

HABITAT MANIPULATIONS TO ENHANCE THE ABUNDANCE AND DIVERSITY OF
POLLINATORS VISITING *HELIANTHUS ANNUUS* L.

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Abstract

HABITAT MANIPULATIONS TO ENHANCE THE ABUNDANCE AND DIVERSITY OF POLLINATORS VISITING *HELIANTHUS ANNUUS* L.

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Insect pollinators face many challenges with the development of modern agriculture. These challenges, such as reduction in habitat range and landscape fragmentation, may be overcome by diversifying the plant community. By planting intercrops alongside a cash crop, there is potential to promote the abundance and richness of pollinator insect species by providing additional resources. Using visual observations and blue vane traps to monitor the bee and fly pollinators of sunflower, buckwheat, and partridge pea, we determined if intercrops can enhance pollinator services in sunflower by promoting abundance and diversity. We found that while the presence of a buckwheat intercrop enhances sunflower seed yields, buckwheat does not enhance pollinator abundance or diversity in the cropping system. Alternatively, the presence of partridge pea enhances the abundance of pollinators within sunflower crops but does not influence sunflower yields. Additionally, when buckwheat and partridge peas are planted together as an intercrop mixture, competition exists and buckwheat presence reduces plant height of partridge peas. These findings are important to understand the role of intercrops and polycultures in cropping systems to assist pollinator conservation.

Chapter 1:

LITERATURE REVIEW

Introduction

The need for pollination services by both native bees and honey bees is growing (Burkle et al., 2013). As the world's population increases, the demand for agricultural land and food crops also increases (Bentrup, 2016). While there are crops that do not require insect pollination for propagation, such as grain crops, there are many fruit, vegetable, and oilseed plants that rely on insect pollinators to spread their genetic information (McGregor, 1976). Both honey bees and native bees are extremely important and necessary as efficient pollinators, with their worth being estimated at over \$10 billion annually (Williams et al., 2014). Unfortunately, with the development of modern agriculture, insect pollinators face many challenges such as fragmented landscapes, reduction in forage ranges, and climate change (Kremen et al., 2002, Bartomeus et al., 2014).

Some of these challenges may be overcome by diversifying crop environments. Planting a variety of crops may increase the diversity of the cropping system (Brody, 1997), but may also diversify pollinator communities and promote the abundance of the bees and flies that are responsible for pollination services by providing multiple additional resources (Hudson, 2009, Herrera, 1987).

Sunflower Cropping

Sunflower, *Helianthus annuus* L. (Asteraceae), production in the United States is over a half billion dollar industry and sunflower is the fifth largest oilseed crop in the

world (National Sunflower Association's 2013 crop report). Confectionary sunflowers are grown for human consumption, but the oilseed variety are grown to produce sunflower oil, protein meal for livestock, and bird seed for game and other backyard birds (Marcy & Martin, 1986). There has also been research on using sunflower oil as a biofuel (Zhao et al., 2015).

Some cultivars of sunflower are able to self-pollinate, but insects play a crucial role in the movement of pollen for seed production (Degrandi- Hoffman, 2006, 2008, Parker, 1981a). Agricultural practices that manipulate the landscape within sunflower fields to provide additional resources and habitat for pollinators can increase pollination services resulting in more profitable yields (Jadhav, 2011). The use of cover crops, and more specifically intercrops, increases biodiversity within an agroecosystem (Clark, 2012). Intercrops also benefit the pollinators by drawing in a diverse assemblage of insects that not only pollinate the intercrop, but also may pollinate the target crop as well (Jones & Gillette, 2005).

Phylogeny and biology of sunflower

Sunflowers are native to North America and were first domesticated and cultivated as food and for plant dyes long before European settlers came to the area (Schneiter, 1997). The Europeans took interest in the plant and cultivated their own varieties in the early 1500s and the Russian variety was marketed in the 1800s then returned back to the United States as an oilseed crop capable of producing large quantities of viable seed (Charles, 2012).

Sunflower is a colorful plant in the Asteraceae family. Different varieties will grow to different heights, but mature plants can be as short as 50 cm while larger cultivars will grow to over 500 cm (Schneiter, 1997). The sunflower is a composite flower that develops a characteristic capitulum, or a seed head containing many small clusters of florets. The outer ray florets are the first to develop and the inner disk florets will develop in rows after the ray florets open (National Sunflower Association). Sunflower seeds will develop within the disk florets. Without proper fertilization, the seeds will not develop (Heiser, 1978).

Pollination requirements

Sunflowers are annuals that are either open pollinated or cross pollinated by insects. The pollen is heavy and sticky so that multiple bee species are able to easily collect and transport large amounts (Parker, 1981b). Sunflowers are not self-compatible under natural conditions, meaning that outcrossing of pollen is required to ensure healthy viable offspring are produced (Schneiter, 1997). Through plant breeding, seed companies have made it easier for the plants to be compatible, but studies show that pollination assistance by bees is still likely to increase seed set and promote healthy propagation (Parker, 1981a, b).

Cover Cropping

Cover crops are sometimes known as short term crop rotations that serve as an addition to the main cash crop of a field (Reeves, 1994). In temperate areas, such as Missouri, cover crops are mainly used for the winter season with small grains and legumes being the most commonly used.

According to Sustainable Agriculture Research and Education (SARE), cover crops are useful for a variety of agricultural needs (Clark, 2012). They are helpful in slowing soil erosion, maintaining soil nutrients and moisture, reducing weed growth, and increasing diversity of plants and animals within the system (Clark, 2012, Snapp et al., 2004).

The use of cover crops to increase plant yields has also been studied. In their 2012 cover crop analysis survey, growers reported corn had a 9.6% increase in yield and soybean had an 11.6% yield increase when planted after the use of a cover crop (Clark, 2012). In another study of soybeans, a crop that is primarily self-pollinating, it has been shown that pollinators are still able to increase yield outputs. Those soybeans exposed to pollinators versus plants that were isolated from the presence of insects were found to show yield improvements (Milfont et al., 2007).

While the use of cover crops has shown benefits, it is important to understand which cover crops are appropriate for the needs of the individual grower. It is also important to know which type of cover cropping may be most beneficial (Rao & Stephen, 2010). Traditionally, cover crops are planted in the “off season” to protect the landscape until the following growing season when the target crop will be planted (Reeves, 1994). Another type of cover crop system is known as intercropping. In this technique, plants are grown alongside the target crop during the main growing season (Lithourgidis et al., 2011). Depending on the species used, the cover crop can be harvested and sold for profit or alternatively serve as a supplement to boost yields and decrease costly inputs into the land (Clark, 2012).

Partridge Pea

Partridge pea, *Chamaecrista fasciculata* Michx (Fabaceae), is native to the southern United States and is grown as a cover crop to help control soil erosion and fix soil nitrogen (Houck & Row, 2006). It is a wildflower that is often grown as an ornamental and can also be a beneficial pioneer species that grows and seeds quickly in disrupted areas (Marcy & Martin, 1986).

Partridge pea is an annual legume that can grow between 0.2 and 0.9 meters tall (Marcy & Martin, 1986, Harshbarger & Perkins, 1971). Yellow flowers grow in clusters and bloom indeterminately through the growing season between early June and late September. The flowers are bright, and the nectaries located near the base of the plant attract many insect species, especially bees (Marcy & Martin, 1986). It is known as an essential honey plant, meaning that honey bees frequent these flowers to gain resources that will create high quality honey, and it also grows in areas where other honey plants may not grow, including road sides and riverbanks (Smith, 2006).

Partridge pea is one of the major food sources for game birds in the Midwest including bobwhite quail, prairie chickens, mallard ducks, and pheasant (Harshbarger & Perkins, 1971). Partridge pea may also be suitable as a food source for white tail deer, but is not recommended for livestock as it contains toxic cathartic substances, which may poison cattle (Houck & Row, 2006).

Buckwheat

Fagopyrum esculentum Moench (Polygonaceae), known as buckwheat, is most commonly grown as a grain but is often used as a cover crop that attracts beneficial

insects (Björkman et al., 2008). It is a short plant mainly used as a ground cover that grows up to approximately one meter tall and can spread up to two meters in area. Buckwheat is native to Asia but has been cultivated in the Americas since its introduction in the early 16th century. Buckwheat has the ability to establish in disturbed environments, giving it a wide range across the United States (Clark, 2012). The seed is converted to flour for human consumption, the foliage is food for small herbivorous mammals or can be used as green manure, and the nectar is attractive to ants and bees (Oplinger et al., 1989). Buckwheat honey is highly valued for its deep color, viscous texture, and robust flavor. After the growing season, buckwheat stem can be mulched to fortify the soil for the next year (Clark, 2012).

Pollinators

Importance of native bees and flies as pollinators

Honey bees (*Apis mellifera*) were brought to the Americas by European settlers to make honey and pollinate the food crops that were brought from the old world. Honey bees are valuable because their pollination service contribution to society is estimated to be worth between \$1.6 and \$5.7 billion annually (Southwick & Southwick, 1992). However, their populations are declining and the need for research in pollination ecology is growing (Oldroyd, 2007).

Honey bee health is of major concern to agronomists, entomologists, and conservationists alike. The challenges that honey bees face such as Colony Collapse Disorder, pesticide usage, infestations of varroa mites, and climate changes contribute to the decline of honey bees in much of the United States (Ellis & Munn, 2005). It is

therefore important not only to focus on honey bees, but to shift focus to the native bee species that have existed in the Americas for centuries (Kremen et al., 2002). It is speculated that native bees provide the same quality and frequency of pollination services as honey bees. In watermelon crops, access by pollinators was manipulated to allow plants to be pollinated by honey bees and native bees or just native bees alone. The study found that the native bees were over 90% effective at pollinating the watermelon plants compared to the treatment with both honey bees and native bees, which pollinated at a frequency of 100% (Winfrey et al., 2007). Therefore, it is necessary that we focus on native bee health as well as honey bee health.

Unfortunately, there are documented declines in North American bumble bee (Apidae) populations as well. A study from the University of Illinois found that the abundances of four different *Bombus* species have declined by over 95% in North America within the last two decades. The reason for these declines has not been heavily studied, but it is known that the *Bombus* in decline have a greater susceptibility to pathogens and have decreased genetic diversity within the sampled populations (Cameron et al., 2010). Not all native bees are equal in their pollination services though. It was found that bees with increased sociality are not as able as solitary species to pollinate in non-natural habitats (Ricketts et al., 2008). Understanding plant and pollinator associations is key in the conservation of pollination services.

Notable Missouri species

Many bee species are present in the agricultural landscapes of Missouri. A few of the most common include the honey bee (*Apis mellifera* L.) and other *Apis* species. In

the same family (Apidae) are the cuckoo bees (*Nomada* and *Triepeolis spp.*), long horned bees (*Mellissodes spp.*), carpenter bees (*Xylocopa spp.*) and the bumble bees (*Bombus spp.*). In Missouri, *Bombus impatiens* Cresson and *Bombus griseocollis* De Geer are both found in rather high abundance. *Bombus pensylvanicus* De Geer, another bumble bee species in Missouri, has been on the decline in recent years (Arduser, 2016). Other families of bees found in Missouri agriculture systems include Megachilidae, Andrenidae, Halictidae, and Colletidae.

The honey bee and native bee interaction

Both honey bees and native bees coexist in the United States, and in Missouri habitats there are many species that interact (Arduser, 2015a). Their levels of sociality and behavior differ, but the importance of all bees as a whole is invaluable to crop production. Before the importation of the European honey bee, native bees of the Americas were responsible for much of the pollination required to produce adequate food. Native bees work in conjunction with honey bees to pollinate all of the necessary crops, so it is incorrect to say that either group takes precedence over the other (Thomson, 2004). It is interesting to note the behavioral differences between the societal levels of bee groups (Woodcock et al., 2013). The eusocial honey bees will forage differently than the solitary bees or bees of lower levels of sociality, like the bumble bees (Ricketts et al., 2008).

Competition for resources also exists among different species of bees. Honey bees that forage on *Agave schottii* are known to choose patches of the highest quality, thus reducing the availability of nectar and foraging sites for other bee species. By

removing the managed honey bees that visit *A. schottii*, the number of feral honey bees and solitary bees increased (Schaffer et al., 1983).

Sampling methods

Some of the most common techniques for collecting pollinators in row cropping systems include the active methods of sweep net sampling and passive methods such as bee bowls and pan traps (Gill et al., 2015). Sweep nets can be useful to sample high numbers of insects in row crops without causing too much damage to the plant itself. With this method, many insect types are captured and the sampling technique is not exclusive to bees (Grundel et al., 2011). For our research, using sweep nets to sample in sunflower plots is not feasible. The height and density of the plants in the field were not conducive to sweep net sampling practices. Hand netting is an alternative method of active sampling but limits the collector's ability to sample in high quantities in a limited amount of time. It is then more useful to have passive traps set in a field to collect insects that may be active throughout the day (Cane et al., 2000).

Bee bowls are the most common traps used to collect pollinators. They are bright florescent bowls painted white, yellow, or blue as an attractant. They are filled with water and a small amount of detergent to keep the bees that land in the bowl from escaping (Arduser, 2015b). This method, while effective, ultimately kills the bee and when the bowl is full, no other insects are able to be caught.

Another way to trap bees, flies, and other insects is by using bucket traps, specifically blue vane traps. This is another passive method of sampling where traps are hung within the field at various heights and distances to collect insects flying through

the cropping system (Stephen & Rao, 2007). The traps are blue to attract insects and the vanes are constructed so that bees are unable to escape once inside the trap. A benefit of this collecting method is that detergent does not have to be used, thus creating a trapping system where bees do not die if there is no solution in the bottom of the trap. This method is effective, but has its limitations as well. Only the bees that fit between vanes are able to be collected. On the other hand, some very small species have the ability to navigate their way and squeeze out of the traps (Kimoto et al., 2012).

Studies show that the most effective way to accurately sample the bee community present in an area is to use multiple methods (Gill et al., 2015, Grundel et al., 2011). This will give the collector a more thorough representation of the insect diversity and abundance of that area.

Study Objectives

My overall objective was to determine if intercrops can enhance pollinator services in sunflower fields by promoting pollinator abundance and diversity. My specific objectives were to:

- 1) Describe the pollinator communities associated with pure stands of sunflowers, buckwheat, and partridge pea.
- 2) Quantify the pollinator community and pollination services in sunflower in response to the presence of intercrops.
- 3) Quantify the pollinator community associated with the intercrop in the presence of sunflowers.

Chapter 2:

DESCRIPTION OF THE POLLINATOR COMMUNITY PRESENT IN PURE STANDS OF SUNFLOWER (*HELIANTHUS ANNUUS* L.), BUCKWHEAT (*FAGOPYRUM ESCULENTUM* MOENCH), AND PARTRIDGE PEA (*CHAMAECRISTA FASCICULATA* MICHX)

Introduction

Intercrops are often used with the goal of enhancing pollinator abundance and diversity in crops and it has been shown that crops benefit from the presence of non-crop flowers that are nearby (Jones & Gillett, 2005). For intercrops to be effective, they need to provide additional resources or diverse resources that are not found in the cash crop itself. In that way, the intercrop can enhance the abundance and diversity of pollinators in the crop system (Nicholls & Altieri, 2012).

Pollen and nectar are two of the most important floral resources for insect pollinators (Thom et al., 2016). Different species of plants produce different types, amounts, and concentrations of these resources (Eberle et al., 2015). Sunflowers, *Helianthus annuus* L. (Asteraceae), attract pollinators not only with their large size and bright coloration, but with their abundance of easily transported pollen and the availability of floral nectaries found in the sunflower head (Martins et. al., 2005). Pollinators associated with sunflower production in Missouri are not thoroughly documented, but it is known that honey bees, a variety of bumble bees, and some fly species are common visitors to sunflowers in other production regions (Arduser, 2016).

Buckwheat, *Fagopyrum esculentum* Moench (Polygonaceae), a common plant grown in Missouri, is a grain plant often used specifically to attract honey bees (Clark,

2012, Bartomeus et al., 2014). The flowers are small and delicate with five white petals each. While buckwheat is known to be a good food source of pollen and nectar for honey bees, other insects rely on these same resources as well (Taki et al., 2009). Many types of insects eat nectar as it is rich in carbohydrates, amino acids and provides energy (Gardener & Gillman, 2002). Buckwheat is a short plant that only grows to about 30-40 cm tall.

Partridge pea, *Chamaecrista fasciculata* Michx (Fabaceae), is another bee attracting plant that is grown as a cover crop and as a soil nitrogen enhancer (Marcy & Martin, 1986). Partridge peas grow to about one meter tall and produce clusters of bright yellow flowers (Clark, 2012). Some of the insects that visit partridge pea include ants, parasitoid wasps, and honey bees (Morris, 2012). Other plants in the Fabaceae family are known to be visited by bees in the families Andrenidae, Megachilidae, and Anthophoridae (Bohart, 1960).

This study described the pollinator community present in pure stands of sunflower, buckwheat, and partridge pea to determine if different taxa are found. The presence of distinct taxa in the different pure stands provides evidence that intercrops could be an effective way of enhancing pollinator diversity. Given the differences in flower appearance and structure and the quality of resources among plants, we predict that there will be observed differences in the pollinator communities that forage in pure stands of sunflower, buckwheat, and partridge pea.

Materials & Methods

We compared the pollinator communities associated with three pure stands of sunflower (*H. annuus* L.), buckwheat (*F. esculentum* Moench), and partridge pea (*C. fasciculata* Michx). Replicate plots (3m x 3m) were established in the summer of 2014 at Bradford Research Center at the University of Missouri. Sunflowers were planted in four, 30 inch rows sown from east to west. Buckwheat and partridge pea were planted by hand in densities recommended by Foundation Seed at the University of Missouri (2 lbs. /acre). Each plant type was replicated four times for a total of 12 plots. The entire 12 plot treatment design was replicated at three different planting times throughout the growing season. The first planting took place the week of May 25, 2014, the second the week of June 15, 2014, and the third the week of July 20, 2014.

The pollinator community was monitored in the three plantings throughout the sunflower growing period. The plots were visually scouted and the identity and abundance of pollinators present in each plot were documented. Five minutes were spent in each plot during each scouting period and any pollinators that landed on a flower were identified to family. Those in the family Apidae were further identified to *Apis mellifera* or *Bombus spp.* Each plot was scouted during peak foraging times in the morning and afternoon, between the hours of 7:00 AM-10: 00 AM and again 2:00 PM-5:00 PM, for a total of three days. The first day of scouting took place during the pre-peak bloom stage of sunflower floral development. The next scouting day occurred during peak bloom for sunflower. The last scouting date was to monitor for pollinators that may still be foraging post bloom when the sunflowers and buckwheat were

senescing. Due to a low number of observed pollinators, observations were summed across replicates, observation dates, and planting dates for each plant type, and pollinator taxon richness and abundance were compared in a qualitative manner. Observations of pollinators were not consistent enough across treatments to allow statistical analysis. The data collected were used to identify any patterns in bee richness, abundance, and identity that exists among pure stands of each flower type.

Results

Unfortunately, partridge pea did not bloom during the growing period of the sunflower and so there were no pollinators present in the partridge pea in any of the observations. Partridge pea, while being an indeterminately blooming plant, takes longer to reach peak bloom than both sunflower and buckwheat. Sunflower and buckwheat had reached full bloom, or a reproductive growth stage of approximately R6/R7 before the partridge pea had time to complete its vegetative growth stage.

Six taxa from 5 different families were found in the buckwheat and sunflower plots (Table 1). The majority of the bee taxa were in the family Apidae, which includes honey bees, bumble bees, and cuckoo bees. The other families observed were Colletidae, Halictidae, and Tenthredinidae. The only observed fly taxon was the family Syrphidae, or the flower flies.

Taxon richness for both the pure stands of sunflower and buckwheat was the same, but the bee community differed both in total abundance and composition (Table 2). Over twice as many individual bees were present in sunflower than were present in buckwheat. For instance, the abundance of honey bees (*Apis mellifera*) in sunflower (80

individuals) was much larger than the number of individuals found in the buckwheat (36 individuals). There were also taxa that were exclusive to sunflower or buckwheat plots. For example, bees in the family Colletidae were found in sunflower but not found in buckwheat and a Tenthredinidae individual was observed foraging in buckwheat but not in sunflower.

Discussion

Pollinator abundance varied across plant types with sunflowers having a higher abundance of pollinators than buckwheat. I was unable to quantify pollinator abundance in the partridge peas because partridge pea plots had not yet reached the reproductive stage of floral production by the time visual observations were collected. The differences found in pollinator abundance between the pure stands of sunflowers and buckwheat is likely due to a variety of factors. Pollinators may have been found in higher numbers in the sunflower plots than in plots containing buckwheat because of the visibility and density of flowers available (Dafni & Kevan, 1997). The sunflowers grew to be quite large both in height and head diameter, meaning there was greater surface area to be found by foraging pollinators. The buckwheat, a short ground cover type plant, only grows to be around 300 mm in height and the florets on each stem are small and sparse. In a study by Dafni and Kevan (1997), smaller pollinators were found to be attracted to different morphological flower characteristics than were larger pollinators. Perhaps by understanding the variability of morphological characteristics between each of the three crops, we can better understand the pollinator community associated with each.

The diversity of pollinators found in sunflower and buckwheat, while not dramatically different from one another, does give rise to more research questions. Perhaps the benefit of adding buckwheat to a crop in the form of an intercrop could solely be to provide resources in the form of more nectar available. Even if a buckwheat intercrop does not increase the taxon richness of pollinators in sunflower cropping systems, it may still benefit yield by enhancing the overall abundance of bee pollinators that frequent sunflowers. Taki et al. (2009), found that over 42% of the observed pollinators of buckwheat were *Apis mellifera*. This information, though taken from a different geographic location than our study site, shows a similar pattern to what the pollinator community was like in our buckwheat plots. Partridge pea may also have a similar effect, but the identity of the bee community in this intercrop is unknown. The information gained through this preliminary experiment serves as a basis to develop further studies. We can use this information to help understand how to enhance abundance and diversity of pollinators in a sunflower cropping system, whether that be by finding alternative intercrops or by composing an intercrop cocktail that uses multiple intercrops that attract diverse assemblages of insect taxa that may be most beneficial to boosting sunflower yields.

Table 1. Pollinator taxa present in pure stands of sunflower (*Helianthus annuus* L.) and buckwheat (*Fagopyrum esculentum* Moench) across four replications at three different plantings throughout the growing season of summer 2014.

	Apidae		Colletidae	Halictidae	Tenthredinidae	Syrphidae
	<i>Apis mellifera</i>	<i>Bombus</i>				
Sunflower	80	6	4	21	0	2
Buckwheat	36	1	0	30	1	8

Table 2. Total pollinator taxon richness and abundance in pure stands of sunflower (*Helianthus annuus* L.) and buckwheat (*Fagopyrum esculentum* Moench) across four replications at three different plantings throughout the growing season of summer 2014.

	Abundance	Taxon richness
Sunflower	230	5
Buckwheat	103	5

Chapter 3:

ABUNDANCE AND DIVERSITY OF BEE POLLINATORS IN *HELIANTHUS ANNUUS* L. IN RESPONSE TO INTERCROP TREATMENTS

Introduction

Sunflower production is a growing industry and factors that increase yields are important to growers. Using cover crops has been shown to increase the presence of pollinators in sunflower (Ellis & Barbercheck, 2015). Buckwheat (*Fagopyrum esculentum*) and partridge pea (*Chamaecrista fasciculata* Michx), both common cover crops in Missouri, are especially effective at attracting bees and other pollinators as they offer pollen and nectar that are valuable food sources for foraging insects (Taki et al., 2009). Though cover crops are often grown in the off season, one type of cover cropping called intercropping allows for a diversity of plants being sown side by side during the growing season (Clark, 2012). The thought behind this is that by increasing floral diversity within a field, a grower will also be increasing beneficial insect diversity due to the availability of multiple floral resources (Mallinger et al., 2016).

Increases in pollinator abundance also correlate with higher seed set in sunflowers. Chambo et al. (2011) found a 43% increase in sunflower seed set in sunflowers that were openly visited by pollinators than flowers that were pollinator limited. Also, in a sunflower production system, it has been shown that crop yields can be negatively impacted by a reduction in pollinator services (Chamer et al., 2015).

We predict that there will be an increase in both the abundance and the richness of pollinator taxa visiting sunflower when we combine both buckwheat and partridge

pea as intercrops in a sunflower cropping system. The combination of two intercrops will also increase sunflower yields and boost seed set in the sunflowers planted alongside both intercrops as opposed to the sunflowers that are only planted with one type of intercrop or no intercrop at all.

Materials & Methods

Experimental design

My field site was located at Bradford Research Center at the University of Missouri. I manipulated the presence of two intercrops in association with sunflower in a factorial design and measured the response of the community of pollinators on sunflower as well as sunflower yield. All observations were collected during the summer of 2015 between August and October. The experiment was repeated once during the summer to account for any variations that may occur at different points during the growing season.

One of four treatments was assigned to each of the plots. Each treatment had three replications for a total of 12 plots during each of the two plantings. Sunflowers were present in all plots and the intercrop treatments were a 2x2 factorial manipulation of the presence or absence of buckwheat and presence or absence of partridge pea. The first treatment was the control, a plot planted with only sunflowers. The next treatment contained sunflowers as well as a buckwheat intercrop. The third treatment had sunflowers and partridge pea as the intercrop, and the final treatment was a combination of both buckwheat and partridge pea planted with sunflowers.

The plots were 3.6 m X 3.6 m with 3.6 m alleys of bare soil between plots (Fig. 1). Sunflowers were grown in two, 30 inch (approximately .75 meter) rows (Fig. 2) in the center of the plot from east to west (Fig. 3). 12- 15 sunflower seeds were planted approximately 0.15-0.20 m apart. Intercrops were planted in the space at the southern end of the plots (Fig. 4). For the control, two more rows of sunflower were planted to the south alongside the center rows. For the buckwheat treatment, buckwheat seeds were broadcast in a 1.2 m x 3.6 m space at a density of 5 lbs. /acre (Fig. 5). For the partridge pea treatment, seeds were broadcast in a 1.2 m x 3.6 m space at a density of 2 lbs. /acre (Fig. 6). In the plots containing the buckwheat + partridge pea treatment, the 1.2 m x 3.6 m space was divided into four equal sections that were perpendicular to the sunflowers. Each section was randomly assigned either buckwheat or partridge pea so that two of each of the four sections contained the same intercrop type.

Visual observations

The plots were scouted to document the types and abundance of pollinators visiting sunflower. Two linear meters of sunflowers were observed for six minutes during each observation period and any pollinators that landed on a plant were identified to the lowest taxonomic unit possible. Observations were recorded in the morning hours between 7:00 and 9:00 AM when bee foraging was likely to occur. Each plot was observed for three days during pre-peak sunflower bloom and three days at peak sunflower bloom. The main and interactive effects of buckwheat presence, partridge pea presence, and bloom period on pollinator abundance and taxon richness

were determined using a 3 way factorial ANOVA with planting date included as a random block factor (PROC MIXED, SAS v.9.4).

Blue vane traps

Blue vane traps were used to supplement the visual data observations. The traps were placed on fence posts at a height of 1.5 m in the center of each plot where the sunflowers and intercrop converged. Since sunflowers were expected to grow between 1.25-1.60 m, the trap was placed to capture any pollinators that were flying at the corresponding height. The traps were set out for 48 hours after pre-peak visual observations were recorded and again for another 48 hours after peak visual observations were recorded. The pollinators in the traps were sorted to family and some were sorted further into genera and species. For analysis, the total of all bees found in the blue vane traps, the main and interactive effects of buckwheat presence, partridge pea presence, and bloom period on pollinator abundance and taxon richness were determined using a 3 way factorial ANOVA with planting date included as a random block factor (PROC MIXED, SAS v.9.4).

Harvest

Once the sunflowers were fully mature and seeds had filled the entire capitulum, plant height was measured and the heads of three random plants from each plot were removed, labeled, and sealed in plastic bags. The diameters of the heads were then measured and all of the seeds were counted. The number of seeds was standardized by head area (number of seeds/cm²). The main and interactive effects of buckwheat

presence and partridge pea presence on sunflower seed set were determined using ANOVA (PROC MIXED, SAS v.9.4).

Results

Visual observations

There was an interactive effect of intercrop presence on the abundance of pollinators visiting sunflowers ($F_{1, 37} = 11.58, p \leq 0.002$, Table 3). All intercrops increased the abundance of pollinators on sunflower. The greatest number of total individuals was observed in plots containing partridge peas. When buckwheat was added to a plot, there was also an increase in the number of visitors to sunflowers. However, the positive effect of partridge pea is reduced when both buckwheat and partridge pea were added to the sunflowers (Fig. 7).

There was a significant interactive effect of the presence of intercrops on pollinator taxon richness in sunflower ($F_{1, 37} = 5.66, p \leq 0.02$, Fig. 8). Partridge pea increased the number of pollinator taxa that were found on sunflowers. However, the addition of buckwheat and partridge pea together had no effect on the total number of taxa, thus leading to the same taxon richness that is seen in the control plots. The taxon richness of pollinators visiting sunflowers in the buckwheat treatment was intermediate and shows that the addition of buckwheat again negated any positive effect of partridge peas to enhance taxon richness in sunflower.

Pollinators from 5 families of Hymenoptera and 1 dipteran family were observed foraging on sunflower heads (Table 4). The most abundant pollinator present in all treatments were Apidae, with *Bombus* being in higher abundance than *A. mellifera*. The

pollinator present at the second greatest density was Colletidae. Interestingly, Andrenidae and Megachilidae were only observed foraging on sunflowers if an intercrop was present, although the numbers observed were low.

While Colletidae is shown to be in great abundance, Apidae appears to have the greatest abundance in treatments containing partridge pea (Table 5). When species of Apidae that were observed were summed, the sunflower + partridge pea treatment had 193 Apidae and 137 Colletidae while the sunflower + buckwheat + partridge pea treatment has 153 Apidae and 119 Colletidae. Other families were in much lower abundance.

Blue vane traps

There was a main effect of buckwheat on the abundance of pollinators found foraging on sunflowers ($F_{1, 40} = 4.37, p \leq 0.04$, Table 6). The abundance of bees found in the control plots did not differ from the abundance of bees found in plots containing the partridge pea only treatment (Fig. 9). However, in the buckwheat only treatment as well as the partridge pea + buckwheat treatment, bee abundance declined relative to the control. The addition of buckwheat decreased the number of pollinators that were collected in the blue vane traps when placed 1.5 m above ground, roughly the same height as the sunflowers. Taxon richness of the various pollinators found in the blue vane traps did not vary among the four different treatments (Fig. 10).

Harvest

There was a main effect of the presence of buckwheat on sunflower seed set ($F_{1, 63} = 6.24, p \leq 0.05$, Table 7). The number of seeds per unit area of sunflower head did not

differ between the control treatment with no intercrop and the partridge pea only (Fig. 11). However, seed set was higher in treatments that contained buckwheat.

Discussion

The addition of intercrops into a sunflower cropping system has the potential to impact the abundance and diversity of pollinators present in the field (Jones & Gillette, 2005). Increases in pollinator abundance have also been shown to increase seed set in sunflowers (Chambo et al., 2011, Chamer et al., 2015). However, our results were not consistent with those previous studies. We observed that partridge pea does increase pollinator abundance and taxon richness, but has no effect on sunflower seed set.

Buckwheat, on the other hand, does not have a positive effect on the abundance and richness of insect pollinators visiting sunflower crops, but does enhance seed set.

Creating floral diversity in a field has been shown to increase the yield of target crops in many different agricultural settings (Mallinger et al., 2016). In our experiment, we see that the addition of two intercrops together, buckwheat and partridge pea, into a sunflower cropping system does not directly increase yield of sunflower seeds. As mentioned, buckwheat does not increase the abundance or diversity of pollinators in a sunflower field, but it does play an important role in enhancing seed set of sunflowers. Alternatively, while partridge peas have been shown to increase the numbers and community richness of pollinators found foraging on sunflowers in an intercropping system, there is no evidence to show that the addition of partridge pea is important for sunflower seed set and does not increase sunflower yields. This could potentially be due to the fact that different insects are attracted to different flowers based on the

resources they provide. Since nectar from different plants contains different concentrations of essential nutrients, it is possible that insects visiting the partridge pea were not looking for the same resources as those visiting buckwheat or the sunflower (Gardener & Gillman, 2002).

In my previous chapter I discussed the differences between sunflower pollinator communities and buckwheat pollinator communities. Those same patterns are seen again when buckwheat is planted as an intercrop alongside sunflowers. Pure stands of sunflower host 5 of the 7 pollinator taxa observed in all plots, but sunflowers planted with a buckwheat intercrop host all of the insect families observed. However, it is interesting to note that the addition of partridge pea into the sunflower + buckwheat system reduces the number of observed insect taxa. The same two taxa that were not observed in pure sunflower stands are again missing with the addition of partridge pea as a second intercrop. Coincidentally, in sunflower + partridge pea where no buckwheat is present, the number of insect taxa observed is again 10.

Now the question is why does this relationship occur? There may be interactions among pollinators occurring in the intercrop itself that were not documented in observing sunflowers alone. The possibility of other interactions at the intercrop level is further discussed in Chapter 4.

Table 3. Results of ANOVA to show the main and interactive effects of two intercrops, buckwheat (*Fagopyrum esculatum* Moench), partridge pea (*Chamaecrista fasciculata* Michx), and bloom period on richness and abundance of pollinators in visual observations.

RICHNESS					ABUNDANCE				
Effect	Num DF	Den DF	F Value	Pr > F	Effect	Num DF	Den DF	F Value	Pr > F
Bloom period	1	37	0.02	0.8856	Bloom period	1	37	0.34	0.5640
Buckwheat	1	37	3.86	0.0569	Buckwheat	1	37	0.00	0.9917
Bloom period * buckwheat	1	37	1.70	0.2004	Bloom period * buckwheat	1	37	0.13	0.7228
Partridge pea	1	37	0.27	0.6073	Partridge pea	1	37	11.27	0.0018
Bloom period * partridge pea	1	37	0.02	0.8856	Bloom period * partridge pea	1	37	0.22	0.6413
Buckwheat * partridge pea	1	37	5.66	0.0226	Buckwheat * partridge pea	1	37	11.58	0.0016
Bloom period * buckwheat * partridge pea	1	37	0.23	0.6369	Bloom period * buckwheat * partridge pea	1	37	0.01	0.9094

Table 4. Total abundance of pollinators observed visiting sunflowers (*Helianthus annuus* L.) in plots with different intercrop treatments during visual observations. Pollinator identity and abundance shown for each individual plot. SF= sunflower, BW= buckwheat, and PP= partridge pea.

Planting	Treatment	Apidae		Andrenidae	Anthophoridae	Colletidae	Megachilidae	Syrphidae
		<i>Apis mellifera</i>	<i>Bombus</i>					
1	control	17	40	0	0	9	0	2
1	control	7	42	0	1	39	0	4
1	control	0	15	0	0	20	0	0
1	SF + BW	15	10	0	0	16	0	3
1	SF + BW	6	31	5	0	42	1	1
1	SF + BW	4	24	3	0	18	0	6
1	SF + PP	20	31	3	6	32	0	2
1	SF + PP	7	53	1	0	37	3	2
1	SF + PP	4	44	0	0	19	1	2
1	SF + BW +PP	13	36	0	0	19	0	0
1	SF + BW +PP	3	33	0	2	24	0	3
1	SF + BW +PP	3	45	0	0	18	0	4
2	control	5	7	0	0	27	0	0
2	control	2	14	0	0	24	0	0
2	control	6	5	0	2	40	0	0
2	SF + BW	5	10	0	0	31	0	0
2	SF + BW	3	7	0	0	19	1	0
2	SF + BW	0	9	0	3	21	0	1
2	SF + PP	3	4	0	0	18	0	0
2	SF + PP	5	18	0	0	31	0	0
2	SF + PP	0	3	0	0	0	0	0
2	SF + BW +PP	0	6	0	0	30	0	0
2	SF + BW +PP	3	3	0	0	19	0	0
2	SF + BW +PP	2	6	0	0	9	0	0

Table 5. Abundance of pollinators observed visiting sunflowers (*Helianthus annuus* L.) in plots with different intercrop treatments across two different plantings throughout the growing season of summer 2015.

	Apidae		Andrenidae	Anthophoridae	Colletidae	Megachilidae	Syrphidae	Total
	<i>Apis mellifera</i>	<i>Bombus</i>						
Sunflower	37	108	0	3	159	0	6	313
Sunflower + buckwheat	33	88	8	3	147	2	11	292
Sunflower + partridge pea	39	154	4	6	137	4	6	350
Sunflower+ buckwheat+ partridge pea	24	129	0	2	119	0	7	281
total	133	479	12	14	562	6	30	1236

Table 6. Results of ANOVA performed on the effects of intercrops, buckwheat (*Fagopyrum esculentum* Moench) and partridge pea (*Chamaecrista fasciculata* Michx), and bloom on richness and abundance of pollinators in traps at the sunflower (*Helianthus annuus* L.) height.

RICHNESS					ABUNDANCE				
Effect	Num DF	Den DF	F Value	Pr > F	Effect	Num DF	Den DF	F Value	Pr > F
Bloom period	1	40	3.24	0.0795	Bloom period	1	40	3.54	0.0673
Buckwheat	1	40	0.94	0.3384	Buckwheat	1	40	4.37	0.0430
Bloom period * buckwheat	1	40	0.17	0.6802	Bloom period * buckwheat	1	40	0.04	0.8355
Partridge pea	1	40	0.17	0.6802	Partridge pea	1	40	0.39	0.5343
Bloom period * partridge pea	1	40	0.48	0.4929	Bloom period * partridge pea	1	40	0.04	0.8355
Buckwheat * partridge pea	1	40	0.48	0.4929	Buckwheat * partridge pea	1	40	0.17	0.6782
Bloom period * buckwheat * partridge pea	1	40	0.48	0.4929	Bloom period * buckwheat * partridge pea	1	40	0.17	0.6782

Table 7. Results of ANOVA performed on the effects of intercrops, buckwheat (*Fagopyrum esculentum* Moench) and partridge pea (*Chamaecrista fasciculata* Michx), on sunflower seed set.

Effect	Num DF	Den DF	F Value	Pr > F
Buckwheat presence	1	63	6.24	0.0151
Partridge pea presence	1	63	0.84	0.3618
Buckwheat presence * partridge pea presence	1	63	0.05	0.8242

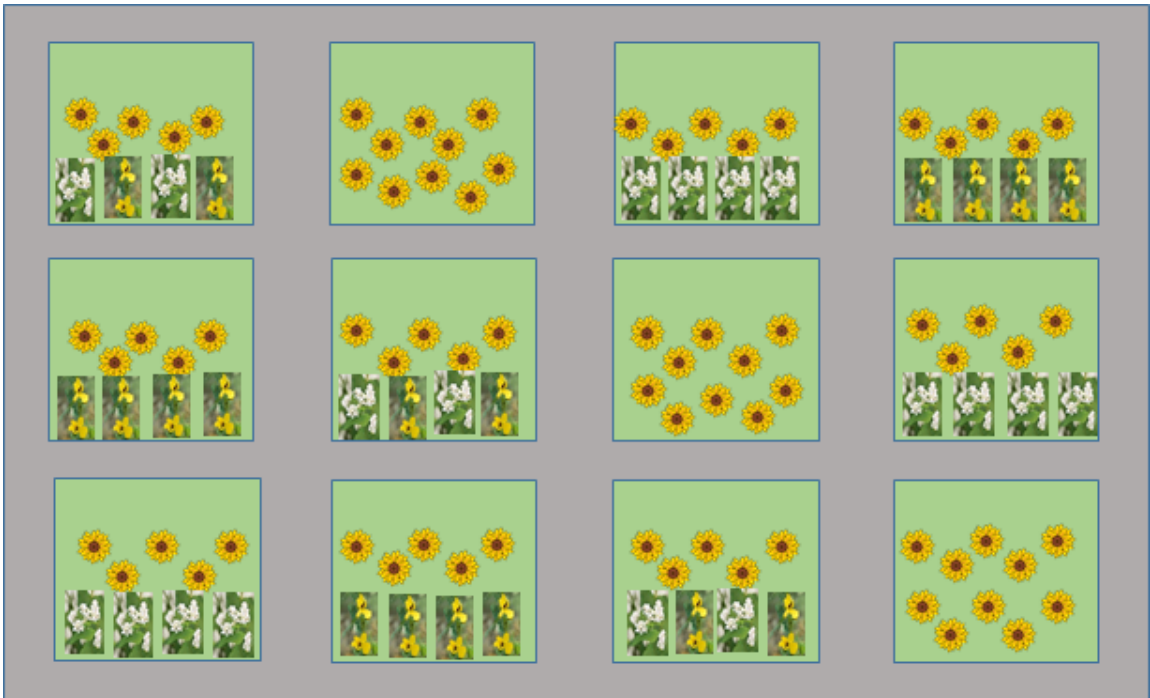


Figure 1. Plot setup at Bradford Research Farm. Sunflowers (*Helianthus annuus* L.) were planted in two rows from east to west and the southern end of each plot contains the intercrop treatment (yellow=partridge pea, white=buckwheat) that was randomly assigned (buckwheat, partridge pea, or buckwheat + partridge pea.) Plots were 3.6m x 3.6m.



Figure 2. View from the east: photograph of sunflower (*Helianthus annuus* L.) plots at peak bloom. Sunflowers planted in two rows from east to west on the northern half of the plot with intercrop treatment planted to the south.



Figure 3. View from the east: photograph of treatment plots. Sunflowers (*Helianthus annuus* L.) grown in two, 30 inch rows on north side of treatment plots with vane trap located in the middle of each plot.



Figure 4. View from the south: photograph of plot containing partridge pea + buckwheat treatment. Strips of intercrops planted perpendicular to sunflower rows.



Figure 5. Photograph of buckwheat (*Fagopyrum esculentum* Moench) prior to bloom. Buckwheat planted at a density of 5 lbs. /acre in treatment plots.



Figure 6. Photograph of partridge peas (*Chamaecrista fasciculata* Michx) at peak bloom. Partridge peas planted at a density of 2 lbs. /acre in treatment plots.

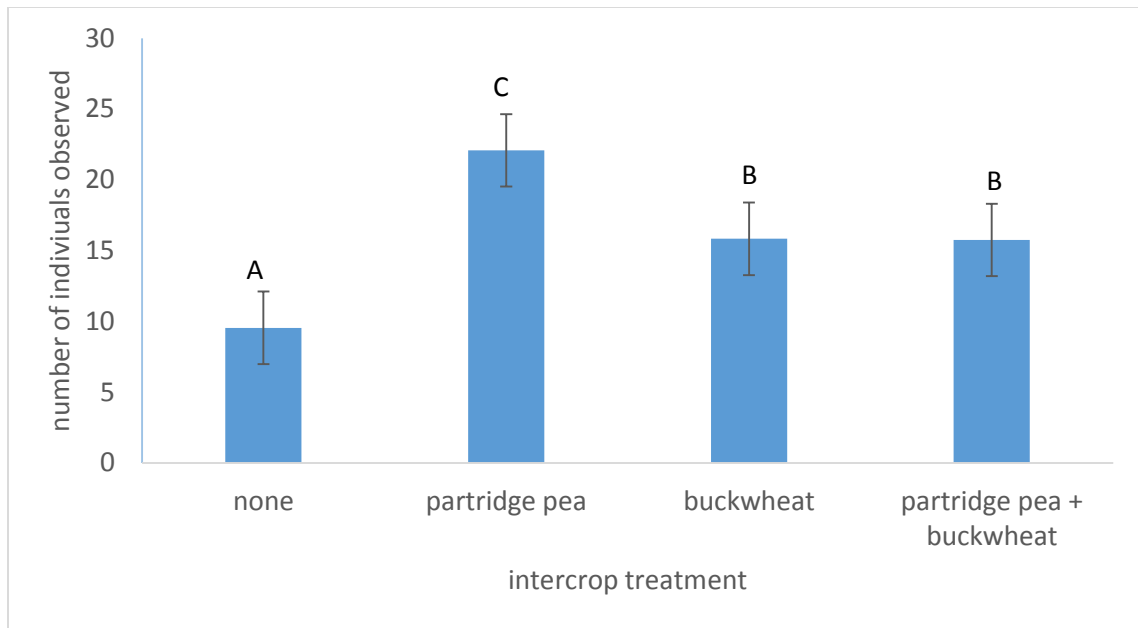


Figure 7. Total abundance of the pollinators observed foraging on sunflowers (*Helianthus annuus* L.) in response to each intercrop treatment during 10 minute visual observations. LS means \pm 1 SE shown. Means with different letters are significantly different at $p \leq 0.05$.

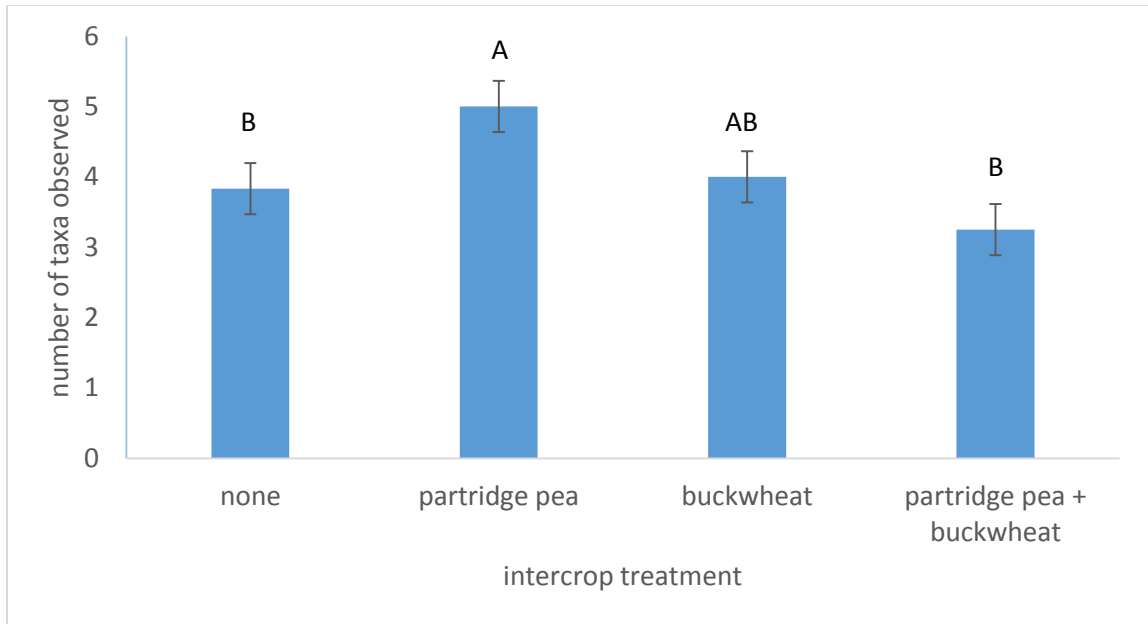


Figure 8. Total taxon richness of the pollinators observed foraging on sunflowers (*Helianthus annuus* L.) in response to each intercrop treatment during 10 minute visual observations. LS means \pm 1 SE shown. Means with different letters are significantly different at $p \leq 0.05$.

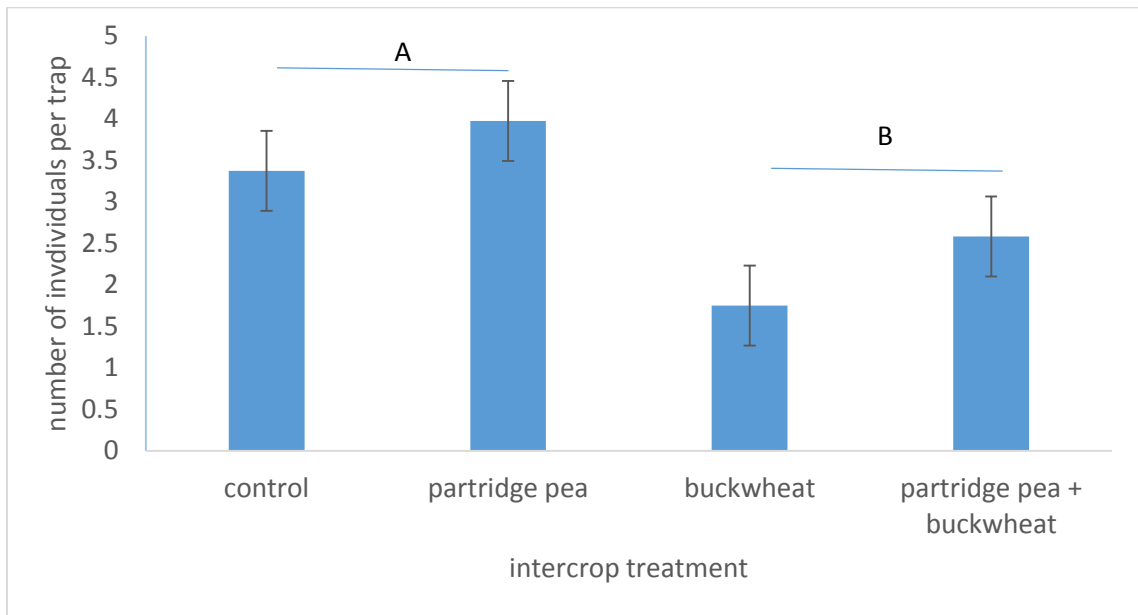


Figure 9. Total abundance of the pollinators observed in blue vane traps placed at the height of sunflower (*Helianthus annuus* L.) heads. Traps deployed for 48 hours during the summer. LS means \pm 1 SE shown. Lines over bars indicate the main effect of intercrop treatment. Lines with different letters are significantly different at $p \leq 0.05$.

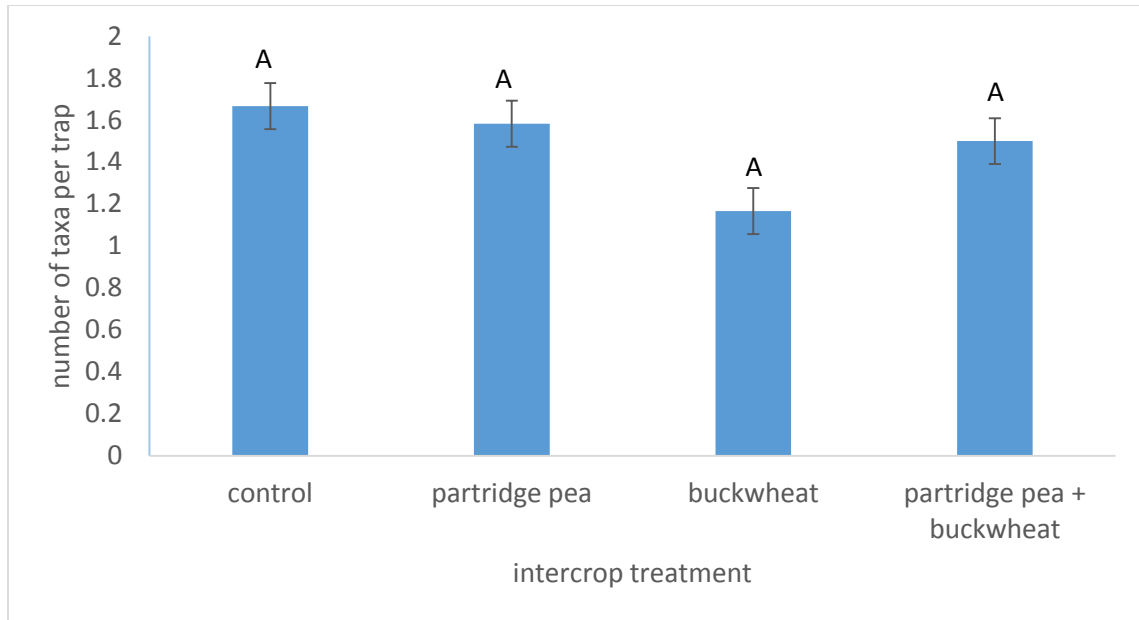


Figure 10. Total taxon richness of the pollinators observed in blue vane traps placed at the height of sunflower (*Helianthus annuus* L.) heads. Traps deployed for 48 hours for during the summer. LS means ± 1 SE shown. Means with different letters are significantly different at $p \leq 0.05$.

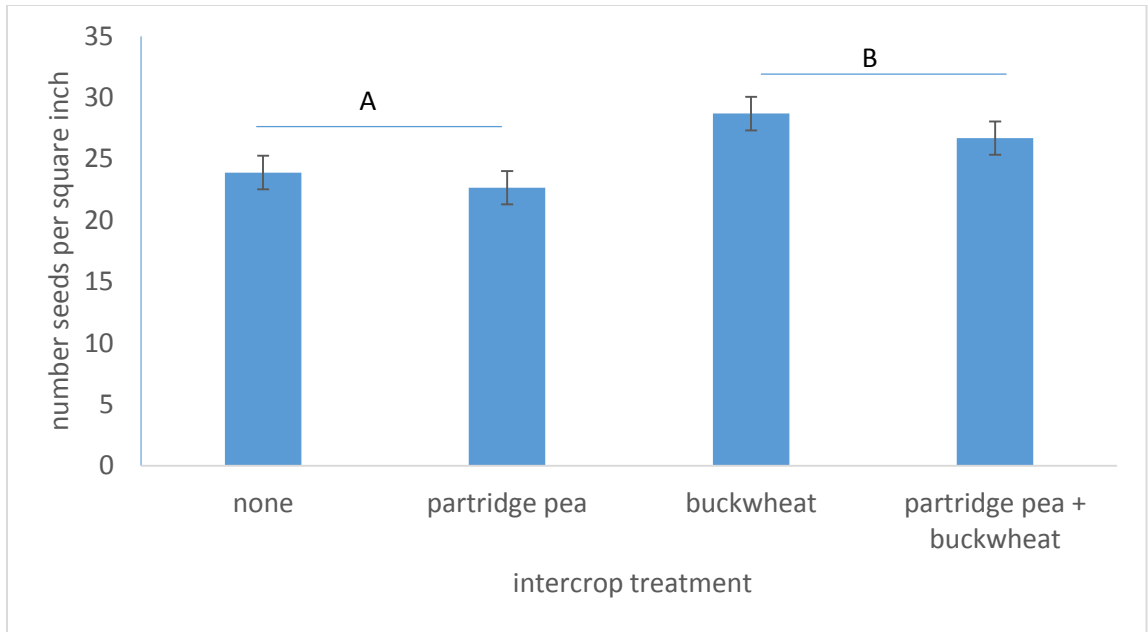


Figure 11. Seed set of mature sunflowers (*Helianthus annuus* L.) in response to intercrop treatment. Average seed set calculated as the number of seeds present per square inch of sunflower head area. LS means \pm 1 SE shown. Lines over bars indicate the main effect of intercrop treatment. Lines with different letters are significantly different at $p \leq 0.05$.

Chapter 4:

ABUNDANCE AND DIVERSITY OF INSECT POLLINATORS IN TWO INTERCROPS ASSOCIATED WITH SUNFLOWER (*HELIANTHUS ANNUUS* L.): BUCKWHEAT (*FAGOPYRUM EXCULENTUM* MOENCH) AND PARTRIDGE PEA (*CHAMAECRISTA FASCICULATA* MICHX)

Introduction

Using cover crops has been shown to increase the presence of pollinators in cover crops such as red clover, canola, and winter pea (Ellis & Barbercheck, 2015). By creating habitat diversity for pollinators, crop yields are increased through the diversification of functional niches that pollinators inhabit (Hoehn et al., 2008). Buckwheat (*Fagopyrum esculentum* Moench) and partridge pea (*Chamaecrista fasciculata* Michx), both common cover crops in Missouri, are especially good at attracting bees and other pollinators as they offer pollen and nectar that are valuable food sources for foraging insects. Though cover crops are often grown in the off season, one type of cover cropping called intercropping allows for a diversity of plants to be sown side by side during the growing season. The thought behind this is that by increasing floral diversity within a field, a grower will also be increasing diversity of beneficial insects due to the availability of multiple floral resources (Bartomeus et al., 2014, Mallinger et al., 2016).

Since it is known that floral diversity increases the abundance and diversity of insects in agroecosystems (Nicholls & Altieri, 2012), the community of pollinators is likely to be different in the intercrop than it is in the cash crop. In a study that compared the bumble bee diversity between grass mixtures and crop mixtures containing grass

plus a cereal grain and a legume, bee diversity and abundance was greater than in the grass mixture alone (Potts et al., 2009). In this study, we examine how the identity and diversity of the intercrop planted alongside sunflowers influences the pollinator community visiting the intercrop. We predict that the community of pollinators attracted to buckwheat and partridge pea will differ and there will be an increase in both pollinator abundance and taxon richness when we combine both buckwheat and partridge pea as intercrops in a sunflower cropping system. Ideally, the combination of the two intercrops will be to benefit pollinator richness and abundance in a sunflower cropping system.

Materials & Methods

Experimental design

My field site was located at Bradford Research Center at the University of Missouri. I manipulated the presence of two intercrops in association with sunflower in a factorial design and measured the response of the community of pollinators on sunflower as well as sunflower yield. All observations were collected during the summer of 2015 between August and October. The experiment was repeated once during the summer to account for any variations that may occur at different points during the growing season.

One of four treatments was assigned to each of the plots. Each treatment had three replications for a total of 12 plots during each of the two plantings. Sunflowers were present in all plots and the intercrop treatments were a 2x2 factorial manipulation of the presence or absence of buckwheat and presence or absence of partridge pea. The

first treatment was the control, a plot planted with only sunflowers. The next treatment contained sunflowers as well as a buckwheat intercrop. The third treatment had sunflowers and partridge pea as the intercrop, and the final treatment was a combination of both buckwheat and partridge pea planted with sunflowers.

The plots were 3.6 m X 3.6 m with 3.6 m alleys of bare soil between plots. Sunflowers were grown in two, 30 inch (approximately .75 meter) rows, apart in the center of the plot from east to west. 12- 15 sunflower seeds were planted approximately 0.15-0.20 m apart. Intercrops were planted in the space at the southern end of the plots. For the control, two more rows of sunflower were planted to the south alongside the center rows. For the buckwheat treatment, buckwheat seeds were broadcast in a 1.2 m x 3.6 m space at a density of 5 lbs. /acre. For the partridge pea treatment, seeds were broadcast in a 1.2 m x 3.6 m space at a density of 2 lbs. /acre. In the plots containing the buckwheat + partridge pea treatment, the 1.2 m x 3.6 m space was divided into four equal sections that were perpendicular to the sunflowers. Each section was randomly assigned either buckwheat or partridge pea so that two of each of the four sections contained the same intercrop type.

Visual observations

Visual observations of pollinators foraging on the intercrop plants were only possible in the three treatments where intercrops were present (buckwheat, partridge pea, and buckwheat + partridge pea). One square meter of the intercrop was observed for six minutes and the number of individuals visiting the intercrop flowers was recorded as well as identity of the pollinators. The sample area was determined for the

buckwheat and partridge pea treatments by randomly placing a 1m² frame into the intercrop. For the combination of buckwheat + partridge pea treatment the frame was centered over the border between the buckwheat and partridge pea subplots, such that an equal area of species was observed. Observations were made in the morning hours between 7:00 AM and 9:00 AM when bee foraging was likely to occur. Each plot was observed on three days when the sunflowers were at pre-peak bloom and on three days when the sunflowers were at peak bloom. The main and interactive effects of the intercrop treatment (buckwheat alone, partridge peas alone, or both buckwheat and partridge pea together) and bloom period (pre-peak or peak) on pollinator abundance and taxon richness was determined using 2-way ANOVA (PROC MIXED, SAS v.9.4).

Blue vane traps

Blue vane traps (SpringStar Inc., Woodinville, WA) were used to capture pollinators foraging at the height of the intercrop. Two traps were placed on a fence post in the center of each plot where the sunflowers and intercrops converged. The traps were placed at the height of one meter and 0.3 meters, which corresponds to the approximate height of the partridge pea and buckwheat plants, respectively. Traps were placed in all treatment plots, including the control plot with no intercrops present. The traps were in the field for 48 hours after pre-peak bloom visual observations and again for another 48 hours after peak bloom visual observations. The pollinators in the traps were sorted to the lowest taxonomic level possible. The main and interactive effects of buckwheat (present/absent) and partridge pea (present/absent), and bloom period (pre-peak or peak) on pollinator abundance and taxon richness was determined for the

one meter traps and the 0.30 meter traps separately using a 3-way ANOVA (PROC MIXED, SAS v.9.4).

Intercrop growth

The height of intercrop plants was compared when grown alone versus when grown in combination with another intercrop species to determine if there was competition between the two intercrops. At the end of the study, the heights of three randomly chosen intercrop plants were measured in each plot and the average plant height for each plot was determined. The height of buckwheat in the presence and absence of partridge pea and the height of partridge pea in the presence and absence of buckwheat were both analyzed by using 1-way ANOVA (PROC MIXED SAS v.9.4).

Results

Visual observations

There was no effect of intercrop treatment on the abundance of pollinators observed foraging in the intercrop ($F_{2, 15} = 0.69, p > 0.5$, Fig. 12). There was also no effect of the intercrop treatment on the taxon richness of pollinators observed foraging in the intercrop ($F_{2, 15} = 0.14, p > 0.5$, Fig. 13, Table 8).

There were 64 total pollinators in the buckwheat treatment with more honey bees (*Apis mellifera*) foraging in this treatment than in either partridge pea or buckwheat + partridge pea. All bees in the family Anthophoridae that were observed in this study were found in the buckwheat treatment. In the partridge pea treatment, there were 105 total individuals observed. Of those 105, over half (67) were *Bombus spp.* (Table 9). The buckwheat + partridge pea intercrop treatment had the highest number of *Bombus spp.*

observed than any other treatments (Table 10). All bees in the family Megachilidae observed in this study were found in partridge pea.

Blue vane traps

At a height of one meter (Table 11), the abundance of pollinators present in blue vane traps was not affected by intercrop treatment (Figure 14), nor was pollinator richness (Figure 15). However, in the 0.30 meter traps (Table 12) there was an interactive effect of the buckwheat and partridge peas together on abundance of pollinators found in the blue vane traps ($F_{1,40}=4.14$, $p \leq 0.0486$, Fig. 16). The abundance of pollinators foraging at 0.30 meters when only partridge pea was present was less than the abundance of pollinators found foraging at 0.30 meters in the control with no intercrop, but when partridge pea was present with buckwheat the number of pollinators foraging at 0.30 meters was equal to that in the control. The buckwheat only treatment did not differ from the control. There was no difference across intercrop treatments in the taxon richness of the pollinators found in 0.30 meter traps (Fig. 17) for any of the treatments.

Intercrop growth

When partridge peas were planted alone, the average plant height was greater than if when they were planted alongside buckwheat plants ($F_{1,9}= 5.53$, $p \leq 0.05$, Fig. 18, Table 13). There is no effect of partridge pea on buckwheat height ($F_{1,10}= 1.31$, $p > 0.05$, Fig. 19, Table 14). The average heights of buckwheat plants when planted alone was not different than the height of buckwheat plants when planted in combination with partridge pea.

Discussion

In my previous chapter I found that while the addition of one intercrop, buckwheat (*Fagopyrum esculentum*) was beneficial for seed yield of the target crops, it had no impact on the pollinator community present in the field. This is also true for the opposite. Some intercrops that are great recruiters for a higher abundance and diversity of pollinators that may forage in the target crop do not host any pollinators that may assist in creating larger yields. In this experiment, we chose to look at this relationship and attempt to understand how the pollinator community foraging at the sunflower level might be different than the community foraging at the intercrop level. The pollinators in the intercrop during visual observations had a tendency to visit the partridge pea in higher abundance than the buckwheat. However, there was a change in the pollinator community diversity of buckwheat visitors that was not seen in the community that visited partridge pea. *Bombus pensylvanicus*, a bee that has been in decline in Missouri (Arduser, 2016), was found to be highly attracted to partridge pea. The resources provided by partridge pea differ from that of buckwheat due to the composition of amino acids and sugars that make up the nectar of each plant. It is possible that bees are utilizing plants that contain the specific resources they require (Gardener & Gilman, 2002).

When we measured plant height, we observed that there may be some competition between plants for resources. In treatments where buckwheat and partridge pea were planted together, the addition of buckwheat decreased the height of partridge pea plants compared to treatments where partridge pea was the only

intercrop. Plants in the partridge pea + buckwheat treatment were not intermingled, but instead planted in separate blocks next to each other. This shows that while buckwheat does have an effect on partridge pea height, the plants are not in direct competition with one another regarding soil resources so this effect is due to other factors. In some cases, having mixtures of cover crops present together in a field creates competition due to the plants being non-complementary. Species competition can result in greater biomass of one cover crop compared to another which can be good to attract more pollinators, but also detrimental to the other cover crop that it competes with (White et al., 2015).

We also observed that the pollinators caught in blue vane traps placed at the height of partridge peas (one meter), did not show an increase in the richness of pollinator taxa that visited the intercrop plants nor did it increase the overall numbers of pollinators that were present foraging on the intercrop. When traps were set at the height of buckwheat (0.3 meters), the change in abundance of pollinators found in partridge pea was greater than that of buckwheat, leading to the belief that there may have been some confounding factors associated with our trapping methods because in plots containing the buckwheat intercrop, traps placed at the height of the buckwheat were supposed to capture more insects than the traps at the partridge pea height due to the absence of that intercrop in the mixture.

Chapter 2 describes the abundance and taxon richness of pollinators observed foraging in pure stands of sunflower, buckwheat, and partridge pea. While there were no recorded observations of pollinators visiting partridge pea due to planting issues,

there were individuals found in buckwheat pure stands that were not observed in the buckwheat intercrop treatments. These include Halictidae and Tenthredinidae. In the pure buckwheat stands, there were no Colletidae found. However, in the buckwheat intercrop, 23 Colletidae were found and when buckwheat was combined with partridge pea, 34 Colletidae were observed.

Understanding pollinator behavior and monitoring their foraging patterns can sometimes be challenging, but it is important to create appropriate pollinator habitat nonetheless. In a recent analysis from the US Forest Service, it was stated that yield increases through increasing bee habitat has not been widely studied in certain agricultural system (Bentrup, 2016). Therefore, it is important to use this study as a building block for future research that can incorporate more focused conservation efforts of pollinator habitats.

Table 8. Results of ANOVA performed on the effects of intercrops, buckwheat (*Fagopyrum esculentum* Moench) and partridge pea (*Chamaecrista fasciculata* Michx), and bloom on richness and abundance of pollinators foraging in the intercrop.

RICHNESS					ABUNDANCE				
Effect	Num DF	Den DF	F Value	Pr > F	Effect	Num DF	Den DF	F Value	Pr > F
Bloom period	1	30	0.12	0.7317	Bloom period	1	30	0.11	0.7386
Intercrop treatment	2	30	0.88	0.4269	Intercrop treatment	2	30	0.41	0.6641
Bloom period * intercrop treatment	2	30	0.37	0.6960	Bloom period * intercrop treatment	2	30	0.09	0.9186

Table 9. Total abundance of pollinators observed visiting intercrop plants during visual observations for each intercrop plot type.

	Apidae		Andrenidae	Anthophoridae	Colletidae	Megachilidae	Syrphidae	total
	<i>Apis mellifera</i>	<i>Bombus</i>						
Sunflower + buckwheat	15	14	5	0	23	0	7	64
Sunflower + partridge pea	5	67	0	6	22	2	3	105
Sunflower+ buckwheat+ partridge pea	8	73	0	8	34	0	6	123
total	28	154	5	8	79	2	16	292

Table 10. Total abundance of all pollinators observed visiting intercrop plants during visual observations. Pollinator identity and abundance shown for each individual plot. SF= sunflower, BW= buckwheat, PP= partridge pea.

Replication	Treatment	Apidae		<i>Andrenidae</i>	<i>Anthophoridae</i>	Colletidae	Megachilidae	Syrphidae	total
		<i>Apis mellifera</i>	<i>Bombus</i>						
1	SF + BW	6	5	0	0	6	0	3	20
1	SF + BW	4	4	2	0	10	0	1	21
1	SF + BW	4	3	3	0	2	0	2	14
1	SF + PP	1	24	0	6	5	0	0	36
1	SF + PP	4	21	0	0	13	1	2	41
1	SF + PP	0	22	0	0	4	1	1	28
1	SF+BW+PP	4	17	0	0	12	0	0	33
1	SF+BW+PP	3	25	0	2	6	0	3	39
1	SF+BW+PP	1	22	0	0	2	0	3	28
2	SF + BW	1	1	0	0	1	0	0	3
2	SF + BW	0	0	0	0	0	0	0	0
2	SF + BW	0	0	0	0	4	0	1	5
2	SF + PP	0	4	0	0	4	0	0	8
2	SF + PP	0	2	0	0	7	0	0	9
2	SF + PP	0	0	0	0	0	0	0	0
2	SF+BW+PP	0	3	0	0	3	0	0	6
2	SF+BW+PP	0	0	0	0	0	0	0	0
2	SF+BW+PP	0	0	0	0	0	0	0	0
	total	28	153	5	8	79	2	16	

Table 11. Results of ANOVA performed on the effects of intercrops, buckwheat (*Fagopyrum esculentum* Moench) and partridge pea (*Chamaecrista fasciculata* Michx), and bloom on richness and abundance of pollinators in traps at the partridge pea height.

RICHNESS					ABUNDANCE				
Effect	Num DF	Den DF	F Value	Pr > F	Effect	Num DF	Den DF	F Value	Pr > F
Bloom period	1	40	11.70	0.0014	Bloom period	1	40	10.28	0.0027
Buckwheat	1	40	0.92	0.3439	Buckwheat	1	40	0.84	0.3652
Bloom period * buckwheat	1	40	0.17	0.6836	Bloom period * buckwheat	1	40	0.47	0.4961
Partridge pea	1	40	2.27	0.1401	Partridge pea	1	40	1.06	0.3090
Bloom period * partridge pea	1	40	0.17	0.6836	Bloom period * partridge pea	1	40	0.01	0.9094
Buckwheat * partridge pea	1	40	0.48	0.4978	Buckwheat * partridge pea	1	40	0.12	0.7331
Bloom period * buckwheat * partridge pea	1	40	0.02	0.8918	Bloom period * buckwheat * partridge pea	1	40	0.64	0.4276

Table 12. Results of ANOVA performed on the effects of intercrops, buckwheat (*Fagopyrum esculentum* Moench) and partridge pea (*Chamaecrista fasciculata* Michx), and bloom on richness and abundance of pollinators in traps at the buckwheat height.

RICHNESS					ABUNDANCE				
Effect	Num DF	Den DF	F Value	Pr > F	Effect	Num DF	Den DF	F Value	Pr > F
Bloom period	1	40	0.49	0.4985	Bloom period	1	40	7.36	0.0098
Buckwheat	1	40	0.05	0.8173	Buckwheat	1	40	0.20	0.6537
Bloom period * buckwheat	1	40	0.05	0.8173	Bloom period * buckwheat	1	40	0.32	0.5752
Partridge pea	1	40	3.46	0.0703	Partridge pea	1	40	4.61	0.0379
Bloom period * partridge pea	1	40	0.86	0.3580	Bloom period * partridge pea	1	40	1.28	0.2652
Buckwheat * partridge pea	1	40	0.86	0.3580	Buckwheat * partridge pea	1	40	4.14	0.0486
Bloom period * buckwheat * partridge pea	1	40	0.86	0.3580	Bloom period * buckwheat * partridge pea	1	40	0.63	0.4336

Table 13. Results of ANOVA examining the effect of buckwheat (*Fagopyrum esculentum* Moench) presence on partridge pea (*Chamaecrista fasciculata* Michx) height.

Response of partridge pea height				
Effect	Number DF	Den DF	F Value	Pr > F
Buckwheat presence	1	9	5.53	0.0432

Table 14. Results of ANOVA examining the effect of partridge pea (*Chamaecrista fasciculata* Michx) presence on buckwheat (*Fagopyrum esculentum* Moench) height.

Response of buckwheat height				
Effect	Number DF	Den DF	F Value	Pr > F
Partridge pea presence	1	10	1.31	0.2792

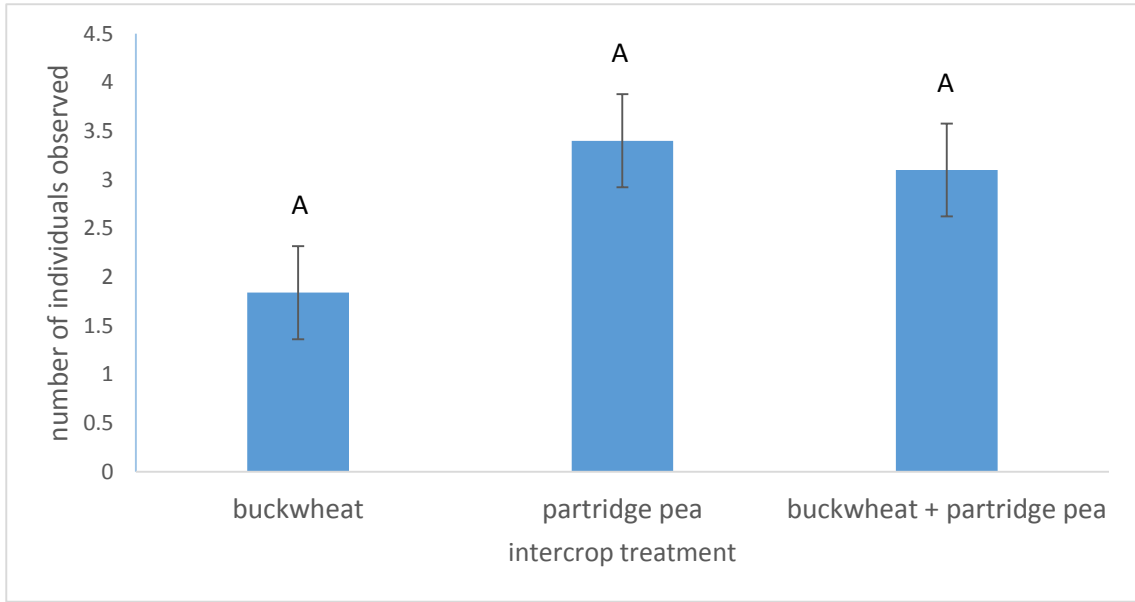


Figure 12. Total abundance of the pollinators observed foraging on either buckwheat (*Fagopyrum esculentum* Moench) or partridge pea (*Chamaecrista fasciculata* Michx) in response to each intercrop treatment during 10 minute visual observations. LS means \pm 1 SE shown. Means with different letters are significantly different at $p \leq 0.05$.

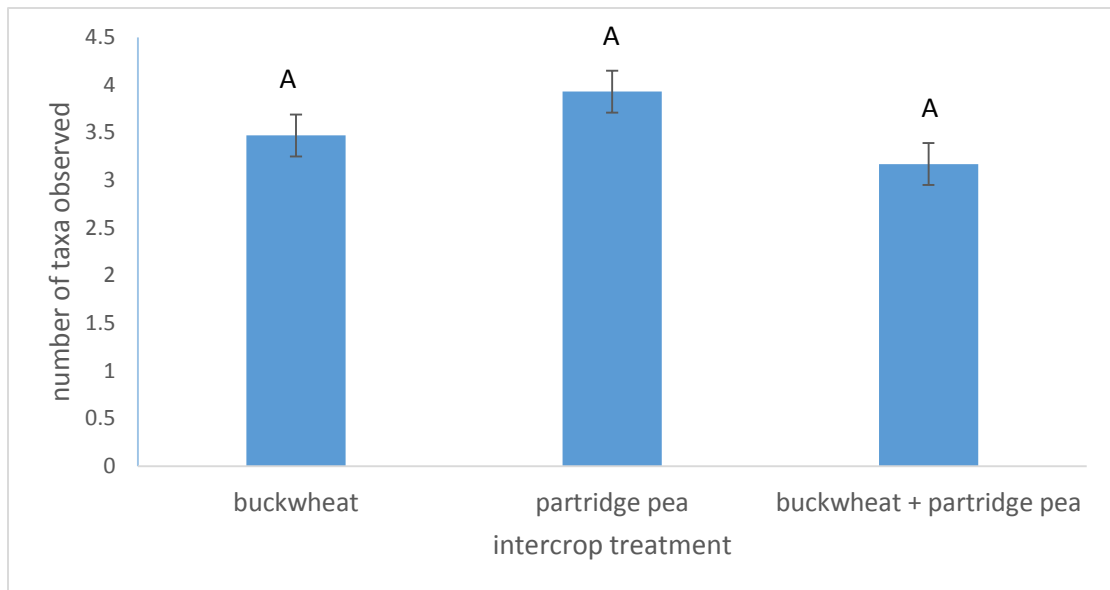


Figure 13. Total taxon richness of the pollinators observed foraging on either buckwheat (*Fagopyrum esculentum* Moench) or partridge pea (*Chamaecrista fasciculata* Michx) in response to each intercrop treatment during visual observations summed across six, 10 minute observations. LS means \pm 1 SE shown. Means with different letters are significantly different at $p \leq 0.05$

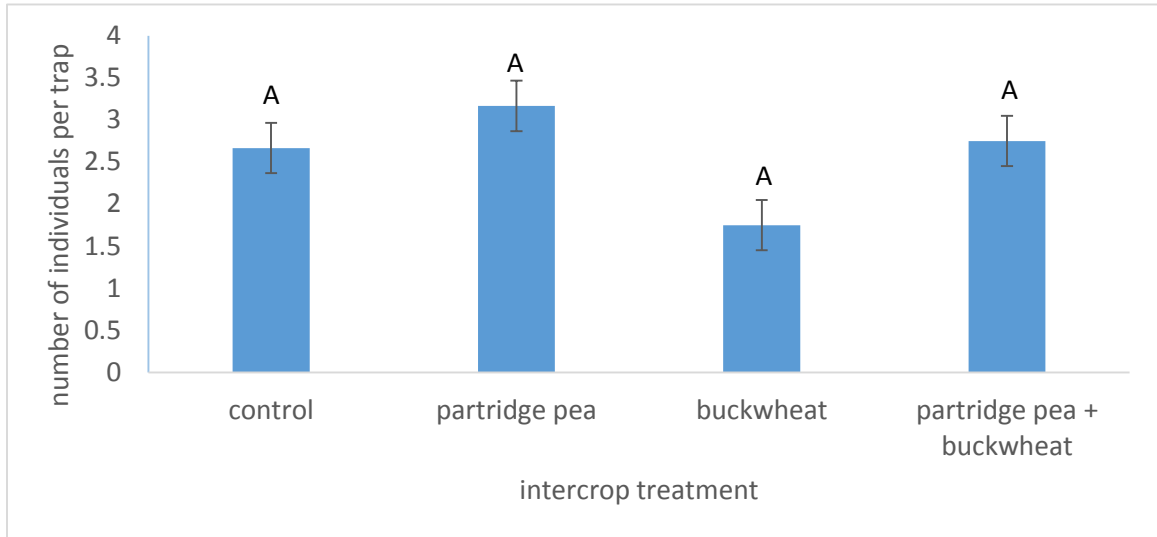


Figure 14. Total abundance of pollinators observed in blue vane traps placed at the approximate height of partridge pea (*Chamaecrista fasciculata* Michx), 1 meter. Traps were deployed for 48 hours for two sampling periods during the summer. LS means \pm 1 SE show. Means with different letters are significantly different at $p \leq 0.05$.

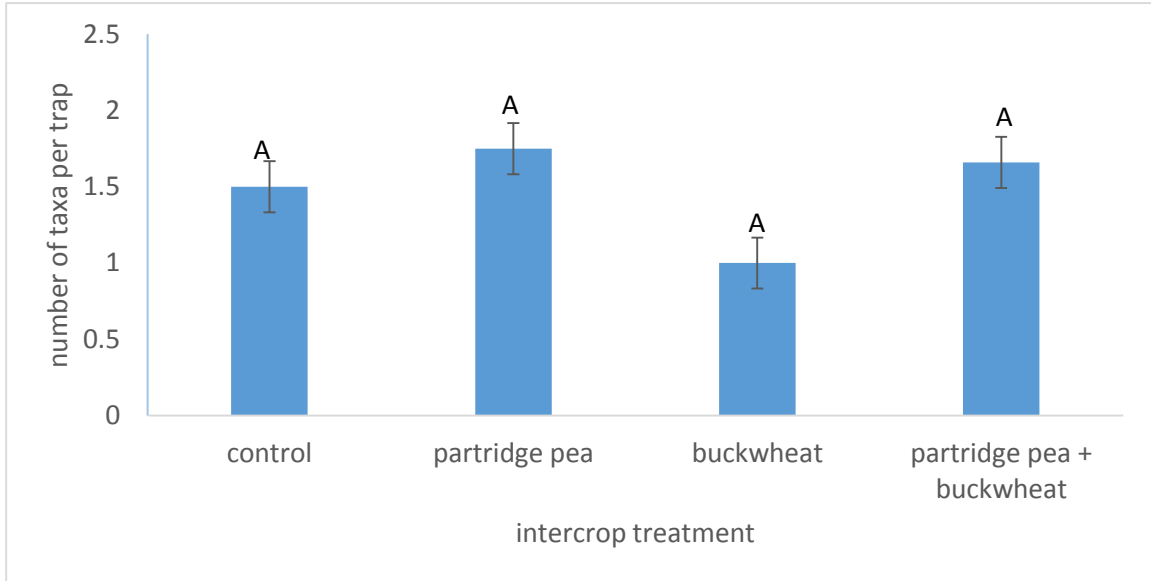


Figure 15. Total taxon richness of pollinators observed in blue vane traps placed at the approximate height of partridge pea (*Chamaecrista fasciculata* Michx), one meter. Traps were deployed for 48 hours for two sampling periods during the summer. LS means ± 1 SE show. Means with different letters are significantly different at $p \leq 0.05$.

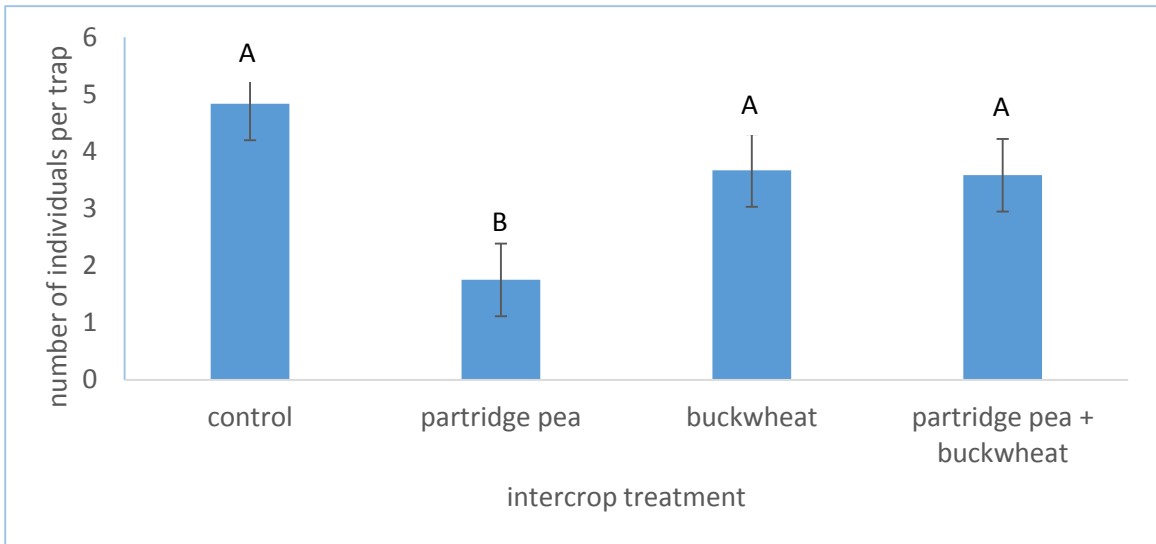


Figure 16. Total abundance of pollinators observed in blue vane traps placed at the approximate height of buckwheat (*Fagopyrum esculentum* Moench), 0.30 meters. Traps deployed for 48 hours for two sampling periods during the summer. LS means \pm 1 SE show. Means with different letters are significantly different at $p \leq 0.05$.

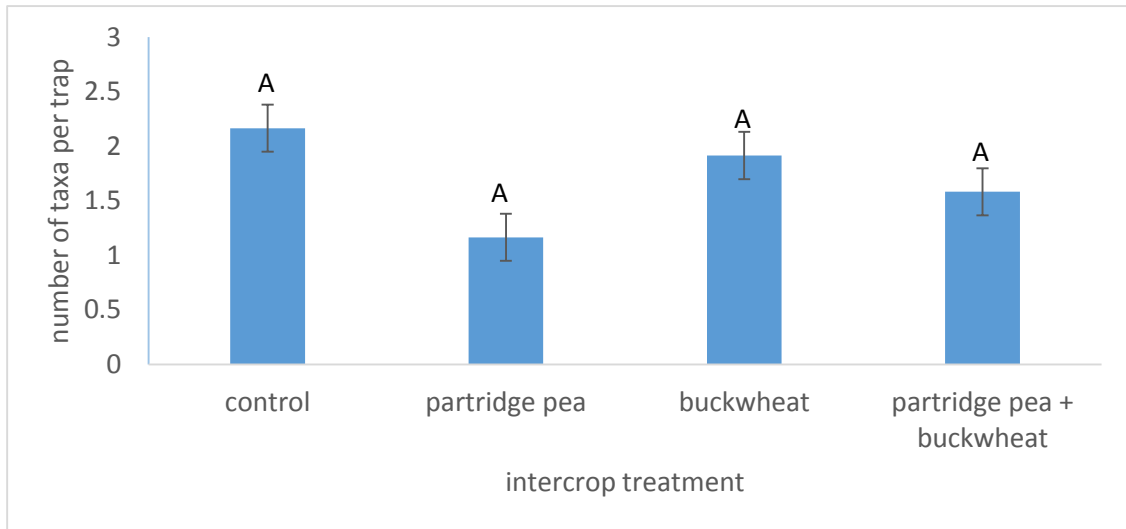


Figure 17. Total taxon richness of pollinators observed in blue vane traps placed at the approximate height of buckwheat (*Fagopyrum esculentum* Moench), 0.30 meters. Traps deployed for 48 hours for two sampling periods during the summer. LS means \pm 1 SE show. Means with different letters are significantly different at $p \leq 0.05$.

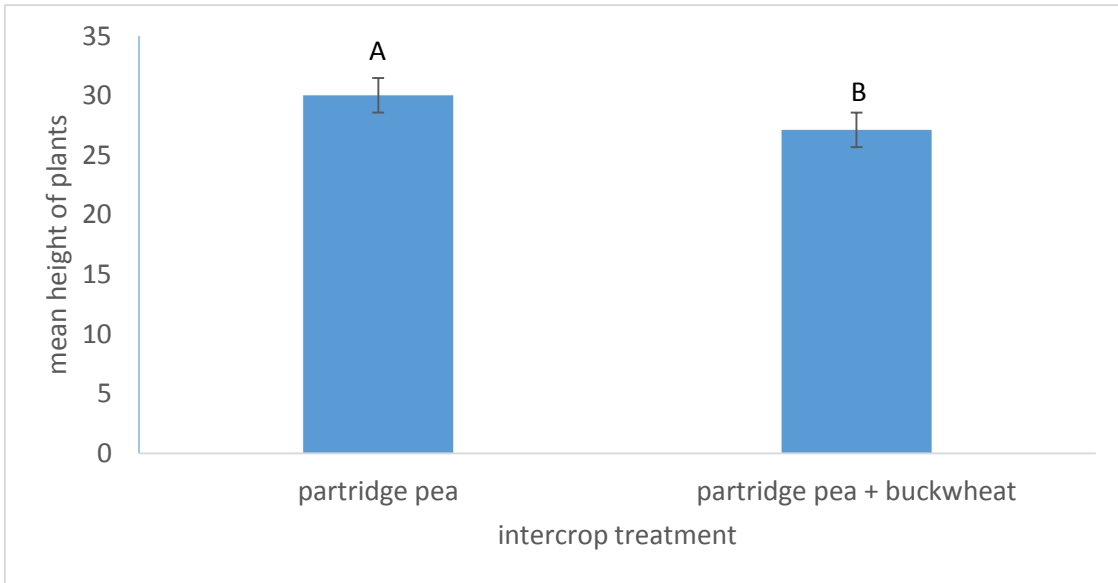


Figure 18. Height of partridge pea (*Chamaecrista fasciculata* Michx) in the presence and absence of buckwheat (*Fagopyrum esculentum* Moench). LS means \pm 1 SE shown. Means with different letters are significantly different at $p \leq 0.05$.

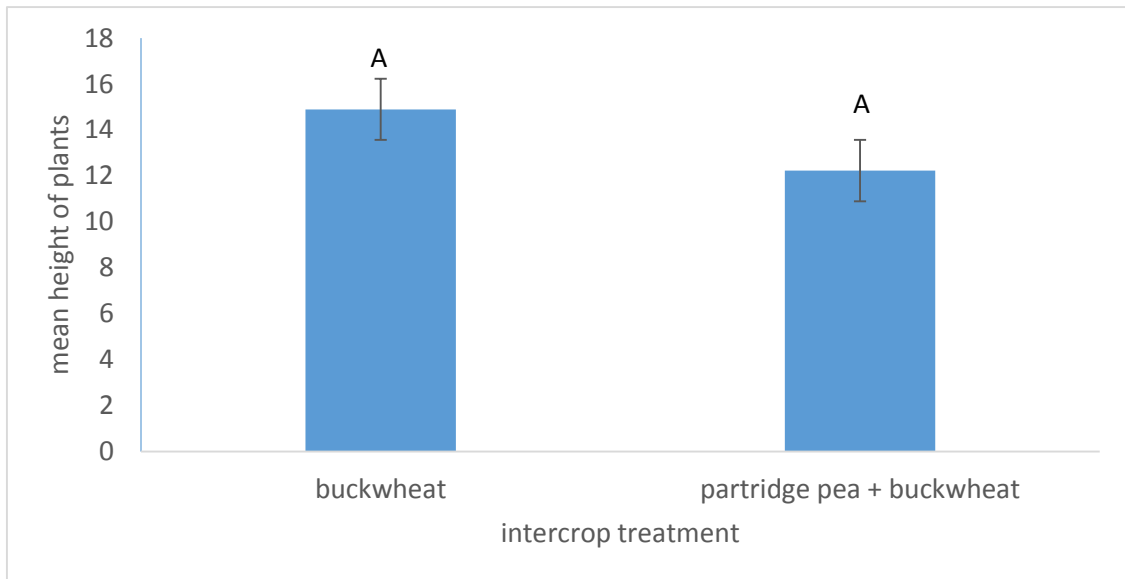


Figure 19. Height of buckwheat (*Fagopyrum esculentum* Moench) in the presence and absence of partridge pea (*Chamaecrista fasciculata* Michx). LS means \pm 1 SE show. Means with different letters are significantly different at $p \leq 0.05$.

Chapter 5:

SUMMARY & CONCLUSIONS

In the first year of our study, pure stands of sunflower (*Helianthus annuus* L.), buckwheat (*Fagopyrum esculentum* Moench), and partridge pea (*Chamaecrista fasciculata* Michx) were planted. While partridge pea did not reach its reproductive growth stage during visual observations, sunflower and buckwheat were both observed to identify the pollinator community foraging in each of the pure stands. We found that insect taxon richness foraging in both of the pure stands of sunflower and buckwheat was the same, but the bee community differed both in total abundance and composition of the insect taxa present. The abundance of bees in sunflower was over twice as many found in buckwheat. Since sunflowers were planted at such a high density and have a longer bloom period than buckwheat, it is possible that pollinators are attracted to plants at a higher density that are accessible for a longer period of time. Also, sunflowers grow much taller and have more surface area than buckwheat, making them more accessible to insects.

In year two, we used the information learned about pure stands to create intercrop treatments containing buckwheat and partridge pea planted alongside sunflower, the cash crop. During visual observations of the pollinators foraging on sunflowers, all intercrop treatments increased the abundance of pollinators on sunflower. The addition of buckwheat with partridge pea may have reduced the positive effect of partridge pea on the sunflower pollinator community because of a reduction in buckwheat quality due to competition between intercrop plants. We found that

partridge pea plants were shorter on average when they were grown alongside buckwheat plants, suggesting competition for access to important nutrients and other resources necessary for growth.

The addition of the two intercrops together also had no effect on the number of taxa present foraging on sunflowers. The lack of an effect on taxon richness may be due to competition for resources and foraging habits of the pollinators. The addition of buckwheat to partridge pea may be causing pollinators to move through the intercrops in search for other sources of pollen and nectar, thus reducing the amount of pollinators that are foraging on sunflowers.

What is fascinating about the use of the intercrop mixture is that the addition of a buckwheat intercrop influences sunflower seed set, the addition of partridge peas as an intercrop did not influence seed set in sunflowers. Having a diverse pollinator community with partridge pea present may have resulted in interference that resulted in reduced seed set. When buckwheat was present, there could have been one very efficient pollinator that enhanced seed set. In the sunflower + buckwheat intercrop, there were more individuals in the family Colletidae foraging than in the other treatments. In another example, individuals in the family Andrenidae were only present foraging in the intercrop if buckwheat was the only intercrop present. Perhaps the addition of partridge pea and the community of insects associated with its introduction are a deterrent for Andrenidae that are foraging. Behaviorally, different bee taxa forage differently. I observed that some of the larger *Bombus* species will displace smaller honey bees if they are foraging together in an area. When one or a few Colletidae are

foraging, they are easily displaced by larger species as well. However, when many Colletidae are present foraging on a sunflower head, larger species do not have the space to land on the sunflower and are therefore unable to utilize the same host plant. Levels of sociality among pollinators, their preference for floral resources, and their ability to procure those resources are all factors that influence the foraging patterns of pollinators.

Looking at this from a practical angle, the addition of one or more of these intercrops can be used to fit the needs of a grower. If a sunflower grower is interested in just yields, it may be beneficial to forego partridge pea and only plant buckwheat. However, buckwheat will not help to create foraging sites and attract a greater number of pollinators. The gradient of plant heights across the intercrop treatments ranges from plants that are 0.3 meters tall (buckwheat) to plants that are one meter tall (partridge pea) to plants almost 2 meters tall (sunflowers), thus creating spatial diversity in the pollinator habitat as well.

One other interesting observation to note is that in the partridge pea treatment, half of the individual pollinators observed foraging on sunflower were *Bombus pensylvanicus*, a bee that has been in decline in Missouri. This was the highest number of *B. pensylvanicus* observed in any treatment and over twice as many *B. pensylvanicus* were observed in this treatment than the other treatments combined.

Understanding pollinator behavior and monitoring their foraging patterns can sometimes be challenging, but it is important to create appropriate pollinator habitat nonetheless. This can be achieved by creating the right intercrop mixture that provides

the resources that pollinators need during the entire growing season. By using multiple intercrops that bloom from early to late in the season, pollinators that forage during the summer will have access to floral resources throughout the growing season.

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Vita

Brittani Nicole Alexander was born in Kansas City, Missouri. In the fall of 2007, she enrolled at Columbia College where she earned a Bachelor of Arts in Biology and a Bachelor of Science in Environmental Science. Her participation in the 2010 Emerging Leadership Program led her to meet Wayne Bailey of the University of Missouri. He offered her a graduate position at the University and she joined the laboratories of Drs. Wayne Bailey and Deborah Finke in the Division of Plant, Insect, and Microbial Sciences in 2013. While at the University of Missouri, Brittani studied the native bee communities of the area and served as president of the CV Riley Entomological Society.