

resulted in poor pollination, or it could have decreased the amount of time available for corn grain fill. All of these factors could have contributed to the smaller yields seen in the annual ryegrass and cereal rye treatments.

CONCLUSIONS

Different cover crop species have different benefit and risk profiles. Benefits can be maximized, and risks minimized, through the careful selection and management of cover crops. Cover crop selection depends upon cropping system needs (e.g., erosion control) and constraints (e.g., limited money and time). In this study, oilseed radish dominated biculture fall biomass production, particularly in the cases of oilseed radish + crimson clover, oilseed radish + hairy vetch, and oilseed radish + winter pea. Biomass relative yield values suggest that oilseed radish competed with the complementary species in the bicultures, to a greater extent with the legumes than the grasses. Thus, the observed small biomass production of the complementary species in biculture was likely not solely the result of the smaller seeding rate in biculture compared to monoculture. Fall competition with oilseed radish, combined with the smaller seeding rates in biculture, explains the small spring biomass of the oilseed radish + annual ryegrass and oilseed radish + hairy vetch treatments. At the rates planted in this study, seed cost for oilseed radish + crimson clover, + annual ryegrass, + cereal rye, or + oats is 16-84% greater than the seed cost of the complementary species grown in monoculture, while oilseed radish + hairy vetch or winter pea biculture seed is only 8-26% less expensive (Table 3.1). Based on cover crop biomass production and seed cost alone, the monocultures were more cost-effective than the bicultures in this study. Oats, with its large fall biomass and lack of winter survival, is an excellent lowmanagement choice. Alternate considerations aside from cost factor in, however. In the case of crimson clover and other cover crops where stand establishment is a concern (e.g., due to planting difficulty or low viable seed), the inclusion of oilseed radish could bolster biomass

production and suppress weeds. Contrary to our hypothesis, oilseed radish + legume bicultures did not have less of a positive impact on corn yield than legume monocultures. Only in one year in one case (oilseed radish + annual ryegrass biculture vs. the annual ryegrass monoculture) was our hypothesis supported that oilseed radish + grass bicultures would have less of a negative effect on corn yield than grass monocultures. Nonetheless, corn yield depression was observed for treatments including annual ryegrass and cereal rye. Producers who grow annual ryegrass or cereal rye should be aware that timely spring cover crop termination, adequate corn seed bed preparation, and N fertilizer application are key to avoiding decreased corn yield. The other cover crops in this study are not without risks, either. Though we did not quantify the risk of these cover crops becoming weeds, we observed oilseed radish and hairy vetch germinating from hard seed during the corn phases of this study. Hairy vetch is of particular concern because it is a contaminant of small grain seed, and should be used cautiously, if at all, in rotations with wheat or barley cash crops.

There are several avenues of research leading from this work. It would be interesting to repeat this experiment as a multifactorial plot design with the added factors of fall fertilizer application to the cover crops and weed control vs. no weed control in the cover crop plots. Cover crop effects on soil properties like bulk density could be measured. A quantification of the ecosystem services provided would be timely and relevant. A study using a substitutive seeding rate design could be performed to optimize the proportion of oilseed radish to other species in biculture mixtures.

APPENDICES

APPENDIX A CHAPTER 3 TABLES AND FIGURES

Tuble 5.1. Estimated Cos	Table 5.1. Estimated cost of cover crop seed in an experiment in Lansing, with							
Cover crop treatment	\$ kg⁻¹†	\$ ha ⁻¹ of	\$ ha ⁻¹ of biculture					
		monoculture‡§						
Oilseed radish	3.53-5.51	50	-					
Crimson clover	2.97-3.75	37	68					
Hairy vetch	4.41-5.51	169	134					
Winter pea	1.32-2.20	118	109					
Annual ryegrass	1.31-1.59	64	82					
Cereal rye	0.55-0.66	76	88					
Oats	0.55-0.73	46	73					

Table 3.1 Estimated cost of cover cron seed in an experiment in Lansing MI

* Seed costs were obtained from suppliers in the mid-Michigan region.
* Estimated seed cost ha⁻¹ based on seeding rates used in this study and average seed price, rounded to the nearest whole dollar.

§ Monoculture: pure stand planting; biculture: grown in combination with oilseed radish

	2011-2012	2012-2013	2013-2014
Field characteristics			
Slope (%)	2-6	2-6	0-3
Soil PH	7.1	5.4	5.8
Soil organic matter (%)	3.2	2.5	3.8
Field activity dates			
Cover crop planting	30 Aug. 2011	13 Aug. 2012	2 Sept. 2013
Fall biomass harvest	14 Nov. 2011	5 Nov. 2012	25 Oct. 2013
Spring biomass harvest	17 Apr. 2012	8 May 2013	14 May 2014
Cover crop termination	27 Apr. 2012	8 May 2013	19 May 2014
Tillage	21 May 2012	20 May 2013	28 May 2014
Corn planting	22 May 2012	20 May 2013	28 May 2014
Spring corn stand	7 Jun. 2012	19 Jun. 2013	29 Jun. 2014
Fall corn stand	30 Oct. 2012	28 Oct. 2013	24 Oct. 2014
Corn harvest	17 Nov. 2012	24 Nov. 2013	14 Nov. 2014

Table 3.2. Field characteristics, field operation and data collection dates for experiments conducted in Lansing, MI.

	2011-2012	2012-2013	2013-2014
Cover crop treatments	-	kg ha ⁻¹	
Oilseed radish	2780	2290 bc	2806 bc
Crimson clover	4498	439 d	523 e
Oilseed radish + crimson clover	3143	2320 bc	2401 bcd
Hairy vetch	2428	910 cd	3147 b
Oilseed radish + hairy vetch	3335	2923 ab	3344 b
Winter pea	2587	1871 bcd	1319 de
Oilseed radish + winter pea	2081	2303 bc	2562 bc
Annual ryegrass	3987	2002 bcd	1676 cde
Oilseed radish + annual ryegrass	3584	2180 bc	2950 b
Cereal rye	2982	2569 abc	4843 a
Oilseed radish + cereal rye	2805	3524 ab	5765 a
Oats	3338	4209 a	2494 bcd
Oilseed radish + oats	2350	3232 ab	2748 bc
P value	0.17	< 0.000	< 0.0001

Table 3.3. Maximum annual dry aboveground biomass produced by cover crop monocultures and bicultures in Lansing, MI (2012-2014).

† Means in the same column followed by the same letter are not significantly different ($\alpha \leq$

	Spring 2012	Fall 2012	Spring 2013	Fall 2013	Spring 2014
Cover crop treatments			kg ha ⁻¹		
Weedy control	1035 a	1586 a	659	105 a	596 ab
Oilseed radish	392 b	155 c	488	42 abc	214 bcd
Crimson clover	170 bcd	1333 a	451	78 ab	565 abc
Oilseed radish + crimson clover	182 bcd	450 c	697	15 bc	136 d
Hairy vetch	21 d	1375 a	941	78 ab	179 cd
Oilseed radish + hairy vetch	112 bcd	376 c	351	7 c	87 d
Winter pea	252 bcd	1268 ab	935	72 abc	663 a
Oilseed radish + winter pea	141 bcd	155 c	245	9 bc	156 d
Annual ryegrass	1 d	469 c	557	29 bc	43 d
Oilseed radish + annual ryegrass	35 cd	253 с	203	4 c	115 d
Cereal rye	4 d	237 с	481	8 bc	0 d
Oilseed radish + cereal rye	15 d	42 c	530	2 c	174 cd
Oats	179 bcd	571 bc	523	5 c	161 d
Oilseed radish + oats	325 bc	101 c	204	47 abc	89 d
P value	< 0.0001	< 0.0001	0.20	< 0.0001	< 0.0001

Table 3.4. Fall and spring total dry aboveground weed biomass in Lansing, MI (2012-2014).

† Means in the same column followed by the same letter are not significantly different ($\alpha \le 0.05$).

	Stand			Grain yield		
	2012	2013	2014	2012	2013	2014
Cover crop treatments		plants 10 m ⁻¹ -			kg ha ⁻¹	
Oilseed radish	57 ab‡	55 ab	52	8596 abc‡	7714	12030 a
Crimson clover	56 abc	53 abc	53	8577 abc	7242	11424 a
Oilseed radish + crimson clover	60 a	53 abc	53	8223 abc	7279	11273 a
Hairy vetch	58 a	53 abc	52	9602 ab	7526	11097 a
Oilseed radish + hairy vetch	60 a	52 abc	53	8769 abc	7352	11100 a
Winter pea	60 a	52 abc	53	9834 a	7285	10583 ab
Oilseed radish + winter pea	59 a	57 a	53	8677 abc	7989	10975 a
Annual ryegrass	49 c	53 abc	52	5091 e	6395	11330a
Oilseed radish + annual ryegrass	56 abc	55 ab	52	6968 cd	6772	11773 a
Cereal rye	52 bc	55 ab	53	5866 de	6622	8095 b
Oilseed radish + cereal rye	60 a	52 bc	51	6722 cde	6431	9703 ab
Oats	59 a	53 abc	53	7422 bcd	6796	11931 a
Oilseed radish + oats	60 a	54 abc	51	8050 abc	7131	11879a
Bare ground control	57 ab	52 abc	53	7315 abcd	6953	11587 a
Weedy control	56 abc	50 c	53	7712 cd	6790	10714a
P value	< 0.0001	0.0007	0.28	< 0.0001	0.26	0.0003

Table 3.5. Mean corn stand[†] and grain yield from experiments conducted in Lansing, MI (2012-2014).

† Data presented are the average of the combined spring and fall corn stand counts for each year. ‡ Means in the same column followed by the same letter are not significantly different ($\alpha \le 0.05$). Means presented were backtransformed from the square root.



Figure 3.1. Monthly and 30-yr mean air temperature and precipitation for Lansing, MI 2011 - 2014. Precipitation is represented on the left y-axis and by bars. Temperature is represented on the right y-axis and by lines.



Figure 3.2. Mean (\pm SE) dry aboveground fall biomass of legume and grass cover crops in monoculture and in biculture with oilseed radish in Lansing, MI in 2011-2013.



Figure 3.3. Mean (\pm SE) dry aboveground spring biomass of legume and grass cover crops in monoculture and in biculture with oilseed radish, weeds, and volunteer wheat in Lansing, MI in 2012-2014.



Figure 3.4. Fall relative yields of oilseed radish and complementary species (species B) in each cover crop biculture treatment. Bars represent one SE from the mean. To the left of the diagonal line $RY_{oilseed radish}=RY_{species B}$, oilseed radish had the competitive advantage over species B while to the right of the diagonal line, the reverse was true. In the upper right quadrant, the species interaction was mutually beneficial. In the lower right quadrant, species B suppressed oilseed radish growth. The lower left quadrant indicates competition or interference between the two species. In the upper left quadrant, oilseed radish suppressed the growth of species B. Figure adapted from Williams and McCarthy (2001).

APPENDIX B

CHAPTER 3 SOIL, WEED, AND CORN SUPPLEMENTARY DATA COLLECTION METHODS

Soil sampling

Pre-side dress nitrate-N testing (PSNT) was performed at corn stage V6-V8 by collecting five soil cores per plot with a soil probe in an H-pattern to a depth of 15-20 cm. Soil samples were aggregated by treatment and ground to pass through a 2 mm sieve. In the 2012 and 2014, the samples were sent to the Michigan State University Soil and Plant Nutrient Laboratory for PSNT. In 2013 samples were extracted and analyzed at Michigan State University Kellogg Biological Station (Hickory Corners, MI) using a flow injector analyzer (NH4⁺ -N via the diffusion colorimetry technique and NO₃-N via cadmium reduction and colorimetry). PSNT data are presented in Table 4.3. After corn harvest, soil samples were collected to determine residual soil nitrate levels. Five cores per plot were collected with a soil probe to a depth of 15-20 cm; soil was dried at 60 °C, ground, and passed through a 4 mm sieve. A KCl extraction was performed as per KBS021 (2015). In 2012, samples were analyzed using cadmium reduction and colorimetry (KBS021, 2015). In years 2013 and 2014, extracts were sent to the Michigan State University Soil and Plant Nutrient Laboratory for nitrate analysis.

Weed counts

In the first year of the study, weeds were counted in two $1-m^2$ quadrats per treatment plot in June at the time of the first herbicide application (Table F.1). Weed densities were so large in the second and third years of the study that weeds were counted in two $0.1-m^2$ quadrats per treatment plot. Weeds were identified to species.

Corn measurements

Corn stand, chlorophyll content, tasseling, and leaf area index (LAI) data were collected on the dates listed in Table 3.6. The height of 10-15 corn plants per plot was measured with a meter stick in June at corn stage V6-V8. In late July to early August the percentage of plants in tassel in each plot was visually estimated by rating the plots as 0-25, 26-50, 51-75, or 76-100 percent in tassel. One non-destructive method of testing corn for relative N status is the use of a SPAD chlorophyll meter. The SPAD meter measures the relative "greenness" of the corn, which is a proxy for leaf chlorophyll content (Bullock and Anderson, 1998; Shapiro et al., 2006). To assess corn N status, a Minolta SPAD-502 chlorophyll meter (Spectrum Technologies, Inc., Aurora, IL) was used to collect leaf chlorophyll content data at corn stages V6 and VT. The meter was placed on the newest fully mature leaf of 15 plants in each experimental unit. An AccuPAR LP-80 photosynthetically-active radiation (PAR) sensor (Decagon Devices, Inc., Pullman, WA) was used to determine corn leaf area indices (LAI) at corn stage R6. Data were collected on days when the sky was clear.

Statistical analysis

Data were analyzed with SAS Enterprise Guide v. 6.1 (SAS Institute Inc., Cary, NC). Analysis of variance was carried out using PROC MIXED and the Tukey adjustment for all pairwise comparisons ($\alpha \le 0.05$). Data were tested for treatment*year interactions. Replication was treated as a random effect. Mean separation was conducted with the PDMIX 800 macro (Saxton, 1998). An average soil bulk density of 1.6 g cm⁻³ was used to convert soil nitrate values from ppm to kg ha⁻¹ using an average soil bulk density of 1.6 g cm⁻³ (USDA-NRCS, 2015). Corn height as a percentage of the control was calculated by dividing the heights of the corn plants in each replicate by the average height of the corn in the bare ground control plots in each replicate and then multiplying by 100. Weed density data were transformed with log base 10 prior to

ANOVA and mean separation. Post-corn harvest soil NO₃ data were transformed using the natural log prior to analysis to improve normality.

APPENDIX C

CHAPTER 3 SUPPLEMENTARY TABLES

Table 3.6. Data collection dates for experiments conducted in Lansing, MI.

	2012	2013	2014
Corn height	18 Jun.	27 Jun.	30 Jun
Soil sampling for PSNT [†]	7 Jun.	18 Jun.	5 Jul.
Weed counts at 1 st herbicide application	15 Jun.	21 Jun.	17 Jun.
Corn SPAD V6	25 Jun.	30 Aug.	30 Jun.
Corn SPAD VT	25 Jul.	9 Aug.	7 Aug.
Corn tasseling	26 Jul.	9 Aug.	4 Aug.
Corn leaf area index	10 Aug.	24 Aug.	17 Aug.
Soil sampling for NO ₃ testing	20 Nov.	6 Dec.	4 Dec.

†PSNT: pre-side dress nitrate nitrogen testing

Cover crop treatmentsRoot: shoot ratioOilseed radish0.82Oilseed radish + crimson clover0.90Oilseed radish + hairy vetch0.76Oilseed radish + winter pea0.82Oilseed radish + annual ryegrass0.85Oilseed radish + cereal rye1.00Oilseed radish + oats0.93P value0.94	Tuble 5.7. Part bliseed radish root. shoot ratios h	Table 5.7. 1 an onseed radish root. shoot ratios in Lansing, wit (2012-2014).					
Oilseed radish0.82Oilseed radish + crimson clover0.90Oilseed radish + hairy vetch0.76Oilseed radish + winter pea0.82Oilseed radish + annual ryegrass0.85Oilseed radish + cereal rye1.00Oilseed radish + oats0.93P value0.94	Cover crop treatments	Root: shoot ratio					
Oilseed radish + crimson clover0.90Oilseed radish + hairy vetch0.76Oilseed radish + winter pea0.82Oilseed radish + annual ryegrass0.85Oilseed radish + cereal rye1.00Oilseed radish + oats0.93P value0.94	Oilseed radish	0.82					
Oilseed radish + hairy vetch0.76Oilseed radish + winter pea0.82Oilseed radish + annual ryegrass0.85Oilseed radish + cereal rye1.00Oilseed radish + oats0.93P value0.94	Oilseed radish + crimson clover	0.90					
Oilseed radish + winter pea0.82Oilseed radish + annual ryegrass0.85Oilseed radish + cereal rye1.00Oilseed radish + oats0.93P value0.94	Oilseed radish + hairy vetch	0.76					
Oilseed radish + annual ryegrass0.85Oilseed radish + cereal rye1.00Oilseed radish + oats0.93P value0.94	Oilseed radish + winter pea	0.82					
Oilseed radish + cereal rye1.00Oilseed radish + oats0.93P value0.94	Oilseed radish + annual ryegrass	0.85					
Oilseed radish + oats0.93P value0.94	Oilseed radish + cereal rye	1.00					
P value 0.94	Oilseed radish + oats	0.93					
	P value	0.94					

Table 3.7. Fall oilseed radish root: shoot ratios in Lansing, MI (2012-2014).

† Data were combined across years because no treatment*year interaction was detected (p = 0.21).

	PSNT		We	ed coun	ts	Post-harvest NO ₃ -N	
	2012	2013	2014	2012	2013	2014	2012-2014§
Cover crop treatments]	kg N ha	a ⁻¹]	plants m	-2	kg N ha ⁻¹
Oilseed radish	38†	14	19	52 abc‡¶	192	327 ab	16
Crimson clover	60	12	19	41 abc	221	227 ab	17
Oilseed radish + crimson clover	38	14	17	44 abc	240	229 ab	15
Hairy vetch	65	14	24	51 ab	343	218 ab	21
Oilseed radish + hairy vetch	26	12	17	44 abc	271	171 bc	21
Winter pea	89	14	14	59 a	285	238 ab	24
Oilseed radish + winter pea	14	17	14	62 a	292	209 abc	14
Annual ryegrass	7	10	19	4 d	364	278 ab	14
Oilseed radish + annual ryegrass	53	14	26	24 c	305	269 ab	15
Cereal rye	14	10	12	29 bc	192	104 c	17
Oilseed radish + cereal rye	62	14	17	35 abc	327	198 abc	16
Oats	31	12	17	36 abc	289	274 ab	19
Oilseed radish + oats	46	22	17	42 abc	325	211 abc	16
Bare ground control	24	14	14	31 abc	228	360 a	16
Weedy control	29	12	22	45 abc	205	217 ab	13
P value	-	-	-	< 0.0001	0.41	< 0.0001	0.25

Table 3.8. Spring pre-side dress nitrate-N (PSNT), weed counts conducted at the time of the first herbicide application, and post corn harvest residual soil nitrate levels in experiments conducted in Lansing, MI (2012-2014).

[†] Soil samples were aggregated by treatment so no statistical analysis was performed.

‡ Means in the same column followed by the same letter are not significantly different ($\alpha \le 0.05$).

¶ Means were back-transformed from log base 10 after ANOVA and mean separation.

§ There was no treatment*year interaction (p = 0.37), so data were combined over years.

	2012	2013	2014
Cover crop treatments		0/	
Oilseed radish	100 (2.4)	98 (2.0)	109 (1.9)
Crimson clover	110 (2.4)	96 (4.3)	114 (2.5)
Oilseed radish + crimson clover	105 (2.1)	92 (3.5)	103 (1.5)
Hairy vetch	113 (1.5)	106 (3.0)	108 (2.0)
Oilseed radish + hairy vetch	115 (1.9)	100 (2.8)	110 (2.0)
Winter pea	114 (2.1)	105 (3.3)	105 (1.8)
Oilseed radish + winter pea	108 (1.3)	90 (2.6)	105 (1.4)
Annual ryegrass	53 (1.7)	85 (2.4)	103 (2.4)
Oilseed radish + annual ryegrass	89 (1.8)	100 (2.2)	104 (1.6)
Cereal rye	93 (2.5)	98 (2.6)	86 (1.8)
Oilseed radish + cereal rye	104 (1.6)	92 (3.4)	98 (1.9)
Oats	99 (1.3)	108 (1.8)	109 (2.5)
Oilseed radish + oats	103 (1.8)	82 (2.9)	111 (1.7)
Bare ground control	100 (1.3)	100 (1.9)	100 (0.9)
Weedy control	103 (2.2)	103 (3.3)	99 (1.5)

Table 3.9. Mean (± SE) corn heights† from experiments conducted in Lansing, MI (2012-2014).

† Heights were collected at corn stages V6-V8 and are presented as percent of the bare ground control.

		V6			VT	
	2012	2013	2014	2012	2013	2014
Cover crop treatments			Relat	ive N status†——		
Oilseed radish	49.1 ab‡	46.8	50.9 ab	42.8 a	27.1	54.6 ab
Crimson clover	50.8 a	44.8	52.3 ab	42.9a	27.8	47.2 abcd
Oilseed radish + crimson clover	48.3 ab	46.2	52.1 ab	41.7a	28.8	51.6 abc
Hairy vetch	49.7 ab	46.3	52.7 ab	43.0a	28.4	47.4 abcd
Oilseed radish + hairy vetch	49.3 ab	46.9	53.3 a	41.3 ab	29.3	48.4 abc
Winter pea	48.9 ab	48.5	52.5 ab	40.9 ab	28.3	48.9 abc
Oilseed radish + winter pea	49.5 ab	47.0	52.7 ab	41.8a	28.3	50.7 abc
Annual ryegrass	43.4 c	45.3	49.8 ab	39.3 ab	25.2	48.7 abc
Oilseed radish + annual ryegrass	46.7 bc	45.2	52.6 ab	39.3 ab	27.1	53.7 abc
Cereal rye	47.5 abc	44.4	47.0b	37.4b	25.2	39.1 d
Oilseed radish + cereal rye	46.6 bc	43.9	50.6 ab	39.0ab	26.0	51.6 cd
Oats	49.0 ab	45.8	51.8 ab	40.8 ab	28.3	54.5 ab
Oilseed radish + oats	49.1 ab	47.0	51.7 ab	40.5 ab	28.9	48.3 abc
Bare ground control	48.1 ab	44.4	49.0 ab	41.0ab	27.3	55.6a
Weedy control	47.9 ab	45.7	50.7 a	39.8 ab	28.2	46.1 bcd
<i>P-value</i>	0.0001	0.55	0.03	0.0004	0.23	< 0.0001

Table 3.10. Relative N status at corn stages V6 and VT from experiments conducted in Lansing, MI (2011-2014).

† Relative N status was based on SPAD chlorophyll meter readings.

‡ Means in the same column followed by the same letter are not significantly different ($\alpha \le 0.05$).

		Tasseling		Leaf area index
	2012	2013	2014	2012-2014¶
Cover crop treatments		%		
Oilseed radish	88(13)§	100(0)	88(7)	2.1
Crimson clover	100(0)	100(0)	88(7)	2.2
Oilseed radish + crimson clover	100(0)	100(0)	100(0)	2.0
Hairy vetch	100(0)	100(0)	94(7)	2.2
Oilseed radish + hairy vetch	100(0)	100(0)	100(0)	2.3
Winter pea	100(0)	100(0)	100(0)	2.1
Oilseed radish + winter pea	100(0)	100(0)	100(0)	2.3
Annual ryegrass	13(7)	100(0)	75(0)	2.0
Oilseed radish + annual ryegrass	88(13)	100(0)	88(13)	2.1
Cereal rye	81(12)	100(0)	31(6)	2.2
Oilseed radish + cereal rye	100(0)	100(0)	100(0)	2.2
Oats	94(6)	100(0)	81(19)	2.5
Oilseed radish + oats	100(0)	100(0)	100(0)	2.2
Bare ground control	100(0)	100(0)	100(0)	2.2
Weedy control	94(6)	100(0)	88(7)	2.2
<i>P-value</i>	-	-	-	0.89

Table 3.11. Average percent corn tasseling in late summer[†] and leaf area index of corn at stage R6[‡] from experiments conducted in 0.0ansing, MI (2012-2014).

[†] Data were collected on 26 Jul. 2012, 9 Aug. 2013, and 4 Aug. 2014.

‡ Data were collected with an AccuPAR LP-80 photosynthetically active radiation (PAR) sensor (Decagon Devices, Inc., Pullman, WA).

¶ Leaf area index data were combined because there was no treatment*year interaction (P = 0.83).

§ Values in parentheses represent one standard error of the mean.

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CHAPTER 4 IMPACT OF FALL-PLANTED COVER CROPS ON NITROUS OXIDE EMISSIONS ABSTRACT

Cover crops may help retain N in agricultural systems and decrease emissions of the potent greenhouse gas (GHG) nitrous oxide (N₂O). The purpose of this research was to investigate the impact annual ryegrass (Lolium multiflorum Lam.) and oilseed radish (Raphanus sativus L. var. oleiformis Pers.) on N2O emissions during the cover crop period and subsequent corn (Zea mays L.) growing season. The experiment was conducted at W.K. Kellogg Biological Station (KBS) (Hickory Corners, MI) and Michigan State University (MSU) (Lansing, MI) from fall 2012 - summer 2014 for a total of four site-years. Treatments included fall-planted oilseed radish, annual ryegrass, a mixture of oilseed radish and annual ryegrass, and a bare ground control. Cover crops were planted after wheat (Triticum aestivum L.) harvest and terminated with herbicides prior to planting field corn. Nitrous oxide emissions were measured three to five times during three periods: fall, spring, and summer. Maximum likely nitrogen inputs from cover crops ranged from 18-49 kg N ha⁻¹ at KBS and 35-83 kg N ha⁻¹ at MSU. After cover crop termination, spring-summer daily N₂O-N emissions at KBS ranged from 1.3-1.9 and 1.6-2.6 g N ha⁻¹ in 2013 and 2014, respectively, and from 1.8-4.2 and 8.1-13.7 g N ha⁻¹ at MSU in 2013 and 2014, respectively. Cumulative spring-summer N₂O-N emissions at KBS were 222-325 and 266-429 g N ha⁻¹ in 2013 and 2014, respectively. At MSU, the 2013 and 2014 spring-summer cumulative N₂O-N emissions were 322-769 and 1385-2361 g N ha⁻¹, respectively. No differences were detected among treatments, including the bare ground control, for cumulative spring-summer N₂O emissions, cumulative emissions scaled to total cover crop biomass, and cumulative emissions scaled to cover crop N. Nitrous oxide emissions did not represent a major pathway for

N loss in our study, and these results suggest that fall-planted non-legume cover crops do not increase N₂O emissions in N-limited corn-based rotations.

INTRODUCTION

Crop production systems impact the global dynamics of three of the major greenhouse gases (GHGS): methane (CH₄), carbon dioxide (CO₂), and nitrous oxide (N₂O) (Robertson et al., 2000). Agriculture accounts for 8.1% of the total greenhouse gas emissions in the U.S., with cropland the source of 61% of direct N₂O emissions and 79% of indirect N₂O emissions (USEPA, 2014). Nitrous oxide is the most potent of the agricultural GHGs. One molecule of N₂O has about 300 times the capacity of one carbon dioxide molecule to trap heat in the atmosphere (IPCC, 2007), therefore even relatively small reductions in N₂O emissions benefit the environment. Soil fertility and physical properties, weather, crop residue quality, and field operations can all influence N₂O fluxes (Millar et al., 2010; Novoa and Tejeda, 2006).

Nitrogen plays a key role in agricultural systems. Soil N availability is one of the major drivers of N₂O emissions (Bouwman et al., 2002a; Bouwman et al., 2002b; Millar et al., 2010; Mosier et al., 1996; Robertson et al., 2000). In particular, N fertilizer management, including amount, formulation, timing, and application method, affect N₂O emissions (Bouwman et al., 2002a; Bouwman et al., 2002b; Drury et al., 2012; Halvorson et al., 2010; Nash et al., 2012; Pelster et al., 2011). From both an environmental and an economic perspective, it is beneficial to maximize nitrogen use efficiency (NUE) of cash crops and retain N in the cropping system between cash crops. Nitrogen which is not retained may be lost via leaching, surface runoff, or gaseous emissions. It has been estimated that 50% of the N applied in agricultural systems is lost through these pathways (Tonitto et al., 2006). Millar et al. (2010) state that a reduction in applied N fertilizer is the most reliable way to reduce N₂O emissions from agronomic systems.

Production practices which alter the amount of mineral N in the soil can affect N₂O emissions by increasing or decreasing the amount of substrate available for the microbial processes of nitrification and denitrification responsible for N₂O production (Robertson and Groffman, 2015). Planting cover crops has been shown to increase, decrease, or have no impact on N₂O emissions by affecting soil N availability (Baggs et al., 2000a; Basche et al., 2014; Wagner-Riddle et al., 1997). A number of factors may be responsible (Basche et al., 2014). The C and N content and ratio in cover crop material helps determine the nature and quantity of substrate available for nitrification and denitrification, and impact the synchrony between N release during cover crop decomposition and N uptake by the cash crop (Millar et al., 2004; Mitchell et al., 2013). Due to its mobility, NO₃ that leaches through the soil profile and into waterways can later contribute to indirect N₂O emissions in locations other than that to which the N was originally applied (Nevison, 2000). Cover crops can reduce N leaching and runoff (Mitchell et al., 2013; Parkin et al., 2006; Syswerda et al., 2012). A meta-analysis by Tonitto et al. (2006) found that non-legume and legume cover crops decreased NO₃ leaching by 70% and 40%, respectively.

Results from the limited literature on cover crops and their impact on greenhouse gas emissions vary. In 40% of the 26 studies selected for meta-analysis by Basche et al. (2014), cover crops decreased N₂O emissions. In the other 60% of the studies evaluated, cover crops increased N₂O emissions. Legume cover crops generally had larger, positive response ratios (a quantification of the effect on N₂O emissions) than grass cover crops, in the absence of applied fertilizer. As fertilizer application rates increased, the legume response ratios declined while the grass response ratios increased a small amount. In one of the few experiments examining greenhouse gas emissions in wheat/corn/soybean [*Glycine max* (L.) Merr.] rotations using cover crops, Parkin and Kaspar (2006) found that while corn plots emitted significantly more N₂O than soybean plots, the inclusion of a cereal rye (*Secale cereale* L.) winter cover crop did not affect N₂O emissions. Robertson et al. (2000) state that the primary driver of N₂O fluxes in agricultural systems is the amount of N available in the soil; the more soil available N, the higher the flux. This is also supported by Gomes et al. (2009), who found leguminous cover crops had larger cumulative N₂O emissions during a 45 day period after cover crop residue management than a grass cover crop in no-till maize rotations in a subtropical climate. During this same period, Gomes et al. (2009) also found that N₂O emissions were correlated with cover crop N added to the soil, after which total soil N content drove N₂O emissions. Nitrous oxide emissions took longer to peak in a legume/grass cover crop mixture than in the legume alone. The use of a cover crop mixture with a balanced C:N ratio may improve N synchrony by delaying N loss from the system (Aulakh et al., 1991; Baggs et al., 2000b; Gomes et al., 2009).

The objective of this study was to quantify the impact of the presence and absence of fallplanted cover crops with varying C:N ratios and biomass on N₂O emissions. We hypothesize that N₂O emissions will: a) be smaller in the cover crop treatment with a high C:N ratio (annual ryegrass) than the bare ground control, and b) be larger in the cover crop treatment with a low C:N ratio (oilseed radish) than the control. We further hypothesize that N₂O emissions in the oilseed radish + annual ryegrass treatment will be intermediate between the annual ryegrass and oilseed radish monoculture treatments.

MATERIALS AND METHODS

Site characteristics and field operations

The experiment was conducted from 2012-2014 at Michigan State University (MSU) (Lansing, MI, USA; latitude 42° 46' N, longitude 84° 34' W) and W.K. Kellogg Biological
Station (KBS) (Hickory Corners, MI, USA; latitude 42° 24' N, longitude 85° 24' W). The soil at KBS both years was a Kalamazoo loam (fine-loamy mixed mesic Typic Hapludalfs). In 2012-2013 the soil at MSU was a Riddles-Hillsdale sandy loam (fine-loamy, mixed, mesic Typic Hapludalfs). In 2013-2014 the soil was a Capac loam (fine-loamy, mixed, mesic Aeric Ochraqualfs). Site characteristics are listed in Table 4.1. The 30-yr mean annual temperatures at KBS and MSU, respectively, are 10.1 and 9.3° C (NOAA, 2015). The 30-yr average yearly precipitation is 1005 and 817 mm, respectively, at MSU and KBS.

The experiment was structured as a randomized complete block design with four replicates per site-year. Experimental units were a minimum of 3 x 12 m in size. The four treatments were a bare ground control, oilseed radish Groundhog[™] (Ampac Seed Co., Tangent, OR) (11.2 kg ha⁻¹), annual ryegrass (44.8 kg ha⁻¹), and a mixture of oilseed radish and annual ryegrass (11.2 and 22.4 kg ha⁻¹, respectively). Field operation dates are listed in Table 4.2. The preceding wheat crop was fertilized as per standard growing practices with 123 kg N ha⁻¹ at green-up. Cover crops were planted in 15-cm rows in August-September following wheat harvest. At MSU the cover crops were planted with a no-till drill while at KBS the field was chisel plowed and cultivated prior to planting. The following spring, plots were sprayed with glyphosate (N-(phosphonomethyl)glycine) and 2,4-D (2,4-dichlorophenoxyacetic acid) to terminate weeds and cover crops which survived the winter. Because N fertilizer rate has been found to explain much of the variability found in other N₂O emissions research (Hoben et al., 2011), no fertilizer was applied to the cover crops in this study to avoid confounding or masking the effect of the cover crops on N_2O emissions. Weeds were controlled in the bare ground treatment during the cover crop phase through the application of glyphosate once each fall.

During the corn phase, weeds were controlled in all plots with glyphosate once or twice per summer as per standard practices.

Data collection

Temperature and precipitation data. Daily temperature and precipitation data for the KBS location were collected from the KBS Long Term Ecological Research (LTER) weather station (<u>http://lter.kbs.msu.edu/datatables/12</u>). Temperature data and precipitation values for 1 May – 31 October for the MSU location were collected from the Lansing/MSUHORT Enviro-weather station (MSUEW, 2015). Because the latter facility is not set up to measure solid precipitation, precipitation data for the MSU site for 1 November – 30 April were collected from the National Oceanic and Atmospheric Administration (NOAA) National Weather Service (NWS) website (NOAA, 2015).

Cover crop data collection. Cover crop aboveground biomass was harvested from two randomly placed 0.25 m² quadrats per experimental unit each fall and again the following spring (Table 4.2). Biomass was separated into its component cover crop fractions before processing. Fall biomass data were collected at the point of peak biomass production before the onset of hard frosts. Spring biomass data were collected immediately prior to cover crop termination. Harvested plants were dried at 70° C for 5-10 d, weighed, and ground using a Wiley mill (Thomas Scientific, Swedesboro, NJ). The C:N ratio was determined via combustion at KBS in 2012-2013 using an Elemental Combustion System 4010 CHNS-O (Costech Analytical Technologies, Inc.; Valencia, CA, USA) and at Midwest Laboratories, Inc. (Omaha, NE, USA) in 2013-2014.

Soil sampling. To determine baseline site soil characteristics, a 2-cm diameter soil probe was used to collect 20 subsamples to a depth of 15-20 cm in each replicate of each field in the fall of each year. The samples were stored at 4° C, ground to pass through a 2-mm mesh screen, and sent to the Michigan State University Soil and Plant Nutrient Laboratory to determine parameters including soil pH and organic matter. Values were averaged to create a composite sample for each field-year. Fall and spring soil NO3⁻ values were determined from soil samples collected to a depth of 15-20 cm (Table 4.2) with a 2-cm diameter soil probe. Five samples were collected in an "H" pattern from each experimental unit. Samples were dried at 60° C and ground to pass through a 2-mm mesh screen. In 2012-2013, extractions were performed and extracts were analyzed for NO₃⁻N and NH₄⁺-N at KBS using a flow injector analyzer using the diffusion colorimetry technique for NH₄⁺ -N and cadmium reduction and colorimetry for NO₃⁻ -N. In 2013-2014, extracts were sent to the Michigan State University Soil and Plant Nutrient Laboratory for analysis. Using the same soil sample collection method as for the NO₃⁻ protocol above, subsamples were collected at corn V6-V8 for pre-side dress NO_3 -N testing (PSNT). Samples were aggregated by treatment, ground to pass through a 2-mm mesh screen, and sent to the Michigan State University Soil and Plant Nutrient Laboratory for NO₃ analysis.

Nitrous oxide sampling and analysis. Cylindrical stainless steel chambers (Kahmark and Millar 2014; <u>http://lter.kbs.msu.edu/citations/3418</u>) were installed to a depth of 5-cm in the soil after cover crop planting. Each chamber was centered over one cover crop row. Chambers were removed prior to tillage and corn planting, and immediately reinstalled between corn rows thereafter. A manual sampling chamber protocol was used to determine greenhouse gas fluxes (Holland et al., 1999). On each sampling date, chamber lids were installed and then headspace

gas was immediately extracted using a 10-mL nylon syringe and a 23-gauge needle. At 20-min intervals over a 60-min period, three more samples were collected from each chamber. Gas samples were placed in 5.9 mL Exetainer® vials (Labco Limited, UK), which had been previously flushed with 10-mL of chamber air. Each vial was over-pressurized to 10-mL to avoid contamination and facilitate analysis. Soil temperature near each experimental unit was collected, along with chamber height to soil surface measured at four points around the circumference. Calculations to determine flux rates (µg N₂O-N m⁻² hour⁻¹) were made using the following equation:

$$N_2O = (\alpha \times V^*W_A^*60)/(A^*MV_{corr})$$

where α represents the change in headspace N₂O concentrations during the period when the chamber is closed, V is the chamber headspace volume in liters, W_A is the atomic mass of the N present in a molecule of N₂O (28), 60 is a conversion factor from minutes to hours, A is the soil surface area covered by the chamber (m⁻²), and MV_{corr} corrects for temperature and pressure mole volume at sampling. N₂O-N fluxes were then converted from µg N₂O-N m⁻² hour⁻¹ to g N₂O-N ha⁻¹ day⁻¹ for each sampling date. Samples were analyzed at the W.K. Kellogg Biological Station using an Agilent 7890A gas chromatograph (Agilent Industries, Inc.; Wilmington, DE, USA) fitted with a 63Ni electron capture detector and a Gerstel MPS2XL autosampler (Gerstel; Linthicum, MD, USA) (Kahmark and Millar, 2008).

There were three intensive gas sampling periods throughout the year. Sampling dates are listed in Table 4.2. Fall samples were collected three times over a 5-wk period. A minimum of three sets of samples were collected over a 7-12 d period during oilseed radish decomposition after winter-kill in early spring (usually in late March). Samples were collected three times over a 4-wk period to estimate spring gas fluxes near the time of corn planting, then once per month

during corn growth. Because of the length of time between fall and early spring gas sampling dates, cumulative N₂O-N emissions were calculated separately for fall and spring-summer periods by interpolating the areas under the lines delimited by the N₂O fluxes. Cumulative N₂O emissions for each period were divided by the number of days in the respective period to derive daily emissions values.

Data analysis

Data were analyzed using analysis of variance (ANOVA) with PROC MIXED in SAS Enterprise Guide v. 6.1 (SAS Institute, Cary, NC, USA) at a significance level of $\alpha \le 0.05$. Data were transformed as necessary to achieve normality. Data for each site-year are presented individually, rather than combined across years, to allow for more nuanced interpretation of the N₂O emissions data. When the F-test was significant, means were separated with the PDMIX 800 macro (Saxton, 1998). Soil NO₃ values were converted from ppm to kg ha⁻¹ using an average soil bulk density of 1.6 g cm⁻³ (Crum and Collins, 1995; USDA-NRCS, 2015). The fall oilseed radish + annual ryegrass C:N ratio was calculated by taking a weighted average using the C:N ratios of the component cover crops and the percentage of dry aboveground biomass of each cover crop in each replicate. Because oilseed radish failed to survive the winter and annual ryegrass survival was low at the KBS site, in each site-year total dry aboveground biomass was calculated using a combination of fall and spring data. In the annual ryegrass treatment, fall biomass values were considered as total biomass if spring biomass production was smaller than fall due to poor winter survival. If spring values were larger than fall, then the spring biomass was taken as total biomass. In the oilseed radish treatment, fall biomass values were used as the total biomass because oilseed radish winter kills and no biomass was present in the spring. In the annual ryegrass + oilseed radish treatment, oilseed radish fall biomass was added to the larger of

the fall or spring annual ryegrass biomass values. The total biomass was used to scale N_2O spring – summer emissions by dividing the total biomass by the cumulative spring – summer N_2O emissions. Spring – summer N_2O emissions were also scaled to cover crop N input using the same procedure to calculate maximum contributed N. Fall sampling cumulative emissions were not included in the calculations because all cover crop treatments were alive during the fall sampling period.

RESULTS AND DISCUSSION

Cover crop biomass

Fall. At KBS, oilseed radish produced 42% more fall aboveground biomass than annual ryegrass in 2012; oilseed radish + annual ryegrass was intermediate between the two (Table 4.3). There were no differences in fall cover crop biomass at KBS in 2013 or at MSU in 2012. At MSU in fall 2013, the oilseed radish and oilseed radish + annual ryegrass treatments produced an average of 63% more biomass than the annual ryegrass treatment. In the oilseed radish + annual ryegrass treatment, 66% of the mixture biomass consisted of oilseed radish each year at KBS (data not shown). At MSU, oilseed radish comprised 70 and 90% of the mixture biomass in fall 2012 and 2013, respectively. Fall KBS cover crop biomass was 44-78% of that at MSU (Tale 4.3), probably as a result of differences in soil type and fertility. The differences were not likely to be the result of different planting dates, since KBS was planted earlier than MSU both years (Table 4.2) and cover crop biomass production is usually larger with earlier fall planting dates. Fall oilseed radish biomass in this study was lower than that found in other studies. Fall-planted oilseed radish has been observed to produce 3,000-5,600 kg ha⁻¹ of aboveground biomass (Allison et al., 1998; Axelsen and Kristensen, 2000; Isse et al., 1999; Thorup-Kristensen, 2001;

Thorup-Kristensen, 2006). On the other hand, annual ryegrass biomass production in this study was similar to that found in other studies. Annual ryegrass has been found to produce 1160–2290 kg ha⁻¹ of dry aboveground biomass in the fall (Dapaah and Vyn, 1998) and 2150-4310 kg ha⁻¹ by the end of March and April, respectively (Odhiambo and Bomke, 2001).

Spring. Across site-years, the annual ryegrass treatment produced 1078-2145 kg ha⁻¹ biomass compared with 549-1601 kg ha⁻¹ biomass in the oilseed radish + annual ryegrass treatment (Table 4.3). The larger spring biomass production was not unexpected since the annual ryegrass seeding rate in mixture was half that of the monoculture treatment and oilseed radish failed to overwinter. In contrast to the fall trend, in the spring the cover crops produced up to three times more biomass at KBS than at MSU. The larger winter precipitation at KBS (Figures 4.1 and 4.2) provided insulation that protected the annual ryegrass from freezing, decreasing the amount of winterkill. In addition, the larger amount of spring biomass at KBS in 2013 compared with 2014 may have been the result of the greater winter precipitation in 2012-2013 (Figure 4.1).

Total biomass. Fall and spring aboveground dry biomass data were used to calculate the total maximum likely amount of biomass contributed by each cover crop treatment from fall through spring, removing the confounding effects of the failure of oilseed radish to survive the winter and low annual ryegrass winter survival at the MSU site. Across fall and spring at KBS, oilseed radish + annual ryegrass produced 1827-2592 kg ha⁻¹ total biomass in 2012-2013 and 2013-2014, more than the 1234-1827 kg ha⁻¹ produced by oilseed radish (Table 4.3). No differences were detected in cover crop total biomass at MSU in 2012-2013. In 2013-2014, the oilseed radish +

annual ryegrass and oilseed radish total biomasses were 2642 and 2806 kg ha⁻¹, respectively, compared with 1676 kg ha⁻¹ annual ryegrass total biomass.

Cover crop C:N ratios, N and soil N

Fall C:N ratios. No differences were detected in fall aboveground biomass C:N ratio in 2012 at KBS (Table 4.4). The C:N ratios ranged from 17:1-20:1. In 2013 at KBS the C:N ratio of annual ryegrass was 13-29% higher than that of the oilseed radish + annual ryegrass and oilseed radish treatments, respectively. In fall 2012 at MSU the annual ryegrass treatment C:N ratio was 24:1, larger than the 16:1 ratio in the oilseed radish treatment. In fall 2013, the oilseed radish + annual ryegrass C:N ratio was 13:1, larger than the 9:1-12:1 observed in the other two cover crop treatments. The C:N ratios of the cover crops studied generally followed the expected pattern. Oilseed radish had the lowest C:N ratio, annual ryegrass the highest ratio, and the oilseed radish + annual ryegrass treatment was intermediate between the two. The exception was MSU in fall 2013, when the oilseed radish + annual ryegrass treatment had the highest C:N ratio. The C:N ratios in that site-year ranged from 9:1 – 13:1 (Table 4.4), unexpectedly low for all treatments. Given how low the C:N ratios were, it is unlikely that the differences were biologically significant.

Spring C:N ratios. At KBS, the annual ryegrass C:N ratio was eight percent higher than the oilseed radish + annual ryegrass treatment in spring 2013 and 12% higher in 2014 (Table 4.4). No differences were detected at the MSU site, though in 2014 annual ryegrass and oilseed radish + annual ryegrass had spring C:N ratios of 10:1 and 16:1, respectively.

The annual ryegrass in our study generally had higher C:N ratios in the fall than have been reported in the literature. For example, Thorup-Kristensen (1994) found annual ryegrass to have a C:N ratio of 12:1 in the fall. However, in that study fertilizer was applied to the cover crops, while in our study it was not. The spring annual ryegrass C:N ratios at the KBS location in our study were similar to those commonly found in the literature. Kuo and Sainju (1998) found fallplanted annual ryegrass to have a C:N ratio of 24:1 in the spring, while Baggs et al. (2000b) found it to have a C:N ratio of 25:1. As with annual ryegrass, oilseed radish C:N ratios in this study were similar to those found in the literature, generally 13:1 to 18:1 (Baggs et al., 2000b; Thorup-Kristensen, 1994). In our study, the C:N ratios for all treatments in both the fall and spring were higher at KBS than MSU (Table 4.3). The typical C:N ratio threshold above which N immobilization is expected to occur is 20-25:1 (Cochran et al., 1980; Kuo and Jellum, 2002). The fall and spring C:N ratios in our study fell within a narrow range around this threshold. While it is possible that net N immobilization occurred as a result of cover crop inclusion in this cropping system, it was unlikely to have been a major factor influencing N₂O-N emissions. The question arises as to whether the statistical significances observed were also biologically significant, given both the generally low C:N ratios and the small percentage differences.

Cover crop N input. To remove the differences caused by varying levels of winter survival, cover crop maximum N input was calculated in the same way as total biomass. No differences were detected in cover crop N input at KBS in 2012-2013, probably because of the variability in the data (Table 4.4). In 2013-2014, the oilseed radish + annual ryegrass treatment had 43-67 % larger N input than the oilseed radish and annual ryegrass treatments, respectively. At MSU, the cover crop N input was 1.4-1.7 times larger in the oilseed radish treatment than in the oilseed

radish + annual ryegrass and annual ryegrass treatments, respectively. No differences at MSU were detected in 2013-2014. The N input calculations suggest that at KBS, cover crops at most could have contributed 33-45 kg N ha⁻¹ in 2012-2013 and 18-30 kg N ha⁻¹ in 2012-2014. At MSU these values were, respectively, 35-60 and 68-83 kg N ha⁻¹ in 2012-2013 and 2013-2014. Given the generally greater biomass production (Table 4.3) and lower C:N ratios (Table 4.4) at MSU compared with KBS, it is unsurprising that the cover crops contributed more N to the cropping system at MSU than KBS.

Soil NO₃ levels. Soil samples were collected in the fall and spring during cover crop growth to test soil NO₃ levels. In fall 2013, NO₃ levels were 2.8-5.1 times greater in the control and oilseed radish treatments than in the annual ryegrass and oilseed radish + annual ryegrass treatments at KBS (Table 4.5), suggesting more soil N was available to undergo denitrification in these plots. At MSU, NO₃ levels were 128-249% greater in the control than in the cover crop treatments. At KBS in the spring, NO₃ levels were higher in the oilseed radish treatment than in the other treatments (8 kg N ha⁻¹ vs. 2.4-5.5 kg N ha⁻¹). Denitrification and subsequent N₂O emissions would thus be expected to be higher in the oilseed radish plots than in other plots. At MSU in spring 2013 oilseed radish and control plots had two times more kg N ha⁻¹ than the annual ryegrass plots. In spring 2014 at MSU no differences were detected.

Soil samples were collected twice during the corn phase of the experiment to test for soil N, once at corn V6-V8 for PSNT and again after corn harvest. PSNT data were pooled, so no statistical analysis was performed (Table 4.1). At KBS, control and oilseed radish plots tested at 20–30 kg N ha⁻¹, while annual ryegrass and oilseed radish + annual ryegrass plots tested at 11–14

kg N ha⁻¹. The range was narrower at MSU in 2013 in the spring, with 11–16 kg N ha⁻¹. In 2014, soil NO₃ values were lower at KBS than MSU, ranging from 9-16 and 16-24 kg N ha⁻¹, respectively. After corn harvest, no differences were generally detected in soil NO₃ after corn harvest (Table 4.5).

The finding in our study that annual ryegrass decreased soil NO₃ levels more than oilseed radish is in line with the results of other studies (Thorup-Kristensen, 1994; Vyn et al., 2000). NO₃ results likely varied between locations at each sampling point due to a combination of different soil fertility levels (Table 4.1), weather (Figures 4.1 and 4.2), and cover crop biomass production and C:N ratios (Tables 4.3 and 4.4).

Nitrous oxide emissions

N₂O fluxes. Nitrous oxide flux emissions were highly variable in our study between sites and years, and were an order of magnitude larger at MSU in spring-summer 2014 than 2013 (Figures 4.1 and 4.2). At both KBS and MSU, N₂O emissions were smallest during the fall sampling period of late October - December, peaked in March – June, and then returned to near-fall levels in July – September. The fluxes followed similar patterns at both locations over the course of each site-year. Cumulative N₂O-N emissions were calculated for the fall and spring-summer sampling periods (Tables 4.6 and 4.7). N₂O-N emissions were between 2 and 40 times higher at MSU than at KBS during both sampling periods each year. This is likely due to different soil types (Table 4.1), amount of cover crop residue (Table 4.3), and cover crop C:N ratios and N inputs (Table 4.4) at the two locations. Soil organic matter (SOM) at MSU was more than twice that of KBS in 2013-2014. Lower SOM typically results in lower net N mineralization rates, leading to less soil N available for nitrification and denitrification. Not only was SOM higher at MSU in 2014, but KBS cover crop total biomass was 60% lower than that of MSU while N

inputs at MSU were 3.2 times larger than at KBS (Tables 4.3 and 4.4). No differences were detected between treatments, including the bare ground control, in cumulative fall or spring-summer N₂O-N emissions in any site-year. Cumulative N₂O-N emissions in the fall were lower than in the spring-summer. During the 28-d fall sampling period in 2012 and the 35-d period in 2013, N₂O-N emissions ranged from 2-11 and 7-14 g N₂O-N ha⁻¹, respectively, at KBS (Table 4.6). At MSU the 2012 fall sampling period was 29-d in length and cumulative emissions were $4-20 \text{ g } N_2\text{O-N} \text{ ha}^{-1}$, while during the 39-d 2013 fall sampling period cumulative emissions were $64-324 \text{ g } N_2\text{O-N} \text{ ha}^{-1}$ (Table 4.7). During spring-summer sampling period, cumulative emissions were $222-325 \text{ g } N_2\text{O-N} \text{ ha}^{-1}$ in 2013 at KBS and 266-429 g N₂O-N ha⁻¹ in 2014 (Table 4.6). At MSU the spring-summer 2013 cumulative emissions were $322-769 \text{ g } N_2\text{O-N} \text{ ha}^{-1}$, while in 2014 they were $1385-2361 \text{ g } N_2\text{O-N} \text{ ha}^{-1}$ (Table 4.7).

Spring-summer cumulative N₂O emissions were scaled to total cover crop biomass and cover crop N inputs. There were no differences in N₂O-N emissions per Mg cover crop biomass between any of the cover crop treatments and the bare ground control for any site-year (Tables 4.6 and 4.7). There were also no differences in N₂O-N emissions per kg cover crop N input detected between treatments in any site-year (Tables 4.6 and 4.7). At the KBS location, 2014 N₂O-N emissions per kg cover crop N were 1.5-2.7 times greater than 2013. At the MSU location, 2014 N₂O-N emissions per kg cover crop N were 1.2-2.1 times greater than 2013. Interestingly, in spite of the differences in 2014 spring-summer N₂O fluxes between the KBS and MSU sites (Figures 4.1 and 4.2), N₂O-N emissions per kg cover crop N were of similar magnitude (Tables 4.6 and 4.7).

As is typical with manual chamber sampling protocols, the discontinuous nature of the sampling may not have captured all large fluxes. However, our sampling method encompassed

periods of time during which a range of fluxes were encountered, enabling unbiased treatment comparisons to be made. Parkin and Kaspar (2006) found that including cover crops had no impact on N₂O emissions when compared to their absence. Cover crop biomass production (Table 4.3) and C:N ratios (Table 4.4) may help explain the difference in the size of N_2O emissions between the KBS and MSU sites. Spring biomass production and C:N ratios were both larger at KBS than MSU. Net immobilization of soil N could have decreased the amount of N available to the microbes that facilitate denitrification and thus also decreased N₂O emissions. At MSU, spring cover crop biomass production was smaller and C:N ratios were under the 20:1 to 25:1 thresholds at which N immobilization becomes probable (Cochran et al., 1980; Kuo and Jellum, 2002). Given the availability of both C and N from the cover crops, and the anaerobic soil conditions typical of a wet Michigan spring, it is likely that most of the N₂O emissions resulted from the denitrification process, corroborating isotopic studies by Ostrom et al. (2010) at nearby sites. Other research has also found N2O fluxes to be correlated with soil N in excess of crop uptake (McSwiney and Robertson, 2005; Van Groeningen et. al., 2010). Based on metaanalysis and modeling done by Bouwman et al. (2002b) and Novoa and Tejeda (2006), about one percent of the N applied to a system would be expected to be lost to N₂O emissions. In a study by Gomes et al. (2009), one percent or less of the N in legume cover crop residue was lost to N₂O emissions. We calculated that in our study, the probable maximum amount of N accumulated in cover crop biomass tissue each year at KBS was 18-49 kg N ha⁻¹ (Table 4.4). Thus, a minimum estimate for N₂O-N emissions would be 180-490 g N ha⁻¹. Based on the probable maximum amount of N accumulated by cover crops at the MSU location (Table 4.4), we estimate that N₂O-N emissions would be in the range of 350-830 g N ha⁻¹. At KBS, springsummer cumulative N₂O-N emissions were within the 180-490 g N ha⁻¹ estimate (Table 4.6). At

MSU, the 2013 spring-summer cumulative N₂O-N emissions were also within the expected 350-830 g N ha⁻¹ range (Table 4.7). In 2014, however, N₂O-N emissions were 1.7-2.8 times larger than the largest typical emissions based solely on cover crop N input. As has previously been discussed, the divergence from literature values was probably the result of soil properties and surplus soil N. Overall, 1-2% of the N contributed to the soil by cover crop biomass was accounted for by N₂O-N emissions; therefore, N₂O emissions did not represent a major pathway for N loss from this cropping system.

CONCLUSIONS

Contrary to our hypotheses and despite varying cover crop biomass and N inputs across site-years, the inclusion of fall-planted cover crops did not increase or decrease N₂O emissions when compared to the bare ground control and each other. Overall, N₂O emissions did not represent a major pathway for N loss. No differences were detected among treatments in terms of cumulative spring-summer N₂O emissions or cumulative emissions scaled to total cover crop biomass and N input. We may have failed to detect differences because leaching was a more prevalent pathway for N loss (leaving less N available for denitrification and emission), or because cover crop N inputs were too low to allow for differentiation between treatments. These results suggest that while farmers may need to balance other advantages and disadvantages associated with fall-planted oilseed radish and annual ryegrass, impact on N₂O emissions need not be a major concern. A future avenue of research to expand upon this work could include the repetition of this experiment with a split-plot design wherein N fertilizer application is a factor and gas and soil sampling are conducted more frequently to better quantify the movement of N

through the cropping system. It would be interesting to create an N budget by combining N_2O and lysimeter sampling to quantify the partitioning between N_2O emissions and NO_3 leaching. **APPENDICES**

APPENDIX A

CHAPTER 4 TABLES AND FIGURES

Table 4.1. Soil characteristics and spring PSNT values[†] in Hickory Corners, MI (KBS) and Lansing, MI (MSU).

	2012-	-2013	2013-20)14
	KBS	MSU	KBS	MSU
Soil characteristics				
Slope (%)	0-3	2-6	0-3	0-3
Soil pH	6.4	5.4	6.1	5.8
Soil organic matter (%)	2.6	2.5	1.7	3.8
Spring PSNT values (kg N ha ⁻¹)				
Oilseed radish	30	15	16	24
Annual ryegrass	11	11	14	22
Oilseed radish + annual ryegrass	14	16	13	31
Control	20	15	9	16

[†] Soil samples were aggregated by treatment prior to KCl extraction so no statistical analysis was performed.

	201	2-2013	2013	3-2014
	KBS	MSU	KBS	MSU
Field management				
Fall tillage	25 Jul. 2012	-	19 Aug. 2013	-
Cover crop planting	25 Jul. 2012	13 Aug. 2012	19 Aug. 2013	2 Sept. 2013
Cover crop termination	7 May 2013	8 May 2013	10 May 2014	19 May 2014
Data collection				
Fall soil sampling	2 Oct. 2012	28 Sept. 2012	20 Nov. 2013	15 Nov. 2013
Spring soil sampling	7 May 2013	8 May 2013	10 Apr. 2014	7 Apr. 2014
Fall biomass harvest	2 Nov. 2012	5 Nov. 2012	29 Oct. 2013	25 Oct. 2013
Spring biomass harvest	7 May 2013	8 May 2013	10 May 2014	13 May 2014
Fall baseline gas sampling	29 Oct. 2012	2 Nov. 2012	28 Oct. 2013	24 Oct. 2013
5 1 5	5 Nov. 2012	9 Nov. 2012	20 Nov. 2013	15 Nov. 2013
	26 Nov. 2012	1 Dec. 2012	2 Dec. 2013	2 Dec. 2013
Spring gas sampling	29 Mar. 2013	29 Mar. 2013	7 Apr. 2014	31 Mar. 2014
	3 Apr. 2013	5 Apr. 2013	10 Apr. 2014	7 Apr. 2014
	5 Apr. 2013	7 Apr. 2013	11 Apr. 2014	11 Apr. 2014
	•	-	18 Apr. 2014	1
Summer gas sampling	6 May 2013	3 May 2013	10 May 2014	11 May 2014
	17 May 2013	26 May 2013	1 Jun. 2014	31 May 2014
	4 Jun. 2013	9 Jun. 2013	9 Jun. 2014	6 Jun. 2014
	10 Jul. 2013	11 Jul. 2013	27 Jun. 2014	30 Jun. 2014
	25 Aug. 2013	20 Aug. 2013	29 Jul. 2014	1 Aug. 2014
	-	-	8 Aug. 2014	21 Aug. 2014
	18 Sep. 2013	27 Sep. 2013	17 Sep. 2014	19 Sep. 2014

Table 4.2. Field operation and data collection dates in Hickory Corners, MI (KBS) and Lansing, MI (MSU).

		Fall biomass				Spring biomass				Total biomass			
	KI	KBS		MSU		KBS		MSU		BS	MSU		
	2012	2013	2012	2013	2013	2014	2013	2014	2013	2014	2013	2014	
		kg ha ⁻¹				kg ha ⁻¹				kg ha ⁻¹			
Oilseed radish	1895 a	1233	2436	2806 a	-	-	-	-	1895b	1234b	2436	2806 a	
Annual ryegrass	1339b	1297	1973	1676b	2145 a	1248 a	1078 a	1629 a	2145 ab	1248b	1973	1676b	
Radish + ryegrass	1472 ab	1221	2052	2642 a	1601 b	936b	549b	637b	2592 a	1827 a	2052	2642 a	
P value	0.01	0.78	0.23	0.006	0.0006	0.02	0.003	0.0002	0.02	0.0004	0.23	0.006	

Table 4.3. Fall, spring, and total contributed dry aboveground cover crop biomass[†] in Hickory Corners, MI (KBS) and Lansing, MI (MSU) (2012-2014).

† The total biomass for the oilseed radish treatment was the fall biomass. The total biomass for the annual ryegrass treatment was the fall or spring biomass, whichever was greater in each site-year. The total biomass for the oilseed radish + annual ryegrass treatment was the fall oilseed radish biomass plus the fall or spring annual ryegrass mixture biomass, whichever was greater in each site-year. ‡ Within each column, values followed by the same letter are not significantly different ($\alpha \le 0.05$).

		Fall C:N				Spring C:N				Cover crop N input			
	K	KBS		MSU		KBS		MSU		KBS		SU	
	2012	2013	2012	2013	2013	2014	2013	2014	2013	2014	2013	2014	
									·	kg h	a ⁻¹		
Oilseed radish	17	21 a	16a	12a	-	-	-	-	45	21 a	60 a	73	
Annual ryegrass	20	27 c	24b	9a	27 a	28 a	20	10	33	18a	35b	68	
Radish + ryegrass	20	24 b	19 ab	13b	25 b	25b	19	16	49	30b	43 ab	83	
P value	0.06	0.0009	0.007	0.005	0.01	0.01	0.55	0.05	0.12	0.009	0.03	0.10	

Table 4.4. Fall and spring cover crop C:N ratios and maximum contributed cover crop N⁺ in Hickory Corners, MI (KBS) and Lansing, MI (MSU) (2012-2014).

[†] The cover crop N input for the oilseed radish treatment was N present in the fall biomass. The cover crop N input for the annual ryegrass treatment was the N present in the fall or spring biomass, whichever was greater in each site-year. The N input for the oilseed radish + annual ryegrass treatment was the N present in the fall oilseed radish biomass plus the N present fall or spring annual ryegrass mixture biomass, whichever was greater in each site-year.

‡ Within each column, values followed by the same letter are not significantly different ($\alpha \le 0.05$).

Fall-cover crop phase Fall-after corn harvest Spring-cover crop phase KBS MSU KBS MSU KBS MSU 2013 2013 2013 2014 2013 2014 2013 2014 2013 2014 -kg ha⁻¹-- kg ha⁻¹-- kg ha⁻¹-Oilseed radish 23.8 4.9†a‡ 11.7b 7.0 10.4 a 25.7b 7.9a 7.0 17.6 9.5 Annual ryegrass 1.3 c 8.3 c 2.4 c 6.9 4.6b 18.5 5.5 16.3 9.7 33.6 a Radish + ryegrass 2.4b 12.7b 3.9bc 7.4 5.0b 21.2 6.5 18.3 8.6 26.8 ab 8.2 24.0 Control 6.6a 29.4 a 5.5b 10.5 a 6.5 15.8 7.8 31.6 ab < 0.0001 P value < 0.0001 0.0002 0.62 0.004 0.68 0.40 0.74 0.15 0.02

Table 4.5. Soil NO₃ values during fall and spring cover crop phases and after corn harvest in Hickory Corners, MI (KBS) and Lansing, MI (MSU) (2012-2014).

[†] Fall cover crop NO₃ means presented were back-transformed after ANOVA.

‡ Within each column, values followed by the same letter are not significantly different ($\alpha \le 0.05$).

	Oilseed	Annual	Radish +	Control	Dyoluo
	radish	ryegrass	ryegrass	Colluloi	1 value
		$-g N_2O$]	N ha ⁻¹ —		
2012 fall cumulative N ₂ O emissions †‡	2	2	7	11	0.11
2013 fall cumulative N ₂ O emissions	14	8	9	7	0.59
2013 spring-summer cumulative N ₂ O emissions	325	222	271	276	0.49
2014 spring-summer cumulative N ₂ O emissions	266	429	349	316	0.35
2013 spring-summer daily N ₂ O emissions	1.9	1.3	1.6	1.6	0.62
2014 spring-summer daily N ₂ O emissions	1.6	2.6	2.1	1.9	0.35
2013 N ₂ O emissions per Mg total contributed cover crop biomass	186	107	103	-	0.21
2014 N ₂ O emissions per Mg total contributed cover crop biomass	212	360	412	-	0.55
2013 N ₂ O emissions per kg maximum contributed cover crop N	8	7	5	-	0.67
2014 N ₂ O emissions per kg maximum contributed cover crop N	12	19	10	-	0.23

Table 4.6. Cumulative, daily, and scaled N₂O-N emissions in Hickory Corners, MI (KBS) in 2012-2014.

[†] The fall sampling period was 28 d in 2012 and 35 d in 2013. The spring-summer sampling period was 173 d in 2013 and 163 d in 2014.

‡ Fall cumulative N₂O emissions were transformed by taking the square root prior to ANOVA. Spring-summer daily N₂O emissions were transformed with log base 10. Means presented are back-transformed.

	Oilseed radish	Annual ryegrass	Radish + ryegrass	Control	P value
		$-g N_2O-1$	N ha ⁻¹ —		
2013 fall cumulative N ₂ O emissions †‡	13	4	10	20	0.22
2014 fall cumulative N ₂ O emissions	324	309	64	89	0.45
2013 spring-summer cumulative N ₂ O emissions	769	322	561	391	0.09
2014 spring-summer cumulative N ₂ O emissions	1669	1720	1385	2361	0.46
2013 spring-summer daily N ₂ O emissions	4.2	1.8	3.1	2.1	0.10
2014 spring-summer daily N ₂ O emissions	9.7	10.0	8.1	13.7	0.46
2013 N ₂ O emissions per Mg total contributed cover crop biomass	323	160	279	-	0.23
2014 N ₂ O emissions per Mg total contributed cover crop biomass	603	1005	521	-	0.06
				-	
2013 N ₂ O emissions per kg maximum contributed cover crop N	13	9	14	-	0.42
2014 N ₂ O emissions per kg maximum contributed cover crop N	21	19	17		0.55

	Table 4.7. Cumulative,	daily, and	l scaled N ₂ O-N	emissions in	Lansing, MI	(MSU)) in 2012-2014.
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[†] The fall sampling period was 29 d in 2012 and 39 d in 2013. The spring-summer sampling period was 182 d in 2013 and 172 d in 2014.

‡ Fall cumulative N₂O emissions were transformed with log base 10 prior to ANOVA. Emissions per total contributed biomass were transformed with log base 10 or by taking the inverse. Means presented are back-transformed.



Figure 4.1. Temperature (°C), daily precipitation (mm), and nitrous oxide emissions (g N₂O-N ha⁻¹ day) from 29 Oct. 2012 – 18 Sep. 2013 (year 1) and 28 Oct. 2013 – 17 Sep. 2014 (year 2) in Hickory Corners, MI (KBS). Each error bar represents one standard error from the mean.



Figure 4.2. Temperature (°C), daily precipitation (mm), and nitrous oxide emissions (g N₂O-N ha⁻¹) from 11 Nov. 2012 – 27 Sep. 2013 and 24 Oct. 2013 – 19 Sep. 2014 in Lansing, MI (MSU). Note the right (2013-2014, year 2) y-axis is one order of magnitude larger than the left y-axis. Each error bar represents one standard error from the mean.

APPENDIX B

CORN GROWTH AND YIELD MATERIALS AND METHODS

Field operations

Field operation dates are listed in Table J.1. After cover crop termination as previously described, the field was prepared with a chisel plow and a soil finisher and 102-d corn 'DeKalb 52-59' (Monsanto, St. Louis, MO) was planted in 76-cm rows at a rate of 74,100 per ha⁻¹ using a four-row planter. Because fertilizer rate has been found to explain much of the variability in N₂O emissions (Basche et al., 2014; Hoben et al., 2011), no fertilizer was applied to the corn in this study to avoid confounding or masking the effect of the cover crops on N₂O emissions. Weeds were controlled with glyphosate as needed at corn stages V4-6.

Corn data collection

Corn heights were measured when corn was at the V6-V8 stage (Table J.1). A minimum of ten plants per experimental units were measured by holding the newest fully mature leaf up against a meter stick. Corn N status was determined after the onset of tasseling by collecting 25 corn ear leaves total (the leaf directly below the lowest ear of corn on each plant) from the two data rows in each experimental unit. Corn ear leaves were dried at 70 °C for 5-7 d and then ground with a Wiley mill (Thomas Scientific, Swedesboro, NJ). Two grams of this ground material were then sent to A&L Great Lakes Laboratories, Inc. (Fort Wayne, IN) for corn ear leaf N analysis. A Minolta SPAD-502 chlorophyll meter (Spectrum Technologies, Inc., Aurora, IL) was used to collect chlorophyll content data at corn stages V6 and VT. The meter was placed on the newest fully mature leaf of 15 plants in each experimental unit. An AccuPAR LP-80 photosynthetically active radiation (PAR) sensor (Decagon Devices, Inc., Pullman, WA) was used to determine corn leaf area indices (LAI) at corn stage R6. Data were collected on days when the sky was clear. To determine corn grain yields, 9 m of each data row (outside of the area

in which corn ear leaves were collected) were harvested using a two-row research combine. Yield was adjusted to 15.5% moisture.

Data analysis

Data were analyzed as previously described in this chapter. Corn heights were converted into a percentage of the control by dividing the heights of the plants in each treatment in each replicate by the average height of the corn in the corresponding replicate control treatment.

APPENDIX C

CORN GROWTH AND YIELD TABLES

Table 4.8. Field operation and corn data collection dates in Hickory Corners, MI (KBS) and Lansing, MI (MSU).

	201	12-2013	2013	-2014
	KBS	MSU	KBS	MSU
Field management				
Spring tillage	13 May 2013	20 May 2013	24 May 2014	28 May 2014
Corn planting	15 May 2013	20 May 2013	24 May 2014	28 May 2014
Data collection				
Height	23 Jun. 2013	27 Jun. 2013	27 Jun. 2014	30 Jun. 2014
V6 SPAD	23 Jun. 2013	30 Jun. 2013	27 Jun. 2014	30 Jun. 2014
VT SPAD	5 Aug. 2013	9 Aug. 2013	4 Aug. 2014	7 Aug. 2014
Grain harvest	4 Nov. 2013	24 Nov. 2013	11 Nov. 2014	14 Nov. 2014

		V6 relativ	e N status†		VT relative N status			
	K	KBS		MSU		KBS		IS U
Treatments	2013	2014	2013	2014	2013	2014	2013	2014
Oilseed radish	44.3 (0.6)	46.1 (0.6)	46.8 (0.5)	51.2 (0.5)	26.3 (0.5)	24.9 (0.6)	27.1 (0.5)	54.6 (0.6)
Annual ryegrass	40.3 (0.5)	44.4 (0.5)	44.7 (0.5)	49.8 (0.6)	21.7 (0.5)	24.5 (0.4)	25.2 (0.6)	49.0 (1.0)
Oilseed radish + ryegrass	42.4 (0.5)	44.8 (0.6)	46.6 (0.7)	52.4 (0.5)	23.7 (0.4)	25.0 (0.5)	27.1 (0.5)	54.5 (0.8)
Control	42.9 (0.6)	45.2 (0.6)	47.4 (0.7)	49.0 (0.6)	25.5 (0.5)	22.6 (0.6)	27.3 (0.6)	55.6 (0.7)

Table 4.9. Mean ± SE relative N status at stages V6 and VT in Hickory Corners, MI (KBS) and Lansing, MI (MSU).

† Relative N status was based on SPAD chlorophyll meter readings.

		Height					LAI		Y	ield	
	K	KBS		MSU		MSU	KBS	MSU	KBS	MSU	
	2013	2014	2013	2014							
		——% of the control——				%—			—kg	—kg ha ⁻¹ —	
Oilseed radish	120 (2)	96 (1)	96 (2)	109 (2)	1.4	2.0	1.1	2.2	1846	3965	
Annual ryegrass	91 (3)	95 (2)	84 (3)	103 (2)	1.3	1.9	1.2	2.2	1789	3431	
Oilseed radish + ryegrass	120 (3)	101 (1)	98 (2)	100(1)	1.4	2.0	1.1	2.5	1892	3708	
Control	100 (2)	100(1)	99 (1)	104 (2)	1.5	2.0	1.0	2.4	1811	3759	
P value					0.70	0.94	0.63	0.72	0.97	0.82	

Table 4.10. Corn height, ear leaf N, leaf area index (LAI), and grain yield in Hickory Corners, MI (KBS) and Lansing, MI (MSU) (2013-2014).

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