

The Pennsylvania State University

The Graduate School

Department of Entomology

**FLOWERING COVER CROPS AS AN EARLY-SEASON FLORAL  
RESOURCE FOR NATIVE BEES IN PENNSYLVANIA AGROECOSYSTEMS**

A Thesis in

Entomology

by

Katherine Elizabeth Ellis

© 2014 Katherine Elizabeth Ellis

Submitted in Partial Fulfillment  
of the Requirements  
for the Degree of

Master of Science

May 2014

The thesis of Katherine Elizabeth Ellis was reviewed and approved\* by the following:

Mary E. Barbercheck  
Professor of Entomology  
Thesis Advisor

David A. Mortensen  
Professor of Weed Ecology/Biology

Edwin G. Rajotte  
Professor of Entomology

David J. Biddinger  
Associate Professor of Entomology

Gary W. Felton  
Professor of Entomology  
Head of the Department of Entomology

\*Signatures are on file in the Graduate School

## ABSTRACT

Conservation of natural processes such as pollination, pest control, and nutrient cycling are essential to maintaining a healthy agroecosystem. The incorporation of cover crops into annual crop rotations is one practice that is used in the Northeast U.S. to manage soil fertility, weed suppression, and erosion control. Additionally, cover crops that have a flowering stage have the potential to support beneficial insect communities, such as native bees. Because of the current decline facing managed honey bee colonies, the conservation of native bee communities is critical to maintaining 'free' pollination services. However, native bees are negatively affected by agricultural intensification and are in decline across North America. This project assesses the potential of flowering cover crop species to act as a conservation resource for native bee communities, in addition to providing benefits to soil fertility and agricultural production. Three flowering cover crop species are evaluated here across various crop rotation schedules and at differing levels of cover crop diversity. Flowering resources were monitored for each of these studies along with native bee and Syrphidae fly visitation.

In conclusion, cover crop species selection, cash crop rotation schedule and plant diversity level all had significant influence on the floral resources available to the native pollinator community. Different cover crop species not only had different blooming schedules and winter survival responses to planting date, but attracted unique native bee communities to their available floral resources. Additionally, flower density was shown to be the driving factor influencing differences in bee visitation frequency across treatments, but within cover crop species, for both the plant diversity and fall planting date experiments. The results from these experiments should be influential in informing future conservation and grower extension recommendations on the applied use of flowering cover crops for pollinator conservation purposes in Pennsylvania annual cropping systems.

## TABLE OF CONTENTS

List of Figures .....	vi
List of Tables .....	xii
Acknowledgements.....	xiii
<b>Chapter 1 INTRODUCTION.....</b>	<b>1</b>
Ecosystem Services and Pollination.....	1
Meet the Pollinators: Bees .....	2
Global Pollinator Decline and Conservation.....	5
Agricultural Conservation Strategies .....	6
Flowering Cover Crops for Pollinator Conservation .....	10
Literature Cited .....	13
<b>Chapter 2 THE EFFECT OF COVER CROP SPECIES DIVERSITY ON NATIVE BEE VISITATION AND DIVERSITY .....</b>	<b>19</b>
<b>INTRODUCTION .....</b>	<b>19</b>
<b>MATERIALS AND METHODS .....</b>	<b>22</b>
Site Description .....	22
Planting and plot establishment.....	25
Blooming phenology .....	26
Observations and netting .....	26
Landscape area traps (passive collection) .....	28
Statistical Data Analysis.....	29
<b>RESULTS .....</b>	<b>30</b>
<b>DISCUSSION .....</b>	<b>42</b>
<b>LITERATURE CITED .....</b>	<b>47</b>
<b>Chapter 3 FALL PLANTING DATE AS IT AFFECTS COVER CROP WINTER SURVIVAL, BLOOM PERIOD, AND NATIVE BEE VISITATION .....</b>	<b>52</b>
<b>INTRODUCTION .....</b>	<b>52</b>
<b>MATERIALS AND METHODS .....</b>	<b>56</b>
Site Description .....	56
Planting and plot establishment.....	58
Fall Biomass .....	59
Observations and Netting .....	59
Landscape area traps (passive collection) .....	60
Statistical Analysis .....	62
<b>RESULTS .....</b>	<b>63</b>
Canola: Planting Date Comparisons .....	77
Austrian Winter Pea: Planting Date Comparisons .....	81
Red Clover: Planting Date Comparisons.....	84

DISCUSSION .....	87
LITERATURE CITED .....	90
Chapter 4 CONCLUSION AND FUTURE DIRECTIONS .....	95
Literature Cited .....	100
Appendix A Experimental Plot Map.....	101
Appendix B Landscape-level Trap Collection Summary Data.....	103

## LIST OF FIGURES

Figure 2-1. Land cover classification map showing the allocation of various cover classes of the landscape in a 250 m buffer zone surrounding the research site (outlined in red). Also shown is the placement of the eight landscape-level trap locations (purple +). .....	24
Figure 2-2. Aerial photograph showing the full project site (outlined in red) and surrounding landscape.....	24
Figure 2-3. Landscape-level passive trapping example setup for one of the project boundary edges. ....	29
Figure 2-4. Species rarefaction curves for the three cover crop mixture treatments. ....	31
Figure 2-5. Average total bee visitation abundance per two minute observation period. Error bars are standard error of the mean; n=16. Bars that do not share the same letter are significantly different at p<0.05 using Tukey’s multiple comparison test.....	33
Figure 2-6. Average total bee visitation abundance by plot location transect per two minute observation period. Error bars are standard error of the mean; n=24. Bars that do not share the same letter are significantly different at p<0.05 using Tukey’s multiple comparison test. ....	33
Figure 2-7. Average bee species richness per one minute post-observation netting period by mixture treatment. Error bars are standard error of the mean; n=16. Bars that do not share the same letter are significantly different at p<0.05 using Tukey’s multiple comparison test. ....	34
Figure 2-8. Average bee species richness per one minute post-observation netting period by plot edge and plot center transects. Error bars are standard error of the mean; n=24. Bars that do not share the same letter are significantly different at p<0.05 using Tukey’s multiple comparison test. ....	34
Figure 2-9. Average number of Syrphidae visits per two minute observation period. Error bars are standard error of the mean; n=16. Bars that do not share the same letter are significantly different at p<0.05 using Tukey’s multiple comparison test.....	35
Figure 2-10. Average number of Syrphidae visits per two minute observation period across plot edges and plot center transects. Error bars are standard error of the mean; n=24. Bars that do not share the same letter are significantly different at p<0.05 using Tukey’s multiple comparison test. ....	35
Figure 2-11. Average canola plant density per 0.25m <sup>2</sup> by mixture treatment. Error bars are standard error of the mean; n= 24. Bars that do not share the same letter are significantly different at p<0.05 using Tukey’s multiple comparison test.....	36

Figure 2-12. Average canola plant density per 0.25m <sup>2</sup> by plot transect location. Error bars are standard error of the mean; n= 36. Bars that do not share the same letter are significantly different at p<0.05 using Tukey’s multiple comparison test.....	36
Figure 2-13. Box and whisker plot of the number of open blooms per 0.25m <sup>2</sup> by cover crop mixture treatment and across the total time span of the experiment (1=Monoculture, 4=4 Species Mix, 6=6 Species Mix). Boxes that do not share the same letter are significantly different at p<0.05 using Tukey’s multiple comparison test. ....	37
Figure 2-14. Box and whisker plot of the number of open blooms per 0.25m <sup>2</sup> by plot location transect across the total time span of the experiment (C=center of plot and E=edge of plot). Boxes that do not share the same letter are significantly different at p<0.05 using Tukey’s multiple comparison test. ....	37
Figure 2-15. Number of open blooms present per 0.25m <sup>2</sup> illustrated over time and as a function of increasing flower density.....	38
Figure 2-16. Box and whisker plot of the average number of open blooms per blooming plant by cover crop mixture treatment (1=Monoculture, 4=4 Species Mix, 6=6 Species Mix). Boxes that do not share the same letter are significantly different at p<0.05 using Tukey’s multiple comparison test. ....	38
Figure 2-17. Box and whisker plot of the average number of open blooms per blooming plant by plot transect locations (C=center of plot and E=edge of plot). Boxes that do not share the same letter are significantly different at p<0.05 using Tukey’s multiple comparison test. ....	39
Figure 2-18. Simple linear regression correlation of the number of bee visits to cover crop mixtures as a function of the average flower density of each treatment plot and transect. $F_{1,46}=11.52$ , $p=0.001$ .....	40
Figure 2-19. Simple linear regression correlation of bee species richness to cover crop mixtures as a function of the average flower density of each treatment plot and transect. $F_{1,46}=15.2$ , $p<0.001$ .....	41
Figure 2-20. Simple linear regression correlation of the number of Syrphidae visits to cover crop mixtures as a function of the average flower density of each treatment plot and transect. $F_{1,46}=0.6994$ , $p=0.41$ .....	41
Figure 3-1. Land cover classification map showing the allocation of various cover classes of the landscape in a 250 m buffer zone surrounding the research site (outlined in red). Also shown is the placement of the eight landscape-level trap locations (purple +). ....	57
Figure 3-2. Aerial photograph showing the project site (outlined in red) and surrounding landscape.....	57

Figure 3-3. Landscape-level passive trapping example setup for one of the project boundary edges. ....	62
Figure 3-4. Species rarefaction curves for the three cover crop species. Ca=canola, AWP= Austrian winter pea, RCl=red clover. ....	67
Figure 3-5. Netted abundance of individual bee species collected from canola plots throughout the total experiment. Asterisk indicates actual relative abundance may be greater than shown; collection was not necessary for identification to species-level and not all bees of these groups seen were collected. Only species that represented >1% of the total catch for the crop are shown. ....	68
Figure 3-6. Netted abundance of individual bee species collected from Austrian winter pea plots throughout the total experiment. Asterisk indicates actual relative abundance may be greater than shown; collection was not necessary for identification to species-level and not all bees of these groups seen were collected. ....	68
Figure 3-7. Netted abundance of individual bee species collected from red clover plots throughout the total experiment. Asterisk indicates actual relative abundance may be greater than shown; collection was not necessary for identification to species-level and not all bees of these groups seen were collected. ....	69
Figure 3-8. Abundance of individual bee species collected from combined landscape-level vane and pan traps throughout the span of the experiment. Only species that represented >1% of the total catch are shown.....	69
Figure 3-9. Multivariate biplot indicating the relative association of observed morphospecies groups to the three cover crop species. XYL= <i>Xylocopa virginica</i> , BOM= <i>Bombus sp.</i> , APIS= <i>Apis mellifera</i> , LGDK=large dark bees, SMDK=small dark bees, GREE=green bees, TINY=tiny bodied bees, SYRP=Syrphidae flies.....	70
Figure 3-10. Multivariate biplot of observation morphospecies groupings and their corresponding responses to variations in environmental temperature and wind speed. XYL= <i>Xylocopa virginica</i> , BOM= <i>Bombus sp.</i> , APIS= <i>Apis mellifera</i> , LGDK=large dark bees, SMDK=small dark bees, GREE=green bees, TINY=tiny bodied bees, SYRP=Syrphidae flies. ....	72
Figure 3-11. Average number of bee visits per two-minute observation period. Error bars are standard error of the mean; n=139 for canola, n=140 for AWP, and n=66 for red clover. Bars that do not share the same letter are significantly different at p<0.05 using Tukey's multiple comparison test. ....	73
Figure 3-12. Average bee species richness by crop per one-minute post-observation netting period. Error bars represent standard error of the mean; n=139 for canola, n=140 for AWP, and n=66 for red clover. Bars that do not share the same letter are significantly different at p<0.05 using Tukey's multiple comparison test.....	73
Figure 3-13. Average number of Syrphidae visits per two-minute observation period by crop. Error bars are standard error of the mean; n=139 for canola, n=140 for AWP,	



and n=66 for red clover. Bars that do not share the same letter are significantly different at $p<0.05$ using Tukey's multiple comparison test.....	74
Figure 3-14. Number of open blooms per $0.25\text{m}^2$ represented over time and as a function of increasing density. RCl=Red clover, AWP=Austrian winter pea, PD= Fall planting date.....	75
Figure 3-15. Average number of bee visits per two-minute observation period by canola planting date. Error bars represent standard error; n=40 for planting dates 1 and 2, n=32 for planting date 3, and n=27 for planting date 4. Bars that do not share the same letter are significantly different at $p<0.05$ using Tukey's multiple comparison test.....	77
Figure 3-16. Average bee species richness per two-minute observation period for canola crop by fall planting date. Error bars represent standard error; n=40 for planting dates 1 and 2, n=32 for planting date 3, and n=27 for planting date 4. Bars that do not share the same letter are significantly different at $p<0.05$ using Tukey's multiple comparison test. ....	78
Figure 3-17. Box and whisker plot of the average number of open blooms per $0.25\text{m}^2$ by canola fall planting date across all weeks of the blooming period. Boxes that do not share the same letter are significantly different at $p<0.05$ using Tukey's multiple comparison test. ....	79
Figure 3-18. Average plant density ( $\pm\text{SE}$ ) for canola plots by planting date. N=12 for planting dates 1-3 and n=9 for planting date 4. Bars that do not share the same letter are significantly different at $p<0.05$ using Tukey's multiple comparison test.....	79
Figure 3-19. Box and whisker plot of the average number of blooms per blooming plant by canola planting date across all weeks of the blooming period. Boxes that do not share the same letter are significantly different at $p<0.05$ using Tukey's multiple comparison test. ....	80
Figure 3-20. Correlation of number of bee visits to canola plots as a function of the average flower density for all planting date treatments. $F_{1,137}=26.02$ , $p<0.001$ . ....	80
Figure 3-21. Correlation of bee species richness collected in canola plots as a function of the average flower density for all planting date treatments. $F_{1,137}=3.46$ , $p=0.065$ . ....	81
Figure 3-22. Average number of bee visits per two-minute observation period. Error bars are standard error of the mean; n=26 for planting date 1, n=40 for planting dates 2 and 3, and n=34 for planting date 4. Bars that do not share the same letter are significantly different at $p<0.05$ using Tukey's multiple comparison test.....	82
Figure 3-23. Average number of bee species per one-minute post-observation netting period. Error bars represent standard error of the mean; n=26 for planting date 1, n=40 for planting dates 2 and 3, and n=34 for planting date 4. Bars that do not share the same letter are significantly different at $p<0.05$ using Tukey's multiple comparison test. ....	82

- Figure 3-24. Box and whisker plot of the average number of open blooms per 0.25m<sup>2</sup> by Austrian winter pea planting date across all weeks of the blooming period. Boxes that do not share the same letter are significantly different at  $p < 0.05$  using Tukey's multiple comparison test. ....83
- Figure 3-25. Correlation of number of bee visits on Austrian winter pea plots as a function of the average flower density for all planting date treatments.  $F_{1,138}=25.38$ ,  $p < 0.001$ . ....83
- Figure 3-26. Correlation of bee species richness collected in Austrian winter pea plots as a function of the average flower density for all planting date treatments.  $F_{1,138}=15.2$ ,  $p < 0.001$ . ....84
- Figure 3-27. Average number of bee visits per two-minute observation period. Error bars shown are standard error of the mean;  $n=32$  for both planting dates. Bars that do not share the same letter are significantly different at  $p < 0.05$  using Tukey's multiple comparison test. ....85
- Figure 3-28. Average bee species richness per one-minute post-observation netting period. Error bars shown are standard error of the mean;  $n=32$  for both planting dates. Bars that do not share the same letter are significantly different at  $p < 0.05$  using Tukey's multiple comparison test. ....85
- Figure 3-29. Box and whisker plot of the average number of blooming red clover heads, or flower clusters, by planting date across all weeks of the blooming period studied. Boxes that do not share the same letter are significantly different at  $p < 0.05$  using Tukey's multiple comparison test. Planting date 4 was not included in the analysis as no flowers were present during study. ....86
- Figure 3-30. Correlation between number of bee visits to red clover plots as a function of average flower density across planting dates one and two only.  $F_{1,62}=14.73$ ,  $p < 0.001$ . ..86
- Figure 3-31. Correlation between bee species richness on red clover plots as a function of average flower density across planting dates one and two only.  $F_{1,62}=12.57$ ,  $p < 0.001$ . ..87
- Figure 4-1. Number of open blooms per 0.25m<sup>2</sup> represented over time and illustrating the flower density that would be achieved prior to cover crop termination in a crop rotation window before spring organic corn planting. RCl=Red clover, AWP=Austrian winter pea, PD= Fall planting date. ....98
- Figure 4-2. Number of open blooms per 0.25m<sup>2</sup> represented over time and illustrating the flower density that would be achieved prior to cover crop termination in a crop rotation window before a summer pumpkin crop. RCl=Red clover, AWP=Austrian winter pea, PD= Fall planting date. ....98
- Figure 5-1. Plot map of the cover crop and plant diversity study discussed in Chapter 2. Shaded plots indicate the cover crop treatments selected for study in this project. Triangles represent the locations of landscape-level passive trapping; filled triangles

are traps on project boundary edges, open triangles are traps on the project interior.  
Crops listed on column headings indicate 2013 summer crops grown in that strip. ....101

Figure 5-2. Plot map of cover crop planting date study discussed in Chapter 3. Blocks are illustrated in rows of each crop, main plots by cover crop species, and split-plots by planting date as indicated on the left-hand edge of the map. Planting dates are consistent in rows across the block. Ca= Canola; AWP= Austrian winter pea; RCl= Red clover. ....102

Figure 6-1. Total abundance of bees collected in the landscape-level passive traps across all project dates (April-July). (A) Bees by trap type and color; (B) by trap type only; (C) by trap color only; and (D) by field location (see Figure 5-1 for specific field locations). Bars that do not share the same letter are significantly different at  $p < 0.05$  using Tukey's multiple comparison test. ....103

Figure 6-2. Total abundance of Syrphidae flies collected in the landscape-level passive traps across all project dates (April-July). (A) Syrphidae flies by trap type and color; (B) by trap type only; (C) by trap color only; and (D) by field location (see Figure 5-1 for specific field locations). Bars that do not share the same letter are significantly different at  $p < 0.05$  using Tukey's multiple comparison test. ....104

## LIST OF TABLES

Table 2-1. Land cover classifications for a 250 meter buffer zone surrounding all edges of the main research site. All values are given in percentage of the total area.....	23
Table 2-2. Seeding rates of cover crop treatments. All values are given in kg/ha.....	25
Table 2-3. Species richness table for the three cover crop mixture treatments and the landscape-level passive trapping. Presence/absence data are listed across all dates of the experiment; asterisk indicates species collected with greater than ten individuals....	30
Table 2-4. Full general linear model statistics for bee visitation, bee species richness and Syrphidae visitation by cover crop mixture treatment and transect location. Asterisks indicate significant ( $p < 0.05$ ) variables in each model.....	32
Table 2-5. Environmental covariates and their respective correlations with each bee and Syrphidae response variable. Only covariates with significant ( $p < 0.05$ ) correlations were included as covariates in the general linear models for the corresponding response.....	32
Table 2-6. Correlation of average flower density per $0.25\text{m}^2$ with each bee and Syrphidae response variable. Asterisks indicate significant ( $p < 0.05$ ) correlations.....	40
Table 3-1. Land cover classifications for a 250 meter buffer zone surrounding all edges of the main research site. All values are given in percentage of the total area.....	56
Table 3-2. Species table of all bees collected via hand netting from cover crop flowers or via the landscape-level passive traps across all weeks of the experiment. Species with an asterisk were collected with greater than ten individuals.....	64
Table 3-3. Environmental covariates and their respective correlations with each bee and Syrphidae response variable. Asterisks indicate those significant ( $p < 0.05$ ) correlations that were included as covariates in the general linear models with the corresponding insect response.....	71
Table 3-4. Dates of first recorded bloom, peak bloom, final bloom and quality of winter survival for each cover crop species and planting date.....	76
Table 3-5. Dry fall plant biomass in grams by crop and planting date. Superscripts that do not share the same letter are significantly different at $p < 0.05$ using Tukey's multiple comparison test.....	76
Table 4-1. Examples of additional information or financial resources available about growing cover crops or general pollinator conservation.....	96

## ACKNOWLEDGEMENTS

There are so many people that I ought to thank for helping me to reach this important milestone of my scientific and educational career. While I know that I can never list every person that supported me along this journey, I wish to acknowledge some of the most recent and significantly influential people here below.

I would like to start by thanking my advisor, Mary Barbercheck, and the other members of my committee, Dave Mortensen, Ed Rajotte, and Dave Biddinger, for their frequent encouragement and guidance as I navigated graduate school and the trial of designing and implementing my own independent research project. Similarly, a giant thanks to all the many members of the Cover Crop Cocktails research team; faculty, graduate students, post-docs, technicians, and field helpers. Each one of you has been a support to me in so many ways, and truly made me feel like a valuable member of the sustainable agriculture community here at Penn State. I know that my cover crop plots would never have even gotten into the ground without the direct guidance and help of Mac Burgess and Al Cook. They provided me with the invaluable farming knowledge, tractor power and plot installation assistance I needed to get this project up and running and probably will never understand how grateful I was to them both in the early months of this project.

I would also like to extend a giant thank you to Jason Gibbs of Michigan State University for allowing me a week of his time and expert knowledge. Without him, I would never have been able to complete the species-level identification of the bees collected during this project.

To all of my friends and second family in the department of Entomology, there are so many of you that have been there for me over the past two and a half years, but especially to my dear friends Ariel Rivers, Deonna Soergel, Loren Rivera Vega and Sheena Sidhu. I learned quickly that having great friends and a strong support system is paramount to successfully navigating the inevitable obstacle course that is life as a graduate student. Thank you for always being there for me, ladies.

Additionally, my unending gratitude to my parents and big brothers for teaching me to persevere, reach for my dreams and never be afraid of new places or experiences.

And finally a huge thank you to Mr. Gregory Tenn for being my rock at the end of every day, for reminding me of what I am capable of achieving, and for providing me with my greatest incentive to reach this final goal.

## Chapter 1

### INTRODUCTION

#### Ecosystem Services and Pollination

The functioning of the earth's many natural ecosystems hinge on the production and services provided by plants, animals, microorganisms and their surrounding abiotic conditions. Such resources produced or provided by natural systems, and that are utilized by the human population, are generally known as ecosystem services (Millennium Ecosystem Assessment 2005). Examples of ecosystem services include vital, yet often overlooked, functions such as nutrient recycling, water purification, and seed dispersal. Even more popular press topics like carbon sequestration, food crops and timber resources fit under the ecosystem services umbrella. Each of these natural services is essential for the function and sustainability of both managed (e.g., agriculture) and unmanaged (e.g., primary forest) systems. The work detailed in this thesis focuses on the service of pollination, specifically pollination that could be enhanced in agricultural landscapes through the use of flowering cover crops.

While plants can be pollinated in many different ways (e.g., wind, water, bats, birds), more than three-fourths of the world's plant species require animal pollination of some form (National Research Council 2007, Ollerton et al. 2011). There are many different animal pollinators; however, the majority of flowering plants are specialized for pollination by insects, and primarily by bees (Hymenoptera: Apoidea) (O'Toole and Raw 1991). Most flowering plants are self-incompatible and thus require outside forces to transfer the pollen of one bloom to the stigma of another. Bees, in their search for pollen and nectar as food, facilitate cross-pollination of the plants (O'Toole and Raw 1991). This symbiosis between the two groups has led to a tightly linked mutualistic relationship, including some of the best examples of co-evolution and specialization.

Pollination as an ecosystem service is vital to the continued reproduction of much of the world's food crops and other flowering plants. While many of the primary staple crops across the globe are wind-pollinated (e.g., corn, rice, wheat), several of our more nutrient rich foods such as fruits and nuts are dependent on insects for pollination. In fact, animal-mediated pollination

(primarily by bees) is required for 35% of the world's total food production (Klein et al. 2007). Additionally, it is estimated that globally 87.5% of all flowering plants are animal pollinated (Ollerton et al. 2011). These values put into perspective the crucial role that pollinators play in the reproduction and continued existence of most angiosperms.

### **Meet the Pollinators: Bees**

While there is a great diversity of animals that encompass the term 'pollinators,' the group of interest in this document are bees. Globally there are approximately 20,000 species of bees with about 4,000 living in North America and approximately 370 in the state of Pennsylvania alone (Michener 2007, Donovall, III and VanEngelsdorp 2010). When discussing bees in the context of agriculture, they are often grouped into two categories, managed bees and wild or native bees.

Managed bees are typically social species that have been domesticated by humans and kept in portable hives for a specific purpose, typically crop pollination or honey production. While some native and introduced social or solitary bees are managed by directing their nesting to locations or containers of our choosing, we usually equate 'managed bees' with domesticated honey bees (Colla et al. 2006, Pitts-Singer and Cane 2011). Managed honey bees have perennial hives and are tended and maintained by human beekeepers. In the United States, the most common managed bee is the European honey bee (*Apis mellifera* L.). European honey bees, as their name implies, are native to Europe and were introduced to North America with the early European settlers. While feral honey bee colonies and other locally naturalized introduced bee species do exist within the United States, no species of the genus *Apis* are native to North America. For this reason, honey bees are not included here in discussions about native or wild pollinators within the United States.

Honey bees are the most common insect pollinator used for agricultural production, however, in recent years managed colonies have been struck with an increase in hive disease and pest pressures, among other issues. The winter of 2006-2007 was characterized by very high (30-40%) and unexplained hive mortality from beekeepers in all regions of the United States. The following years showed a continuation of this trend, which has been termed Colony Collapse Disorder (CCD). CCD is characterized not only by the loss of honey bee colonies but also by the unexplained disappearance of the (presumed) dead worker bees. To date, no single causal

mechanism can explain this decline in bee health (VanEngelsdorp et al. 2009, Williams et al. 2010). With the continued occurrence of CCD in honey bee hives and continued expansion of pollinator-dependent crops, there is now a deficit of colonies available to pollinate North American agricultural crops (Aizen and Harder 2009). This not only adds extra stress on the bees and their keepers, but also greatly increases the cost of honey bee hive rentals for growers (Sumner and Boriss 2006). Currently the honey bee research community has identified a series of possible causes of decreased honey bee health with no single source consensus. It is likely, rather, a combination of multiple in-hive and environmental issues. Candidates leading to this decline include increased pesticide use, hive parasites, viruses and other diseases, poor hive nutrition, and other stresses such as long distance transportation of hives for cross-continental pollination needs and general habitat loss (National Research Council 2007, Oldroyd 2007, Blacquière et al. 2012). Honey bee colony decline and all the related economic effects have not only increased the public awareness for the importance of pollinators, but have also spurred an increase in public and research interest in the ‘free’ pollination service that native, unmanaged bees can provide.

Several studies have demonstrated the overall importance of native bees for pollination of a diversity of crops (Kremen et al. 2002, Ricketts 2004, Morandin and Winston 2005, Greenleaf and Kremen 2006a, 2006b). Native bees are not only important pollinators, some are more efficient crop pollinators than managed bees (Winfree et al. 2007, Garibaldi et al. 2013). This enhanced efficiency can arise from greater bee activity under adverse weather conditions (Vicens and Bosch 2000), successful pollen release via buzz pollination (Greenleaf and Kremen 2006b), or by a greater level of pollen deposition per visit in crops like blueberries and watermelon (Javorek et al. 2002, Winfree et al. 2007). Interestingly, behavioral interactions between native bees and honey bees can increase the efficiency of honey bee pollination by altering honey bee behavior (Greenleaf and Kremen 2006a, Brittain et al. 2013). Most native or wild bees, however, are solitary species that cannot be directly managed by humans. For this reason, some farmers are wary to depend solely on native bees for their crop pollination as they are unable to control when and how much pollination will occur. However, given the proper habitat requirements and crop area, native bees have been shown to provide all the pollination necessary for watermelon crops (Winfree et al. 2007). While further scientific study is necessary for other pollinator-dependent crops, it is likely that native bees provide full pollination for many areas as not all fruit and vegetable farmers rent honey bee colonies, yet they often get successful crop production. However, dependence on native pollination is more prevalent in diversified landscapes such as



central Pennsylvania than in more agriculturally dominated landscapes with minimum surrounding natural habitat.

Native bees are very diverse in appearance, behavior, and resource requirements. They are found in colors ranging from golden brown, black to metallic blue or green. Body size and shape also vary widely, from a few millimeters to over an inch in length (Michener 2007). Bees also use a variety of nesting sites. Social bees, such as honey bees or bumble bees (*Bombus spp.*), search for empty caverns as nesting locations (e.g., abandoned rodent burrows or dead logs), while solitary bee species often excavate nesting holes in the ground or in dead wood, or search for tight tubular spaces such as old wood-boring beetle galleries or inside dead, pithy plant stems (O'Toole and Raw 1991).

Bees can be classified into categories based on their foraging styles. Some species specialize in the plants that they visit, while others are generalists. Bees that are specialized for collecting pollen from only a single species or closely related group of plants are known as oligolectic bees (O'Toole and Raw 1991). While they may collect nectar from multiple species, they are specific in their pollen collection. In contrast, polylectic bees are generalists and collect nectar and pollen from a variety of plant species (O'Toole and Raw 1991). Oligolectic bees, because of their restricted foraging requirements, are phenologically closely matched in timing of adult activity with the blooming phenology of their host plants. Polylectic bees, however, are often active for the blooming periods of multiple flowering species and are typically more adaptable to changes in their surrounding environment than their specialist counterparts (O'Toole and Raw 1991). Differences in foraging strategies are also important when considering plant species choices for pollinator gardens or other conservation refuges. Specialist bees are less likely to benefit from a non-native plant species than their generalists counterparts (Hinnert and Hjelmroos-Koski 2009).

Another aspect of bee biology that is of great interest for agricultural pollination as well as general bee conservation is the difference in foraging distances between bee species. While all bees are central place foragers, meaning they radiate from the nesting location daily in search of food, there are significant differences in foraging distance between bees of different body size. Smaller bees tend to fly shorter total distances than larger bees, although this relationship is nonlinear (Greenleaf et al. 2007). Because of a general increase in native habitat fragmentation, particularly in agricultural landscapes, an increase in pollinator-friendly resources spaced evenly throughout a landscape can be crucial to native bee foraging success. Understanding the diversity in behavior, foraging preferences, habits, and nesting requirements found within an area's native

pollinator community is paramount to the successful implementation of any conservation strategy.

### **Global Pollinator Decline and Conservation**

While the decline in managed honey bee colonies is now well-documented, there is also evidence for a global decline in other pollinator groups as well as many pollinator-dependent plants (Potts et al. 2010). Because little information is known about the ecology and life history of many native bee species, there is a deficit of historical datasets necessary to provide solid evidence of decline for some species or regions (National Research Council 2007, Potts et al. 2010). However, bumble bees, being social and one of the more well-studied bee groups, show clear declines of many species across the globe (Goulson et al. 2008, Grixti et al. 2009, Cameron et al. 2011). In areas of the U.S. that do have substantial historical datasets, changes in overall pollinator community structure and diversity can be detected. For example, a recent study in central Illinois found a 50% loss of bee species in a single community over a 120-year period (Burkle et al. 2013). A study focusing on museum specimens in the northeastern U.S. found similar declines of bee species over time, although the decline was only statistically significant for bumble bees (Bartomeus et al. 2013). To improve our ability to monitor the abundance and biodiversity of wild bees across different regions, standardized bee community monitoring protocols are currently under development (Lebuhn et al. 2013).

Presently, there are many threats to the world's pollinators, wild and managed alike. Among these threats are habitat loss, agricultural pesticides, pathogens, disease, and climate change (Potts et al. 2010). These possible causes of pollinator decline have been hypothesized to work independently as well as synergistically. However, it is most likely the combined interaction of these factors that has led to much of the widespread global pollinator decline that we see today. Habitat loss and fragmentation, in particular, is often listed as one of the greatest and most common threats to wild pollinators, particularly bees (Kremen et al. 2002, Ricketts 2004, Goulson et al. 2008, Winfree et al. 2009). Indeed, habitat loss has been shown to have a significant negative relationship for both bee abundance and species richness (Winfree et al. 2009). Overall, the change in terrestrial land-use from natural habitat to managed systems like urban or agricultural environments are responsible for most habitat loss and fragmentation. In fact, both historical datasets mentioned above for central Illinois and the northeastern US showed

a corresponding change in land-use during the time period of the study. Because total land-use change has been predicted to have the greatest effect on global biodiversity of terrestrial ecosystems over the next 100 years (Sala et al. 2000), determining alternative scenarios that limit the effects of habitat change on native pollinators is a significant consideration for the pollinator research and conservation community.

In general, conserving native pollinators is essential for the preservation of pollination services in agriculture and for native plant communities. Maintaining high pollinator diversity is beneficial for total pollination services through a combination of species complementarity and functional resilience (Hoehn et al. 2008). However, because we cannot directly manage most of our native bees, conservation strategies should be focused on preserving the natural habitat and resources that are required for survival. Successful pollinator conservation strategies should include a focus on sufficient floral resources, both spatially across the landscape and throughout the full growing season; adequate nesting habitat for ground and cavity nesting bees; and protection from pesticides and other pollutants. Additionally, because native bees are so diverse, different bee groups are affected by human influences such as habitat destruction and fragmentation in different ways. For example, oligolectic and bees with small foraging ranges are more affected by fragmentation of their habitat than polylectic bees due to increased difficulty of locating adequate specialized resources in the surrounding environment (O'Toole and Raw 1991, Zurbuchen et al. 2010, Bartomeus et al. 2013). Because of this, no single strategy will work for all bee groups, but by having a strong understanding of the needs of, and threats to, local pollinator communities, successful conservation strategies can be found and adopted for all landscapes and habitat types.

### **Agricultural Conservation Strategies**

While pollinator conservation strategies can be necessary in all types of landscapes, agricultural systems, due to their prevalence and distribution across terrestrial environments, as well as frequent dependence on pollination services, are often important and ideal focal points for conservationists. Historically, agriculture is often associated with negative influences on biodiversity and increased land simplification (Matson et al. 1997, Tilman et al. 2001). The transition of landscapes from their natural, undisturbed status to a more managed, cultivated system typically involves the destruction of wildlife resources and natural habitats, including

those of beneficial and pollinating insects. Indeed, this degradation is often so severe that some scientists predict that over the next few decades the global conversion of land from natural habitat to farmland will be one of the leading causes of species extinctions and subsequent loss of ecosystem services (Tilman et al. 2001). Moreover, an increase in cultivated land area not only threatens the stability of natural ecosystems, but also the health of the agroecosystems. Studies show a diminishing return in crop yield and stability with a greater farmer input, which tends to increase the amount of land in cultivation to fill this yield gap. Pollinator-dependent crops are especially sensitive to this diminishing return in land area to yield trend (Garibaldi et al. 2011). Because bees are central-place foragers, this result is likely due to the increased difficulty of pollinators to reach the crop as distance from nesting habitat grows. In fact, bees are the insect group shown to be the most negatively affected by agricultural intensification (Hendrickx et al. 2007). Because farming is significant for global food supply as well as economic profits, this cycle of agricultural land expansion is likely to continue into the future. However, without a change in current agricultural practice standards, this land-use change cycle endangers native pollinator habitats and is likely to lead to increased pollination deficiencies in the future (Aizen et al. 2009).

On the other hand, even though the need for food production and other more short-term material goods and supplies is likely to increase with the growing global population, it is important to consider the need for trade-offs between total agricultural production and sustainable ecosystem services. Many scientists believe that identifying a balance between short-term and long-term sustainability goals is not only possible, but is the only real strategy for counteracting the negative influences of agricultural intensification. To achieve this, researchers need to work towards finding the optimal balance where managed landscapes (e.g., agriculture) can also sustain natural ecosystem functions and valuable wildlife habitat. It is only by linking different disciplines such as agronomy and entomology that the global change in land-use structure can be positively managed and controlled (Foley et al. 2005). This research project highlights the threat to wild pollinator communities that traditional agriculture often presents while focusing on strategies for integrating conservation and cultivated landscapes.

It is important to note that not all agricultural landscapes are created equal. In fact, different agricultural systems vary in levels of natural habitat disturbance. Agricultural practices and ideals exist on a continuum from the most intensive and simplified landscape to a much lower-intensity farming with emphasis is on maintaining biodiversity and natural ecosystem functioning. The agriculture of the last century is often characterized by increased intensification

and expansion. Typically such agriculture is associated with high-yielding monocultures, chemical fertilizers and pesticides, and mechanized production. Indeed, this intensive agriculture has also been shown to negatively affect ecosystem services beyond just the destruction of natural habitat (Matson et al. 1997).

However, not all agricultural land-use has a negative effect on biodiversity. In contrast to conventional agriculture, diversified farming and organic agriculture has been shown to have improved biodiversity and ecosystem services (Hole et al. 2005, Kremen and Miles 2012). For some insect-based services, such as pest control and pollination, diversified farming alone is not sufficient to maintain the service and must be considered in coordination with other land management and conservation strategies (Kremen and Miles 2012). Nevertheless, low-intensity farming and pasturelands in Europe contain many of the most biodiverse habitats of the region (Bignal and McCracken 1996). In the United States, organic agriculture certification, while not exclusive to diversified farming systems, requires more than the elimination of synthetic chemical pesticides or fertilizers; it also is based on the idea of a holistic, whole-farm approach to agriculture (USDA 2013). Conserving habitat for native wildlife is one of the obligations of organic growers, but also one that occasionally comes with its own set of constraints. For example, it is not often desirable for a grower to set-aside large areas of land from cultivation as a habitat refuge when it may be more economic in production.

However, a variety of agricultural conservation strategies and options exist for wildlife refuges and supplemental resources. Additionally, not all strategies require that land be used only for conservation or production; it is also possible to institute a practice that splits an area of land for conservation and production either spatially or temporally. Even though the studies discussed in this thesis were conducted in central Pennsylvania, an area dominated by relatively small and more-diversified farms compared other regions of the US (US Census Bureau 2012), and was performed using USDA certified organic growing practices, it is the hope that the principles and ideas presented and discussed here can be applied to a wide range of temperate agricultural landscape and management systems provided that the conservation needs of native pollinators are properly considered.

The many different conservation strategies that could be incorporated into agricultural lands depend on the location, needs, and goals of the farmer and focal species to be conserved. Many strategies focus on a single species or type of plant or animal, but often conservation strategies are more functional when total biodiversity and species complementarity are taken into consideration (Tschamtko et al. 2005). Two such strategies that can be implemented to the

benefit of many species, but that are often debated against each other in terms of optimal effectiveness, are the concepts of land-sparing and land-sharing.

There is evidence that suggests that some species, including total native plant diversity, is best conserved in agricultural landscapes through the preservation of natural habitat parcels which are maintained separately from the intensively-cultivated land surrounding it (Phalan et al. 2011, Egan and Mortensen 2012). This strategy is commonly referred to as land-sparing. However, true land-sparing as an agricultural conservation scheme must be implemented more intelligently and efficiently than just setting aside parcels of unused land. Other considerations should include a network of corridors between conservation areas and local wildlife communities, attention to detail on wildlife needs and habitat ranges, and functional management and protection of the conserved landscape. Additionally, as high-yielding agriculture is significantly important to this approach, an efficient and sustainable strategy through context-specific farming techniques focused on local growers and less dependent on international agrochemical companies is essential (Fischer et al. 2011, Phalan et al. 2011). To achieve an ideal land-sparing state can be difficult in some areas. However, with or without the infrastructure or space available to implement a fully successful conservation reserve strategy, other agricultural conservation techniques can be applied.

While land-sparing conservation techniques such as the preservation of hedgerows, old fields, and non-tillable land patches has demonstrated evidence of positive success for wildlife, including pollinators (Rands and Whitney 2011), many growers are concerned with creating habitat for pests such as insects and weeds and may be reluctant to implement land-sparing practices. Additional grower concerns may include loss of space, time, and money necessary to grow and maintain such refuges. Within-field strategies, however, may be easier for growers to adopt due to the benefit gained directly to their fields as well as to the local wildlife and the ecosystem services they provide. This is where a different focus of 'land-sharing' comes into play.

Land-sharing is an agricultural conservation term used to identify any form of sustainable farming where practices are modified to increase the biodiversity within the cultivated landscape. Such practices are either modified crop management practices, such as conservation tillage practices, or separated from crop production temporally, such as planting cover crops during otherwise fallow rotation windows. Other examples include polyculture and intercropping. All of these examples are practices commonly found on organic farms due to their direct influence on field-level ecosystem functioning. Advocates of land-sharing argue that wildlife-friendly farming

need not have negative effects on total yield. In fact, a direct positive relationship between cultivated land and biodiversity may be achieved, especially in complex, heterogeneous landscapes (Perfecto and Vandermeer 2010, Hodgson et al. 2010, Egan and Mortensen 2012).

Agricultural landscapes can be deceptively complex in nature. For example, there is often a hidden heterogeneity within most agricultural systems: the dynamic temporal changes that occur within a single landscape. Such seasonal differences in resource availability or location can significantly affect arthropod populations across a growing season (Vasseur et al. 2013). All in all, by focusing on an array of techniques that take into account a combination of practices including preservation of habitat refuges, wildlife-friendly farming, and the seasonal variations across the landscape, an optimal conservation strategy may be found for a wide range of farm types or locations (Hodgson et al. 2010).

For Pennsylvania, where agricultural landscapes are often heterogeneous with remnant forest patches (Egan and Mortensen 2012), the urgency of conservation strategies of any nature is less than perhaps would be considered in more homogeneous, monoculture-dominated landscapes. However, this does not lessen the importance of incorporating conservation strategies to improve the natural systems in heterogeneous landscapes. Pollinators in particular, due to their overall decline and sensitivity to fragmentation and habitat loss, should be of major conservation focus. Because of the already diverse landscape found throughout Pennsylvania, focusing on the within-farm and temporal availability of resources may be the most efficient strategy. For this, we consider the incorporation of winter cover crops into organic farming systems as a combination of these two factors.

### **Flowering Cover Crops for Pollinator Conservation**

Cover crops are plant species grown within a cultivated field during fallow periods in annual cash crop rotation schedules, or intermixed within cash crop plantings. They can be almost any species of plant, but are most commonly grasses and legumes, and can be planted almost anytime during the year depending on the crop rotation and local climate. Most farmers plant cover crops for within-field erosion control, soil fertility management, or weed suppression (Lal et al. 1991, Clark 2007). However, because the addition of cover crops into an annual crop rotation potentially increases spatial and temporal plant diversity levels, it can also act as an agricultural conservation strategy. Such innovative within-field conservation and management

techniques benefit crop productivity as well as supplement resources to native wildlife populations.

Most cover crop research focuses on the benefits to agricultural yield and productivity through soil fertility management or weed suppression (Snapp et al. 2005). Even previous research on insects and cover crops has focused on the impact of cover crops on agricultural pests (e.g., insects, weeds) or on the natural enemies of pests (Bugg 1991, Bond and Grundy 2001). Additionally, much of the focus of such studies is on summer or perennial cover cropping systems such as for vegetable crops (Wyland et al. 1996, Hooks et al. 1998, 2012) orchard crops (Bugg and Waddington 1994), or vineyards (English-Loeb et al. 2003). Vegetable crop research concentrates on the effect of the cover crop on pests or natural enemies during the later months that the vegetable is growing. Additionally, orchards and vineyards typically use cover crops during the growing season when insect communities are most abundant. While this information is important for the production of certain crops and understanding the general interactions between cover crops and insect communities, there seems to be a knowledge gap regarding the influence of cover crops on springtime insect communities, especially those that are not crop pests, such as pollinators.

In temperate climate agriculture, over-wintering cover crops are used most often in annual crop rotations that have fallow periods from mid-fall to spring or early summer (Clark 2007). In Pennsylvania, species chosen for this window may winter-kill and leave the ground fallow in the spring, or be winter-hardy and act as ground cover for both the fall and spring periods. The choices growers make when selecting cover crop species depends on management decisions and desired purpose for the cover crop or cover crop mixture. One variable that is often not discussed in the literature, but which is of great importance when considering cover crops as an agricultural conservation scheme, is the timing, density, and general availability of flowers.

Because a winter cover crop species that also produces flowers has the potential to bloom at either the early or late parts of the growing season (fall and spring), these flowers have the potential to supplement the nutrient resources of bees during times when other flowers are scarce on the landscape. These transitional periods are often crucial in the establishment of bees' foraging and nesting habitats. For example, in California, an increase in early-season floral resources was shown to positively affect the total size of native bumble bee colonies throughout the remainder of the season (Williams et al. 2012). However, floral resource availability in agricultural landscapes is often low in the springtime compared to other times of the season



(Mandelik et al. 2012). For this reason, flowering winter cover crops have the potential to substantially increase the early-season resource availability in agroecosystems.

This thesis extends the effort of typical cover crop research by focusing on the conservation potential of blooming cover crop species for native pollinators, a benefit beyond what is typically considered when planting cover crops. While the discussion about benefits to pollinators from flowering cover crops is not necessary novel (e.g., Mader et al. 2011), evidence given is often anecdotal and supporting research reports in the scientific literature are limited. Additionally, given the many intricacies involved in agricultural production, increased knowledge of how factors such as weather, planting and termination date, and inter-species competition affect the phenology of flower production and density is beneficial to understand how to best implement cover crops as a conservation strategy for native bee communities.

### Literature Cited

- Aizen, M. A., L. A. Garibaldi, S. A. Cunningham, and A. M. Klein. 2009. How much does agriculture depend on pollinators? Lessons from long-term trends in crop production. *Annals of Botany* 103:1579–88.
- Aizen, M. A., and L. D. Harder. 2009. The global stock of domesticated honey bees is growing slower than agricultural demand for pollination. *Current biology* : CB 19:915–8.
- Bartomeus, I., J. S. Ascher, J. Gibbs, B. N. Danforth, D. L. Wagner, S. M. Hedtke, and R. Winfree. 2013. Historical changes in northeastern US bee pollinators related to shared ecological traits. *Proceedings of the National Academy of Sciences of the United States of America* 110:4656–60.
- Signal, E. M., and D. I. McCracken. 1996. Low-intensity farming systems in the conservation of the countryside. *Journal of Applied Ecology* 33:413–424.
- Blacquière, T., G. Smagghe, C. a M. van Gestel, and V. Mommaerts. 2012. Neonicotinoids in bees: a review on concentrations, side-effects and risk assessment. *Ecotoxicology (London, England)* 21:973–92.
- Bond, W., and A. C. Grundy. 2001. Non-chemical weed management in organic farming systems. *Weed Research* 41:383–405.
- Brittain, C., N. Williams, C. Kremen, and A.-M. Klein. 2013. Synergistic effects of non- Apis bees and honey bees for pollination services. *Proceedings of the Royal Society B: Biological Sciences* 280.
- Bugg, R. L. 1991. Cover crops and control of arthropod pests of agriculture. Pages 157–165 in W. L. Hargrove, editor. *Cover Crops for Clean Water*. Soil and Water Conservation Society.
- Bugg, R. L., and C. Waddington. 1994. Using cover crops to manage arthropod pests of orchards : A review. *Agriculture, Ecosystems and Environment* 50:11–28.
- Burkle, L. A., J. C. Marlin, and T. M. Knight. 2013. Plant-Pollinator Interactions over 120 Years: Loss of Species, Co-Occurrence, and Function. *Science* 339:1611–1615.
- Cameron, S. a, J. D. Lozier, J. P. Strange, J. B. Koch, N. Cordes, L. F. Solter, and T. L. Griswold. 2011. Patterns of widespread decline in North American bumble bees. *Proceedings of the National Academy of Sciences of the United States of America* 108:662–7.
- Clark, A. (Ed.). 2007. *Managing Cover Crops Profitably* 3rd ed. Sustainable Agriculture Research and Education (SARE).
- Colla, S. R., M. C. Otterstatter, R. J. Gegear, and J. D. Thomson. 2006. Plight of the bumble bee: Pathogen spillover from commercial to wild populations. *Biological Conservation* 129:461–467.

- Donovall, III, L. R., and D. VanEngelsdorp. 2010. A Checklist of the Bees (Hymenoptera : Apoidea) of Pennsylvania. *Journal of the Kansas Entomological Society* 83:7–24.
- Egan, J. F., and D. A. Mortensen. 2012. A comparison of land-sharing and land-sparing strategies for plant richness conservation in agricultural landscapes. *Ecological applications : a publication of the Ecological Society of America* 22:459–71.
- English-Loeb, G., M. Rhainds, T. Martinson, and T. Ugine. 2003. Influence of flowering cover crops on *Anagrus* parasitoids (Hymenoptera: Mymaridae) and *Erythroneura* leafhoppers (Homoptera: Cicadellidae) in New York vineyards. *Agricultural and Forest Entomology* 5:173–181.
- Fischer, J., P. Batary, K. S. Bawa, L. Brussaard, M. J. Chappell, Y. Clough, G. C. Daily, J. Dorough, T. Hartel, L. E. Jackson, A. M. Klein, C. Kremen, T. Kuemmerle, D. B. Lindenmayer, H. A. Mooney, I. Perfecto, S. M. Philpott, T. Tschardt, J. Vandermeer, T. C. Wanger, and H. Von Wehrden. 2011. Conservation: Limits of Land Sparing. *Science* 334:593.
- Foley, J. A., R. Defries, G. P. Asner, C. Barford, G. Bonan, S. R. Carpenter, F. S. Chapin, M. T. Coe, G. C. Daily, H. K. Gibbs, J. H. Helkowski, T. Holloway, E. a Howard, C. J. Kucharik, C. Monfreda, J. a Patz, I. C. Prentice, N. Ramankutty, and P. K. Snyder. 2005. Global consequences of land use. *Science (New York, N.Y.)* 309:570–4.
- Garibaldi, L. A., M. A. Aizen, A. M. Klein, S. A. Cunningham, and L. D. Harder. 2011. Global growth and stability of agricultural yield decrease with pollinator dependence. *Proceedings of the National Academy of Sciences of the United States of America* 108:5909–14.
- Garibaldi, L. A., I. Steffan-Dewenter, R. Winfree, M. A. Aizen, R. Bommarco, S. A. Cunningham, C. Kremen, L. G. Carvalheiro, L. D. Harder, O. Afik, I. Bartomeus, F. Benjamin, V. Boreux, D. Cariveau, N. P. Chacoff, J. H. Dudenhöffer, B. M. Freitas, J. Ghazoul, S. Greenleaf, J. Hipólito, A. Holzschuh, B. Howlett, R. Isaacs, S. K. Javorek, C. M. Kennedy, K. M. Krewenka, S. Krishnan, Y. Mandelik, M. M. Mayfield, I. Motzke, T. Munyuli, B. A. Nault, M. Otieno, J. Petersen, G. Pisanty, S. G. Potts, R. Rader, T. H. Ricketts, M. Rundlöf, C. L. Seymour, C. Schüepp, H. Szentgyörgyi, H. Taki, T. Tschardt, C. H. Vergara, B. F. Viana, T. C. Wanger, C. Westphal, N. Williams, and A. M. Klein. 2013. Wild pollinators enhance fruit set of crops regardless of honey bee abundance. *Science* 339:1608–11.
- Goulson, D., G. C. Lye, and B. Darvill. 2008. Decline and conservation of bumble bees. *Annual review of entomology* 53:191–208.
- Greenleaf, S. S., and C. Kremen. 2006a. Wild bees enhance honey bees' pollination of hybrid sunflower. *Proceedings of the National Academy of Sciences of the United States of America* 103:13890–5.
- Greenleaf, S. S., and C. Kremen. 2006b. Wild bee species increase tomato production and respond differently to surrounding land use in Northern California. *Biological Conservation* 133:81–87.

- Greenleaf, S. S., N. M. Williams, R. Winfree, and C. Kremen. 2007. Bee foraging ranges and their relationship to body size. *Oecologia* 153:589–96.
- Grixti, J. C., L. T. Wong, S. a. Cameron, and C. Favret. 2009. Decline of bumble bees (*Bombus*) in the North American Midwest. *Biological Conservation* 142:75–84.
- Hendrickx, F., J.-P. Maelfait, W. Van Wingerden, O. Schweiger, M. Speelmans, S. Aviron, I. Augenstein, R. Billeter, D. Bailey, R. Bukacek, F. Burel, T. Diekötter, J. Dirksen, F. Herzog, J. Liira, M. Roubalova, V. Vandomme, and R. Bugter. 2007. How landscape structure, land-use intensity and habitat diversity affect components of total arthropod diversity in agricultural landscapes. *Journal of Applied Ecology* 44:340–351.
- Hinners, S. J., and M. K. Hjelmroos-Koski. 2009. Receptiveness of Foraging Wild Bees to Exotic Landscape Elements. *The American Midland Naturalist* 162:253–265.
- Hodgson, J. A., W. E. Kunin, C. D. Thomas, T. G. Benton, and D. Gabriel. 2010. Comparing organic farming and land sparing: optimizing yield and butterfly populations at a landscape scale. *Ecology letters* 13:1358–67.
- Hoehn, P., T. Tschardtke, J. M. Tylianakis, and I. Steffan-Dewenter. 2008. Functional group diversity of bee pollinators increases crop yield. *Proceedings. Biological sciences / The Royal Society* 275:2283–91.
- Hole, D. G., A. J. Perkins, J. D. Wilson, I. H. Alexander, P. V. Grice, and a. D. Evans. 2005. Does organic farming benefit biodiversity? *Biological Conservation* 122:113–130.
- Hooks, C. R. R., J. Hinds, E. Zobel, and T. Patton. 2012. Impact of crimson clover dying mulch on two eggplant insect herbivores. *Journal of Applied Entomology*:1-11.
- Hooks, C. R. R., H. . Valenzuela, and J. Defrank. 1998. Incidence of pests and arthropod natural enemies in zucchini grown with living mulches. *Agriculture, Ecosystems & Environment* 69:217–231.
- Javorek, A. S. K., K. E. Mackenzie, and S. P. Vander Kloet. 2002. Comparative Pollination Effectiveness Among Bees ( Hymenoptera : Apoidea ) on Lowbush Blueberry ( Ericaceae : Vaccinium angustifolium ). *Annals of the Entomological Society of America* 95:345–351.
- Klein, A.-M., B. E. Vaissière, J. H. Cane, I. Steffan-Dewenter, S. a Cunningham, C. Kremen, and T. Tschardtke. 2007. Importance of pollinators in changing landscapes for world crops. *Proceedings of the Royal Society B: Biological Sciences* 274:303–13.
- Kremen, C., and A. Miles. 2012. Ecosystem Services in Biologically Diversified versus Conventional Farming Systems : Benefits , Externalities , and Trade-Offs. *Ecology and Society* 17.

- Kremen, C., N. M. Williams, and R. W. Thorp. 2002. Crop pollination from native bees at risk from agricultural intensification. *Proceedings of the National Academy of Sciences of the United States of America* 99:16812–6.
- Lal, R., E. Regnier, D. J. Eckert, W. M. Edwards, and R. Hammond. 1991. Expectations of cover crops for sustainable agriculture. Pages 1–11 in W. L. Hargrove, editor. *Cover Crops for Clean Water*. Soil and Water Conservation Society.
- LeBuhn, G., S. Droege, E. F. Connor, B. Gemmill-Herren, S. G. Potts, R. L. Minckley, T. Griswold, R. Jean, E. Kula, D. W. Roubik, J. Cane, K. W. Wright, G. Frankie, and F. Parker. 2013. Detecting insect pollinator declines on regional and global scales. *Conservation biology : the journal of the Society for Conservation Biology* 27:113–20.
- Mader, E., M. Shepherd, M. Vaughan, S. H. Black, and G. LeBuhn. 2011. *Attracting Native Pollinators: Protecting North America's Bees and Butterflies*. Storey Publishing, North Adams, MA.
- Mandelik, Y., R. Winfree, T. Neeson, and C. Kremen. 2012. Complementary habitat use by wild bees in agro-natural landscapes. *Ecological applications : a publication of the Ecological Society of America* 22:1535–46.
- Matson, P. A., W. J. Parton, A. G. Power, and M. J. Swift. 1997. Agricultural Intensification and Ecosystem Properties. *Science* 277:504–509.
- Michener, C. D. 2007. *The Bees of the World* 2nd Ed. The John Hopkins University Press, Baltimore, MD.
- Millennium Ecosystem Assessment. 2005. *Ecosystems and Human Well-being: Synthesis*. Pages 1–155. Island Press, Washington, DC.
- Morandin, L. A., and M. L. Winston. 2005. Wild bee abundance and seed production in conventional, organic, and genetically modified canola. *Ecological Applications* 15:871–881.
- National Research Council. 2007. *Status of Pollinators in North America*. The National Academies Press, Washington, DC.
- O'Toole, C., and A. Raw. 1991. *Bees of the World*. Facts on File, New York, NY.
- Oldroyd, B. P. 2007. What's killing American honey bees? *PLoS biology* 5:e168.
- Ollerton, J., R. Winfree, and S. Tarrant. 2011. How many flowering plants are pollinated by animals? *Oikos* 120:321–326.
- Perfecto, I., and J. Vandermeer. 2010. The agroecological matrix as alternative to the land-sparing/agriculture intensification model. *Proceedings of the National Academy of Sciences of the United States of America* 107:5786–91.

- Phalan, B., M. Onial, A. Balmford, and R. E. Green. 2011. Reconciling food production and biodiversity conservation: land sharing and land sparing compared. *Science* 333:1289–91.
- Pitts-Singer, T. L., and J. H. Cane. 2011. The alfalfa leafcutting bee, *Megachile rotundata*: the world's most intensively managed solitary bee. *Annual review of entomology* 56:221–37.
- Potts, S. G., J. C. Biesmeijer, C. Kremen, P. Neumann, O. Schweiger, and W. E. Kunin. 2010. Global pollinator declines: trends, impacts and drivers. *Trends in Ecology & Evolution* 25:345–53.
- Rands, S. A., and H. M. Whitney. 2011. Field margins, foraging distances and their impacts on nesting pollinator success. *PloS one* 6:e25971.
- Ricketts, T. H. 2004. Tropical Forest Fragments Enhance Pollinator Activity in Nearby Coffee Crops. *Conservation Biology* 18:1262–1271.
- Sala, O. E., F. S. Chapin, J. J. Armesto, E. Berlow, J. Bloomfield, R. Dirzo, E. Huber-Sanwald, L. F. Huenneke, R. B. Jackson, A. Kinzig, R. Leemans, D. M. Lodge, H. A. Mooney, M. Oesterheld, N. L. Poff, M. T. Sykes, B. H. Walker, M. Walker, and D. H. Wall. 2000. Global biodiversity scenarios for the year 2100. *Science* 287:1770–4.
- Snapp, S. S., S. M. Swinton, R. Labarta, D. Mutch, J. R. Black, R. Leep, J. Nyiraneza, and K. O'Neil. 2005. Evaluating Cover Crops for Benefits, Costs and Performance withing Cropping System Niches. *Agronomy Journal* 97:322–332.
- Sumner, D. A., and H. Boriss. 2006. Bee-economics and the Leap in Pollination Fees. *Agricultural and Resource Economics Update* 9:9–11.
- Tilman, D., J. Fargione, B. Wolff, C. D'Antonio, a Dobson, R. Howarth, D. Schindler, W. H. Schlesinger, D. Simberloff, and D. Swackhamer. 2001. Forecasting agriculturally driven global environmental change. *Science* 292:281–4.
- Tscharntke, T., A. M. Klein, A. Kruess, I. Steffan-Dewenter, and C. Thies. 2005. Landscape perspectives on agricultural intensification and biodiversity - ecosystem service management. *Ecology Letters* 8:857–874.
- US Census Bureau. 2012. The 2012 Statistic Abstract: Agriculture. <http://www.census.gov/compendia/statab/cats/agriculture.html>
- USDA. 2013. National Organic Program: Organic Standards. [http://www.ams.usda.gov/AMSV1.0/ams.fetchTemplateData.do?template=TemplateN&rightNav1=NOSBlinkNOSBMeetings&topNav=&leftNav=&page=NOPOrganicStandard&resultType=.](http://www.ams.usda.gov/AMSV1.0/ams.fetchTemplateData.do?template=TemplateN&rightNav1=NOSBlinkNOSBMeetings&topNav=&leftNav=&page=NOPOrganicStandard&resultType=)
- VanEngelsdorp, D., J. D. Evans, C. Saegerman, C. Mullin, E. Haubruge, B. K. Nguyen, M. Frazier, J. Frazier, D. Cox-Foster, Y. Chen, R. Underwood, D. R. Tarpy, and J. S. Pettis. 2009. Colony collapse disorder: a descriptive study. *PloS one* 4:e6481.

- Vasseur, C., A. Joannon, S. Aviron, F. Burel, J.-M. Meynard, and J. Baudry. 2013. The cropping systems mosaic: How does the hidden heterogeneity of agricultural landscapes drive arthropod populations? *Agriculture, Ecosystems & Environment* 166:3–14.
- Vicens, N., and J. Bosch. 2000. Weather-Dependent Pollinator Activity in an Apple Orchard , with Special Reference to *Osmia cornuta* and *Apis mellifera* ( Hymenoptera : Megachilidae and Apidae ). *Environmental Entomology* 29:413–420.
- Williams, G. R., D. R. Tarpy, D. vanEngelsdorp, M.-P. Chauzat, D. L. Cox-Foster, K. S. Delaplane, P. Neumann, J. S. Pettis, R. E. L. Rogers, and D. Shutler. 2010. Colony Collapse Disorder in context. *BioEssays : news and reviews in molecular, cellular and developmental biology* 32:845–6.
- Williams, N. M., J. Regetz, and C. Kremen. 2012. Landscape-scale resources promote colony growth but not reproductive performance of bumble bees. *Ecology* 93:1049–58.
- Winfree, R., R. Aguilar, D. P. Vazquez, G. LeBuhn, and M. A. Aizen. 2009. A meta-analysis of bees' responses to anthropogenic disturbance. *Ecology* 90:2068–2076.
- Winfree, R., N. M. Williams, J. Dushoff, and C. Kremen. 2007. Native bees provide insurance against ongoing honey bee losses. *Ecology letters* 10:1105–13.
- Wyland, L. J., L. E. Jackson, W. E. Chaney, K. Klonsky, S. T. Koike, and B. Kimple. 1996. Winter cover crops in a vegetable cropping system : Impacts on nitrate leaching , soil water , crop yield , pests and management costs. *Agriculture, Ecosystems and Environment* 59:1–17.
- Zurbuchen, A., L. Landert, J. Klaiber, A. Müller, S. Hein, and S. Dorn. 2010. Maximum foraging ranges in solitary bees: only few individuals have the capability to cover long foraging distances. *Biological Conservation* 143:669–676.

## Chapter 2

# THE EFFECT OF COVER CROP SPECIES DIVERSITY ON NATIVE BEE VISITATION AND DIVERSITY

## INTRODUCTION

Pollination as an ecosystem service is vital to the reproduction of much of the world's food crops and other flowering plants. In fact, animal-mediated pollination (primarily by bees) is required for 35% of the world's total food production and 87.5% of all flowering plants (Klein et al. 2007, Ollerton et al. 2011). While managed honey bee colonies are most often used for agricultural pollination, native bees are also known to play an important role in crop pollination (Kremen et al. 2002, Ricketts 2004, Morandin and Winston 2005, Greenleaf and Kremen 2006a, 2006b). However, despite the fact that pollination services are often necessary in agricultural production, the importance of native bee communities extends far beyond this purpose.

Unfortunately, the world's pollinators are in decline. While the recent decrease in managed honey bee colonies is now well-documented (VanEngelsdorp et al. 2009, Williams et al. 2010), there is also evidence for a global decline in other pollinator groups as well as many pollinator-dependent plants (National Research Council 2007, Potts et al. 2010). The possible causes of this decline include loss of natural habitat, agricultural pesticides, pathogens, disease, and climate change (Potts et al. 2010). However, it is most likely the combined interaction of these factors that has led to much of the widespread global pollinator decline that we see today. Habitat loss and fragmentation, in particular, are often listed as some of the greatest and most common threats to wild pollinators, particularly bees (Kremen et al. 2002, Ricketts 2004, Goulson et al. 2008, Winfree et al. 2009).

Historically, agriculture is often associated with negative influences on biodiversity and increased land simplification (Matson et al. 1997, Tilman et al. 2001). This is because the transition of landscapes from their natural, undisturbed status to cultivated and managed systems typically involves the destruction of previously-established wildlife resources and natural habitats, including those of beneficial and pollinating insects. Indeed, bees are the insect group



shown to be the most negatively affected by agricultural intensification (Hendrickx et al. 2007). Additionally, because total land-use change has been predicted to have the greatest effect on global biodiversity of terrestrial ecosystems over the next 100 years (Sala et al. 2000), determining alternative scenarios that limit the effects of habitat change on native pollinators is a significant consideration for the pollinator research and conservation community. However, because we cannot directly manage most of our native bees, conservation strategies should be focused on preserving the natural habitat and resources that are required for their survival.

One strategy for increasing agricultural conservation and ecosystem health is a trend toward organic or diversified farming. Compared to conventional farming, organic agriculture can increase biodiversity and ecosystem services (Hole et al. 2005, Kremen and Miles 2012) and support a greater diversity of native bees (Holzschuh et al. 2006). In the United States, organic agriculture certification, while not exclusive to diversified farming systems, requires more than the elimination of synthetic chemical pesticides or fertilizers; it also is based on the idea of a holistic, whole-farm approach to agriculture (USDA 2013). For this, wildlife-friendly farming and conserving some aspect of habitat for native wildlife is one of the obligations of organic growers.

Additionally, agricultural landscapes can be deceptively complex in nature. For example, the dynamic temporal changes that occur over the course of the growing season often illustrate a type of hidden heterogeneity in agricultural systems. Such seasonal differences in resource availability or location can significantly affect arthropod populations (Vasseur et al. 2013). All in all, by focusing on an array of techniques that take into account a combination of practices including preservation of habitat refuges, wildlife-friendly farming, and the seasonal resource variations across the landscape, an optimal conservation strategy may be found for a wide range of farm types or locations (Hodgson et al. 2010). As a partial solution to this conservation need, we consider the incorporation of winter cover crops into organic farming systems to help enhance floral resources both spatially and temporally across the landscape.

Cover crops are plant species grown within a cultivated field during fallow periods in annual cash crop rotation schedules, or intermixed within cash crop plantings. They can be almost any species of plant, but are mostly commonly grasses and legumes, and can be planted almost anytime during the year depending on the crop rotation and local climate. Most farmers plant cover crops for within-field erosion control, soil fertility management, or weed suppression (Lal et al. 1991, Snapp et al. 2005, Clark 2007). However, because the addition of cover crops into an annual crop rotation potentially increases spatial and temporal plant diversity levels, it can

also act as an agricultural conservation strategy. Additionally, by selecting cover crops that also produce insect-visited flowers attractive to native pollinators, this technique can benefit crop productivity as well as supplement resources to native wildlife populations.

This study was conducted as a component of a larger, interdisciplinary field experiment studying the benefits and trade-offs of cover crop species and trait diversity on field-level agronomic and ecosystem services. Services studied include annual and perennial weed suppression, nitrogen fixation and retention, soil microbial activity, drought buffering, insect pest reduction, and a combination of the above on plant productivity and farm profitability. The cover crop species chosen for this experiment were selected for a range of plant functional traits and included an annual grass, oats (*Avena sativa* L.), perennial grass, cereal rye (*Secale cereale* L.), two legumes, red clover (*Trifolium pratense* L.) and Austrian winter pea (*Pisum sativum subsp. arvense* L.), and two brassicas, forage radish (*Raphanus sativus* L.) and canola (*Brassica napus* L. 'Wichita'). In addition, all cover crop species studied are commonly used or appropriate for use in the central Pennsylvania study area.

Cover crops can be important not only for increasing spatial resource availability, but also for creating temporal plant diversity in agricultural systems. Many flowering cover crops (e.g., buckwheat, cowpeas) are used in the summertime and are planted in spring or early-summer (Clark 2007). While these may produce summer flowers and be beneficial to native bee communities, our study focuses on overwintering cover crops and early-spring cover crop blooming potential. Additionally, as the prevailing agricultural practice in the region is dairy production, local cropping systems are dominated by dairy forage crops (e.g., corn, soybean, wheat, and hay) (USDA 2007). Therefore, this experiment was conducted within cover crop windows appropriate for these crop rotations. All treatments were planted in late August after winter wheat harvest and terminated in mid-May prior to planting of organic field corn.

The addition of supplemental flowering resources to an agricultural landscape has been shown to be beneficial to native bee communities and is often used as a pollinator conservation strategy (Tuell et al. 2008, Winfree 2010). Our study focuses on whether the addition of a spring-blooming cover crop species could achieve the same results. This timeframe is especially important because some native bee species benefit from an increase in springtime floral resources (Elliott 2009, Williams et al. 2012). However, agricultural landscapes are often lacking these early-season flowers compared with other natural or fallow areas (Winfree et al. 2007, Williams et al. 2012, Mandelik et al. 2012). It is for this reason that increasing spring flowering resources

within cultivated fields would likely have a large influence on overall resource availability for native bees during this time of the year.

However, as cover crops are grown for a multitude of agronomic benefits, many farmers have demonstrated recent interest in planting diverse cover crop mixtures in an effort to maximize the combination of ecosystem services from different plant groups. For example, some cover crops are best at increasing or maintaining soil nutrients, some produce high biomass and are better at suppressing weed growth, and others still have the main role of using deep root systems to naturally aerate the soil (Clark 2007). By combining several of these cover crop groups into a single planting, farmers may help expand the overall ecosystem service benefits provided. However, if these cover crop mixtures also include a flowering species in the hopes of providing pollinator conservation benefits, it is important to have an understanding of the effect of increased cover crop plant diversity on the potential visiting pollinator community. This experiment aims to provide knowledge on what the influence of increased cover crop plant diversity may be on subsequent flower density on native bee visitation abundance and species richness. A good understanding of the applied use and effects of these cover crop mixtures and management practices is critical to informing recommendations on the benefits of flowering cover crops and cover crop mixtures on native bee conservation efforts.

## **MATERIALS AND METHODS**

### **Site Description**

This experiment was conducted at a single site established on approximately 11 ha of land at the Penn State University Russell E. Larson Research and Education Center (RELREC) near Rock Springs, Pennsylvania. The dominant soil type at the site is Hagerstown silt loam with soil texture being predominantly clay loam with variability in silt, clay and sand. This land is in transition to organic certification and was managed in accordance with the USDA National Organic Standards (USDA 2013).

A geographic information systems (GIS) analysis of the site and surrounding habitat types were performed using base map satellite imagery, hand-digitized land cover classifications, and ground-truthing (ArcMap version 10.1, 2012 ESRI Inc.). A buffer zone of 250 m from the

edge of the total experiment site was chosen to accommodate the foraging range of many of the smaller, ground nesting bees collected during the study. All cropland within this boundary is part of the RELERC and was separated into tilled, arable crop land and non-tilled farm land which includes areas such as grassy drive rows and field edges (Table 2-1, Figures 2-1, 2-2). The primary crops grown in this area are field grains such as corn, soybeans, and winter wheat, although some small patches of spring canola for oilseed production are also grown. Additionally, some neighboring experiments grow cover crops in their rotation including some flowering species such as hairy vetch and red clover.

Table 2-1. Land cover classifications for a 250 meter buffer zone surrounding all edges of the main research site. All values are given in percentage of the total area.

<b>Land Cover Classification</b>	<b>250 m</b>
Arable cropland	67.2
Non-tilled cropland (grass alleyways, roadsides, other less-disturbed areas)	16.7
Semi-natural habitat (tree lines, un-managed grassland)	0.4
Residential	1.1
Managed lawn	6.2
Industrial surface (roads, farm buildings)	8.4

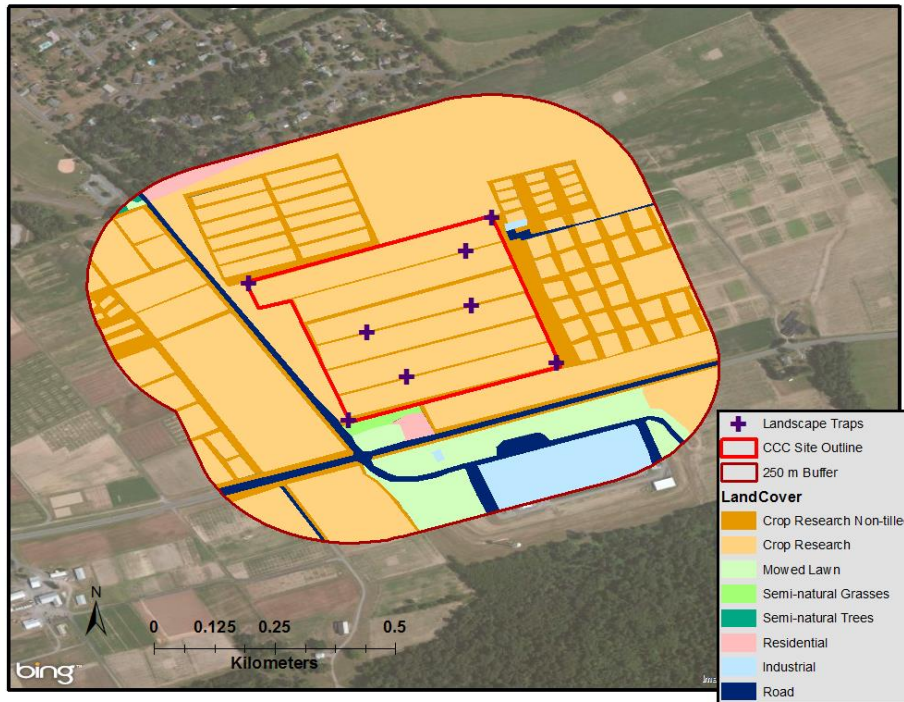


Figure 2-1. Land cover classification map showing the allocation of various cover classes of the landscape in a 250 m buffer zone surrounding the research site (outlined in red). Also shown is the placement of the eight landscape-level trap locations (purple +).



Figure 2-2. Aerial photograph showing the full project site (outlined in red) and surrounding landscape.

## Planting and plot establishment

To study the benefits and trade-offs of cover crop diversity on a suite of ecosystem functions, a full-entry, stripped, randomized complete block design field experiment was established at RELREC (see Appendix A for plot map). This experiment is part of a larger project studying the benefits and trade-offs of cover crop species and mixtures on several ecosystem services, including soil nutrient management, weed suppression, and insect pest regulation. The twelve treatments in the larger experiment are comprised of six fall-planted cover crop species grown in monoculture, five cover crop mixture treatments, and a fallow treatment embedded in a winter wheat (*Triticum aestivum* L.), corn (*Zea mays* L.), and soybean (*Glycine max* (L.) Merr.) rotation. Treatments were each replicated four times.

The experiment described here focused on three treatments within the larger experiment; canola (*Brassica napus* L. 'Wichita') in monoculture, a four species mix including canola, medium red clover (*Trifolium pratense* L.), Austrian winter pea (*Pisum sativum* subsp. *arvense* L.), and cereal rye (*Secale cereale* L.), and a six species mix that included canola, medium red clover, Austrian winter pea, cereal rye, forage radish (*Raphanus sativus* L.) and oats (*Avena sativa* L.). Cover crop plots were established after summer barley harvest which replaced wheat in the rotation for the first year of the study and planted on 25-26 August 2012. Seeding rates are given in Table 2-2 below.

Table 2-2. Seeding rates of cover crop treatments. All values are given in kg/ha.

	<b>Monoculture</b>	<b>4 Species Mixture</b>	<b>6 Species Mixture</b>
Canola ( <i>Brassica napus</i> 'Wichita')	12.7	6.3	3.1
Austrian Winter Pea ( <i>Pisum sativum</i> subsp. <i>arvense</i> )	-	43.4	21.7
Medium Red Clover ( <i>Trifolium pratense</i> )	-	6.7	3.4
Cereal Rye ( <i>Secale cereale</i> )	-	27.8	27.8
Forage Radish ( <i>Raphanus sativus</i> )	-	-	3.6
Oats ( <i>Avena sativa</i> )	-	-	25.8

Planting of cover crops was completed using a no-till cone plot drill with double disc openers (Great Plains 3P605NT) which planted nine rows of seed, with row spacing of 19 cm. Total treatment plot size measured approximately 24 m by 27 m. Planting depth was consistent at

approximately 1 cm depth for all cover crop species. Treatment plots were managed without irrigation and in accordance to organic production standards with no synthetic chemical herbicides or pesticides applied (USDA 2013). Some manual weed suppression techniques were implemented in the fall and early spring in attempt to limit annual weed populations during vulnerable, young cover crop and cash crop growing stages.

### **Blooming phenology**

To assess cover crop bloom over time and across treatments, two randomly located 0.25m<sup>2</sup> quadrats were flagged in each plot prior to the onset of flowering. One quadrat was located along a transect at the interface between the crop and permanent drive row (edge) and one was located along an inner, center-plot transect (center) approximately 12 m from, and parallel to, the drive row edge. All plots were monitored for open canola flowers at least once per week and the total number of open blooms were recorded for each quadrat from the onset of plot flowering until the day prior to cover crop termination.

Six additional quadrats per plot were monitored within 24 hours of the pollinator observations. These were randomly located along the edge and center transects, with three quadrats on each transect. For these observations, the total number of open blooms per 0.25m<sup>2</sup> was recorded to serve as a measure of average bloom density for the plot across time.

### **Observations and netting**

All bee visitation, species richness and Syrphidae data values analyzed are from a single collection date of May 2, 2013. Visitation and netting data was additionally collected on May 13, 2013 but was excluded from all analyses considering observation rates as the average temperature during morning data collection was 5.4C with an average of 8.5C in the afternoon. These temperatures were deemed too cold for sufficient bee foraging behavior and corresponded with very low insect data collected, and was excluded from analysis. However, data from May 13 was included in the total species richness by mixture data (Table 2-3), and all plant and flower analyses.

As a method of quantifying abundance of visitation to the blooming crop, visual pollinator observations were conducted on two occasions after the canola began to bloom and prior to cover crop termination in mid-May. Each plot was visually monitored for bee floral visitation for two minutes per transect, twice per day; once in the morning and once in the early afternoon. The observer walked at a slow and steady rate along the transect recording all bees and Syrphidae flies that visited the open cover crop blooms during the two minute period. Each bee or fly that was observed landing on the reproductive parts of the flower was recorded to the lowest taxonomic level possible from visual estimations (modified from Westphal et al. 2008). Groups that were easy to determine on-the-wing were identified to genus (e.g., *Bombus*, *Apis*, *Xylocopa*), whereas those that were smaller or more difficult to identify in motion were grouped into morphospecies categories (e.g., large dark bee, green bee, small dark bee, syrphid fly).

After completing the observations on the edge and center transect of each plot, both transects were walked for an additional 60 seconds and all bees and Syrphidae flies observed landing on the reproductive parts of the cover crop flowers were collected with an aerial insect net. Netted specimens were killed by placing them in a glass collecting jar with an ethyl acetate-soaked plaster bottom and returned to the lab. All bees were identified to species level. These specimens served as a reference for the morphospecies categories of the preceding observation period as well as an overall indicator of the bee species richness associated with each treatment. As richness and not abundance was the goal of netted specimens, bees that were obviously of the same species (e.g., *Xylocopa virginica* L.) and were collected already once during the netting period on that plot were not collected in duplicate even if observed on the flowers of interest. *Apis mellifera* (L.) specimens were not collected often during netting periods as species identification was confident during the visual observations.

Bee species were identified using a series of online or hard-copy taxonomic keys, as appropriate by genus or subgenus (“Discover Life” n.d., Bouseman and LaBerge 1979, McGinley 1986, Gibbs 2011, Gibbs et al. 2013). Some of the more difficult species of the genus *Andrena* or *Lasioglossum* were identified by Jason Gibbs of Michigan State University. Previously identified species were also confirmed by J. Gibbs. Voucher specimens have been submitted to the Frost Museum at the Pennsylvania State University.

Weather data including air temperature, thirty second average wind speed, and sky condition were collected twice for each session, before and after each morning and afternoon observation and netting period using a Kestrel 2000™ thermo-anemometer



(www.kestrelmeters.com). This was repeated for both the morning and afternoon observation sessions.

### **Landscape area traps (passive collection)**

To compare the community of bees that were collected from the flowering cover crops to the total bee community in the landscape during the same time period we placed two types of passive traps, pan and plastic vane traps, across the site on a weekly basis from April 22, 2013 until the completion of the study. Traps were set for 48 hr with collected specimens removed from the traps every 24 hr. Traps were placed in groups comprised of three pan traps (1 each white, yellow, blue) and two vane traps (one each blue and yellow). In total, 8 groups of pan and vane traps were deployed across the 11 ha study area. Four of the trap groups were set on the edge of the field site, one at each corner (edge traps), and four were placed in the interior of the project boundaries (interior traps) (see plot map in Appendix A). All traps were located along grass access roads surrounding the study plots, and were as evenly distributed across the study area as was possible given road spacing constraints and other field operation concerns.

Methodology used for pan trapping was adapted from Westphal et al. (2008) and from *The Bee Inventory Plot* report (LeBuhn et al. 2002). The pan traps, also referred to as bee bowls, were constructed of 96 ml Dart brand soufflé cups spray painted in white (Krylon® Fusion for plastic), florescent yellow (Krylon®), or florescent blue (ACE® Glo Spray). All yellow and blue bowls were also painted with a primer of the white plastic-bonding paint. All three colors were used as different bee groups have been shown to be attracted to differently colored traps; most commonly white, blue and yellow (Leong and Thorp 1999). Bowls were mounted above the ground on 1.2 m tall, 2 cm diameter wooden dowels. Atop each dowel one painted bowl was attached using a single thumb tack. The final setup consisted of another bowl of the same color placed within the support, thumb-tacked bowl. The sample bowl was filled three-fourths full with soapy water. The soapy water mix was created using 2 L and approximately 1 ml of blue, original non-scented Dawn® dish soap. The soap reduced the water surface tension to increase insect catch without additional odors.

The plastic vane traps (SpringStar, Inc.) are constructed of yellow and florescent blue perpendicular vanes and a collecting tub attached below the vanes. All vane traps were used in their unaltered form. Each trap was suspended from a 1.2m galvanized steel shepherd's hook

purchased from a local garden supply store. Approximately 200 ml of soapy water mixture were added to the collection tub of each vane trap to act as an insect killing agent.

One trap of each type and color were set at each of the 8 site locations. The group of five different traps (pan and vane traps) were randomly ordered along a linear transect on the edge of the nearest plot and spaced approximately two meters from the nearest trap (Figure 2-3). All insects collected in the traps that were neither bee nor Syrphidae flies were considered bycatch and discarded. Summary data for landscape-level trap type and color efficiencies is given in Appendix B.



Figure 2-3. Landscape-level passive trapping example setup for one of the project boundary edges.

### **Statistical Data Analysis**

Data analysis was performed with the R statistical language (R Core Team 2012). Additional R packages used for linear mixed-effects models (Bates et al. 2013), Tukey's multiple comparison post-hoc test (Hothorn et al. 2008), and species richness rarefaction curves (Jacobs 2011, Oksanen et al. 2013). Generalized linear mixed-effect models with a Poisson distribution were used for all bee-based data analyses using random errors appropriate for blocked designs and with significantly correlated temperature or wind covariates as appropriate (Table 2-5). Plant-based data analysis was performed using linear mixed models considering the random errors necessary for blocked designs and repeated measures analysis over the time span of each

cover crop mixture. Correlations between individual variables were performed using simple linear regression.

## RESULTS

A total of 23 bee species were collected throughout the span of this experiment, 13 from the cover crop mixture treatments and 18 from the passive landscape-level traps (Table 2-3). However, as is shown from the lack of asymptotic reach of any mixture treatment on the species rarefaction curves shown in Figure 2-4, the limited sampling periods and sizes collected during this experiment did not likely sample the total species richness of any treatment. It can be assumed, therefore, that the potential species richness of bees on canola flowers in each treatment is greater than shown in this study.

Table 2-3. Species richness table for the three cover crop mixture treatments and the landscape-level passive trapping. Presence/absence data are listed across all dates of the experiment; asterisk indicates species collected with greater than ten individuals.

	Monoculture	4 Species Mix	6 Species Mix	Landscape
<i>Agapostemon texanus</i> (Cresson, 1872)				X
<i>Andrena arabis</i> (Robertson, 1897)	X	X	X	
<i>Andrena carlini</i> (Cockerell, 1901)	X			
<i>Andrena forbesii</i> (Robertson, 1983)		X	X	
<i>Andrena hippotes</i> (Robertson, 1895)	X	X		X
<i>Andrena imitatrix</i> (Cresson, 1872)	X			X*
<i>Andrena miserabilis</i> (Cresson, 1872)	X			X
<i>Andrena rugosa</i> (Robertson, 1891)	X	X		
<i>Apis mellifera</i> (Linnaeus, 1758)	X	X		
<i>Bombus bimaculatus</i> (Cresson, 1863)				X
<i>Bombus impatiens</i>	X			X

(Cresson, 1863)				
<i>Bombus ternarius</i> (Say, 1837)				X
<i>Halictus confusus</i> (Smith, 1854)				X
<i>Halictus rubicundis</i> (Christ, 1791)	X			X
<i>Lasioglossum coriaceum</i> (Smith, 1853)			X	X
<i>Lasioglossum forbseii</i> (Robertson, 1890)				X
<i>Lasioglossum hitchensi</i> (Gibbs, 2012)	X			X
<i>Lasioglossum paradmirandum</i> (Knerer and Atwood, 1966)	X	X		X
<i>Lasioglossum pilosum</i> (Smith, 1853)				X
<i>Lasioglossum truncatum</i> (Robertson, 1901)				X
<i>Osmia cornifrons</i> (Radoszkowski, 1902)				X
<i>Osmia taurus</i> (Smith, 1873)				X
<i>Xylocopa virginica</i> (Linnaeus, 1771)				X
<b>Total Species Richness</b>	<b>11</b>	<b>6</b>	<b>3</b>	<b>18</b>

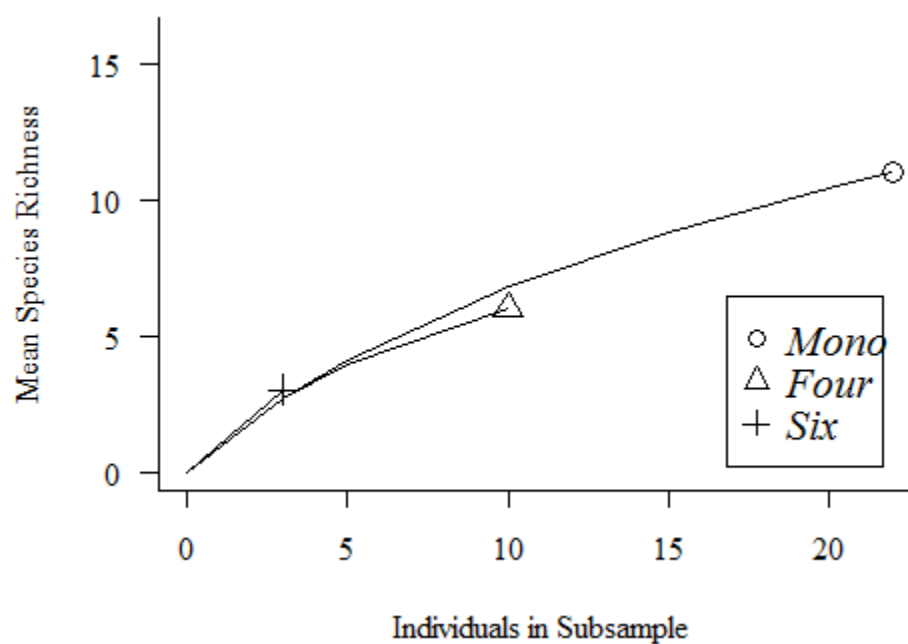


Figure 2-4. Species rarefaction curves for the three cover crop mixture treatments.

There were significant effects of both mixture and transect location (edge or center of the plot) on number of bee visits per two minute observation period (Table 2-4, Figures 2-5 and 2-6) with greater visitation in canola monocultures than mixtures as well as on center transects than on plot edges. In contrast, while mixture treatment was a significant predictor for bee species richness per two minute observation period with the greatest species richness observed in the monoculture, no differences were observed in species richness across plot transects (Table 2-4, Figures 2-7 and 2-8). For the number of Syrphidae visitors per two minute observation, no differences were observed across mixture treatment or transect location (Table 2-4, Figures 2-9 and 2-10). The environmental variables of temperature and wind speed were shown to have a non-significant influence on bee visitation abundance, bee species richness, or Syrphidae visitation (Table 2-5) and thus were not included in the previously mentioned models.

Table 2-4. Full general linear model statistics for bee visitation, bee species richness and Syrphidae visitation by cover crop mixture treatment and transect location. Asterisks indicate significant ( $p < 0.05$ ) variables in each model.

	Bee Visitation Abundance		Bee Species Richness		Syrphidae Visitation Abundance	
	<i>Chi-sq(DF)</i>	<i>p-value</i>	<i>Chi-sq(DF)</i>	<i>p-value</i>	<i>Chi-sq(DF)</i>	<i>p-value</i>
Mix	13.009(2)	0.001*	8.8037(2)	0.012*	4.208(2)	0.122
Transect	5.6795(1)	0.017*	0.143(1)	0.705	1.9918(1)	0.158

Table 2-5. Environmental covariates and their respective correlations with each bee and Syrphidae response variable. Only covariates with significant ( $p < 0.05$ ) correlations were included as covariates in the general linear models for the corresponding response.

	Bee Visitation Abundance			Bee Species Richness			Syrphidae Visitation Abundance		
	<i>F-stat</i> ( <i>DF<sub>num</sub></i> , <i>DF<sub>den</sub></i> )	<i>p-value</i>	<i>R<sup>2</sup></i>	<i>F-stat</i> ( <i>DF<sub>num</sub></i> , <i>DF<sub>den</sub></i> )	<i>p-value</i>	<i>R<sup>2</sup></i>	<i>F-stat</i> ( <i>DF<sub>num</sub></i> , <i>DF<sub>den</sub></i> )	<i>p-value</i>	<i>R<sup>2</sup></i>
Average Temperature	0.89(1,46)	0.351	0.019	3.03(1,46)	0.088	0.062	2.67(1,46)	0.109	0.055
Average Wind Speed	0.89(1,46)	0.351	0.019	3.03(1,46)	0.088	0.062	2.67(1,46)	0.109	0.055

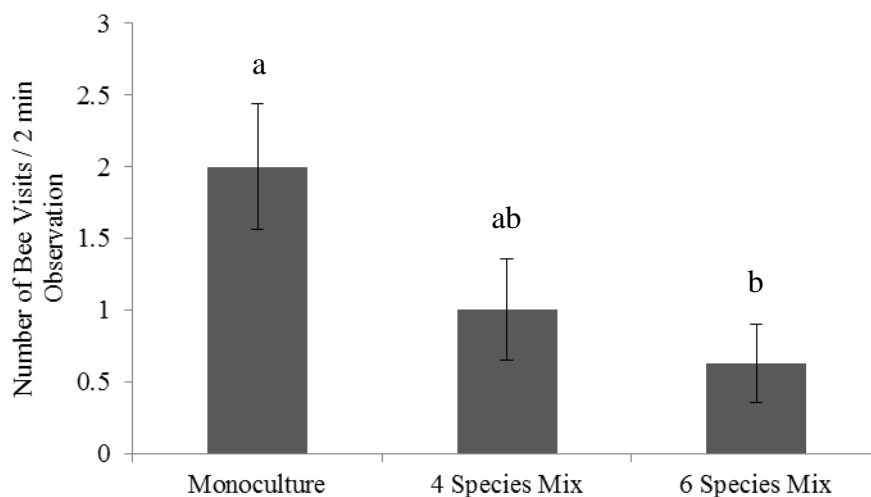


Figure 2-5. Average total bee visitation abundance per two minute observation period. Error bars are standard error of the mean; n=16. Bars that do not share the same letter are significantly different at  $p < 0.05$  using Tukey's multiple comparison test.

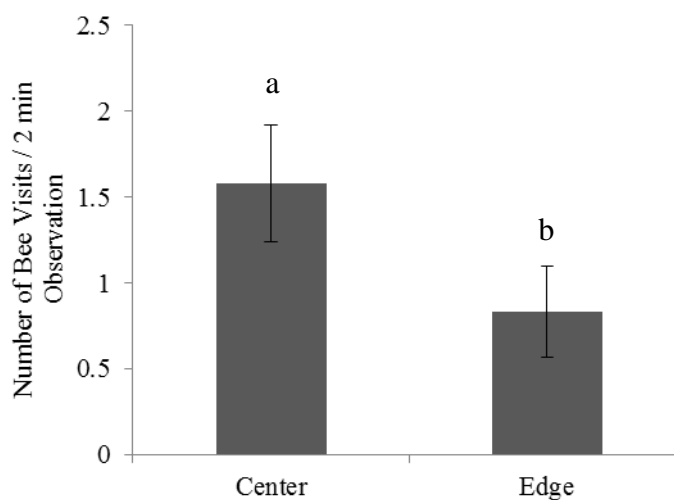


Figure 2-6. Average total bee visitation abundance by plot location transect per two minute observation period. Error bars are standard error of the mean; n=24. Bars that do not share the same letter are significantly different at  $p < 0.05$  using Tukey's multiple comparison test.

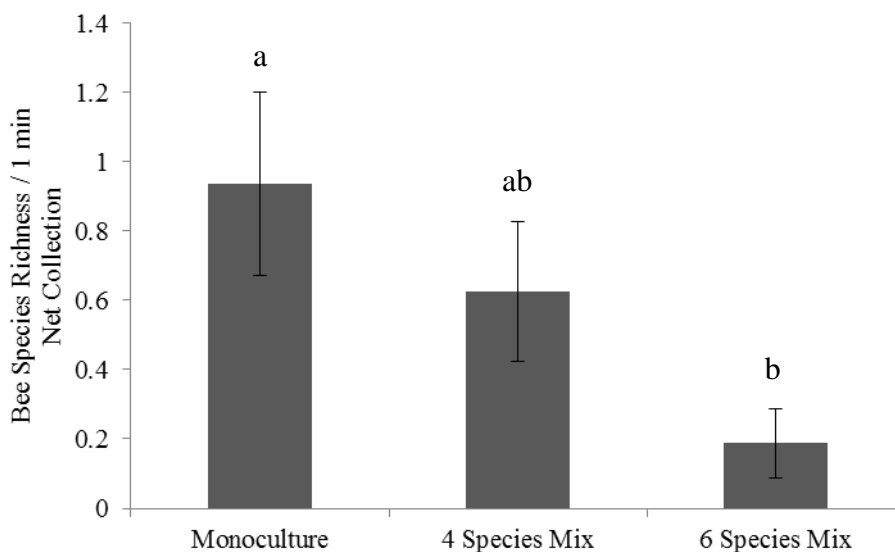


Figure 2-7. Average bee species richness per one minute post-observation netting period by mixture treatment. Error bars are standard error of the mean;  $n=16$ . Bars that do not share the same letter are significantly different at  $p<0.05$  using Tukey's multiple comparison test.

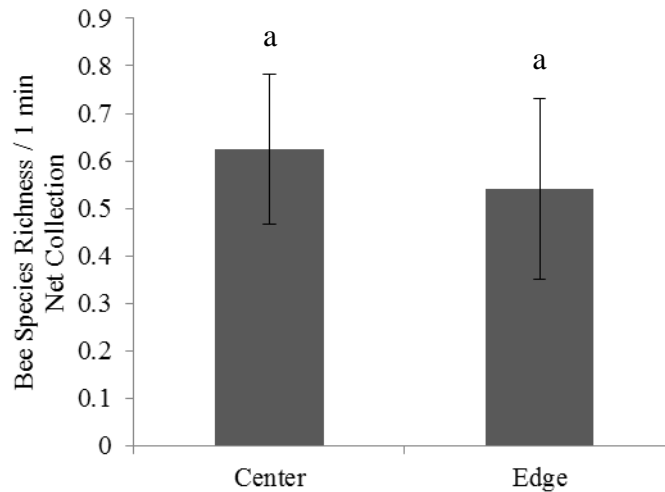


Figure 2-8. Average bee species richness per one minute post-observation netting period by plot edge and plot center transects. Error bars are standard error of the mean;  $n=24$ . Bars that do not share the same letter are significantly different at  $p<0.05$  using Tukey's multiple comparison test.

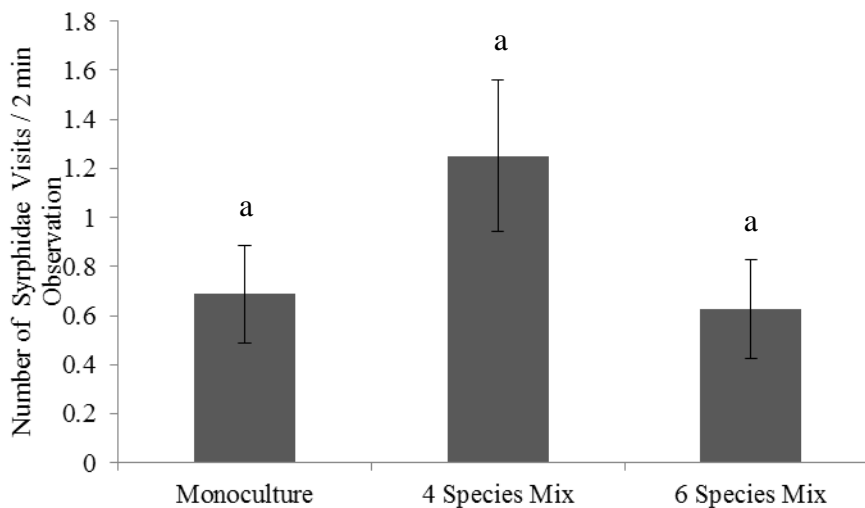


Figure 2-9. Average number of Syrphidae visits per two minute observation period. Error bars are standard error of the mean; n=16. Bars that do not share the same letter are significantly different at  $p < 0.05$  using Tukey's multiple comparison test.

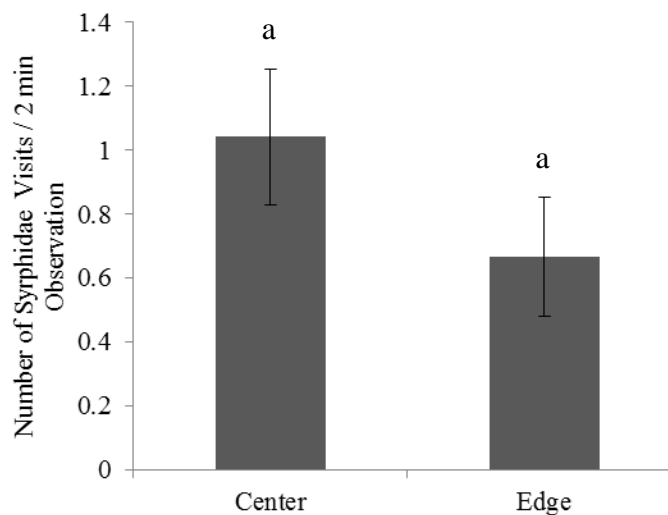


Figure 2-10. Average number of Syrphidae visits per two minute observation period across plot edges and plot center transects. Error bars are standard error of the mean; n=24. Bars that do not share the same letter are significantly different at  $p < 0.05$  using Tukey's multiple comparison test.

There were significant differences between total canola plant density in all mixture treatments (Figure 2-11) but not by plot transect location (Figure 2-12). In general, canola plant density was greatest in the canola monoculture, intermediate in the four species mix, and least in the 6 species mix. Similar trends were found for the average density of canola flowers which



were significantly different by mixture (Figures 2-13 and 2-15) but not by transect location (Figure 2-14). In contrast, only the monoculture treatment had significantly more average blooms per blooming plant than did either the four or six species mixtures (Figure 2-16), but this trend was also not significant by transect location (Figure 2-17).

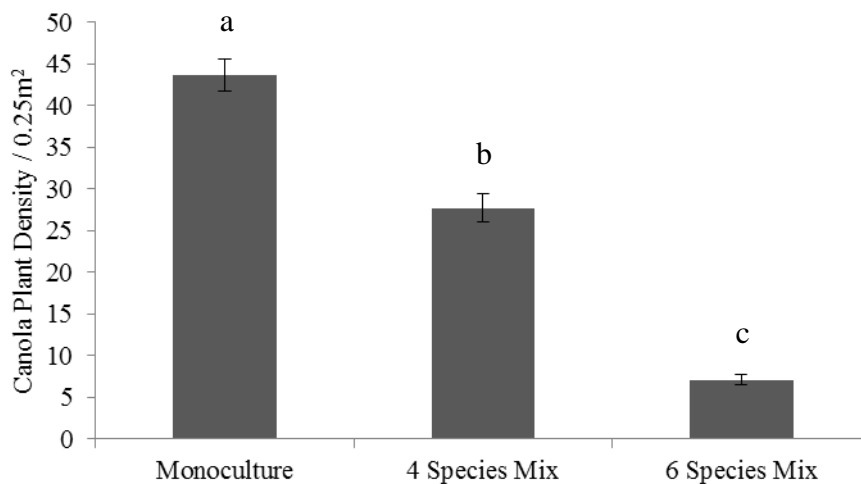


Figure 2-11. Average canola plant density per 0.25m<sup>2</sup> by mixture treatment. Error bars are standard error of the mean; n= 24. Bars that do not share the same letter are significantly different at  $p < 0.05$  using Tukey's multiple comparison test.

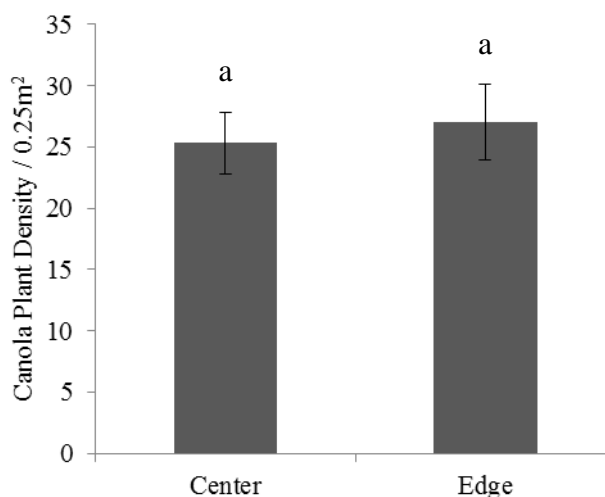


Figure 2-12. Average canola plant density per 0.25m<sup>2</sup> by plot transect location. Error bars are standard error of the mean; n= 36. Bars that do not share the same letter are significantly different at  $p < 0.05$  using Tukey's multiple comparison test.

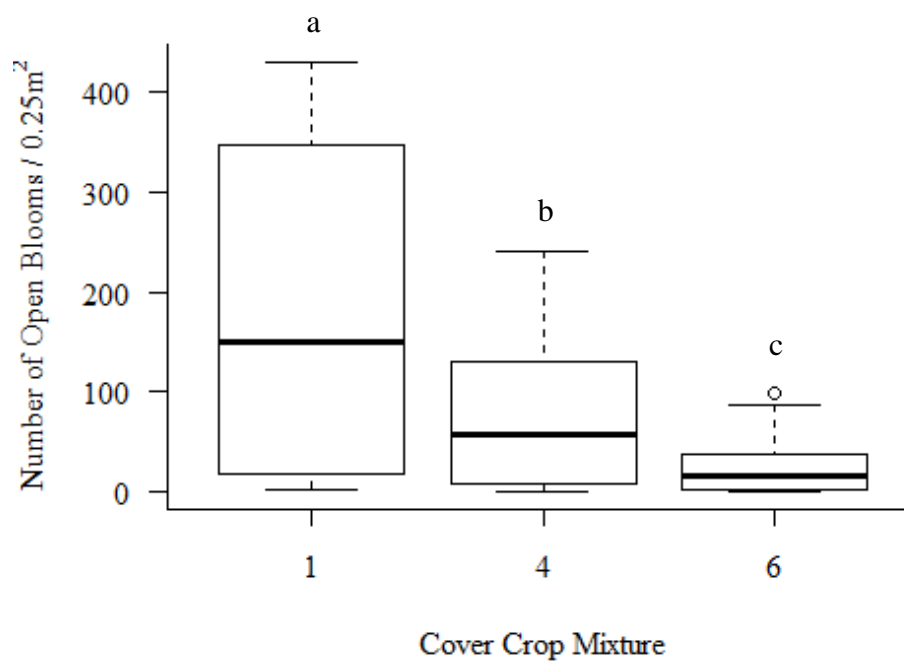


Figure 2-13. Box and whisker plot of the number of open blooms per 0.25m<sup>2</sup> by cover crop mixture treatment and across the total time span of the experiment (1=Monoculture, 4=4 Species Mix, 6=6 Species Mix). Boxes that do not share the same letter are significantly different at  $p < 0.05$  using Tukey's multiple comparison test.

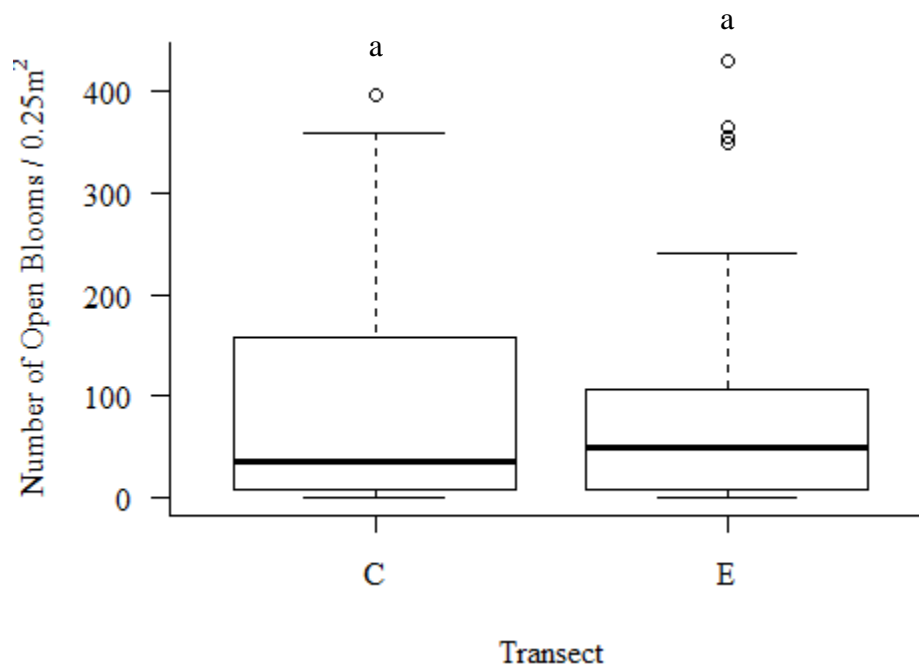


Figure 2-14. Box and whisker plot of the number of open blooms per 0.25m<sup>2</sup> by plot location transect across the total time span of the experiment (C=center of plot and E=edge of plot). Boxes that do not share the same letter are significantly different at  $p < 0.05$  using Tukey's multiple comparison test.

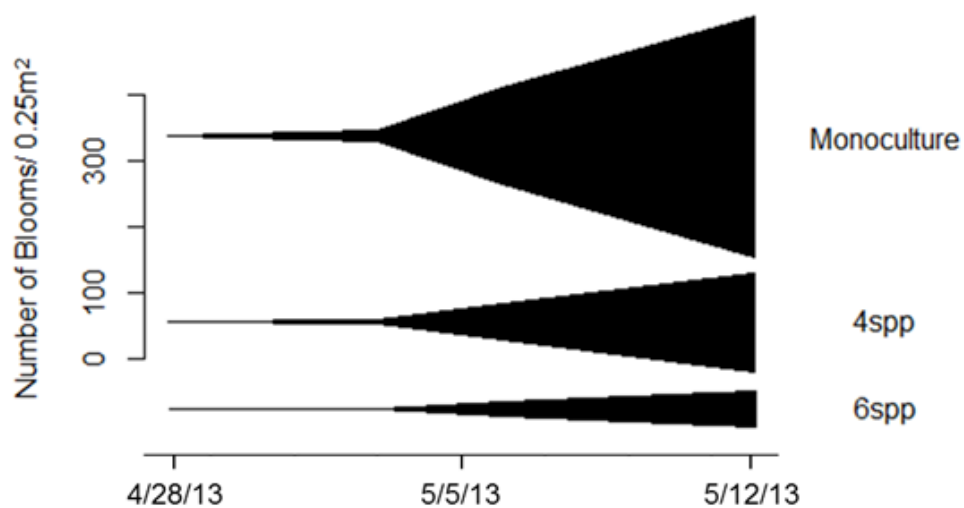


Figure 2-15. Number of open blooms present per 0.25m<sup>2</sup> illustrated over time and as a function of increasing flower density.

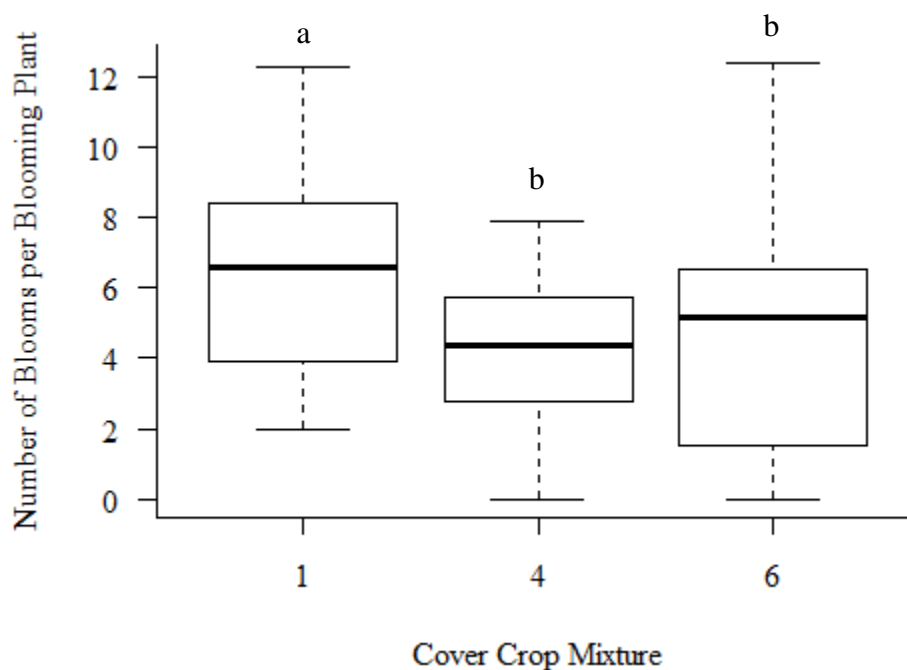


Figure 2-16. Box and whisker plot of the average number of open blooms per blooming plant by cover crop mixture treatment (1=Monoculture, 4=4 Species Mix, 6=6 Species Mix). Boxes that do not share the same letter are significantly different at  $p < 0.05$  using Tukey's multiple comparison test.

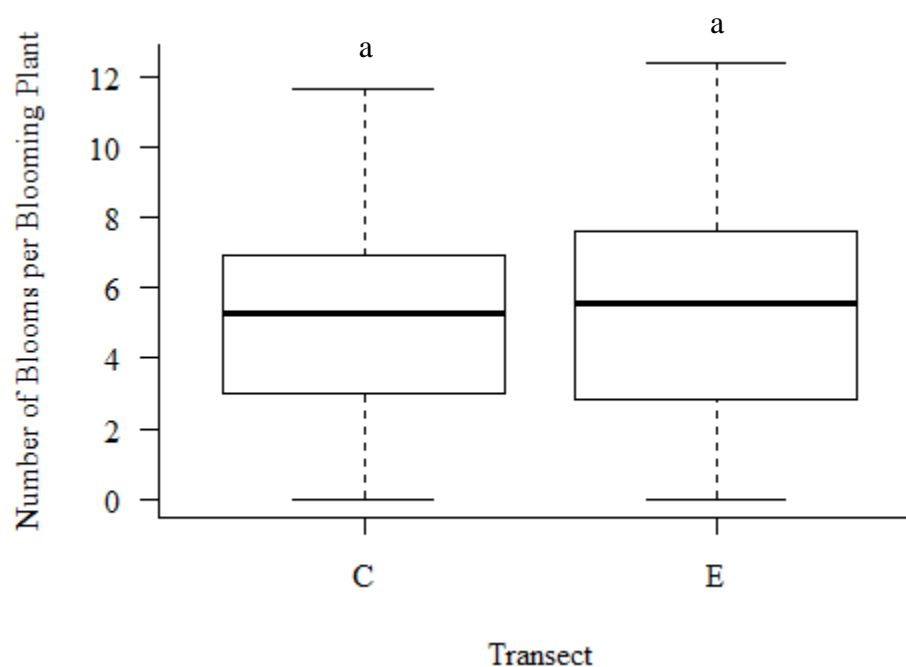


Figure 2-17. Box and whisker plot of the average number of open blooms per blooming plant by plot transect locations (C=center of plot and E=edge of plot). Boxes that do not share the same letter are significantly different at  $p < 0.05$  using Tukey's multiple comparison test.

Average flower density per  $0.25\text{m}^2$  was significantly correlated with both average bee visitation abundance and bee species richness but not with average number of Syrphidae visits (Table 2-6; Figures 2-18, 2-19 and 2-20 respectively). Flower density was not included as a covariate in the general linear model for any response variable however, due to the high correlation found between average flower density and mixture treatments ( $F_{1,45}=30.83$ ,  $p < 0.001$ ,  $R^2=0.58$ ). This was not the case for flower density and plot transect location ( $F_{1,46}=1.16$ ,  $p=0.288$ ,  $R^2=0.025$ ) so differences in behavioral response of the insects to plot location is likely not related to floral resource variations.

By the date of termination (13 May 2013), an average of 95.5% ( $\pm 0.01$ ) plants in the canola monoculture were in bloom, with 83.5% ( $\pm 0.04$ ) and 71.9% ( $\pm 0.10$ ) canola plants in bloom for the four and six species mixtures, respectively.

Table 2-6. Correlation of average flower density per 0.25m<sup>2</sup> with each bee and Syrphidae response variable. Asterisks indicate significant ( $p < 0.05$ ) correlations.

	Bee Visitation Abundance			Bee Species Richness			Syrphidae Visitation Abundance		
	<i>F-stat</i> ( <i>df<sub>num</sub></i> , <i>df<sub>den</sub></i> )	<i>p-value</i>	<i>R</i> <sup>2</sup>	<i>F-stat</i> ( <i>df<sub>num</sub></i> , <i>df<sub>den</sub></i> )	<i>p-value</i>	<i>R</i> <sup>2</sup>	<i>F-stat</i> ( <i>df<sub>num</sub></i> , <i>df<sub>den</sub></i> )	<i>p-value</i>	<i>R</i> <sup>2</sup>
Average Flower Density	11.5(1,46)	0.001*	0.200	15.2(1,46)	<0.001*	0.248	0.70(1,46)	0.407	0.015

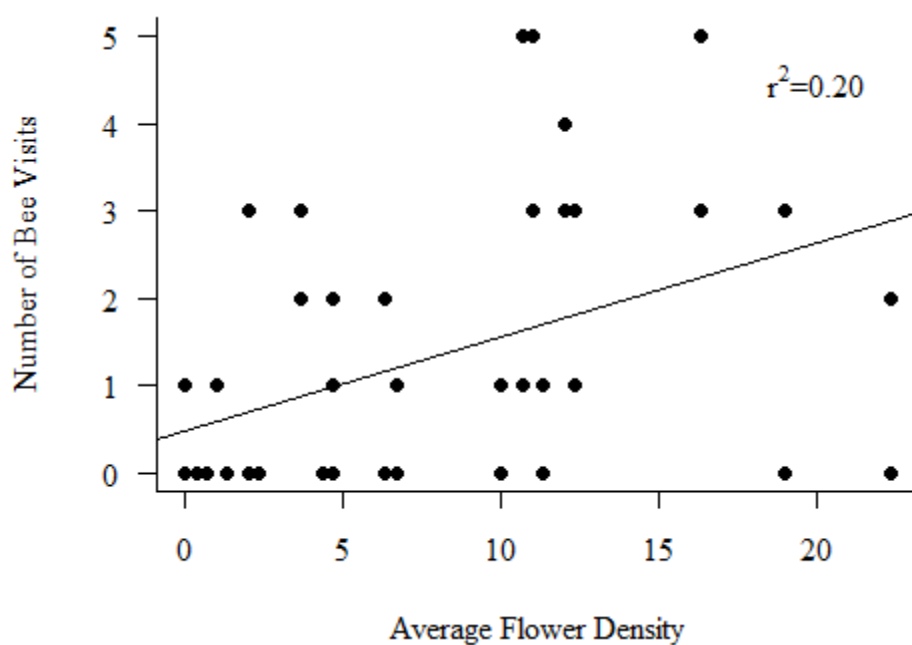


Figure 2-18. Simple linear regression correlation of the number of bee visits to cover crop mixtures as a function of the average flower density of each treatment plot and transect.  $F_{1,46} = 11.52$ ,  $p = 0.001$ .

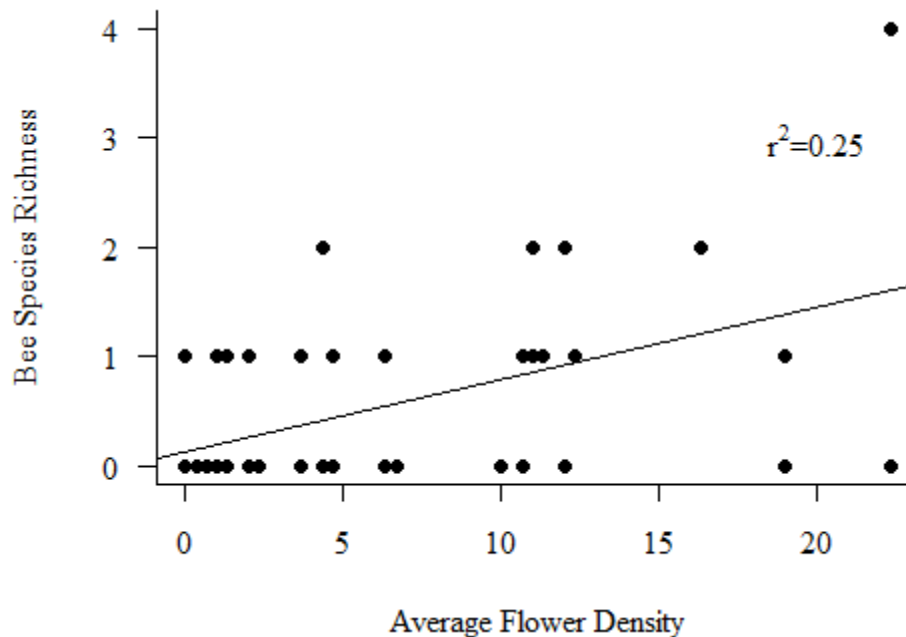


Figure 2-19. Simple linear regression correlation of bee species richness to cover crop mixtures as a function of the average flower density of each treatment plot and transect.  $F_{1,46}=15.2$ ,  $p<0.001$ .

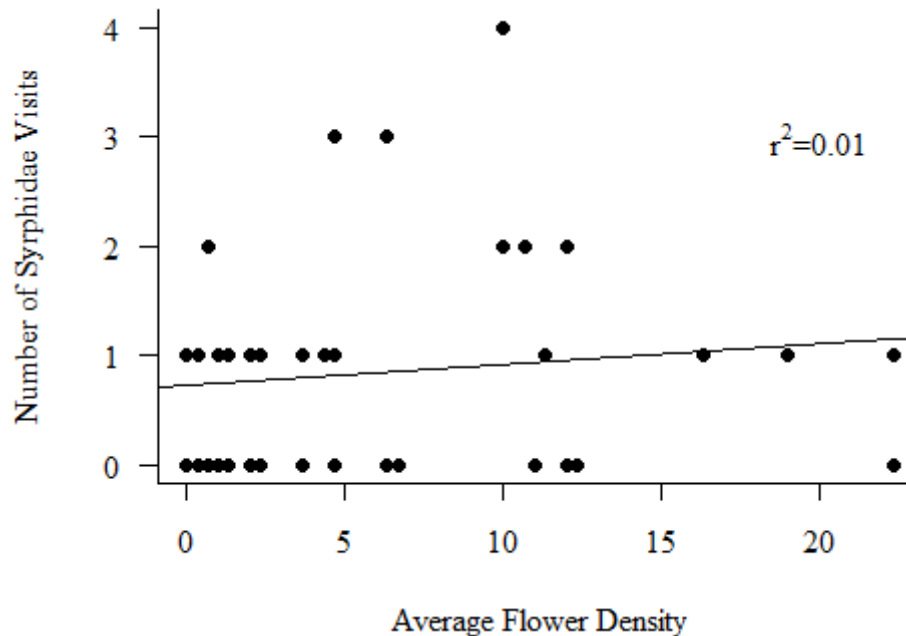


Figure 2-20. Simple linear regression correlation of the number of Syrphidae visits to cover crop mixtures as a function of the average flower density of each treatment plot and transect.  $F_{1,46}=0.6994$ ,  $p=0.41$ .

## DISCUSSION

Because many growers already plant cover crops to enhance a variety of ecosystem services, it is logical that capitalizing on a previously established farmer practice could help to increase adoption of flowering cover crops for pollinator conservation. In this scenario, farmers need only to adjust the species of cover crops used to include those that produce flowers that are visited by beneficial insects. This can be done without negatively affecting the other field-level benefits achieved by planting cover crops. In addition, as cover cropping is beneficial for a variety of ecosystem services, a farmer may choose to plant a diverse mixture of cover crop species on a field to maximize within-field benefits. While increased flower diversity has been shown to positively impact pollinator communities, this relationship is also associated with flower density and not total plant diversity directly (Potts et al. 2003, 2009, Holzschuh et al. 2006).

In order to consider the use of flowering cover crops as a within-field conservation strategy for native bee communities, it is important to understand how cover crop species grow and produce floral resources within real life rotation windows and field growing conditions. This study focused on the effect of diverse cover crop mixtures on floral resource quantity and subsequent bee and Syrphidae fly visitation. The crop rotation window that was used, a common rotation for central Pennsylvania feed grain rotations, was between harvest of winter wheat and planting of corn in a corn, soybean, and wheat rotation schedule. Because of the restrictive nature of the timing of corn planting in the spring, cover crops were terminated and incorporated into the soil in mid-May. While this time period was sufficient to achieve many of the standard, field-level cover crop benefits such as retention and provision of soil nutrients, weed suppression, or control of winter erosion, the rotation created a limited timeframe for cover crop flower production and their subsequent bee visitors to be present. Out of the six cover crop species in the experiment, three (red clover, Austrian winter pea, and canola) were considered potential spring pollinator-friendly species as they are all winter-hardy and produce insect-attractive flowers. However, the only cover crop to flower in any of the studied treatments prior to termination was canola. Austrian winter pea and red clover, although present in both the four and six species mixtures, were terminated before flowering and thus provided no floral resource benefit to the pollinator community within this rotation window.

As canola crops did produce a dense array of blooms, and because we observed a diversity of insect visitors to their flowers, canola can be considered a successful cover crop

species choice for providing essential early-season resources for beneficial insects in this organic field crop rotation window. For this reason and because canola flowers are generalist accessible due to a combination of open petals, short corolla, and readily available pollen and nectar, canola was considered an adequate study system for this experiment which is focused on the effect of cover crop plant diversity on native bee visitation (Viik et al. 2012). Indeed, as a mass-flowering oilseed crop, canola has been shown to be visited by, and attractive to, a wide diversity of pollinators including managed honey bees, native bees, and flies of the Syrphidae family (Free and Nuttall 1968, Jauker and Wolters 2008, Mänd et al. 2010, Viik et al. 2012, Woodcock et al. 2013). Canola plants and their flower resources have also been shown to be effected by intraspecific crop density (Cresswell 2001). While its popularity as a cover crop species is still growing, this bright yellow, dense flowering plant has great potential as an early-season pollinator resource, especially in otherwise fragmented, dairy forage crop dominated landscapes such as central Pennsylvania (USDA 2007).

It is important also to consider variations in weather patterns from year to year and their consequences on winter survival, plant growth, flowering, and insect use. For example, based on preliminary data collected on cover crop bloom and bee visitation in the spring of 2012, canola bloomed as early as the first week of April in central Pennsylvania. In contrast, the first blooms of 2013 did not appear until the fourth week of April. These few weeks of difference would have likely been significant in terms of total quantity of canola bloom in the environment prior to termination, assuming that corn would have been planted on the same date both years. Indeed, other studies have shown great differences between canola oilseed crop production between study years, which were attributed mostly to variations in weather across multiple growing seasons (e.g., Lutman et al. 2000).

Weather variations do not only affect plant growth, however, they also influence the timing of insect emergence and foraging activity. During the flowering period of canola within this study, bee visitation was observed on canola flowers on two days. Favorable weather conditions for pollinator foraging were present on the first date, and many species and individuals were observed in the treatment plots, although the crop was not yet in full bloom. However, the second observation date corresponded to an increase in canola bloom density, but experienced record low temperatures. Because of this great difference in conditions between dates on which observations were made, very few insects were observed foraging on the second date, and those that were observed were primarily bumble bees and other larger bodied insects that are more adapted to foraging in colder temperatures than smaller bodied insects (Heinrich 1979, Vicens



and Bosch 2000). We determined that this limited dataset was not indicative of the pollinator community that would typically be foraging on the canola at this time and the insect visitation data was excluded from analysis. In general, suitable weather conditions for pollinator activity are considered to be low wind, no rain, dry vegetation, and above 15C (Westphal et al. 2008). Differences in springtime temperature or rainfall, therefore, have the potential to reduce total bee use of a cover crop resource if foraging is limited by ambient weather conditions. Because cover crop bloom in this rotation will always be subject to fluctuations in spring weather, it is possible that cooler or wetter spring seasons may not provide the same potential resource use as other warmer, dryer years.

This rotation window, however confined in scope due to limits in flower blooming time and weather conditions, did support the goal of providing early-season resources to the springtime native pollinator community. In fact, only ten of the twenty-three bee species collected in all trap types during this experiment were not found within any of the canola treatment plots (Table 2-4). The first native bees were collected in the passive landscape-level traps on 22 April 2013, which corresponded closely to the date of first bloom of the canola cover crop. In total, nine species of bee were collected during the first week of bloom. This observation provides evidence of the co-occurrence of active bee communities in the environment and the early cover crop resource.

In addition to the occurrence of appropriate floral resources in the environment during this early-season time period, this study also focused on the effect of cover crop plant diversity on floral resources and pollinator visitation. We observed a significant decrease in flower density per quarter-meter-squared in the two mixture treatments compared to the monoculture treatment (Figure 2-13). This corresponded with an equally significant decrease in canola plant density (Figure 2-11), but a different pattern in the number of canola flowers per plant (Figure 2-16). Canola flower density decreased linearly with plant density, but the canola grown in monoculture had significantly more flowers per plant than in either the four or six species mixtures. Other studies examining the influence of competition by weedy plant species with canola oilseed crops found a decrease in yield with increased weed pressure (Bijanazadeh et al. 2010). We suggest that competition with other cover crops in the mixture treatments had a negative influence on canola plant growth, as evidenced by the decrease in per-plant flower production observed in this experiment.

Corresponding with differences in canola flower density across mixtures treatments, we also observed a decrease in bee visitation and bee species richness with an increase in cover crop plant diversity (Figures 2-5, 2-7) as well as significant, positive correlations between average

flower density and each bee response (Figures 2-18, 2-19). We conclude that floral resource density was the driving factor in the differential response of bee community use across treatments corresponding with a dilution of canola blooms per unit area with increased plant diversity. This conclusion is supported by studies of pollination services in canola fields (Viik et al. 2012). While the authors found differences in bee visitation related to variations in pollen and nectar resources across canola fertilizer treatments, they concluded that total flower density was the main factor influencing the occurrence of bee visitation. Because we did not monitor pollen or nectar resources in this study, we cannot discuss whether this may also have contributed to the differences observed across mixture treatments. However, as fertilization and other management procedures were consistent across all treatments, we suggest that flower density was the most likely cause of differences observed in bee visitation to the cover crop mixtures.

Beyond canola-only pollinator experiments, similar results regarding the positive relationship between flower density and native bee visitation have been shown for other agricultural conservation and pollination studies (Tuell et al. 2008, Potts et al. 2009). Potts *et al.* (2009) showed, specifically, that an increase in total plant diversity (including many non-flowering plant and grass species) did not positively influence bumble bee abundance, but mixtures with an increased flowering plant density did. Considering that the cover crop mixtures studied in this experiment contained other grass or non-flowering species, this further supports the conclusion of flower density, and not plant diversity directly, driving the observed levels of bee visitation.

Syrphidae visitation, however, was neither different across mixture treatments nor correlated to average flower density (Figures 2-9, 2-20). Although syrphid flies have been shown to be effective pollinators of canola crops (Jauker and Wolters 2008), they do not appear to be resource driven in terms of foraging preference as are bee pollinators.

We found significantly higher levels of bee visitation on center plot transects than on plot edges (Figure 2-6). This trend, however, was not evident for bee species richness or Syrphidae visitation abundance (Figures 2-8, 2-10). This suggests a behavioral influence on bee visitation preference for interior patches of canola flowers, although there were no observed differences in flower density across plot transect locations (Figure 2-14).

Understanding the combination of factors which are influenced by plant diversity and floral density and how they affect bee visitation within these resource patches is particularly important for informing the adoption of flowering cover crops in agricultural landscapes for pollinator conservation. While plant diversity may be beneficial in general for pollinator

communities, considering the confined timeframe of the cover crop rotation window being studied in this experiment, it is important to recognize that with an increase in plant diversity, we observed a decrease or dilution in canola blooms per square meter, and thus a decrease in floral resource availability. We do not conclude, however, that cover crop mixtures are not beneficial for conservation purposes, especially if the ultimate goal of cover cropping is to maximize total ecosystem service benefits to both field-level crop productivity as well as ecosystem health and conservation needs. Rather, we are advocating for informed considerations of how agronomic management choices influence the complete cover cropping system and its multifunctional services.

## LITERATURE CITED

- Bates, D., M. Maechler, B. Bolker, and S. Walker. 2013. lme4: Linear Mixed-Effects Models Using Eigen and S4. <http://cran.r-project/package=lme4>.
- Bijanzadeh, E., R. Naderi, and A. Behpoori. 2010. Interrelationships between oilseed rape yield and weeds population under herbicides application. *Australian Journal of Crop Science* 4:155–162.
- Bouseman, J. K., and W. E. LaBerge. 1979. A Revision of the Bees of the Genus *Andrena* of the Western Hemisphere. Part IX. Subgenus *Melandrena*. *Transactions of the American Entomological Society* 104:275–389.
- Clark, A. (Ed.). 2007. *Managing Cover Crops Profitably* 3rd ed. Sustainable Agriculture Research and Education (SARE).
- Cresswell, J. 2001. Attributes of Individual Flowers of *Brassica napus* L. are Affected by Defoliation but not by Intraspecific Competition. *Annals of Botany* 88:111–117.
- Discover Life. (n.d.). . [www.discoverlife.org](http://www.discoverlife.org).
- Elliott, S. E. 2009. Subalpine Bumble Bee Foraging Distances and Densities in Relation to Flower Availability. *Environmental Entomology* 38:748–756.
- Free, J. B., and P. M. Nuttall. 1968. The pollination of oilseed rape (*Brassica napus*) and the behaviour of bees on the crop. *The Journal of Agricultural Science* 71:91–94.
- Gibbs, J. 2011. Revision of the metallic *Lasioglossum* (*Dialictus*) of eastern North America (Hymenoptera: Halictidae: Halictini). *Zootaxa* 3073:1–216.
- Gibbs, J., L. Packer, S. Dumes, and B. N. Danforth. 2013. Revision and reclassification of *Lasioglossum* (*Evylaeus*), L. (*Hemihalictus*) and L. (*Sphecodogastra*) in eastern North America (Hymenoptera: Apoidea: Halictidae). *Zootaxa* 3672:001–117.
- Goulson, D., G. C. Lye, and B. Darvill. 2008. Decline and conservation of bumble bees. *Annual Review of Entomology* 53:191–208.
- Greenleaf, S. S., and C. Kremen. 2006a. Wild bees enhance honey bees' pollination of hybrid sunflower. *Proceedings of the National Academy of Sciences of the United States of America* 103:13890–5.
- Greenleaf, S. S., and C. Kremen. 2006b. Wild bee species increase tomato production and respond differently to surrounding land use in Northern California. *Biological Conservation* 133:81–87.
- Heinrich, B. 1979. *Bumblebee Economics*. Harvard University Press, Cambridge, MA.

- Hendrickx, F., J.-P. Maelfait, W. Van Wingerden, O. Schweiger, M. Speelmans, S. Aviron, I. Augenstein, R. Billeter, D. Bailey, R. Bukacek, F. Burel, T. Diekötter, J. Dirksen, F. Herzog, J. Liira, M. Roubalova, V. Vandomme, and R. Bugter. 2007. How landscape structure, land-use intensity and habitat diversity affect components of total arthropod diversity in agricultural landscapes. *Journal of Applied Ecology* 44:340–351.
- Hodgson, J. A., W. E. Kunin, C. D. Thomas, T. G. Benton, and D. Gabriel. 2010. Comparing organic farming and land sparing: optimizing yield and butterfly populations at a landscape scale. *Ecology letters* 13:1358–67.
- Hole, D. G., A. J. Perkins, J. D. Wilson, I. H. Alexander, P. V. Grice, and A. D. Evans. 2005. Does organic farming benefit biodiversity? *Biological Conservation* 122:113–130.
- Holzschuh, A., I. Steffan-Dewenter, D. Kleijn, and T. Tscharntke. 2006. Diversity of flower-visiting bees in cereal fields: effects of farming system, landscape composition and regional context. *Journal of Applied Ecology* 44:41–49.
- Hothorn, T., F. Bretz, and P. Westfall. 2008. Simultaneous Inference in General Parametric Models. *Biometrical Journal* 50:346–363.
- Jacobs, J. 2011. Individual Based Rarefaction Using R-package. <http://www.jennajacobs.org/R/rarefaction.html>.
- Jauker, F., and V. Wolters. 2008. Hover flies are efficient pollinators of oilseed rape. *Oecologia* 156:819–23.
- Klein, A.-M., B. E. Vaissière, J. H. Cane, I. Steffan-Dewenter, S. A. Cunningham, C. Kremen, and T. Tscharntke. 2007. Importance of pollinators in changing landscapes for world crops. *Proceedings of the Royal Society B: Biological Sciences* 274:303–13.
- Kremen, C., and A. Miles. 2012. Ecosystem Services in Biologically Diversified versus Conventional Farming Systems : Benefits , Externalities , and Trade-Offs. *Ecology and Society* 17(4):40.
- Kremen, C., N. M. Williams, and R. W. Thorp. 2002. Crop pollination from native bees at risk from agricultural intensification. *Proceedings of the National Academy of Sciences of the United States of America* 99:16812–6.
- Lal, R., E. Regnier, D. J. Eckert, W. M. Edwards, and R. Hammond. 1991. Expectations of cover crops for sustainable agriculture. Pages 1–11 in W. L. Hargrove, editor. *Cover Crops for Clean Water*. Soil and Water Conservation Society.
- LeBuhn, G., S. Droege, N. Williams, B. Minckley, T. Griswold, C. Kremen, O. Messinger, J. Cane, T. Roulston, F. Parker, V. Tepedino, and S. Buchmann. 2002. The Bee Inventory Plot. [online.sfsu.edu/beeplot/](http://online.sfsu.edu/beeplot/).

- Leong, J. M., and R. W. Thorp. 1999. Colour-coded sampling: the pan trap colour preferences of oligolectic and nonoligolectic bees associated with a vernal pool plant. *Ecological Entomology* 24:329–335.
- Lutman, P. J. W., P. Bowerman, G. M. Palmer, and G. P. Whytock. 2000. Prediction of competition between oilseed rape and *Stellaria media*. *Weed Research* 40:255–269.
- Mänd, M., I. H. Williams, E. Viik, and R. Karise. 2010. Oilseed Rape, Bees and Integrated Pest Management. Pages 357–379 in I. H. Williams, editor. *Biocontrol-Based Integrated Management of Oilseed Rape Pests*. Springer Netherlands, Dordrecht.
- Mandelik, Y., R. Winfree, T. Neeson, and C. Kremen. 2012. Complementary habitat use by wild bees in agro-natural landscapes. *Ecological applications : a publication of the Ecological Society of America* 22:1535–46.
- Matson, P. A., W. J. Parton, A. G. Power, and M. J. Swift. 1997. Agricultural Intensification and Ecosystem Properties. *Science* 277:504–509.
- McGinley, R. J. 1986. Studies of Halictinae (Apoidea: Halictidae), I: Revision of New World *Lasioglossum* Curtis. *Smithsonian Contributions to Zoology* 429.
- Morandin, L. A., and M. L. Winston. 2005. Wild bee abundance and seed production in conventional, organic, and genetically modified canola. *Ecological Applications* 15:871–881.
- National Research Council. 2007. *Status of Pollinators in North America*. The National Academies Press, Washington, DC.
- Oksanen, J., F. G. Blanchet, R. Kindt, P. Legendre, P. R. Minchin, R. B. O'Hara, G. L. Simpson, P. M. Solymos, H. H. Stevens, and H. Wagner. 2013. *vegan: Community Ecology Package*. <http://cran.r-project.org/package=vegan>.
- Ollerton, J., R. Winfree, and S. Tarrant. 2011. How many flowering plants are pollinated by animals? *Oikos* 120:321–326.
- Potts, S. G., J. C. Biesmeijer, C. Kremen, P. Neumann, O. Schweiger, and W. E. Kunin. 2010. Global pollinator declines: trends, impacts and drivers. *Trends in Ecology & Evolution* 25:345–53.
- Potts, S. G., B. Vulliamy, A. Dafni, G. Ne'Eman, and P. Willmer. 2003. Linking Bees and Flowers : How Do Floral Communities Structure Pollinator Communities? *Ecology* 84:2628–2642.
- Potts, S. G., B. A. Woodcock, S. P. M. Roberts, T. Tscheulin, E. S. Pilgrim, V. K. Brown, and J. R. Tallowin. 2009. Enhancing pollinator biodiversity in intensive grasslands. *Journal of Applied Ecology* 46:369–379.

- R Core Team. 2012. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. <http://www.r-project.org/>.
- Ricketts, T. H. 2004. Tropical Forest Fragments Enhance Pollinator Activity in Nearby Coffee Crops. *Conservation Biology* 18:1262–1271.
- Sala, O. E., F. S. Chapin, J. J. Armesto, E. Berlow, J. Bloomfield, R. Dirzo, E. Huber-Sanwald, L. F. Huenneke, R. B. Jackson, A. Kinzig, R. Leemans, D. M. Lodge, H. A. Mooney, M. Oesterheld, N. L. Poff, M. T. Sykes, B. H. Walker, M. Walker, and D. H. Wall. 2000. Global biodiversity scenarios for the year 2100. *Science* 287:1770–4.
- Snapp, S. S., S. M. Swinton, R. Labarta, D. Mutch, J. R. Black, R. Leep, J. Nyiraneza, and K. O’Neil. 2005. Evaluating Cover Crops for Benefits, Costs and Performance withing Cropping System Niches. *Agronomy Journal* 97:322–332.
- Tilman, D., J. Fargione, B. Wolff, C. D’Antonio, a Dobson, R. Howarth, D. Schindler, W. H. Schlesinger, D. Simberloff, and D. Swackhamer. 2001. Forecasting agriculturally driven global environmental change. *Science* 292:281–4.
- Tuell, J. K., A. K. Fiedler, D. Landis, and R. Isaacs. 2008. Visitation by Wild and Managed Bees ( Hymenoptera : Apoidea ) to Eastern U . S . Native Plants for Use in Conservation Programs. *Environmental Entomology* 37:707–718.
- USDA. 2007. Census of Agriculture County Profile: Centre County, Pennsylvania. [www.agcensus.usda.gov](http://www.agcensus.usda.gov).
- USDA. 2013. National Organic Program: Organic Standards. <http://www.ams.usda.gov/AMSV1.0/ams.fetchTemplateData.do?template=TemplateN&rightNav1=NOSBlinkNOSBMeetings&topNav=&leftNav=&page=NOPOrganicStandards&resultType=>.
- VanEngelsdorp, D., J. D. Evans, C. Saegerman, C. Mullin, E. Haubruge, B. K. Nguyen, M. Frazier, J. Frazier, D. Cox-Foster, Y. Chen, R. Underwood, D. R. Tarpy, and J. S. Pettis. 2009. Colony collapse disorder: a descriptive study. *PloS one* 4:e6481.
- Vasseur, C., A. Joannon, S. Aviron, F. Burel, J.-M. Meynard, and J. Baudry. 2013. The cropping systems mosaic: How does the hidden heterogeneity of agricultural landscapes drive arthropod populations? *Agriculture, Ecosystems & Environment* 166:3–14.
- Vicens, N., and J. Bosch. 2000. Weather-Dependent Pollinator Activity in an Apple Orchard , with Special Reference to *Osmia cornuta* and *Apis mellifera* ( Hymenoptera : Megachilidae and Apidae ). *Environmental Entomology* 29:413–420.
- Viik, E., M. Mänd, R. Karise, P. Lääniste, and I. H. Williams. 2012. The impact of foliar fertilization on the number of bees ( Apoidea ) on spring oilseed rape. *Zemdirbyste=Agriculture* 99:41–46.

- Westphal, C., R. Bommarco, G. Carre, E. Lamborn, N. Morison, T. Petanidou, S. G. Potts, S. P. M. Roberts, H. Szentgyörgyi, T. Tscheulin, B. E. Vaissière, M. Woyciechowski, J. Biesmeijer, W. E. Kunin, J. Settele, and I. Steffan-Dewenter. 2008. Measuring Bee Diversity in Different European Habitats and Biogeographical Regions. *Ecological Monographs* 78:653–671.
- Williams, G. R., D. R. Tarpy, D. vanEngelsdorp, M.-P. Chauzat, D. L. Cox-Foster, K. S. Delaplane, P. Neumann, J. S. Pettis, R. E. L. Rogers, and D. Shutler. 2010. Colony Collapse Disorder in context. *BioEssays : news and reviews in molecular, cellular and developmental biology* 32:845–6.
- Williams, N. M., J. Regetz, and C. Kremen. 2012. Landscape-scale resources promote colony growth but not reproductive performance of bumble bees. *Ecology* 93:1049–58.
- Winfree, R. 2010. The conservation and restoration of wild bees. *Annals of the New York Academy of Sciences* 1195:169–97.
- Winfree, R., R. Aguilar, D. P. Vazquez, G. LeBuhn, and M. A. Aizen. 2009. A meta-analysis of bees' responses to anthropogenic disturbance. *Ecology* 90:2068–2076.
- Winfree, R., T. Griswold, and C. Kremen. 2007. Effect of human disturbance on bee communities in a forested ecosystem. *Conservation biology : the journal of the Society for Conservation Biology* 21:213–23.
- Woodcock, B. A., M. Edwards, J. Redhead, W. R. Meek, P. Nuttall, S. Falk, M. Nowakowski, and R. F. Pywell. 2013. Crop flower visitation by honeybees, bumblebees and solitary bees: Behavioural differences and diversity responses to landscape. *Agriculture, Ecosystems & Environment* 171:1–8.



## Chapter 3

# FALL PLANTING DATE AS IT AFFECTS COVER CROP WINTER SURVIVAL, BLOOM PERIOD, AND NATIVE BEE VISITATION

## INTRODUCTION

Pollination as an ecosystem service is vital to the reproduction of much of the world's food crops and other flowering plants. In fact, animal-mediated pollination (primarily by bees) is required for 35% of the world's total food production and 87.5% of all flowering plants (Klein et al. 2007, Ollerton et al. 2011). While managed honey bee colonies are most often used for agricultural pollination, native bees are also known to play an important role in crop pollination (Kremen et al. 2002, Ricketts 2004, Morandin and Winston 2005, Greenleaf and Kremen 2006a, 2006b). However, despite the fact that pollination services are often necessary in agricultural production, the importance of native bee communities extends far beyond this purpose.

Unfortunately, the world's pollinators are in decline. While the recent decrease in managed honey bee colonies is now well-documented (VanEngelsdorp et al. 2009, Williams et al. 2010), there is also evidence for a global decline in other pollinator groups as well as many pollinator-dependent plants (National Research Council 2007, Potts et al. 2010). The possible causes of this decline include loss of natural habitat, agricultural pesticides, pathogens, disease, and climate change (Potts et al. 2010). However, it is most likely the combined interaction of these factors that has led to much of the widespread global pollinator decline that we see today. Habitat loss and fragmentation, in particular, are often listed as some of the greatest and most common threats to wild pollinators, particularly bees (Kremen et al. 2002, Ricketts 2004, Goulson et al. 2008, Winfree et al. 2009).

Historically, agriculture is often associated with negative influences on biodiversity and increased land simplification (Matson et al. 1997, Tilman et al. 2001). This is because the transition of landscapes from their natural, undisturbed status to cultivated and managed systems typically involves the destruction of previously-established wildlife resources and natural habitats, including those of beneficial and pollinating insects. Indeed, bees are the insect group

shown to be the most negatively affected by agricultural intensification (Hendrickx et al. 2007). Additionally, because total land-use change has been predicted to have the greatest effect on global biodiversity of terrestrial ecosystems over the next 100 years (Sala et al. 2000), determining alternative scenarios that limit the effects of habitat change on native pollinators is a significant consideration for the pollinator research and conservation community. However, because we cannot directly manage most of our native bees, conservation strategies should be focused on preserving the natural habitat and resources that are required for their survival.

One strategy for increasing agricultural conservation and ecosystem health is a trend toward organic or diversified farming. Compared to conventional farming, organic agriculture can increase biodiversity and ecosystem services (Hole et al. 2005, Kremen and Miles 2012) and support a greater diversity of native bees (Holzschuh et al. 2006). In the United States, organic agriculture certification, while not exclusive to diversified farming systems, requires more than the elimination of synthetic chemical pesticides or fertilizers; it also is based on the idea of a holistic, whole-farm approach to agriculture (USDA 2013). For this, wildlife-friendly farming and conserving some aspect of habitat for native wildlife is one of the obligations of organic growers.

Additionally, agricultural landscapes can be deceptively complex in nature. For example, the dynamic temporal changes that occur over the course of the growing season often illustrate a type of hidden heterogeneity in agricultural systems. Such seasonal differences in resource availability or location can significantly affect arthropod populations (Vasseur et al. 2013). All in all, by focusing on an array of techniques that take into account a combination of practices including preservation of habitat refuges, wildlife-friendly farming, and the seasonal resource variations across the landscape, an optimal conservation strategy may be found for a wide range of farm types or locations (Hodgson et al. 2010). As a partial solution to this conservation need, we consider the incorporation of winter cover crops into organic farming systems to help enhance floral resources both spatially and temporally across the landscape.

Cover crops are plant species grown within a cultivated field during fallow periods in annual cash crop rotation schedules, or intermixed within cash crop plantings. They can be almost any species of plant, but are mostly commonly grasses and legumes, and can be planted almost anytime during the year depending on the crop rotation and local climate. Most farmers plant cover crops for within-field erosion control, soil fertility management, or weed suppression (Lal et al. 1991, Snapp et al. 2005, Clark 2007). However, because the addition of cover crops into an annual crop rotation potentially increases spatial and temporal plant diversity levels, it can

also act as an agricultural conservation strategy. Additionally, by selecting cover crops that also produce insect-visited flowers attractive to native pollinators, this technique can benefit crop productivity as well as supplement resources to native wildlife populations.

The addition of supplemental flowering resources to an agricultural landscape has been shown to be beneficial to native bee communities and is often used as a pollinator conservation strategy, although these are typically installed along field edges or in uncultivated farm areas (Tuell et al. 2008, Winfree 2010). Our study focuses on whether the addition of a spring-blooming cover crop species could achieve the same results. This timeframe is especially important because some native bee species benefit from an increase in springtime floral resources (Elliott 2009, Williams et al. 2012). However, agricultural landscapes are often lacking these early-season flowers compared with other natural or fallow areas (Winfree et al. 2007, Williams et al. 2012, Mandelik et al. 2012). It is for this reason that increasing spring flowering resources within cultivated fields would likely have a large influence on overall resource availability for native bees during this time of the year.

Incorporating flowering cover crops into a grower's rotation schedule is an agricultural conservation strategy with the opportunity to consider the needs of both the grower and the native pollinator community. However, to make appropriate recommendations to farmers interested in achieving these dual benefits, it is important to select the appropriate plants for a specific production system. Some of the factors that need to be considered in the selection of cover crops include: when will these species bloom, how will that bloom be influenced by fall planting or spring termination dates (crop rotation windows), and what bee species will visit each of these cover crop flowers? For example, one grower may be interested in a cover crop species that benefits the greatest diversity of pollinators, while another may want to focus on those bees that are needed to pollinate a summer cash crop. This project focuses on pursuing answers to some of these questions in order to better inform future decisions on the pollinator conservation potential of a variety of cover crop species and crop rotation schedules for central Pennsylvania.

The three commonly used, or locally appropriate, cover crop species that were chosen for study in this experiment were winter canola (*Brassica napus* 'Wichita' L.), Austrian winter pea (*Pisum sativum* subsp. *arvense* L.), and red clover (*Trifolium pratense* L.). Each of these species was selected because they produce insect-visited flowers in addition to providing other agronomic cover crop benefits, e.g., regulating soil nutrients or suppressing weed growth. These three species vary in floral morphology, color, and blooming time. Differences in flower morphology and physiology such as color, petal shape, pollen and nectar resources make each of these plants

more or less attractive or accessible to bee species depending on the evolutionary adaptations and specialization of the bee (O'Toole and Raw 1991, Potts et al. 2003). Because individual bee species respond differently to variations in flower resources and morphology, we expected the pollinator communities to vary across the cover crop species.

Additionally, because corresponding research on flowering cover crop mixtures and native bee visitation illustrated the limiting effect of field crop rotation schedules and restrictive cover cropping windows on cover crop flowering in central Pennsylvania, we choose to study these three cover crop species outside of a standard cash crop rotation. By creating a gradient of fall planting dates and unrestricted spring blooming opportunity, each of these cover crops and their associated pollinator communities could be studied and modeled across a variety of potential cash crop rotation windows. The goal was to observe what influences these changes in fall planting times would have on cover crop winter survival and subsequent springtime bloom as these both would influence prospective pollinator floral resource use.

Several studies have shown that planting date affects the flowering phenology of many agricultural crops, including canola for oilseed production (Major et al. 1975, Alessi et al. 1977, Teasdale et al. 2004, Adamsen and Coffelt 2005). Differences in day length, temperature, and accumulated growing degree-days are attributed as the major influences of crop growth across planting date gradients. As pollinator community structure is also influenced by seasonal phenology, we expected differences in cover crop blooming time to influence bee visitation abundance and diversity on each cover crop and planting date treatment (Bartomeus et al. 2011, Kimoto et al. 2012).

Accordingly, we examined the effects of fall planting date on bloom onset, duration and intensity for three overwintering cover crop species; winter canola, Austrian winter pea, and red clover. We also surveyed the native bee community of each cover crop, focusing on the total abundance and diversity of bees that visited each cover crop during their complete spring blooming period.

## MATERIALS AND METHODS

### Site Description

The experiments were conducted at a single site established on approximately 11 hectares of land at the Penn State University Russell E. Larson Research and Education Center (RELREC) near Rock Springs, Pennsylvania. The dominant soil type at the site is Hagerstown silt loam with soil texture being predominantly clay loam with variability in silt, clay and sand. This land is in transition to organic certification and was managed in accordance with the USDA National Organic Standards (USDA 2013).

A geographic information systems (GIS) analysis of the site and surrounding habitat types were performed using base map satellite imagery, hand-digitized land cover classifications, and ground-truthing (ArcMap version 10.1, 2012 ESRI Inc.). A buffer zone of 250 m from the edge of the total experiment site was chosen to accommodate the foraging range of many of the smaller, ground nesting bees collected during the study. All cropland within this boundary is part of the RELERC and was separated into tilled, arable crop land and non-tilled farm land which includes areas such as grassy drive rows and field edges (Table 3-1, Figures 3-1, 3-2). The primary crops grown in this area are field grains such as corn, soybeans, and winter wheat, although some small patches of spring canola for oilseed production are also grown. Additionally, some neighboring experiments grow cover crops in their rotation including some flowering species such as hairy vetch and red clover.

Table 3-1. Land cover classifications for a 250 meter buffer zone surrounding all edges of the main research site. All values are given in percentage of the total area

<b>Land Cover Classification</b>	<b>250 m</b>
Arable cropland	67.2
Non-tilled cropland (grass alleyways, roadsides, other less-disturbed areas)	16.7
Semi-natural habitat (tree lines, un-managed grassland)	0.4
Residential	1.1
Managed lawn	6.2
Industrial surface (roads, farm buildings)	8.4

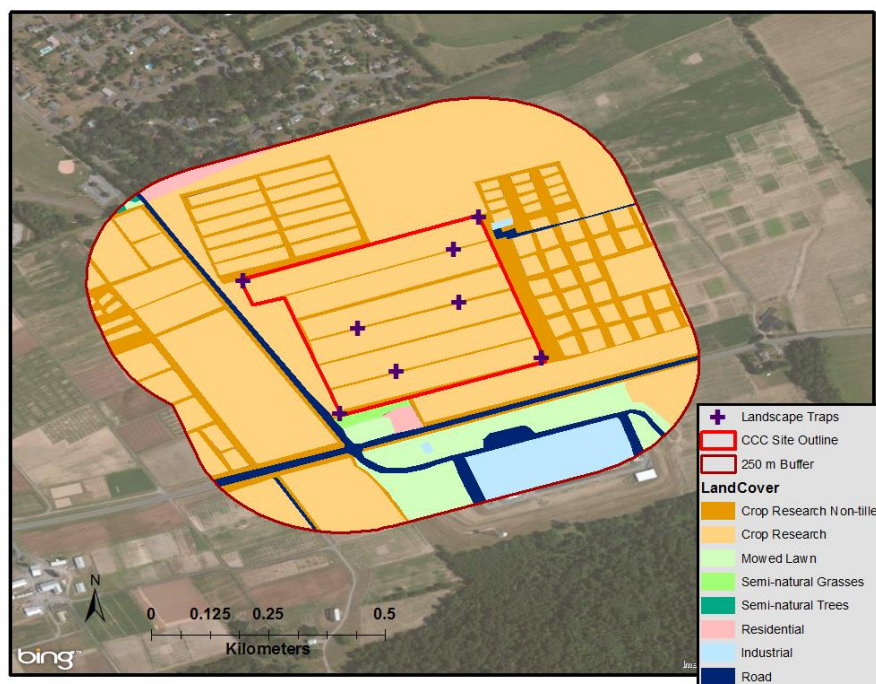


Figure 3-1. Land cover classification map showing the allocation of various cover classes of the landscape in a 250 m buffer zone surrounding the research site (outlined in red). Also shown is the placement of the eight landscape-level trap locations (purple +).



Figure 3-2. Aerial photograph showing the project site (outlined in red) and surrounding landscape.

## Planting and plot establishment

To determine the effect of fall planting date on spring cover crop flowering time, flower density, and native bee visitation, an experiment was established on 0.25 hectares of land at the Penn State University Russell E. Larson Research and Education Center (RELREC) near Rock Springs, Pennsylvania. All data was collected from fall of 2012 through July 2013. Three species of cover crop, canola (*Brassica napus* 'Wichita' L.), medium red clover (*Trifolium pratense* L.), and Austrian winter pea (*Pisum sativum* subsp. *arvense* L.) were each planted in on four dates during the fall of 2012, each three weeks apart. The experiment utilized a split-block or strip-block design with crop and planting date as the main effects. Each main plot was approximately 9 m by 11 m in size, with crop type as a main plot, and planting date subplots of approximately 2 meters by 11 meters stripped within the main plot (Appendix A). Each treatment was replicated four times with a total of 12 main plots (cover crop type) and 48 subplots (planting date by cover crop type). The four planting dates used were 2 August, 24 August, 13 September, and 5 October, 2012. These variations in fall planting date are representative of a range of possible cover crop planting dates for use in common agronomic cash crop rotations of the Mid-Atlantic region.

Seeds were weighed and measured to provide a seeding rate that was representative of common farmer practices for cover crop monocultures of each species (Clark 2007) . Canola was planted at a rate of 12.7 kg/ha (11.3 lb/ac), Austrian winter pea at 87.3 kg/ha (78 lb/ac), and medium red clover at 13.4 kg/ha (12 lb/ac). Planting was completed using a no-till cone plot drill with double disc openers (Great Plains 3P605NT) which planted nine rows of seed, each 19 cm apart. Planting depth was varied by crop with canola and red clover at 1 cm depth and the Austrian winter pea at 2 cm depth. Plots were managed without irrigation and in accordance to organic growing standards with no synthetic chemical herbicides or insecticides applied (USDA 2013). Some manual weed suppression techniques were implemented in the fall and early spring to limit annual and perennial weed growth. However, once cover crops achieved a growth stage where competition from weeds was unlikely to interfere with crop growth or bloom, most weed control efforts were stopped.

## Fall Biomass

Fall biomass samples were collected from each treatment to quantify the effects of planting date on winter survival, spring bloom and plant density. All 48 subplots were sampled between November 8, 9, and 12, 2012, approximately five weeks after the fourth planting date and as late as possible before the first hard winter freeze. Samples were taken in a 0.25m<sup>2</sup> quadrat randomly located within each cover crop by planting date subplot. Each quadrat spanned three crop rows. Living, aboveground cover crop mass was cut at soil level and dried in an oven at 55 C for 1-2 weeks until completely dry. Weed biomass was not sampled. Because of variations in crop emergence, small size of each plot, and the destructive nature of sampling, only one 0.25m<sup>2</sup> area was sampled per subplot.

## Observations and Netting

To quantify visitation to the blooming crop, visual observations of pollinators were completed on a weekly basis through the complete blooming cycle in all treatment plots, or through the first week of July, whichever came first. Red clover, which did not initiate bloom until mid-June, was only monitored until early July, the date that was determined to be beyond the scope of most overwintering cover crop rotation windows in the Mid-Atlantic region (Clark 2007). Each subplot was monitored for bee floral visitation for two minutes twice per day; once in the morning and once in the early afternoon. The observer walked at a slow and even rate along the edge of each plot to cover as much of the perimeter as possible during the two minute period (modified from Westphal et al. 2008). Each bee or Syrphidae fly that was observed landing on the reproductive parts of the flower was recorded to the lowest taxonomic level possible from visual estimations. Groups that were easy to determine on-the-wing were identified to genus (e.g., *Bombus*, *Apis*, *Xylocopa*) whereas those that were smaller or more difficult to identify in motion were grouped into morphospecies categories (e.g., large dark bee, green bee, small dark bee, syrphid fly).

After each morning and afternoon observation period each subplot was monitored for an additional 60 sec using an active collection technique. During these observations, every bee or Syrphidae fly that was observed landing on the reproductive parts of the cover crop flowers was



collected with an aerial insect net, killed by placing them in a glass collecting jar with an ethyl acetate-soaked plaster bottom and returned to the laboratory for identification to species. Netted specimens acted as a reference to those grouped to morphospecies during the preceding observation period. Bees that were obviously of the same species (e.g., *Xylocopa virginica* L.) and were collected already once during the netting period in the subplot being observed were not collected in duplicate even if observed on the flowers of interest. *Apis mellifera* (L.) specimens were not collected often during netting periods as species identification was confident during the visual observations.

Bee species were identified using a series of online or hard-copy taxonomic keys, as appropriate by genus or subgenus (“Discover Life” n.d., Bouseman and LaBerge 1979, McGinley 1986, Gibbs 2011, Gibbs et al. 2013). Some of the more difficult species of the genus *Andrena* or *Lasioglossum* were identified by Jason Gibbs of Michigan State University. Previously identified species were also confirmed by J. Gibbs. Voucher specimens have been submitted to the Frost Museum at the Pennsylvania State University.

Weather data including air temperature, 30 sec average wind speed, and sky condition were collected three times per observation period; once at the beginning of visual observations, after observations for all subplots were completed, and after netting all plots. Weather data was collected using a Kestrel 2000™ thermo-anemometer ([www.kestrelmeters.com](http://www.kestrelmeters.com)). This was repeated for both the morning and afternoon observation sessions.

### **Landscape area traps (passive collection)**

To relate the community of bees that were collected from the flowering cover crops to the general bee community in the landscape during the same time period, we placed two types of passive traps, pan and plastic vane traps, across the site on a weekly basis from April 22, 2013 until the completion of the study on July 3, 2013. Traps were deployed for 48 hr with collected specimens removed from the traps every 24 hr. Traps were placed in groups comprised of three pan traps (1 each white, yellow, blue) and two vane traps (one each blue and yellow). In total, 8 groups of pan and vane traps were deployed across the 11 ha study area. Four of the trap groups were set on the edge of the field site, one at each corner (edge traps), and four were placed in the interior of the project boundaries (interior traps) (see plot map in Appendix A). All traps were located along grass access roads surrounding the study plots, and were as evenly distributed

across the study area as was possible given road spacing constraints and other field operation concerns.

Methodology used for pan trapping was adapted from Westphal et al. (2008) and from *The Bee Inventory Plot* report (LeBuhn et al. 2002). The pan traps, also referred to as bee bowls, were constructed of 96 ml Dart brand soufflé cups spray painted in white (Krylon® Fusion for plastic), florescent yellow (Krylon®), or florescent blue (ACE® Glo Spray). All yellow and blue bowls were also painted with a primer of the white plastic-bonding paint. Three colors were used as different bee groups have been shown to be attracted to differently colored traps; most commonly white, blue and yellow (Leong and Thorp 1999). Bowls were mounted above the ground on 1.2 m tall, 2 cm diameter wooden dowels. Atop each dowel one painted bowl was attached using a single thumb tack. The sample bowl consisted of another bowl of the same color placed within the support bowl. The sample bowl was filled three-fourths full with soapy water. The soapy water mix was created using 2 L and approximately 1 ml of blue, original non-scented Dawn® dish soap. The soap reduced the water surface tension to increase insect catch without additional odors.

Plastic vane traps (SpringStar, Inc.) were constructed out of yellow and florescent blue perpendicular vanes and a collecting tub attached below the vanes. All vane traps were used in their unaltered form. Each trap was suspended from a 1.2m galvanized steel shepherd's hook purchased from a local garden supply store. Approximately 200 ml of soapy water mixture were added to the collection tub of each vane trap to act as an insect killing agent.

One trap of each type and color were set at each of the 8 site locations. The group of five different traps (pan and vane traps) were randomly ordered along a linear transect on the edge of the nearest plot and spaced approximately two meters from the nearest trap (Figure 3-3). All insects collected in the traps that were neither bee nor Syrphidae flies were considered bycatch and discarded. Summary data for landscape-level trap type and color efficiencies is given in Appendix B.



Figure 3-3. Landscape-level passive trapping example setup for one of the project boundary edges.

### Statistical Analysis

Analysis of bee visitation abundance and diversity was performed with the R statistical language (R Core Team 2012). Additional R packages used for linear mixed-effects models (Bates et al. 2013), Tukey's multiple comparison post-hoc test (Hothorn et al. 2008), and species richness rarefaction curves (Jacobs 2011, Oksanen et al. 2013). Generalized linear mixed-effect models with a Poisson distribution were used for all bee-based data analysis using random errors appropriate for blocked designs and with significantly correlated temperature or wind covariates as appropriate (Table 3-3). Plant-based data analysis was performed using linear mixed models considering the random errors necessary for blocked designs and repeated measures analysis over the time span of each cover crop mixture. Correlations between individual variables were performed using simple linear regression.

Syrphidae visits to individual cover crop planting dates per two minute observation period were excluded from analysis as few individual flies were collected from any particular planting date thus increasing the probability of type I statistical errors.

Because the response of individual taxa to a treatment may not be independent from the response of other taxa in a community, a multivariate direct gradient analysis was performed to explore the distribution of the measured pollinator community to response variables including cover crop species and average ambient temperature and wind speed measured during the same time period in relation to experimental treatments. Preliminary detrended correspondence analysis using all taxa indicated that community composition gradients were short, therefore, a

linear gradient procedure, redundancy analysis (RDA), was performed with 'CANOCO' for Windows version 4.5 (Ter Braak and Smilauer 2002, Leps and Smilauer 2003, Ter Braak 2003). Experimental factors (cover crop species) were treated as nominal (0,1) environmental (explanatory) variables, and block was treated as a nominal covariable. Explanatory environmental factors (wind speed, temperature) were treated as continuous variables. Numbers of pollinators were summed across sample dates in each treatment, and were used to calculate mean number of pollinator species or species groups per treatment in each block. Pollinators comprising over 1% of the population were included in the RDA. A Monte Carlo permutation option was employed to determine the significance of the first axis and of all canonical axes combined, constrained by block. RDA results are displayed graphically with bi-plot scaling focused on centered and standardized inter-taxon distances, where points depict nominal variables and response variables, including pollinator taxa, with a fit to the model of at least 1%, are represented as vectors.

## RESULTS

A total of 61 bee species were collected between the landscape-level passive traps and the crop-level netting throughout the course of the experiment. The greatest number of bee species was collected in canola, followed by Austrian winter pea and red clover respectively (Table 3-2). In contrast, there were 49 bee species collected in the passive landscape-level traps, 12 of which were unique to the traps and not collected on any of the three cover crops. No single crop or trapping method collected all species observed during this project. The lack of asymptote on the species richness rarefaction curve for canola, and to some degree Austrian winter pea, suggest that the full bee community of these cover crops may not be fully represented in the samples collected (Figure 3-4).

Table 3-2. Species table of all bees collected via hand netting from cover crop flowers or via the landscape-level passive traps across all weeks of the experiment. Species with an asterisk were collected with greater than ten individuals.

	Canola	Austrian Winter Pea	Red Clover	Landscape Traps
<i>Agapostemon sericeus</i> (Forster, 1771)	X			X
<i>Agapostemon texanus</i> (Cresson, 1872)				X
<i>Agapostemon virescens</i> (Fabricius, 1775)				X*
<i>Andrena arabis</i> (Robertson, 1897)	X*			
<i>Andrena carlini</i> (Cockerell, 1901)	X			
<i>Andrena commoda</i> (Smith, 1879)	X			X
<i>Andrena crataegi</i> (Robertson, 1893)	X			X
<i>Andrena cressonii</i> (Robertson, 1891)	X			
<i>Andrena forbesii</i> (Robertson, 1891)	X			X
<i>Andrena hipotes</i> (Robertson, 1895)	X*			X
<i>Andrena imitatrix</i> (Cresson, 1872)	X			X*
<i>Andrena integra</i> (Smith, 1853)				X
<i>Andrena miserabilis</i> (Cresson, 1872)	X			X
<i>Andrena nasonii</i> (Robertson, 1895)	X*			X
<i>Andrena perplexa</i> (Smith, 1853)	X			X
<i>Andrena rugosa</i> (Robertson, 1891)	X			
<i>Andrena vicina</i> (Smith, 1853)	X			
<i>Andrena wilkella</i> (Kirby, 1802)	X		X	
<i>Apis mellifera</i> (Linnaeus, 1758)	X	X		X
<i>Augochlorella aurata</i> (Smith, 1853)	X*			X
<i>Bombus bimaculatus</i> (Cresson, 1863)	X	X*	X*	X*

<i>Bombus fervidus</i> (Fabricius, 1798)		X		X
<i>Bombus griseocollis</i> (DeGreer, 1773)		X	X	
<i>Bombus impatiens</i> (Cresson, 1863)	X	X	X	X
<i>Bombus perplexus</i> (Cresson, 1863)				X
<i>Bombus ternarius</i> (Say, 1837)				X
<i>Bombus vagans</i> (Smith, 1854)		X	X	X
<i>Ceratina calcarata</i> (Robertson, 1900)	X			
<i>Ceratina mikmaqi</i> (Rehan and Sheffield, 2011)				X
<i>Eucera atriventris</i> (Smith, 1854)				X
<i>Eucera hamata</i> (Bradley, 1942)				X*
<i>Halictus confusus</i> (Smith, 1853)	X			X
<i>Halictus ligatus</i> (Say, 1837)	X			X*
<i>Halictus rubicundis</i> (Christ, 1791)	X		X	X
<i>Hoplitis pilosifrons</i> (Cresson, 1864)				X
<i>Lasioglossum albipenne</i> (Robertson, 1890)				X
<i>Lasioglossum cinctipes</i> (Provancher, 1888)				X
<i>Lasioglossum coriaceum</i> (Smith, 1853)	X			X
<i>Lasioglossum forbseii</i> (Robertson, 1890)				X
<i>Lasioglossum foxii</i> (Robertson, 1895)	X			
<i>Lasioglossum hitchensi</i> (Gibbs, 2012)	X	X		X
<i>Lasioglossum imitatum</i> (Smith, 1853)	X			
<i>Lasioglossum leucozonium</i> (Schrank, 1781)				X
<i>Lasioglossum nymphaearum</i> (Robertson, 1895)	X			X
<i>Lasioglossum obscurum</i> (Robertson, 1892)	X			
<i>Lasioglossum paradmirandum</i>	X	X		X*

(Knerer and Atwood, 1966)				
<i>Lasioglossum pectorale</i> (Smith, 1853)	X			X
<i>Lasioglossum perpunctatum</i> (Ellis, 1913)	X			X
<i>Lasioglossum pilosum</i> (Smith, 1853)		X		X*
<i>Lasioglossum tegulare</i> (Robertson, 1901)	X	X		
<i>Lasioglossum timothyi</i> (Gibbs, 2010)				X
<i>Lasioglossum truncatum</i> (Robertson, 1901)	X			X*
<i>Lasioglossum versatum</i> (Robertson, 1902)	X			X
<i>Melissodes sp.</i>				X
<i>Nomada sp.</i>				X
<i>Osmia bucephala</i> (Cresson, 1864)				X
<i>Osmia cornifrons</i> (Radoszkowski, 1887)				X
<i>Osmia sp.</i>				X
<i>Osmia taurus</i> (Smith, 1873)				X
<i>Sphecodes dichrous</i> (Smith, 1853)				X
<i>Xylocopa virginica</i> (Linnaeus, 1771)	X	X*		X
<b><i>Total Species Richness</i></b>	<b>36</b>	<b>11</b>	<b>6</b>	<b>49</b>

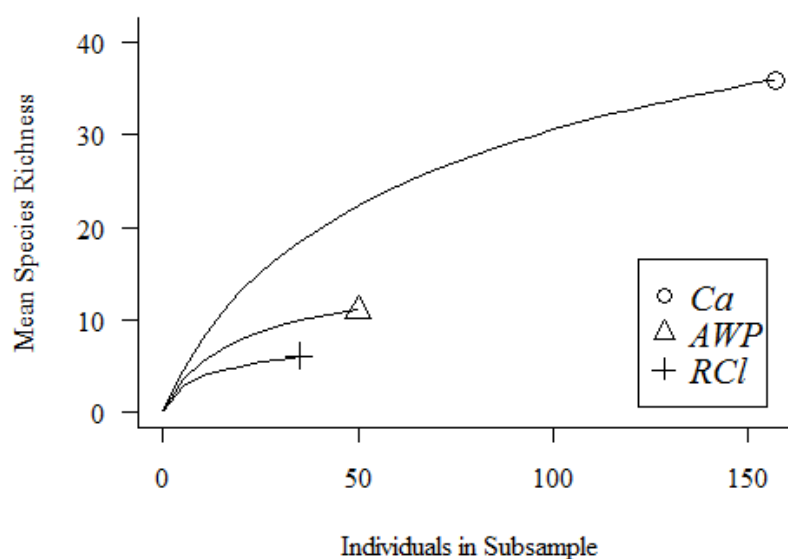


Figure 3-4. Species rarefaction curves for the three cover crop species. Ca=canola, AWP=Austrian winter pea, RCl=red clover.

The three cover crop species not only varied in total bee species richness but also in bee community composition. Canola was dominated by *Andrena* species and other small-bodied bee groups as those included in the small dark, large dark, tiny, and green bee morphospecies observation categories as well as Syrphidae flies (Figures 3-5 and 3-9). Austrian winter pea was visited by larger-bodied bumble bees (*Bombus sp.*), carpenter bees (*Xylocopa virginica*), and a select few tiny *Lasioglossum* species (Figures 3-6 and 3-9). Red clover, which supported the lowest total diversity, was primarily visited by bumble bees and *Andrena wilkella*, a species known to collect pollen primarily from clover flowers (J. Gibbs, *personal communication*) (Figures 3-7 and 3-9). In contrast, the most abundant species collected in the landscape-level traps were not only different than the most abundant species found on any cover crop, but two of the species collected in highest numbers, *Eucera hamata* and *Agapostemon virescens*, were not observed visiting any of the cover crops in this study (Figure 3-8).

In this experiment, the first four axes resulting from RDA accounted for 44.8% of the variation in the measured pollinator community (Figure 3-9). The first, second, third and fourth axes accounted for 12.1, 1.9, 17.4, and 13.4% of the species group variance, respectively. The first canonical axis was significant (Eigen value = 0.119,  $F = 28.28$ ,  $P = 0.002$ ), as was the total of all canonical variables (Trace = 0.138,  $F = 16.7$ ,  $P = 0.002$ ).



## Canola

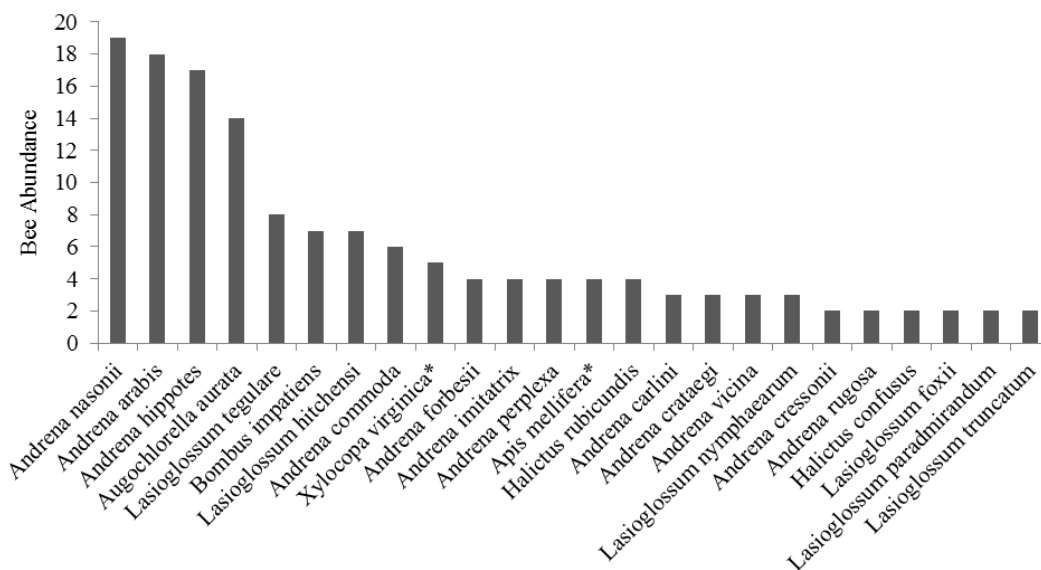


Figure 3-5. Netted abundance of individual bee species collected from canola plots throughout the total experiment. Asterisk indicates actual relative abundance may be greater than shown; collection was not necessary for identification to species-level and not all bees of these groups seen were collected. Only species that represented >1% of the total catch for the crop are shown.

## Austrian Winter Pea

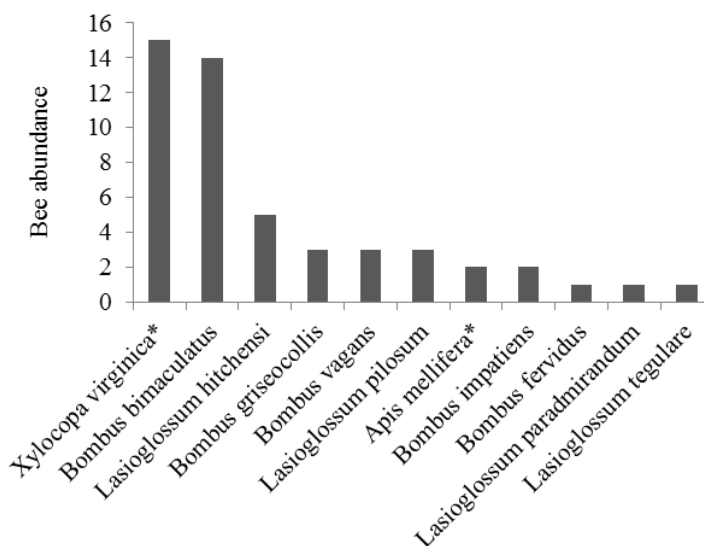


Figure 3-6. Netted abundance of individual bee species collected from Austrian winter pea plots throughout the total experiment. Asterisk indicates actual relative abundance may be greater than shown; collection was not necessary for identification to species-level and not all bees of these groups seen were collected.

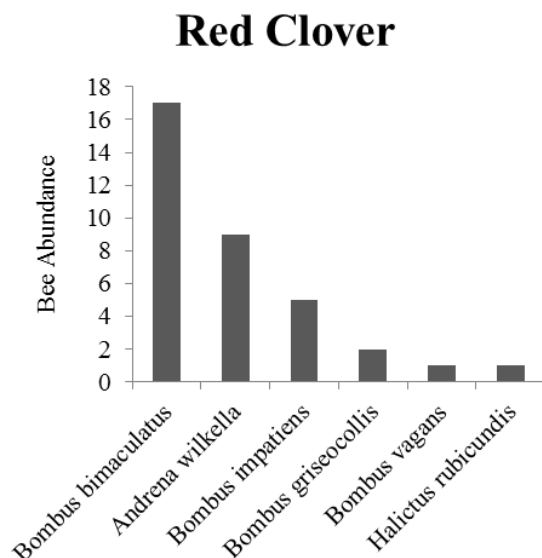


Figure 3-7. Netted abundance of individual bee species collected from red clover plots throughout the total experiment. Asterisk indicates actual relative abundance may be greater than shown; collection was not necessary for identification to species-level and not all bees of these groups seen were collected.

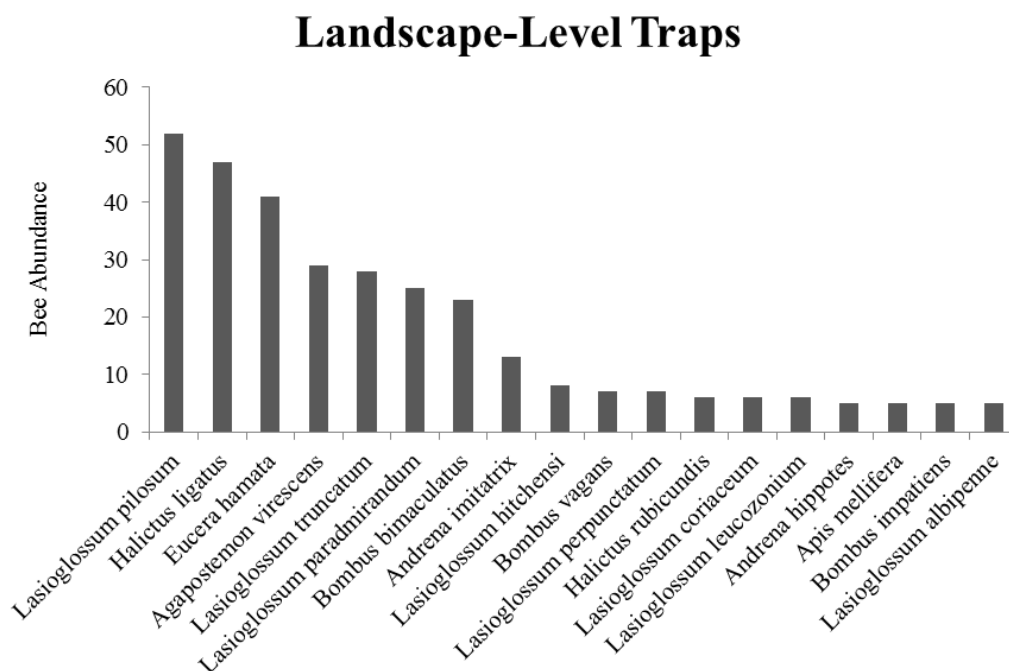


Figure 3-8. Abundance of individual bee species collected from combined landscape-level vane and pan traps throughout the span of the experiment. Only species that represented >1% of the total catch are shown.

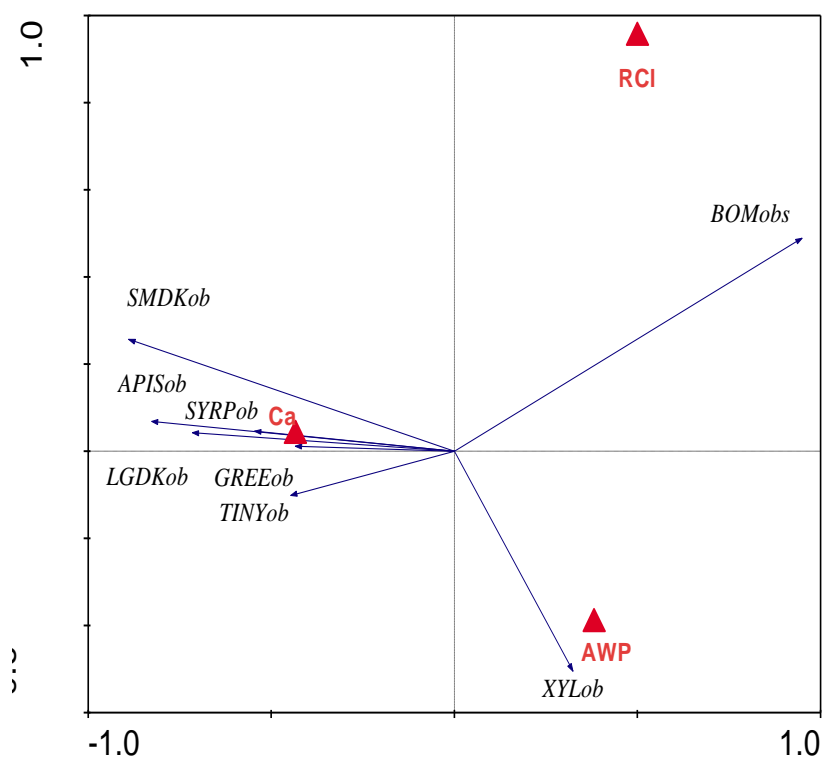


Figure 3-9. Multivariate biplot indicating the relative association of observed morphospecies groups to the three cover crop species. *XYL*=*Xylocopa virginica*, *BOM*= *Bombus sp.*, *APIS*=*Apis mellifera*, *LGDK*=large dark bees, *SMDK*=small dark bees, *GREE*=green bees, *TINY*=tiny bodied bees, *SYRP*=Syrphidae flies.

In addition to the divergences in bee communities across cover crop species, we observed differences in response of bee species and species groups to environmental variables of ambient temperature and average wind speed. Crops with significant, or nearly significant, correlations between environmental variables and insect responses of abundance or diversity included the environmental variables as covariates in all corresponding crop-specific analyses (Table 3-3). These variations in temperature and wind speed responses by cover crop is likely related to differences in the visiting insect community composition of each crop as different bee observation groups responded uniquely to variations in temperature and average wind speed (Figure 3-10). The resulting RDA for the wind and temperature environmental factors accounted for 53.5% of the variation in the measured pollinator community (Figure 3-10). The first, second, third and fourth axes accounted for 2, 1.5, 37.4, and 12.6% of the species variance, respectively. The first canonical axis was significant (Eigen value = 0.020,  $F = 6.983$ ,  $P = 0.002$ ), as was the total of all canonical variables (Trace = 0.035,  $F = 6.98$ ,  $P = 0.002$ ). Lower wind speed at the

time of sampling was also associated with the abundance of Syrphidae flies, tiny bees, and green bees, as lower temperatures were associated with small dark bees. However, as small dark bees were also more greatly associated with canola crops (Figure 3-9) which bloomed earlier in the season, the temperature effect here is likely related to seasonal differences rather than bee behavior.

Table 3-3. Environmental covariates and their respective correlations with each bee and Syrphidae response variable. Asterisks indicate those significant ( $p < 0.05$ ) correlations that were included as covariates in the general linear models with the corresponding insect response.

	Bee Visitation Abundance			Bee Species Richness		
	<i>F-stat</i> ( <i>DF<sub>num</sub></i> , <i>DF<sub>den</sub></i> )	<i>p-value</i>	<i>R</i> <sup>2</sup>	<i>F-stat</i> ( <i>DF<sub>num</sub></i> , <i>DF<sub>den</sub></i> )	<i>p-value</i>	<i>R</i> <sup>2</sup>
<b>Canola</b>						
Average Temperature	3.73(1,137)	0.056 <sup>+</sup>	0.027	1.14(1,137)	0.289	0.008
Average Wind Speed	0.04(1,137)	0.846	<0.001	2.95(1,137)	0.088	0.021
<b>Austrian Winter Pea</b>						
Average Temperature	0.42(1,138)	0.520	0.003	0.00(1,138)	0.987	<0.001
Average Wind Speed	10.3(1,138)	0.002*	0.070	0.40(1,138)	0.529	0.003
<b>Red Clover</b>						
Average Temperature	1.88(1,62)	0.175	0.030	0.002(1,62)	0.965	<0.001
Average Wind Speed	6.18(1,62)	0.016*	0.091	6.14(1,62)	0.016*	0.090

<sup>+</sup> Temperature was included in this model even though  $p > 0.05$ .

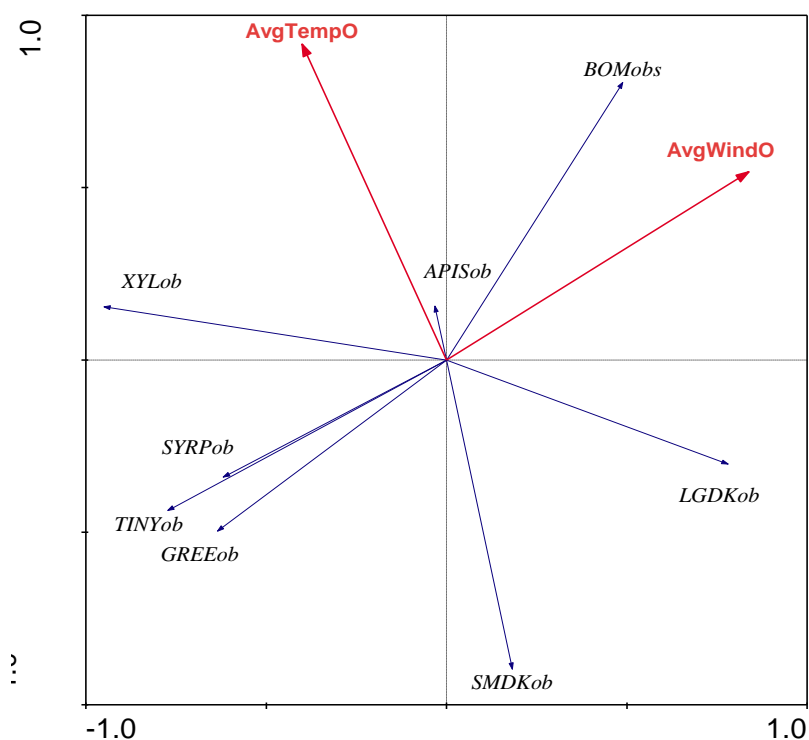


Figure 3-10. Multivariate biplot of observation morphospecies groupings and their corresponding responses to variations in environmental temperature and wind speed. XYL=*Xylocopa virginica*, BOM= *Bombus sp.*, APIS=*Apis mellifera*, LGDK=large dark bees, SMDK=small dark bees, GREE=green bees, TINY=tiny bodied bees, SYRP=Syrphidae flies.

Furthermore, the three cover crop species differed significantly in average number of bee visits and species diversity per observation and netting period with canola attracting significantly greater bee abundances than either Austrian winter pea ( $p < 0.001$ ) or red clover ( $p < 0.001$ ) (Figure 3-11). The same trend was also true for bee species diversity per observation period with canola attracting more species than either Austrian winter pea ( $p < 0.001$ ) or red clover ( $p < 0.001$ ) (Figure 3-12). Red clover and winter pea did not differ however in either visitation abundance ( $p = 0.965$ ) or species richness ( $p = 0.441$ ) (Figures 3-11 and 3-12). Syrphidae visitor abundance was also significantly higher in canola than in Austrian winter pea ( $p < 0.001$ ) or red clover ( $p = 0.013$ ) but no difference was found between red clover and Austrian winter pea ( $p = 0.315$ ) (Figure 3-13).

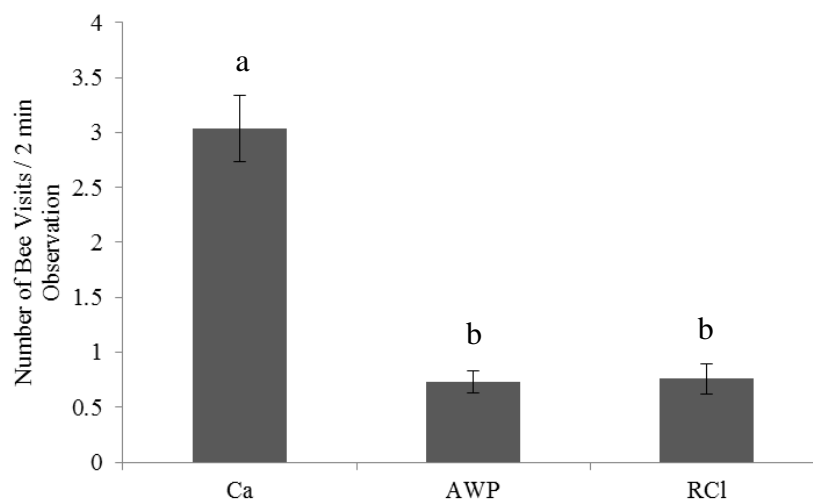


Figure 3-11. Average number of bee visits per two-minute observation period. Error bars are standard error of the mean; n=139 for canola, n=140 for AWP, and n=66 for red clover. Bars that do not share the same letter are significantly different at  $p < 0.05$  using Tukey's multiple comparison test.

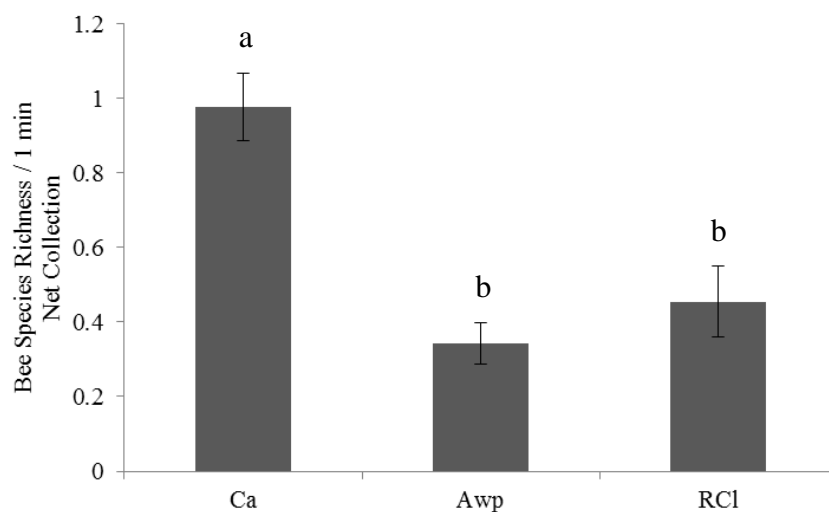


Figure 3-12. Average bee species richness by crop per one-minute post-observation netting period. Error bars represent standard error of the mean; n=139 for canola, n=140 for AWP, and n=66 for red clover. Bars that do not share the same letter are significantly different at  $p < 0.05$  using Tukey's multiple comparison test.

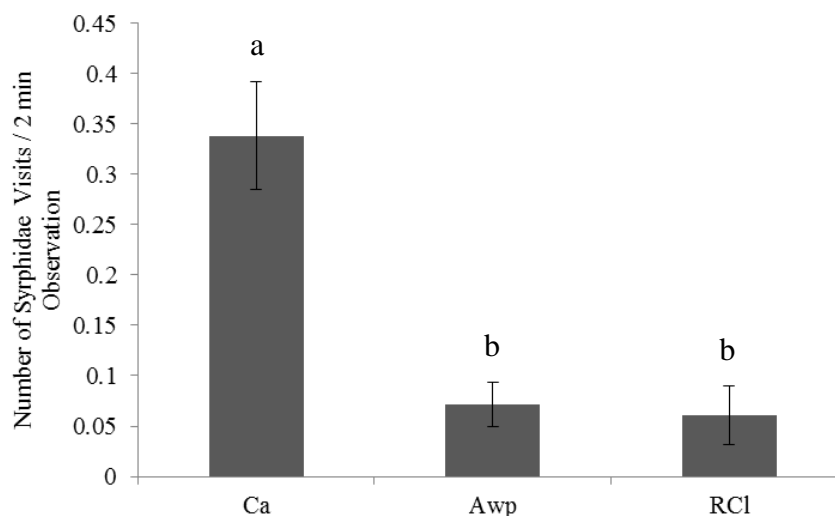


Figure 3-13. Average number of Syrphidae visits per two-minute observation period by crop. Error bars are standard error of the mean; n=139 for canola, n=140 for AWP, and n=66 for red clover. Bars that do not share the same letter are significantly different at  $p < 0.05$  using Tukey's multiple comparison test.

The three cover crop species differed in springtime flowering start and ending dates. Canola was the earliest cover crop to bloom, followed by Austrian winter pea and red clover. Flowering dates did overlap a few days between canola and Austrian winter pea, and between the peas and red clover (Figure 3-14). The dates of first, peak and final flowering for each crop and planting date are given in Table 3-4. Each cover crop had at least one planting date treatment that had poor winter survival (Table 3-4). Because no data was collected past July 8, 2013, the true peak blooming dates for red clover planting dates 1 through 3 are unknown. Additionally the few surviving plants in red clover planting date 4 did not produce any flowers prior to the completion of the study. For canola, the fourth planting date with lowest pre-winter biomass production also had the lowest winter survival (Table 3-5). Only three of the four replicate blocks for canola planting date four overwintered, with only an approximate 1-2% total plant survival in those blocks. However, the remaining fourth planting date canola plants did produce flowers within the time frame of the experiment, albeit delayed compared to the other three planting dates (Figure 3-14). A similar trend was found for red clover where the third and fourth planting dates, with lowest recorded fall biomass, also showed limited winter survival (Table 3-5). The third planting date had some surviving plants in all four blocks, although only one plot produced any flowers prior to the end of the study period. However, the fourth planting date of red clover only had surviving plants in one of the four blocks and did not produce any flowers during the study.

In contrast, Austrian winter pea had poor winter survival for the first planting date with greatest fall biomass (Table 3-5). Springtime flowers for this treatment were only observed on a few small areas of plot edge regrowth.

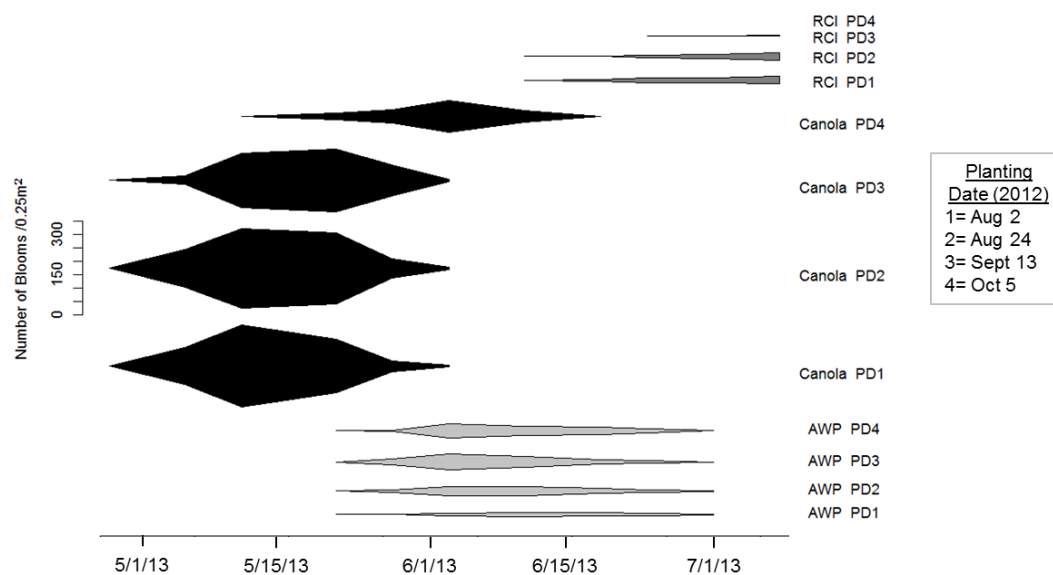


Figure 3-14. Number of open blooms per 0.25m<sup>2</sup> represented over time and as a function of increasing density. RCI=Red clover, AWP=Austrian winter pea, PD= Fall planting date.



Table 3-4. Dates of first recorded bloom, peak bloom, final bloom and quality of winter survival for each cover crop species and planting date.

Crop	Planting Date	First Recorded Flowers	Peak Flowering	Last Recorded Flowers	Winter Survival
Canola	1	April 21	May 12	June 3	Good
	2	April 28	May 12	June 3	Good
	3	May 6	May 22	June 3	Good
	4	May 21	June 3	June 19	Poor
AWP	1	May 28	June 11	July 1	Poor
	2	May 21	June 11	July 1	Good
	3	May 21	June 3	June 24	Good
	4	May 28	June 3	June 24	Good
Red clover	1	June 3	July 8*	n/a	Good
	2	June 3	July 8*	n/a	Good
	3	July 1	July 8*	n/a	Poor
	4	n/a	n/a	n/a	Poor

\* Red clover was not monitored, and no data was collected, after July 8. Therefore actual peak and end flowering dates are unknown.

Table 3-5. Dry fall plant biomass in grams by crop and planting date. Superscripts that do not share the same letter are significantly different at  $p < 0.05$  using Tukey's multiple comparison test.

Crop	Planting Date	Fall Biomass (g) ( $\pm$ SE)
Canola	1	113.76 ( $\pm$ 15.74) <sup>a</sup>
	2	89.48 ( $\pm$ 18.38) <sup>a</sup>
	3	10.04 ( $\pm$ 2.47) <sup>b</sup>
	4	0.97 ( $\pm$ 0.16) <sup>c</sup>
Austrian winter pea	1	115.22 ( $\pm$ 24.03) <sup>a</sup>
	2	52.63 ( $\pm$ 17.98) <sup>a</sup>
	3	5.64 ( $\pm$ 1.60) <sup>b</sup>
	4	1.58 ( $\pm$ 0.26) <sup>b</sup>
Red clover	1	23.07 ( $\pm$ 10.63) <sup>a</sup>
	2	11.87 ( $\pm$ 5.88) <sup>a</sup>
	3	0.30 ( $\pm$ 0.10) <sup>b</sup>
	4	0.22 ( $\pm$ 0.09) <sup>b</sup>

### Canola: Planting Date Comparisons

There were significantly fewer bee visits per two-minute observation period for the fourth planting date of canola than planting date one ( $p=0.002$ ), planting date two ( $p<0.001$ ) or planting date three ( $p<0.001$ ) (Figure 3-15). This trend does not hold for average bee species richness, however, as no significant differences were found between any planting date in terms of collected diversity (Figure 3-16).

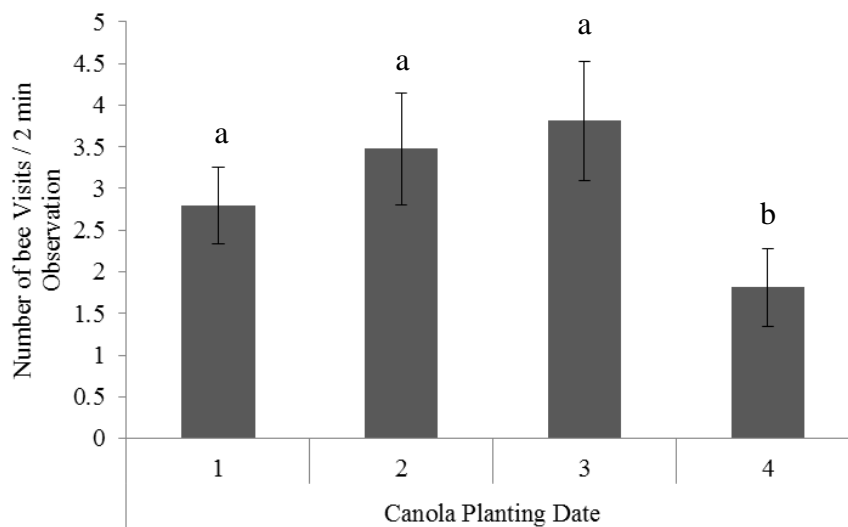


Figure 3-15. Average number of bee visits per two-minute observation period by canola planting date. Error bars represent standard error;  $n=40$  for planting dates 1 and 2,  $n=32$  for planting date 3, and  $n=27$  for planting date 4. Bars that do not share the same letter are significantly different at  $p<0.05$  using Tukey's multiple comparison test.

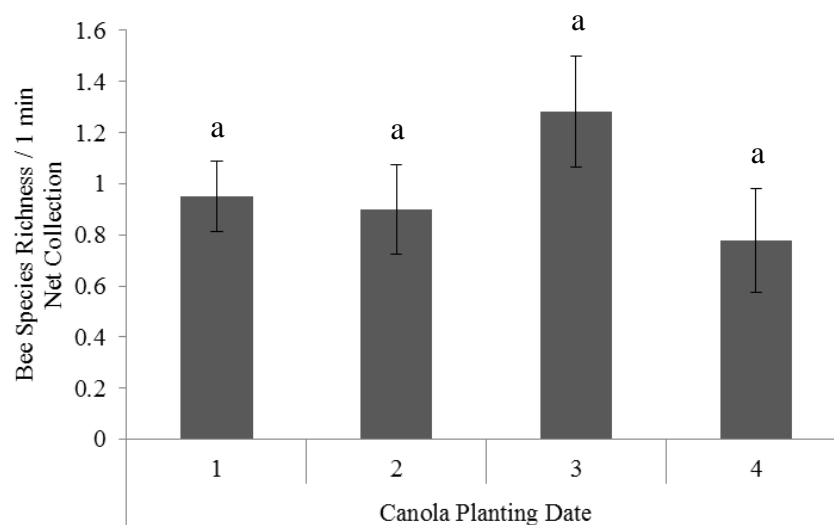


Figure 3-16. Average bee species richness per two-minute observation period for canola crop by fall planting date. Error bars represent standard error;  $n=40$  for planting dates 1 and 2,  $n=32$  for planting date 3, and  $n=27$  for planting date 4. Bars that do not share the same letter are significantly different at  $p<0.05$  using Tukey's multiple comparison test.

There were also differences found between plant growth properties across the four canola planting dates. These included significantly lower bloom density per  $0.25\text{m}^2$  in the fourth planting date than planting date one ( $p=0.003$ ), planting date two ( $p<0.001$ ) or planting date three ( $p=0.023$ ) (Figure 3-17). There was also significantly lower total canola plant density in planting date one than in planting dates two ( $p<0.001$ ) or three ( $p<0.001$ ) and between planting dates four and two ( $p<0.001$ ) and three ( $p<0.001$ ). However, no difference was found between planting dates one and four ( $p=0.633$ ) or two and three ( $p=0.110$ ) (Figure 3-18). While planting date four had low levels of bloom and plant density, planting date one showed total bloom density equal to that of the second and third plantings. This difference between low plant density and high flower density of planting date one is illustrated by a significantly greater number of blooms per plant in planting date one than in either of the other three planting dates ( $p=0.006$ ,  $p<0.001$ , and  $p<0.001$  for two, three and four respectively) (Figure 3-19). In addition we observed a significant, positive correlation in bee visitation with increasing canola flower density (Figure 3-20), although this trend was not significant for species richness (Figure 3-21).

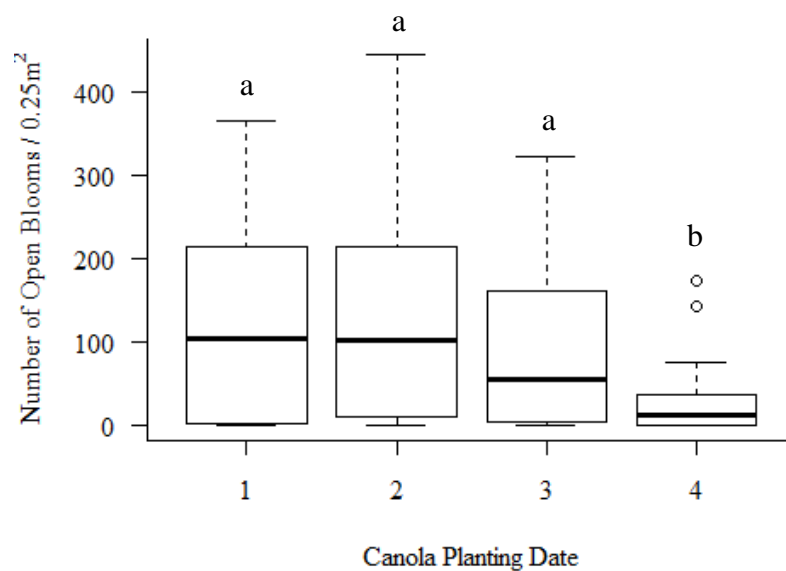


Figure 3-17. Box and whisker plot of the average number of open blooms per 0.25m<sup>2</sup> by canola fall planting date across all weeks of the blooming period. Boxes that do not share the same letter are significantly different at p < 0.05 using Tukey's multiple comparison test.

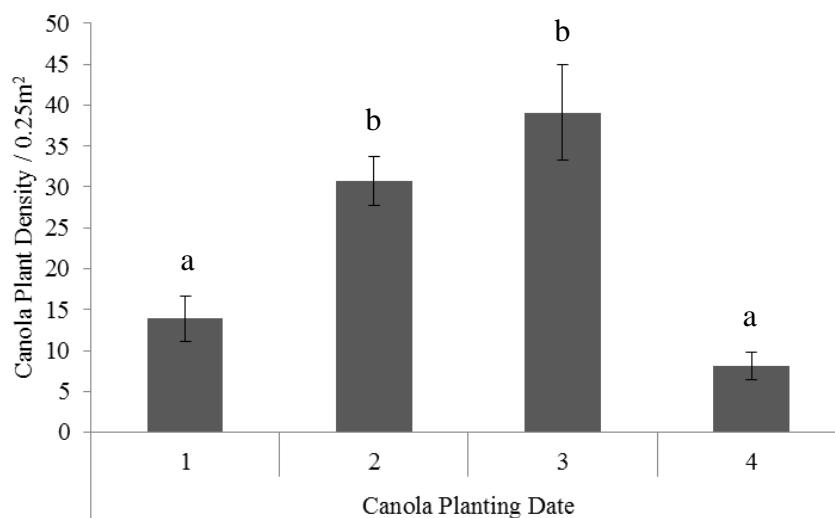


Figure 3-18. Average plant density (±SE) for canola plots by planting date. N=12 for planting dates 1-3 and n=9 for planting date 4. Bars that do not share the same letter are significantly different at p < 0.05 using Tukey's multiple comparison test.

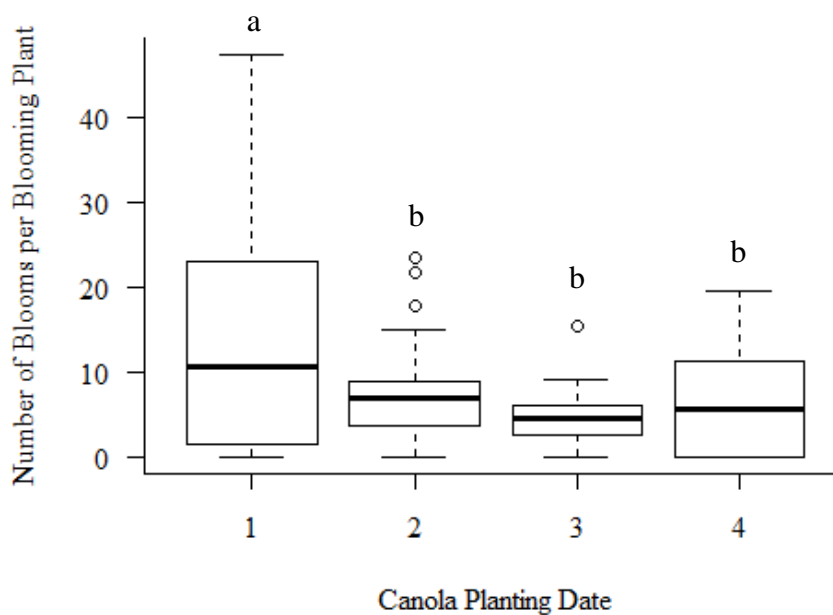


Figure 3-19. Box and whisker plot of the average number of blooms per blooming plant by canola planting date across all weeks of the blooming period. Boxes that do not share the same letter are significantly different at  $p < 0.05$  using Tukey's multiple comparison test.

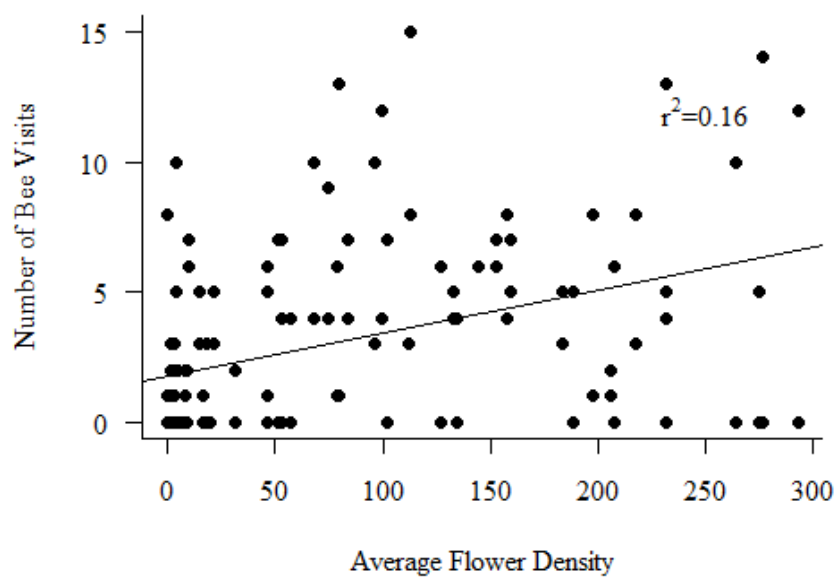


Figure 3-20. Correlation of number of bee visits to canola plots as a function of the average flower density for all planting date treatments.  $F_{1,137} = 26.02$ ,  $p < 0.001$ .

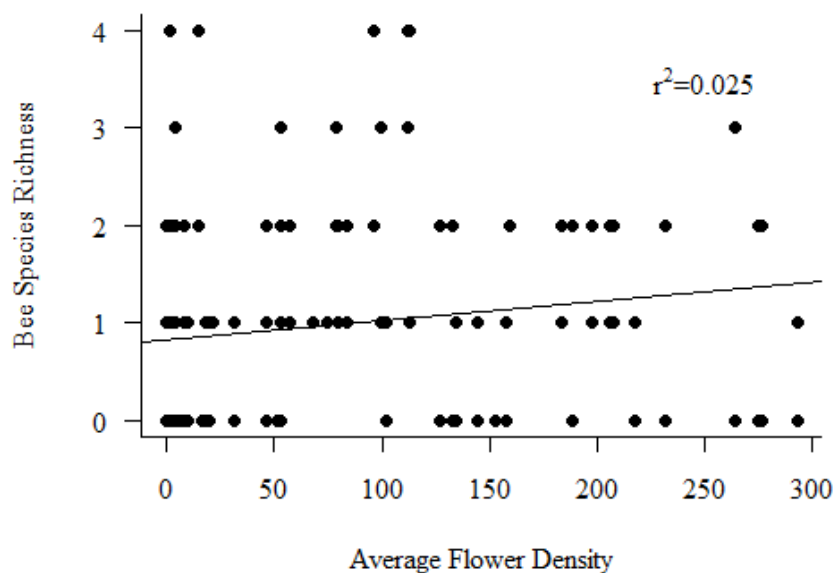


Figure 3-21. Correlation of bee species richness collected in canola plots as a function of the average flower density for all planting date treatments.  $F_{1,137}=3.46$ ,  $p=0.065$ .

### Austrian Winter Pea: Planting Date Comparisons

For Austrian winter pea, there were significantly fewer bee visits per two-minute observation period for the first planting date than in planting date two ( $p=0.008$ ), planting date three ( $p=0.002$ ), or planting date four ( $p=0.003$ ) (Figure 3-22). This trend is also seen across average bee species richness per post-observation netting period, however, only the difference between planting date one and four was statistically significant ( $p=0.045$ ), although marginal significance was observed for the comparison of planting dates one and two ( $p=0.079$ ) (Figure 3-23).

In addition, the first planting date of Austrian winter pea also had significantly lower flowers per  $0.25\text{m}^2$  than planting dates two ( $p=0.002$ ), three ( $p<0.001$ ) or four ( $p<0.001$ ) (Figure 3-24). This relationship between lower bee visitation in the first planting date and average flower density is also demonstrated via a significant, positive correlation between number of bee visits and bee species richness per observation period and the average flower density (Figures 3-25 and 3-26).

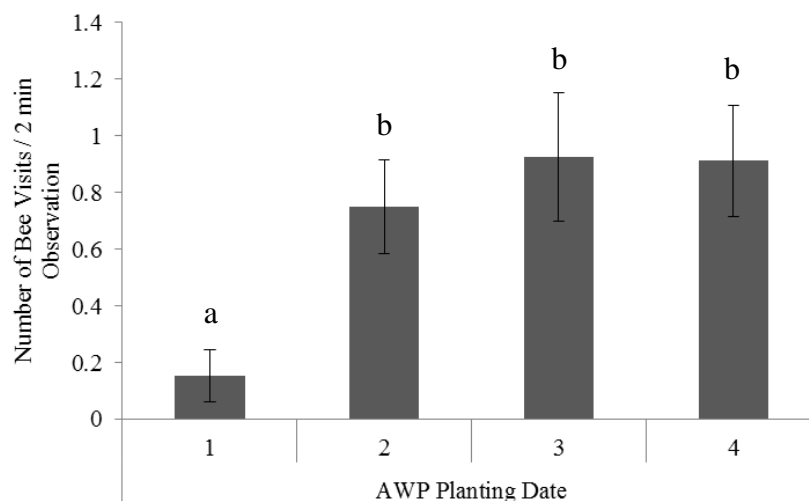


Figure 3-22. Average number of bee visits per two-minute observation period. Error bars are standard error of the mean;  $n=26$  for planting date 1,  $n=40$  for planting dates 2 and 3, and  $n=34$  for planting date 4. Bars that do not share the same letter are significantly different at  $p<0.05$  using Tukey's multiple comparison test.

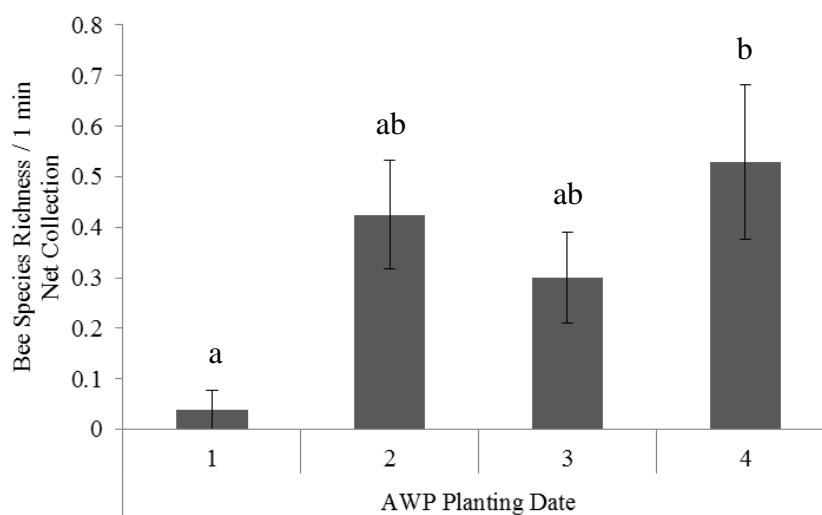


Figure 3-23. Average number of bee species per one-minute post-observation netting period. Error bars represent standard error of the mean;  $n=26$  for planting date 1,  $n=40$  for planting dates 2 and 3, and  $n=34$  for planting date 4. Bars that do not share the same letter are significantly different at  $p<0.05$  using Tukey's multiple comparison test.

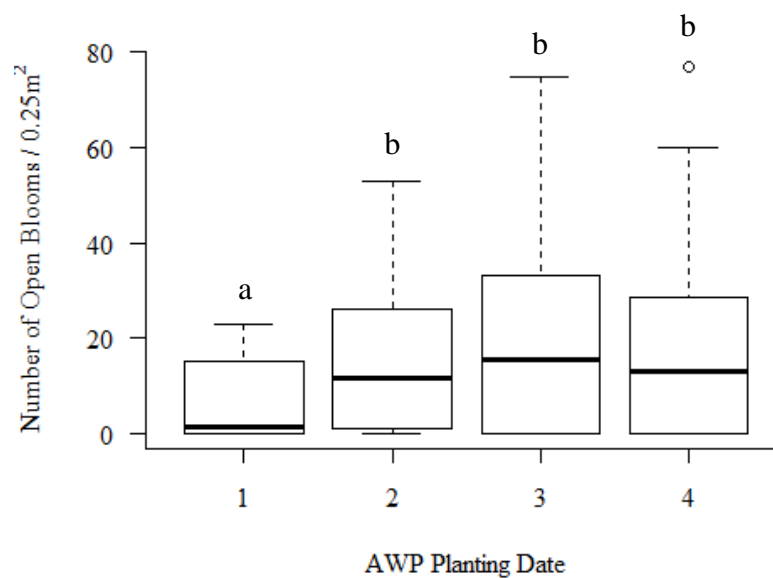


Figure 3-24. Box and whisker plot of the average number of open blooms per 0.25m<sup>2</sup> by Austrian winter pea planting date across all weeks of the blooming period. Boxes that do not share the same letter are significantly different at  $p < 0.05$  using Tukey's multiple comparison test.

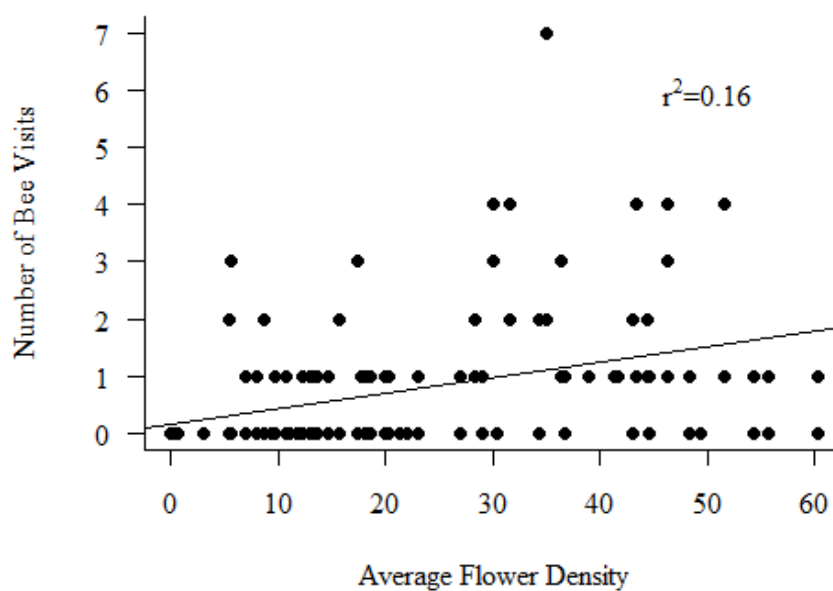


Figure 3-25. Correlation of number of bee visits on Austrian winter pea plots as a function of the average flower density for all planting date treatments.  $F_{1,138} = 25.38$ ,  $p < 0.001$ .



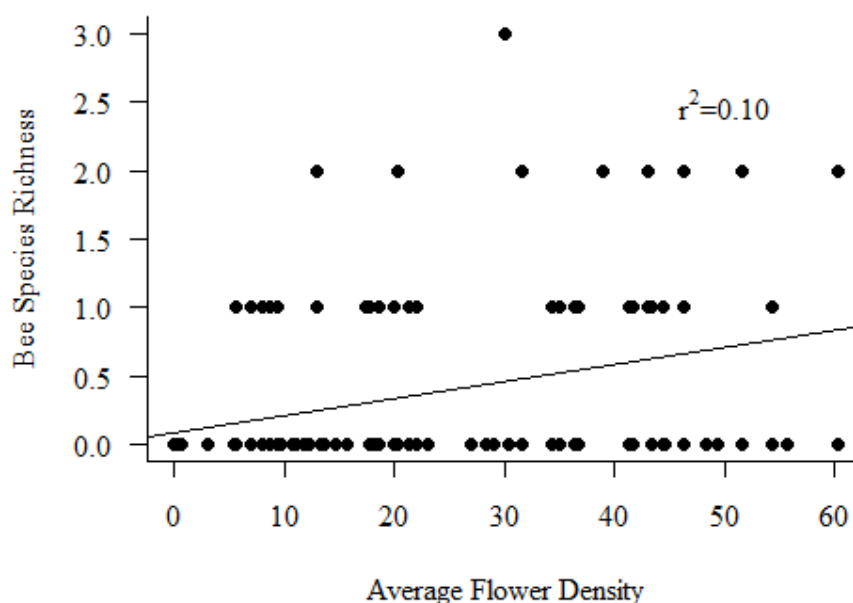


Figure 3-26. Correlation of bee species richness collected in Austrian winter pea plots as a function of the average flower density for all planting date treatments.  $F_{1,138}=15.2$ ,  $p<0.001$ .

### Red Clover: Planting Date Comparisons

In red clover, only the first two planting dates produced flowers during our study period. There were more bees observed visiting the flowers of the first planting date of red clover than the second, although this difference was only marginally significant ( $p=0.0523$ ) (Figure 3-27). However, statistical significance was observed for bee species richness per netting period with the first planting date having significantly greater species richness than the second planting date ( $p=0.015$ ) (Figure 3-28).

Although the third planting date of red clover only produced flowers during the final week of the study (see Table 3-4), there were significantly fewer total clover heads in the third planting date than the first or second ( $p<0.001$  and  $p<0.001$ ) (Figures 3-29). Planting date four was not included in these analyses as no flowers were produced during the study. Although there were no significant differences in flower density between planting dates one and two ( $p=0.306$ ), the mean flower density for planting date one ( $13.95$  clover heads/ $0.25\text{m}^2$ ) was greater than planting date two ( $10.6$  clover heads/ $0.25\text{m}^2$ ). As with canola and Austrian winter pea, red clover also showed a significant, positive relationship between average flower density and bee visitation abundance and species richness (Figures 3-30 and 3-31).

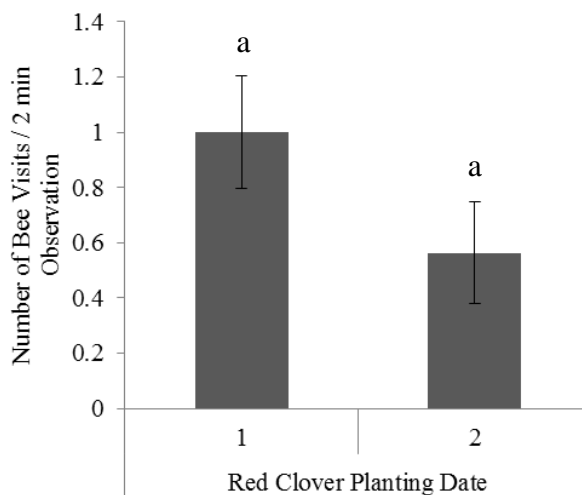


Figure 3-27. Average number of bee visits per two-minute observation period. Error bars shown are standard error of the mean;  $n=32$  for both planting dates. Bars that do not share the same letter are significantly different at  $p<0.05$  using Tukey's multiple comparison test.

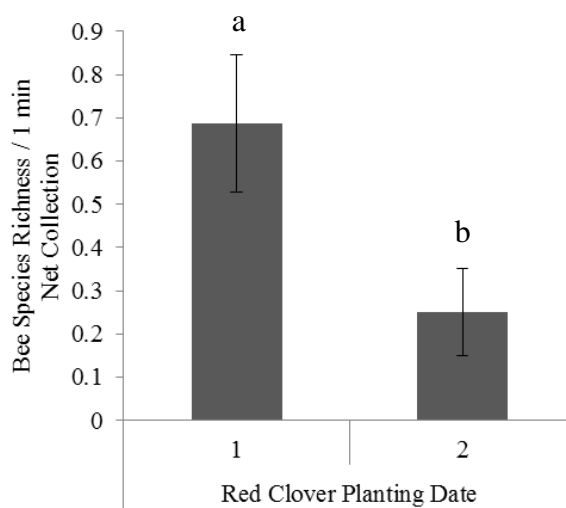


Figure 3-28. Average bee species richness per one-minute post-observation netting period. Error bars shown are standard error of the mean;  $n=32$  for both planting dates. Bars that do not share the same letter are significantly different at  $p<0.05$  using Tukey's multiple comparison test.

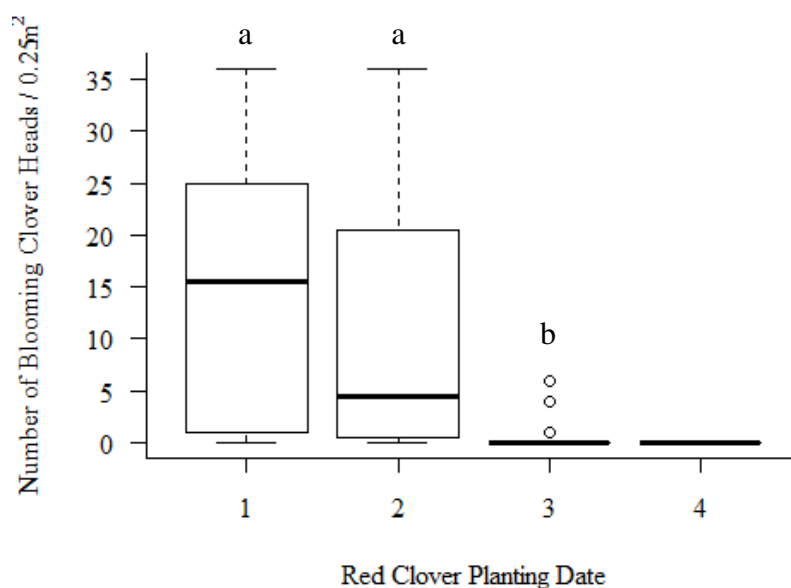


Figure 3-29. Box and whisker plot of the average number of blooming red clover heads, or flower clusters, by planting date across all weeks of the blooming period studied. Boxes that do not share the same letter are significantly different at  $p < 0.05$  using Tukey's multiple comparison test. Planting date 4 was not included in the analysis as no flowers were present during study.

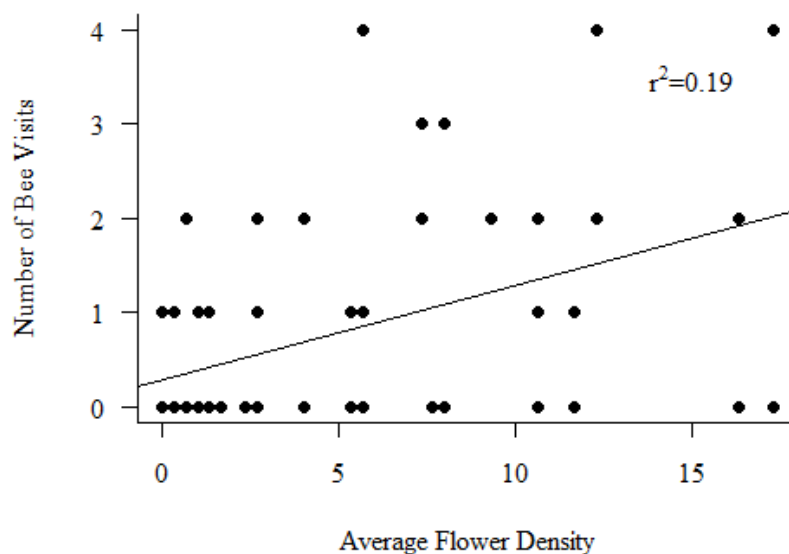


Figure 3-30. Correlation between number of bee visits to red clover plots as a function of average flower density across planting dates one and two only.  $F_{1,62} = 14.73$ ,  $p < 0.001$ .

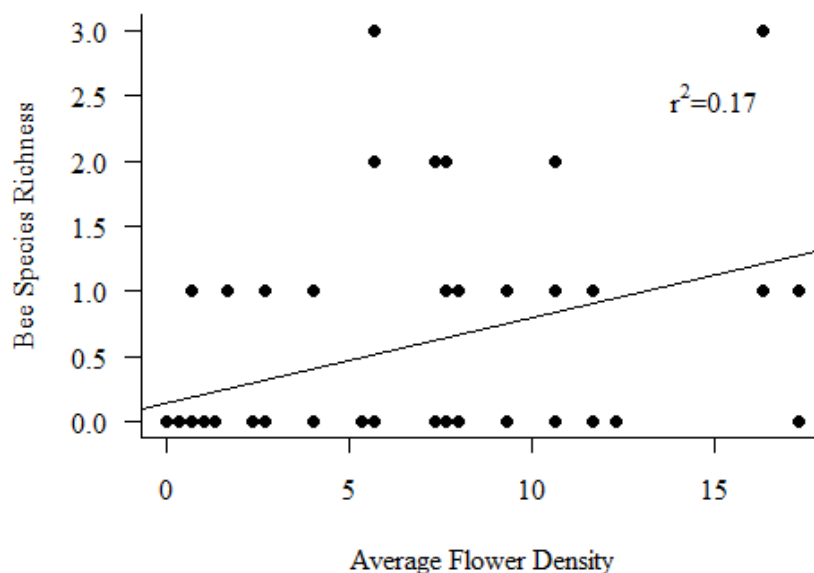


Figure 3-31. Correlation between bee species richness on red clover plots as a function of average flower density across planting dates one and two only.  $F_{1,62}=12.57$ ,  $p<0.001$ .

## DISCUSSION

The adoption of flowering overwintering cover crops in agricultural landscapes has the potential to not only increase floral resource availability for springtime native pollinator communities, but also to provide other valuable benefits to field quality and overall agronomic productivity. However, in order to encourage the increased use of flowering cover crops for their conservation potential, research on the influence of cover crop species selection and cash crop rotation schedules is essential for making accurate recommendations to farmers and other land managers. This study sought to fill in some of these research gaps by focusing on three common central Pennsylvanian flowering cover crop species and their floral resource provisioning across a gradient of fall planting dates.

The three species studied (canola, Austrian winter pea and red clover) each attracted unique native bee communities. Canola was the earliest cover crop to bloom and attracted the greatest diversity of bee species and the highest quantity of Syrphidae flies compared to the other two plant species. For these reasons, out of the three cover crops studied, canola is likely to be of highest conservation potential to the widest diversity of bee species in the greatest number of crop rotations. Given that Austrian winter pea and red clover did not flower until late-May to mid-June, these cover crops would require either rotation windows with summer cash crop planting

times or else require that portions of the cover crop be left in-field to achieve any floral resource benefit. Bee community composition was not, however, the only difference observed between cover crop species. We also observed significant differences between cover crop species in bee visitation abundance with more bees visiting canola than either Austrian winter pea or red clover. This was also true for Syrphidae visitation abundance across cover crops. This further supports the conclusion of canola being of greatest total pollinator benefit given the species studied and the timeframes considered in this study.

We also found differences across species and planting dates for both winter survival and subsequent spring blooming density. Although in most cases the planting dates with lowest flower density were also the treatments with lowest winter survival (e.g., canola planting date four, Austrian winter pea planting date one). However, for most cover crop species the major influence of bee visitation abundance or species richness was floral resource density. This density-driven influence has also been shown in other pollinator resource studies (Potts et al. 2003, 2009, Holzschuh et al. 2006, Tuell et al. 2008). While we also observed differences in canola plant physiology by planting date, such measurements could not be conducted in Austrian winter pea or red clover due to the difficulty in separating individual plants among the dense crop mat of intertwining stems. Overall, canola and red clover showed greatest winter survival and spring flower density with the earliest fall planting dates, while Austrian winter pea had higher survival for the later fall planting dates. From this we see that fall growth and winter survival is species dependent and should be considered along with spring flowering time for determining ideal cash crop rotation windows for each cover crop species given the time constraints of cash crop fall harvest and spring planting.

As indicated with variations in fall biomass between cover crop planting dates, accumulated growing degree-days influenced plant growth by planting date, especially in the fall. However, with the exception of canola planting date four, planting date did not have a significant effect on spring flowering time for the cover crop treatments. Instead, we saw the highest influence of planting date on winter survival rather than blooming time. As a result, observations of plant growth throughout the spring saw a convergence of crop growth and flowering densities across planting dates as the warm season progressed. This diminishing difference in plant growth across establishment gradients has also been observed in canola oilseed yield and hairy vetch cover crop planting date trials (Lutman et al. 2000, Teasdale et al. 2004).

We did not monitor pollen or nectar levels for any of the treatments and thus did not directly quantify resource availability to the pollinator community. Instead, we focused on flower

density as a representation of this resource. However, pollen or nectar differences may have contributed to the variations in visitation observed across cover crop species. Additionally, floral resource quantity or quality may have influenced the variations of bee visitation observed between the first and second planting dates of red clover. Red clover was the only crop to show variations in bee visitation and species richness across planting dates that did not correspond with similar patterns in flower density (Figures 27-29).

Another important result from this study is that no single trapping method or cover crop species was representative of the total bee community observed during this study. Instead, the 61 bee species found were distributed across cover crops and various passive trapping types and colors (see Appendix B for more information). This further supports the results of other studies that highlight the need for multiple collection methods in order to sample complete pollinator communities in a given area (Cane et al. 2000, Westphal et al. 2008, Wilson et al. 2008).

In conclusion, it is important to consider winter survival, spring blooming time, and visiting bee community composition when making decisions about cover crop species selection for pollinator conservation purposes. For example, careful consideration of cash crop rotation limits (i.e., required planting date for summer cash crop) would be necessary prior to recommending any of these cover crop species for springtime pollinator benefit based on cover crop blooming time. This experiment does not provide definitive answers to the question of which flowering cover crop is best for a particular rotation, but rather highlights the importance of all these factors in cover crop species selection as well as to inform the future the study of other flowering cover crop species and their associated pollinators. The influence of planting date, for example, was not uniform across cover crops (e.g., winter survival was greatest for early or late plantings depending on species) and further study is required before the potential of overwintering cover crops as a conservation tool for native pollinators is fully understood. In total, the knowledge accumulated in this study will help to educate conservationists and land managers on selecting the appropriate cover crop species for a variety of crop rotation schedules used in central Pennsylvania.

## LITERATURE CITED

- Adamsen, F. J., and T. A. Coffelt. 2005. Planting date effects on flowering, seed yield, and oil content of rape and crambe cultivars. *Industrial Crops and Products* 21:293–307.
- Alessi, J., J. F. Power, and D. C. Zimmerman. 1977. Sunflower Yield and Water Use as Influenced by Planting Date, Population, and Row Spacing. *Agronomy Journal* 69:465–469.
- Bartomeus, I., J. S. Ascher, D. Wagner, B. N. Danforth, S. Colla, S. Kornbluth, and R. Winfree. 2011. Climate-associated phenological advances in bee pollinators and bee-pollinated plants. *Proceedings of the National Academy of Sciences of the United States of America* 108:20645–9.
- Bates, D., M. Maechler, B. Bolker, and S. Walker. 2013. lme4: Linear Mixed-Effects Models Using Eigen and S4. <http://cran.r-project/package=lme4>.
- Bouseman, J. K., and W. E. LaBerge. 1979. A Revision of the Bees of the Genus *Andrena* of the Western Hemisphere. Part IX. Subgenus *Melandrena*. *Transactions of the American Entomological Society* 104:275–389.
- Ter Braak, C. J. F. 2003. CANOCO Version 4.5A 1988-2003. Biometris - quantitative methods in the life and earth sciences. Plant Research International, Wageningen, The Netherlands.
- Ter Braak, C. J. F., and P. Smilauer. 2002. CANOCO reference manual and CanoDraw for Windows User's Guide, Software for Canonical Community Ordination (version 4.5). 500 pp. Microcomputer Power, Ithaca, NY.
- Cane, J. H., R. L. Minckley, and J. Kervin. 2000. Sampling Bees (Hymenoptera: Apiformes) for Pollinator Community Studies : Pitfalls of Pan- Trapping. *Journal of the Kansas Entomological Society* 73:225–231.
- Clark, A. (Ed.). 2007. *Managing Cover Crops Profitably* 3rd ed. Sustainable Agriculture Research and Education (SARE).
- Discover Life. (n.d.). . [www.discoverlife.org](http://www.discoverlife.org).
- Elliott, S. E. 2009. Subalpine Bumble Bee Foraging Distances and Densities in Relation to Flower Availability. *Environmental Entomology* 38:748–756.
- Gibbs, J. 2011. Revision of the metallic *Lasioglossum* (*Dialictus*) of eastern North America (Hymenoptera: Halictidae: Halictini). *Zootaxa* 3073:1–216.
- Gibbs, J., L. Packer, S. Dumes, and B. N. Danforth. 2013. Revision and reclassification of *Lasioglossum* (*Evylaeus*), L. (*Hemihalictus*) and L. (*Sphecodogastra*) in eastern North America (Hymenoptera: Apoidea: Halictidae). *Zootaxa* 3672:001–117.

- Goulson, D., G. C. Lye, and B. Darvill. 2008. Decline and conservation of bumble bees. *Annual Review of Entomology* 53:191–208.
- Greenleaf, S. S., and C. Kremen. 2006a. Wild bees enhance honey bees' pollination of hybrid sunflower. *Proceedings of the National Academy of Sciences of the United States of America* 103:13890–5.
- Greenleaf, S. S., and C. Kremen. 2006b. Wild bee species increase tomato production and respond differently to surrounding land use in Northern California. *Biological Conservation* 133:81–87.
- Hendrickx, F., J.-P. Maelfait, W. Van Wingerden, O. Schweiger, M. Speelmans, S. Aviron, I. Augenstein, R. Billeter, D. Bailey, R. Bukacek, F. Burel, T. Diekötter, J. Dirksen, F. Herzog, J. Liira, M. Roubalova, V. Vandomme, and R. Bugter. 2007. How landscape structure, land-use intensity and habitat diversity affect components of total arthropod diversity in agricultural landscapes. *Journal of Applied Ecology* 44:340–351.
- Hodgson, J. A., W. E. Kunin, C. D. Thomas, T. G. Benton, and D. Gabriel. 2010. Comparing organic farming and land sparing: optimizing yield and butterfly populations at a landscape scale. *Ecology letters* 13:1358–67.
- Hole, D. G., A. J. Perkins, J. D. Wilson, I. H. Alexander, P. V. Grice, and A. D. Evans. 2005. Does organic farming benefit biodiversity? *Biological Conservation* 122:113–130.
- Holzschuh, A., I. Steffan-Dewenter, D. Kleijn, and T. Tscharntke. 2006. Diversity of flower-visiting bees in cereal fields: effects of farming system, landscape composition and regional context. *Journal of Applied Ecology* 44:41–49.
- Hothorn, T., F. Bretz, and P. Westfall. 2008. Simultaneous Inference in General Parametric Models. *Biometrical Journal* 50:346–363.
- Jacobs, J. 2011. Individual Based Rarefaction Using R-package. <http://www.jennajacobs.org/R/rarefaction.html>.
- Kimoto, C., S. J. Debano, R. W. Thorp, S. Rao, and W. P. Stephen. 2012. Investigating temporal patterns of a native bee community in a remnant north american bunchgrass prairie using blue vane traps. *Journal of insect science (Online)* 12:108.
- Klein, A.-M., B. E. Vaissière, J. H. Cane, I. Steffan-Dewenter, S. a Cunningham, C. Kremen, and T. Tscharntke. 2007. Importance of pollinators in changing landscapes for world crops. *Proceedings of the Royal Society B: Biological Sciences* 274:303–13.
- Kremen, C., and A. Miles. 2012. Ecosystem Services in Biologically Diversified versus Conventional Farming Systems : Benefits , Externalities , and Trade-Offs. *Ecology and Society* 17(4):40.



- Kremen, C., N. M. Williams, and R. W. Thorp. 2002. Crop pollination from native bees at risk from agricultural intensification. *Proceedings of the National Academy of Sciences of the United States of America* 99:16812–6.
- Lal, R., E. Regnier, D. J. Eckert, W. M. Edwards, and R. Hammond. 1991. Expectations of cover crops for sustainable agriculture. Pages 1–11 in W. L. Hargrove, editor. *Cover Crops for Clean Water*. Soil and Water Conservation Society.
- LeBuhn, G., S. Droege, N. Williams, B. Minckley, T. Griswold, C. Kremen, O. Messinger, J. Cane, T. Roulston, F. Parker, V. Tepedino, and S. Buchmann. 2002. The Bee Inventory Plot. [online.sfsu.edu/beeplot/](http://online.sfsu.edu/beeplot/).
- Leong, J. M., and R. W. Thorp. 1999. Colour-coded sampling: the pan trap colour preferences of oligolectic and nonoligolectic bees associated with a vernal pool plant. *Ecological Entomology* 24:329–335.
- Leps, J., and P. Smilauer. 2003. *Multivariate analysis of ecological data using CANOCO*. 269 pp. University Press, Cambridge, MA.
- Lutman, P. J. W., P. Bowerman, G. M. Palmer, and G. P. Whytock. 2000. Prediction of competition between oilseed rape and *Stellaria media*. *Weed Research* 40:255–269.
- Major, D. J., D. R. Johnson, J. W. Tanner, and I. C. Anderson. 1975. Effects of Daylength and Temperature on Soybean Development. *Crop Science* 15:174–179.
- Mandelik, Y., R. Winfree, T. Neeson, and C. Kremen. 2012. Complementary habitat use by wild bees in agro-natural landscapes. *Ecological applications* : a publication of the Ecological Society of America 22:1535–46.
- Matson, P. A., W. J. Parton, A. G. Power, and M. J. Swift. 1997. Agricultural Intensification and Ecosystem Properties. *Science* 277:504–509.
- McGinley, R. J. 1986. Studies of Halictinae (Apoidea: Halictidae), I: Revision of New World *Lasioglossum* Curtis. *Smithsonian Contributions to Zoology* 429.
- Morandin, L. A., and M. L. Winston. 2005. Wild bee abundance and seed production in conventional, organic, and genetically modified canola. *Ecological Applications* 15:871–881.
- National Research Council. 2007. *Status of Pollinators in North America*. The National Academies Press, Washington, DC.
- O’Toole, C., and A. Raw. 1991. *Bees of the World*. Facts On File, New York, NY.
- Oksanen, J., F. G. Blanchet, R. Kindt, P. Legendre, P. R. Minchin, R. B. O’Hara, G. L. Simpson, P. M. Solymos, H. H. Stevens, and H. Wagner. 2013. *vegan: Community Ecology Package*. <http://cran.r-project.org/package=vegan>.

- Ollerton, J., R. Winfree, and S. Tarrant. 2011. How many flowering plants are pollinated by animals? *Oikos* 120:321–326.
- Potts, S. G., J. C. Biesmeijer, C. Kremen, P. Neumann, O. Schweiger, and W. E. Kunin. 2010. Global pollinator declines: trends, impacts and drivers. *Trends in Ecology & Evolution* 25:345–53.
- Potts, S. G., B. Vulliamy, A. Dafni, G. Ne'Eman, and P. Willmer. 2003. Linking Bees and Flowers : How Do Floral Communities Structure Pollinator Communities? *Ecology* 84:2628–2642.
- Potts, S. G., B. A. Woodcock, S. P. M. Roberts, T. Tscheulin, E. S. Pilgrim, V. K. Brown, and J. R. Tallowin. 2009. Enhancing pollinator biodiversity in intensive grasslands. *Journal of Applied Ecology* 46:369–379.
- R Core Team. 2012. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. <http://www.r-project.org/>.
- Ricketts, T. H. 2004. Tropical Forest Fragments Enhance Pollinator Activity in Nearby Coffee Crops. *Conservation Biology* 18:1262–1271.
- Sala, O. E., F. S. Chapin, J. J. Armesto, E. Berlow, J. Bloomfield, R. Dirzo, E. Huber-Sanwald, L. F. Hueneke, R. B. Jackson, A. Kinzig, R. Leemans, D. M. Lodge, H. A. Mooney, M. Oesterheld, N. L. Poff, M. T. Sykes, B. H. Walker, M. Walker, and D. H. Wall. 2000. Global biodiversity scenarios for the year 2100. *Science* 287:1770–4.
- Snapp, S. S., S. M. Swinton, R. Labarta, D. Mutch, J. R. Black, R. Leep, J. Nyiraneza, and K. O'Neil. 2005. Evaluating Cover Crops for Benefits, Costs and Performance within Cropping System Niches. *Agronomy Journal* 97:322–332.
- Teasdale, J. R., T. E. Devine, J. A. Mosjidis, R. R. Bellinder, C. E. Beste, and M. June. 2004. Growth and Development of Hairy Vetch Cultivars in the Northeastern United States as Influenced by Planting and Harvesting Date. *Agronomy Journal* 96:1266–1271.
- Tilman, D., J. Fargione, B. Wolff, C. D'Antonio, A. Dobson, R. Howarth, D. Schindler, W. H. Schlesinger, D. Simberloff, and D. Swackhamer. 2001. Forecasting agriculturally driven global environmental change. *Science* 292:281–4.
- Tuell, J. K., A. K. Fiedler, D. Landis, and R. Isaacs. 2008. Visitation by Wild and Managed Bees (Hymenoptera: Apoidea) to Eastern U. S. Native Plants for Use in Conservation Programs. *Environmental Entomology* 37:707–718.
- USDA. 2013. National Organic Program: Organic Standards. [http://www.ams.usda.gov/AMSv1.0/ams.fetchTemplateData.do?template=TemplateN&rightNav1=NOSBlinkNOSBMeetings&topNav=&leftNav=&page=NOPOrganicStandards&resultType=.](http://www.ams.usda.gov/AMSv1.0/ams.fetchTemplateData.do?template=TemplateN&rightNav1=NOSBlinkNOSBMeetings&topNav=&leftNav=&page=NOPOrganicStandards&resultType=)

- VanEngelsdorp, D., J. D. Evans, C. Saegerman, C. Mullin, E. Haubruge, B. K. Nguyen, M. Frazier, J. Frazier, D. Cox-Foster, Y. Chen, R. Underwood, D. R. Tarpy, and J. S. Pettis. 2009. Colony collapse disorder: a descriptive study. *PloS one* 4:e6481.
- Vasseur, C., A. Joannon, S. Aviron, F. Burel, J.-M. Meynard, and J. Baudry. 2013. The cropping systems mosaic: How does the hidden heterogeneity of agricultural landscapes drive arthropod populations? *Agriculture, Ecosystems & Environment* 166:3–14.
- Westphal, C., R. Bommarco, G. Carre, E. Lamborn, N. Morison, T. Petanidou, S. G. Potts, S. P. M. Roberts, H. Szentgyörgyi, T. Tscheulin, B. E. Vaissière, M. Wojciechowski, J. Biesmeijer, W. E. Kunin, J. Settele, and I. Steffan-Dewenter. 2008. Measuring Bee Diversity in Different European Habitats and Biogeographical Regions. *Ecological Monographs* 78:653–671.
- Williams, G. R., D. R. Tarpy, D. vanEngelsdorp, M.-P. Chauzat, D. L. Cox-Foster, K. S. Delaplane, P. Neumann, J. S. Pettis, R. E. L. Rogers, and D. Shutler. 2010. Colony Collapse Disorder in context. *BioEssays : news and reviews in molecular, cellular and developmental biology* 32:845–6.
- Williams, N. M., J. Regetz, and C. Kremen. 2012. Landscape-scale resources promote colony growth but not reproductive performance of bumble bees. *Ecology* 93:1049–58.
- Wilson, J. S., T. Griswold, and O. J. Messinger. 2008. Sampling Bee Communities ( Hymenoptera : Apiformes ) in a Desert Landscape : Are Pan Traps Sufficient ? *Journal of the Kansas Entomological Society* 81:288–300.
- Winfree, R. 2010. The conservation and restoration of wild bees. *Annals of the New York Academy of Sciences* 1195:169–97.
- Winfree, R., R. Aguilar, D. P. Vazquez, G. LeBuhn, and M. A. Aizen. 2009. A meta-analysis of bees' responses to anthropogenic disturbance. *Ecology* 90:2068–2076.
- Winfree, R., T. Griswold, and C. Kremen. 2007. Effect of human disturbance on bee communities in a forested ecosystem. *Conservation biology : the journal of the Society for Conservation Biology* 21:213–23.

## Chapter 4

### CONCLUSION AND FUTURE DIRECTIONS

The data collected and presented in these two cover crop experiments has helped to shed light on several agronomic properties of noteworthy importance when considering the application of flowering cover crops for native pollinator conservation. Because the primary farmer use of cover crops is likely to continue to be for field-based agronomic benefits like soil quality enhancement, the adoption of cover crops for pollinators is an extra, auxiliary ecosystem benefit. However, because the focus of growers is not likely to be on maximizing cover crop flower production, it is important to have a full understanding on how management adjustments such as cover crop selection, fall planting date, spring termination time, and cover crop plant diversity affect blooming time and concentration as well as the pollinator community that utilizes them. While this may in some ways provide more questions than answers, it does help to move us in a positive direction for understanding the pollinator conservation potential of flowering cover crops in agricultural landscapes.

Because of the myriad of benefits that can be gained through the planting of cover crops for field productivity and soil protection, careful selection of cover crop plant species to those that have the potential of producing flowers in the chosen crop rotation window is a small adjustment that may have large positive pollinator conservation effects. Indeed, the theory behind the use of flowering cover crops for pollinator conservation is that a farmer can implement these strategies with limited additional effort, especially if they are already using cover crops in their rotation. In contrast, other commonly used pollinator conservation methods, such as hedgerows or field edge pollinator plantings, require committing areas of land or other inputs solely for the purpose of attracting bees or beneficial insects. In these single purpose scenarios, pollination or pest reduction services are the only direct return for farm productivity, whereas cover crops also have increased soil enhancement value.

Successful conservation initiatives include not only enhancing habitat and decreasing risks to threatened insect communities, but also spreading the word and increasing awareness of the issue. In fact, public outreach is considered one of the most important tools for current native pollinator conservation strategies (National Research Council 2007, Mader et al. 2011). Unlike

some other agricultural pollinator conservation approaches that involve removing land from cultivation, the incorporation of flowering cover crops requires little more than careful planning and consideration for flower production and pollinator use. For this reason, increased public education and farmer extension on the benefits of cover crops to the farm as well as to local pollinator communities will hopefully greatly increase their adoption and provide conservation services to agricultural landscapes across the country. In many cases there may even be financial incentive programs or assistance available for conserving farmland natural resources. Such incentives can often be utilized for cover crop adoption and/or pollination plantings (see examples in Table 4-1).

Table 4-1. Examples of additional information or financial resources available about growing cover crops or general pollinator conservation.

Name of Agency or Organization	Type of Organization	Type of Assistance Available	Website
Natural Resources Conservation Service (NRCS)	US Government (USDA)	Informative and Financial Assistance	<a href="http://www.nrcs.usda.gov/wps/portal/nrcs/main/national/plantsanimals/pollinate/">http://www.nrcs.usda.gov/wps/portal/nrcs/main/national/plantsanimals/pollinate/</a> or see 'Using Farm Bill Programs for Pollinator Conservation' <a href="http://plants.usda.gov/pollinators/Using_Farm_Bill_Programs_for_Pollinator_Conservation.pdf">http://plants.usda.gov/pollinators/Using_Farm_Bill_Programs_for_Pollinator_Conservation.pdf</a>
University Extension Offices (i.e., Penn State Extension)	University	Informative	Cover crops: <a href="http://extension.psu.edu/plants/crops/soil-management/cover-crops">http://extension.psu.edu/plants/crops/soil-management/cover-crops</a> Native Pollinators: <a href="http://extension.psu.edu/pests/ipm/native-pollinators">http://extension.psu.edu/pests/ipm/native-pollinators</a>
The Xerces Society for Invertebrate Conservation	Non-Profit	Informative	<a href="http://www.xerces.org">www.xerces.org</a>
The Pollinator Partnership	Non-Profit	Informative	<a href="http://www.pollinator.org">www.pollinator.org</a>

Unfortunately, the studies discussed here do not directly measure conservation services. To do so would require either a studied comparison of pollinator communities between landscapes with and without flowering cover crops across habitat gradients, or at least consideration for increasing local bee communities over multiple generations at a single location. Bee communities are often quite different in species composition or dominance from year to year. For this reason the species collected and observed during this study may be different from those observed in future years were this study to be repeated. Additionally, as the true benefit of providing these additional floral resources to the bee community is in increasing the reproductive success of the bees, univoltine species will not demonstrate the success of this effort until their

offspring emerge the following year. For this reason, it would take a study of multiple years to determine whether flowering cover crops are actually increasing bee population sizes in the area.

Another valuable future research avenue would be studying the impact of spring-flowering cover crops on the direct pollination services provided to following, or nearby, pollinator-dependent cash crops. Would flowering cover crops that attract your main cash crop pollinators (e.g., bumble bees needed for tomato production) actually increase the populations of those bees on your farm? Is it possible that the cover crops are competing for pollinators with neighboring spring-flowering crops (e.g., spring apple bloom)? Could attracting pollinators to a high concentration resource like canola and then abruptly terminating said resource trap or kill the bees during the process of cover crop termination? All of these are important questions that ought to be answered and considered for future research as well as during the promotion of increased flowering cover crop adoption.

However, what these studies do provide is valuable information on the blooming potential and visiting pollinator communities present on the three common flowering cover crop species studied. Examples for how the data collected from these projects can be applied to real life crop rotations and farmer timing requirements are shown in figures 4-1 and 4-2 below. These illustrations demonstrate how the limitations of a particular cash crop rotation window would either allow or prohibit cover crop flower production depending on fall planting date and spring cover crop termination timing. In a winter cover crop window prior to spring organic corn planting (such as was the rotation in the plant diversity experiment discussed in Chapter 2), only earlier fall planting dates of canola would be able to produce flowers before termination (Figure 4-1). In contrast, if the same cover crop species were grown prior to pumpkin planting, which would not typically happen until early-summer, canola crops would have completed flowering and the Austrian winter pea would also have provided floral resources in that timeframe (Figure 4-2). One suggestion on how to achieve cover crop flowering during more restricted rotations would be to leave a section or strip of the cover crop in the field for an extended period. This would not only allow for more flower production, but may help enhance pollination services to the cash crop if additional pollinators are attracted to the field. While other flowering cover crop species may also have potential for this purpose, the information given can be used to initiate discussion, inform research and educate extension recommendations for the alternative uses and benefits of cover crops for native pollinators. The goal of this thesis was to provide a baseline and research framing from which future examinations could expand, and I believe these objectives were successfully met.

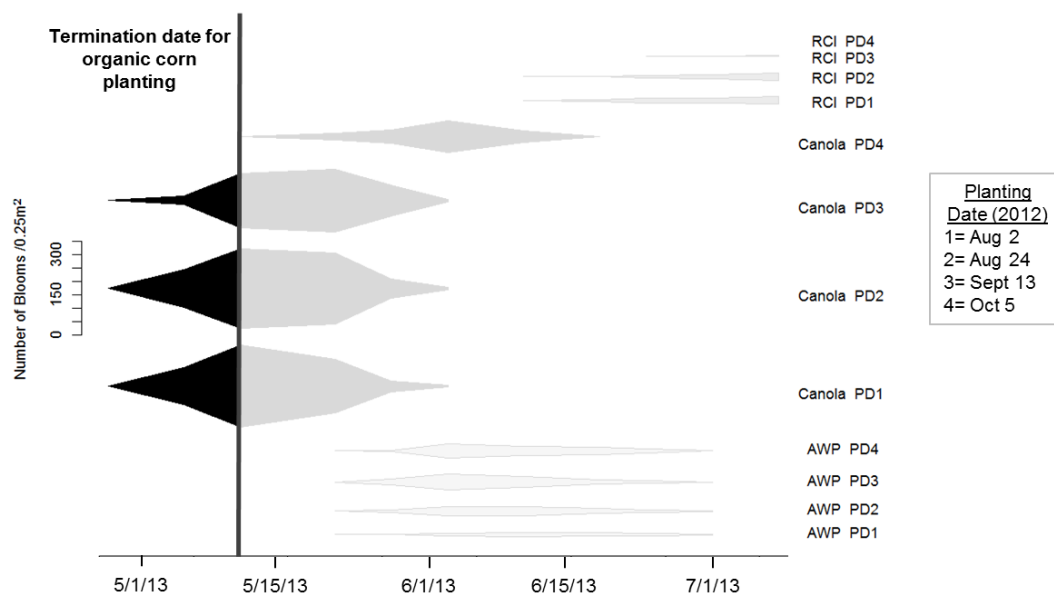


Figure 4-1. Number of open blooms per 0.25m<sup>2</sup> represented over time and illustrating the flower density that would be achieved prior to cover crop termination in a crop rotation window before spring organic corn planting. RCI=Red clover, AWP=Austrian winter pea, PD= Fall planting date.

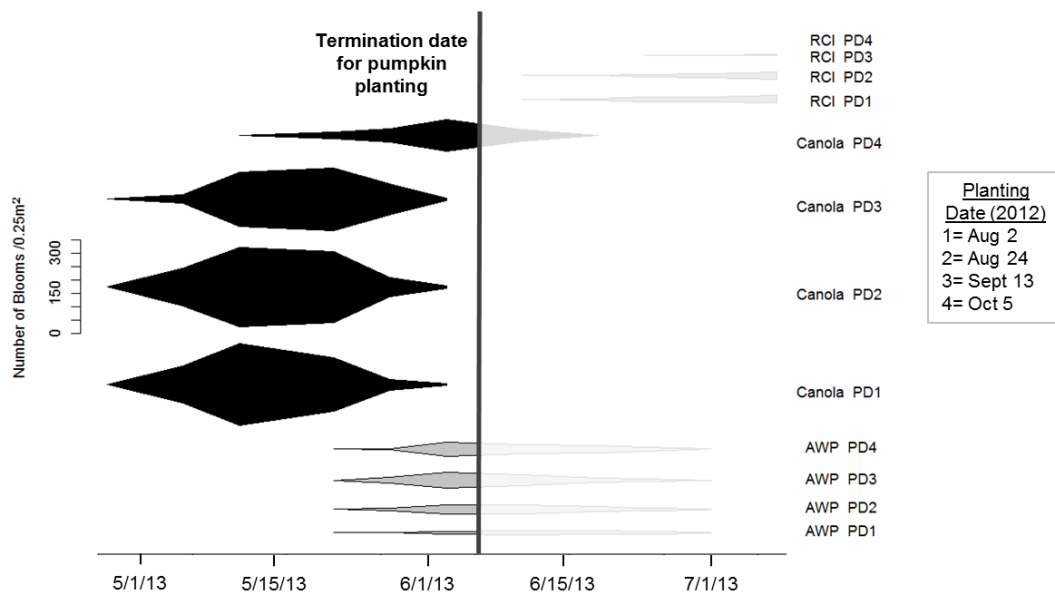


Figure 4-2. Number of open blooms per 0.25m<sup>2</sup> represented over time and illustrating the flower density that would be achieved prior to cover crop termination in a crop rotation window before a summer pumpkin crop. RCI=Red clover, AWP=Austrian winter pea, PD= Fall planting date.

Overall, this thesis extends the range of standard cover crop research by focusing on the potential of blooming cover crop species to act as a conservation potential for native pollinators. While the discussion about benefits to pollinators from flowering cover crops is not necessary novel (e.g., Mader et al. 2011), the evidence given in such cases is often lacking in supporting scientific research. The information provided in this thesis herein hopefully enhances the literature on this topic by providing evidence that cover crop species selection, planting and termination dates, and species diversity can all have significant effects on cover crop blooming potential and their associated native pollinator communities.



**Literature Cited**

Mader, E., M. Shepherd, M. Vaughan, S. H. Black, and G. LeBuhn. 2011. *Attracting Native Pollinators: Protecting North America's Bees and Butterflies*. Storey Publishing, North Adams, MA.

National Research Council. 2007. *Status of Pollinators in North America*. The National Academies Press, Washington, DC.

### Appendix A

### Experimental Plot Map

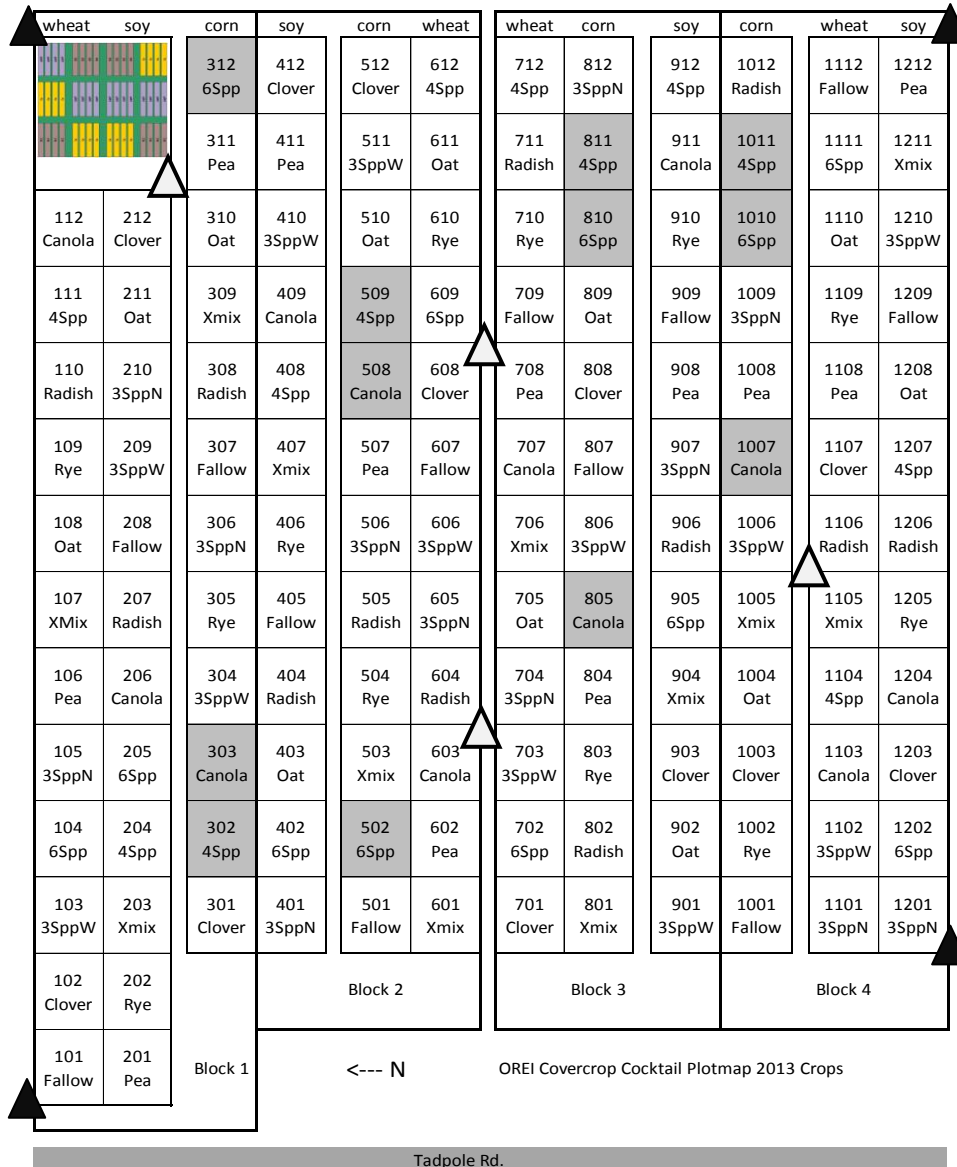


Figure 5-1. Plot map of the cover crop and plant diversity study discussed in Chapter 2. Shaded plots indicate the cover crop treatments selected for study in this project. Triangles represent the locations of landscape-level passive trapping; filled triangles are traps on project boundary edges, open triangles are traps on the project interior. Crops listed on column headings indicate 2013 summer crops grown in that strip.

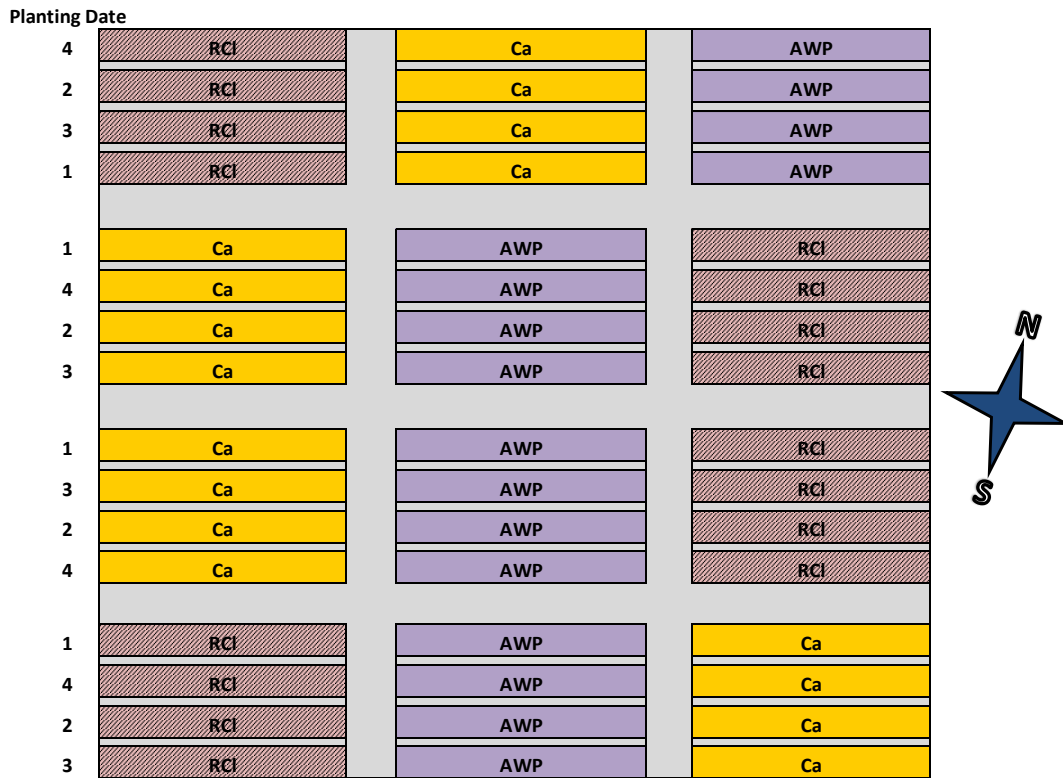


Figure 5-2. Plot map of cover crop planting date study discussed in Chapter 3. Blocks are illustrated in rows of each crop, main plots by cover crop species, and split-plots by planting date as indicated on the left-hand edge of the map. Planting dates are consistent in rows across the block. Ca= Canola; AWP= Austrian winter pea; RCI= Red clover.

## Appendix B

### Landscape-level Trap Collection Summary Data

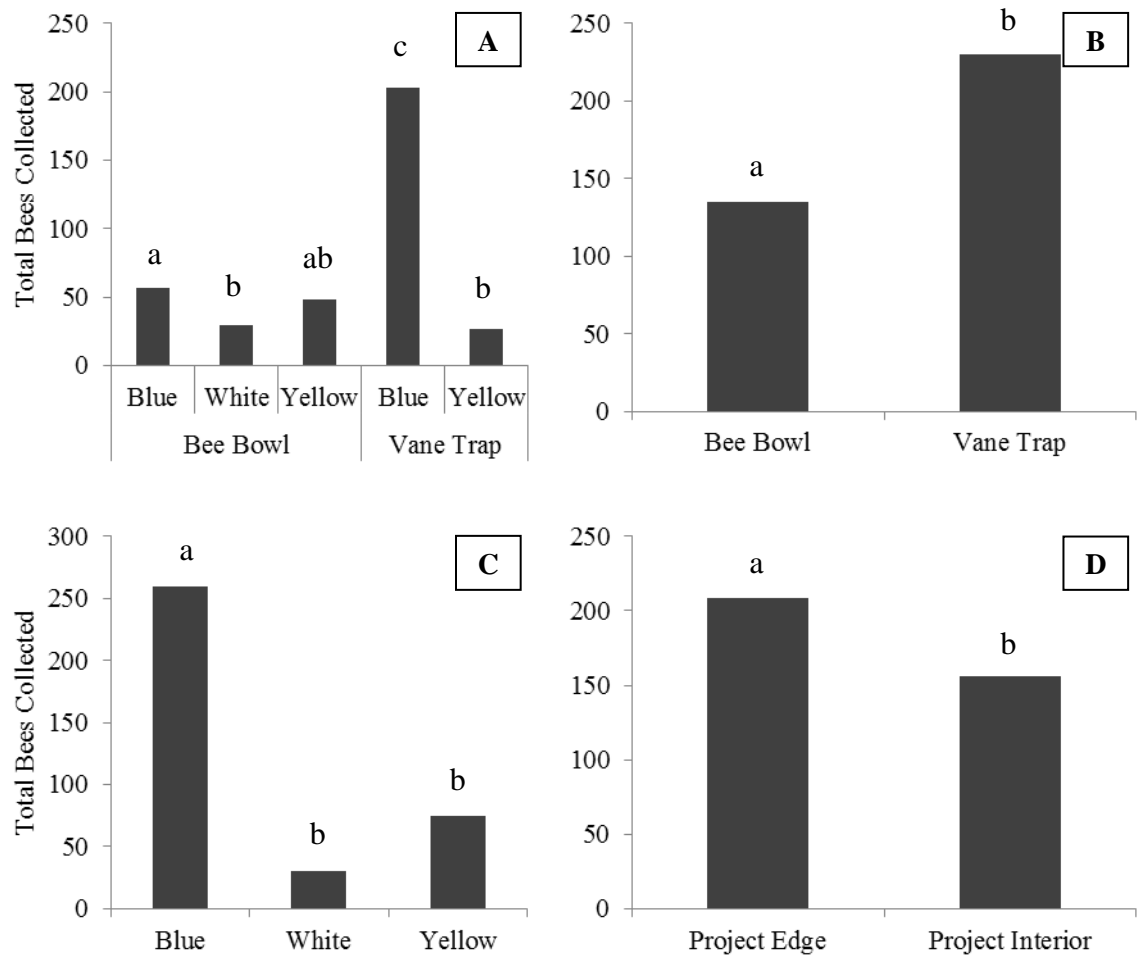


Figure 6-1. Total abundance of bees collected in the landscape-level passive traps across all project dates (April-July). (A) Bees by trap type and color; (B) by trap type only; (C) by trap color only; and (D) by field location (see Figure 5-1 for specific field locations). Bars that do not share the same letter are significantly different at  $p < 0.05$  using Tukey's multiple comparison test.

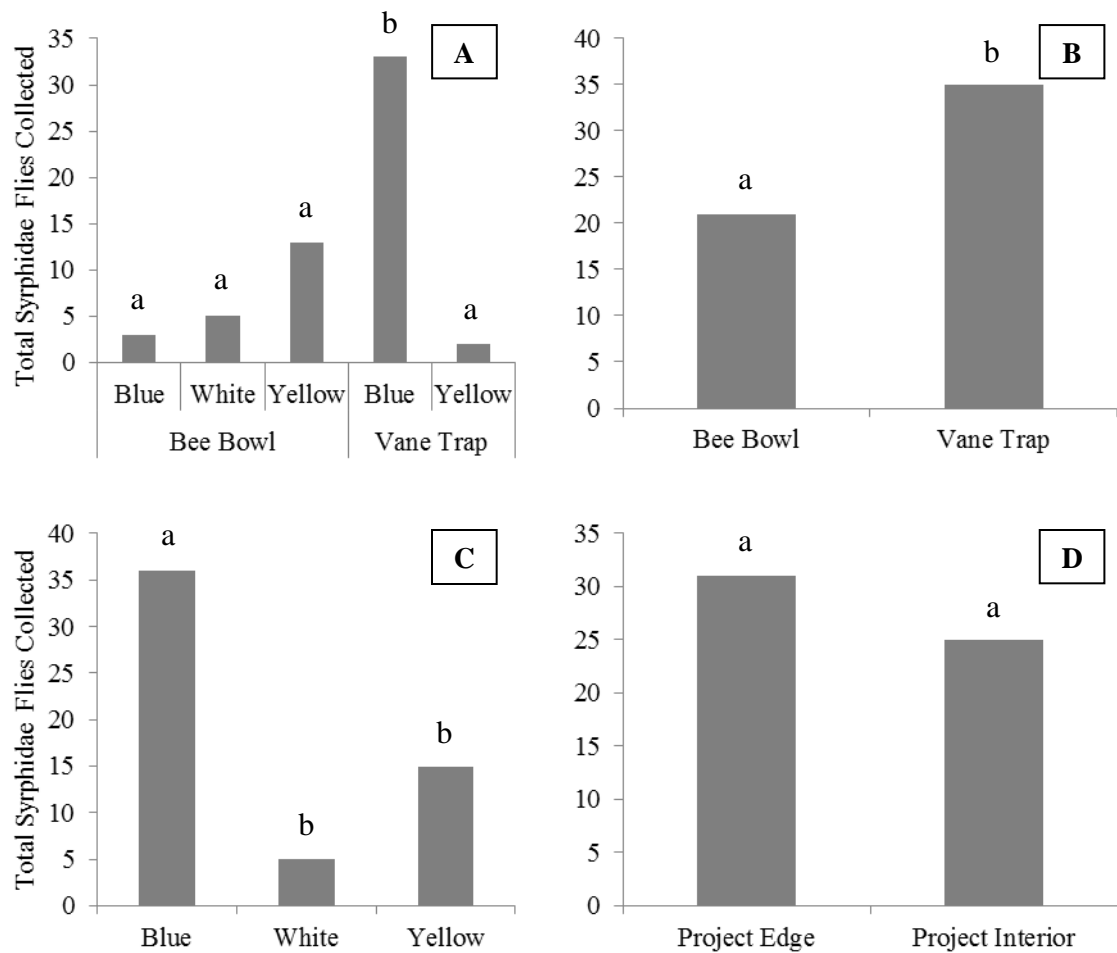


Figure 6-2. Total abundance of Syrphidae flies collected in the landscape-level passive traps across all project dates (April-July). (A) Syrphidae flies by trap type and color; (B) by trap type only; (C) by trap color only; and (D) by field location (see Figure 5-1 for specific field locations). Bars that do not share the same letter are significantly different at  $p < 0.05$  using Tukey's multiple comparison test.