Parasitic Hymenopteran Food and Habitat Preferences in Urban Agriculture

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ABSTRACT

Since the first introduction of parasitoids to control pests in the 1920s, various species of parasitoids have been used as biological control agents in agricultural pest management. This research investigates the theory that parasitic Hymenoptera have food and habitat preferences, and by supporting them with floral resources, biological control can be strengthened. From collection data based in San Francisco Bay Area, three major groups of parasitoids were present on the urban farms including, Chalcidoidea, Cynipoidea, and Braconidae. Based on the parasitoids' visiting frequencies of various plants, it is apparent that these parasitoids have food and habitat preferences, and different subfamilies and families of wasps visit different types of flowers. Chalcidoidea were present in large numbers on many flowering plants because of their wide variety of feeding behavior. Cynipoidea: Alloxystinae are hyperparasitoids and were found on certain flowers with nectar. Braconidae: Aphidiinae are parasitoids and Aphidiinae were found on some flowers with nectar as well. By recognizing the different wasp subfamilies' varying presence on different flowering plants, biological control can be reinforced when taking into account of parasitoid's floral habitat preferences, and types of flowers to plant to best support sustainable agriculture.

KEYWORDS

parasitoids, biological control, chalcidoidea, cynipoidea, and braconidae

INTRODUCTION

In sustainable cities, organic urban agriculture takes an increasingly active role. Urban agriculture provides socioeconomic benefits such as fresh food and recreational values such as green space. Some of the challenges faced by urban community farms include pests such as aphids, snails, and different types of caterpillars. Through biological control, pests can be reduced, and as a sustainable and organic agroecological approach, pesticide use is decreased and eliminated. Since these farms are located in urban areas, the use of pesticides would otherwise increase the risks of harming the local community's environmental health due to proximity. The common adverse health and ecological effects include cancer, neurological and reproductive harms for nearby communities, respiratory problems caused by breathing in toxic pesticides, and the increased risks of crops becoming pesticide resistance as the pesticide dosage increases each year. Therefore, it is crucial to examine sustainable pest reduction approaches towards urban agriculture. Biological control can help organic gardeners or farmers achieve their goals of reducing pests and pesticide use.

Biological control is one possible way to minimize insect pest densities (Jervis and Heimpel 2005). One method in applying biological pest control is through parasitic Hymenoptera (Downes and Dahlem 1987). The presence of natural enemies enhances organic agriculture crop yield. Hymenopteran superfamily parasitoid wasps naturally control unwanted pests by laying their eggs in at times unwanted host arthropods. The parasitism later causes the death of the hosts, eliminating the pests. Studies about parasitoids have been done on select parts of the world, such as recording of the genus *Agathis* Latreille (Hymenoptera, Braconidae, Agathidinae) from Spain (Belokobylskij and Jervis 1998), but more research can be done on parasitoids in the San Francisco Bay Area. Specifically, given the parasitoid's contributions to ecosystem services, more research can be done on parasitic Hymenoptera's food and habitat preferences to attract and support the parasitoids more. Although natural enemies are essential, not all habitats support them in the same way.

Modifying natural habitats to attract and support natural enemies improves biological pest reduction. Plant species composition has dramatic effects on wasps (Jervis et al. 1993). Parasitoids are known to be attracted to habitats with flowering plants (Jervis and Kidd, 1986). These plants provide them with food, including floral nectar, extrafloral nectar, pollen, and honeydew. A study related to wasps and flowers examined in this research showed that the presence of alyssum (*Lobularia maritima*) with leafrollers increased Braconidae's longevity and fecundity (Berndt 2005) because alyssum provided the wasps with necessary floral resources to increase the average lifetime. Moreover, female parasitoids with the support of alyssum flowers lived seven times longer than those without floral support, and male parasitoids lived three times longer than those without floral resources. For Hymenopteran reproduction, the sex ratio was equal when female parasitoids had access to flowers (Henneicke et al. 1992). Enhancing and favoring natural enemies are desirable for increasing the natural enemy population (Siekmann et al. 2004). Providing flowering plants as alternative food sources such as pollen and non-host food improves parasitoid longevity and fecundity, which are desirable for increasing the natural enemy population (Siekmann et al. 2004). More research is needed to find out what floral species can better attract parasitoid Hymenoptera for biological control in urban agriculture.

This study examines the difference in food and habitat preferences for parasitic Hymenoptera species in urban gardens of the San Francisco Bay Area in California. I hypothesize that parasitic Hymenoptera species have variations in preferences for food and habitat sources. I predict that in comparison to other species of Hymenoptera clade, parasitoids have food and habitat preferences, and can provide positive effects on biodiversity, pollination, and biological control through natural enemies.

METHODS

Study site

We conducted our study in 12 urban community farms in the eastern San Francisco Bay Area of Northern California. The farms were selected based on similar farm sizes (Arnold and Altieri 2016). I conducted my agroecology research with Professor Miguel Altieri and mentor Joshua Arnold.

Study species

We focused our study on parasitic Hymenoptera Chalcidoidea (superfamily), Cynipoidea (superfamily), and Braconidae (family). Parasitoids wasps with very large groups include Chalcidoidea and Braconidae. Hymenoptera parasitism is unique to parasitoid wasps, and parasitoids can vary in methods of parasitism. The first method is koinobiont, and as an endoparasite, the wasp lays its larvae inside the host. While its larvae are still developing, hosts continue to feed and grow. The second method is idiobiont, where the wasp paralyzes the host immediately.

Chalcidoidea superfamily

Parasitoid wasps in Chalcidoidea parasitize on a wide range of hosts and feeding behaviors. They lay eggs on the hosts that consume the hosts. By parasitizing aphids, caterpillars, and other pests, Chalcidoidea's long-term survival improves and biological control is improved (Heinz 1990).

Cynipoidea superfamily

Parasitic Hymenopteran wasps from Cynipoidea were mostly identified to Charpidae Alloxystinae, which are hyperparasitoids.

Braconidae family

Parasitoid wasps from the Braconidae family were identified to the Aphidiinae subfamily. Aphidiinae is koinobiont endoparasitoids that parasitize aphids. They parasitize over forty-one host aphid species as a cosmopolitan specie (Charles and Paine 2016). The wasps complete their life cycle's reproduction phase by parasitizing aphids and laying their eggs on the hosts (Souza 2018).

Plant species

I observed plants that were common across all sites and restricted my analysis to specific plant species that had at least fifteen individual wasps present.

The following are the plants observed:

1. Alyssum (*Lobularia maritima)*
2. Amaranth (*Amaranthus spp*.)
3. Borage/starflower (*Borago officinalis*)
4. Buckwheat (*Fagopyrum esculentum*)
5. Calendula (*Calendula offiicinalis*)
6. California lilac (*Ceanothus spp.*)
7. Celery (*Apium graveolens*)
8. Comfrey (*Symphytum spp*.)
9. Coriander (*Coriandrum sativum*)
10. Cornflower (*Centaurea cyanus*)
11. Coyote bush (*Baccharis pilularis*)
12. Dill (*Anethum graveolens*)
13. Hemlock (*Conium maculatum*)
14. Marigold (*Tagetes erecta*)
15. Mexican marigold (*Tagetes lucida*)
16. Mullein (*Verbascum Thapsus*)
17. Nasturtium (*Tropaeolaceae spp*.)
18. Nettles (*Urtica spp*.)
19. Oregano (*Origanum vulgare*)
20. Parsnip (*Pastinaca sativa*)
21. Purple sage (*Salvia spp*.)
22. Sage Hot lips (*Salvia microphylla spp*.)
23. Sunflower (*Helianthus*)
24. Yarrow (*Achillea millefolium*)

Wasp collection

Insect sampling

My mentor Joshua Arnold visited the farms every two weeks for two years from May to October in 2018 and 2019 and took samples of the plant groups. He vacuumed each plant for a total of 15 seconds with a TORO brand leaf vacuum. The records included plant density, floral diversity and abundance, ground cover types (mulch, leaf litter or bare soil), crop diversity, farm size, and surrounding percentage of the impervious surface.

Wasp sorting

To begin sorting the wasps, I took the wasps out of the freezer and placed them in the Relaxing Chamber (Appendix A). After leaving the wasps in the Relaxing Chamber for 20 minutes, I brushed out the contents and labeled them wasp and non-wasp. I marked information such as the farm, plant, and the date the wasps were collected from in the labels (Appendix A).

Wasp identification

To determine the wasp identities, I used books to manually identify the wasps under the microscope, using small paint brush and needle point clay tool to help us turn the wasps. The books used include: Hymenoptera of the World: An Identification Guide to Families by Goulet, H. and Huber, J.T. (1993), Annotated Keys to the Genera of Nearctic Chalcidoidea (Hymenoptera) by Gary A. Gibson, James B. Woolley, John T. (1997), and Manual of the New World Genera of the Family Braconidae (Hymenoptera) by Wharton, R.A., P.M. Marsh, M.J. (1997). The parasitic Hymenoptera are identified to subfamilies. I recorded the Wasp ID, and related data such as farm, plant, and the date the wasps were collected.

Analysis

From the data collection (Appendix B), I narrowed the observations to plants that had at least fifteen individual wasps present across the different groups of wasps, and performed my analysis based on these observations.

Anova: Single Factor. To first find out if there were any variance for the wasps' presence across all flowers, I did this test to determine the variance in the wasp collection and plant relationship (Table 1).

Chi-squared Test and Post Hoc Analysis with Bonferroni Correction. To determine if parasitic Hymenoptera had floral preferences, I performed a Chi-squared test in R (R Development Core Team 2014) (Fox 2005). The Chi-squared test shows if there's an association between two categorical variables. From the test, it is shown that there is significant data (Table 2). However, since Chi-squared arguments are made for 2x2 contingency tables and my data is larger than 2x2, I ran a post hoc test with a Bonferroni correction to get significant p-values and determine the relationship between specific taxa groups and different flowers (Table 3).

RESULTS

Preference by taxonomic group

Figure 1-4. Summary of parasitic Hymenoptera collection used in the study. The parasitoids' presence on plants were divided into four groups: (1) Cynipoidea Superfamily, (2) Chalcidoidea Superfamily, (3) Braconidae Family and (4) Other Wasps.

From the wasp collection results, the plants with the most Chalcidoidea found are Coriander (*Coriandrum sativum*), Marigold (*Tagetes erecta*), Fennel (*Foeniculum vulgare*), and Yarrow (*Achillea millefolium*) (Figure 1). The y-axis depicts the number of Chalcidoidea wasps found. The x-axis denotes the plants species and their scientific names.

1.

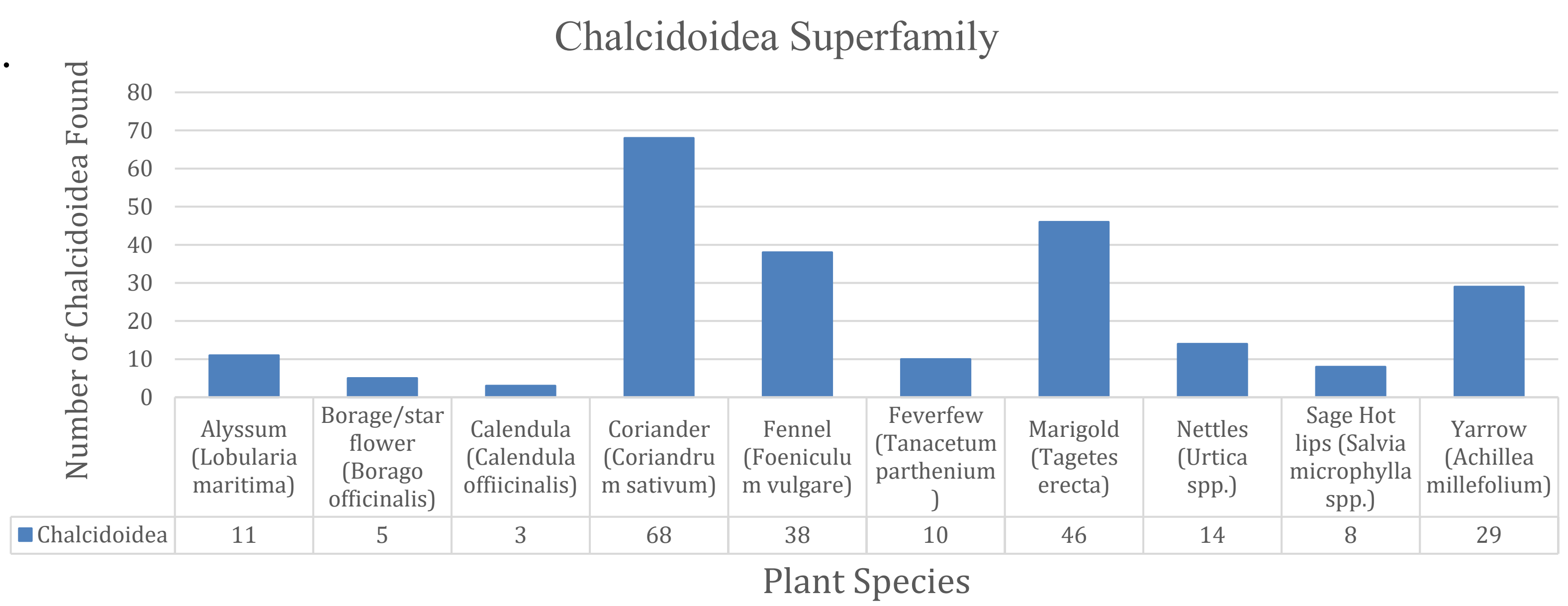


Figure 1. Graph showing the results of Chalcidoidea. Coriander had the greatest number of Chalcidoidea found (68), Marigold had the second most number of Chalcidoidea found (46), Fennel had the third most Chalcidoidea found (38) and Yarrow had the fourth most Chalcidoidea found (29).

From the wasp collection results, the plants with the most Cynipoidea found are Coriander (*Coriandrum sativum*), Borage (*Borago officinalis*), Fennel (*Foeniculum vulgare*), and Alyssum (*Lobularia maritima*) (Figure 2). The y-axis depicts the number of Cynipoidea wasps found. The x-axis denotes the plants species and their scientific names.

2.

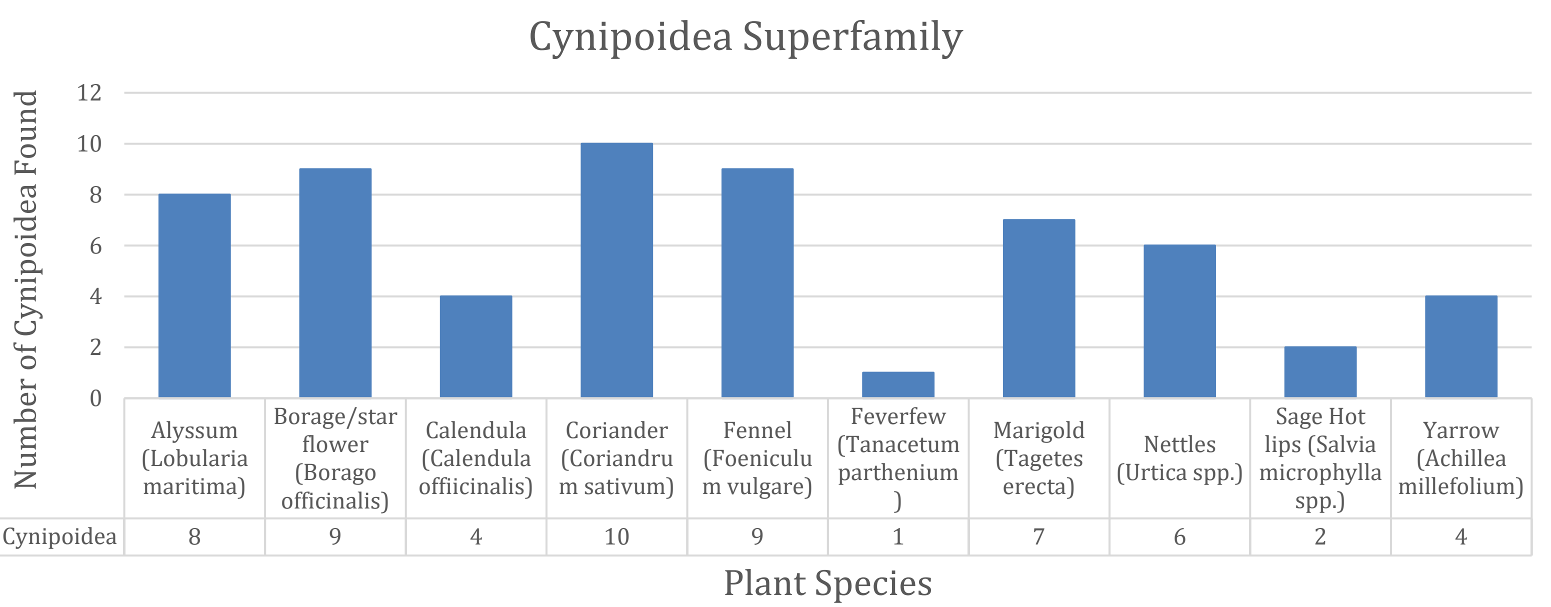


Figure 2. Graph showing the results of Cynipoidea. Coriander had the greatest number of Cynipoidea found (10), Borage had the second most number of Cynipoidea found (9), Fennel had the third most Cynipoidea found (9), Alyssum had the fourth most Cynipoidea found (8), and Marigold had the fifth most Cynipoidea found (7). However, the numbers were similar and were in the range of (7-10).

From the wasp collection results, the plants with the most Braconidae found are Yarrow (*Achillea millefolium*), Marigold (*Tagetes erecta*), Coriander (*Coriandrum sativum*) and Fennel (*Foeniculum vulgare*) (Figure 3). The y-axis depicts the number of Braconidae wasps found. The x-axis denotes the plants species and their scientific names.

3.

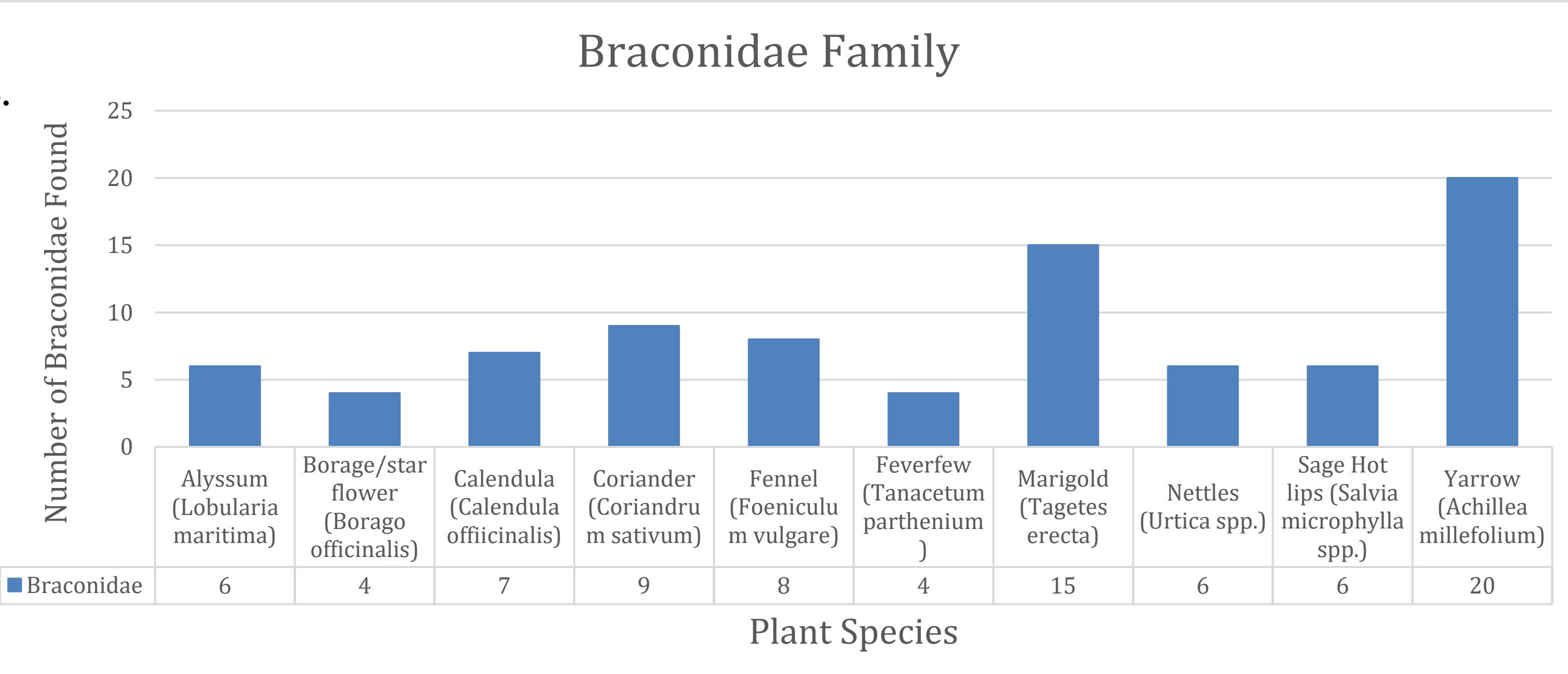
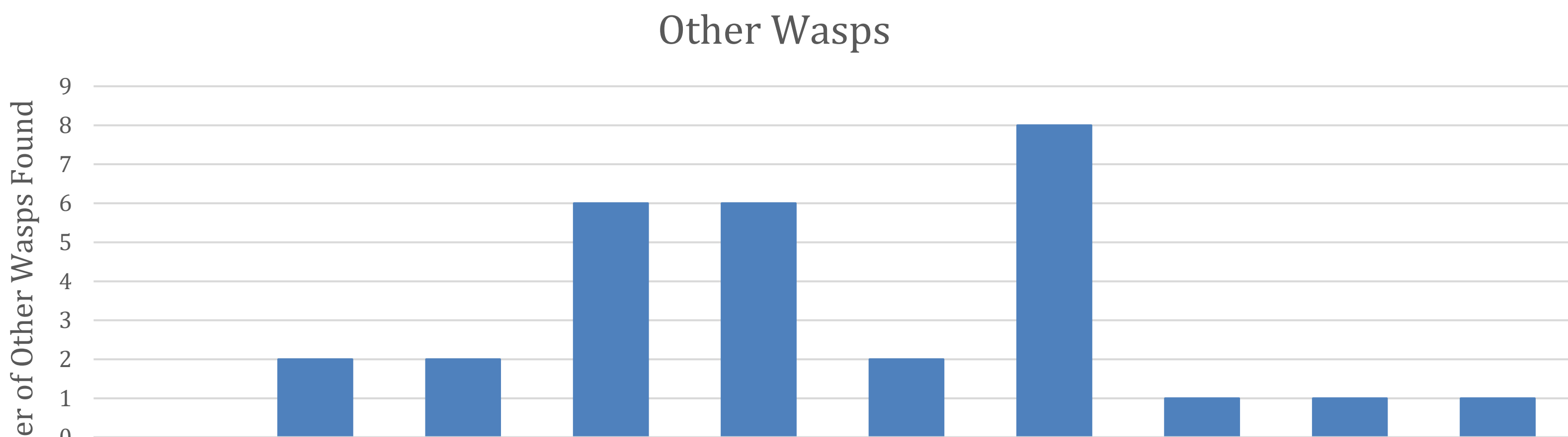


Figure 3. Graph showing the results of Braconidae. Yarrow had the greatest number of Braconidae found (20), Marigold had the second most number of Braconidae found (15), Coriander had the third most Braconidae found (9), Fennel had the fourth most Braconidae found (8), and Calendula had the fifth most Braconidae found (7). This was similar to that of Chalcidoidea and Cynipoidea in that the plants with the most wasps found included Coriander, Fennel, Marigold, and Yarrow.

From the wasp collection results, the plants with the greatest number of other wasps found are Marigold (*Tagetes erecta*), Coriander (*Coriandrum sativum*) and Fennel (*Foeniculum vulgare*) (Figure 4). The y-axis depicts the number of other wasps found. The x-axis denotes the plants species and their scientific names.

4.



	Alyssum (Lobularia maritima)	Borage/star flower (Borago officinalis)	Calendula (Calendula offiicinalis)	Coriander (Coriandru m sativum)	Fennel (Foeniculu m vulgare)	Feverfew (Tanacetum parthenium)	Marigold (Tagetes erecta)	Nettles (Urtica spp.)	Sage Hot lips (Salvia microphylla spp.)	Yarrow (Achillea millefolium)
Other	0	2	2	6	6	2	8	1	1	1

Plant Species

Figure 4. Graph showing the results of other wasps. Marigold had the greatest number of other wasps found (8), Fennel had the second most number of other wasps found (6), Coriander had the third most other wasps found (6).

The data summary showed that Chalcidoidea were found across many flowers in varying counts (Table 1), and that Chalcidoidea's presence varied the most across the flowers they were found on. The data summary also represented Braconidae's appearance across the different flowers and it varied less in counts when compared to Chalcidoidea's, clearly showing that depending on the family or subfamily of parasitoids, they have different preferences.

SUMMARY

Groups	Count	Sum	Average	Variance
Chalcidoidea	10	232	23.2	461.9556
Cynipoidea	10	60	6	9.7778
Braconidae	10	85	8.5	26.2778
Unknown	10	29	2.9	7.4333

Table 1. The wasps' presence varied in numbers and across the flowers. Chalcidoidea had the largest average number of wasps found on the plants (23.2), and with high variance (461.95). The data depicted a wide range of presence on different flowers. Braconidae had a moderate sample variance (26.28).

For the relationships between the wasps and the top six flowers with the most wasps present (more than 15 individual wasps present), Chi-squared showed that there's an association between two categorical variables (Table 2), the wasps and the flowers. The flowers with more than 15 wasps present included Alyssum (*Lobularia maritima*), Borage (*Borago officinalis*), Fennel (*Foeniculum vulgare*), Marigold (*Tagetes erecta*), Yarrow (*Achillea millefolium*), and Nettles (*Urtica spp*.).

Category	Calculation
X-squared	46.187
Degrees of freedom	15
p-value	4.966e-05

Table 2. There is an association between the wasps and the flowers. Wasps have floral preferences, $p<0.01$. Chi-squared test showed statistically highly significant p-value<0.001 between the wasps and flowers.

Data analysis also showed that there were associations between specific wasp taxa and plants. The first pair of association are Cynipoidea and Borage (*Borago officinalis*). The second pair of association is Braconidae with Fennel (*Foeniculum vulgare*) and Yarrow (*Achillea millefolium*).

	Type of Wasps			
Flowers	**Cynipoidea**	**Chalcidoidea**	**Braconidae**	**Other**
Alyssum (*Lobularia maritima*)	0.6271	1	1	1
Borage (*Borago officinalis*)	0.0075*	0.1458	1	1
Fennel (*Foeniculum vulgare*)	1	1	0.0491*	0.38
Marigold (*Tagetes erecta*)	1	1	1	1
Yarrow (*Achillea millefolium*)	1	1	0.0182*	1
Nettles (*Urtica spp.*)	1	1	1	1

Table 3. Chisq. with Post hoc Bonferroni correction showed significant P-values for parasitic Hymenoptera floral preferences**.** Asterisks denote significant P-values. There is an association between the two variables, Cynipoidea and Borage (*Borago officinalis*). Cynipoideas are more likely to be found on Borage $p<0.01$. There is also an association between the two variables, Braconidae and Fennel (*Foeniculum vulgare*), as well as Braconidae and Yarrow (*Achillea millefolium*). Braconidae are more likely to be found on Fennel and Yarrow $p<0.05$.

DISCUSSION

Introduction

With urban areas as the fastest growing landscape in the present day, sustainable urban farms are important in that they provide the community with many ecosystem services and goods. These benefits include crop yield for food security, green space, and recreation space. The urban farms also improve biodiversity and pollination. My research hypothesized that by planting certain types of flowers, gardens could attract parasitoid wasps with the goal to eliminate pests. The results showed that there were associations between Cynipoidea and borage (*Borago officinalis*), as well as Braconidae with fennel (*Foeniculum vulgare*), and yarrow (*Achillea millefolium*). Different wasp subfamilies have different floral preferences. Gardeners can plant flowers to strengthen biological control agent parasitoids, including attracting more parasitoids and supporting parasitoids with floral resources that improve their longevity and fecundity (Jervis and Heimpel 2005).

Preferences for floral resources according to wasp taxa

From the research results, Chalcidoidea superfamily had a higher average (23.2) and a wider range (65) with a large sample variance (461.96), meaning that although more Chalcidoidea wasps were observed from the plants, Chalcidoidea's numbers varied widely depending on the plant (Figure 1 and Table 1). Chalcidoidea were found in large numbers on many of the flowers. These wasps preferred flowering plants that offered floral food resources. These flowers include fennel (*Foeniculum vulgare*), and yarrow (*Achillea millefolium*) (Figure 1). My data included phytophagous Chalcidoidea families Eurytomidae, Eulophidae, and Pteromalidae in large numbers across the flowering plants. Phytophagous Chalcidoidea associate with forty-four different plant families (Noyes, J.S. 2019) and are herbivore wasps. However, my data also included Aphelinidae and Encyrtidae, which are useful in biological control. The two families were found on a variety of flowers, including marigold (*Tagetes erecta*), sage hot lips (*Salvia microphylla spp.*), borage/starflower (*Borago officinalis*), yarrow (*Achillea millefolium*), calendula (*Calendula offiicinalis*) and purple sage (*Salvia spp*.). Aphelinidae and Encyrtidae wasps parasitize pests including aphids and variety of caterpillars to complete their reproduction and life history cycle. However, we do not know if the flowers had host insects on them. This research does confirm that Chalcidoidea's appearances varied based on floral preference and confirmed the alternative hypothesis that parasitic Hymenoptera has food/habitat preferences.

The Cynipoidea superfamily showed a significant association with borage ($p\text{-value} < 0.01$), confirming my alternative hypothesis that parasitoid wasps have floral preferences (Table 3). The identification process further identified the wasps from Cynipoidea superfamily as Charpidae: Alloxystinae. Charpidae: Alloxystinae are hyperparasitoids. Cynipoidea had floral preferences and preferred borage (*Borago officinalis*) with nectar, confirming the hypothesis of parasitic Hymenoptera having food/habitat preferences.

The Braconidae family showed significant association with fennel and yarrow ($p\text{-value} < 0.05$), which confirmed the alternative hypothesis that parasitic Hymenoptera has food/habitat preferences (Table 3). Through wasp identification, I further identified wasps from the Braconidae family as Aphidiinae. The Aphidiinae subfamily uses aphids as their hosts. Aphidiinae were found on fennel and yarrow, acting as parasitoids, confirming my alternative hypothesis that different subfamilies and families of wasps visit different types of flowers.

My research data found floral preferences among different families and subfamilies of wasps. This was evident in the varying Chalcidoidea presence on different plant species. My alternative hypothesis was also confirmed when the data analysis showed association between Cynipoidea superfamily and borage (*Borago officinalis*), as well as the Braconidae family with fennel (*Foeniculum vulgare*) and yarrow (*Achillea millefolium*). Therefore, it is evident that parasitic Hymenoptera has food/ habitat preferences.

Limitations and future directions

Limitations of this research include research locations and farm sizes. This research was based in San Francisco Bay Area, California, USA. In some cases of parasitoid wasp research done in other locations and countries, different subfamilies of wasps could be found. Additionally, our study focuses on the flower species common in urban gardens of the area; however, there may be other types of flowers that also attract parasitoid wasps not included in this research. For future directions, parasitoid preferences can be used in the management of urban agriculture.

Management implications

For economic value and societal benefits, urban vegetation is valued at $33 billion annually with projected annual food production of 100–180 million tons (Clinton et al. 2018). Through urban agriculture, it is also expected to contribute to energy savings ranging from 14 to 15 billion kilowatt-hours, and nitrogen sequestration between 100,000 and 170,000 tons, with the benefits of avoiding stormwater runoff between 45 and 57 billion cubic meters annually (Clinton et al. 2018). In total, it is estimated to save $80–160 billion annually (Clinton et al. 2018). Furthermore, urban agriculture also increases biodiversity patterns, including crop diversity, and improves essential ecosystem services such as pollination (Lin et al. 2015). Urban agriculture can take many forms from larger community farms to school gardens in cities. One common concern is pest management.

Chalcidoidea and Cynipoidea parasitoids are known as classical biological control agents introduced to eliminate pests. My research has confirmed the appearance of parasitoids in the San Francisco Bay Area and that parasitic Hymenoptera has floral/food preferences. Management implications and potentials indicate that urban farmers can utilize the knowledge of parasitic Hymenoptera floral/food preferences in enhancing biological control experiences. Among parasitic Hymenoptera, Chalcidoidea is one of the most successful examples of applied biological control, and over eight-hundred species of Chalcidoidea have been used for biological control (Noyes 2003). The Aphelinidae and Encyrtidae families appeared in large numbers in my research and are also known to parasitize crop pests (Clausen 1978). Therefore, my research results can support the biological control of crop pests in the San Francisco Bay Area's urban agriculture. With sufficient alternative food sources such as alternative prey, crop residue, and organic matter, predators can establish in the crop before seasonal pests increase, which effectively enhances the natural enemy population (Settle et al. 1996). Given what we know now, we can plant flowers to strengthen biological control agent parasitoids by attracting and supporting more parasitoids with floral resources that improve their longevity and fecundity.

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APPENDIX A: Wasp Sorting

Procedures

1. To preserve the wasps, I placed the wasps in the freezer. To begin sorting, I took the nets wrapped by plastic bags out of the freezer, then placed the nets in the warming Relaxing Chamber to unfreeze the wasps.
2. Relaxing Chamber Set Up: The relaxing chamber has an empty ice chest as the foundation, a plate to place the wasps, and beneath the plate, two glass jars of boiled water. The wasps are in the relaxing chamber for 20 minutes.
3. After taking the nets out of the relaxing chamber, I brushed the wasps out of the nets using a small paintbrush. Then I placed the contents from the nets under the microscope and sorted them into two vials where one vial is labeled as wasp, and the other is labeled as non-wasp. Also, I marked information such as the farm, plant, and the date the wasps were collected from in the labels.

APPENDIX B: Research Results

Total Number of Identifications

In 2018, 1200 individual wasps were identified, and in 2019, 1400 individual wasps were identified.