Biodiversity Boost: Unleashing Ecosystem Services for a Thriving Planet

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Abstract

Because conventional agriculture does not often involve practices that increase biodiversity, we ignore the extent to which biological control and pollination can be supported by non-crop plants such as trap crops and insectary plants in agro-ecosystems. Increasing the biodiversity of a wildflower garden may increase the diversity and/or abundance of pollinators, predators, and parasitoids present, all of which are essential in creating a balanced ecosystem that can contribute to agricultural productivity. To gauge this, we hypothesized that a mixture of five insectary and trap crop plants will attract a greater number of predators, parasitoids, and pollinators than (1) single plant species and (2) wildflower plants already present next to an apple tree block. We evaluated plots of Mighty Mustard (*Brassica juncea)*, Buckwheat (*Fagopyrum esculentum*), Dwarf Sunflowers (*Helianthus Annus*), Sweet Alyssum (*Lobularia maritima*), Partridge Pea (*Chamaecrista fasciculate)*, and a mixed crop of all five species, as well as two wildflower plots, which were analyzed jointly. In each plot, we actively (via visual counts) and passively (via sticky traps) monitored insect visitations. Further, we prepared, deployed, and retrieved sentinel (frozen) Brown Marmorated Stink Bug eggs, which we assessed for signs of predation and parasitoid emergence. By monitoring egg consumption and the difference in beneficial insect prevalence across plant types, we determined that biodiversity can influence important ecosystem services. The Sunflower, Buckwheat, and the Partridge Pea alone seemed to outperform the mixed plot. We found no evidence to suggest that a mixture of plants increases the prevalence or diversity of insect residents when compared to wildflower plants. Our results motivate further studies to clarify the effect of plant biodiversity on the prevalence of ecosystem service providers in the apple agroecosystem.

1 Introduction

As of 2021, there are over two million farms in the United States, equalling about 895,300,000 acres of land used for multiple agricultural purposes (USDA 2021). Approximately 4.9 million acres of this land are exclusively occupied by fruit orchards (USDA, 2021). The majority of crop-producing farms use conventional practices to increase yield, quality, and profit, usually

relying on monocultures– the cultivation of a single crop in one area (Sumberg et al., 2022), and synthetic inputs such as fertilizers, herbicides, and pesticides, all of which are known to impact the ecosystem and lead to a loss of biodiversity (Simon et al., 2011). As an alternative or supplement to chemical controls, biological controls have been successfully used in many cropping systems to reduce pest populations (Reido et al., 2019).
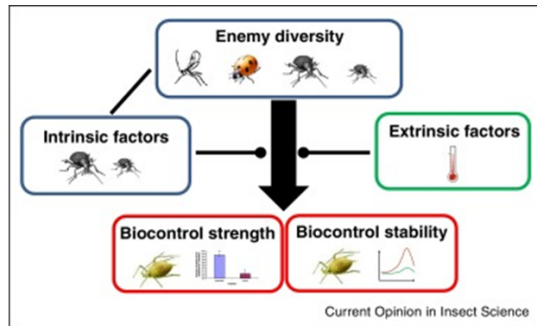

Figure 1. The effects of increasing enemy diversity based on intrinsic and extrinsic factors. "The effect of increasing enemy species diversity on the strength and stability of biological control depends on the traits in the enemy community (intrinsic factors), and on prey traits and environmental conditions (extrinsic factors)." (Jonsson et al. 2017)

Biological control can be described as controlling a pest(s) by the introduction, promotion, or retention of natural enemies like parasites, predators, herbivores or pathogens (Van Driesche et al., 2009). The term "pest", in an agricultural context, can be defined as any organism (insects, mites, nematodes, plant pathogens, weeds and vertebrates, among others) that is pernicious or damaging to human activities (OTA, 1979). However, all insects contribute to a balanced ecosystem. For instance, increasing plant biodiversity can enhance important ecosystem services provided by organisms that occupy these ecological niches, such as increased pollination, increased microbes in the soil (which can promote soil health), and increased biological control of economically-significant pests (Erisman et al., 2016) (Fig. 1).

Ecosystem services, like pollination, are crucial to any crop-production system. Among pollinators, bees are one of the most important in both agricultural and wild ecosystems (Potts et al., 2010). Despite this, bee populations have been declining over the years due to habitat loss, chronic insecticide use, climate change, and species invasion (Winfree, 2010). Human-induced habitat loss is one of the largest threats to not just pollinator species, but other beneficial insects like predators and parasitoid wasps (Sanchez-Bayo et al., 2019).

Parasitoid wasps play an important role in biological pest control and, given that they rely on insect hosts for nutrition and reproduction, are essential in insect food chains (Fei et al. 2022). Insect food chains show the relationships between insects and how competition, predation, and parasitism occur at different trophic levels (Fei et al., 2022).
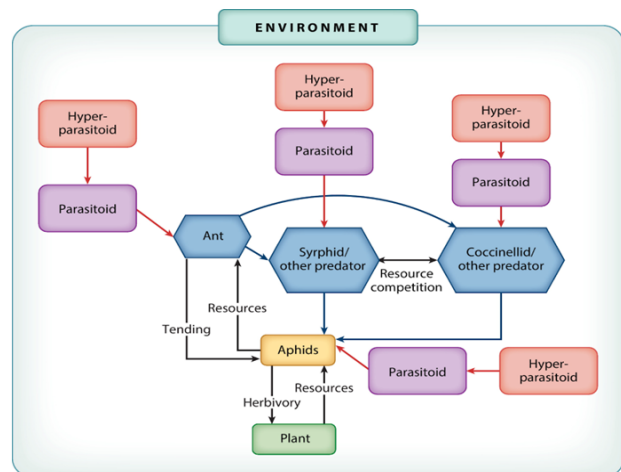

Figure 2. Aphid food web extended to the fifth trophic level. "Aphid food web extended to the fifth trophic level. Different colors refer to the various trophic levels or natural enemy status [predators in blue and (hyper)parasitoids in purple and red], with the plant (first trophic level) in green and aphids (second trophic level) in yellow. Parasitoids of aphids are in the third trophic level, whereas those of aphid predators are in the fourth. All parasitoids can be attacked by their own parasitoids (hyperparasitoids). Direct interactions between food-web members are indicated by arrows, with predation in blue and parasitism in red. Other types of direct interactions are indicated by dark gray arrows. Interactions are not restricted to direct ones, but for simplicity, indirect interactions are not considered in this figure." (Fei at al. 2022)

Predatory arthropods lie on the third trophic level (Fig. 2), while parasitoids lie on the fourth trophic level (Fig. 2; Fei et al., 2022). Understanding these relationships can help create a sustainable ecosystem that promotes the success of biological controls at all trophic levels.

Increasing the abundance of beneficial insects first requires habitat resources that are optimal for their survival and that include rich sources of nutrition and shelter. To meet these criteria, non-crop plants such as insectary plants and selected trap crop plants can be strategically chosen to attract certain types of pests (in the case of trap crops) and beneficial arthropods. On the one hand, insectary plants can attract, feed and shelter parasitoids and predators to enhance biological pest control (Morandin et al., 2016). On the other hand, trap crop plants are sacrificial plants that are extremely attractive to herbivores and can trump their attraction response to the cash crop (Westerfield et al., 2022). Interestingly, some plant species can act as both trap crops for an insect pest, and insectary plants for the pest's enemies (Shrestha et al., 2019).

Sweet Alyssum (*Lobularia maritima*) and Buckwheat (*Fagopyrum esculentum*) are examples of insectary crops that have been shown to attract parasitoid wasps (Piñero and Manandhar, 2015); Shrestha et al., 2019) and, for some species, increase their longevity and fecundity when compared to alternative nectar diets (Nafziger et al., 2011). Buckwheat in particular has an open, dish shaped flower with a strong fragrance that is very attractive to a wide variety of pollinators and natural enemy species (Shrestha et al., 2019; Lui et al., 2020).

Sunflower (*Helianthus annuus*) also acts as a powerful insectary plant and trap crop (Jones et al. 2005) that confers health benefits to its visitors. Specifically, sunflower pollen has been found to greatly reduce the intensity of infection by the trypanosome *Crithidia Bombi* in bumble bee (*Bombus impatiens)* workers (Giacomini et al., 2021). Sunflower has also been successfully used as a trap crop to enhance an attract and kill system for the Brown Marmorated Stink Bug (BMSB; *Halyomorpha Halys)* in an orchard setting with buckwheat (*F. esculentum*) (Pinero et al., 2022. In addition, sunflower acts as a trap crop in pepper plants (Soergel et al., 2015).

Partridge pea (*Chamaecrista fasciculate)* is known to be an insectary plant and very attractive to beneficial insects (Moore, 2010). Partridge Pea has also been found to be one of the more important hosts for the brown stink bug (*Euchistis Servus)* and can be used as a trap crop (Jones and Sullivan, 1982). Another known trap crop that has also been utilized is mighty mustard, which also serves as an insectary plant (*Brassica juncea;* Shrestha et al., 2019).

While many studies have tested individual plants, in the context of apple production to our knowledge only a few studies have addressed the question of whether mixing multiple plant species show increased abundance and diversity in pollinators, predators, and parasitoids compared to individual plant species and plants naturally growing. To address these questions, we conducted a field study at the University of Massachusetts Amherst Cold Spring Orchard Research Facility (CSO). We hypothesized that the mixed plot with sunflower, buckwheat, mighty mustard, partridge pea and sweet alyssum will show a greater number of parasitoids, predators and pollinators than the two wildflower plots or any one of the individual plant types. In order to test the notion that increased abundance of predatory and parasitic insects

contribute to pest control, we utilized sentinel egg masses (SEMs) of the Brown Marmorated Stink Bug (BMSB). Brown Marmorated Stink Bugs are native to China, but invaded the United States in the early 1990s (Lee et al. 2013), and Massachusetts in 2007 (MDAR, 2021). The BMSB has recently become a problem for some growers in MA, who have begun to spray chemical insecticides to deal with apple infestations (personal correspondence). Creating alternatives that increase natural biological controls may help prevent further spread of this pest (and others) in MA agroecosystems.

2 Materials and Methods

2.1 Study Site and Plot Preparation

We conducted our experiment at the University of Massachusetts Amherst Cold Spring Orchard Research Facility (CSO) in Belchertown, Massachusetts from July 14th to July 31st, 2023. Specifically, we prepared a 38 meter long and one meter wide area of the orchard perimeter next to a new apple cider tree block. Along this area, eight 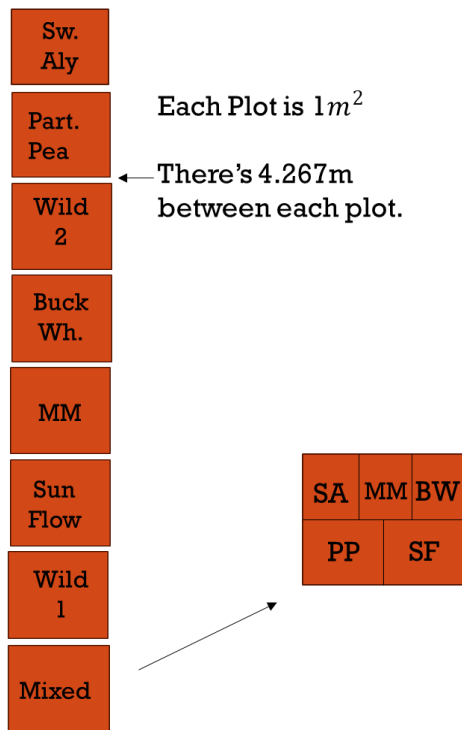separate one meter by one meter plots (delimited using PCV square frames) were selected. Each plot was approximately four meters apart from each other (Fig. 3). Two PVC squares were placed over existing vegetation that included multiple flower species. The rest of the area was prepared, via mowing and rototilling, to plant the experimental plants.

The following treatments were evaluated: (1) sunflower alone, (2) buckwheat alone, (3) sweet alyssum alone, (4) mighty mustard alone, (5) Partridge pea alone, (6) an even mix of all five plant species, and (7) multiple flowering plant species growing naturally in the experimental location. Buckwheat, sweet alyssum, and mighty mustard were directly seeded. Sunflower and Partridge Pea were transplanted a month after they were seeded in May at the University of Massachusetts Amherst College of Natural Sciences (CNS) Educational Greenhouses. After the experimental plants were in, the plots were checked at least once per week for flower blossoming, after which we began our monitoring protocol.

Figure 3. Diagram of plot placement at Cold Spring Orchards and measurements. Starting from the top down, we planted Sweet Alyssum, Partridge Pea, Wildflower 2, Buckwheat, Mighty Mustard, Sunflower, Wildflower 1 and then the mixed plot. The mixed plot contained an even mixture of Partridge Pea, and Sunflower, and the other half of the plot was split in thirds planted with Sweet Alyssum, Mighty Mustard and Buckwheat.

2.2 Identification of Preexisting Flower Species

The plant species that were in the existing vegetative plots were identified using Google Lens and iNaturalist. All plants in both wildflower plots were in bloom when the first deployment was initiated. We found that there were only two plants that were identified as native plants: *Erigeron strigosus* (Daisy Fleabane) and *Oxalis stricta* (Common Yellow Oxalis), the latter of which was identified only in the second wild plot (Wild 2). One exotic plant identified in the first wildflower (Wild 1) plot that was not in the second one was identified as *Hieracium lachenalii,* also known as Yellow Hawkweed. Further, two other exotic plants identified in the second wildflower plot were not present in the first wildflower plot: *Leucanthemum vulgare* (Oxeye Daisy), and *Rumex obtusifolius* (Bitter Dock). The rest of the plants identified were found in both wildflower plots: *Galium mollugo* (False Baby's Breath), *Vicia cracca* (Bird Vetch), *Trifolium pratense* (Red Clover), *Plantago lanceolata* (Ribwort Plantain), and *Daucus carota* (Queen Anne's Lace).

2.3 Monitoring Biocontrol Services and Flower Visitors

In order to directly quantify biological control services at the plots, we deployed sentinel Brown Marmorated Stink Bug (BMSB) eggs. The eggs were purchased from Phillip Alampi Beneficial Insect Laboratory (New Jersey Department of Agriculture, NJ). To prepare the sentinel egg masses (SEMs), the eggs were kept at -80°C for at least 24 hours before deployment (Tillman et al., 2020). After the eggs were frozen, we superglued two egg masses on a small piece of wax paper. The eggs sat for about ten minutes to dry before deploying them on each plot. To affix the eggs, we used paper clips on a leaf or stem in the plot, with the eggs facing the soil to minimize weathering. The first set of egg masses and sticky cards were all deployed on July 14th. They were retrieved and redeployed again on July 20th, again on July 25th, and for one last time on July 31st. Every time egg masses were retrieved from the field, they were taken back to the lab to assess predation (e.g. chewing, sucking) damage using a dissecting (light) microscope (Tillman et al., 2020). After this, the eggs from each plot were isolated into cups and placed in a temperature-controlled rearing chamber for four to five weeks to monitor parasitoid emergence.

In order to categorize the insects recorded they were separated into pollinators, predators, parasitoids, and a pests/other category and identified to the family level or order level, other than parasitica, which we defined as a group encompassing all specimens belonging to the multiple hymenopteran superfamilies that include parasitoid wasps. The order diptera (e.i. Tachinidae) was also included under parasitica. The insect groupings can be seen in Table 1.

Table 1. Breakdown of insect groups included into Parasitica, Pollinators, Predators and Pests/Other and identified to the family or order level.

Parasitica	Parasitoid Wasps, *Tachinidae*
Pollinators	*Syrphidae, Vespidae, Halicitidae, Hesperiidae, Apidae, Nymphalidae, Lycanenidae, Papilionidae*
Predators	*Araneae, Coccinellidae, Vespidae, Dolichopodidae, Asilidae, Carabidae, Stratiomyidae, Cicindelidae, Nabidae, Coenagrionidae, Reduviidae, Libellulidae*
Pests/Other	*Muscidae, Cicadellidae, Miridae, Thyreocoridae, Cydnidae, Lygaeidae, Formicidae, Tenebrionidae, Curculionidae, Membracidae, Scarabaeidae, Coreidae, Pieridae, Latridiidae, Pentatomidae*

We set up one yellow sticky card and a clear sticky card within each plot space to monitor for plot visitors (i.e. parasitica, predators, pollinators and other/pest insects). The yellow card was originally 11in x 9in but was cut in half to be 9in x 5.5in and then was folded in half so both sides of the card would catch insects. The clear card was originally 11.75in x 6in and was sticky on either side but was cut in half (6.25in x 6in). The cards were assessed once per week and trapped insects were also recorded. Further, at least once per week, we visually recorded insect visitations on each individual plot for ten minutes. The plots were monitored in a different order every time to minimize the effect of position. Each insect was identified at least to the order level (e.g. Araneae), but mostly to the family level (e.g. Coccinellidae). Microsoft Excel (v. 2307; Microsoft Office) was used to digitize, analyze, and visually represent all of our data.

3 Results

3.1 Sentinel Eggs

We observed the highest mean number of chewed SEMs in the Buckwheat plot ($\bar{x}$=5.75; Fig. 4). Following the Buckwheat was the combination of both wildflower plots ($\bar{x}$=4.88). We observed that the Sunflower plot had a mean chewing pressure of 3 and the remaining two plots, Partridge Pea ($\bar{x}$=0.5) and the mixed ($\bar{x}$=0.25) plot had the lowest amount of chewing damage (Fig. 4). In Figure 4, we show the mean number of sucked sentinel BMSB eggs. We observed that the combination of both the wildflower 1 and wildflower 2 plots ($\bar{x}$=5.5) had a very similar mean to Sunflower ($\bar{x}$=5.25). The Partridge Pea and the Mixed plot both had the same mean number of sucked eggs ($\bar{x}$=4.75). The plot with the least amount of sucked eggs was Buckwheat with a mean of 3.75.
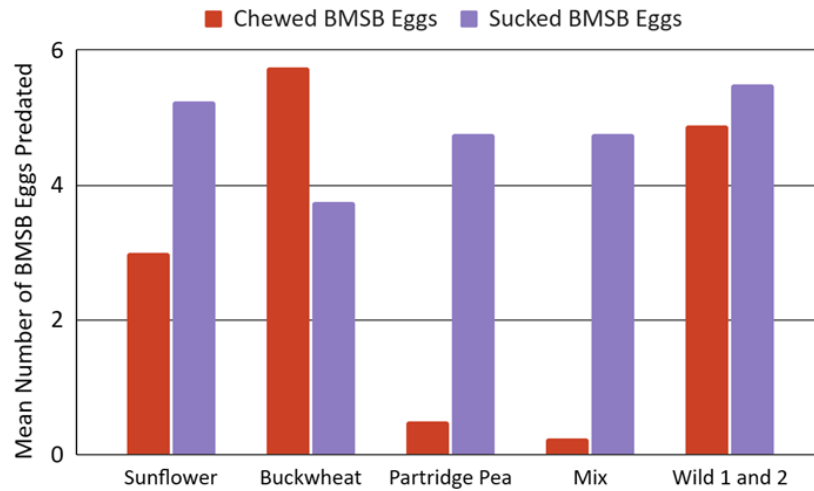
Figure 4. Mean number of chewed sentinel Brown Marmorated Stink Bug (BMSB) eggs compared to the mean number of sucked BMSB eggs. Buckwheat has the highest mean of chewed eggs while the mixed plot has the lowest chewing pressure. The combination of Wild 1 and Wild 2 have the closest comparison of chewed eggs versus the sucked eggs, while the mixed plot and Partridge Pea have the furthest comparison of mean values.

3.2 Arthropod Counts in Yellow and Clear Sticky Cards

In Figure 5, we compare the mean number of predatory, parasitic, and pollinator insect catches on sticky traps (yellow and clear sticky card data are combined) across six different flower treatments. In terms of predatory insects, we observed that Buckwheat ($\bar{x}$=4.5) had the highest mean of predators compared to the other treatments. Following Buckwheat was Partridge Pea with a mean of 4.34. Sunflower follows Partridge Pea with a mean of 3.25. The treatment with the lowest mean of predator catches was the mixed plot ($\bar{x}$=2.13) followed by the combination of both wildflower plots with a mean of 2.69 predators.

As for recorded parasitica, the combination of both wildflower ($\bar{x}$=11.44) plots had the highest mean values. Following closely behind were Sunflower ($\bar{x}$=10.75) and the mixed plot with a mean of 10.375. The treatment with the lowest mean of parasitica recorded was Buckwheat ($\bar{x}$=8.63).

The treatments that attracted the most pollinators were Sunflower ($\bar{x}$=3.13), Buckwheat ($\bar{x}$=3) and Partridge Pea ($\bar{x}$=3). We observed that the results of the combination of both wildflower treatments had a mean of 2.06 and the treatment with the lowest mean of recorded pollinators was the mixed plot ($\bar{x}$=1.63).

We observed that Sunflower ($\bar{x}$=14.63) had the highest mean of pests and/or other insects, followed by Partridge Pea with a mean of 14.13 and Buckwheat ($\bar{x}$=13.75). The results of the combination of the two wildflower plots had a mean of 5.38 and the treatment that had three times less than the other treatments is observed as the mixed plot ($\bar{x}$=3.63).
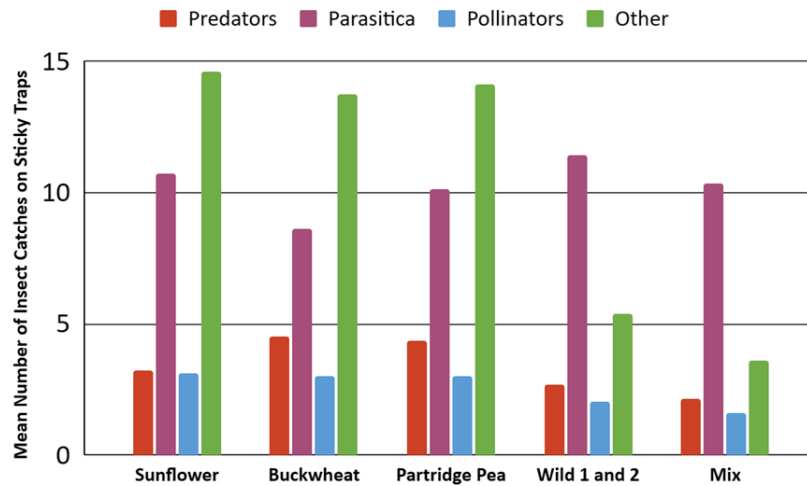
Figure 5. The mean number of insect catches on yellow and clear sticky traps across six different flower treatments. The Buckwheat had the highest mean of predators closely followed by Partridge Pea. The combination of Wild 1 and Wild 2 have the highest amount of parasitica, followed by Sunflower. The mixed plot was found to have the least number of pollinators while the Sunflower had a slightly higher count than the other treatments.

Figure 6 shows the mean number of insect catches (by functional group) on yellow sticky cards versus the clear sticky cards. Across all treatments, we observed that the yellow cards recorded more insect catches, except in parasitica, there was no recorded difference. The most insect catches on the yellow sticky cards were of other insects and/or pests ($\bar{x}$=14.58), following the pests/other category is observed as parasitica ($\bar{x}$=10.13). Following parasitica were predators on the yellow sticky cards, with a mean of insect catches of 6.08. The least amount of insects caught on the yellow sticky cards were pollinators ($\bar{x}$=5.04).
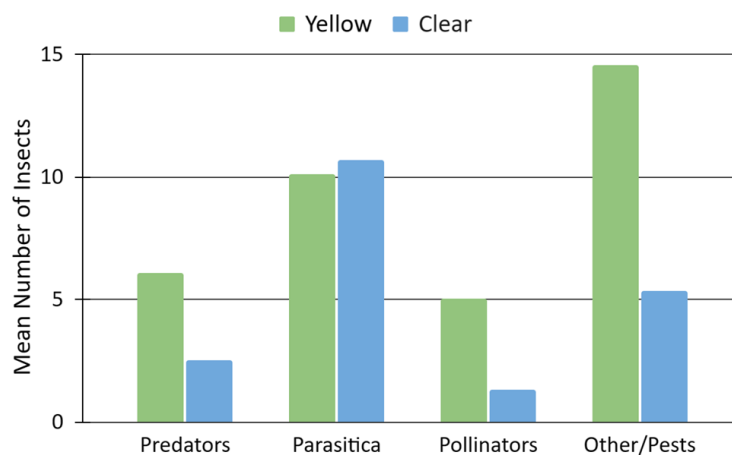

Figure 6. The mean number of insects found on the yellow sticky cards compared to the mean number of insects found on the clear sticky cards. For predators, pollinators, and the pests/other category the yellow cards had more insects than the clear cards. There was no difference between the yellow or clear in the parasitica category.

3.3 Visual Arthropod Counts:

In Figure 7, we show the mean number of visual insect observations across six different treatments. In the Sunflower plot, the highest mean number of recorded insect visitations was observed as pests/other ($\bar{x}$=3.25). In the same plot, the second highest recorded visual visitations were done by pollinators ($\bar{x}$=3). Also in the Sunflower treatment, the lowest amount of visual visitations is observed as predators with a mean of 1.5.

In the Buckwheat treatment we observed that the highest mean of insect visitations came from pollinators ($\bar{x}$=4.25). Following closely behind in the same treatment was other/pests with a mean of 4. The lowest amount of insect visitations recorded in the Buckwheat treatment was predators ($\bar{x}$=2.25).

The Partridge Pea had the highest mean number of pollinators recorded across all treatments with a mean of 4.5. While having the highest recorded mean of pollinators, the Partridge Pea also has the lowest number of recorded predators ($\bar{x}$=0.75) compared to the other treatments. Partridge Pea also has the lowest number of recorded visitations by pests/other ($\bar{x}$=1.5) compared to the other treatments.
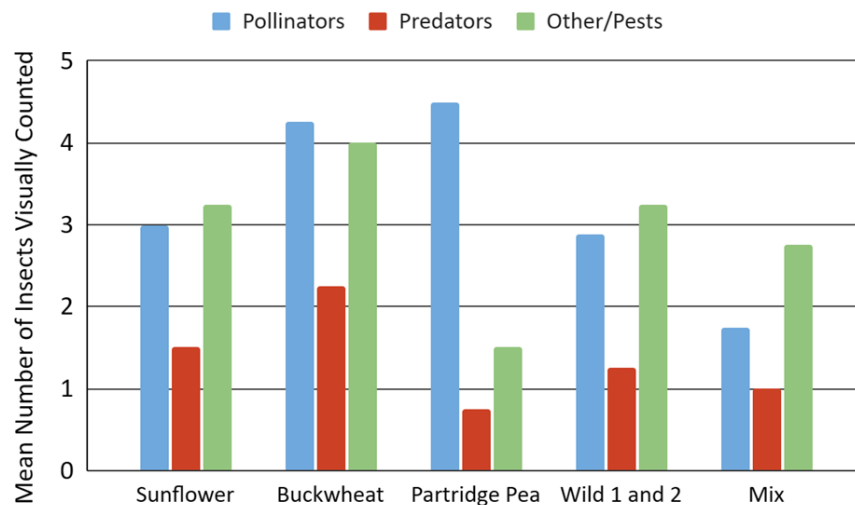

Figure 7. The mean number of insects visually identified across six treatments. The highest number of visual visitations is pollinators in the Partridge Pea. The lowest mean number of visual visitations was recorded in the Partridge Pea and were observed as predators. Partridge Pea also lowest number of recorded pests/other. Buckwheat had the highest number of observed predators.

4 Discussion

We had hypothesized that the mixed plot with the Sweet Alyssum, Buckwheat, Mighty Mustard, Sunflower, and Partridge Pea would have a higher number of parasitoids, predators, and pollinators than the wildflower plots. After reviewing the results, we observed that the mixed

plot had the least amount of chewed eggs compared to the other treatments, and the mixed plot also had the least amount of pollinators, predators and pests/other, compared to the other treatment plots. As seen in the results, there is no data for the Mighty Mustard and the Sweet Alyssum. This is because the plants did not grow due to the heavy rains that occurred when the seeds were planted and failed to germinate and grow successfully. Buckwheat was also planted by seed but had no problem with germination and flowering, while Sunflower and Partridge Pea were planted by transplants. This could have potentially skewed the results because two of the treatments didn't grow, leaving a lot of room for weeds and the mixed plot did not grow to be as thick and full as the other treatment plots. Interestingly, however, because two out of five plants in the mixed plot failed to flower, the plots with the highest number of successful species were the two wildflower plots, with at least seven native and exotic plant species each. Therefore, when considering the wildflower plot as the most diverse group of plants, the notion that increased biodiversity contributes to pest control is supported by the higher number of BMSB sentinel eggs predated by chewing and sucking.

We observed an uptrend in visually-identified pollinator visitations with Sunflower, Buckwheat and Partridge Pea and a downtrend across the combined effects of wildflower plots and the mixed plot. There is no observed trend with the visually identified predators or pests/other. It is important to highlight that it was difficult to conduct the weekly visual visitations due to the weather being unfavorable on some weeks. The weather being so rainy, we believe, skewed some results because there were a lot of insects that weren't out during the storms. The combination of insects on the sticky cards did not show a strong trend with any insects across the six treatments, even with the results of the two wildflower plots combined. This can be ein part explained by the high mobility of flying insects, as found in other studies (e.g., Shrestha, 2017). There was also no trend with the sentinel eggs possibly due to Buckwheat and the Wild 2 plot having an entire egg mass that was chewed off which threw off the results as well.

Comparing yellow and clear sticky cards, it was clear (no pun intended) that the yellow cards had a higher mean number of insect catches. This could be likely due to the fact that the color yellow has been known to be more attractive to pollinators, predators, parasitoids and other insects. The clear cards have no biases with color therefore they catch anything that happens to be flying, or crawling by, although we have noticed some insect pests, such as stink bugs and tarnished plant bugs, to be more commonly caught via clear sticky cards. In this instance there were no biases for the parasitoids or predators when it came down to card color. Using clear cards could be more beneficial for this study because it doesn't attract any pollinators, risking their lives.

When considering the mixed plot as the most diverse group of plants, virtually none of our results supported our initial hypothesis given that single plant species were very effective at attracting beneficial insects such as predators, parasitoids and pollinators. It still was surprising to see that the combined results of wild 1 and wild 2 plots had some higher results than the mixed plot for some of the insect residents recorded there. Although, it was even more surprising to see that Sunflower, Buckwheat and Partridge Pea alone in their plots had higher

mean values than the mixed plot as well. It should be noted that the experiment was executed in what was previously existing pollinator garden area (now a young apple cider tree block), resulting in a potentially established ecosystem with highly attractive host plants, at least five of which comprised the wildflower plots in our study.

Overall, a lack of comprehensive statistical analyses due to the use of a single location limits our ability to draw major conclusions. If this experiment were to be carried out again, the most important thing would be to replicate the treatments in different locations for stronger statistical power and to reduce variability. It would also be more beneficial to use all transplants or all seeds in order to get more balanced results.

5 Conclusion

In this study we hypothesized that the mixed plot with Sunflower, Buckwheat, Partridge Pea, Mighty Mustard, and Sweet Alyssum would have a greater number of parasitoids, predators, and pollinators than the wildflower plots. It was observed that the mixed plot had the least amount of visually identified insect visitations aside from predators. It was also found that the mixed plot had the lowest amount of sticky trap catches for pollinators, predators and pests/other insects. However, the wildflower plots had the highest number of species and would have to be objectively considered the most diverse plot. In this light, we found partial support to the notion that increased biodiversity contributes to pest control.

6 Acknowledgements

7. References

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