OPTIMIZING RYE-VETCH COVER CROP MIXTURE MANAGEMENT IN VEGETABLE CROPPING SYSTEMS: OPPORTUNITIES AND TRADEOFFS

By

Zachary D. Hayden

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ABSTRACT

OPTIMIZING RYE-VETCH COVER CROP MIXTURE MANAGEMENT IN VEGETABLE CROPPING SYSTEMS: OPPORTUNITIES AND TRADEOFFS

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Cereal-legume cover crop mixtures composed of cereal rye (*Secale cereale* L.) and hairy vetch (*Vicia villosa* Roth) have the potential to combine the unique strengths of the component species while taking advantage of interspecific synergies. However, management practices and environmental conditions can influence both species interactions and residue decomposition. The overarching objective of this research was to evaluate strategies for optimizing rye-vetch mixture performance within vegetable cropping systems.

The main component of this research evaluated 1) how the relative proportions of rye and vetch sown in mixtures influenced cover crop biomass production, winter annual weed suppression, vetch winter survival, and vetch N fixation; and 2) the interactive effects of rye-vetch residue characteristics and the use of black polyethylene mulch (PM) on soil N dynamics, microbial communities, and vegetable yields during production of bell pepper (*Capsicum annuum* L.) and slicing cucumber (*Cucumis sativus* L.). Following a replacement series design, main plot treatments consisted of a gradient of seven rye-vetch mixture proportions ranging from 100% vetch to 100% rye, in addition to a no-cover-crop control. Following cover crop termination, subplots consisted of pepper and cucumber grown either with or without PM. Density and biomass composition in the mixtures were highly correlated with rye and vetch seeding rates, with little evidence of substantial interspecific interference. Total shoot biomass in all mixtures was equal to or greater than that of either monoculture, but no differences were detected in vetch winter survival or the efficiency of N fixation. Changing the proportions of rye and vetch in the mixtures resulted in important tradeoffs among some of the services provided by the cover crop. Increasing vetch in mixtures generally led to greater fixed N accumulation, soil inorganic N levels, and vegetable yields, but also led to increased seed costs, pore water NO_3^- concentrations below the crop root zone, and reduced winter annual weed suppression. Yields and soil N levels were generally higher when PM was used, but the differences between PM and bare ground treatments varied between the two years due in part to differences in the magnitude and timing of precipitation events. Lower microbial biomass was observed under PM compared to bare ground 3 wk after plastic application, but cover crop and mulching treatments did not explain the majority of variation observed in patterns of carbon substrate utilization from Biolog-Ecoplate^{TM} data.

In a separate field experiment, we also investigated how fall planting dates influenced rye-vetch cover crop biomass quantity and quality in the spring, and evaluated whether staggering (delaying) rye seeding could improve vetch performance in mixtures. Treatments consisted of a two-way factorial of three vetch planting dates (late August, mid September, and late September) and three lengths of rye seeding stagger (co-seeded, short stagger, and long stagger). Later planting of co-seeded mixtures generally led to reduced total shoot biomass and lower proportions of vetch biomass, resulting in cover crop residues with less fixed N and a higher total C:N. For earlier planting dates, delaying rye seeding until vetch emergence (short stagger) increased vetch shoot biomass by 760 - 1,060 kg ha^{-1} (30-36 kg vetch N ha^{-1}) relative to co-seeding. Staggered seeding provided no benefit to vetch biomass at later planting dates, and delaying rye seeding until the vetch 3-4 leaf stage (long stagger) resulted in significant reductions in vetch winter survival compared with co-seeding.

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INTRODUCTION

INTRODUCTION

Cereal-legume cover crop mixtures can combine the unique functional strengths of the component species while also taking advantage of potential interspecific synergies. Winter annual mixtures composed of cereal rye (*Secale cereale* L.) and the legume hairy vetch (*Vicia villosa* Roth) have been well-studied for their potential to accumulate significant fixed N while also providing services more characteristic of rye, including more effective weed suppression, erosion control, scavenging of residual soil N, greater biomass production, and lower overall seed costs than a vetch monoculture. In addition, cereal-legume complementarity may contribute to advantages in rye-vetch mixtures, including greater resource-use efficiency, improved N availability for rye, and enhanced N fixation, light interception, and winter survival for vetch. Furthermore, the moderation of total residue quality in mixtures may alter patterns of net N mineralization following cover crop termination— alleviating challenges with N tie-up associated with rye residues, while potentially improving the synchrony of N release with crop demand relative to monoculture vetch. The winter hardiness and grower familiarity of the two species make them well suited for broad applicability in American agriculture, supporting the value of additional research to better understand species interactions and improve mixture management.

The services provided by rye-vetch cover crops and the extent to which possible facilitative benefits are realized depend on both environmental conditions and management factors that influence species interactions during cover crop growth, the final composition of mixture stands, and the decomposition of residues following cover crop termination. Evaluating cover crop performance requires an understanding of how the cover crops influence multiple components of the agroecosystem, from weed suppression to N availability, so that management decisions can take into account a farmer's principal goals and crop/soil management practices. The unifying objective of this work was to document the performance of rye-vetch mixtures relative to rye and vetch monocultures while leveraging our understanding of ecological principles to develop and evaluate strategies for improving rye-vetch mixture management within vegetable cropping systems.

Although generally considered less of a management priority than summer annual weeds, winter annual weeds can serve as alternative hosts for important pests and diseases, and can interfere directly with summer cash crops in reduced tillage systems. In **Chapter I**, we report results from two studies evaluating the effectiveness of rye and vetch cover crops in suppressing the biomass and density of components of winter annual weed communities, comparing the performance of a rye-vetch mixture with rye and vetch monocultures.

Recognizing that seeding rates were a logical tool for managing rye-vetch mixture composition, the next two chapters present results from a large systems-level experiment evaluating how the proportion of rye and vetch sown in a mixture influences species interactions and cover crop performance. **Chapter II** describes the effects of species proportions on the living cover crops, focusing on stand establishment, biomass productivity, vetch winter survival and N fixation, and tradeoffs among seed costs and cover crop services like fixed N accumulation and winter annual weed suppression. **Chapter III** follows with the impact of incorporated residues on vegetable production after cover crop termination, evaluating the interactive effects of rye-vetch residue quality across mixture proportions and the use of black polyethylene mulch on cash crop yields and quality, soil N dynamics, and microbial communities during production of both bell pepper (*Capsicum annuum* L.) and slicing cucumber

(*Cucumis sativus* L.).

Finally, inclement weather and late harvests of previous cash crops can force delays in planting of rye and vetch cover crops past recommended dates in some years, and fall establishment is an important determinant of spring biomass production. Furthermore, cereals tend to be the more-competitive component in cereal-legume mixtures, and depending on conditions, interference from rye can suppress vetch productivity in mixtures. **Chapter IV** presents results from an additional experiment that investigated how fall planting dates influenced rye-vetch cover crop biomass quantity and quality in the spring, and tested a unique approach for improving vetch productivity and N fixation in mixtures by staggering (delaying) the seeding of rye after vetch in the fall. This research also provided the opportunity to explore interactions between vetch developmental stage and the presence of rye on vetch overwinter survival in the Great Lakes region.

CHAPTER I

WINTER ANNUAL WEED SUPPRESSION IN RYE-VETCH COVER CROP MIXTURES

Winter Annual Weed Suppression in Rye-Vetch Cover Crop Mixtures

Zachary D. Hayden*, Daniel C. Brainard, Ben Henshaw, and Mathieu Ngouajio

Department of Horticulture, Michigan State University, Plant and Soil Sciences Building, 1066 Bogue St., East Lansing, MI 48824.

*Corresponding author (haydenza@msu.edu).

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ABSTRACT

Winter annual weeds can interfere directly with crops and serve as alternative hosts for important pests, particularly in reduced tillage systems. Field experiments were conducted on loamy sand soils at two sites in Holt, Michigan between 2008 and 2011 to evaluate the relative effects of cereal rye, hairy vetch, and rye-vetch mixture cover crops on the biomass and density of winter annual weed communities. All cover crop treatments significantly reduced total weed biomass compared to a no cover crop control, with suppression ranging from 71 - 91% for vetch, to 95 - 98% for rye. In all trials, the density of non-mustard family broadleaf weeds was either not suppressed or suppressed equally by all cover crop treatments. In contrast, the density of mustard family weed species was suppressed more by rye and rye-vetch mixtures than by vetch. Cover crops were more consistently suppressive of weed dry weight per plant than of weed density, with rye-containing cover crops generally more suppressive than vetch. Overall, rye was most effective at suppressing winter annual weeds; however, rye-vetch mixtures can match the level of control achieved by rye, in addition to providing a potential source of fixed nitrogen for subsequent cash crops.

INTRODUCTION

Crop production losses in the United States due to agricultural weeds may be as high as $33 billion annually, and American farmers spend an estimated $6 billion each year on herbicides, tillage, and cultivation for weed control (Liebman et al., 2001; Pimentel et al., 2005). Summer annual weed species are undoubtedly the greatest contributors to these costs, because they have life cycles that facilitate direct interference with most agronomic and vegetable crops. As a result, comparatively little research in summer annual cropping systems has focused on *winter annual* weeds, which establish prior to warm-season production windows and are typically controlled by pre-planting tillage or herbicide applications. However, due in part to increasing adoption of reduced tillage systems (CTIC, 2008), reductions in residual herbicide usage (Young, 2006), and trends toward milder winter temperatures in North America (Hayhoe et al., 2006), winter annuals may become a greater management priority in the future (Creech et al., 2008).

In the absence of an established forage or cover crop, winter annual weeds may provide valuable services in fields during the off-season, including erosion control and recycling of residual nutrients (Jordan and Vatovec, 2004). However, the significant challenges posed by increasingly prevalent winter annual populations likely outweigh any potential benefits. In no-till and other reduced tillage systems, winter annual and perennial weeds not controlled in the fall or spring can interfere directly with early-season or summer cash crops, and their previous establishment can make them highly competitive (Liebman et al., 2001; Brainard et al., 2012b). This is particularly problematic in organic reduced tillage systems, where the prohibition of synthetic herbicides leaves few options for effective pre-planting control. Failure to effectively control winter annual weeds will also increase soil seedbanks over time, which can lead to costly infestations in winter annual cash crops (e.g., winter cereals) grown later in a rotation (Mirsky et al., 2010). Moreover, many "winter annual" weed species are actually facultative winter annuals, which can germinate in both the fall and spring (Cici and Van Acker, 2009), and can reduce yields, interfere with harvest, and serve as costly contaminants in summer annual as well as winter annual crops. For example, mayweed chamomile (*Anthemis cotula* L.) is a weed in winter wheat and can also interfere with combining during pea (*Pisum sativum* L.) harvest in Washington and Northern Idaho (Ogg et al., 1993). Furthermore, as alternative hosts for economically important pests and diseases, overwintering weeds can also serve as "biological bridges" from one growing season to the next (Wisler and Norris, 2005; Norris and Kogan, 2005), fostering plant parasitic nematodes and plant pathogenic viruses, as well as their insect vectors in some cases (Creech et al., 2007; Duffus 1971; Groves et al., 2001). Since many common winter annuals belong to the *Brassicaceae* (mustard family), vegetable crops in this family may be particularly vulnerable to pests and diseases where winter annuals are not controlled (Schaad and Dianese, 1981; Chen et al., 2009).

Cover cropping is an important component of integrated approaches to weed management. Residues from winter cover crops are well-studied for their potential to suppress summer weed populations, particularly when maintained as a surface mulch in reduced tillage systems (Carrerra et al., 2004; Teasdale, 1996; Teasdale and Mohler, 1993). However, in addition to the provision of agroecosystem services like erosion control, organic matter addition, nutrient recycling, and nitrogen fixation, winter cover crops also have significant potential for managing winter annual weeds. Displacing winter annual populations with cover crop species that are unsuitable or less-suitable alternative hosts may reduce overwintering reservoirs of important pests and diseases, as well as draw down weed seedbanks over time. Cover crops are more likely to be adopted, however, if they can combine effective weed suppression with other desirable services.

Winter cover crops composed of mixtures of cereal and legume species, such as cereal rye and hairy vetch, have been studied for their potential to provide significant fixed nitrogen with greater weed suppression and lower overall seed costs than monoculture legumes (Brainard et al., 2012a). Mixtures are often more efficient than monocultures in the capture of light, water, and nutrients (Liebman and Dyck, 1993), which may contribute to greater biomass productivity in cover crop mixtures vs. monocultures, and suggests that mixtures may be more competitive with weeds. In practice, weed suppression in cereal-legume mixtures may be more closely related to the presence of the competitively dominant cereal species, rather than on the diversity of the mixture, *per se* (Liebman and Dyck, 1993). Accordingly, many studies suggest that cereal-legume mixtures often suppress weeds better than a monoculture of the legume, but less than or equivalent to a monoculture of the cereal (Akemo et al., 2000; Brainard et al., 2011; Brennan and Smith, 2005; Mohler and Liebman, 1987; Poggio, 2005).

While cereal rye and hairy vetch are well-researched both as winter cover crop monocultures and in mixture, few studies have documented the effects of rye-vetch mixtures on winter annual weeds. In monoculture, the notable ability of rye to suppress weeds, reduce nitrate leaching, and control erosion is often tempered by the high C:N ratio of its residues and the threat of subsequent yield losses due to nitrogen immobilization (McCracken et al., 1994; Shipley et al., 1992; Wagger et al., 1998). Vetch, on the other hand, can fix large amounts of nitrogen, but generally provides less effective weed suppression than rye (Clark, 2007; Clark et al., 2007; Mennan et al., 2009). In mixture, rye-vetch stand characteristics vary among studies, but total N release from mixture residues can approach the amount released from vetch monocultures, and total dry matter yields of rye-vetch cover crops can be greater than yields of either species in monoculture (Ranells and Wagger, 1996; Sainju et al., 2005). These qualities suggest that rye-vetch mixtures could be effective cover crops for controlling winter annual weeds, in balance with providing other important services.

The objective of this research was to evaluate the relative effects of rye, hairy vetch, and rye-vetch mixture cover crops on the biomass and density of winter annual weed communities.

MATERIALS AND METHODS

Two separate experiments were conducted at the Michigan State University (MSU) Horticulture Teaching and Research Center (HTRC) in Holt, MI (42°40” N, 84°28” W) at two sites within 2 km of each other (hereafter referred to as College Rd and Jolly Rd), both on Spinks loamy sand soil (sandy, mixed, mesic Lamellic Hapludalf). Initial soil chemical characteristics at College Rd included pH 6.6; CEC 7.7 cmol kg^{-1}; and P, K, and Mg levels of 117, 146, and 42 mg kg^{-1}, respectively. Initial soil chemical characteristics at Jolly Rd included pH 7.9; CEC 7.5 cmol kg^{-1}; and P, K, and Mg levels of 71, 83, and 266 mg kg^{-1}, respectively. Experiments at both sites were repeated for two seasons, alternating between adjacent fields, from 2008-2011. Summer cover crops of sorghum sudangrass (*Sorghum bicolor* x *S. bicolor* var. sudanense) were grown on each field prior to seeding winter cover crops in the fall.

Experiments at both sites investigated the effects of rye and hairy vetch cover crops on winter annual weed populations using a randomized complete block design with four replications. Cover crop treatments common to the College Rd and Jolly Rd experiments included: hairy vetch in monoculture (V), cereal rye in monoculture (R), a rye-vetch mixture (RV), and a control treatment with no winter cover crop (C). Vetch seeding rates were 42 and 45 kg ha^{-1} in monoculture and 21 and 22.5 kg ha^{-1} (50% of monoculture rate) in mixture at College Rd and Jolly Rd, respectively, while rye was sown at 94 and 125 kg ha^{-1} in monoculture and 47 and 62.5 kg ha^{-1} in mixture. “Variety not stated” (VNS) vetch seed from Oregon was used at both sites, while VNS rye from Minnesota and ‘Wheeler’ rye were used at the College Rd and Jolly Rd sites, respectively (Albert Lea Seed House, Albert Lea, MN). At College Rd, plot sizes were 6.7 x 8.5 m in 2009-2010 and 6.1 x 7.6 m in 2010-2011, while at Jolly Rd, plots were 3.8 x 18.3 m in 2008-2009 and 3.0 x 12.2 m in 2009-2010.

The dates of key field operations and data collection are summarized in Table 1. Previous sorghum sudangrass summer cover crops were flail mowed and incorporated at least 2 wk prior to winter cover crop seeding in all trials. Incorporation was accomplished with a rototiller at College Rd and with a moldboard plow and rotary spader at Jolly Rd in 2008 and 2009, respectively, followed by secondary tillage with a disk. The College Rd experiment was managed organically with no additional soil amendments applied. The Jolly Rd experiment was managed conventionally, and fall fertilizer applications included 224 kg ha^{-1} 19-19-19 (N-P-K) in 2008 and 45 kg ha^{-1} urea plus 67 kg ha^{-1} potash in 2009, in addition to 336 kg ha^{-1} elemental S applied in both years to lower soil pH. All amendments were broadcast and incorporated using a harrow. At College Rd, winter cover crop treatments were sown on September 1 in both 2010 and 2011. Rye and vetch seeds were broadcast by hand using a grid system to ensure uniformity, and then incorporated to a depth of roughly 5 cm using a field cultivator. Cover crops at Jolly Rd were drilled on September 2, 2008 using a Moore Unidrill no-till seeder (County Antrim, Northern Ireland). In 2009, rye was drilled using a John Deere 750 no-till grain drill (Deere and Company, Moline, IL) on September 4, while vetch was seeded using a Jang push seeder (JP-3, Chungbuk, South Korea). All vetch seed was inoculated with N-DURE *Rhizobium leguminosarum* inoculant prior to seeding (INTX Microbials LLC, Kentland, IN) at a rate of approximately 10 g inoculant kg^{-1} seed.

Cover crop densities and aboveground biomass were sampled in the spring from four 25 x 50 cm (0.125 m^2) quadrats in each plot at College Rd on May 10, 2010 and May 14, 2011, and from two 0.5 m^2 quadrats per plot at Jolly Rd on May 29, 2008 and May 27, 2009. At College Rd, biomass and densities of weed populations were sampled from those same quadrats at the time of cover crop sampling, and weeds sampled from C, V, RV, and R treatments were subsequently sorted by species in the laboratory. At Jolly Rd, weed populations were not sampled in 2009, and weed biomass was not collected in 2010. However, densities of weed populations were sampled on April 2, 2010 using two 0.5 m^2 quadrats in each plot. In addition, individuals of field pennycress were transplanted into Jolly Rd cover crop plots in order to evaluate the effects of cover crop treatments on the growth of this problematic mustard family weed while also controlling for variability in field population densities and possible confounding effects of cover crops on field pennycress emergence. A total of 12 (in 2008) and 5 (in 2009) field pennycress individuals that emerged following pre-planting tillage (growing outside but adjacent to the experimental area) were transplanted into two locations in each plot on September 29, 2008 and October 16, 2009. Field pennycress rosettes were 2.5 to 5 cm in diameter at the time of transplanting, and were transplanted at a spacing of 30 cm within rows. Following overwintering and spring growth periods, the number of surviving transplants was recorded and their biomass sampled on May 13, 2009 and May 7, 2010. Average survival of transplants was 82 and 99 percent in 2009 and 2010, respectively, and cover crop treatment did not significantly affect transplant survival in either year (data not shown). For both trials, all cover crop and weed biomass samples were dried to constant weight at 38 °C.

Weeds were identified to species, and the data were then grouped into the following categories for analysis: mustard family weeds, other broadleaves, grasses, and total weeds. The fixed effect of rye-vetch cover crop treatment on all variables was evaluated using the Proc MIXED procedure in SAS (Version 9.2, SAS Institute, Cary, NC) with block (replicate) included as a random effect in the model. Data were natural-log or square-root transformed as necessary to meet ANOVA assumptions of normality and homogeneity of variances. Data were analyzed separately by trial location and year due to differences in management and the composition of weed communities among the experimental fields. Where the global *F* test was significant ($p < 0.05$), treatment means were separated using Fisher's protected LSD.

RESULTS AND DISCUSSION

Figure 1 presents aboveground biomass produced by rye and vetch in cover crop monoculture and mixture treatments in the College Rd and Jolly Rd experiments. Average vetch biomass in monoculture varied between 356 and 563 g m^{-2} across sites and years, while rye biomass ranged from 330 to 587 g m^{-2}, both typical ranges for these cover crop species in the Great Lakes region (Clark, 2007; Sarrantonio, 1994). Seeding dates were similar across trials and years, with cover crops at Jolly Rd seeded 3 to 4 days later in September than at College Rd (Table 1). However, cover crops were sampled on average 2 weeks later in May at Jolly Rd than at College Rd (Table 1), resulting in a longer period of cover crop growth for the Jolly Rd experiment. Total rye-vetch mixture biomass tended to be dominated by rye at Jolly Rd (60 and 69 percent rye by dry weight in 2009 and 2010, respectively), while the proportions of rye to vetch biomass in mixtures at College Rd were closer to 1:1 (46 and 55 percent rye in 2010 and 2011, respectively). Higher rye seeding rates, higher soil N fertility as a result of fall fertilization, and slightly later fall planting dates at Jolly Rd could all favor rye over vetch in mixture (Clark et al., 2007; Jannink et al., 1997; Jensen, 1996; Shipley et al., 1992).

Total aboveground biomass production among the cover crop treatments did not differ significantly at College Rd in 2010 or 2011 (Figure 1A). In contrast, total biomass production in the rye-vetch mixture was significantly greater than either monoculture at Jolly Rd in 2009, and rye and rye-vetch mixture treatments produced similar biomass in 2010, both significantly greater than the vetch monoculture (Figure 1B). The large amounts of total biomass produced by the rye-vetch mixtures relative to the monoculture treatments at Jolly Rd were driven largely by the rye component; although seeded at half the monoculture rate, rye biomass in mixture was 94 and 74 percent of that produced in monoculture in 2009 and 2010, respectively.

Weed community composition in experimental fields varied with location and year. Table 2 lists dominant weed species at College Rd and Jolly Rd in 2010 and 2011. Collectively, mustard family species made up 52 to 55 percent of the total weed density in control treatments at College Rd in 2010 and 2011, respectively, but accounted for only 22 to 29 percent of the total weed biomass, reflecting their relatively small size in the spring compared to other winter annual broadleaf and grass species present in the fields. Common chickweed was an abundant broadleaf weed in all years at both sites. Other broadleaf species present at College Rd included henbit in 2010 and corn chamomile (*Anthemis arvensis* L.) in 2011, while at Jolly Rd, broadleaf weeds other than chickweed were not identified to the species level. No grass weeds were abundant at College Rd in 2011, and annual bluegrass (*Poa annua* L.) was the only grass species abundant in 2010; however, annual bluegrass distribution across the field tended to be more variable than other weed species at College Rd, likely reducing the power of statistical inference regarding cover crop effects on annual bluegrass. At Jolly Rd, grasses comprised nearly 40 percent of the total weed community by density, but data was not collected to the species level. Total weed pressure in experimental fields, based on control treatments, was lowest at Jolly Rd in 2010 (139 plants m^{-2}), followed by College Rd in 2010 (431 plants m^{-2}), and highest at College Rd in 2011 (1120 plants m^{-2}).

In both years at College Rd, all winter cover crop treatments reduced weed biomass (dry weight, g m^{-2}) compared to the no cover crop control, although the reduction was not significant for annual bluegrass (Table 3A). In 2010, suppression of total weed biomass ranged from 91 percent in vetch monoculture to 95 and 98 percent in the rye-vetch mixture and rye monoculture, respectively. Rye provided significantly greater suppression of weed biomass than vetch monoculture for mustard species, but not for other broadleaf species (mostly common chickweed). In 2011, total weed biomass suppression was 71 percent in vetch monoculture, compared to 94 and 95 percent in the rye-vetch mixture and rye monoculture, respectively. Rye suppressed both mustard and other broadleaf species significantly more than vetch. In both years, the rye-vetch mixture provided suppression of weed biomass equivalent to that of the rye monoculture.

Our results are generally consistent with previous studies showing greater weed suppressive ability of cereal species compared to legumes (Brainard et al., 2011; Ofori and Stern, 1987), which may be a result of more effective resource competition or allelopathy (Barnes and Putnam, 1986). Suppression by rye-containing cover crops was also less variable between 2010 and 2011 compared to suppression by vetch in monoculture, suggesting that the suppressive ability of rye may be more robust in the face of year-to-year variability in environmental conditions than vetch. We speculate that cooler spring temperatures in 2011 (data not shown) may have limited vetch growth relative to both weeds and rye that year, resulting in less effective weed suppression by vetch, and perhaps a greater proportion of rye in the rye-vetch mixture (Figure 1A). The relative proportion of the component species in cereal-legume mixtures is likely a key determinant of mixture performance. In a study of rye-pea cover crop mixtures, Akemo et al. (2000) observed that weed biomass decreased with increasing proportion of rye in the mixture, despite decreasing total cover crop biomass. Therefore, the weed suppressiveness of cereal-legume mixtures is likely closely related to the relative proportion of the cereal species.

Although total weed biomass in the field is a relevant measure of actual weed pressure, it is also a function of both the density of weeds present and their average individual biomass production. Separately evaluating effects on weed density and weed dry weight per plant can provide greater insight into the potential mechanisms behind suppression by cover crops. For example, Kumar et al. (2008) found that both the inhibitory effects of cover crops on weeds (including corn chamomile and shepherd's-purse) and the mechanisms responsible for these effects (nitrogen and fungal pathogens) differed for emergence and growth life stages.

At College Rd, all cover crops reduced the density of weeds (with the exception of annual bluegrass) compared to the control in 2010 (Table 3B). Rye was significantly more suppressive of mustard weed density than both the vetch monoculture and the rye-vetch mixture, but the density of other broadleaf species was reduced equally by all cover crop treatments. In 2011, mustard weed density was suppressed by all cover crop treatments, with the rye-vetch mixture and rye monoculture providing equivalent control, both greater than the vetch monoculture. The density of other broadleaf species, however, was not significantly affected by any of the cover crops.

Comparable results were obtained at Jolly Rd in 2010, where the rye-vetch mixture and rye monoculture both significantly reduced mustard and grass species density compared to the control and vetch monoculture, but did not have an effect on other broadleaf density (Table 4). In contrast to the results from College Rd, however, the vetch monoculture did not reduce weed densities compared to the control at Jolly Rd. This difference may in part be attributable to the early timing of weed sampling at Jolly Rd (April 2, compared to mid-May at College Rd). Vetch often doesn't achieve more than 30 percent ground cover in the fall, and produces the majority of its biomass during the warmest months of the spring (Shipley et al., 1992; Teasdale et al., 2004).

Cover crops that can close their canopy earlier in the season are often better at suppressing weeds (Brennan et al., 2011), and vetch biomass at the time weeds were sampled was likely close to only half the amount observed in May. Rye, on the other hand, generally provides more extensive soil cover in the fall (Boyd et al., 2009), in addition to its capacity to deplete soil moisture and nutrients (particularly N) early in the season (Brainard et al., 2012c; Shipley et al., 1992). The potential for rye to effectively interfere with weeds earlier than vetch may explain why the rye monoculture and rye-vetch mixture (69 percent rye biomass) at Jolly Rd still exhibited suppression of mustard and grass weed density at the earlier sampling date. Furthermore, the suppression of grass species by rye-containing cover crops at Jolly Rd suggests that the lack of significant cover crop effects for annual bluegrass at College Rd is not reflective of grasses in general.

Interestingly, in all trials, the density of non-mustard broadleaf species (common chickweed, corn chamomile, and henbit) was suppressed equally by all cover crop treatments, while that of mustards was suppressed more by rye than by vetch (Tables 3B and 4). In a broad sense, cover crops reduce weed density through a combination of inhibiting seed germination or emergence, and promoting post-emergence mortality through interference or environmental effects. Although we can only speculate, the greater suppression of mustards by rye compared to vetch may have been due in part to mustard sensitivity to nitrogen depletion or shade. Many members of the mustard family are highly sensitive to soil N levels in both their germination (Kumar et al., 2008) and growth (Blackshaw et al., 2003) responses, so reductions in soil N due to rye may have weakened these species and contributed to their mortality. Conversely, several of the non-mustard species present in our trials, including common chickweed and corn chamomile, are known to be relatively shade tolerant (Turkington et al., 1980) and less sensitive to nitrogen depletion (Blackshaw et al., 2003; Kumar et al., 2008), and thus perhaps better able to withstand competition from rye and vetch cover crops than mustards. In a study comparing the effects of twelve weed species on kale and sugar beets under different levels of nitrogen, Welbank (1963) found that the competitive effect of chickweed—one of the primary non-mustard broadleaf species in our trials—was relatively unaffected by nitrogen level. The greater inhibitory effects of rye relative to vetch are likely to be most pronounced for weed species that germinate and grow primarily in the fall when rye growth typically exceeds that of hairy vetch. Conversely, for facultative winter annuals that have significant flushes of emergence in the spring—potentially including common chickweed, henbit and corn chamomile, but also shepherd's purse and field pennycress (Cici and Van Acker, 2009; Kay, 1971)—fewer differences in density suppression due to cover crops might be expected since growth of rye and vetch is more similar at that time.

Cover crop treatments, particularly those containing rye, were more consistently suppressive of weed biomass production (as measured by dry weight per plant) than of weed density. All cover crop treatments significantly reduced the dry weight per plant of other broadleaf species compared to the control in trials at College Rd, with rye monoculture consistently providing the highest level of suppression (Table 3C). The rye-vetch mixture equaled rye in 2011, but not in 2010. Mustard dry weight per plant was not significantly lower in vetch monoculture than in the control in 2011, but otherwise, all cover crop treatments suppressed the growth of mustard species as well. Rye monoculture and the rye-vetch mixture provided equivalent suppression of mustards in both years at College Rd, a trend that was also present for annual bluegrass in 2010, though not significant. Transplanted field pennycress responded similarly to cover crop treatments at Jolly Rd in 2009 and 2010 (Figure 2). Both the rye monoculture and the rye-vetch mixture provided over 95 percent suppression of field pennycress dry weight per plant in each year, compared to less than 70 percent suppression by vetch monoculture. Cereals are generally better competitors than legumes (Ofori and Stern, 1987), and as our results support, this quality tends to be manifest more in the suppression of weed growth than weed density (Boyd et al., 2009).

This research supports that winter cover crops composed of rye and/or vetch can significantly suppress winter annual weeds. Reductions in the biomass of winter annuals present in fields may decrease the size of overwintering populations of important pests and diseases, which could in turn lower disease pressure during the following season. However, hairy vetch is also known to be a reproductive host for several species of plant parasitic nematodes, so growing vetch before susceptible crops should be avoided (Aarssen et al., 1986; Rich et al., 2009; Timper et al., 2006). Despite suppressing weed biomass production across taxonomic groups, the cover crops failed to consistently reduce the density of non-mustard broadleaf weeds, including common chickweed, henbit, and corn chamomile. Particularly in organic reduced tillage systems, dense populations of these low-growing species could become a problem in subsequent crops if they are able to survive mechanical kill of the cover crops. The persistence of these weeds highlights the importance of integrating winter cover crops with other weed management strategies to provide more complete control and avoid increases in problematic weeds over time (Liebman and Gallandt, 1997).

Overall, cereal rye was the most effective weed suppressor. Therefore, where winter annual weed control is a primary objective, rye would likely be the most effective and inexpensive cover crop option. However, our results demonstrate that rye-vetch mixtures can match the level of suppression achieved by rye monoculture, in addition to providing a potential source of fixed nitrogen—a benefit particularly relevant for organic production. The weed suppressiveness of the mixtures is likely tied to relative species composition, and mixtures containing less than 50 percent rye biomass may sacrifice winter annual weed control. Additional research is needed to relate rye-vetch seeding rates to resulting stand characteristics, and to investigate how species proportions in rye-vetch mixtures influence the provision of other agroecosystem services.

APPENDIX

Table 1.1. Dates of key field operations and data collection at College Rd and Jolly Rd.

	College Rd		Jolly Rd	
Activity	2009-10	2010-11	2008-09	2009-10
Cover crops seeded	1 Sept	1 Sept	4 Sept	5 Sept
THLAR† transplanted	—	—	29 Sept	16 Oct
Ambient weeds sampled	10 May	14 May	—	2 Apr
THLAR biomass sampled	—	—	13 May	7 May
Cover crop biomass sampled	10 May	14 May	29 May	27 May

† Field pennycress, *Thlaspi arvense* L.

Table 1.2. Dominant weed species present at College Rd and Jolly Rd in 2010 and 2011, including percent composition based on density and biomass calculated from control treatments.

	% Density			% Biomass	
	College Rd		Jolly Rd	College Rd	
Species	2010	2011	2010	2010	2011
	%				
Mustard family species	**52**	**55**	**30**	**22**	**29**
Field pepperweed - *Lepidium campestre* (L.) R. Br.	12	29	—	6	20
Mouse-ear cress - *Arabidopsis thaliana* (L.) Heynh.	8	21	—	5	5
Shepherd's purse - *Capsella bursa-pastoris* (L.) Medik.	20	3	7	10	4
Spring whitlowgrass - *Draba verna* L.	11	2	—	2	< 1
Field pennycress - *Thlaspi arvense* L.	—	—	20	—	—
Hoary alyssum - *Berteroa incana* (L.) DC.	—	—	3	—	—
Other broadleaf species	**30**	**45**	**30**	**61**	**71**
Common chickweed - *Stellaria media* (L.) Vill.	22	20	12	51	18
Henbit - *Lamium amplexicaule* L.	7	—	—	9	—
Corn chamomile - *Anthemis arvensis* L.	—	25	—	—	53
Other†	—	—	18	—	—
Grass species	**19**	—	**39‡**	**18**	—
Annual bluegrass - *Poa annua* L.	19	—	—	18	—

† With the exception of *Stellaria media*, "Other broadleaf species" data were not collected to the species level at Jolly Rd.

‡ Data for "Grass species" were not collected to the species level at Jolly Rd.

Table 1.3. Effect of cover crop treatment on (A) total shoot dry weight, (B) density, and (C) shoot dry weight per plant of dominant winter annual weeds present in the College Rd experiment in 2010 and 2011.

	Mustards		Other broadleaves		Grasses†		Total	
Cover Crop	2010	2011	2010	2011	2010	2011	2010	2011
A. Dry weight	g m^{-2}							
None	25.9 a‡	45.5 a	72.3 a	110.8 a	21.1 a	—	119.3 a	156.3 a
Vetch	1.5 b	19.2 b	3.3 b	27.0 b	5.5 a	—	10.3 b	46.2 b
Rye-Vetch	0.6 bc	2.6 c	3.2 b	6.6 c	2.6 a	—	6.3 bc	9.2 c
Rye	0.1 c	1.9 c	1.4 b	6.7 c	0.9 a	—	2.4 c	8.6 c
B. Density	plants m^{-2}							
None	223 a	616 a	128 a	504 a	81 a	—	431 a	1120 a
Vetch	38 b	275 b	14 b	226 a	56 a	—	108 b	500 b
Rye-Vetch	37 b	138 c	15 b	185 a	72 a	—	123 b	323 b
Rye	6 c	105 c	17 b	259 a	25 a	—	48 b	364 b
C. Dry weight per plant	mg plant^{-1}							
None	118.1 a	75.8 a	589.5 a	235.1 a	195.8 a	—	279.3 a	137.5 a
Vetch	37.0 b	75.1 a	225.5 b	128.7 b	129.2 a	—	102.0 b	96.8 a
Rye-Vetch	14.1 c	16.7 b	217.1 b	34.9 c	71.9 a	—	85.0 bc	26.8 b
Rye	18.1 c	15.4 b	67.9 c	24.7 c	34.0 a	—	46.5 c	22.2 b

† Annual bluegrass (*Poa annua* L.) was the only abundant grass species present in 2010.

‡ For a given measurement, means within a column followed by the same letter are not significantly different (α=0.05).

Table 1.4. Effect of cover crop treatment on density of dominant winter annual weeds present in the Jolly Rd experiment in 2010†.

Cover Crop	Mustards	Other broadleaves	Grasses	Total
	plants m^{-2}			
None	42 a‡	42 a	55 a	139 ab
Vetch	42 a	47 a	70 a	159 a
Rye-Vetch	11 b	50 a	5 b	66 c
Rye	11 b	50 a	11 b	72 bc

† Density data do not include transplanted field pennycress (Figure 1.2).

‡ Means within a column followed by the same letter are not significantly different (α=0.05).

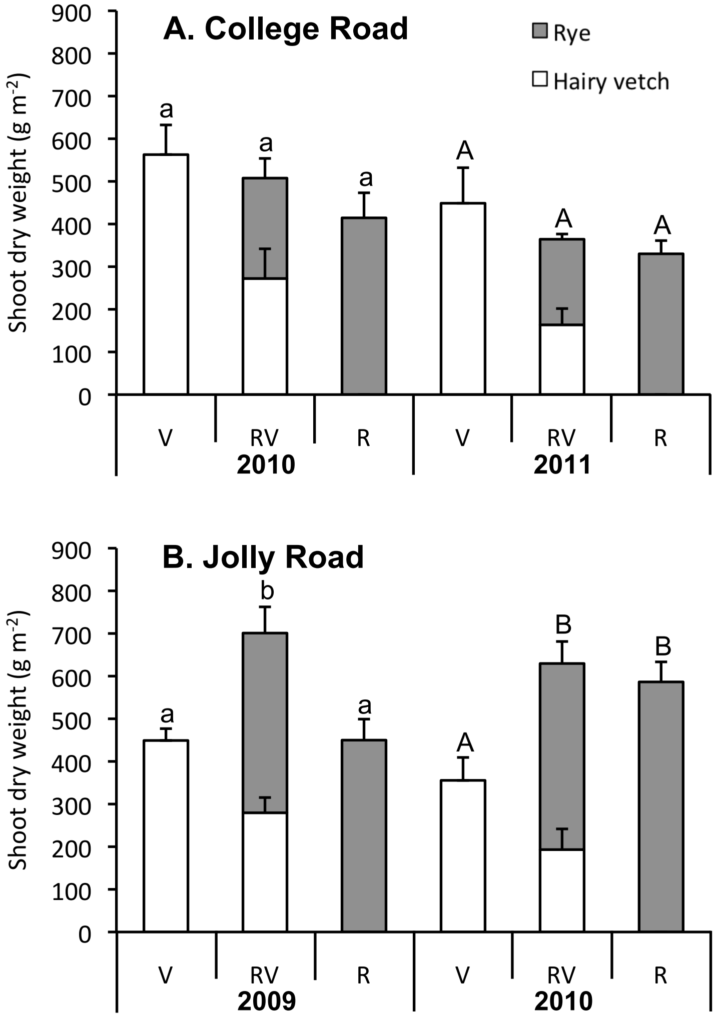

Figure 1.1. Mean (±SE) shoot dry weight of vetch (V), rye-vetch mixture (RV), and rye (R) winter cover crops in the spring at (A) College Rd in 2010 and 2011, and (B) Jolly Rd in 2009 and 2010. For RV treatments, the height of the stacked bar represents total cover crop biomass (rye+vetch), while depicted standard errors correspond to the mean biomass of each component species in the mixture. Within a given site and year, total cover crop biomass is not significantly different for treatments labeled with the same letter (α=0.05).

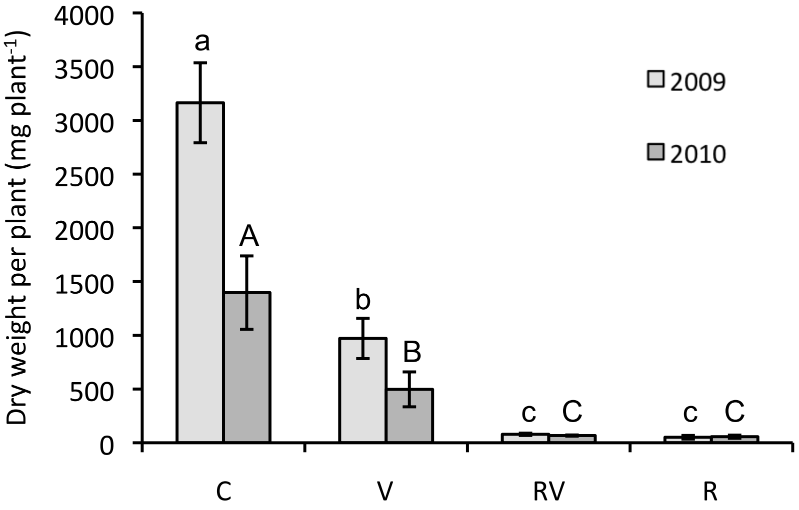

Figure 1.2. Mean (±SE) dry weight per plant of transplanted field pennycress (*Thlaspi arvense* L.) in control (C), vetch (V), rye-vetch mixture (RV), and rye (R) treatments at Jolly Rd in 2009 and 2010. Within a given year, means labeled with the same letter are not significantly different (α=0.05).

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CHAPTER II

RYE-VETCH MIXTURE PROPORTION TRADEOFFS: COVER CROP PRODUCTIVITY, NITROGEN ACCUMULATION, AND WEED SUPPRESSION

Rye-vetch mixture proportion tradeoffs: Cover crop productivity, nitrogen accumulation, and weed suppression

Zachary D. Hayden*, Mathieu Ngouajio, and Daniel C. Brainard

Department of Horticulture, Michigan State University, Plant and Soil Sciences Building, 1066 Bogue St., East Lansing, MI 48824.

*Corresponding author (haydenza@msu.edu).

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ABSTRACT

Cereal-legume cover crop mixtures have the potential to combine the unique strengths of the component species while taking advantage of interspecific synergies. However, the relative proportion of each species in mixture is likely to influence species interactions and entail important tradeoffs in cover crop performance. The objective of this study was to evaluate how the relative proportions of cereal rye (*Secale cereale* L.) and hairy vetch (*Vicia villosa* Roth) sown in mixtures influenced cover crop biomass production, winter annual weed suppression, vetch winter survival, and vetch N-fixation as measured by the ^{15}N natural abundance method. Following a replacement series design, treatments consisted of a gradient of seven rye-vetch mixture proportions ranging from 100% vetch to 100% rye. Density and biomass composition in the mixtures were highly correlated with rye and vetch seeding rates, with little evidence of substantial interspecific interference. Total shoot biomass in all mixtures was equal to or greater than that of either monoculture, but no differences were detected in vetch winter survival or the efficiency of N fixation. Changing the proportions of rye and vetch in the mixtures resulted in tradeoffs related to nitrogen and weed management goals. Increasing vetch in mixtures led to greater fixed N accumulation, but also increased seed costs and reduced winter annual weed suppression. A greater understanding of how rye-vetch mixture proportions influence cover crop performance can support more-informed decision-making regarding cover crop selection and mixture seeding rates.

INTRODUCTION

The grass-legume association has long been of interest to researchers working in both natural and managed ecosystems. Mixtures of plants with complementary functional traits can benefit from more efficient capture of light, water, and nutrients compared with monocultures, and when interspecific interference is low, grass-legume mixtures can also benefit from potential facilitative interactions (Vandermeer, 1992). In particular, reductions in soil inorganic N by the grass can increase legume nodulation and N fixation (Izaurralde et al., 1992; Streeter, 1988), while the presence of the legume can correspondingly increase N availability to the grass, through biomass turnover or possible direct N transfer (Eaglesham et al., 1981; Ledgard and Steele, 1992; van Kesse et al., 1985). Light interception by vining legumes can also be enhanced by the opportunity to climb into the upright canopy of a companion grass or cereal (Keating and Carberry, 1993). By reducing frost heaving or buffering temperature extremes at the soil surface through reduced air movement or increased snow cover retention, the presence of a hardier grass species may also improve legume winter survival in northern climates (Jannink et al., 1997; Smith, 1975).

Cereal rye and the legume hairy vetch are widely studied as winter annual cover crop species, in part because their cold hardiness makes them suitable for production across a broad geographic range, including the northern United States (Clark, 2007). Rye is a rapid-growing, N-responsive grass that has demonstrated substantial capacity to provide erosion control, conservation of residual soil N, and weed suppression—both as a living cover crop and as a thick surface mulch in reduced or no-tillage production systems (Clark, 2007; Ditsch et al., 1993; Peachey et al., 2004). These strengths, in addition to inexpensive and widely available seed, have made rye one of the most commonly grown cover crops in the United States. However, while rye residues can build soil organic matter, they are not a significant source of available N for subsequent cash crops, and depending on the stage at which rye is terminated, residue incorporation can result in net N immobilization (Sainju et al., 2000; Clark et al., 2007a). In contrast, hairy vetch is a legume known for its ability to accumulate considerable amounts of N (up to 190 kg ha^{-1}, even in northern temperate climates), much of which is rapidly available to cash crops in the first season after termination (Ranells and Wagger, 1996; Teasdale, 2004). However, vetch seed is relatively expensive, and its slower growth and readily decomposable residues generally make it less effective than rye at soil conservation and weed suppression, both before and after termination (Mohler and Teasdale, 1993; McCracken et al., 1994; Mennan et al., 2009). Vetch can also become a problematic weed in winter annual and perennial cropping systems due to hard seed (Aarssen et al., 1986), and in reduced tillage systems as a result of incomplete kill and regrowth (Creamer and Dabney, 2002; Mischler et al., 2010).

Rye-vetch mixtures are often proposed as a way to combine the strengths of the two species while moderating their individual weaknesses, in addition to taking advantage of potential synergies that arise from the grass-legume association. In practice, the characteristics of cereal-legume cover crop mixtures tend to reflect the relative proportions of each species present, with the qualities of a given species becoming more prominent with greater proportion in the mixture. Accordingly, many studies find that qualities such as weed suppression, total dry matter production, and N availability in cereal-legume mixtures tend to be intermediate to the corresponding monocultures (e.g., Ranells and Wagger, 1997c; Akemo et al., 2000b; Hauggaard-Nielsen et al., 2003; Clark et al., 2007b; Benincasa et al., 2010; Dordas and Lithourgidis, 2011). However, studies have also demonstrated that mixtures of vetch with a cereal can accumulate more total dry matter and N than either species in monoculture (Clark et al., 1994; Sainju et al., 2005), exhibit more efficient N accumulation by the components (Tosti et al., 2010), mineralize N at rates approaching that of a vetch monoculture (Ranells and Wagger, 1996), and provide weed suppression equivalent to that of a cereal monoculture (Teasdale and Abdul-Baki, 1998;

Hayden et al., 2012). In addition, because legumes are generally sown at reduced rates in mixtures, overall seed costs for cereal-legume mixtures tend to be lower than for monoculture legumes.

The performance of cereal-legume cover crop mixtures relative to monocultures depends heavily on both environmental conditions and management decisions that influence the competitive balance between species and final stand characteristics. Cereals, with their extensive root systems and higher relative growth rates, tend to be stronger competitors for below-ground resources than legumes (Mariotti et al., 2009). Therefore, when resources other than N, including moisture, are limiting, cereals are likely to suppress legumes in mixtures (Ofori and Stern, 1987). However, N limitation often shifts the competitive balance to benefit legumes in mixtures, while high N fertility generally favors non-legumes (Jensen, 1996), including rye in rye-vetch mixtures (Shipley et al., 1992; Clark et al., 2007a). In addition, rye can germinate and grow at cooler temperatures than vetch (Nuttonson, 1958; Teasdale et al., 2004), which contributes to its ability to establish later in the fall and accumulate biomass faster in the spring (Shipley et al., 1992). As a result, colder weather, later planting dates, and earlier cover crop termination are all likely to shift the balance toward rye over vetch in mixtures.

Considering the importance of relative stand composition in influencing the properties of cereal-legume mixtures, seeding rates are a logical tool for managing mixture performance. The fundamental importance of component species density is recognized and often thoroughly investigated in studies of plant competition (Firbank and Watkinson, 1985) and agronomic intercropping (Willey, 1979b). However, few experiments systematically evaluate the influence of sown species proportions on the characteristics of cover crop mixtures (Akemo et al., 2000b; Karpenstein-Machan and Stuelpnagel, 2000; Tosti et al., 2010), and most draw conclusions from only a single mixture, often where the species are sown at 50% of their recommended monoculture seeding rates. When multiple seeding rates of cereal-legume cover crops have been evaluated side-by-side, the differences in stand characteristics among the mixtures are often enough to significantly influence the provision of agroecosystem services by the cover crops, such as weed suppression, soil N fertility, and effects on subsequent crop yields (Clark et al., 1994; Akemo et al., 2000a, 2000b; Tosti et al., 2012).

Replacement series experiments, in which treatments consist of a monoculture of each species and a gradient of species mixtures, provide one approach for investigating species proportions in cover crop mixtures (Jolliffe, 2000). Replacement series are widely used in the study of plant competition, and are commonly applied within agricultural contexts to evaluate crop-weed interactions and intercrop productivity. While total plant density is held constant in a traditional replacement series, a replacement series is *proportional* when the relevant monoculture seeding rates differ for the component species, and total density therefore varies across the mixtures. The limitations to inference from such designs have been thoroughly reviewed (Snaydon, 1991; Jolliffe, 2000), but over-interpretation remains common in the literature. In particular, the densities of component species are confounded with each other and with total plant density in proportional designs. Therefore, without data on density-dependent yield responses for each species (Firbank and Watson, 1985), the design is not suited for drawing definitive conclusions regarding relative competitive abilities or the causal mechanisms contributing to species performance in mixtures. Despite these caveats, proportional replacement series are still effective in providing applied insights that can inform improvements in cover crop mixture management.

A greater understanding of how species proportions influence mixture stands is an important step toward maximizing potential benefits from rye-vetch cover crop mixtures to better serve specific goals within cropping systems. Therefore, the objective of this study was to evaluate how the relative proportions of rye and vetch sown in a winter annual cover crop mixture influenced cover crop stand characteristics and performance with respect to establishment, biomass productivity, winter annual weed suppression, and N accumulation, fixation, and costs relative to rye and vetch monocultures.

MATERIALS AND METHODS

Site Description

The study was conducted at the Michigan State University (MSU) Horticulture Teaching and Research Center in Holt, MI (42°40” N, 84°28” W) over the course of two seasons, alternating between adjacent fields. Both fields were on level terrain and had a Spinks loamy sand soil (sandy, mixed, mesic Lamellic Hapludalf). In each field, a summer cover crop of sorghum sudangrass (*Sorghum bicolor* x *S. bicolor* var. Sudanense) was flail mowed and incorporated using a rototiller at least 2 wk prior to sowing of winter cover crops in the fall. Initial soil chemical characteristics for the fields in 2009 and 2010 were similar, and included on average pH 6.6; CEC 7.1 cmol kg^{-1}; and P (Bray P1 extract), K, and Mg levels of 133, 145, and 47 mg kg^{-1}, respectively. The fields were managed according to National Organic Program guidelines (Agricultural Marketing Service, 2013), and had been in organic transition with a cropping history of warm-season vegetables under conventional tillage since 2008. No fertilizers or soil amendments were applied in either year of this study.

Treatment and Experimental Design

Following a proportional replacement series design (Snaydon, 1991; Jolliffe, 2000), winter cover crop treatments included rye and vetch sown in monocultures at rates of 94 and 42 kg ha^{-1}, respectively, and a gradient of species mixtures containing the following sown proportions of rye:vetch relative to their monoculture seeding rates— 83:17, 67:33, 50:50, 33:67, 17:83. Rye and vetch seeding rates and seed cost estimates for the cover crop treatments are listed in Table 1. A no-cover-crop control was also included for comparison. Experimental plots were 6.7 by 8.5 m in 2009-2010 and 6.1 by 7.6 m in 2010-2011, arranged in a randomized complete block design with four replications.

Field Management and Data Collection

The dates of key field activities and data collection are summarized in Table 2. Rye and vetch cover crops were broadcast sown by hand using a grid system that divided plots into quarters to help ensure uniformity, and then incorporated to a depth of about 5 cm using a field cultivator (Perfecta II, Unverferth Manufacturing Co. Inc., Kalida, OH). Variety not stated (VNS) vetch seed grown in Oregon and VNS rye seed grown in Minnesota were used in both years (Albert Lea Seed House, Albert Lea, MN). Vetch seed was inoculated with N-DURE *Rhizobium leguminosarum* inoculant (INTX Microbials LLC, Kentland, IN) at a rate of approximately 10 g inoculant kg^{-1} seed.

Rye and vetch plant densities were assessed in the fall from four 25- by 50-cm (0.125 m^{-2}) quadrats established in each plot. Spring cover crop densities, shoot biomass, and total weed biomass were sampled later from the same quadrats, shortly before mowing and incorporation. At the time of spring sampling in both years, vetch was pre-bloom and rye was between ear-emergence and anthesis. Potential differences in vetch winter survival across treatments were evaluated by calculating the percentage change in vetch population density between fall and spring sampling dates.

Shortly before sampling cover crop biomass in the spring, photosynthetically active radiation (PAR) was measured both above the canopy and at the soil surface at four locations in each plot at least 1 m from a plot edge. Measurements were taken midday under clear skies with a 70-cm long quantum flux sensor (MQ-301, Apogee Instruments Inc., Logan, UT), and used to calculate the percentage of PAR penetrating the cover crop canopies.

Rye, vetch, and total weed biomass samples were dried to a constant weight at 38°C prior to taking dry weights. Cover crop dry weights per plant were calculated using rye and vetch spring densities. Cover crop shoot biomass samples were then ground to pass through a 1 mm screen, and subsamples were submitted to Midwest Laboratories Inc. (Omaha, NE) for analysis of percent total C and N using a LECO TruSpec elemental analyzer (LECO Corp., St. Joseph, MI). Subsamples were also submitted to the University of California-Davis Stable Isotope Facility (SIF) for analysis of δ^{15}N using a PDZ Europa ANCA-GSL elemental analyzer interfaced to a PDZ Europa 20-20 continuous flow isotope ratio mass spectrometer (Sercon Ltd., Cheshire, UK).

Corrections for Soil Contamination on Vetch Shoots

Near-saturated soil moisture conditions at the time of cover crop sampling in 2011 resulted in considerable amounts of soil adhering to vetch shoots that could not be completely removed from samples prior to drying and analysis. Therefore, vetch dry weight and percent N data were corrected for the presence of soil contamination following equations found in Hunt et al. (1999). Briefly, the fraction by weight of vetch tissue (F_v) in a contaminated sample (vetch tissue plus contaminating soil) was estimated using the following equation:

[1] $F_v = (A_0 - A_s) / (A_0 - A_v)$

where A_s is the ash fraction of the contaminated sample, A_v is the ash fraction of uncontaminated vetch tissue, and A_0 is the ash fraction of the contaminating soil. The ash fraction of uncontaminated vetch tissue (A_v) was estimated as 0.2, based on vetch samples taken from 2010 and 2011 that were not visually contaminated with soil and did not exhibit outlier $\delta^{15}N$ values. The ash fraction of the contaminating soil (A_o) was estimated from soil sampled shortly before cover crop sampling in 2011, and sieved to remove large particles that were less likely to have adhered to vetch tissue. Ash fractions were determined for the soil and for all vetch tissue samples using a muffle furnace (2 h drying period at 105°C, followed by 4 h at 500°C).

The corrected vetch tissue dry weight (M_v) was then derived from the dry weight of the contaminated sample (M_s) using Eq. 2:

[2] $M_v = M_s \text{ x } F_v$

Finally, the concentration of N in vetch tissue (N_v) was estimated from the sample N concentration (N_s) and the concentration of N in the contaminating soil (N_0) by:

[3] $N_v = (N_s - N_0 \text{ x } (1 - F_v)) / F_v$

For consistency, the correction was applied to vetch samples from both years of the study; however, it did not significantly affect the 2010 data due to the lack of soil contamination in that year. No correction was necessary for rye data in either year because the upright nature of the shoots minimized the risk of soil contamination during sampling.

Land Equivalent Ratios

Partial and total land equivalent ratios (LER) were calculated for each mixture on the basis of rye and vetch dry weight (de Wit and Van der Bergh, 1965; Willey, 1979a). Partial LERs (relative yields) for rye (L_r) and vetch (L_v) were calculated as the ratio of the dry weight of each species in mixture to its dry weight in monoculture. The total LER for mixtures was then calculated as the sum of the partial LERs. Relative yields greater than relative sown proportions indicate greater biomass productivity in mixtures per unit rye or vetch seed sown, while total LER > 1.0 demonstrates greater overall biomass productivity (dry weight per seed sown) for the mixture relative to the monocultures.

Vetch N Fixation Estimates

Vetch N fixation was estimated using the ^{15}N natural abundance method, which relies on the slight enrichment of ^{15}N generally present in soil N relative to atmospheric N_2 (Shearer and Kohl, 1986). Since biological N fixation exhibits little isotopic fractionation, legumes tend to have a lower proportion of ^{15}N in their tissues than plants that derive their N entirely from the soil (Hogberg, 1997). Isotopic composition is expressed relative to atmospheric N_2 using $\delta^{15}N$ values in parts per thousand (‰), with higher $\delta^{15}N$ indicating greater ^{15}N enrichment.

The percentage of vetch N derived from the atmosphere (%Ndfa) was calculated from Eq. 4 (Rochester and Peoples, 2005),

$$[4] \quad \%\mathrm{Ndfa} = (\delta^{15}N_{ref} - \delta^{15}N_{leg})/(\delta^{15}N_{ref} - B) \times 100$$

where $\delta^{15}N_{ref}$ is the shoot $\delta^{15}N$ value for rye in monoculture (used as the non-N-fixing reference plant), $\delta^{15}N_{leg}$ is the shoot $\delta^{15}N$ value for vetch, and B is the estimated $\delta^{15}N$ of vetch when grown entirely dependent on atmospheric sources of N. Following Hansen et al. (2002) and Hansen and Vinther (2001), the lowest $\delta^{15}N_{leg}$ value in each study year was used to approximate B, resulting in conservative estimates of vetch %Ndfa. Rye monoculture $\delta^{15}N_{ref}$ values were 2.52 and 1.49 ‰ in 2010 and 2011, respectively.

Vetch shoots contaminated with soil (ash content >0.2) in 2011 exhibited unreasonably high $\delta^{15}N$ values (1.10 – 5.58 ‰) that could not be explained by contributions from soil ^{15}N alone. We speculate that microbial N transformations (such as denitrification) may have occurred preferentially on contaminated vetch shoots prior to sampling in the field or during sample processing before drying, potentially resulting in significant isotope fractionation (Mariotti et al., 1981; Shearer and Kohl, 1986). Therefore, contaminated vetch tissue was excluded from the analysis. As a result, 2011 mean %Ndfa estimates are reported only for the three cover crop treatments where at least 3 field replicates of uncontaminated vetch tissue (ash content < 0.2) were available.

The amount of vetch shoot N derived from N fixation (SNF, in kg ha^{-1}) was estimated using the following formulas:

[5] $\mathrm{ShootN} = (\mathrm{shootdryweight})\mathrm{x}(N_v)$

[6] $\mathrm{SNF} = (\mathrm{shootN})\mathrm{x}(\%\mathrm{Ndfa})$

where N_v is the contamination-corrected concentration of N in the vetch shoots. Estimates of SNF for 2011 were calculated assuming a consistent %Ndfa across the cover crop treatments equal to the average %Ndfa obtained from the uncontaminated samples in that year.

Cost of Nitrogen Fixed

In order to estimate the relative costs incurred by growers for the additional fixed N contributed to the system by vetch, the cost of N fixed (CNF) was estimated for each vetch-containing cover crop treatment following Brainard et al. (2012), using the equation:

[7] $\mathrm{CNF} = [(P_v \times Q_v) + (P_r \times Q_r) - (P_r \times 94)] / \mathrm{SNF}$

where P_v and P_r are the prices ($ kg^{-1}) of conventional vetch and rye seed, respectively; Q_v and Q_r are the seeding rates (kg ha^{-1}) of vetch and rye, respectively; and SNF is the vetch shoot N fixed (kg ha^{-1}) estimated from Eq. [6]. CNF is an estimate of the price paid for the N fixed by vetch as influenced by legume performance and cover crop seed costs. The calculation controls for costs associated with cover crop establishment by assuming that it applies to growers who already seed rye at 94 kg ha^{-1}, and therefore just have to substitute legume seed in their planting equipment. Since rye is a common cover crop in many agronomic and vegetable cropping systems, CNF provides a reasonable estimate of both the additional costs a grower may incur to gain fixed N from vetch in their system, and the relative effects of rye-vetch mixture proportion on the estimated price of additional fixed N. Because some fixed N also accumulates in root tissue, calculations of CNF based on Eq. [7] likely overestimate the cost of total fixed N in vetch biomass.

Statistical Analysis

The fixed effects of year, cover crop mixture proportion, and their interaction were analyzed using mixed model ANOVA with the MIXED procedure of SAS (Version 9.2, SAS Institute, Cary, NC). Block (replication) was included as a random factor in all models. Assumptions of normality and equality of variances were evaluated, and unequal variance models were used when necessary. Unless otherwise noted, effects where judged significant when $P < 0.05$. Where the effect of cover crop mixture proportion was significant, the nature of the response in each year was investigated using linear or polynomial regression with the REG procedure of SAS. In the case of PAR, the effects of year and the interaction between year and cover crop were not significant, so data were pooled over years and fitted with a single regression equation. The significance and nature of the relationship between rye shoot N concentration and cover crop mixture proportion were evaluated in the context of analysis of covariance (ANCOVA) using the MIXED procedure of SAS, since the response was linear and had a common slope in both years. Individual partial and total LER estimates for cover crop mixtures were compared to their respective critical values using *t*-tests. Finally, due to missing data in 2011, vetch %Ndfa data were analyzed separately by year.

RESULTS AND DISCUSSION

Weather Conditions

Table 3 presents total growing degree day (GDD) base 4 °C accumulation and rainfall at the experiment station during the periods of cover crop growth. Heat accumulation and total rainfall were similar in the fall of both study years, but the spring of 2011 saw 113 fewer GDD and 125 more mm of rainfall than spring 2010. Previous research has demonstrated good correlation between GDD base 4 °C accumulation and winter cover crop biomass in the spring (Teasdale et al., 2004). While wetter soil conditions in 2011 contributed to difficulties with vetch biomass sampling that season (see Materials and Methods), rainfall was sufficient in both years to make water deficit during cover crop growth unlikely, suggesting that compared to heat accumulation, soil moisture was probably a minor contributor to variability in cover crop growth across study years.

Cover Crop Density and Vetch Winter Survival

Rye and vetch spring population densities across treatments were similar in the two years of the study, with rye and vetch monocultures averaging 146 and 271 plants m^{-2}, respectively (Figure 1). The densities of both species in mixture were highly correlated with their relative sown proportions, and the relationships were predominantly linear. This suggests that inter-specific interactions had little effect on the germination and establishment of rye and vetch under the conditions of our field study, and that seeding rates can be good predictors of final population composition in rye-vetch mixtures.

The influence of mixture proportions on vetch winter survival, in particular, was investigated by observing the percent change in vetch population density between counts taken in the fall and spring from the same quadrats. There were no significant differences in vetch population change among the treatments in either year (Table 4); however, contrary to expectation, in most cases our estimates of vetch density actually increased between the fall and spring, resulting in apparent overwinter survival estimates greater than 100%.

This has been observed in previous studies (Brainard et al., 2012), and may in part be attributed to delayed germination from vetch hard seed (Aarssen et al., 1986). In addition, individual vetch plants are increasingly difficult to distinguish from each other *in-situ* as the shoots sprawl and intertwine. Therefore, we speculate that the *in-situ* counts taken in the fall may have underestimated vetch density compared with the spring counts, in which quadrats were destructively sampled and individual plants could more easily be identified by their roots. Later fall sampling (and more advanced vetch growth) in 2009 (Table 3) may also have contributed to greater under-counting that year, suggesting the larger population change estimates in 2009 relative to 2010 ($P = 0.006$) could be an artifact.

Assuming delayed vetch germination and potential sampling biases were consistent across treatments, we found no evidence that vetch winter survival was improved by mixing with rye. In contrast, Jannink et al. (1997) found that the presence of rye reduced overwinter mortality of vetch at two locations in Maine, and Brainard et al. (2012) observed a similar response in one out of two years in Michigan. While increased legume winter survival is an often-stated benefit of cereal-legume cover crop mixtures, its significance will likely vary depending on both winter weather conditions and the establishment (and hardiness) of the legume. The presence of rye may have a greater influence on vetch winter survival, for example, when the vetch is seeded late in the fall or when winter temperatures are particularly harsh.

Cover Crop Biomass Production and Land Equivalent Ratios

Vetch shoot biomass in monoculture was 551 and 305 g m^{-2} in 2010 and 2011, respectively, while rye monocultures produced 414 and 330 g m^{-2} in those two years (Figure 2). Cooler spring temperatures in 2011 (Table 3) likely contributed to the overall lower biomass production observed that year, particularly for vetch (rye, $P = 0.03$; vetch, $P = 0.004$). As with population density, relative seeding rates were strongly correlated with rye and vetch shoot dry weight in mixtures, demonstrating that seeding rates can be a good predictor of biomass composition as well. Over the course of the study, the total shoot biomass produced in mixtures was generally equal to or greater than that produced in either monoculture. This was reflected in the total LER values for the mixtures, which ranged from 1.13 to 1.42, and in all cases were either equal to or significantly greater than one (Table 5). Furthermore, relative yields of both rye (L_r) and vetch (L_v) met, and in some cases significantly exceeded, their relative sown proportions in all mixtures, indicating that both species contributed to the equivalent or greater efficiency of biomass production (dry weight per seed sown) observed in the mixtures relative to the monocultures. In other words, just as much or more biomass was produced per dollar spent on rye and vetch seed in mixtures as in monocultures.

Inference regarding the nature of interactions between species is limited from proportional replacement series designs because total plant density is not constant across the treatments (Jolliffe, 2000). In the case of this study, total density increased with higher proportions of rye in the mixture (Figure 1). Without data on the yield response of each species to changes in density, the balance between inter- and intra-specific interactions in mixtures cannot be determined with confidence (Firbank and Watkinson, 1985; Jolliffe, 2000). However, given that neither rye nor vetch productivity (dry weight per seed sown) was suppressed in any of the mixtures, interspecific interference between the two species appears to have been minimal under the conditions of this experiment. Furthermore, plants generally exhibit an inverse relationship between density and biomass production per individual (Avci and Akar, 2006; Boyd et al., 2009). Therefore, increased rye productivity in mixtures could simply be due primarily to the decrease in total plant density associated with decreasing proportions of rye in the mixtures. Conversely, vetch productivity was not reduced at the higher total plant densities associated with higher rye proportions (in opposition to the expected density-dependent response), suggesting that vetch may have benefitted from either weaker interspecific competition or possible facilitation from rye.

Decades of intercropping research have demonstrated that cereals are usually the dominant component in cereal-legume mixtures, suppressing the growth of the legume and contributing a greater proportion of biomass to the total mixture yield (Ofori and Stern, 1987). This competitive imbalance may be more severe for low-growing legumes than for those with climbing growth habits (Davis et al., 1984; Fukai and Trenbath, 1993), but the performance of vetch in mixture with cereals still varies across studies. Kurdali et al. (1996) and Tosti et al. (2010) both saw reduced vetch dry matter productivity in mixture with barley (*Hordeum vulgare* L.), while Dhima et al. (2007) also observed suppression of vetch by barley and oat (*Avena sativa* L.), but not by wheat (*Triticum aestivum* L.) or triticale (*xTriticosecale* Wittmack). As with our results, Ranells and Wagger (1996) observed limited suppression of vetch in mixture with rye; however, Brainard et al. (2012) observed one out of two seasons where the productivity of three vetch varieties was reduced on average by rye, which yielded over 400 g m^{-2} in the 50:50 mixtures. By comparison, rye yields were relatively low in our study. The sandy soils, prior growth and incorporation of sorghum-sudangrass, and absence of N fertilization likely contributed to low soil N conditions that favored vetch growth relative to rye, reducing the potential for competitive inhibition in the mixtures.

An important qualification to note is that measures of yield advantage or efficiency depend not only on the performance of the species in mixture, but also on the characteristics of their respective monocultures. An implicit assumption in intercrop or mixture evaluation is that the monocultures chosen are the optimal ones for sole-cropping of each species (Willey, 1979b). In this study, monoculture seeding rates were chosen with the goal of maximizing cover crop biomass production, especially for vetch. However, due to the high cost of seed, it is common to see vetch sown at 28 kg ha^{-1} in monoculture, particularly in agronomic cropping systems (e.g., Clark et al., 1994; Sainju et al., 2005; Parr et al., 2011). Furthermore, rye monocultures are often sown at rates of 125 kg ha^{-1} or more, which may better maximize biomass production, particularly under conditions where soil N is not limiting (Clark, 2007; Boyd et al., 2009). Outcomes in replacement-type mixtures and conclusions regarding mixture performance may vary when different monoculture seeding rates are used (Willey, 1979b).

Winter Annual Weed Biomass and PAR

The percentage of PAR penetrating the cover crop canopies in early May ranged from 6 to 47% for vetch and rye monocultures, respectively, averaged over the two years of the study. In the mixtures, PAR penetration increased with decreasing proportion of vetch and increasing proportion of rye (Figure 3). All cover crop treatments significantly suppressed winter annual weed biomass relative to control treatments in 2010 and 2011, and suppression increased with increasing rye in the cover crops (Figure 4). Total weed biomass in the control treatments was 144 and 162 g m^{-2} in 2010 and 2011, respectively, and dominant species included common chickweed [*Stellaria media* (L.) Vill.], annual bluegrass (*Poa annua* L.) and shepherd’s purse [*Capsella bursa-pastoris* (L.) Medik.] in 2010, and corn chamomile (*Anthemis arvensis* L.), field pepperweed [*Lepidium campestre* (L.) R. Br.], and common chickweed in 2011. While suppression by high rye treatments was comparable between the two years of the study, variability in weed suppression between 2010 and 2011 increased with greater proportion of vetch sown. Although generally less of a management priority than summer annuals, winter annual weeds can interfere directly with crops in reduced tillage systems and serve as alternative hosts for important pests and diseases (Hayden et al., 2012).

Our results are consistent with previous studies demonstrating that weed suppression in cereal-legume mixtures is generally driven by the presence and relative proportion of the more competitive cereal component (Liebman and Dyck, 1993; Akemo et al., 2000b; Hayden et al., 2012). The increasingly lower weed suppression in 2011 relative to 2010 with greater vetch in the cover crops was likely a result of the cooler spring temperatures and lower biomass production that year, and also suggests that the weed suppressiveness of vetch is less robust in the face of year-to-year environmental variability than rye. While both rye and vetch are known to exhibit allelopathic properties (Barnes and Putnam, 1986; White et al., 1989), shading is likely a more dominant mechanism of competition for vetch than it is for rye, which suppresses neighboring plants more predominately through competition for water and soil nutrients (Teasdale and Daughtry, 1993; Mariotti et al., 2009). This dominance of below-ground interference in the weed suppressiveness of rye is supported by the fact that weed biomass decreased with increasing rye in the cover crops, despite a corresponding increase in the amount of PAR reaching the soil surface in the spring. However, it is important to note that canopy architecture changes substantially during rye growth, and PAR readings taken in the spring may not reflect the level of competition for light exerted by rye on weeds the previous fall.

Cover Crop Nitrogen Content

Total shoot N content across the cover crop stands in 2010 and 2011 was largely driven by rye and vetch biomass production. As expected, vetch biomass contributed the majority of N to rye-vetch mixtures, and total N content generally increased with increasing proportion of vetch sown (Figure 5). In 2010, total N content ranged from 3.8 g m^{-2} in the rye monoculture to 17.5 g m^{-2} in the vetch monoculture, while in 2011, total N content ranged from 4.2 g m^{-2} in rye monoculture to 11.6 g m^{-2} in the 83:17 vetch:rye mixture.

In both study years, a modest but significant increase in rye tissue N concentration was observed in mixtures with increasing proportions of vetch (Figure 6). Combined with the equivalent or greater rye biomass productivity observed in mixtures relative to the rye monoculture (Table 5), this trend provides added support that the productivity of N accumulation by rye in mixtures was greater as well. Similarly, Tosti et al. (2010, 2012) saw increases in the tissue N concentration of barley with increasing vetch sown in replacement-style mixtures. In both cases, the mechanism behind the increase cannot be clearly established. Improved cereal N economy in cereal-legume mixtures is consistent with a facilitative increase in N availability due to the legume. However, in proportional replacement series designs, greater legume proportions in mixtures are also accompanied by a corresponding reduction in cereal density. Because rye is a stronger competitor for soil N resources than vetch, reduced rye intra-specific competition for soil N could also account for the increase in rye tissue N concentration across the gradient of rye-vetch mixtures. The overall higher rye N concentrations in 2011 compared with 2010 ($P < 0.001$) likely reflect a more immature rye growth stage at sampling due to the lower GDD accumulation during the 2010-2011 season.

Vetch Nitrogen Fixation

Estimates of vetch shoot %Ndfa were relatively high in both years, ranging from 81 to 88%. Although %Ndfa was marginally lower for vetch in monoculture than for vetch in mixture with rye on average in 2010 (linear contrast, $P = 0.065$), estimates of N fixation did not differ significantly across the rye-vetch mixtures. In 2011, soil contamination compromised N fixation estimates from cover crop treatments containing greater than 50% vetch; however, for the three mixtures where reliable data were available, vetch %Ndfa averaged 82% and did not differ among the mixture rates.

Previous studies have observed increases in vetch N fixation efficiency in mixture with cereals (Kurdali et al., 1996; Brainard et al., 2012). We expected that higher proportions of rye in mixture might lead to greater inter-specific competition for available soil N and promote greater N fixation by vetch. However, the relatively sandy soils, low organic matter content, and absence of N fertilization at our study site may have reduced the potential for observing such an effect. The presence of rye and its relative proportion in mixture may be more likely to influence vetch N fixation efficiency under conditions where soil N availability is high. On the other hand, excessive soil N may also shift the competitive balance in favor of the cereal, resulting in inhibition of legume growth and potentially N fixation capacity. While Brainard et al. (2011) observed increased nodulation of soybean when grown in combination with Japanese millet [*Echinochloa frumentacea* (Roxb.) Link], the effect was not observed when composted manure was added to the system.

The amount of shoot N fixed (SNF) was driven predominately by vetch biomass productivity. Across the gradient of rye-vetch cover crops, SNF varied from zero in rye monoculture to 142 and 85 kg N ha^{-1} in the 2010 and 2011 vetch monocultures, respectively (Table 6). However, the total cost for conventional cover crop seed increased from \$37 to \$176 ha^{-1} over that same range of treatments (Table 1). As a result, the estimated cost of the N fixed by vetch (CNF) ranged from \$0.79 to \$1.90 kg^{-1} N over the two years of the study, and despite a numerical trend toward decreasing costs in mixture with greater rye, CNF was not affected significantly by rye-vetch mixture rate (Table 6). A similar range of CNF estimates was established by Brainard et al. (2012) for three vetch varieties in monoculture and in mixture with rye. For conventional growers in particular, acknowledging the value of fixed N as a potential fertilizer replacement may help justify the substantially higher seed costs of cover crops that contain greater proportions of vetch. Comparisons of CNF with the cost of synthetic N fertilizers are confounded by the incomplete first-season availability of N from organic residues and by the potential value of additional services that are provided by cover crops beyond N fertility. But as a reference, the average cost of urea fertilizer over the past five years was fairly comparable to CNF from vetch in this study, ranging from \$1.10 to \$1.35 kg^{-1} N (USDA-Economic Research Service, 2013). Furthermore, estimates of CNF in this study did not include fixed N accumulated in root tissue. Since vetch roots can account for 10% or more of total tissue N (Rogers and Sturkie, 1939), our figures likely overestimate CNF. Nevertheless, while seed costs vary with time, source, and whether conventional or organic seed is used, our results suggest that under the conditions of this study, the relative cost for the N fixed by vetch was not significantly influenced by mixing with rye at any proportion.

CONCLUSIONS

The objective of this research was to evaluate the effects of rye-vetch species proportions on cover crop mixture stand characteristics and performance relative to rye and vetch monocultures. Seeding rates were good predictors of rye and vetch stand density and biomass composition in mixtures, with little evidence of substantial interference between the two species under the conditions of the study. While outcomes may vary depending on environment and management, we also saw little evidence of synergistic benefits in the mixtures, such as improved vetch winter survival, increased N fixation, or reduced costs per unit of N fixed. However, changing the proportions of rye and vetch sown resulted in tradeoffs among some of the agroecosystem services provided by the living cover crops—for example, greater fixed N accumulation but higher seed costs and reduced winter annual weed suppression with increasing vetch. A greater understanding of the tradeoffs among cover crop services, as well as costs, in rye-vetch mixtures will support more-informed decision-making regarding cover crop selection and mixture seeding rates.

APPENDIX

Table 2.1. Rye (R) and vetch (V) seeding rates and estimated seed costs across cover crop mixture proportions.

	Seeding rate		Cost of seed†		
Cover crop treatment	Vetch	Rye	Vetch	Rye	Total
	kg ha^{-1}			\$ ha^{-1}	
100 V : 0 R	42	0	176	0	176
83 V : 17 R	35	16	146	6	152
67 V : 33 R	28	31	118	12	130
50 V : 50 R	21	47	88	18	106
33 V : 67 R	14	63	58	25	83
17 V : 83 R	7	78	30	30	60
0 V : 100 R	0	94	0	37	37

† Seed costs were calculated using average prices paid for conventional vetch and rye seed at the time of this experiment (\$4.19 and \$0.39 kg^{-1}, respectively). Average prices paid for the organic seed sown in the experiment were \$4.63 and \$0.48 kg^{-1} for vetch and rye, respectively. Actual seed costs will vary with time, seed source, and whether conventional or organic seed is used.

Table 2.2. Dates of key field activities and data collection.

Activity	2009-2010	2010-2011
Cover crop seeding	1 Sept.	1 Sept.
Cover crop fall density sampling	19-21 Oct.	27-29 Sept.
PAR† light penetration readings	6 May	11 May
Spring cover crop and weed biomass and density sampling	10 May	14 May

† Photosynthetically active radiation

Table 2.3. Monthly growing degree day (GDD) and rainfall totals in Holt, MI during cover crop growth, 2009-2011.

	GDD†		Rainfall	
Month	2009-10	2010-11	2009-10	2010-11
			mm	
September‡	393	364	24	92
October	173	228	92	34
November	99	81	24	50
March	81	28	16	66
April	233	125	74	135
May§	91	139	52	65
Sept. to Nov.‡	665	673	141	176
Mar. to May§	405	292	141	266

† Calculated according to Baskerville-Emin method (Baskerville and Emin, 1969) using base 4°C.

‡ Cumulative GDD beginning at cover crop seeding (see Table 2.2). Data for Dec. through Feb. not presented due to negligible GDD accumulation and unreliability of measurements of frozen precipitation.

§ Cumulative GDD ending at cover crop biomass sampling (Table 2.2).

Table 2.4. Vetch apparent overwinter survival (population change) across cover crop mixture proportions.

	Overwinter survival	
Cover crop treatment	2010	2011
	%	
100 V : 0 R	137†	94
83 V : 17 R	119	98
67 V : 33 R	127	106
50 V : 50 R	157	105
33 V : 67 R	123	91
17 V : 83 R	126	95
F‡	ns§	ns

† See text for possible explanations for apparent overwinter survival estimates > 100%.

‡ Significance of fixed effect of cover crop mixture proportion within years.

§ ns, nonsignificant at the 0.05 probability level.

Table 2.5. Partial land equivalent ratios for vetch (L_v) and rye (L_r) and total land equivalent ratios (LER) in rye-vetch cover crop mixtures.

	2010			2011		
Cover crop treatment	L_v	L_r	LER	L_v	L_r	LER
83 V : 17 R	0.76	0.51*†	1.27	0.98	0.39*	1.38‡
67 V : 33 R	0.72	0.52	1.25	0.91*	0.51*	1.42*
50 V : 50 R	0.54	0.66	1.20	0.53	0.62	1.15
33 V : 67 R	0.37	0.90	1.27	0.55‡	0.82‡	1.37‡
17 V : 83 R	0.22	1.00	1.23	0.31	0.82	1.13

* Significant at the 0.05 probability level.

† Indications of significance refer to whether individual L_v and L_r means are significantly different from vetch and rye sown proportions in mixture, respectively, and whether LER means are significantly different from 1.0 (based on *t*-tests).

‡ Significant at the 0.10 probability level.

Table 2.6. Vetch shoot N concentration, percentage of N derived from the atmosphere (Ndfa), estimated shoot N fixed (SNF) and cost of N fixed (CNF) across cover crop mixture proportions.

	N concentration		Ndfa†		SNF‡		CNF§	
Cover crop treatment	2010	2011	2010	2011¶	2010	2011	2010	2011
	%		%		kg ha^{-1}		\$ kg^{-1}	
100 V : 0 R	3.17	3.37	80.9	--	142.4	84.6	1.05	1.68
83 V : 17 R	3.18	3.30	87.9	--	110.8	80.1	1.13	1.47
67 V : 33 R	3.13	3.31	86.5	--	102.3	74.9	0.92	1.29
50 V : 50 R	3.12	3.36	88.4	84.0#	74.9	43.6	1.15	1.90
33 V : 67 R	3.28	3.37	87.5	82.8#	56.9	46.0	0.84	1.16
17 V : 83 R	3.18	3.36	84.5	80.1	31.5	25.5	0.79	1.11
F††	ns‡‡	ns	ns	ns	***	**	ns	ns

** Significant at the 0.01 probability level.

*** Significant at the 0.001 probability level.

† Calculated from Eq. [4].

‡ Calculated from Eq. [6].

§ Calculated from Eq. [7].

¶ Means are reported for treatments only where at least 3 replicates of uncontaminated vetch tissue were available in 2011 (see Materials and Methods).

Means calculated from three replicates (*n*=3)

†† Significance of fixed effect of cover crop mixture proportion within years.

‡‡ ns, nonsignificant at the 0.05 probability level.

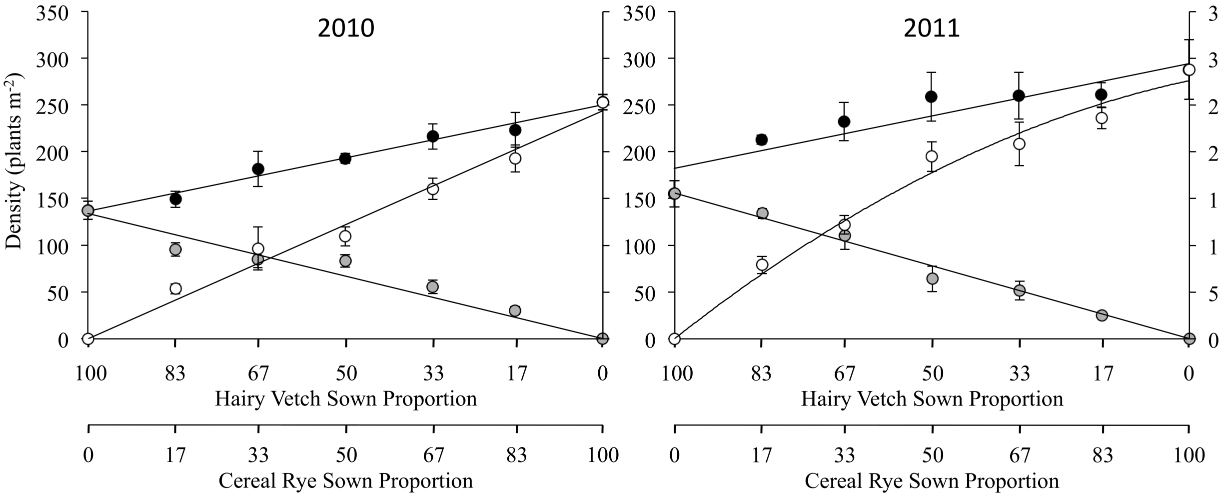

Figure 2.1. Spring density of vetch (gray), rye (white), and vetch+rye (black) across cover crop mixture proportions in 2010 and 2011. 2010 responses of vetch: y=1.34x, r^2=0.81***; rye: y=2.44x, r^2=0.87***; and vetch+rye: y=1.14x+136.23, r^2=0.73***. 2011 responses of vetch: y=1.56x, r^2=0.84***; rye: y=0.006x^2+0.46x, r^2=0.80***; and vetch+rye: y=1.12x+182.26, r^2=0.47***. Means (+/- SE). Single, double, and triple asterisks following r^2 values denote regression model significance at the 0.05, 0.01, and 0.001 levels, respectively.

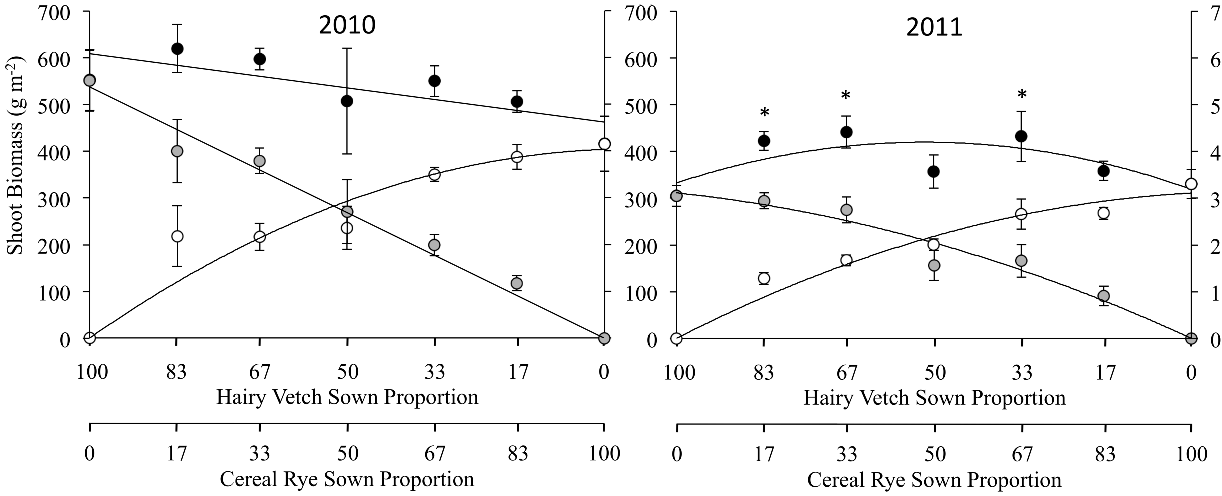

Figure 2.2. Spring shoot biomass dry weights of vetch (gray), rye (white), and vetch+rye (black) across cover crop mixture proportions in 2010 and 2011. 2010 responses of vetch: y=5.38x, r^2=0.71***; rye: y=-0.036x^2+7.67x, r^2=0.50***; and vetch+rye: y=-1.46x+607.32, r^2=0.16*. 2011 responses of vetch: y=-0.020x^2+5.07x, r^2=0.69***; rye: y=-0.025x^2+5.62x, r^2=0.76***; and vetch+rye: y=-0.038x^2+3.62x+332.52, r^2=0.23*. Means (+/- SE). Single, double, and triple asterisks following r^2 values denote regression model significance at the 0.05, 0.01, and 0.001 levels, respectively. Asterisks in the figure indicate where total biomass (vetch+rye) in a mixture was significantly greater than in either monoculture in that year at the 0.05 probability level.

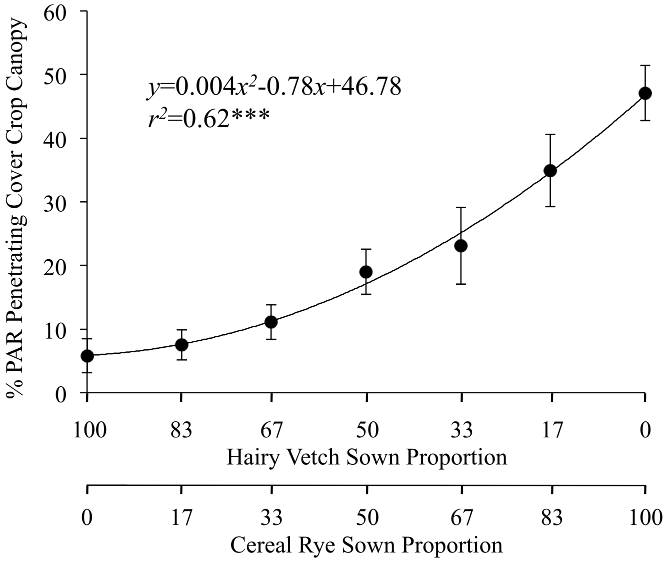

Figure 2.3. Percentage of photosynthetically active radiation (PAR) penetrating the cover crop stand canopy across mixture proportions. Means are for 2010 and 2011 data combined (*n*=8), presented +/- SE. The triple asterisks following the r^2 value in the figure denote regression model significance at the 0.001 level.

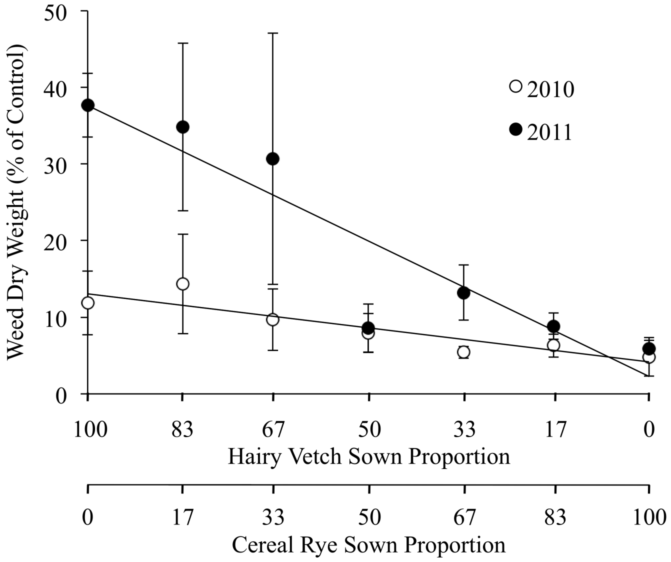

Figure 2.4. Winter annual weed biomass (dry weight) across cover crop mixture proportions in 2010 and 2011, expressed as mean percentage of the no-cover-crop control treatment (+/- SE). 2010 response: y=0.089x+4.18, r^2=0.18*. 2011 response: y=0.35x+2.27, r^2=0.40***. Single, double, and triple asterisks following r^2 values denote regression model significance at the 0.05, 0.01, and 0.001 levels, respectively.

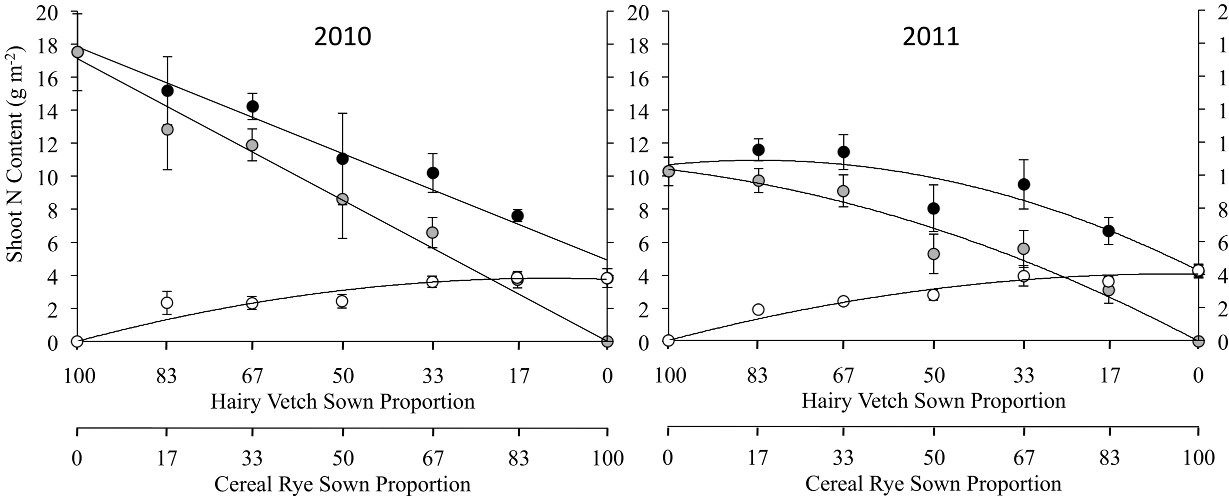

Figure 2.5. Shoot N accumulated in vetch (gray), rye (white), and vetch+rye (black) across cover crop mixture proportions in 2010 and 2011. 2010 responses of vetch: $y=0.92x$, $r^2=0.66$***; rye: $y=-0.00048x^2+0.086x$, $r^2=0.36$***; and vetch+rye: $y=-0.13x+17.83$, $r^2=0.67$***. 2011 responses of vetch: $y=-0.00066x^2+0.17x$, $r^2=0.66$***; rye: $y=-0.00044x^2+0.084x$, $r^2=0.61$***; and vetch+rye: $y=-0.00097x^2+0.032x+10.69$, $r^2=0.58$***. Means (+/- SE). Single, double, and triple asterisks following r^2 values denote regression model significance at the 0.05, 0.01, and 0.001 levels, respectively.

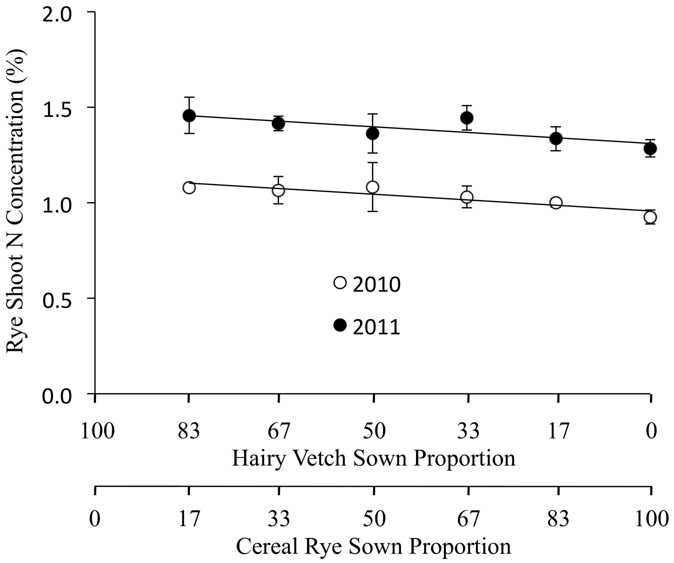

Figure 2.6. Rye shoot N concentration across rye-vetch cover crop mixture proportions in 2010 and 2011. The slope of the response to rye-vetch proportion was equal in both years and significant overall (analysis of covariance, $P < 0.001$). 2010 equation: y=0.0018x+0.96, r^2=0.15. 2011 equation: y=0.0018x+1.31, r^2=0.13. Means (+/- SE).

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CHAPTER III

RYE-VETCH COVER CROP RESIDUE QUALITY AND PLASTIC MULCH AFFECT SOIL NITROGEN DYNAMICS, MICROBIAL COMMUNITIES, AND YIELDS IN VEGETABLE PRODUCTION SYSTEMS

Rye-vetch cover crop residue quality and plastic mulch affect soil nitrogen dynamics, microbial communities, and yields in vegetable production systems

Zachary D. Hayden*, Mathieu Ngouajio, and Daniel C. Brainard

Department of Horticulture, Michigan State University, Plant and Soil Sciences Building, 1066 Bogue St., East Lansing, MI 48824.
*Corresponding author (haydenza@msu.edu).

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ABSTRACT

Cover crops can be an important source of nitrogen (N) for organic vegetable production, but N availability depends heavily on both the characteristics of the cover crop residues and crop management practices following cover crop termination. A study was conducted over two seasons (2009-2010 and 2010-2011) in Michigan to investigate 1) how the proportions of cereal rye (*Secale cereale* L.) and hairy vetch (*Vicia villosa* Roth) sown in a winter cover crop mixture influence cover crop residue quantity and quality in the spring, and 2) how rye-vetch residue characteristics and the use of black polyethylene mulch (PM) interact to affect soil N dynamics, microbial communities, and yields of bell pepper (*Capsicum annuum* L.) and slicing cucumber (*Cucumis sativus* L.) the following summer. Cover crop main plot treatments consisted of a gradient of 7 sown proportions of rye:vetch, from 100% rye (94 kg/ha) to 100% vetch (42 kg/ha), in addition to a no cover crop control. Subplots consisted of pepper and cucumber grown either with or without PM. In both years, cover crop stands with higher sown proportions of vetch generally contained more total N and had lower total residue C:N. Marketable vegetable yields, soil inorganic N levels, and pore water nitrate concentrations below the root zone during the summers were positively correlated with the total N content of cover crop mixtures. Yields and soil N levels were generally higher when PM was used, but the differences between PM and bare ground treatments were much less pronounced in 2011 than in 2010. Differences in the magnitude and timing of summer precipitation events likely contributed to the discrepancy between the two years, with potentially greater N leached from bare ground treatments in 2010 than in 2011. Lower microbial biomass was observed under PM compared to bare ground 3 wk after plastic application, but cover crop and mulching treatments did not explain the majority of variation observed in patterns of carbon substrate utilization across soil microbial communities

from Biolog-Ecoplate^{TM} data. Our results demonstrate that PM can be an important tool for preserving N fertility benefits derived from high N cover crop residues, particularly on sandy soils. While higher proportions of vetch in the cover crop mixtures and the use of PM generally resulted in higher marketable vegetable yields, system performance must ultimately be based on a balanced consideration of all the services provided, their costs, and their relationship to primary production goals.

INTRODUCTION

The potential benefits of cereal rye (*Secale cereale* L.) and the legume hairy vetch (*Vicia villosa* Roth) as winter annual cover crop species have been well documented in both agronomic and vegetable cropping systems. Rye seed is inexpensive, and it possesses superior capabilities in suppressing weeds, conserving residual soil N, and controlling erosion (Shipley et al., 1992; Ditsch et al., 1993; Clark, 2007; Hayden et al., 2012). However, with increasing maturity, rye residues can cause net N immobilization and yield loss in subsequent crops (Clark et al., 2007). In contrast, the defining strengths of vetch are its considerable capacity for fixed N accumulation and the rapid mineralization of that residue N following cover crop termination (Ranells and Wagger, 1996; Teasdale et al., 2004). While outcomes vary depending on management and environment, rye-vetch mixtures can combine these strengths while benefitting from potential grass-legume functional complementarities (Ofori and Stern, 1987; Clark et al., 1994; Teasdale and Abdul-Baki, 1998; Sainju et al., 2005). However, in most cases, cereal-legume cover crop characteristics reflect relative species composition (Akemo et al., 2000; Karpenstein-Machan and Stuelpnagel, 2000; Tosti et al., 2010). In Hayden et al. (2014), we demonstrated that relative seeding rates can accurately govern the final proportions of rye and vetch in cover crop mixture stands, and that changing the relative proportions of the species resulted in tradeoffs between total fixed N accumulation, seed costs, and winter annual weed suppression. Understanding how differences in total residue characteristics across the mixture proportions influence the soil-crop system following residue incorporation is also essential in evaluating overall mixture performance.

The availability of N from cover crop residues is governed by complex interactions between soil biological activity, residue characteristics, and environment. Temperature, moisture, soil texture, whether residues are incorporated or left as a surface mulch, and the timing and method of cover crop termination are all factors that can influence residue mineralization (Creamer and Dabney, 2002; Reberg-Horton et al., 2012). In addition, losses of N through ammonia volatilization, denitrification, weed uptake, and perhaps most notably, leaching, can reduce N availability for subsequent crops. Net mineralization from high N legume residues typically occurs rapidly and out of sync with crop demand, resulting in excess soil inorganic N early in the season that is susceptible to loss (Palm et al., 2001). In addition to reducing the potential for N tie-up from cereal residues, the moderation of total residue quality in cereal-legume mixtures may alter patterns of net N mineralization in ways that can reduce risks for early-season N losses and improve synchrony with crop demand relative to a monoculture legume (Kuo and Sainju, 1998; Clark et al., 2007; Crews and Peoples, 2005). Few studies have evaluated the effects of cereal-legume mixture proportions on N dynamics and crop yields in a field setting (Tosti et al., 2012), and results may vary in intensive vegetable production systems for crops with differing durations and nutrient requirements, like bell pepper (*Capsicum annuum* L.) and slicing cucumber (*Cucumis sativus* L.).

The use of black polyethylene (plastic) mulch is an industry standard for commercial organic and conventional production of many warm-season vegetable crops in cooler climates. Depending on color and chemical composition, plastic mulches can distinctly alter the soil and surface microclimate in the crop row, while providing a barrier to weed emergence, precipitation, soil splash, and the spread of soil borne diseases (Kasirajan and Ngouajio, 2012). Under drip-irrigated black plastic, higher soil temperatures, more consistent soil moisture conditions, and protection from leaching precipitation can have substantial effects on N mineralization and loss from organic residues (Clarkson, 1960; Tarara, 2000; Schonbeck and Evanylo, 1998). However, concerns over the economic and environmental costs associated with plastic mulch disposal, and challenges with the performance and price of biodegradable alternatives, have discouraged the use of plastic mulch in some agricultural sectors (Kyrikou and Briassoulis, 2007). Evaluating the effects of cover crop residue quality on N dynamics and vegetable production both with and without plastic mulch can provide insights into the influence of mulching on rye-vetch mixture performance while producing results relevant to both production systems.

The size, activity, and functional characteristics of soil microbial communities are important determinants of many agroecosystem processes, particularly residue decomposition and nutrient cycling (Paul, 2007). Although innate soil characteristics and climate often have greater effects, agricultural management practices—such as tillage, compost additions, and cover cropping—can result in changes to microbial communities in both the short- and long-term (Insam, 1990; Bossio et al., 1998; Nair and Ngouajio, 2012). Plastic films, sometimes in combination with organic additions, are employed in practices such as soil solarization (Katan,1981) and anaerobic disinfestation (Noriaki, 2008) to create extreme temperatures and/or anoxic conditions for a short period with the goal of reducing populations of soil-borne pests and pathogens. Under these conditions, plastic mulches reduce total microbial biomass and can influence community composition (Scopa and Dumontet, 2007; Noriaki, 2008). However, few studies (Moreno and Moreno, 2008; Carrera et al., 2007) have evaluated the influence on microbial community function of the more modest but continuous increases in soil temperature, and changes to aeration and moisture conditions (Khan et al., 2000), that result from standard black plastic mulching during crop production, particularly in combination with residues of varying quality.

The objectives of this research were 1) to investigate how the proportions of cereal rye and hairy vetch sown in a mixture influence cover crop residue quantity and quality in the spring, and 2) how the characteristics of incorporated rye-vetch residues and the use of black plastic mulch interact to affect vegetable yields and fruit quality, soil inorganic N dynamics, and soil microbial communities in bell pepper (*Capsicum annuum* L.) and slicing cucumber (*Cucumis sativus*) production systems.

MATERIALS AND METHODS

Site Description

The study was conducted over two seasons (2009-2010 and 2010-2011), alternating between adjacent fields on a level Spinks loamy sand soil (sandy, mixed, mesic Lamellic Hapludalf) at the Michigan State University Horticulture Teaching and Research Center in Holt, Michigan (42°40” N, 84°28” W). A summer cover crop of sorghum sudangrass (*Sorghum bicolor* x *S. bicolor* var. Sudanense) was flail mowed and incorporated into each field with a rototiller at least 2 wk prior to seeding rye and vetch in the fall. The fields had been in organic transition with a history of warm-season vegetable production under conventional tillage since 2008. Initial soil chemical characteristics for the fields averaged pH 6.6; CEC 7.1 cmol kg^{-1}; 903

ppm Ca; and P (Bray-P1), K, and Mg levels of 133, 145, and 47 mg kg^{-1}, respectively. Production practices during the experiment complied with National Organic Program guidelines (Agricultural Marketing Service, 2013). No fertilizers or soil amendments were applied in either year.

Treatment and Experimental Design

The treatment design was a 3-way factorial, with the factors including cover crop mixture, cash crop grown, and plastic mulch use. The experiment was arranged as a split-split plot RCBD with 4 replications, where the whole plot factor was rye-vetch winter cover crop treatment. Levels followed a proportional replacement series design (Jolliffe, 2000; Hayden et al., 2014), and included a gradient of the following sown proportions of rye:vetch (relative to their monoculture seeding rates): 100:0, 83:17, 67:33, 50:50, 33:67, 17:83, 0:100, in addition to a no cover crop control (0:0). The rye and vetch monocultures (100:0 and 0:100) were sown at 94 and 42 kg ha^{-1}, respectively. Following cover crop kill and incorporation in the spring, bell pepper and slicing cucumber were grown during the summer (as subplots) in each main plot, both on black plastic mulch and bare ground (sub-subplots).

Field Management

Table 1 summarizes the dates of key field activities and data collection during the study. Rye and vetch seed was broadcast sown by hand and incorporated to a depth of about 5 cm using a field cultivator. Whole plots were 6.7 by 8.5 m in 2009-2010 and 6.1 by 7.6 m in 2010-2011. Organic-certified variety not stated (VNS) vetch seed grown in Oregon and VNS rye seed grown in Minnesota were used in both years (Albert Lea Seed House, Albert Lea, MN). Vetch seed was inoculated with N-DURE *Rhizobium leguminosarum* inoculant (INTX Microbials LLC, Kentland, IN) at a rate of approximately 10 g inoculant kg^{-1} seed.

Cover crop stands were flail mowed and incorporated using a rototiller to a depth of about 15 cm in the spring, taking care to drive tractors slow enough to minimize residue movement. In 2010, residue incorporation was delayed by 1 wk following mowing due to heavy rainfall and subsequent wet soil conditions.

Approximately 2 wk after residue incorporation, fields were rototilled again prior to bed preparation. For plastic beds, black polyethylene mulch (1.25 mil thickness; Mid South Extrusion Inc., Monroe, LA) was laid on 75 cm wide raised beds (15 cm height) with a single drip irrigation line (30 cm emitter spacing) using a combined plastic mulch layer and bed shaper. Bare ground beds were similarly drip irrigated, but in keeping with general grower practices for un-mulched systems on sandy soils, beds were not raised. Plastic and bare ground beds within subplots were spaced on 1.22 m centers, while adjacent beds in separate main or subplots were on 1.83 m centers. All beds were at least 91 cm inside any main plot (cover crop treatment) border.

Bell pepper (var. ‘Paladin’) and slicing cucumber (var. ‘Cobra’) were grown from organic-certified seed (when available) in a greenhouse over the course of 9 and 2 wk prior to field transplanting, respectively. Pepper and cucumber transplants were grown in 98- and 72-cell plug trays, respectively, using a media composed of 50% peat moss, 25% vermiculite, and 25% plant-based compost by volume. Fish emulsion was applied for supplemental fertility as needed during transplant production. Transplants were hardened outdoors for 1 wk in a lath house prior to being transplanted into the field.

Peppers were grown in staggered double rows (46 cm in-row plant spacing and 30 cm inter-row spacing) with each treatment bed containing 20 data plants. Cucumbers were grown in single rows (46 cm plant spacing) with each treatment bed containing 12 data plants. All beds were drip irrigated simultaneously on a weekly basis and as needed, receiving approximately 2.5 cm per week during the summer. Crops were not fertilized, relying solely on N fertility derived from soil organic matter and incorporated cover crop residues.

Bare ground beds were maintained weed-free during the summer through hoeing every 2-3 wk within and between rows, and transplant holes in plastic beds were hand-weeded at those times as well. Preventative sprays of *Bacillus thuringiensis* (Dipel® DF, Valent BioSciences Corp., Libertyville, IL) were applied to peppers to limit damage from cutworms (family Noctuidae) and tomato hornworm (*Manduca quinquemaculata*), and pyrethrin (Pyganic®, McLaughlin Gormley King Company, Minneapolis, MN) applications were made to cucumbers to manage cucumber beetles (*Diabrotica spp.* and *Acalymma spp.*) prior to harvest.

Data Collection and Analysis

Cover Crop Residue Quantity and Quality

Rye and vetch shoot biomass was sampled in the spring from four 25- by 50-cm (0.125 m^{-2}) quadrats in each main plot, dried to a constant weight at 38 °C before measuring dry weights, and then ground to pass through a 1-mm screen. Subsamples were submitted to Midwest Laboratories (Omaha, NE) for analysis of total C and N concentrations using a Leco TruSpec elemental analyzer (Leco Corp., St. Joseph, MI). Vetch tissue dry weight and nutrient concentration data were corrected to account for soil adherence to vetch shoot samples in 2010, the details of which are discussed in Hayden et al. (2014). More extensive analysis of the influence of rye-vetch mixture proportions on cover crop stand establishment, biomass productivity, N fixation, and winter annual weed suppression are also presented.

Vegetable Crop Yields and Fruit Quality

Bell peppers were picked five times at approximately 1 wk intervals in both years. Fruit were counted and weighed after being categorized as either marketable (including U.S. Fancy, U.S. No. 1, and U.S. No. 2 grades) or non-marketable (deformed or damaged by sunscald, disease, decay, or insects) according to USDA standards (Agricultural Marketing Service, 2005). Cucumbers were picked nine times at approximately 3 d intervals in both years, and were similarly counted, weighed, and categorized. Marketable cucumber fruit included U.S. Fancy, U.S. Extra No. 1, U.S. No. 1, U.S. No. 1 Small, U.S. No. 1 Large, and U.S. No. 2 grades, while non-marketable fruit were deformed or damaged by cuts, scarring, disease, decay, or insects (Agricultural Marketing Service, 1958). Yields from individual harvests were summed to obtain total pepper and cucumber yields for each season. In the event that one or more data plants in a bed died at random prior to harvest but more than 2 wk after transplanting (6% of pepper beds lost a maximum of one plant and 27% of cucumber beds lost a maximum of up to two plants), total yield values were adjusted to a per 20- or 12- plant basis for pepper and cucumber, respectively. The average weight of marketable fruit was calculated by dividing total marketable fruit weights by total marketable fruit counts, and the percentage non-marketable yield by weight was obtained by dividing the total weight of non-marketable fruit by the combined weight of total marketable and non-marketable fruit. Pepper leaf absorbance readings were also taken at the time of flowering using a SPAD-502 Chlorophyll Meter (SPAD; Minolta, Osaka, Japan) as an estimate of chlorophyll content and plant N status. Readings were taken consistently on mature lower leaves that were axilary to the first branching of the plant stem.

Soil Inorganic Nitrogen Dynamics

Composite soil samples were collected from each whole plot prior to cover crop seeding in the fall and 1 wk after cover crop incorporation in the spring. Beginning 1 wk after vegetable bed preparation and plastic mulch application, composite soil samples (12 cores per bed) were taken from pepper beds every 2 wk during the growing season through mid-August. Cucumber beds were not sampled in the interest of time and cost of analysis. All soil samples were taken to a depth of 15 cm using a soil probe and stored at 4 ºC until analyzed. Soil was collected from beneath plastic mulch by cutting small slits prior to inserting the probe, resulting in only superficial damage to the mulch in terms of soil coverage, even by the end of the season. Post-cover-crop-incorporation soil samples were analyzed for inorganic N content. Soils were dried at 38 ºC for 36 h, 10 g dry soil were extracted in 50 ml 1M KCl, and extracts were analyzed for NO_3^- and NH_4^+ concentrations using a Lachat injection flow autoanalyzer (Lachat QuickChem, Hach Company, Loveland, CO).

Suction lysimeters were also installed in pepper beds as a way to estimate relative differences in nitrate leaching potential during the summer among plastic mulch and bare ground management following select cover crop treatments (100 V: 0 R, 50 V: 50 R, 0 V: 100 R, and 0:0). Each lysimeter consisted of a porous ceramic cup (4.8 cm outside diameter and 5.8 cm height) at the base of a 91 cm long PVC tube sealed at the top with a one-hole rubber stopper (Model 1900, SoilMoisture Equipment Corp., Santa Barabara, CA). Lysimeters were installed within the crop row roughly 15 cm from the drip line and with the porous cup at a depth of 0.6 m from the bed surface. Installation involved auguring a hole to the desired depth, inserting the lysimeter into a slurry of silica flour to surround the porous cup and improve hydraulic contact with the surrounding soil, adding bentonite clay pellets to form a seal around the PVC tube above the ceramic cup to limit preferential flow down the tube into the sampling area, and backfilling the remaining height around the lysimeter with reserved soil. Pore water samples were collected weekly during the summer beginning 1 wk after plastic mulch application. To collect the samples, a 35 cb vacuum was applied to each lysimeter using a vacuum hand pump generally 1 d following an irrigation event. The total volume of accumulated pore water was then removed and measured 18 h after vacuum application, and subsamples were frozen at -20 ºC until analysis for NO_3^- concentration on a Lachat injection flow autoanalyzer.

Soil Microbial Communities

Subsamples of soil collected 3 wk following bed preparation (21 and 20 June in 2010 and 2011, respectively) from plastic mulch and bare ground pepper beds following select cover crop treatments (100 V: 0 R, 50 V: 50 R, 0 V: 100 R, and 0:0) were sieved (4 mm) and used for microbial analyses. Soil microbial biomass carbon (MBC) was estimated by the chloroform fumigation-incubation (CFI) method (Jenkinson and Paulson, 1976; Paul et al., 1999). For each treatment replicate, 20 g of soil were weighed into six 50-ml beakers, three of which were fumigated with $CHCl_3$ (stabilized with non-polar hydrocarbons) for 24 h, and three of which served as non-fumigated controls. Soils in beakers were then re-inoculated with approximately 1 g of soil from their respective treatments and adjusted to 55% water holding capacity. Each beaker was incubated at 23 ºC for 10 d in a 1 L air-tight mason jar with a rubber septum in the lid. Headspace samples where extracted with a 1 ml syringe following incubation and analyzed for CO_2 concentration on an infrared gas analyzer (Qubit Model S-151, Qubit Systems Inc., Kingston, ON, Canada). Following Horwath et al. (1996), MBC was calculated as:

[1] $MBC = 1.73C_F - 0.56C_C$

where C_F and C_C are the evolved CO_2-carbon from fumigated and non-fumigated control beakers per g dry soil, respectively (average of three lab replicates). Soil respiration was taken as C_C, and metabolic quotient (qCO_2) was calculated by dividing MBC by soil respiration.

Differences in the functional diversity of soil microbial communities were evaluated based on patterns of substrate utilization by populations cultured on Biolog EcoPlates^{TM} (Biolog Inc., Hayward, CA). Each 96-well plate contains 31 distinct carbon sources and a control well (water) replicated three times. Following a procedure adapted from Garland and Mills (1991), 10 g field-moist soil from each treatment field replicate sample were shaken in 90 ml of a sterilized saline solution (0.85% NaCl, w/v) for 60 min, pre-incubated for 18 h at 23 ºC, and then brought to a 10^{-3} final dilution before adding 150 µl aliquots to each of the 96 wells (1 plate per treatment field replicate). Carbon source utilization is indicated by color change (clear to purple) due to the reduction of tetrazolium dye in the wells by respiratory CO_2. Plates were incubated at 23 C for 7 d, during which color development in each well was measured at 24 h intervals as absorbance at 590 nm using a microplate reader (Model 680, Bio-Rad Laboratories, Hercules, CA). The overall degree of substrate utilization was expressed as average well color development (AWCD), calculated as the mean difference between carbon source absorbances (R_i) and the absorbance reading for the control well (C) within plate replicates (Garland and Mills, 1991):

[2] $$AWCD = \frac{\sum R_i - C}{31}$$

Patterns of substrate utilization were investigated after first dividing the control-corrected absorbances (R_i - C) by AWCD within plate replicates to reduce potential bias due to differences in inoculum density among samples (Garland and Mills, 1991). Carbon source responses were then averaged based on categorization of the substrates into 6 chemical classes (Preston-Mafham et al., 2002) prior to principal component analysis.

Statistical Analysis

For cover crop residue variables, the fixed effects of year, cover crop mixture proportion, and their interaction were analyzed using mixed model ANOVA with the MIXED procedure of SAS (Version 9.2, SAS Institute, Cary, NC), where block (nested in year) was included as a random factor. Where the effects of cover crop mixture proportion on total residue N content and C:N were significant, the nature of each response within years was investigated using polynomial or linear regression with the REG procedure of SAS. For vegetable yield and quality variables and all univariately-analyzed microbial community variables, the fixed effects of year, cover crop treatment, plastic mulch use, and all interactions were analyzed as a split-plot design using mixed model ANOVA with the MIXED procedure of SAS. Block(year) and block(year)*cover crop treatment were included as random factors in the model. Where mulch*cover crop and higher order interactions were not significant ($\alpha = 0.05$), only main effects and marginal means of mulch use and cover crop treatment within years are presented (Tables 3, 4, and 5). Total soil inorganic N and pore water NO_3^- concentrations during the summer were analyzed using repeated measures mixed model ANOVA with the MIXED procedure of SAS. Model structure was similar to above, but with years analyzed separately due to model complexity, and sample date included as the repeated factor. Due to the even temporal spacing of the measurements, a first-order autoregressive variance-covariance structure was used. For all of the above analyses, residuals were inspected to evaluate consistency with ANOVA assumptions of normality and equality of variances, and data transformations (natural log or square root) and unequal variance models were employed when necessary. Effects were judged significant when $P < 0.05$.

Principal components analysis (PCA) was conducted on Biolog^{TM} data using the correlation matrix of the six substrate class absorbance variables. Data for each year were analyzed separately, and plate replicates were considered subsamples and averaged prior to analysis. The first two principal component (PC) axes accounted for a majority of the total variance in both years, and PC scores labeled by treatments were plotted on these axes to evaluate patterns of substrate utilization. Correlations (loadings) between the PC axes and the original substrate class variables were plotted as vectors, with correlations > |0.50| considered significant.

RESULTS

Weather Conditions

Daily air temperature and rainfall data from April through September during the two years of the experiment are presented in Figure 1. Spring temperatures during cover crop growth were cooler in 2011 than in 2010. However, during the period of vegetable production, average temperatures were similar in the two years (21.7 and 21.5 ºC in 2010 and 2011, respectively), with 1284 and 1232 growing degree days (GDD) base 10 ºC accumulating between pepper transplanting and last harvest in the two seasons. While cumulative rainfall during that period was higher in 2011 than in 2010 (204 and 286 mm in 2010 and 2011, respectively), 56% of the summer 2011 precipitation fell over a period of just 3 d near the start of pepper harvests. In contrast, during the first month following plastic application, fields received 106 mm of rainfall in 2010, compared with only 44 mm in 2011.

Rye-vetch Mixture Residues

Rye and vetch shoot dry weights were highly correlated with the proportion of each species sown in mixture (Table 2). Biomass production was highest for each species when grown in monoculture, with vetch yielding 551 and 305 g m^{-2} in 2010 and 2011, respectively, and rye yielding 415 and 330 g m^{-2} (Table 2). In 2010, total shoot biomass (vetch+rye) in mixtures were equivalent or intermediate to yields in the two monocultures, while in 2011, three mixtures (83 V: 17R, 67V: 33R, and 33V: 17R) produced significantly greater total biomass than either monoculture (Table 2).

The shifts in cereal-legume composition across mixtures influenced the quality of the combined cover crop residues. Total residue N content generally increased with greater proportions of vetch in mixture (Figure 2). The increase was linear in 2010, when vetch monoculture residues contained 175 kg N ha^{-1}. In 2011, increases in total N leveled off for cover crops containing greater than 50% vetch, due in part to reduced vetch biomass productivity and greater rye tissue N concentrations that year (Hayden et al., 2014). Correspondingly, total residue C:N increased with greater proportions of rye in mixture in both years, but still remained below 25:1 for all cover crop treatments except the rye monoculture.

Vegetable Yields and Fruit Quality

Marketable yields (weight) of both bell pepper and slicing cucumber were generally higher following cover crop mixtures that contained greater proportions of vetch (Figure 3). Yield increases were predominately linear in 2011, while in 2010, the relative gains diminished or ceased for mixtures containing more than 33 or 50% vetch. With the exception of treatments containing greater than 83% rye, cover crops generally provided a significant yield benefit relative to the no-cover-crop control. However, for vegetables grown on bare ground in 2010, yield benefits even from high vetch mixtures were comparably modest and not statistically significant. Overall, yields were higher for peppers and cucumbers grown on plastic mulch than on bare ground, but the magnitude of the yield benefits differed between the two study years. In 2010, plastic mulch yields were 155% and 152% higher than bare ground on average ($P<0.001$) for pepper and cucumber, respectively, but only 77% ($P<0.001$) and 9% ($P = 0.04$) higher in 2011.

Differences in the number of marketable pepper (Table 3) and cucumber (Table 4) fruit produced largely mirrored the trends observed with marketable weights. The use of plastic mulch and cover crops containing greater proportions of vetch were associated with modest increases in average pepper and cucumber fruit weights in some cases (Tables 3 and 4). While cover crop treatment had a significant effect in 2011 on the percentage of pepper yields that were non-marketable, it had no significant influence on cucumber non-marketable percentage in either year (Tables 3 and 4). Furthermore, plastic mulch use reduced pepper non-marketable percentage from 9.2 to 4.7% in 2010 and from 19.6 to 15.3% in 2011 (Table 3). In contrast, cucumber non-marketable percentages actually increased when grown on plastic, from 15.6 to 20.9% in 2010 and from 19.1 to 22.5% in 2011 (Table 4).

Soil Inorganic Nitrogen

Average soil inorganic N availability during the summer in pepper beds was highly correlated with both marketable yields (Figure 4a) and SPAD chlorophyll content readings (Figure 4b) over the course of the study. Cover crops containing greater proportions of vetch generally resulted in higher soil inorganic N concentrations, with levels for rye monocultures remaining less than or equivalent to the no-cover-crop control (Figure 5). Soil N levels increased during the first several weeks after residue incorporation and then fell as pepper uptake increased in late June. No significant differences in the timing of N release could be detected among the varying cover crop residues (Figure 5). Plastic mulch was overall associated with higher soil N levels, but the effect varied considerably between years. In 2010, the average inorganic N concentration during the summer under bare ground management was 56% less than under plastic mulch (Figure 5 a, b; $P<0.001$). In contrast, average bare ground soil N was only 19% less than plastic mulch in 2011 ($P<0.001$), with no significant differences between the two practices on 3 of the 6 sampling dates following plastic application (June 6, July 6, and July 18;

Figure 5 c, d), compared with no dates in 2010 (Figure 5 a, b).

Lysimeter Pore Water Nitrate Concentrations

Similar to concentrations of total inorganic N in the top 15 cm of soil, the concentration of nitrate in pore water extracted from 0.6 m depth with lysimeters was generally higher following cover crop residues containing greater proportions of vetch (Figure 6). Nitrate concentrations following rye monocultures were less than or equivalent to no-cover-crop controls. Under plastic mulch, the changes in pore water nitrate over the course of the summer displayed a pattern comparable to N availability in the overlying soil—increasing early in the season and then decreasing with greater pepper uptake later (Figure 6 a, b). Under bare ground, pore water nitrate concentrations showed influence from precipitation events, particularly in 2011 (Figure 6 b, d). As with soil N levels, plastic mulch had higher pore water nitrate concentrations at depth than bare ground. The effect varied between years as well, but in a manner opposite to that observed with the upper layer soil N. In 2010, average bare ground pore water nitrate concentrations were 10% less than plastic mulch (Figure 6 a, b; $P<0.001$), while in 2011, nitrate concentrations were 24% lower under bare ground—56% lower if sampling dates following the 3 d, 160 mm rainfall event in late July are excluded (Figure 6 c, d; $P<0.001$).

Microbial Communities

The incorporation of rye-vetch cover crops resulted in equivalent or greater MBC, soil respiration, and metabolic quotient than the no-cover-crop control on the dates microbial communities were sampled in 2010 and 2011 (Table 5). Where differences among the cover crop treatments were significant, values tended to be higher following the rye monoculture or the rye-vetch mixture than following the vetch monoculture. In both years, MBC and soil respiration were higher for soils under bare ground management than under plastic mulch (Table

5). However, the metabolic quotient was not influenced by the mulching practice.

Principal component analyses of data from Biolog Ecoplates^{TM} revealed variation in microbial community substrate utilization across soils sampled during the experiment (Figure 7). The percentage of total variance captured on PC 1 was 39 and 38% in 2010 and 2011, respectively, and PC 2 represented 21 and 24% in the two years. In both years, PC 1 was positively correlated with utilization of substrates classified as polymers and miscellaneous, while negatively correlated with carboxylic acids. The predominant loadings for PC 2 were carbohydrates (r=-0.67) and amines/amides (r=0.73) in 2010, but included carbohydrates (r=0.89) and amino acids (r=-0.79) in 2011 (Figure 7 b, d). Despite variation across samples, considerable overlap in principal component scores was observed among cover crop treatments and between plastic mulch and bare ground management, with no clear distinctions in microbial community substrate utilization resolved in either year. However, microbial communities in no-cover-crop control soils exhibited greater variability than cover crop treatments along PC 2 in 2010 (Figure 7a) and along PC 1 in 2011 (Figure 7c).

DISCUSSION

Rye-Vetch Residue Effects on N Dynamics and Vegetable Yields

The availability of N from cover crop biomass depends in large part on the characteristics of the residues. While the prevalence of specific classes of compounds (i.e., cellulose, hemicellulose, lignins, polyphenols) can have important impacts on N mineralization (Crews and Peoples, 2005), first-season N availability is broadly related to total residue N and C content. Laboratory incubation studies suggest that the percentage of total N in incorporated vetch and vetch-cereal residues that becomes available as inorganic N in the first season is likely at maximum 50-60%, and decreases with increasing total residue C:N in mixtures (Vigil and Kissel, 1991; Honeycutt et al., 1993; Garrett, 2009), with residues that have total C:N > 25 potentially resulting in short term net N immobilization (Allison, 1966). Our results generally conformed to these principles, with rye-vetch residues that contained more total N and had lower C:N leading to higher early-season soil inorganic N levels, and inorganic N following rye monocultures remaining less than or equivalent to the background levels found in the no-cover-crop control. Not surprisingly, increasing inorganic N levels in the plow layer also translated into higher pore water nitrate concentrations below the root zone (Figure 6). Similarly, Rosecrance et al. (2000) and Tosti et al. (2014) found higher N leaching from vetch residues than from rye or barley alone or in mixture with vetch, and Campiglia et al. (2010) observed a linear relationship between total N content of vetch and vetch-oat residues and the amount leached under the cover crops when used as mulches during pepper production.

Mineralization from high N cover crop residues occurs rapidly following incorporation, typically resulting in an excess of soil inorganic N vulnerable to leaching in the weeks prior to peak warm-season crop demand (Ranells and Wagger, 1996). It is often proposed that the combination of contrasting residue qualities in cereal-legume mixtures may alter patterns of N release to mitigate this asynchrony (Crews and Peoples, 2005). Without information on pepper N uptake during the growing season, data on soil inorganic N concentrations during the summer within the pepper rows alone are insufficient for resolving differences in temporal patterns in net N mineralization following the various rye-vetch mixtures. Nevertheless, rapid mineralization leading to excess early-season N was evident following all cover crop treatments in this study that exhibited net N mineralization (Figure 5). Research suggests that combinations of residues of contrasting quality most often exhibit patterns of net N mineralization that are more or less the weighted average of the patterns exhibited by the high and low quality residues individually, rather than resulting in release that is notably more synchronous with crop demand (Palm et al., 2001). Over the course of a 30 wk laboratory incubation, Kuo and Sainju (1998) tracked net N mineralization from mulched rye-vetch residue combinations along a gradient from 100% vetch to 100% rye. While early season mineralization rates varied within a narrow range between the two extremes, the cumulative amount of N mineralized followed the total N content in the mixtures. In replacement-style mixtures (where a proportion of the high N component is replaced with an equivalent proportion of the low N component), the resulting reductions in total N and lower percent availability in the first season will likely have much greater relevance for warm-season crop N nutrition than the magnitude of any potential changes in patterns of net N mineralization. However, in the event that species interactions or management result in a rye-vetch mixture accumulating significantly greater total N than a vetch monoculture (Sainju et al., 2005), the relative reduction in net N mineralization could have the benefit of increasing soil N retention without limiting potential N fertility (Lawson et al., 2013).

The correlations between pepper marketable yields, SPAD readings, and average soil inorganic N support that N fertility was a key driver of treatment effects on vegetable yields (Figure 4). In 2011, both pepper and cucumber exhibited predominately linear increases in marketable yield following cover crops containing greater proportions of vetch. Despite cover crop residues containing more total N than in 2011, relative yield gains in 2010 diminished with increasing vetch proportion. A number of possibilities could account for this difference. Nitrogen requirements may have been largely met by levels following the 50:50 rye:vetch mixture within the context of the 2010 field, or another limiting factor not measured may have prevented yield gains with increasing N availability that year. Alternatively, differences in cover crop management between the timing of termination and plastic application in the two years could have led to greater N loss from high vetch residues in 2010. Cover crops were mowed and incorporated 2 wk before plastic application in 2011, while in 2010, cover crops were mowed 3 wk before plastic application and the mowed residues were left on the soil surface during the first wk after termination because heavy rainfall prevented tillage. Preferential N leaching, denitrification (Rosecrance et al., 2000), or ammonia volatilization (Quemada and Cabrera, 1995; Crews and Peoples, 2004) from higher vetch cover crop residues during this extended time could have reduced the relative total season N availability from these treatments. However, the close association between vetch proportion and soil inorganic N levels still observed early in the 2010 season (Figure 5a) would appear to contradict this explanation.

Plastic Mulch Effects

The higher soil inorganic N concentrations observed under plastic, as well as the close correspondence between relative yield benefits and differences in N levels between plastic and bare ground treatments during the two study years, suggest that N fertility was an important factor behind the yield increases observed with plastic mulch. In regions with cooler summers, black polyethylene mulch has the potential to promote greater N mineralization by warming the soil, and when paired with drip irrigation, by maintaining more consistent soil moisture conditions (Tarara, 2000; Agehara and Warncke, 2005). The process of soil redistribution in forming raised beds in a plastic mulch system may also serve to concentrate incorporated residues from a wider area into the crop root zone, providing proportionally greater cover-crop-derived N fertility localized beneath the crop relative to a flat bed. In addition, plastic mulch protects inorganic N in the crop root zone from leaching during precipitation events, which can be considerable on sandy soils like those present at the experimental site (Clarkson, 1960).

Differences in the amount and timing of precipitation in 2010 and 2011 may account for the differences in N availability and yields between plastic mulched and bare ground pepper beds in the two years. In 2010, twice as much precipitation fell in the first month after plastic application than in 2011, likely leading to relatively greater leaching from bare ground beds that year. Indeed, a substantial drop in soil N concentrations occurred only in bare ground beds following 56 mm of rain in the 6 d after plastic application in 2010, and levels remained lower than plastic mulch for the rest of the summer. Correspondingly, pore water N concentrations below bare ground beds were relatively higher in 2010 than in 2011.

Interestingly, pore water nitrate concentrations at depth were generally equivalent or higher under plastic mulch than bare ground management. This indicates that there was still considerable potential for leaching losses from plastic beds under our drip irrigation regime. In this study, irrigation was applied uniformly across the fields to meet the requirements of crops grown on bare ground, but a lower rate for plastic mulch beds may have been able to satisfy crop demands while reducing the depth of irrigation (Quemada et al., 2013). The potential for leaching due to precipitation events is clearly greater in the absence of plastic mulch, as evinced by nitrate concentrations under bare ground following heavy rains in 2011 (Figure 6d). However, data from suction lysimeters provide an incomplete picture of total leaching losses. Even with accurate estimates of drainage through the soil profile, suction lysimeters generally sample resident pore water solute concentrations rather than flux concentrations (Brandi-Dohrn et al., 1996), with the potential to miss high concentrations of N leached below the sampling depth prior to pore water extraction. As a result, suction lysimeters tend to underestimate quantities of leached N compared with other methods, including drainage lysimeters (Zotarelli et al., 2007). This bias should be more pronounced for soils exposed to heavy rain events that infiltrate below the root zone, suggesting our data may underestimate the N leaching potential of bare ground relative to plastic mulch management.

Even when N is not limiting, the temperature and moisture microclimate effects of black plastic mulch can promote more rapid growth and development of warm-season vegetable crops leading to higher and earlier yields (Kasirajan and Ngouajio, 2012), although reaching temperatures above crop-specific thresholds can be detrimental (Tarara, 2000). Plastic mulches can also influence yield quality by promoting cleaner fruit and reducing the incidence of soilborne disease, in addition to quality benefits related to water and fertility (Kasirajan and Ngouajio, 2012). The lower percentage of non-marketable pepper fruit with plastic mulch and with greater proportions of vetch in cover crop mixtures suggest that quality improvement in pepper was related to N availability. However, the relationship may have been indirect—solar injury (sunscald) was the predominant pepper fruit defect during the study, and shading from fuller plant canopies under higher N fertility may have reduced incidence (Roberts and Anderson, 1994). The increase in non-marketable percentage for cucumbers grown on plastic in both years was surprising, particularly for a crop whose fruit rest on the soil surface. The lack of cover crop effect on cucumber quality suggests that differences were not driven by N fertility. The predominant cucumber defects were deformed or misshapen fruit and excessive scarring from cucumber beetle damage. Surface temperature highs may be elevated by up to 20 ºC on black plastic mulch relative to bare ground (Tarara, 2000), and we speculate that these higher temperatures may have contributed to more incomplete pollination in cucumbers, either by directly harming pollen growth or survival or by discouraging pollinators, leading to higher incidence of deformed fruit (Maestro and Alvarez, 1988; Swiader and Ware, 2002). The less extreme temperature increases typical >2.5 cm above the surface of black plastic may also have promoted greater reproduction and/or activity of cucumber beetles (Radin and Drummond, 1994; Tarara, 2000).

Responses of Soil Microbial Communities

Soil microbes are typically carbon limited, and the addition of organic residues generally prompts an increase in microbial activity and growth proportional to the quantity of labile C added, which slows as the materials are decomposed over time (Jackson, 2000; Paul, 2007). The tendency towards higher MBC and soil respiration following rye or rye-vetch cover crops, particularly in 2011, is most likely related to larger total C additions. The metabolic quotient is a measure of the amount of C respired per unit microbial biomass, or the efficiency of microbial C utilization. Higher metabolic quotients are often considered an indication of stress, suggesting that more carbon is being allocated to energy and maintenance functions (and thus respired) rather than contributing to growth (Anderson and Domsch, 1993), although this interpretation has been questioned (Wardle and Ghani, 1995). Larger metabolic quotients following rye or rye-vetch mixtures than vetch alone may reflect a greater degree of N limitation for microbes relative to the quantity of carbon added (Wardle and Ghani, 1995). Nair and Ngouajio (2012) saw a similar response to increasing carbon additions with incorporation of rye and vetch cover crops and compost.

Overall, plastic mulch exhibited higher MBC than bare ground, which likely accounted for the correspondingly higher soil respirations also observed (Table 5). Moreno and Moreno (2008) also found reduced indicators of microbial biomass following black polyethylene mulch, which they suggested may be a result of inhibition due to higher temperatures. Alternatively, one might expect that moderate soil warming under plastic would lead to increased microbial activity and population growth (Nicolardot et al., 1994), and the lack of differences in metabolic quotient between plastic and bare ground treatments in our study do not support that microbes were under particular stress under plastic. Li et al. (2004) documented higher MBC with plastic mulching relative to bare ground, although differences varied depending on the time after plastic application in one year. Another possible explanation is that conditions under plastic mulching may alter the temporal dynamics of microbial growth, such that comparisons of MBC with bare ground may vary depending on the date of sampling.

We hypothesized that incorporation of cover crop residues of differing quality and the fertility and microclimate effects related to plastic mulch use would result in shifts in the functional diversity of microorganisms present. While variation in substrate utilization by the communities on Biolog Ecoplates^{TM} was evident, distinct treatment effects were not resolved by our analysis (Figure 7). By contrast, previous studies have documented changes in microbial community composition in the first season as a result of cover cropping and plastic mulching (Carrera et al., 2007; Wu et al., 2009; Nair and Ngouajio, 2012). However, results from Buyer et al. (2010) comparing in part a vetch residue mulch with black and white plastic mulches during tomato production suggest that organic inputs from cover cropping are more important than management induced variations in temperature and moisture for controlling microbial community structure. Utilizing fatty acid and DNA marker techniques, distinctions in community composition have been identified specifically between soils following rye and vetch cover cropping as well (Buyer et al., 2010; Maul et al., 2014). The greater variability we observed for substrate utilization in no-cover-crop treatments in both years was an interesting finding. Those treatments also tended to have the lowest MBC, so one possible explanation for the greater variability is that there was a higher probability of losing rare species of functional significance during dilution of the no-cover-crop relative to the cover crop treatments (Preston-

Mafham et al., 2002).

The Biolog^{TM} assay is a culture-based technique, and has been demonstrated to select for certain components of the microbial community over others—particularly favoring rapidly-growing (r-strategist) species (Ros et al., 2008). In addition, due to the great diversity of microbes in the soil, communities can exhibit considerable functional redundancy (Strickland et al., 2009), which may further limit the ability of substrate utilization approaches to resolve differences in microbial communities even if structural changes are present, particularly in the short term. For example, Carrera et al., (2007) found relatively weaker cover cropping effects with substrate utilization data than with analysis of phospholipid fatty acids. These results and our data support that potential changes in microbial community composition resulting from rye and vetch cover crop incorporation or mulching with black plastic film in the first season (short term) may have limited significance for actual agroecosystem functioning.

Management Implications

Rye-vetch cover crops containing greater proportions of vetch can be expected to result in greater soil N availability following incorporation, and when N is limiting, to lead to higher yields of bell pepper and slicing cucumber. Along the gradient of rye-vetch proportions evaluated in this study, changes in total N content appeared to have a larger influence on vegetable yields than did potential changes in patterns of N mineralization. However, Hayden et al. (2014) demonstrated that greater proportions of vetch also resulted in increased seed costs and tradeoffs with other agroecosystem services, such as winter annual weed suppression. The optimal rye-vetch mixture proportion will ultimately depend on cover crop objectives, crop management practices, and site-specific characteristics of the field.

Regardless of rye-vetch proportion, net N that mineralized from mixtures accumulated in the soil prior to peak vegetable demand, leaving it vulnerable to leaching from early-season precipitation, particularly on the sandy soils used in this study. Our results demonstrate that plastic mulch can be an important tool for preserving N fertility benefits from high N cover crop residues, contributing substantially to the yield increases observed relative to bare ground management. Where mulch film is used following cover crop incorporation, it should be applied as soon as residue conditions allow to maximize N preservation. Although the benefits of plastic mulch must be weighed against the costs associated with its use and disposal, the importance of protecting soils against N leaching will likely grow over time due to predicted increases in extreme precipitation events in many regions (Hayhoe et al., 2006).

APPENDIX

Table 3.1. Dates of key field activities and data collection.

Activity	2009-2010	2010-2011
Cover crop seeding	1 September	1 September
Cover crop biomass sampling	10 May	14 May
Cover crop termination (by flail mower)	10 May	17 May
Cover crop incorporation (by rototiller)	17 May	17 May
Plastic mulch application	1 June	31 May
Bell pepper transplanting	3 June	2 June
Slicing cucumber transplanting	3 June	6 June
Suction lysimeter installation	8 June	7 June
Bell pepper SPAD† readings	12-13 July	15 July
Slicing cucumber harvests (9)	12 July - 2 August	18 July - 5 August
Bell pepper harvests (5)	27 July - 31 August	27 July - 29 August

† Minolta SPAD-502 Chlorophyll Meter

Table 3.2. Shoot residue dry weight of vetch, rye, and vetch+rye (total) across cover crop mixture proportions in 2010 and 2011.

Cover Crop Treatment	Vetch	Rye	Total
		g m^{-2}	
2010 Season			
100 V : 0 R	550.9 (64.9)†	--	550.9 (64.9)
83 V : 17 R	400.5 (67.5)	217.7 (65.0)	618.2 (51.3)
67 V : 33 R	379.5 (27.2)	216.2 (29.1)	595.7 (23.0)
50 V : 50 R	270.6 (68.2)	235.4 (46.2)	506.0 (113.2)
33 V : 67 R	199.2 (22.6)	349.4 (14.9)	548.6 (32.9)
17 V : 83 R	117.8 (16.1)	387 (26.2)	504.8 (22.9)
0 V : 100 R	--	414.5 (58.8)	414.5 (58.8)
Prob > F‡	< 0.001	0.002	0.30
2011 Season			
100 V : 0 R	305.0 (22.7)	--	305.0 (22.7)
83 V : 17 R	294.4 (16.8)	128.3 (12.5)	422.7 (20.3)
67 V : 33 R	274.9 (27.5)	167.0 (11.5)	441.9 (34.4)
50 V : 50 R	156.3 (32.0)	200.7 (12.3)	357.0 (35.5)
33 V : 67 R	165.8 (34.6)	266.3 (32.4)	432.1 (53.8)
17 V : 83 R	90.8 (20.6)	267.8 (13.0)	358.5 (20.8)
0 V : 100 R	--	330.2 (31.0)	330.2 (31.0)
Prob > F‡	< 0.001	< 0.001	0.01

† Treatment mean (standard error)

‡ Significance of fixed effect of cover crop mixture proportion within years (*P* value).

Table 3.3. Bell pepper total number of marketable fruit harvested, average weight of marketable fruit, and percentage non-marketable yield by weight when grown either on black plastic mulch or bare ground (no plastic) following 7 rye-vetch cover crop mixture proportions or a no cover crop control in 2010 and 2011.

Treatment	Number of Marketable Fruit		Average Weight of Marketable Fruit		Percentage Non-Marketable Weight	
	2010	2011	2010	2011	2010	2011
	number 20-plants^{-1}		g		%	
Mulch Main Effect†						
No plastic	28 (3)‡	54 (4)	184 (2)	185 (2)	9.2 (1.1)	19.6 (1.7)
Plastic Mulch	68 (4)	73 (4)	190 (2)	190 (2)	4.7 (0.5)	15.3 (1.1)
Prob > *F*§	< 0.001	< 0.001	< 0.001	< 0.001	0.006	0.009
Cover Crop Main Effect†						
Control	38 (8)	39 (6)	182 (2)	173 (4)	8.3 (1.5)	22.2 (2.7)
100 V : 0 R	57 (12)	86 (8)	191 (3)	197 (4)	6.0 (1.7)	12.4 (1.4)
83 V : 17 R	62 (9)	75 (10)	196 (3)	197 (3)	4.4 (0.7)	17.1 (4.6)
67 V : 33 R	52 (10)	83 (6)	186 (2)	196 (3)	6.8 (2.2)	13.0 (1.0)
50 V : 50 R	50 (10)	55 (6)	186 (4)	186 (4)	4.3 (1.0)	15.8 (2.0)
33 V : 67 R	56 (11)	65 (8)	186 (3)	189 (6)	5.6 (1.2)	18.3 (4.0)
17 V : 83 R	44 (8)	51 (6)	189 (3)	184 (1)	7.2 (1.7)	20.9 (1.8)
0 V : 100 R	25 (7)	51 (5)	181 (4)	178 (4)	13.1 (3.2)	19.8 (2.7)
Prob > *F*§	0.03	< 0.001	0.17	< 0.001	0.21	< 0.001

† Main effects and marginal means are presented because mulch*cover crop and higher order interactions were not significant at the 0.05 probability level.

‡ Marginal mean (standard error)

§ Significance of fixed main effects within years (*P* value).

Table 3.4. Slicing cucumber total number of marketable fruit harvested, average weight of marketable fruit, and percentage non-marketable yield by weight when grown either on black plastic mulch or bare ground (no plastic) following 7 rye-vetch cover crop mixture proportions or a no cover crop control in 2010 and 2011.

Treatment	Number of Marketable Fruit		Average Weight of Marketable Fruit		Percentage Non-Marketable Weight	
	2010	2011	2010	2011	2010	2011
	number 20-plants^{-1}		g		%	
Mulch Main Effect†						
No plastic	29 (2)‡	71 (4)	241 (4)	283 (2)	15.6 (1.6)	19.1 (0.8)
Plastic Mulch	66 (4)	76 (4)	268 (2)	290 (2)	20.9 (1.4)	22.5 (0.7)
Prob > *F*§	< 0.001	0.10	< 0.001	0.07	0.01	< 0.001
Cover Crop Main Effect†						
Control	32 (7)	50 (4)	251 (7)	283 (3)	22.3 (3.8)	21.2 (1.3)
100 V : 0 R	59 (8)	97 (6)	256 (7)	284 (3)	17.5 (2.1)	21.5 (1.3)
83 V : 17 R	60 (9)	90 (5)	253 (11)	292 (4)	15.5 (2.5)	19.5 (1.4)
67 V : 33 R	55 (9)	92 (5)	265 (7)	291 (2)	15.6 (2.4)	22.4 (1.8)
50 V : 50 R	57 (10)	66 (7)	254 (7)	281 (6)	16.7 (2.9)	19.0 (1.3)
33 V : 67 R	49 (9)	80 (8)	253 (7)	292 (3)	16.4 (2.5)	19.8 (2.7)
17 V : 83 R	41 (7)	60 (4)	254 (6)	286 (5)	16.8 (2.7)	20.8 (1.7)
0 V : 100 R	26 (7)	55 (3)	252 (11)	283 (6)	25.0 (4.9)	22.0 (1.3)
Prob > *F*§	< 0.001	< 0.001	0.23	0.27	0.33	0.86

† Main effects and marginal means are presented because mulch*cover crop and higher order interactions were not significant at the 0.05 probability level.

‡ Marginal mean (standard error)

§ Significance of fixed main effects within years (*P* value).

Table 3.5. Microbial biomass carbon (MBC), soil respiration, and metabolic quotient in soils sampled from pepper rows grown either on black plastic mulch or bare ground (no plastic) following select rye-vetch cover crop mixture proportions or a no cover crop control. Soils were sampled on 21 and 20 June in 2010 and 2011, respectively.

Treatment	MBC		Soil Respiration		Metabolic Quotient	
	2010	2011	2010	2011	2010	2011
	ug CO_2-C g^{-1} dry soil		ug CO_2-C g^{-1} dry soil			
Mulch Main Effect†						
No plastic	105 (4)‡	118 (5)	67 (2)	76 (4)	0.65 (0.02)	0.65 (0.03)
Plastic Mulch	95 (4)	105 (4)	63 (3)	69 (3)	0.66 (0.02)	0.66 (0.03)
Prob > F§	0.003	< 0.001	0.12	0.02	0.58	0.78
Cover Crop Main Effect†						
Control	91 (6)	103 (7)	57 (4)	64 (5)	0.63 (0.02)	0.63 (0.04)
100 V : 0 R	102 (5)	117 (5)	62 (4)	69 (4)	0.61 (0.03)	0.59 (0.03)
50 V : 50 R	102 (6)	100 (7)	69 (4)	73 (3)	0.68 (0.03)	0.74 (0.04)
0 V : 100 R	103 (9)	126 (6)	71 (5)	85 (4)	0.71 (0.03)	0.68 (0.03)
Prob > F§	0.65	0.07	0.13	0.02	0.10	0.007

† Main effects and marginal means are presented because mulch*cover crop and higher order interactions were not significant at the 0.05 probability level.

‡ Marginal mean (standard error)

§ Significance of fixed main effects within years (*P* value).

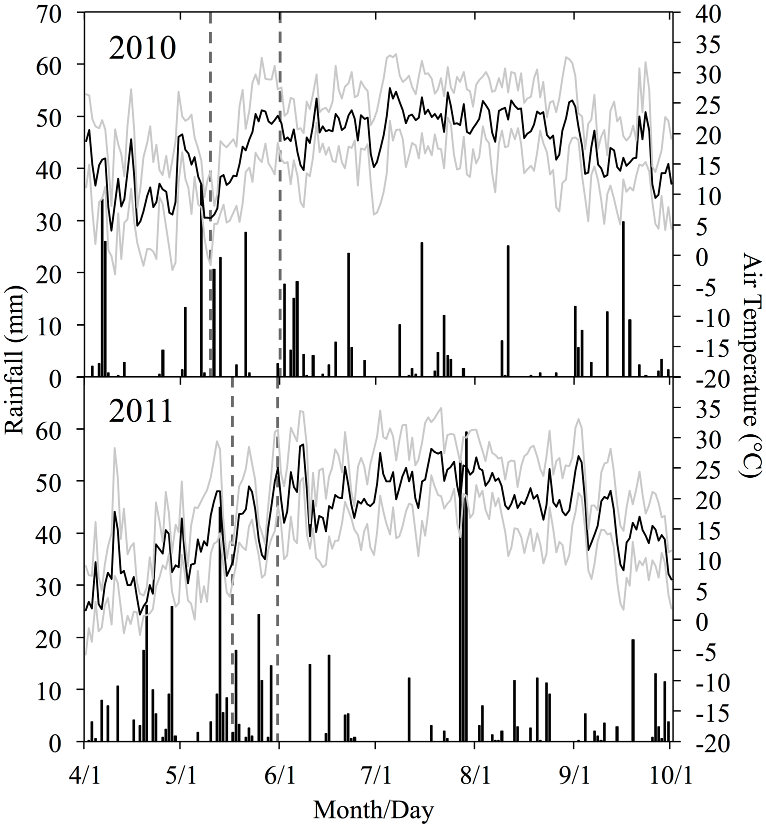

Figure 3.1. Daily rainfall amounts (bars) and minimum, maximum, and average daily air temperatures (lines) at the Michigan State University Horticulture Teaching and Research Center (Holt, Michigan) from April through September in 2010 and 2011. The first and second dotted lines denote the dates of cover crop termination and plastic mulch application, respectively, in each year.

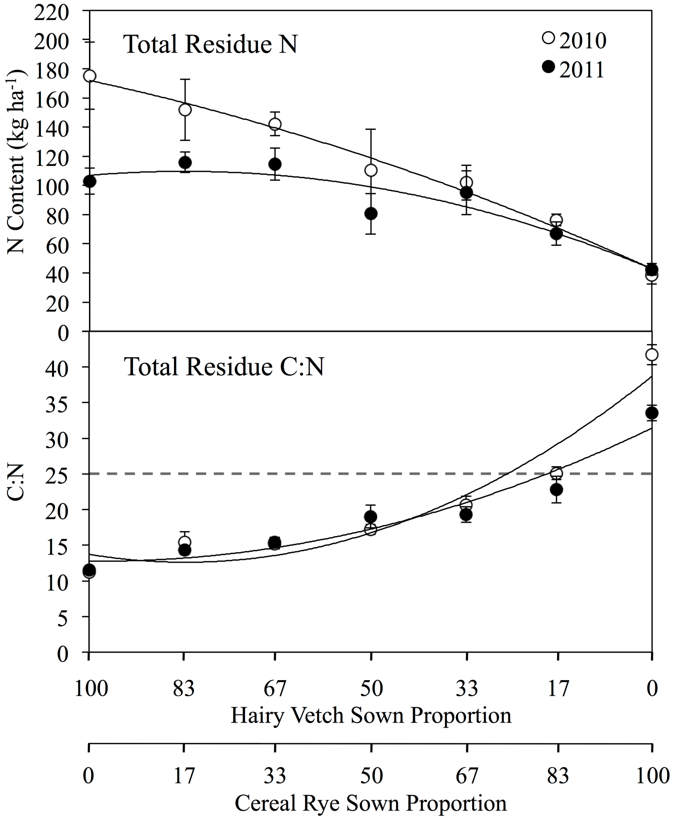

Figure 3.2. Total (vetch+rye) shoot residue N content (a) and C:N ratio (b) across cover crop mixture proportions in 2010 (open circles) and 2011 (closed circles). The horizontal dotted line references a C:N value of 25, a common estimate for the cutoff between expectations of net N mineralization and immobilization in soils following residue incorporation. Means +/- SE. The responses of total N content in 2010: y=-1.3x+178.3, r^2=0.67, P<0.001; and 2011: y=-0.0097x^2+0.32x+106.9, r^2=0.58, P<0.001. The responses of C:N in 2010: 0.0038x^2-0.13x+13.73, r^2=0.90, P<0.001; and 2011: 0.0019x^2-0.01x+12.77, r^2=0.85, P<0.001.

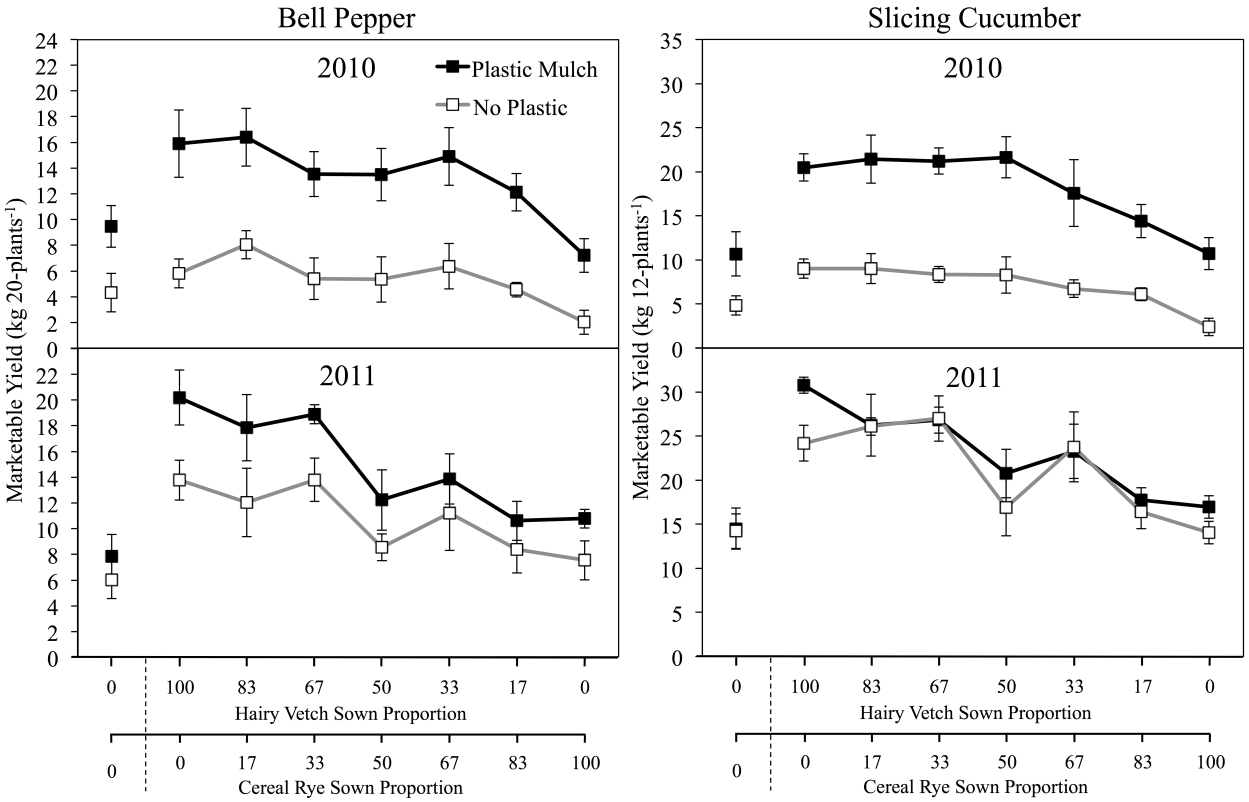

Figure 3.3. Total marketable yields of bell pepper (a, c) and slicing cucumber (b, d) when grown either on black plastic mulch (black squares) or bare ground (white squares) following 7 rye-vetch cover crop mixture proportions or a no cover crop control (0:0) in 2010 (a, b) and 2011 (c, d). Means +/- SE.

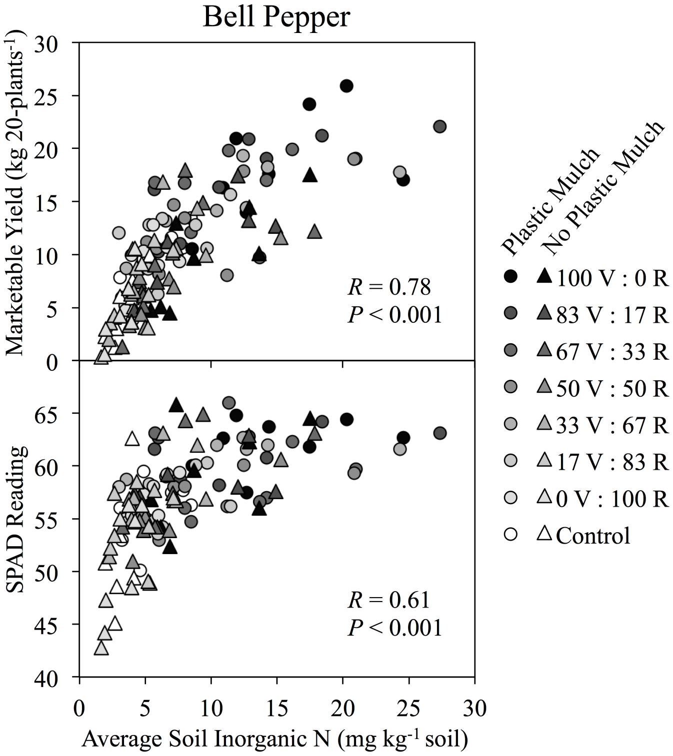

Figure 3.4. Correlations between the average soil inorganic N concentration in bell pepper rows during the summer and a) total bell pepper marketable yield and b) bell pepper SPAD meter leaf absorbance readings (dimensionless units) for 2010 and 2011 data combined. Rye-vetch cover crop mixture treatments are designated by grayscale gradient, while black plastic mulch and bare ground treatments are differentiated by shape. Pearson correlation coefficients (*R*) and their significance are indicated on the plots.

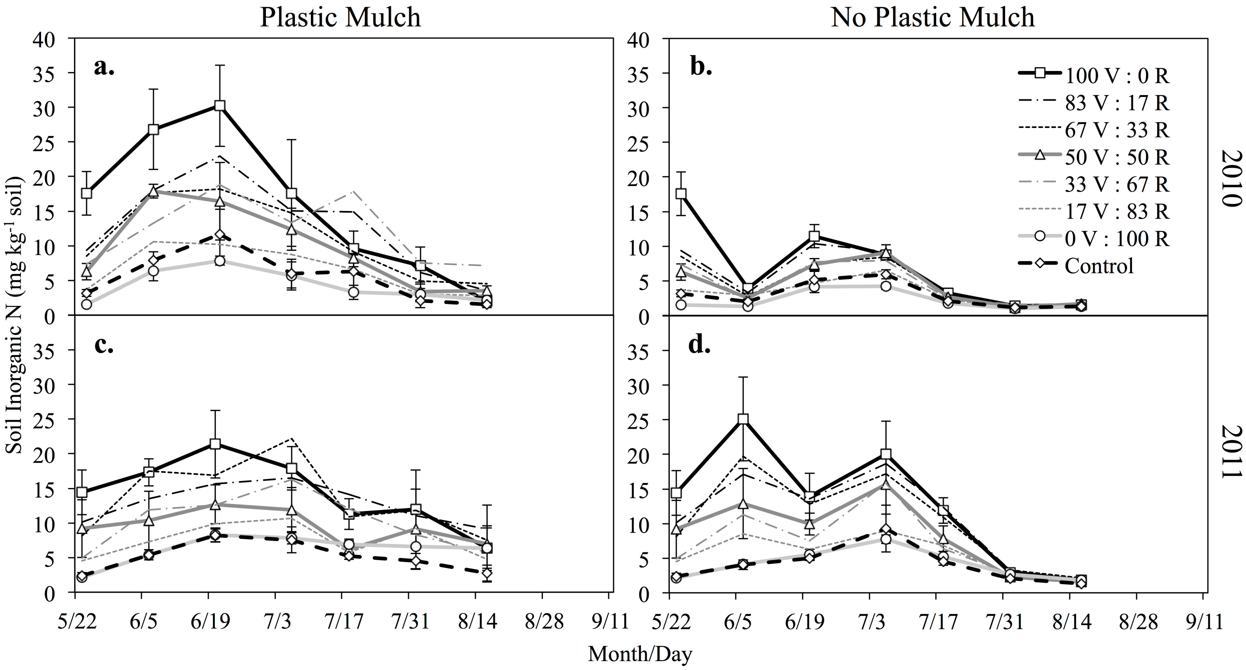

Figure 3.5. Total soil inorganic N concentrations during the summer in bell pepper rows grown either on black plastic mulch (a, c) or bare ground (b, d) in 2010 (a, b) and 2011 (c, d) following 7 rye-vetch cover crop mixture proportions or a no cover crop control. Means +/- SE. For clarity, error bars are only presented for select cover crop treatments (Control, 100 V: 0 R, 50 V: 50 R, and 0 V: 100 R).

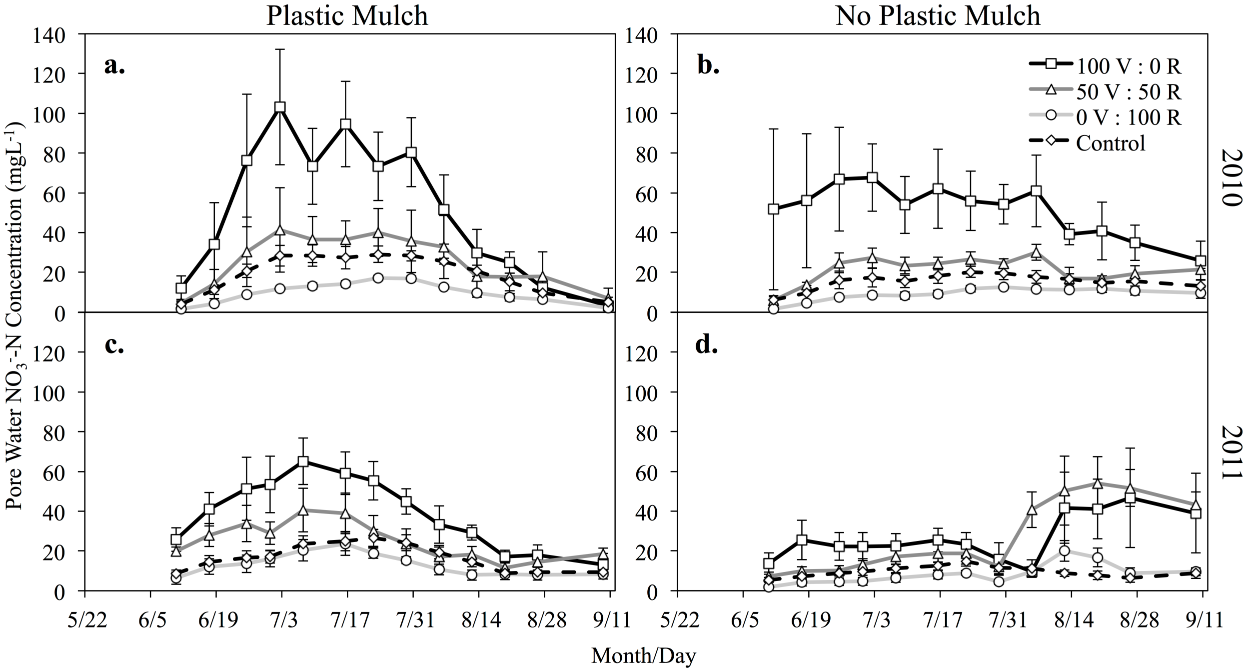

Figure 3.6. NO_3^--N concentrations of pore water extracted during the summer from suction lysimeters installed to a depth of 61 cm below bell pepper rows grown either on black plastic mulch (a, c) or bare ground (b, d) in 2010 (a, b) and 2011 (c, d) following select rye-vetch cover crop mixture proportions or a no-cover-crop control. Means +/- SE.

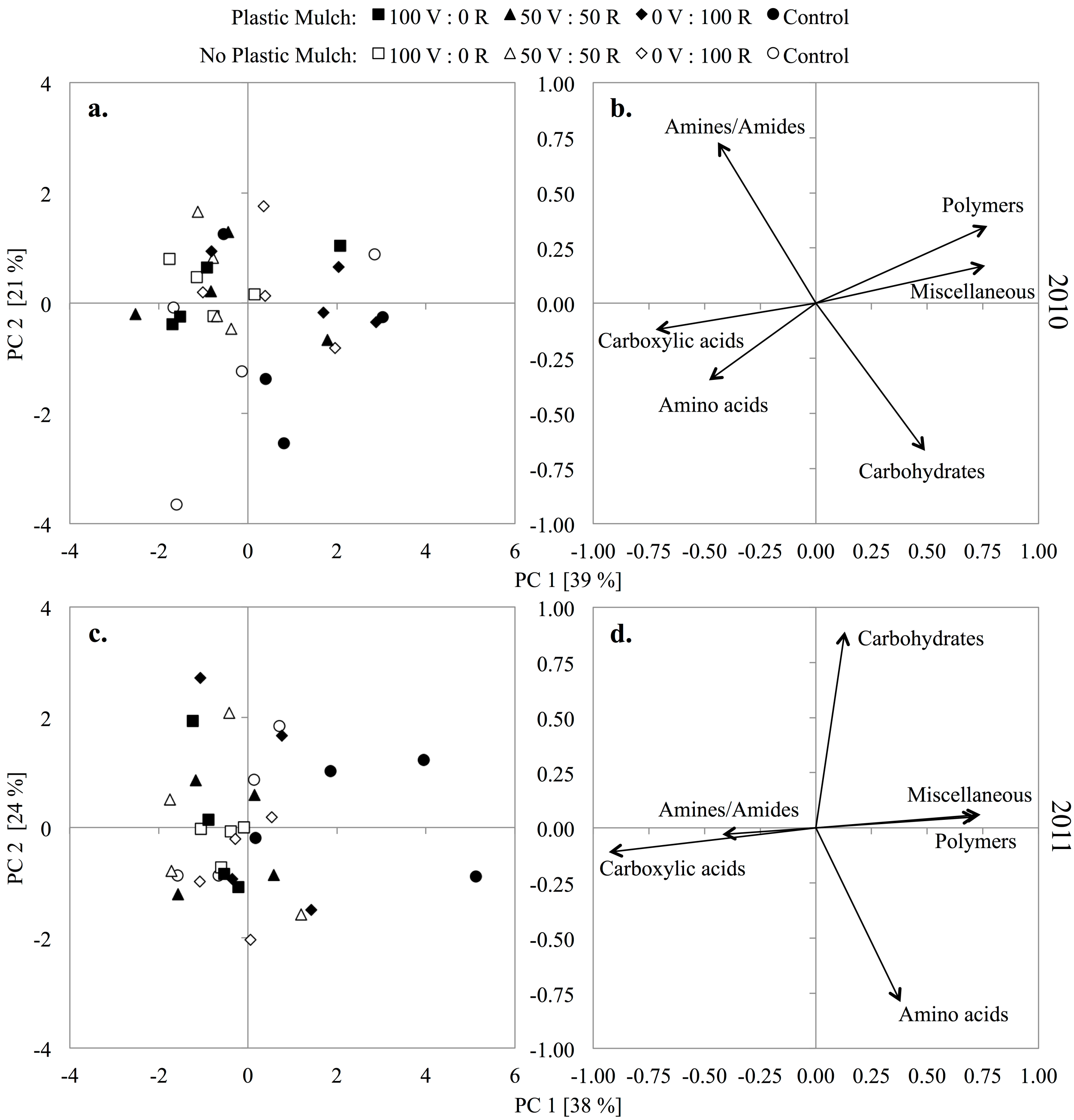

Figure 3.7. Principal component analysis of soil microbial substrate utilization data from Biolog-EcoPlates^{TM} in 2010 (a, b) and 2011 (c, d). Component scores for black plastic mulch (black) and bare ground (white) management following select rye-vetch cover crop mixture treatments (shapes) are plotted for the first 2 principal components (a, c), as well as the correlations (loadings) between the 2 principal components and the original 6 substrate class variables (b, d). The percentages of the total variance accounted for by each principal component in a given year are indicated in parentheses in axis titles.

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CHAPTER IV

PLANTING DATE AND STAGGERED SEEDING OF RYE-VETCH MIXTURES: BIOMASS, NITROGEN, AND LEGUME WINTER SURVIVAL

Planting date and staggered seeding of rye-vetch mixtures: Biomass, nitrogen, and legume winter survival

Zachary D. Hayden*, Mathieu Ngouajio, and Daniel C. Brainard

Department of Horticulture, Michigan State University, Plant and Soil Sciences Building, 1066 Bogue St., East Lansing, MI 48824.
*Corresponding author (haydenza@msu.edu).

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ABSTRACT

Mixtures of cereal rye (*Secale cereale* L.) and the legume hairy vetch (*Vicia villosa* Roth) are used to provide fixed N in balance with other important cover crop services, but late planting and interference from rye can limit vetch productivity. This two-year study in Michigan investigated how fall planting dates influence rye-vetch cover crop biomass quantity and quality in the spring, and evaluated whether staggering (delaying) rye seeding could improve vetch performance (biomass production, N_2 fixation, and winter survival) in mixtures. Treatments consisted of a two-way factorial of three vetch planting dates (late August, mid September, and late September) and three lengths of rye seeding stagger (co-seeded, short stagger, and long stagger). Later planting of co-seeded mixtures generally led to reduced total shoot biomass and lower proportions of vetch, resulting in cover crop residues with less fixed N and a higher total C:N. For earlier planting dates, delaying rye seeding until vetch emergence (short stagger) increased vetch shoot biomass by 760 - 1,060 kg ha^{-1} (30-36 kg vetch N ha^{-1}) relative to co-seeding. Staggered seeding provided no benefit to vetch biomass at later planting dates, and delaying rye seeding until the vetch 3-4 leaf stage (long stagger) resulted in significant reductions in vetch winter survival compared with co-seeding. Additional research would be needed to determine whether potential on-farm benefits of rye-vetch staggered seeding justify the additional management complexity involved and possible tradeoffs with other cover crop services.

INTRODUCTION

Winter annual cover crop mixtures composed of cereal rye and the legume hairy vetch have been well-studied for their potential to accumulate significant fixed N (Ranells and Wagger,

1996) while providing more effective weed suppression (Teasdale and Abdul-Baki, 1998; Hayden et al., 2012), erosion control (Clark, 2007), N scavenging (Shipley et al., 1992), and greater biomass production (Clark et al., 1994; Sainju et al., 2005) at lower overall seed costs (Hayden et al., 2014) than a vetch monoculture. Cereal-legume functional complementarity may contribute to mixture advantages, including greater resource-use efficiency (Fukai and Trenbath, 1993), improved N availability for the cereal (Ledgard and Steele, 1992), and enhanced N fixation (Streeter, 1988; Izaurralde et al., 1992), light interception (Keating and Carberry, 1993), and winter survival (Jannink et al., 1997) for the legume. The moderation of total residue quality in mixtures may also alter patterns of net N mineralization following cover crop termination, alleviating problems with N tie-up associated with cereal cover crops while potentially improving the synchrony of N release with crop demand compared to a monoculture legume (Kuo and Sainju, 1998; Clark et al., 2007). However, interference between the components can limit mixture benefits—most often resulting in reductions in legume performance (Ofori and Stern, 1987).

Both management decisions and environmental conditions can have considerable influence on species interactions and outcomes in cereal-legume mixtures. As winter annuals, rye and vetch are sown in the late summer or early fall, and after overwintering, put on the majority of their growth the following spring. The popularity of rye and vetch as cover crops can be attributed in part to their cold hardiness, which makes production possible across a broad geographic range, including the Northeast United States and the Great Lakes region (Clark, 2007). Fall planting dates influence the developmental stage reached before the onset of winter conditions, with later planting reducing spring biomass production (Teasdale et al., 2004; Farsad et al., 2011) and increasing the potential for overwinter mortality, particularly for vetch

(Brandsaeter et al., 2008). Rye can germinate, grow, and survive at lower temperatures than vetch (Nuttonson, 1957; Teasdale et al., 2004), contributing to its ability to establish later in the fall and accumulate biomass faster in the spring (Shipley et al., 1992). Later planting dates and colder winter conditions are therefore likely to favor rye growth over that of vetch in mixtures. However, the presence of rye may also improve vetch winter survival by reducing frost heaving or buffering low temperature extremes through greater snow cover retention or reduced air movement at the soil surface (Smith, 1975; Jannink et al., 1997). Despite a body of research on these two species individually, little information exists on how planting dates influence rye and vetch in mixture. Since inclement weather and the harvest of previous cash crops often delay planting past recommended dates, such information is important for helping farmers understand what they can expect from rye-vetch mixtures, particularly in terms of biomass production and N fixation.

Owing in part to extensive root systems and high relative growth rates, cereals tend to be stronger competitors than legumes (Mariotti et al., 2009). Suppression of the legume component is a common challenge in cereal-legume mixtures, particularly under conditions of excess soil N, moisture stress, or non-N nutrient limitation (Ofori and Stern, 1987; Jensen, 1996). Previous studies have investigated seeding rates (e.g. Clark et al., 1994), termination dates (e.g. Clark et al., 1997), and planting arrangements (e.g. Brennan et al., 2009) in rye-vetch and other cereal-legume mixtures as means to manage species interactions for improved cover crop performance.

Relay intercropping, in which the seeding of mixture components is staggered temporally, has also been used in cereal-legume cash crop and cash crop-cover crop combinations as a strategy for moderating competition to improve yields of one or both components or to achieve other system benefits (Francis et al., 1982; Ofori and Stern, 1987; Amosse et al., 2013). While the concept is rarely considered for cover crop mixtures, staggered seeding of rye and vetch could foster advantages of agronomic significance over the standard co-seeded practice. In particular, delayed seeding of rye after vetch could reduce interference from rye during both fall establishment and spring growth periods, with the potential to improve vetch establishment, productivity, and N fixation in mixtures. Alternatively, less rye growth in the fall could negatively impact vetch winter survival (Smith, 1975), and reductions in the uptake of rhizosphere N by stagger-sown rye could limit possible improvements to vetch N fixation efficiency compared with a co-seeded mixture (Streeter, 1988). Staggered seeding also entails additional management complexity, including the labor, fuel, and soil compaction costs associated with an added tractor pass. However, if the practice resulted in significant improvements in the total amount of N fixed per vetch seed sown in mixtures while also maintaining services more characteristic of rye (such as weed suppression, N scavenging, and erosion control), staggered seeding of rye-vetch mixtures may provide a net benefit.

The objectives of this study were 1) to investigate the influence of fall planting date on spring biomass quantity and quality in a co-seeded rye-vetch cover crop mixture, and 2) to evaluate the interactive effects of staggered (delayed) rye seeding and planting date on cover crop mixture outcomes, with particular emphasis on possible improvements in vetch biomass production, N fixation, and winter survival.

MATERIALS AND METHODS

The study site was located at the Michigan State University (MSU) Horticulture Teaching and Research Center in Holt, MI (42°40” N, 84°28” W) on a level Spinks loamy sand soil (sandy, mixed, mesic Lamellic Hapludalf). Prior to the start of the experiment, the field had been fallow for two summers with fall and spring rototilling to control weeds. Initial soil chemical characteristics in fall 2009 included pH 6.4; CEC 5.3 cmol kg^{-1}; 602 ppm Ca; and P (Bray-P1), K, and Mg levels of 131, 85, and 101 mg kg^{-1}, respectively. No fertilizers or soil amendments were applied during the study.

The treatment design was a two-way factorial consisting of three vetch planting dates (late August, mid September, and late September) and three lengths of rye seeding stagger (co-seeded, short stagger, and long stagger), resulting in nine rye-vetch cover crop mixture treatments. Rye and vetch were sown at the same time in co-seeded mixtures, whereas rye seeding was delayed in staggered mixtures. Table 1 presents the dates of rye and vetch seeding for each mixture in 2009 and 2010. To ensure that rye was sown at a consistent vetch growth stage in each stagger treatment, the delay in rye seeding was based on growing degree day (GDD) base 4°C accumulation following vetch seeding. In short and long stagger treatments, rye was sown approximately 120 and 230 GDD after vetch, respectively, corresponding to the timing of vetch emergence and the appearance of the first runner at the base of vetch seedlings (3-4 leaf stage).

Rye-vetch mixture treatments were sown into permanent 1.5 x 1.5m experimental plots arranged in a randomized complete block design with four replications. Adjacent plots were separated by 0.6m wide paths. Rye and vetch seeds were planted by hand using custom-made grids for each species to ensure uniform seed densities and spacings. Rye was sown at 266 seeds m^{-2} and vetch at 82 seeds m^{-2} in all mixtures, approximating field seeding rates of 47 and 21 kg ha^{-1} for rye and vetch, respectively. All seeds were planted to a depth of about 3 cm, and care was taken not to disturb vetch seedlings during staggered rye seedings. In both years of the study, variety not stated (VNS) vetch seed grown in Oregon and VNS rye seed grown in Minnesota were used (Albert Lea Seed House, Albert Lea, MN). Vetch seed was inoculated with N-DURE *Rhizobium leguminosarum* inoculant (INTX Microbials LLC, Kentland, IN) prior to each seeding event at a rate of approximately 10 g inoculant kg^{-1} seed.

Rye and vetch densities and shoot biomass were sampled from two 25 x 50 cm quadrats established in each plot, oriented such that they encompassed a consistent number of sown rye and vetch seeds. Fall counts of rye and vetch seedlings within the quadrats were taken following emergence of each species in a given treatment, and plant counts were taken again in the spring from the same quadrats immediately prior to biomass harvest, on 14 and 17 May in 2010 and 2011, respectively. Potential differences in vetch winter survival were evaluated by calculating the percentage change in vetch plant counts between fall and spring samplings. Rye and vetch shoot biomass was harvested from quadrats by clipping at the soil surface, and dry weights were obtained after drying to a consistent weight at 38 ºC. Cover crop biomass remaining in the plots after sampling in 2010 was mowed with a line trimmer and incorporated into the soil using a walk-behind rototiller. Weeds were managed during the summer with a line trimmer, and plots were rototilled twice more at 2 wk and 1 d prior to sowing rye and vetch for the second year of the study. Treatment locations were re-randomized.

Rye and vetch tissue samples were ground to pass through a 1mm screen, and submitted to the University of California-Davis Stable Isotope Facility (SIF) for analysis of percentage total C and N and $\delta^{15}N$ using a PDZ Europa ANCA-GSL elemental analyzer interfaced to a PDZ Europa 20-20 continuous flow isotope ratio mass spectrometer (Sercon Ltd., Cheshire, UK). Cover crop shoot C and N content was calculated as the product of shoot dry weight and percentage shoot C or N. Vetch N fixation was estimated using the ^{15}N natural abundance method, which relies on the slight enrichment of ^{15}N generally present in soil N relative to atmospheric N_2 (Shearer and Kohl, 1986; Hogberg, 1997). Isotopic composition is expressed relative to atmospheric N_2 using $\delta^{15}N$ values in parts per thousand (‰), with higher $\delta^{15}N$ indicating greater ^{15}N enrichment. The percentage of vetch N derived from the atmosphere (%Ndfa) was calculated from Eq. 1 (Rochester and Peoples, 2005),

$$[1] \quad \%Ndfa = (\delta^{15}N_{ref} - \delta^{15}N_{leg})/(\delta^{15}N_{ref} - B) \times 100$$

where $\delta^{15}N_{ref}$ is the shoot $\delta^{15}N$ value for rye in monoculture (used as the non-N-fixing reference plant), $\delta^{15}N_{leg}$ is the shoot $\delta^{15}N$ value for vetch, and B is the estimated $\delta^{15}N$ of vetch when grown entirely dependent on atmospheric sources of N. Following Hansen et al. (2002) and Hansen and Vinther (2001), the lowest $\delta^{15}N_{leg}$ value in each study year was used to approximate B, resulting in conservative estimates of vetch %Ndfa. Rye monoculture $\delta^{15}N_{ref}$ values were 2.08 and 1.44 ‰ in 2010 and 2011, respectively.

Statistical Analysis and Data Presentation

In keeping with the factorial design of the experiment, the fixed effects of year, planting date, stagger, and their interactions were analyzed through mixed model ANOVA using the MIXED procedure of SAS, where block (replication) was included as a random factor. Unequal variance models and/or square root transformations were used when necessary to satisfy ANOVA assumptions of normality and equality of variances. Where slicing indicated significant differences ($P < 0.05$) between levels of stagger within a given year and planting date, means were separated using Fisher's Protected LSD at the 0.05 probability level.

Differences in planting dates and weather conditions between the two years of the study resulted in variable heat accumulation during cover crop growth in each treatment (Table 1), with important implications for interpreting the results. Data for both years were therefore combined in figures on the basis of total GDD base 4 ºC accumulation during the period of vetch growth in mixtures, in order to visualize the effects of later fall planting and staggered seeding within the context of seasonal temperature variability. With years combined, the nature of the overall effect of heat accumulation (GDD, as influenced by planting date and weather) on cover crop mixture outcomes within each level of stagger was investigated through linear or polynomial regression using the REG procedure of SAS (Version 9.2, SAS Institute, Cary, NC).

RESULTS AND DISCUSSION

Weather and GDD Accumulation

Table 1 presents the influence of planting dates and lengths of rye stagger on the total GDD accumulated during vetch and rye growth in the various mixture treatments during the 2009-2010 and 2010-2011 seasons. Within years, relative differences in heat accumulation among treatments were driven by planting dates, while between years differences in heat accumulation for a given treatment were influenced by both planting date and variable weather conditions. Due to later planting dates and cooler temperatures, cover crops grown during the 2010-2011 season experienced 51 fewer GDD in the fall and 123 fewer GDD in the spring than those grown during the 2009-2010 season. Furthermore, minimum winter temperatures were lower overall in 2010-2011 than in 2009-2010. The average and absolute minimum air temperatures, respectively, during the coldest month (January) were -8.5 and -20.8 ºC in 2009-2010 and -11.2 and -22.7 ºC in 2010-2011. Final spring biomass production by rye and vetch cover crops is highly correlated with GDD base 4 ºC accumulation during both the fall and spring (Teasdale et al., 2004; Farsad et al., 2011). While colder winter conditions will tend to result in lower overall heat accumulation, the magnitude and frequency of minimum temperature extremes, which are not directly captured in the calculation of GDD, may be a more influential factor in cover crop overwinter survival.

Effect of Planting Date on Co-seeded Mixture Biomass

Later fall planting dates and lower overall GDD accumulation were generally associated with lower total shoot biomass of co-seeded rye-vetch mixtures by the time of cover crop harvest in May, but the responses differed for the component species (Figure 1A). Vetch biomass production decreased substantially in mixtures planted later in the fall, with reductions from 355.0 to 82.9 and 153.2 to 20.6 g m^{-2} between the late August and late September planting dates in 2009-2010 and 2010-2011, respectively. In contrast, rye biomass production in later planted mixtures increased or remained unchanged relative to earlier planting dates for all but the mid September planting in 2010-2011. As a consequence, spring stands of mixtures planted later in the fall contained increasingly greater proportions of rye over vetch biomass. The percentage of total mixture biomass that was vetch decreased from 58 to 21% between the late August and late September planting dates in 2009-2010, and from 42 to 12% between the same planting date treatments in 2010-2011. This compositional change was accompanied by a corresponding decrease in total N accumulated by the cover crops. While mixtures planted in late August accumulated 16.8 and 8.7 g N m^{-2} in 2010 and 2011, respectively, delaying planting until late September reduced total N by 55 and 57% in the two years (Figure 1B). Furthermore, the total residue C:N ratio of the mixture stands exhibited a general increase with later planting and lower GDD accumulation, ranging from 15.2 to 24.0 in 2010 and from 17.2 to 19.9 in 2011 (Figure 1C).

These results are consistent with physiological and growth response differences between rye and vetch. While reduced heat accumulation in the fall or spring will limit the biomass production of both species (Guldan and Martin, 2003; Teasdale et al., 2004; Farsad et al., 2011), rye productivity is generally more robust in the face of delayed planting or colder temperatures than vetch (Shipley et al., 1992). The increases in rye biomass observed in later-planted mixtures, particularly in 2010, were likely a result of reduced competition from the legume.

The reduced overall productivity and shifts in biomass composition that accompanied delayed planting of rye-vetch mixtures have important implications for the green manure benefits that can be expected from the cover crop following termination. Not only do potential fixed N additions decline in tandem with reduced vetch productivity, but the increasing biomass C:N of later planted mixtures may negatively impact the first season availability of any accumulated residue N as well (Vigil and Kissel, 1991; Kuo and Sainju, 1998). Total C:N was below 25 for all mixtures in this study, suggesting that the residues would not have caused extended net N immobilization if incorporated at the time of sampling (Allison, 1966). However, the quality of the mixed residues at the time of harvest will be influenced by the maturity of rye and vetch in addition to the relative species composition of the mixture, and may vary considerably depending on when the cover crop is terminated (Clark et al., 1997; Clark et al., 2007).

Given the relatively high cost of vetch seed, farmers should consider whether seeding a mixture is worth the investment in the legume after a certain point in the fall, when compared with sowing a less expensive cereal like rye alone (Hayden et al., 2014). Seeding a rye-vetch mixture, even later than recommended, does maintain the possibility of significant fixed N additions in the event that winter and/or spring temperatures are warmer than average. Farmers may also be able to compensate by altering mixture management to promote greater vetch biomass production— such as by increasing the proportion of vetch sown in the mixture (Hayden et al., 2014), or if possible, by delaying cover crop termination to allow more time for cover crop growth in the spring (Teasdale et al., 2004; Clark et al., 2007). The potential for delayed rye seeding to improve vetch biomass production in mixtures was also a consideration in designing this experiment.

Staggered Seeding Effects

Staggered seeding of rye resulted in greater vetch shoot biomass production than co-seeding in mixtures planted in late August and mid September in 2009 and in late August in 2010 (Figure 2A). Delaying rye seeding on those planting dates until vetch emergence (short stagger) increased vetch biomass by between 30 - 62% (76 - 106 g m^{-2}) relative to the co-seeded mixtures. The effect on vetch biomass of further delaying rye seeding until the vetch 3-4 leaf stage (long stagger) did not differ significantly from the shorter stagger.

Vetch productivity and stand establishment may have benefitted from reduced interference during seedling establishment, though biomass production more likely increased as a result of lower competitive inhibition from the delayed rye component during fall and especially spring growth periods. Total GDD accumulation during cover crop growth appeared to regulate the interactions between vetch planting date and the effects of staggered seeding over the two seasons of the study.

No significant benefits to vetch biomass production were observed with staggered seeding in mixtures where vetch experienced fewer than 850 GDD before harvest, which included the late September planting in both years and the mid September planting in 2010 (Figure 2A). Below that threshold, vetch productivity may have been more limited by heat accumulation than by interference from rye, or was perhaps tempered by greater stand loss from overwinter mortality in the staggered mixtures (Figure 3C). In most cases, rye biomass declined in mixtures with increasing length of stagger due to the shorter period available for growth (Figure 2B). The magnitude of the stagger-driven reductions in rye biomass generally increased with later planting dates and more limited GDD accumulation, as well as greater overwinter stand loss (data not shown). As a result, where vetch accumulated greater than 950 GDD, stagger-sown mixtures yielded equivalent total biomass to co-seeded mixtures, while in the case of lower GDD accumulation, staggering seeding led to significant reductions in the total biomass produced (Figure 2C).

The shoot N content of the cover crop mixtures was driven predominately by rye and vetch biomass yields. For the earlier planting date treatments where >950 GDD accumulated during vetch growth, staggered seeding resulted in the equivalent of an additional 30-36 kg N ha^{-1} in vetch shoots relative to co-seeding due to an increase in vetch biomass (Table 2). While the average effect of delayed rye seeding on vetch N did not differ between the two lengths of stagger, the change in the total N content of mixtures generally decreased from short to long staggers due to greater reductions in rye biomass (Table 2). However, by increasing vetch and/or decreasing rye biomass, the compositional shifts in mixture biomass fostered by longer delays in rye seeding typically resulted in lower total residue C:N at the time of sampling (Table 2). Predicting the dynamics of inorganic N availability from mixed organic materials can be complex, but both N content and residue quality will influence net N mineralization in the first season following cover crop incorporation (Kuo and Sainju, 1998).

Neither planting date nor staggered seeding significantly influenced vetch N fixation efficiency (%Ndfa) under the conditions of this study (Table 2). The estimated percentage of vetch N derived from fixation was 88% in 2009-10, and 77% in 2010-11 but did not vary by treatment within years. Depending on genetic and environmental contexts, the presence of a grass species in mixture may alternately enhance legume N fixation by depleting soil inorganic N (Jensen, 1996) or impair it as a result of competitive inhibition (Brainard et al., 2011). Previous studies have documented increases in vetch %Ndfa when vetch is grown in mixture with a cereal species rather than in monoculture (Kurdali et al., 1996; Brainard et al., 2012), but the impact of relatively small compositional changes in rye-vetch mixtures (such as those resulting from staggered seeding) on vetch N fixation are probably minor or difficult to detect, especially under conditions of low soil N availability. Working on land adjacent to this experiment with similar sandy and unfertilized soils, Hayden et al. (2014) observed no significant differences in vetch %Ndfa across a gradient of different rye and vetch proportions in mixture.

Vetch Winter Survival

In co-seeded mixtures, vetch winter survival was consistent across the planting dates and study years, with 90% of vetch seedlings surviving through the winter on average (Figure 3C). Staggered seeding did not influence winter survival in mixtures where >950 GDD accumulated during vetch growth; however, for later planting dates where mixtures experienced more limited GDD accumulation, delaying rye seeding resulted in significant reductions in vetch winter survival. For the late September seeding in 2010-11, delaying rye seeding until the vetch 3-4 leaf stage (long stagger) resulted in winter survival of 46% compared to 90% with co-seeding (Figure 3C).

Hairy vetch is the hardiest of winter annual legume cover crops, but its cold tolerance varies depending on cultivar and developmental stage (Wilke et al., 2008). Although freezing resistance declines after flower initiation (Brandsaeter et al., 2002), vetch stands generally experience greater winter mortality when planted later in the fall (Brandsaeter et al., 2008; Guldan and Martin, 2003), suggesting that in the vegetative phase younger plants or seedlings are more cold sensitive. For example, during a season in New York when the average and absolute minimum temperatures in January were -13 and -25 ºC, respectively, Teasdale et al. (2004) observed 85% winter survival for hairy vetch sown on 25 August versus 39% survival when sown 14 September.

The likely mechanisms by which rye growth may improve legume winter survival in mixtures include reducing frost heaving of the soil, slowing boundary layer air movement, and increasing snow cover retention—all of which lower effective minimum temperature extremes or otherwise limit legume exposure (Smith, 1975). Jannink et al. (1997) observed reduced vetch overwinter mortality in mixture with rye at two locations in Maine, and Brainard et al. (2012) found a similar response for three vetch cultivars in one out of two years in Michigan. In contrast, Hayden et al. (2014) found no evidence that the presence of rye influenced vetch winter survival in mixtures that were sown on 1 September over two years in Michigan. Our staggered seeding results demonstrate that the presence of rye can provide a clear benefit to vetch winter survival, particularly when vetch is planted late or is otherwise exposed to cold temperatures at a less freeze-resistant developmental stage. Delaying rye seeding at later planting dates reduced rye biomass production in the fall (Figure 2B), such that it was likely no longer effective at moderating soil surface conditions and/or microclimate during the winter months. However, from a practical standpoint, it is worth noting that the conditions under which rye is likely to have the greatest influence on vetch winter survival, including exceptionally cold winter temperatures or late fall planting, are also conditions that tend to greatly limit vetch biomass production at mid-May termination dates, irrespective of stand density.

CONCLUSIONS

Rye-vetch cover crop stands will vary considerably depending on when the mixture is planted. Later planting and less GDD accumulation during cover crop growth generally led to reduced total shoot biomass and a greater proportion of rye biomass in mixtures. As a consequence, later planted co-seeded mixtures accumulated less fixed and total N, and produced residues with a higher combined C:N.

A novel finding from this study was that for earlier planted rye-vetch mixtures where >950 GDD accumulated during vetch growth, staggering (delaying) rye seeding until vetch emergence increased vetch shoot biomass by 760 - 1,060 kg ha^{-1} and vetch N content by 30 - 36 kg ha^{-1} compared with co-seeding. For later plantings, staggered seeding provided no benefit to vetch biomass production, only reductions in rye and total biomass. In addition, staggering rye seeding until the vetch 3-4 leaf stage at these later planting dates resulted in lower vetch winter survival compared with co-seeded mixtures.

Although staggered seeding resulted in increased fixed N accumulation in some mixtures planted earlier in the season, this study did not evaluate possible tradeoffs in the provision of other important services by the cover crop. For example, weed suppression has been shown to decrease with reductions in rye biomass in rye-legume mixtures (Akemo et al., 2000; Hayden et al., 2014). Furthermore, any benefits of the practice must ultimately be weighed against the costs associated with additional management complexity, including the time, labor, and fuel of an added tractor pass for rye seeding. In this regard, it is important to note that the seeding methodology used in this study did not reflect conditions that would result from the field-scale application of staggered seeding, such as the effects of additional traffic on young vetch seedlings or the establishment of broadcast rye without incorporation.

Several alternative approaches to realizing the benefits of staggered seeding seen in this study may be worth further exploration. Seed coatings have been used to delay germination of cash crops to allow for simultaneous seeding in relay intercropping systems, but coating seeds is itself expensive and the resulting germination kinetics can be challenging to manage (Stendahl, 2005). Alternatively, selection for more rapid germinating vetch varieties, or use of seed-priming techniques, may be useful for increasing the rate of germination and emergence of vetch relative to rye. Seed priming is commonly used to speed germination (Parera and Cantliffe, 1994), although its efficacy for vetch has not been studied, and the costs associated with priming can be substantial. Additional research would be required to determine whether potential on-farm benefits of rye-vetch staggered seeding would justify additional management costs or possible tradeoffs in cover crop services.

APPENDIX

Table 4.1. Seeding schedule and accumulated growing degree days (GDD) during rye and vetch growth in cover crop mixture treatments.

	2009-2010				2010-2011			
	Date sown		Accumulated GDD†		Date sown		Accumulated GDD†	
Treatment	Vetch	Rye	Vetch	Rye	Vetch	Rye	Vetch	Rye
Late Aug. vetch seeding								
Co-seeded mixture	8/25/09	8/25/09	1189	1189	8/31/10	8/31/10	1020	1020
Short rye stagger‡	8/25/09	9/3/09	1189	1078	8/31/10	9/8/10	1020	898
Long rye stagger§	8/25/09	9/11/09	1189	961	8/31/10	9/17/10	1020	800
Mid Sept. vetch seeding								
Co-seeded mixture	9/9/09	9/9/09	993	993	9/14/10	9/14/10	832	832
Short rye stagger	9/9/09	9/18/09	993	864	9/14/10	9/23/10	832	720
Long rye stagger	9/9/09	9/24/09	993	780	9/14/10	10/7/10	832	599
Late Sept. vetch seeding								
Co-seeded mixture	9/22/09	9/22/09	815	815	9/28/10	9/28/10	665	665
Short rye stagger	9/22/09	10/2/09	815	702	9/28/10	10/12/10	665	540
Long rye stagger	9/22/09	10/27/09	815	571	9/28/10	10/29/10	665	417

† Total growing degree day (GDD) base 4°C accumulation between date of vetch or rye seeding and cover crop sampling on 14 or 17 May in 2010 and 2011, respectively.

‡ In short stagger mixtures, rye seeding was delayed 120 GDD following vetch seeding, approximately at vetch seedling emergence.

§ In long stagger mixtures, rye seeding was delayed 230 GDD following vetch seeding, approximately at the vetch 3-4 leaf stage.

Table 4.2. Percentage vetch shoot N derived from the atmosphere (Ndfa), vetch and rye shoot N content, and total (vetch+rye) shoot N content and C:N ratio in co-seeded, short stagger, and long stagger mixtures sown in late August, Mid September, and Late September in 2009 and 2010.

	Vetch Ndfa		Vetch shoot N content		Rye shoot N content		Total shoot N content		Total shoot C:N	
	2009-10	2010-11	2009-10	2010-11	2009-10	2010-11	2009-10	2010-11	2009-10	2010-11
	%		g m^{-2}							
Late Aug. vetch seeding										
Co-seeded mixture	92 (0.4) ns†‡	74 (6) ns	12.7 (1.3) a§	5.9 (0.6) a	4.1 (0.2) ns	2.9 (0.1) ab	16.8 (1.2) a	8.7 (0.6) a	15.2 (1.0) ns	17.2 (0.3) a
Short rye stagger	74 (12)	78 (4)	16.3 (2.0) b	9.2 (0.9) b	4.3 (0.5)	3.7 (0.5) a	20.6 (2.4) b	12.9 (1.2) b	14.2 (0.4)	15.8 (0.6) ab
Long rye stagger	67 (17)	76 (4)	13.1 (0.8) a	9.5 (1.9) b	3.2 (0.3)	2.2 (0.2) b	16.3 (1.0) a	11.7 (2.0) ab	14.9 (0.3)	14.5 (0.7) b
Mid Sept. vetch seeding										
Co-seeded mixture	94 (2) ns	86 (8) ns	6.1 (0.9) a	4.2 (0.7) ns	4.1 (0.4) a	4.9 (0.9) a	10.2 (1.2) a	9.0 (1.4) ns	20.8 (0.7) a	19.9 (2.1) a
Short rye stagger	93 (2)	79 (3)	9.1 (1.2) ab	3.0 (0.7)	3.7 (0.3) a	3.6 (0.6) b	12.8 (1.5) ab	6.6 (0.5)	17.0 (0.5) b	18.0 (0.9) a
Long rye stagger	93 (3)	72 (6)	12.2 (2.2) b	4.1 (1.6)	2.7 (0.3) b	1.6 (0.3) c	14.9 (2.1) b	5.7 (1.6)	14.3 (0.6) c	13.2 (0.6) b
Late Sept. vetch seeding										
Co-seeded mixture	95 (2) a	74 (5) ns	3.0 (0.4) ns	0.8 (0.3) ns	4.6 (1.2) a	2.8 (0.1) a	7.6 (0.9) a	3.7 (0.3) ns	24.0 (1.0) a	18.4 (2.2) a
Short rye stagger	97 (1) a	77 (5)	3.5 (0.6)	1.1 (0.3)	3.1 (0.2) b	1.7 (0.2) b	6.6 (0.4) a	2.8 (0.4)	20.6 (1.2) a	16.4 (1.3) a
Long rye stagger	91 (3) b	79 (2)	1.8 (0.5)	1.1 (0.8)	1.3 (0.2) c	0.6 (0.04) c	3.1 (0.6) b	1.7 (0.8)	16.5 (0.9) b	10.1 (0.5) b

† Mean (SE), *n* = 4.

‡ ns, not significant at the 0.05 probability level.

§ Within a given year and planting date, means followed by the same letter indicate levels of stagger are not significantly different at the 0.05 probability level.

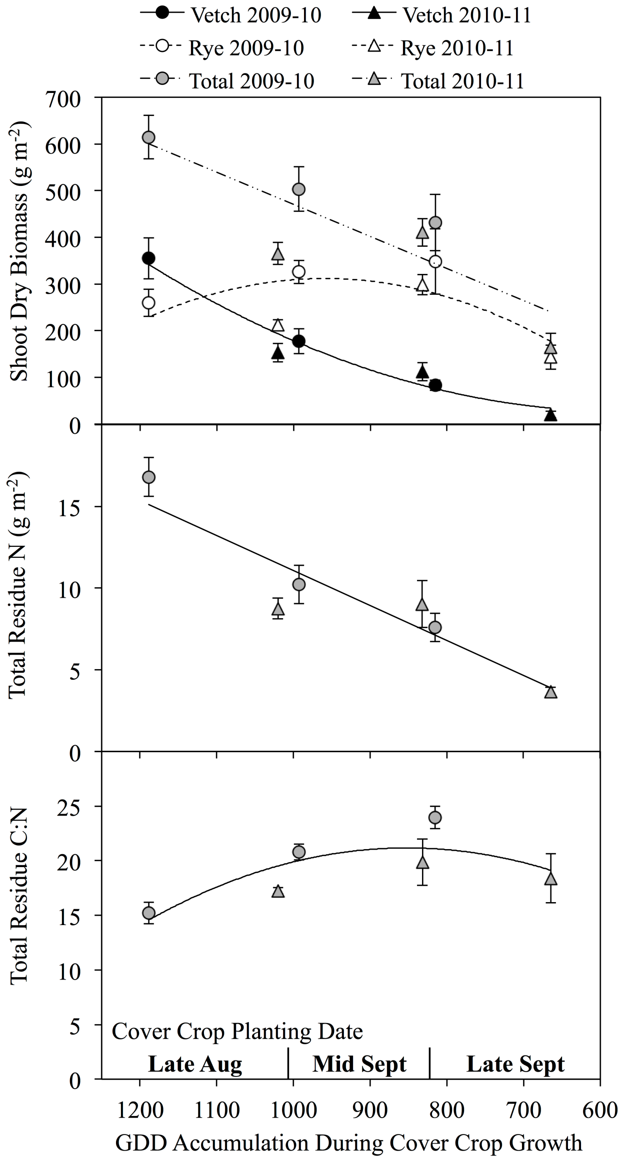

Figure 4.1. Vetch (black), rye (white), and total (vetch+rye, gray) shoot dry biomass (A), total residue N content (B), and total residue carbon:nitrogen ratio (C) in co-seeded mixtures sown in late August, Mid September, and Late September in 2009 (circles) and 2010 (triangles). Data from the two years are plotted based on total growing degree days (GDD) base 4 ºC accumulated during vetch growth on a reversed scale, with planting date treatments indicated above the x-axis. Means +/- SE. Responses to GDD accumulation (years combined) of vetch shoot biomass: $y=0.0008x^2-0.95x+298.53$, $r^2= 0.82$***; rye shoot biomass: $y=-0.0016x^2+2.99x-1122.92$, $r^2= 0.26$*; total shoot biomass: $y=0.69x-216.93$, $r^2= 0.56$***; total N content: $y=0.02x-10.32$, $r^2= 0.71$***; and total C:N: $y=-0.0001x^2+0.10x-21.08$, $r^2= 0.36$**. Single, double, and triple asterisks following r^2 values denote regression model significance at the 0.05, 0.01, and 0.001 levels, respectively.

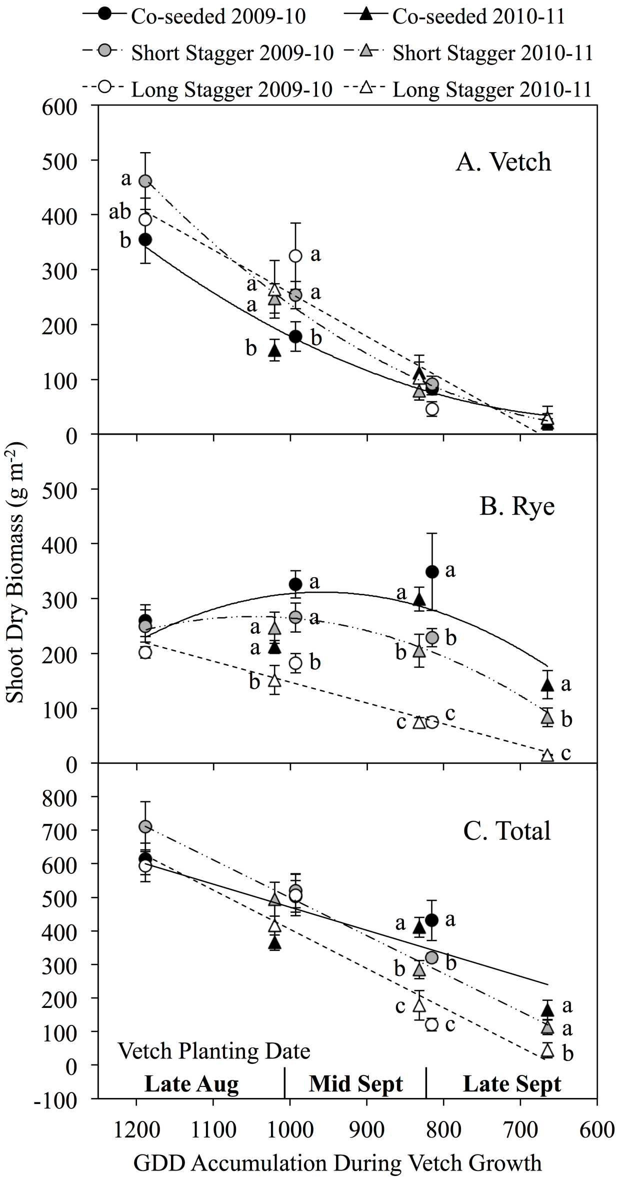

Figure 4.2. Vetch (A), rye (B), and total (vetch+rye, C) shoot dry biomass in short rye stagger (gray) and long rye stagger (white) mixtures relative to co-seeded mixtures (black) sown in late August, Mid September, and Late September in 2009 (circles) and 2010 (triangles). Data from the two years are plotted based on total growing degree days (GDD) base 4 ºC accumulated during vetch growth on a reversed scale, with planting date treatments indicated above the x-axis. Actual GDD accumulation for rye is less than that for vetch in staggered mixtures (Table 1). Within a given year and planting date treatment, levels of stagger are not significantly different when means (+/- SE) are followed by the same letter (α = 0.05). Responses to GDD accumulation (years combined) of vetch co-seeded: $y=0.0008x^2-0.95x+298.53$, $r^2= 0.82$***; vetch short stagger: $y=0.0011x^2-1.22x+342.41$, $r^2= 0.89$***; vetch long stagger: $y=0.79x-531.85$, $r^2= 0.71$***; rye co-seeded: $y=-0.0016x^2+2.99x-1122.92$, $r^2= 0.26$*; rye short stagger: $y=-0.0012x^2+2.51x-1048.44$, $r^2= 0.62$***; rye long stagger: $y=0.38x-232.79$, $r^2= 0.81$***; total co-seeded: $y=0.69x-216.93$, $r^2= 0.56$***; total short stagger: $y=1.13x-630.00$, $r^2= 0.85$***; total

Figure 4.2 (cont'd)

long stagger: y=1.17x-764.63, r^2= 0.80***. Single, double, and triple asterisks following r^2 values denote regression model significance at the 0.05, 0.01, and 0.001 levels, respectively.

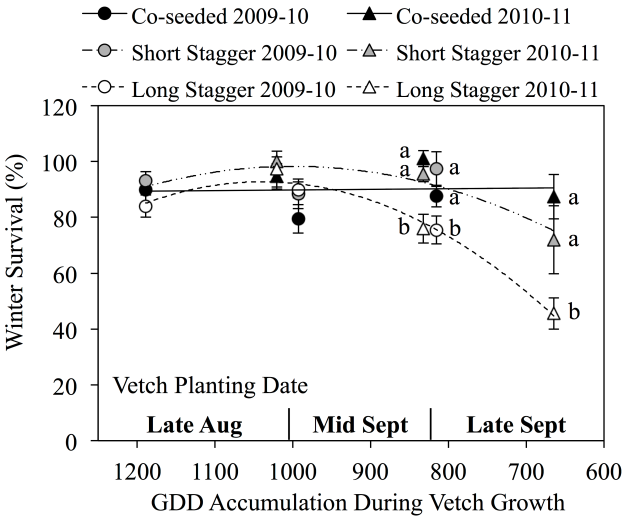

Figure 4.3. Percentage vetch winter survival in co-seeded (black), short rye stagger (gray), and long rye stagger (white) mixtures sown in late August, Mid September, and Late September in 2009 (circles) and 2010 (triangles). Data from the two years are plotted based on total growing degree days (GDD) base 4 ºC accumulated during vetch growth on a reversed scale, with planting date treatments indicated above the x-axis. Within a given year and planting date treatment, levels of stagger are not significantly different when means (+/- SE) are followed by the same letter (α=0.05). Responses to GDD accumulation (years combined) of co-seeded: y=-0.0023x+92.10, r^2=0.0011; short stagger: y=-0.00021x^2+0.41x-107.20, r^2=0.29*; and long stagger: y=-0.00034x^2+0.71x-277.40 , r^2=0.77***. Single, double, and triple asterisks following r^2 values denote regression model significance at the 0.05, 0.01, and 0.001 levels, respectively.

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CHAPTER V

CONCLUSIONS

CONCLUSIONS

Cover crop mixtures composed of cereal rye and the legume hairy vetch have the potential to combine the unique strengths of the component species while taking advantage of interspecific synergies. However, outcomes depend on both environmental and management factors that influence species interactions during cover crop growth, the final composition of mixture stands, and the decomposition of residues following cover crop termination. The unifying objective of this work was to document the performance of rye-vetch mixtures relative to rye and vetch monocultures while leveraging our understanding of ecological principles to develop and evaluate strategies for improving mixture management within vegetable cropping systems.

In evaluating how the proportion of rye and vetch sown in a mixture influenced species interactions and cover crop performance, seeding rates were found to be good predictors of rye and vetch stand density and biomass composition in the spring. While outcomes may vary depending on environment and management, we observed little evidence of substantial interference between the two species in mixture, but also saw no significant evidence of facilitative benefits, such as improved vetch winter survival, increased N fixation efficiency, or reduced costs per unit of N fixed. However, changing the proportions of rye and vetch sown resulted in important tradeoffs among some of the agroecosystem services provided by the living cover crops—for example, greater fixed N accumulation, but higher seed costs and reduced winter annual weed suppression, with increasing vetch. While greater rye content was associated with greater suppression of winter annual weed density and biomass, differences in the relative effects of rye and vetch on the density of specific components of the weed community were documented.

Following cover crop termination, N content was a dominant factor in the effects of incorporated rye-vetch residues and the use of black plastic mulch on subsequent vegetable production. Cover crops containing greater proportions of vetch generally resulted in higher soil N availability following incorporation, and under conditions where N was limiting, led to higher yields of bell pepper and slicing cucumber. However, greater N availability in the plough layer also translated into higher pore water NO_3^- concentrations below the crop root zone.

Along the gradient of rye-vetch proportions evaluated in this experiment, changes in total cover crop N content appeared to have a larger influence on vegetable yields than did any potential changes in temporal patterns of N mineralization. Regardless of rye-vetch proportion, net N mineralized from mixtures accumulated in the soil prior to peak vegetable demand, leaving it vulnerable to leaching from early-season precipitation, particularly on the sandy soils present at the experimental site. Although the benefits of plastic mulch must be weighed against the costs associated with its use and disposal, our results demonstrate that plastic mulch can be an important tool for preserving N fertility benefits from high N cover crop residues, contributing substantially to the yield increases observed with plastic mulch relative to bare ground management. Although lower microbial biomass was also observed under plastic mulch relative to bare ground 3 wk after plastic application, changes to the microbial communities resulting from cover crop and plastic mulch treatments would likely have limited practical significance for agroecosystem functioning in the short term.

Results from a separate experiment demonstrated that fall planting dates and staggered (delayed) seeding of rye after vetch can have important impacts on biomass composition of rye-vetch mixture stands in the spring. Later planting and less GDD accumulation during cover crop growth generally led to reduced total shoot biomass and a greater proportion of rye biomass in mixtures. As a consequence, delayed planting of rye-vetch mixtures resulted in spring stands containing less fixed and total N, and residues having a higher combined C:N. Staggering rye seeding just until vetch emergence in the fall was successful in increasing vetch shoot biomass production by 760 - 1,060 kg ha^{-1} and vetch N content by 30-36 kg ha^{-1} compared with co-seeded mixtures for earlier planting dates, but benefits were not observed when mixtures were planted later in the fall. Staggering rye seeding until the vetch 3-4 leaf stage at these later planting dates resulted in higher vetch winter morality compared with co-seeded mixtures, demonstrating a clear benefit to vetch winter survival due to the presence of rye, particularly when vetch is planted late or is otherwise exposed to cold temperatures at a less freeze-resistant developmental stage. Additional research would be needed to determine whether potential on-farm benefits of rye-vetch staggered seeding justify the additional management complexity involved and possible tradeoffs with other cover crop services.

Future improvements in the economic and environmental sustainability of agriculture will depend heavily on our ability to translate understanding of complex ecological interactions into applied management solutions. Further optimizing the performance of cover crops within agricultural production systems will rest broadly on our understanding and application of the biotic and abiotic controls on plant competition and decomposition of organic materials. The results of this research have the potential to support more-informed decision making regarding rye and vetch cover crop selection, mixture seeding rates, and management practices both before and after cover crop termination, which can take into account a farmer's principal objectives for the cover crop, as well as the contexts of the cropping system and environment.