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How Weeds Affect Insects in Mango Cultivation of South Florida

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FLORIDA INTERNATIONAL UNIVERSITY

Miami, Florida

HOW WEEDS AFFECT INSECTS IN MANGO CULTIVATION OF SOUTH FLORIDA

A thesis submitted in partial fulfillment of the

requirements for the degree of

MASTER OF SCIENCE

in

ENVIRONMENTAL STUDIES

by

Blaire Mallory Kleiman

2021

To: Dean Michael R. Heithaus
College of Arts, Sciences and Education

This thesis, written by Blaire Mallory Kleiman, and entitled How Weeds Affect Insects in Mango Cultivation of South Florida, having been approved in respect to style and intellectual content, is referred to you for judgment.

We have read this thesis and recommend that it be approved.

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Florida International University, 2021

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DEDICATION

Dedicated to my family, my father, and my sister for their continued support and allowing storage of field equipment and specimens in our home, as well as to my father for his weed-whacking efforts in the field. Thank you to my many volunteers both in the field and lab who dedicated many hours, effort, and ideas to my thesis- Nisrine Toury, Emie Marin, Helen Martinez, Daphne Zapsas, and Guthsa Altena, who all enabled this project to fruition. Special thanks to Nisrine for her invaluable work and support both in the lab and field. In memory of my sweet golden and best friend, Odie Kleiman.

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ABSTRACT OF THE THESIS
HOW WEEDS AFFECT INSECTS IN MANGO CULTIVATION OF SOUTH
FLORIDA

by

Blair Mallory Kleiman

Florida International University, 2021

Miami, Florida

Professor Krishnaswamy Jayachandran, Major Professor

The use of weeds as insectary plants is an emerging management tactic by agroecologists and entomologists to sustain beneficial insect species. Fallow lands have always been used by insects and are an important part of their diet in fragmented ecosystems. Weeds provide floral resources to beneficial insects such as pollinators, parasitoids, and predators and resources to keep them within a field in between crop flowering. Using weeds as a tool in tropical fruit production reliant on pollination like Mango (*Mangifera indica*) allows farmers to reduce herbicide use, increases the biodiversity of both plants and insects, and increases pollination of crops by native insects. This study examines the plant-insect ecological interactions when weeds are left within a farm and finds that the presence of weeds strongly correlated with increased mango yield, flower visitors and parasitoid insects on mango trees, and the insect orders Hymenoptera, Diptera, Coleoptera, Neuroptera, and Thysanoptera on mango trees. The species of weeds encountered in mango farms of South Florida were identified, and weeds were found to support more pollinators, predators, and parasitoids than pest insects. Weeds also increased soil carbon and decreased soil pH.

TABLE OF CONTENTS

SECTION	PAGE
1. Introduction.....	1
1.1 Research Objective	3
1.2 Research Questions.....	3
1.3 Hypotheses.....	4
2. Literature Review.....	5
2.1 Pollinators	8
2.2 Floral rewards	16
2.2.1 Nectar.....	16
2.2.2 Pollen	17
2.3 Pollinator Networks	21
2.4 Parasitoid Insects	23
2.5 Predatory Insects.....	24
2.6 Debate	25
2.7 Mango	30
3. Methodology.....	36
3.1 Site Description.....	36
3.2 Field Data Collection	37
3.2.1 Statistical Analyses	38
3.2.2 Farm Maintenance	39
3.2.3 Weed Species.....	41
3.3 Pollen Analysis	42
3.4 Rearing.....	43
3.5 Fruit Yield.....	44
3.6 Soil Analysis	45
3.6.1 Total Carbon and Nitrogen (Soil)	45
3.6.2 Total Phosphorus (Soil)	45
3.6.3 Soil pH	46
3.7 Chlorophyll Analysis	46
4. Results.....	47
4.1 Insects on Mango	47

4.1.1 Parasitoids	54
4.1.2 Lacewings	54
4.1.3 Flower Visitors of Mango	55
4.1.4 Predators	59
4.1.5 Insect Behavior and Mango Diseases	61
4.1.6 Insects on Weeds and Mango	64
4.2 Pollen Analysis	72
4.3 Rearing	77
4.4 Fruit Yield	79
4.5 Soil Analysis	81
4.5.1 Total Carbon and Nitrogen	81
4.5.2 Total Phosphorous	83
4.5.3 Soil pH	84
4.6 Chlorophyll Analysis	85
5. Discussion	86
6. Conclusion	88
References	91

LIST OF TABLES

TABLE	PAGE
Table 1 Weed Family and Species in Mango Farm.....	47
Table 2 Multivariate ANOVA Tests for effects of treatment and tree age on arthropod types.....	50
Table 3 Insect Types and Fruit Yield on Mango With or Without Weeds- significant differences have p-value in bold. GLM Multivariate ANOVA (MANOVA).	50
Table 4: GLM- effects of treatment on counted fruit yield and main types of arthropods associated with mango trees.....	51
Table 5 Multivariate ANOVA Tests for effects of tree age and treatment on arthropod orders.....	52
Table 6 Insect Orders on Mango With or Without Weeds- significant differences have p-value in bold. GLM Multivariate ANOVA (MANOVA).....	52
Table 7 Lacewing (Neuroptera) Adults/Larvae /Eggs on Mango Trees With/Without Weeds T-Test.....	55
Table 8 Spiders on Mango Trees T-Test.....	61
Table 9 MANOVA Mango Diseases & Insect Behavior.....	62
Table 10 T-test Mango Diseases & Insect Behavior	63
Table 11 Chi-Square Table Insect Types on 4 Common Weed Species.	65
Table 12 T-test Insect Families on Mango With/ Without Weeds	71
Table 13 Pollen Carrying Insects in Mango and Weed Flowers	73
Table 14 Soil Carbon and Nitrogen Weedy or No-Weed Trees, T-Test.	82
Table 15 Total Phosphorous compared by treatment at the beginning and end of study, T-Test.....	83
Table 16 One-way Anova Comparing Total Soil Phosphorous Weeds vs. No-Weeds, Beginning and End Samples Combined	84
Table 17 Soil pH Between Treatments and Over Time, Univariate ANOVA.....	85

Table 18 SPAD Chlorophyll Readings for Mango Leaves, Weeds vs. No Weeds.
Univariate ANOVA. 86

LIST OF FIGURES

FIGURE	PAGE
Figure 1 Types of Insects	4
Figure 2 Farm Treatment Map of Weed and No-Weed Trees	37
Figure 3 Left: <i>Macroptilium lathyroides</i> , "Phasey Bean" purple flowers, non-native.	41
Figure 4 Right: <i>Oxalis</i> , "Wood Sorrel", non-native.....	41
Figure 5 Left: <i>Ceratina</i> bee with Pollen	43
Figure 6 Right: <i>Ornidia obesa</i> Hoverfly with Pollen	43
Figure 7 Caterpillar Rearing from Weeds.....	43
Figure 8 Lacewing Eggs on Mango Inflorescence	55
Figure 9 Top left Halictidae in <i>Bidens alba</i> , top right Skipper (Hesperiidae) in <i>Lantana camara</i> , bottom: Honey bees (<i>Apis mellifera</i>) in <i>Bidens alba</i>	57
Figure 10 Left: Blow Fly (Calliphoridae), Right: Ants (Formicidae), in Mango Bloom ..	58
Figure 11 Coccinellidae (Lady Beetle) eggs on <i>Bidens alba</i> with Aphididae Prey.....	60
Figure 12 Hopper Nymph and Adult on Fruit Stem	63
Figure 13 Left: Parasitoid Wasp Feeding in <i>Conoclinium coelestinum</i> with pollen, Middle: Syrphidae (<i>Copestylum mexicanum</i>) Feeding in <i>Bidens alba</i> / <i>Conoclinium coelestinum</i> , Right: Syrphidae (<i>Copestylum mexicanum</i>) pollen on thorax	74
Figure 14 Left: <i>Sarcophaga haemorrhoidalis</i> Feeding in Mango Flower carrying multi- species pollen, Right: pollen on tongue	74
Figure 15 Left: Apidae (<i>Ceratina</i>) feeding in <i>Conoclinium coelestinum</i> , Right: Pollen on abdomen.....	75
Figure 16 Left: Megachilidae feeding in <i>Bidens alba</i> flower, Right: Pollen on Body	75
Figure 17 Honey bee <i>Apis Mellifera</i> feeding in Mango Flower, Right: Blue Mist Flower (<i>Conoclinium coelestinum</i>) Pollen.....	76
Figure 18 Left: Mango Pollen from Halictidae feeding in Mango Flower in Weed Treatment. Right: Pollen from Halictidae: Mango and <i>Bidens alba</i>	76

Figure 19 Left: *Crotolaria incana* Pollen. Right: Red Morning Glory (*Ipomoea hederifolia*) Pollen77

Figure 20 Scale (Coccidae) Parasitoids Reared from Mango Leaf Scale78

Figure 21 Chalcidae Parasitoid Reared from Moth Caterpillar78

Figure 22 Membracidae Nymph Reared.....79

Figure 23 Hawkmoth Caterpillar Reared from Spermacoce.....79

Figure 24 Mango “Keitt” Harvest.....81

LIST OF GRAPHS

GRAPH	PAGE
Graph 1 Mean Number of Types of Insects on Mango by Treatment ± SE. Significant Difference Indicated Above Each Type: NS P>0.05, * P ≤ 0.05, ** P ≤ 0.01, **** P ≤ 0.001, **** P ≤ 0.0001.....	51
Graph 2 Mean Number of Pollinator Insect Orders on Mango by Treatment ± SE. Significant Difference Indicated Above Each Type: NS P>0.05, * P ≤ 0.05, ** P ≤ 0.01, *** P ≤ 0.001, **** P ≤ 0.0001. Diptera *: P=0.057 weedy mango trees.	56
Graph 3 Mean Number of Insect Orders on Mango by Treatment + SE. Significant Difference Indicated Above Each Type: NS P>0.05, * P ≤ 0.05, ** P ≤ 0.01, **** P ≤ 0.001, **** P ≤ 0.0001.....	58
Graph 4 Expected and Actual Counts of Insect Types on the No-Weed Mango Trees and <i>Bidens alba</i> reported from a Chi-Square Test. $X^2_3 = 1845.806$; P<0.0001.....	65
Graph 5 Proportions of Insect Types on all Mango Trees and <i>Bidens alba</i> reported from a Chi-Square Test. Different letters within plant species denotes significantly different numbers of those types. $X^2_3 = 2206.804$; P<0.0001.....	66
Graph 6 Percentage of Types of Insects on <i>Bidens alba</i> (Spanish needles) and Mango trees.....	67
Graph 7 Proportion of Less Frequent Insect Types on Mango and <i>Bidens alba</i> . Number of arthropod counts per plant species were 6256 for <i>Bidens alba</i> and 11,342 for <i>Mangifera indica</i>	67
Graph 8 Expected and Actual Proportions of Insect Types on Common Weeds, reported from Chi-Square Test.....	68
Graph 9 Proportions of Insect Types on Common Weeds. Different letters within plant species denotes significantly different numbers of those types reported from a Chi-Square Test. $X^2_9 = 38.065$; P<0.0001.	69
Graph 10 Insect Types on Less Frequent Weeds <i>Parthenocissus quinquefolia</i> (Virginia Creeper), <i>Cissus verticillata</i> (Possum Grape Vine), and <i>Ipomoea hederifolia</i> (Scarlet Morning Glory). Since there were few observations, counts are reported rather than proportions.....	70
Graph 11 Mean Number of Mango Fruit Harvested or Counted by Treatment ± SE. Significant Difference Indicated Above Each Type: NS P>0.05, * P ≤ 0.05, ** P ≤ 0.01, *** P ≤ 0.001, **** P ≤ 0.0001.	80

Graph 12 Soil Carbon Mean by Treatment at the Beginning and End of Study.	82
Graph 13 Mean Soil pH by Treatment at the Beginning and End of Study.	85
Graph 14 Mean SPAD Chlorophyll Readings for Mango Leaves, Weeds vs. No Weeds, for New Leaves, Green Leaves, and Old Leaves.....	86

1. Introduction

Cultivated crops are often subject to pest pressure, a major focus of agricultural entomological research for the last century. There has been a growing interest in environmentally sound pest control, using beneficial insects rather than pesticides, and this approach holds much promise for increasing food production and growing healthy crops without harmful chemicals in foods and the environment (Blaauw and Isaacs 2014). The presence of non-crop plants may be very useful in this approach, and weeds can provide resources that attract and maintain populations of beneficial insects- parasitoids, predators, and pollinators. Weeds - wild plants growing where they are not wanted – are seen as detrimental to crop production in agriculture and are often treated with herbicides. This lack of weeds diminishes beneficial insects, through the loss of floral and prey resources (Altieri & Nicholls, 2018). The role of weeds in agriculture as a tool for insect management is an emerging topic of inquiry in agroecology. The benefits of using insectary plants in farms is well known (Hogg, Bugg, & Daane, 2011), however, using weeds as such in tropical fruit production is an expansion upon previous research. Just as insects disappear with the disappearance of their weeds, they can also reappear when their weeds return (Pickett & Bugg, 1998).

There are various hypotheses that can help in understanding the interactions of weeds and insects, and why on a case-by-case instance, results vary from pest reduction to exacerbating pest populations. The “Resource Concentration Hypothesis” states the relative attractiveness of a habitat to a particular insect is based on the concentration of resource host plants or prey species. Weeds can dilute the concentration of the predominant crop plant, and therefore the attractiveness of the crop to its pests. This hypothesis is based on the concept of “Apparency”- hosts that are more apparent are more likely to be attacked (Castagneyrol et al. 2013). Crop host

plants more apparent to herbivores are more likely to be fed upon, and therefore weeds can alter crop “apparency”, and act as a sort of camouflage against pests.

The “Enemies Hypothesis” states that having more diverse plant habitats supports a greater diversity of prey insects, and thus more stable populations of natural enemies. Monocultures of crop plants are easily detected and exploited by their herbivores, which are more easily diverted and confused in a varied environment (Andow, 1991). The “Diversity Stability Hypothesis” states that increasing species diversity in an ecosystem results in increased stability. Pest outbreaks are less likely to occur in highly diverse ecosystems due to increased diversity and numbers of enemies (Burkes and Philpott, 2017; Philpott and Bichier 2017). Weeds increase biodiversity, which increases the diversity of natural enemy insects available to prey on crop pests. Increasing diversity, therefore, is a pest management method, one increasingly studied in crop management (Altieri 1991; Ratnadass et al. 2012; Busch et al. 2020). Parasitic wasps of pests, for example, have increased fecundity due to nectar obtained from weeds, and are supported by immature arthropods living on the weeds (Pavuk and Stinner, 2017). These beneficial insects naturally suppress pest populations and may enhance agricultural output and quality.

Both theoretical and empirical evidence indicate that habitat complexity enhances ecosystem functioning and stability by promoting coexistence through resource partitioning among species (Loreau et al. 2003; Tschardt et al. 2012; Cardinale et al. 2000). Weeds within and around fields supply vegetative cover which supports higher densities and evenness of natural enemies, enhancing biological control by predatory and parasitoid insects (Letourneau et al. 2011; Diehl et al. 2012; Blubaugh et al. 2021, 2016). Weeds also modify the microclimate such as soil

ground temperature, and may buffer against environmental disturbances such as heavy rain/heat waves and pesticide sprays (Gontijo 2019).

These hypotheses support the premise to study the utility of weeds and warrant future research to investigate their potential benefit in various crop systems. Previous work has shown increased success of beneficial parasitoid insects in the presence of weeds, as beneficial insects use nectar or pollen during their adult life stage to increase life span and fecundity (Norris and Kogan, 2000). Similarly, pollinators can have their populations greatly bolstered in the presence of weeds and have been shown to have a unique relationship with them (Kremen et al. 2002).

1.1 Research Objective

To examine increasing biodiversity with weeds as refuge resource plants to enhance the abundance and diversity of beneficial insect species, to benefit Mango (*Mangifera indica*) crop production in South Florida.

1.2 Research Questions

1. How do the abundance and diversity of insect species (both beneficial and pest) differ on mango in the presence or absence of weeds?
2. What is the impact on mango fruit yield from the presence of weeds?

Types of Insects

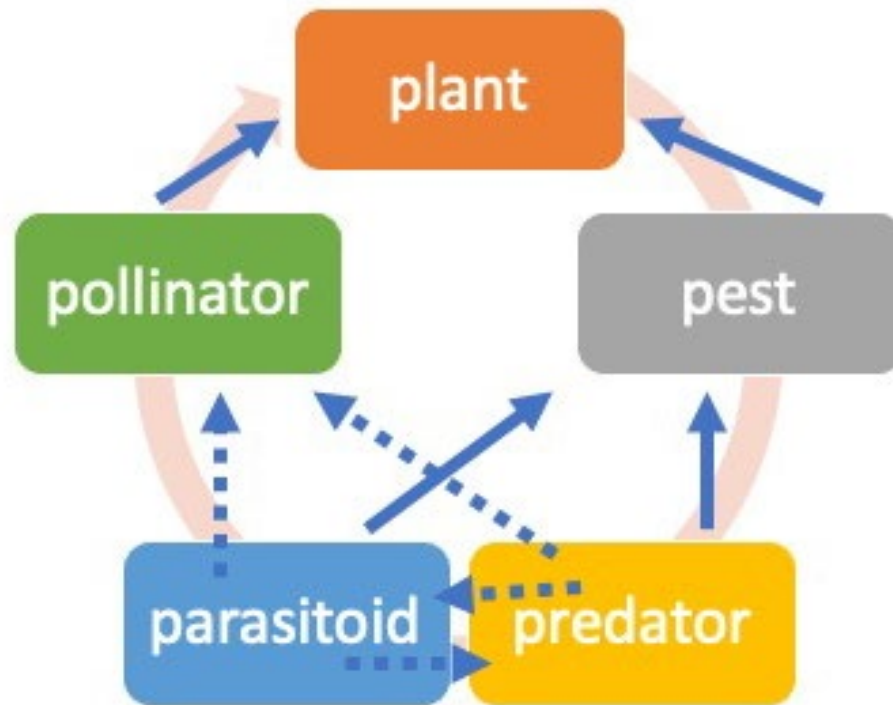


Figure 1 Types of Insects. Direct Interactions (solid line) and Indirect Interactions (dashed line).

1.3 Hypotheses

1. There will be a higher abundance and diversity of beneficial insect species around the mango trees where weeds have been left to grow (weed treatment) in comparison to mango trees that have been cleared of all weeds (no weed treatment).

2. There will be higher fruit count per tree in the weed treatment than the no-weed treatments.

If the data do not support the hypothesis, this will indicate that weeds are more a source of pests and detract from the crop rather than providing resources to beneficial insects and should be completely or selectively removed to maximize crop yield.

2. Literature Review

Field edges and other natural habitats in agricultural communities and urban landscapes help sustain beneficial insects. Weeds increase biodiversity, which increases the diversity of natural enemy insects available to prey on crop pests, and of native pollinators. Beneficial insects may provide biological control of crop pests; weeds also provide alternative floral resources for pollinators, encouraging pollinator species to remain in a local area in the interval between crop flowering events (Melin et al., 2018). This is especially important in crops that require pollination by insects, such as mango, which is known to benefit from the presence of a diversity of weeds (Melin et al., 2018).

Beneficial insects use nectar or pollen during their adult life stage to increase life span and fecundity. Previous work has shown increased success of beneficial parasitoid insects, which use pests as hosts, ultimately killing them, in the presence of weeds adapted to local environments at equal rates to common insectary plants (Araj et al. 2019). Parasitoid insects, many of which are wasps, are used as biological control of pests as they lay their eggs inside or on a host to feed on and ultimately kill. Establishment of parasitoids in farms is enhanced by the presence of weeds that provide nectar to adult female wasps and pest outbreaks are generally less common in the presence of weeds due to increased mortality by natural enemies (Altieri & Nicholls, 2018).

Tolerable weed levels, therefore, enhance these beneficial insects, without reducing crop yield (Altieri et al. 1984). Weeds can also provide many other resources to parasitoids, other than

nectar and pollen, such as oviposition sites. The Colorado potato beetle, *Leptinotarsa decemlineata*, an economic pest of potato, prefers to oviposit on the weed hairy nightshade (*Solanum sarrachoides* Sendtner) rather than on potato (*S. tuberosum* (L.)), and eggs are less abundant on potato in the presence of nightshade (Horton and Capinera 1990). A study on the parasitoid of blackberry leafhoppers showed that they overwinter on weeds in the absence of the crop plant (Doutt & Nakata, 1973). The weeds provide alternate prey for the parasitoid that isn't a crop pest and keep this parasitoid in the field between seasons. In-crop weeds can also indirectly increase crop yields by harboring alternate insect hosts that support natural enemies of the European corn borer, for example (DiTommaso et al. 2016). Weeds, therefore, can be a reservoir of alternative prey and oviposition sites, and by living on weeds, parasitoids also protect crop yields by reducing pests.

Weeds have been seen to greatly bolster the establishment and success of parasitoids. Weeds adapted to local environments were found to provide the same resources as common insectary plants, like alyssum, to significantly increase whitefly parasitoids longevity, egg load, and fecundity (Araj et al., 2019). Native weeds, then, have the potential to act as insectary plants when growing companion plants isn't possible, or can add to the variety of diets for parasitoids.

The eradication of weeds limits the availability of nectar provided by plants for pollinators (Altieri & Nicholls, 2018). Tropical crops, such as mango, pollinated by social bees may be most susceptible to pollination failure from habitat loss (Ricketts et al. 2008). Pollinators can use weeds as alternative resources before, during, and after the bloom of a crop, and increase crop yields if given these resources. Resident pollinators are healthiest with 15 or more flowering species providing a season-long food supply (Willmer, 2011) and refuges with weeds can provide this floral diversity, while helping alleviate the pollinator decline crisis in areas of

intensive farming. During seasonal fluctuations of crops and pollinator needs, native pollinators can provide a substantial portion of crop pollination, when supplied adequate habitat. In farms near untouched adjacent areas, native bee communities were found to provide full pollination services, even for watermelon, a crop with heavy pollination requirements (Kremen et al., 2002). The use of local weeds in farmed land safeguards this pollinator diversity and the specialized links between pollinators and weed species. It also buffers against possible lapses in pollination by the European honey bee, a troubled species (Paudel et al., 2015), by ensuring native bee health and range in farmland.

The economic value of field margins as refuge for pollinators to agricultural productivity is relatively unknown, and few farmers manage these areas to enhance beneficial insects (Nicholls and Altieri 2013). Therefore, managing flowering weeds at tolerable levels to provide alternative resources for pollinators within farms is a neglected habitat management tactic. Native pollinators can provide free pollination services, and further studies on their requirements and success can provide solutions to the pollinator decline crisis.

The use of herbicides to reduce weeds limits the availability of nectar provided by plants for pollinators (Altieri & Nicholls, 2018). Agroecosystems have thwarted the opportunity for co-evolution of insects and plants, with massive synchronous blooms of a single species, and vegetational simplification of large expanses of land. This lack of wild plant floral resources within a farm or adjacent to it before and after the crop blooms can cause a decline in pollinators, due to a lack of support when the crop isn't in bloom. Pollinators can use weeds as alternative resources before, during, and after the bloom of a crop, and increase crop yields if given these resources (Carol & David, 1997). Decline in pollinators is interlinked with weed and habitat

decline, through increased applications of pesticides and fertilizers (Nicholls & Altieri, 2013; Vogel 2017), and the expansion of monocultures.

Weeds are resilient, hardy plant species. Agricultural intensification leads to decreased landscape biodiversity for plants and insects, making weeds a significant part of the remaining floral diversity. Weeds are in general ambophilous, both insect- and wind-pollinated, both of which promotes genetic diversity and adaptation to environmental disturbances. This generation of gene flow and environmental plasticity allows successful persistence of weeds in arable landscapes. Increased habitat diversity and patches of unmanaged habitat reduces extinction rates of weeds, through increased genetic variability and species richness (Rollin et al., 2016) There is an evolutionary trend in agroecosystems of de-specialization of plant-pollinator networks, lowering the risk of pollinator absence due to disturbance. Mutualistic pollination networks are key ecological processes, and their stability depends on many links between species. The presence of rare weeds in farmlands is an indicator of the stability of a community, as their presence is in part due to pollinators, which are the slowest to recover after high levels of agricultural intensity (Rollin et al., 2016).

Mass flowering of crops alters floral availability temporarily, changing pollinator preferences and the stability of wild networks. The link between plants and insects, and the presence of native weeds, can serve as indicators of the biodiversity of arable lands.

2.1 Pollinators

Almost 35% of crops depend on pollinators globally (Klein et al. 2007). Pollination of at least 63 crops is vulnerable to agricultural intensification, which may reduce the diversity and abundance of pollinators (Klein et al. 2007). There is a widespread pattern of loss in pollinator richness and abundance resulting from agricultural intensification and habitat loss. With less pollination, more land will be needed for agriculture to produce these crops, resulting in demand

for natural habitat destruction, especially in the developing world (Aizen et al. 2009). The global annual economic value of insect pollination was estimated to be 9.5% of the value of global agricultural production, or upwards of \$173 billion worldwide (Potts et al. 2010).

Pollination by bees and other animals increases the size, quality, and stability of harvests for 70% of leading global crops (Ricketts et al. 2008), including blueberries (Nicholson and Ricketts 2019), coffee (Klein et al. 2003), oilseed rape (Bommarco et al. 2012), strawberries (Klatt et al. 2014), tomatoes (Greenleaf and Kremen 2006), and mango (Carvalho et al. 2012), to name just a few. Because native species pollinate many of these crops effectively, conserving habitats for wild pollinators within agricultural landscapes can help promote pollination services (Garibaldi et al. 2013). Relationships between pollination services and distance from natural or semi-natural habitats have strong exponential declines in both pollinator richness and native visitation rate (Ricketts et al. 2008). Nesting pollinators travel relatively short distances from the nest to forage: most species of bee are known to travel less than two kilometers away (Rands and Whitney 2011). Small scale practices can have major effects on pollination services, then, especially for insects with short flight ranges (Garibaldi et al. 2014)

For 17 crops in agricultural landscapes around the globe, a significant negative effect of distance from natural habitat- due to habitat loss and conversion- was found on the richness and abundance of wild bees (Ricketts et al. 2008). Visitation rates by pollinators on crops decline as distance from natural areas increases, reaching half of its maximum at just 0.6km away from natural habitats (Ricketts et al. 2008). Pollination services decline exponentially with distance from plantings, and perennial and older flower strips with higher flowering plant diversity are found to enhance pollination most effectively (Albrecht et al. 2020).

Most wild plant species (80%) are directly dependent on insect pollination for fruit and seed set, and many (62–73%) plant populations show pollination limitations (Ricketts et al. 2008). Tropical crops pollinated primarily by social bees may be most susceptible to pollination failure from habitat loss (Ricketts et al. 2008). Exposures to pesticides and pathogens, coupled with food stressors, can impair immune responses, rendering bees more vulnerable to parasites (Goulson et al. 2015). Removing stressors by increasing floral availability in farmlands through retaining or restoring areas of semi-natural habitat can improve nest site availability and reduce dietary stress.

Climate change also poses a threat to both pollinators and crops. Heat stress results in lower yields of crops, such as with faba bean (*Vicia faba*) (Bishop et al. 2016). However, insect pollination partially recovered faba bean yield loss following heat stress, with significantly lower yield losses in plants pollinated by the buff-tailed bumblebee *Bombus terrestris* with increased transfer of pollen to damaged flowers (Bishop et al. 2016). Insect pollination may increase production stability in changing environments, becoming more important in crop production as the probability of heat waves increases because of climate change. Therefore, maintaining pollination services requires the conservation of native plants for wild pollinators within agricultural landscapes, such as suitable nesting and floral resources (Kremen et al. 2002)

Proximity of nesting habitats to agricultural fields is critical for insect pollinated crops. On average, fields 1.5km away from natural habitat patches can be expected to contain 50% of the pollinator diversity of fields closest to these patches (Ricketts et al. 2008). As distances from natural habitats increase, fewer pollinator species forage to that distance or nest in fields isolated from their natural resources. Field margin manipulation can enhance the proportion of land available to longer-distance nesting foragers such as honey bees, and short-distance foragers such as solitary bees for foraging, regardless of the distance over which they normally travels to find

food (Rands and Whitney 2011). However, many solitary bees prefer to nest in exposed soil; therefore, the disturbance created in many agricultural fields may actually create preferred nesting substrate for some of these ground-nesting species (Ricketts et al. 2008).

Limited flight ranges of insects can cause a loss of connectivity between habitat types; however, this loss is partially mitigated by the abilities of some bee species to nest and reproduce within agricultural landscapes (Kremen et al. 2002). Several species found in isolated farms readily nest in the ground within agricultural fields or fallow farm borders (Kim et al. 2006). These small species have relatively low resource demands per individual, and therefore may be able to survive and reproduce on the resources provided by flowering weeds when crops are not blooming (Crooks and Sanjayan 2006). Bees usually nest in one habitat type, most frequently in the ground or dead plant stems, including tree cavities, but require other types of habitats for forage, like natural prairies, forests, and wildflower meadows (Ricketts et al. 2006). In California watermelon fields, for example, the species most often absent from isolated fields also tend to be the most efficient pollinators, exacerbating the effect of declining richness on pollination services (Larsen et al. 2005). Connectivity among different habitats in a landscape can affect these important ecosystem services, many of which provide economic benefits to human populations (Ricketts et al. 2006).

High aculeate, or stinging hymenopteran diversity, including species of conservation concern, can persist in agricultural environments containing 21–22% semi-natural habitat (Wood et al. 2015). Adding floral resources significantly increases aculeate diversity more so in simpler, intensively farmed landscapes, with around 5–10% semi-natural habitat (Wood et al. 2015). One way to provide for more habitat in which to conserve native bees and other species biodiversity, as well as nonrenewable resources, would be to improve the yields on existing crop lands. This

can be accomplished, in part, by introducing the most efficient pollinators for these crops (Batra 1995).

Adding plant and insect diversity to the barest agricultural landscapes ensures efficient pollination and productivity, which in turn reduces the need for converting natural areas into agriculture to feed growing populations. Pollinator supporting plants are more successful when background floral resources are scarce. In South Africa, for example, the importance of weed species richness for enhancing crop sunflower seed set increased with larger distances from natural areas, underscoring their importance in isolated homogenous landscapes (Garibaldi et al. 2014). A heterogeneous environment also reduces competition between species through niche partitioning, allowing diverse assemblages that perform their ecosystem functions at elevated rates (Tylianakis et al. 2008).

There is a pollinator decline crisis in areas of intensive human land use and landscape simplification, including farmlands (Dainese et al. 2019). Pollinators are sensitive to the presence or absence of floral resources, with their populations fluctuating in turn. A number of insect groups and farmland birds have shown marked population declines over the past 30 years, with the average decline of terrestrial insect abundance about 9% per decade (van Klink et al. 2020). Correlational studies indicate that many of these declines are associated with changes in agricultural practices (Marshall et al. 2003). Many of the weed species that are known to support farmland birds or insects have decreased significantly over the last few decades, and Storkey et al. (2012) attribute the decline of weed abundance and diversity in arable fields to increased intensity of management, including increased crop plant density, decreased crop diversity, increased fertilizer and herbicide use, and more efficient seed cleaning. However, local-scale diversification can change overall population trends, providing hope for directed conservation tactics.

Decline in pollinators is interlinked with weed and habitat decline, through increased applications of pesticides (van der Valk et al. 2013), fertilizers (Nicholls and Altieri 2013), and the expansion of monocultures. Herbicidal eradication of weeds removes nectar that weeds can provide for pollinators (Nicholls and Altieri 2018). With large synchronous blooms of a single crop and vegetational simplification, mutualisms between insects and flowering plants are greatly diminished. The lack of floral resources within or around a farm before and after the crop blooms can cause a decline in pollinators between seasons (Carol and William, 1997). Pollinators can use weeds as alternative or additional resources in a farm (Batra 1979) before a crop blooms, during, and after. This increases pollinator health and proximity to crops, and in turn increases crop yields (Carol and William, 1997).

Native bees and other insects are important contributors to global crop pollination (Batra 1967, 1995; Rader et al. 2016). The contribution of wild, free-living bee species in California alone is between \$937 million to \$2.4 billion per year in economic value of crop pollination, with no cost to farmers for this ecosystem service (Chaplin-Kramer et al. 2011). These native bees can provide full pollination services for free, without the use of managed honey bees, in farms near natural areas (Kremen et al. 2002). However, pollinators require 15 or more flowering species providing a season-long food supply to be healthy and remain in a farm (Willmer, 2011). Weedy refuges with wildflowers, then, provide this floral diversity and palliate the pollinator decline crisis, especially in monocultures (Pickett and Bugg, 1998).

Patches of flowering habitats work as refuges; however, if the farm is too large (>5 ha) native pollinators cannot spill-over and spread into farms (Nicholls and Altieri 2013). Similarly, arable weeds can play an important role in maintaining and restoring invertebrate populations, but 10% weed cover is needed to support invertebrates that provide ecosystem services (Smith et al.

2020). A landscape management scenario designed to maximize crop production with weeds is to maximize biodiversity with 25% of the landscape devoted to non-crop area or grass strips at 10% of the landscape, resulting in high crop production and medium biodiversity at the landscape scale. Land-sharing scenarios attempting to maximize both productivity and biodiversity within the same field always produced less biodiversity and less production (Colbach et al. 2018).

On-farm diversification may be an important refuge for both specialist bees and other pollinator species that are vulnerable to floral resource simplification resulting from land development (Guzman et al. 2019). Increasing diversity of native bees can assuage low populations of agricultural European honey bees (Kremen et al. 2002), and recent studies have highlighted the importance of overall biodiversity on pollination success and crop yield (Kremen et al. 2007, Garibaldi et al. 2016). There is evidence that artificially bringing in hives of agricultural honey bees can increase crop pollination temporarily (Rader et al. 2013), but that during fluctuating crop and pollinator needs, native pollinators can provide significant crop pollination when near natural areas. Honey bees negatively affect the mutualistic interactions between native bees and plants, and there is evidence that when they dominate the landscape, pollinator-dependent crops will be less productive than with a more diverse pollinator array (Aizen et al. 2020).

Native bee communities, for example, can provide full pollination services for watermelon, a crop with heavy pollination requirements (Kremen et al. 2002, Njoroge et al. 2010, Rader et al. 2013). Native bees also can buffer against the negative impact of climate warming on honey bee pollination of watermelon crops, exemplifying how biodiversity can stabilize ecosystem services against environmental change. Similarly, diversified organic farming increases insect functional diversity (Goded et al. 2019), enhancing pollinator diversity, abundance, crop pollination, and yield (Woodcock et al. 2019). Ecosystem services arguments and conserving biodiversity concerns

both concur, therefore, on the potential benefits of increasing diversity with weedy species. Such connectivity may be provided by deliberately planted strips of native plants, wildflowers, or simply uncleared ground that has been colonized by weeds.

Weeds are by definition tenacious, hardy plant species: they grow without being cultivated, and often where they are not wanted. Weeds are classically defined as plants that spontaneously grow on land modified by humans, while arable weeds are those that occur in regularly cultivated fields, yet there is still no definitive answer to “what makes a weed a weed?” (Bourgeois et al. 2019). Arable weeds have specific functional traits that make them tolerant to arable fields, such as soil disturbances and fertilization, making a large overlap in the weedy potential of non-weed species. Most weeds are therophytes with earlier and longer flowering, larger leaf area, and affinity for nutrient rich sunny dry environments compared to non-weeds (Bourgeois et al. 2019). Many weeds can be non-native, but some native plants also have weedy propensities. With decreasing landscape diversity, weedy floral resources are often all that remains. Weeds may be insect- or wind- pollinated, aiding genetic variation and adaptation to disturbances. They are often self-compatible and may automatically produce seed without pollinators (Baker 1974), ensuring their continued presence in the seed bank. This environmental plasticity allows successful persistence of “weeds” in disturbed habitats (Baker 1991). Weeds are an essential resource for pollen because of their continuous flowering phenology (especially during the late spring period) and their high species richness, which contributes directly to the pollen diversity dietary needs of insects (Requier et al. 2015).

Several studies have examined the benefits of planting strips of wildflowers near crops to enhance pollination (Ouvrard et al. 2018; Tschumi et al. 2016; Feltham et al. 2015). Similarly, when comparing wildflower plantings with weeds to support wild bees, in one site the numbers of

honey bees did not differ significantly between wildflowers and weedy control plots (Williams et al. 2015), showing the economic significance of both kinds of floral resources to pollinators.

2.2 Floral rewards

2.2.1 Nectar

Bees, wasps, flies, butterflies, and some moths utilize nectar, a common floral reward. While all flowers have pollen, not all flowers have nectar, but many of those visited by insects do (Baker and Hurd 1967; Goulson 1999; Faegri and Van Der Pijl 2013). Nectar is used for insect's daily energy intake. It is the metabolic precursor for beeswax and is processed into honey, the food reserve for overwintering bees who do not forage in winter (Bretagnolle and Gaba 2015).

Honey bees collect nectar from flowering plants, and the different types of honey are named for the dominant flowering plant resource present in certain locations and seasons. High honey bee densities may have an impact on other pollinators via competition for floral resources (Torné-Noguera et al. 2016). A well-developed suction pump in the head represents an important adaptation for nectar-feeding insects, such as Hymenoptera, Lepidoptera, and Diptera. This pumping organ creates a pressure gradient along the proboscis, responsible for nectar uptake (Karolyi et al. 2013). Large-body flies such as *Syrphidae* have pollen collecting hairs, long, spirally grooved bristles, and elongate mouthparts to ingest nectar and pollen from flowers.

Butterflies use amino acids in nectar to enhance their fitness and fecundity, acting as agents of natural selection on nectar composition in plants, supporting the existence of a relationship between nectar preferences and fitness benefits (Jervis and Boggs 2005). Interestingly, Map butterflies (*Araschnia levana*) can use nectar as adults to override impacts of poor larval food (Mevi-Schütz and Erhardt 2005). Similarly, the provision of adequate nectar resources and larval host plants within intensively managed arable landscapes is likely to be essential to the successful conservation of butterfly species. Weedy patches in the crop have a direct benefit to butterfly

richness, with mobile species taking advantage of these spatially and temporally unpredictable nectar resources (Pywell et al. 2004). Adequate shelter, floristically diverse field margins, and availability of nectar resources can increase the abundance of immobile butterfly species in arable landscapes, as well as presence of larval host plants and the abundance of mobile species (Pywell et al. 2004).

2.2.2 Pollen

In addition to nectar, pollinators need pollen from a variety of plants, and rely on a wide diversity of plants for their pollen needs throughout the season, even during crop flowering (Requier et al. 2015). Pollen is used for brood development, as it contains proteins, fats, mineral salts, amino acids and vitamins, and is stored in small quantities as it deteriorates rapidly (Bretagnolle and Gaba 2015). Pollen collected and stored in hives can provide a record of what species the honey bees have collected (Anderson et al. 2014).

Weeds represent a substantial part of the honey bee annual diet. To overcome the need to feed colonies artificially during a period of food supply depletion, it is necessary to increase either the diversity of crops or the abundance and diversity of floral resources in association with crops: weeds, grasslands, hedgerows, or field margins (Requier et al. 2015). This provides free resources to agricultural bee colonies, as well as native insect species. Pollen resource diversity and quality also enhances resistance to fungal diseases, tolerance to pesticides, and immunity in honey bees to parasites, diseases, and pathogens (Di Pasquale et al. 2013; Requier et al. 2015).

Agricultural honey bees have a high botanical richness in their pollen diet, mostly from trees and weeds, and are influenced by local landscape composition. Weed species constitute the bulk of honey bee diets between mass flowering crop periods (up to 40%) and are therefore suspected to play a critical role at this time period (Requier et al. 2015). This is especially critical during late spring, where there is a food supply depletion period of both pollen and nectar and a

peak in honey bee populations. Early in the season, similarly, crop species were less used for pollen (11%) than other floral resources. In contrast, honey bees relied heavily on woody and herbaceous plants like weeds from semi-natural habitats to meet their pollen requirements, totaling more than 60% of their annual pollen diet (Requier et al. 2015). At the first pollen peak, for example, honey bees massively foraged on floral species from adjacent habitats rather than the crop rapeseed for pollen, possibly due to better nutrition of the weed pollen for their needs at that time (Requier et al. 2015).

Honey bees use a wide variety of resources for their pollen diets, then, to ensure the health of the entire colony. This may explain why bees foraged on more plant species than expected during rapeseed blooming, because larvae are more numerous during this period, with high quality pollen requirements (Keller et al. 2005). For 85% of 41 different bee species examined, the whole pollen content of more than 30 flowers is required to rear just a single larva (Müller et al. 2006). As only about 40% of the pollen contained in a flower is available to a single female bee, however, these estimates must be multiplied by a factor of about 2.5 to correct for pollen that has already been removed and for pollen that will later be removed by other flower visitors (Müller et al. 2006). With the exception of honey bees, *Apis spp.*, most of the Apidae appear to forage at a maximum of 2km from their nests, and a majority under 1km (Rands and Whitney 2011). The survival and development of honey bee colonies is influenced by the regularity, quality, and quantity of nectar and pollen after overwintering for the replacement of workers, during spring and summer when the population has peaked, and in autumn for the storage of winter food (Wratten et al. 2012).

Pollen foraging plays an important role in pollination and in the life of all bee colonies that adjust their foraging to natural variation in pollen protein quality and temporal availability (Jha et al. 2013). *Bombus vosnesenskii*, the yellow-faced bumblebee, collects pollen from a wide range of

plant families and does not exhibit a significant preference for native versus non-native species (Jha et al. 2013). Similarly, it was found that bumblebees forage further in pursuit of species-rich floral patches and in landscapes where patch-to-patch variation in floral resources is less, regardless of habitat composition. This demonstrates extreme foraging plasticity in wild pollinators, and that floral diversity, not density, drives bee foraging distance (Jha and Kremen 2013). Non-native wild plant species flowering in late summer, then, can fill a forage gap for the diet of both honey bees and generalist wild bees during this time (Wood et al. 2018).

Additionally, native bees such as *Hylaeus spp.* are more likely to carry less pollen and exhibit higher pollinator fidelity compared with the non-native honey bee *Apis mellifera* (Miller et al 2015). By contrast, honey bees are more likely to carry mixed pollen and forage on invasive plant species (Miller et al 2015). Honey bees tend to be found more frequently and in greater abundance in mass-flowering crops whereas wild, solitary bees are more abundant in semi-natural areas, grasslands, and weedy strips (Bretagnolle and Gaba 2015).

For both wild and honey bees, weeds are a limiting resource: quantitatively for honey bees, especially between mass-flowering periods, and qualitatively for the more selective wild bee foragers (Bretagnolle and Gaba 2015). The decline in wild bee diversity is, therefore, strongly correlated with the decline in weeds and wildflowers.

Wild flowers are strongly dependent on pollinating insects for their reproduction: 78–94 % of flowering species rely on pollination (Ollerton et al. 2011; Winfree et al. 2011). Native and rare weed species and wildflowers are pollinated by wild bees, with honey bees actively collecting pollen from wildflowers, helping to ensure the conservation of floral biodiversity. The presence of wild bees on flowers also can cause behavioral changes in honey bees, which, when disturbed, forage on other flowers, helping to pollinate crop plants and increase the success of pollination

(Riedinger et al. 2014). For instance, when weeds are present in sufficient numbers or close to crops, the wild bee community is more abundant, pushing honey bees away to pollinate crop flowers which in turn increase crop production (Carvalheiro et al. 2011). This can be an asset in mango pollination, whose flowers are less attractive to honey bees.

Pollen abundance is also important for wild bees (Müller et al. 2006). Oligolectic, or specialized pollinator species demand a great abundance of their preferred plant resources, and populations of some species with a narrow diet have declined more so than generalists, in line with declines of their preferred plants (Kleijn and Raemakers 2008). Therefore, over-representation of a single source of resource in the bees' diet could have detrimental effects upon development of the colony for certain species, and the lack of dietary diversity may lead to a lack of micronutrients essential to larval development (Rands and Whitney 2011). Wild and honey bee networks are interconnected through the weeds, on which they both depend as a limiting resource, either in space (wild bees) or time (honey bees) (Bretagnolle and Gaba 2015).

Pollinators are often associated with field margins and their associated hedgerows as remnants of semi-natural habitat to provide food, overwintering and/or nesting resources. When examining spill-over between oilseed rape and unmanaged margins, wild plant species all overlapped in flower-visitor niche (Stanley and Stout 2014). Oilseed rape overlaps in terms of species of flower-visitor (pollinator niche), and in terms of individual flower visitors, with co-flowering wild species that grow in the field margins and hedgerows (Stanley and Stout 2014). Enhancing these areas, then, can lead to an increase in the availability of forage to pollinators that nest within the landscape (Rands and Whitney 2011).

The pollinator niche of wild plant species shared with oilseed rape was 26 % (Stanley and Stout 2014). In general, insects did not specialize on a single flower type, and carried pollen from

a range of different wild plant species. Very little oilseed rape pollen was found on the stigmas of wild plants, indicating that the deposition of crop pollen may not be a mechanism for interference to pollination services. Insects foraged on both the crop and wild plants in the margins, the majority of which carried pollen from both the crop and wild species (Stanley and Stout 2014). Seven wild plant species growing in field margins and hedgerows around mass-flowering oilseed rape in Ireland, for example, overlap with the crop in terms of the individual insects that visit their flowers (Stanley and Stout 2014). As floral richness increases, then, so does variety in bloom periods and thus the overall temporal availability of pollen and nectar resources in field margins (Morrison et al. 2017).

2.3 Pollinator Networks

Ecologists have described pollinator networks as maps of all the documented associations between and among pollinator and flowering plant species in a given area or habitat (Lewinsohn et al. 2006; Fortuna et al. 2010). In agroecosystems, there is de-specialization of plant-pollinator networks, lowering the risk of pollinator absence due to environmental disturbance (Rollin et al. 2016). These mutualistic pollination networks are essential ecological processes, and their stability depends on many species' interactions (Parra-Tabla et al. 2017; Jauker et al. 2019). Habitat loss affects the diversity of wild bee communities, with social bees and small generalist bees substantially affected (Bommarco et al. 2010). Habitat fragmentation has profound effects on pollinator networks, creating the extinction of ecological interactions and networks (Janzen 1971, Wilson et al. 2016).

Rare weeds in farmlands, then, can act as an indicator as to the stability of a community, since their presence is in part due to pollinators, the slowest to recover after high levels of agricultural intensity (Rollin et al. 2016). Community level studies have shown that maintaining the structure of the entire food web is important, because a greater diversity of pollinators, and of

pollination guilds, can improve yield and stability (Hoehn et al. 2008), and because rare plants may be linked to common plants through shared pollinators (Wratten et al. 2012).

Mass flowering plants can act as ‘pollinator hogs’, which can reduce the pollination success of adjacent co-flowering neighbors by drawing pollinators from these plants (Ghazoul 2006; Koptur and Barrios 2020). However, very attractive mass-flowering plants can also act as magnets, producing pollination ‘spillover-effects’ through increased pollinator movements to adjacent co-flowering taxa, potentially either increasing pollination (Thomson 1978) or impacting it through the transfer of mixed-species pollen (Gilpin et al. 2019). In 10 of 18 comparisons, co-flowering species supported a slightly higher diversity of pollinators than magnets, with honey bees comprising a significantly lower proportion of flower visitors in 14 comparisons, with no evidence of pollinator spillover effects (Gilpin et al. 2019).

Additionally, florally enhanced field edges harbor more taxonomically and functionally abundant, diverse, and compositionally different bee communities compared to bare edges (Nicholson et al. 2020). However, enhancements did not increase the abundance or diversity of bees visiting crops, indicating that the supply of pollination services was unchanged following enhancement. Promoting crop pollination, therefore, improves multiple dimensions of biodiversity, underscoring their conservation value, but these benefits may not be spilling over to crops. Floral plantings have great potential to benefit ecosystem service provision, but to do so will need to be carefully tailored for functioning at specific spatial scales. Increasing flower diversity and the age of these plantings are important drivers through which this can be achieved (Albrecht et al. 2020)

Temporal availability of mass flowering crops can change pollinator preferences, as well as the stability of wild pollinator networks. Pollinator abundance increases with flower abundance,

vegetation height, and floral diversity (Morrison et al. 2017). The conservation of plant diversity safeguards native pollinator diversity, and the specialized links between pollinators and specific weed species, as well as enhances overall biodiversity and ecosystem services (Requier et al. 2015). This safeguarding also buffers against possible lapses in pollination by agricultural European honey-bees, whose populations are declining at a rapid rate (Paudel et al. 2015), by ensuring native bee health and range in farmlands. The connections between plants and insects, and the presence of weed species, can serve as indicators of the biodiversity in arable lands.

2.4 Parasitoid Insects

Parasitoid insects, the majority of which are wasps, are used as biological control of pests as they lay their eggs inside or on a host to feed on and ultimately kill. Establishment of parasitoids in farms is enhanced by the presence of weeds that provide nectar to adult female wasps (Altieri and Nicholls, 2018), and pest outbreaks are generally less common in the presence of weeds due to increased mortality by natural enemies. Tolerable weed levels enhance these beneficial insects, without reducing crop yield.

Some studies have shown the success of parasitoids with more floral resources. Parasitism rates of armored scales by *Encarsia citrina* increased over time in the presence of floral resources, through incremental growth of parasitoid populations and immigration in response to increased floral resources (Rebek et al. 2006). Similarly, while both hosts and parasitoids feed on shared floral resources, when exposed to common flowering plants, parasitoids benefited eight times more than their leaf-mining hosts (Kehrli and Bacher, 2008). In maize fields, parasitism by *Trichogramma chilonis* of *Helicoverpa armigera* eggs was positively correlated to the proportion of non-crop habitat diversity and other host crops (Liu et al. 2016). Increasing agricultural intensity and loss of biologically diverse habitat would have great reductions in the presence and parasitism of *T. chilonis*.

Weeds adapted to local environments were found to provide similar resources to common insectary plants, like alyssum, to significantly increase whitefly parasitoids longevity, egg load, and fecundity (Araj et al. 2019). Native weeds, therefore, have the potential to act as insectary plants when growing companion plants isn't possible, or can add to the variety of diets for parasitoids. Weeds, then, can greatly bolster the establishment and success of parasitoids.

Weeds can provide alternative prey, that are not crop pests, to parasitoids as well. A study on the parasitoid of grape leafhoppers, *Anagrus*, showed that they overwinter on adjacent habitat to vineyards (Provost and Pedneault, 2016). The vegetation within and around the vineyard provided alternate prey for the parasitoid that isn't a crop pest, and kept this parasitoid in the field between seasons. Similarly, European corn borer infestations were decreased in the presence of weeds. Parasitoids of this pest were supported by moth species living on the weeds in corn fields (Pavuk and Stinner, 2017).

Weeds both provide food for beneficial insects as well as provide oviposition (egg laying) sites. There are better egg survival rates when oviposited on weeds than the crop. In the absence of prey for the larvae of predatory lady beetles, *Coleomegilla maculata* oviposits on weeds rather than the crop, and as a result the eggs had better survival through less predation and parasitism (Cottrell and Yeargan, 1998). Weeds, therefore, can be a reservoir of alternative prey, and by living on weeds, parasitoids also protect crop yields by reducing pests.

2.5 Predatory Insects

60 studies found that in-field plant diversity strongly increased arthropod abundance and richness but had weaker effects on evenness (Lichtenberg et al. 2017). Weed-seed predatory ground beetle activity-density is higher in systems with greater above ground biomass and plant diversity (Ward et al. 2011). Weedy patches in cropped areas contribute to biodiversity by

conserving populations of granivorous carabids and weed seed predation (Kulkarni et al. 2017; Blubaugh and Kaplan 2015).

Vegetative cover provided by weeds can also enhance arthropod numbers. In a study of weed-invertebrate relationships across seven studies of winter-sown wheat spanning 18 years, Smith et al. (2020) found that herbivores showed a stronger positive relationship with weed cover than did predators, but that arthropod predator abundance was related to total weed cover in wheat, *Triticum aestivum* L.

2.6 Debate

An area of debate that needs further evaluation is that in some situations, weed–insect interactions supply disservices such as increases in weed fecundity and abundance, crop and cover crop seed consumption, maintenance of phytopathogens, and insect pest populations (Barbercheck and Wallace 2021). Additionally, the concept that weeds providing nectar to beneficial insects may also provide resources to crop damaging pests, and even attract beneficial insects away from the crop is a potential issue (Capinera 2005). Although vegetative cover can support higher densities of natural enemies by providing a favorable microclimate and provisions of non-pest food (Diehl et al. 2012), alternate foods may distract natural enemies from focal pest suppression (Frank et al. 2011). If weed species are preferred over an adjacent crop, the weeds may act as a barrier or a trap crop (Andow 1991).

In addition, because pathogens can infect a wide variety of plants over many plant families, weeds can act as vectors of pathogens for crops (Wisler and Norris 2005). Weeds can serve as an overwintering or in-season reservoir for phytopathogens that are transmitted from outside or within the crop field to crop plants by insects (Barbercheck and Wallace 2021). Some weeds may serve as an obligate alternate host for phytopathogens that require them as part of the

phytopathogen life cycle. Perennial weed species may serve as long-lasting inoculum acquisition sources of Tomato Spot Wilt Virus (TSWV) for spread to susceptible crops (Groves et al. 2002). Insect transmitted crop viruses are mediated by both within-field weeds and surrounding areas. For vectors of nonpersistent stylet-borne viruses, field edge composition has a greater influence on aphid vector rates than in-field weed cover (Angellella et al. 2016). Therefore, in-field weed management may not be an effective method of crop virus prevention, and management approaches should focus on optimal crop placement relative to surrounding land use (Angellella et al. 2016; Barbercheck and Wallace 2021). Similarly, there is the question of “superweeds”, resulting from genetic drift of genes from genetically engineered crops into surrounding weed populations (Bain et al. 2017).

Using weeds as an insect management tool is a relatively new area of study, and there is still much debate as well as unanswered questions to be evaluated further. The issue of hyperparasitism underscores biological control programs, illuminated through the resource concentration hypothesis. Increased concentrations of crops (host plants) in weed-free plots leads to a greater density of pests. This may send signals and attract parasitoids and hyperparasitoids into weed-free plots, where their host resource (the pest arthropods) is more concentrated. This effect could perhaps negate the benefits of weeds, should weed-free plots have increased parasitism of crop pests. Without the presence of a hyperparasitoid, *Aphidius ervi*, a biological control agent of aphids, eliminated their populations in a controlled test. However, *A. ervi* itself was eliminated by a hyperparasitoid, *Asaphes suspensus*, within seven generations (Schooler et al. 2011). This phenomenon, however, contrasts with what happens in field surveys, in which the hyperparasitoid doesn't entirely eliminate the primary parasitoid, due to environmental

disturbances. Small primary parasitoid populations, however, are particularly susceptible to hyperparasitism (Schooler et al. 2011).

Additionally, as insects are mobile organisms, tracking their movements and activities between weeds and crops is difficult to quantify (Norris and Kogan 2000). There is also the issue of the benefits of leaving weeds as a source of nectar for beneficial insects when increased fecundity of pests may arise due to nectar obtained by the adults (Shields et al. 2019). Additionally, weeds need to provide alternative prey sources (arthropods) that are not crop pests, seen successfully done in the study of leafhopper parasitoids of vineyards (Provost and Pedneault, 2016). A similar study found some genera of aphid pests on weeds attack crop plants, while most other aphid species did not. They did, however, represent a good source of food for aphid eating predators and parasitoids, and can act as alternative prey when crop aphid populations are low (Pickett and Bugg, 1998). Alternatively, monitoring of insect pests hosted by weeds can allow managers to anticipate problems and selectively support beneficial species. This approach to understand insect behavior may prove a useful management technique in agriculture. Similarly, practices that break the taxonomic link between weeds and crop plants can reduce the movement of specialist and oligophagous insects from weeds to crop plants (Barbercheck and Wallace 2021).

Additionally, the economic value of field margins as refuge for pollinators to increase agricultural productivity is difficult to quantify, and therefore few farmers manage these areas to enhance beneficial insects. Native pollinators, predominantly bees, may be responsible for almost \$3.07 billions of fruits and vegetables produced in the United States; however, the specific added value of field margin resources to their success has not been calculated (Losey and Vaughan 2006). The total contribution of wild pollinators was valued between \$49.1 million and \$310.9 million,

for which there is no direct payment from producers (Allsopp et al., 2008). These values illustrate the importance of maintaining natural and other forage areas for the conservation of insect pollinators. It is difficult to successfully link the enhancement of pollinator habitat adjacent to crop fields with increased yield, a factor that may affect widespread adoption of such practices by farmers (Wratten et al. 2012). Despite positive effects of adjacent natural habitats and records of increased pollinator abundance and flower visitation, there is a lack of research documenting pollination spill-over in the other direction- into crops from flower rich margins or from ‘pollinator-enhancement’ areas (Wratten et al. 2012). The presence of insects on weeds however may support populations of beneficial insects that can spill over onto crops and help suppress pests (Barbercheck and Wallace 2021). Therefore, managing flowering weeds at tolerable levels to provide alternative resources for beneficial insects within farms is a relatively neglected habitat management tactic.

Additionally, the number of observed pollinators may increase initially with total open flower richness, but may then decrease with more than nine open flower species (Morrison et al. 2017). This decrease in observed pollinator abundance may be as species richness increases, density of each individual species decreases. Native pollinators can provide full and free pollination services, but further studies as to their behaviors and requisites for success can help provide solutions to the pollinator decline crisis.

Furthermore, since wild pollinators and honey bees forage on similar resources between the mass-flowering periods of crops, pollinators may compete for resources. This raises the question as to how focusing on honey bees for crop or honey production may be detrimental to wild bees. This is an important issue because reducing wild bee communities may reduce the abundance of weeds that are not pollinated by honey bees, and consequently reduce the abundance

of honey bees. Understanding the dynamics of this complex network and how the spatiotemporal composition of landscapes affects relationships within this network remains a challenge for agroecosystem management (Bretagnolle and Gaba 2015).

The effects of granivorous beetles like carabid beetles on weed seed banks is similarly unknown (Collins et al. 2002). Carabid, or ground beetles are primarily used as insect predators, but they can also have negative impacts on weed populations. They consume the weed seedbank in the soil, decreasing the number of subsequent weeds. Beetles, however, are effective predators that can easily move over long distances, meaning weed strips cannot be seen as a crucial pest reservoir (Pickett and Bugg, 1998). This should also be evaluated when assessing carabids use in insect pest management. Many studies have recorded the removal and consumption of applied weed seeds or from weed seed ‘cafeterias’ by insects, but relatively few studies of weed seed predation include data on predation of naturally dispersed weed seed or the actual decrease of weed pressure due to seed predation under commercial growing conditions (Barbercheck and Wallace 2021). Similarly, predation-related services will not necessarily translate into net benefits for agricultural production in some cases (Tschumi et al. 2018).

Further quantification must be addressed before farmers can readily use weeds to manipulate insects. For example, how does increasing the numbers of beneficial insects affect certain pest species, and what is the overall economic impact of insect manipulation? It is difficult to model any potential increase in agricultural production in relation to broader adoption of habitat enhancements. Assessing such economic impacts requires distinct valuations of component parts, and there is still uncertainty about the interconnected nature of ecosystem services (Diaz et al. 2007, Wratten et al. 2012). Pollinators and other organisms do not distinguish between field boundaries, making counting units difficult to discern, and some species typically perform more

than one function, such as hoverflies whose larvae consume insect pests and as adults are pollinators (Wratten et al. 2012).

The critical period of interference between specific weeds and crops, also, is likely to vary, and is still not well quantified (Altieri 2018). Determining when the benefits of added pollination of crops outweighs crop interference of weeds for certain species is essential. Judging when there are enough weeds to support pollinators and beneficial insects, but to not pull nutrients and interfere with the crop's production, is crucial to successfully employing weeds as insectary plants. A weed-management strategy is required that provides sufficiently abundant weeds to support beneficial species but not so abundant that crop yield is lost to weed competition (Barbercheck and Wallace 2021).

2.7 Mango

Effective insect pollination is essential for good fruit set and yield in mango (*Mangifera indica* L.) (Dag and Gazit 2000). Mango flowers are unspecialized, enabling pollination by most insects (Heard 1999). These pollinators are critical for successful fruit set of mango flowers. Managed pollinators are unsuitable (e.g. Kevan 1999) or insufficient when acting alone (Greenleaf & Kremen 2006; Carvalheiro et al. 2010; Breeze et al. 2011); and honey bees are not attracted to mango flowers (Free and Williams 1976; Carvalheiro et al. 2010). Furthermore, hand pollination is seldom economically viable (Allsopp, de Lange & Veldtman 2008). Pollination of one of the main cultivars (Kent) of mango *Mangifera indica* L. (Anacardiaceae) is highly dependent on a diverse assemblage of flying visitors, which is strongly negatively affected by distance to natural habitat (Carvalheiro et al. 2010).

Most pollinators of mango belong to order Hymenoptera, however insects in the order Diptera have been suggested as the dominant pollinators (Dag and Gazit 2000). Nectar

production for the attraction of insects indicates entomophilous pollination of mango trees (Kumar et al. 2016). Additionally, mango does not show adaptations for wind pollination morphologically or physiologically as a single anther produces comparatively few pollen-grains (200-300) and the stigma is small to catch pollen grains.

The insect orders Diptera, Hymenoptera, Lepidoptera and Coleoptera are the most common insect visitors to mango flowers, with pollen-grains observed adhering to the bodies of many species belonging to these orders. The biology of different kinds of pollinators of mango have been studied, with dipteran and hymenopteran insects found to play major roles in pollination (Dag and Gazit 2000; Kumar et al. 2016). For dipterans, or flies, 71 of the 150 families depend for feeding on flowers. Dipteran species are good pollinators of more than 550 species of flowering plants, and the family Calliphoridae (blowflies) are the main pollinators of mango (Bhatia et al. 1995). *Chrysomya*, *Lucilia* and *Musca sp.* (Diptera) were reported as the most efficient pollinators in mango orchards because of their visiting frequency and abundance (Bhatia et al. 1995). Many plant species are pollinated by flower flies (Syrphidae) reported as good pollinators of mango (Evenhuis et al. 2008) as they are capable of transporting pollens for long distances, reproducing rapidly, and producing large numbers of eggs with up to five generations per year (Ssymank et al. 2001)

Insects visiting mango bloom were collected for 3 years in 10 commercial orchards located in all major mango-growing areas in Israel. Forty-six distinct species or types were found; most belonged to the orders Diptera (26), Hymenoptera (12) and Coleoptera (6) (Dag and Gazit 2000). Two blow flies (*Chrysomya albiceps* and *Lucilia sericata*); the honey bee (*Apis mellifera*) and the housefly (*Musca domestica*) played a significant role in mango pollination in most orchards. Found in only one or two orchards, in medium to large numbers, were: the hover

fly *Episyrphus balfeatus*, the wasp *Bembecinus tridens*, and two beetles- *Cantharis atropoveolatus* and *Omophilus syriacus*.

The effectiveness of 12 pollinators were assessed in one orchard. Blow flies were found to be as effective as the honey bee, whereas the housefly was less so. Yield of small caged 'Keitt' mango trees was minuscule (1 kg/tree), whereas open-pollinated trees carried a good crop of 61 kg/tree. The introduction of three pollinators - the honey bee, the bumblebee (*Bombus terrestris*) and the housefly - resulted in higher yield. Several species of flies, particularly common blow flies *Chrysomya albiceps* and *Lucilia sericata*, may serve as very effective mango pollinators, and are less likely than honey bees to abandon the mango orchard for more attractive blooms, in Israel as well as the United States (Dag and Gazit 2000).

When looking at arthropods associated with mango leaves and weeds during different seasons and management systems, Cabrera-Mireles et al. 2011 found weeds had the greatest arthropod abundance compared with leaves of mango trees during winter. In weeds, the greatest abundance, diversity, and richness was found in the “technified” (conventional) farms followed by the minimum traditional system. In mango leaves, the greatest abundance was registered in the transitional system while the technified had the lowest abundance. Major diversity was found in the minimum traditional system, and major richness in the system substituted by sugarcane. The system substituted by sugarcane had a negative impact on abundance, diversity, and richness of arthropods in weeds and foliage during the dry season. The less technified system (minimum traditional) always maintained higher diversity and richness, independently of the season of the year.

The effect of management systems was significantly different with greater values during the rainy season, yet similar in the dry and winter seasons. This could be potentially due to the

increased weed biomass found in the rainy seasons. On the contrary, the dry and winter seasons affected drastically arthropod abundance, explained by the scarcity of living resources compared with those in the rainy season, where there was greater richness, abundance, and biomass of plant species. All this allows the establishment and survival of arthropods with different habitats, leading to a greater abundance of organisms.

Mango habitat maintains a broad diversity of organisms, but this has not been properly documented (Plan Veracruzano de Desarrollo, 2010). Mango agroecosystems might be a good example of arthropod biodiversity reduction, due to the application of different levels of technology in the same area. This study made clear the importance of crop weeds as the habitat of several arthropods, being the substrate with greater abundance of organisms, compared to mango and sugarcane leaves. The difference in arthropod abundance among the substrate studied could have been related with the heterogeneity of weeds given that weed heterogeneity favors habitat differentiation, and increases opportunities of coexistence and interaction among species, leading to a greater resource efficacy.

Traditional agriculture utilizes selective weed control, even promoting weed presence for its beneficial role in the crop or additional utility for the grower (Altieri, 1999). Most of the time it is easier to try to manage the weeds than eliminating them; they can be useful in preventing soil erosion or helping in reduction of pest populations. In mango agroecosystems, weeds might serve as an appropriate habitat for predatory ants, as well as Staphylinidae and Histeridae beetles which act directly on *Anastrepha* fruit flies (Cabrera-Mireles et al. 2011).

Surrounding vegetation of mango trees may serve as refuge or feeding source for wasps that are parasitoids of pests, such as scales (Hemiptera: Coccoidea and Diaspididae), and *Anastrepha* fruit flies. However, the potential of natural enemies can be diminished by

intensification of the agricultural management system, similar to what was found in several trials with natural enemies in disturbed systems (Platt et al., 1999; Gámez-Virués et al., 2009; Mailafiya et al., 2010). In all systems, insects with the greatest richness were from Hymenoptera, Hemiptera, Coleoptera and Diptera, compared to several arthropod orders. Mango foliage also favored dipterans and thysanopterans.

Small patches of native flora, planted in nonproductive margins of large mango orchards, enhanced abundance and diversity of mango flower visitors, ameliorating the negative effects of isolation from natural habitat. These increases were associated with significantly higher mango production in all three major cultivars, including Keitt (Carvalho et al. 2012). When looking at native flowers to facilitate crop pollination in mango, pesticide use and isolation from natural habitat were associated with declines in flying visitors and mango production (kg of marketable fresh fruit) (Carvalho et al. 2012). However, presence of Native Flower Conservation Areas (NFCAs) ameliorated these declines, and NFCAs did not harbor any mango pests. The most abundant flower visitors to mango -ants- or initial fruit set was significantly affected by distance, pesticides or NFCAs, suggesting that although fertilization is associated with factors unaffected by isolation from natural habitat and pesticide use (i.e. self- and ant-pollination), viable fruit set (and ultimately, production) requires cross-pollination, for which flying visitors are essential (Carvalho et al. 2012).

To study native flower conservation areas, two perennial native species that are present within the regional natural vegetation, flower before and during mango flowering season, and have different floral structures were used. However, NFCAs with more plant diversity could have achieved a larger effect than found. Two co-flowering species will compete with or facilitate each other for flower visitors, some studies suggest that facilitation is more likely

between plants with unequal flower abundance (Ghazoul 2006) or that attractive species may facilitate less attractive species, as is the case with mango.

The presence of native flowers significantly increased both species richness and abundance of mango flying visitors in orchards far from natural habitat (Carvalho et al. 2012). Diversity declined with isolation from natural habitat for all cultivars, being on average 47% lower at 300m from natural habitat, (i.e. on average c. 2 species were lost) than near natural habitat. In orchards with NFCAs, diversity was only 7% lower. A negative effect was detected for pesticide use, on both abundance (41% decline) and diversity (40% decline) of mango flying visitors for all cultivars. Honey bees more than doubled near NFCAs and the proportion of honey bees among mango flower visitors increasing significantly.

Early fruit set of cultivars Kent and Keitt tended to decline with distance from natural habitat, and mangoes had a high rate of fruit drop/abortion (80–100% per inflorescence) and final production of both Kent and Keitt showed significant declines with distance from natural pollinator habitat. Production was significantly higher near NFCAs, with an average increase of 15 kg of commercially suitable mango per tree.

Low-abundance flowering plants within agricultural lands can significantly increase flower visitors to crops, observed in crop fields with variable weed abundance (e.g. sunflower, Carvalho et al. 2011; watermelon and pepper, Winfree et al. 2008). Honey bees were markedly influenced by the presence of NFCAs, suggesting that although bees tend not to be attracted to mango (Carvalho et al. 2010), if enticed to forage within mango fields they can contribute to mango pollination.

Farmers often remove weeds from agricultural fields through fear of competition for soil nutrients and flower visitors. However, the presence of flower diversity before and during crop

flowering facilitates, rather than reduces, pollination of the hyperabundant crop flower resource (Winfree et al. 2008; Carneiro et al. 2011; Carneiro et al. 2012). For a perennial pollinator-dependent crop like mango, creation of flower conservations can be profitable, improving production within existing areas and reducing the need for agricultural expansion, contribute to the conservation of biodiversity within a region, and increase the habitat and resources for insects within farms (Carneiro et al. 2012). For perennial tree crops such as mango, negative effects of nonparasitic flowering low growth form perennials are unlikely.

3. Methodology

3.1 Site Description

The experiment was installed at a conventional mango farm (20 acres), variety “Keitt”, within the major agricultural area of Homestead, Florida (25°29'42.9"N 80°29'30.2"W). Trees were evenly spaced in 20 x 20 feet intervals. Distance between rows was approximately 20 feet. Average yearly production is approximately 7000 bushels. Two treatments were applied to the trees using a randomized design: weeds vs. no weeds. Three sections of 10 trees were assigned per treatment, with buffer rows and trees surrounding the treatments. For the weed treatment, unadulterated weed growth was allowed between the trees, and weed species identified. For the no-weed treatment, no weed growth was allowed around the crop, using mechanical removal techniques such as mower and string trimmer, a tool for cutting grasses, small weeds, and vegetation near obstacles or on irregular terrain. All weed specimens within the weed treatment were vouchered and identified.

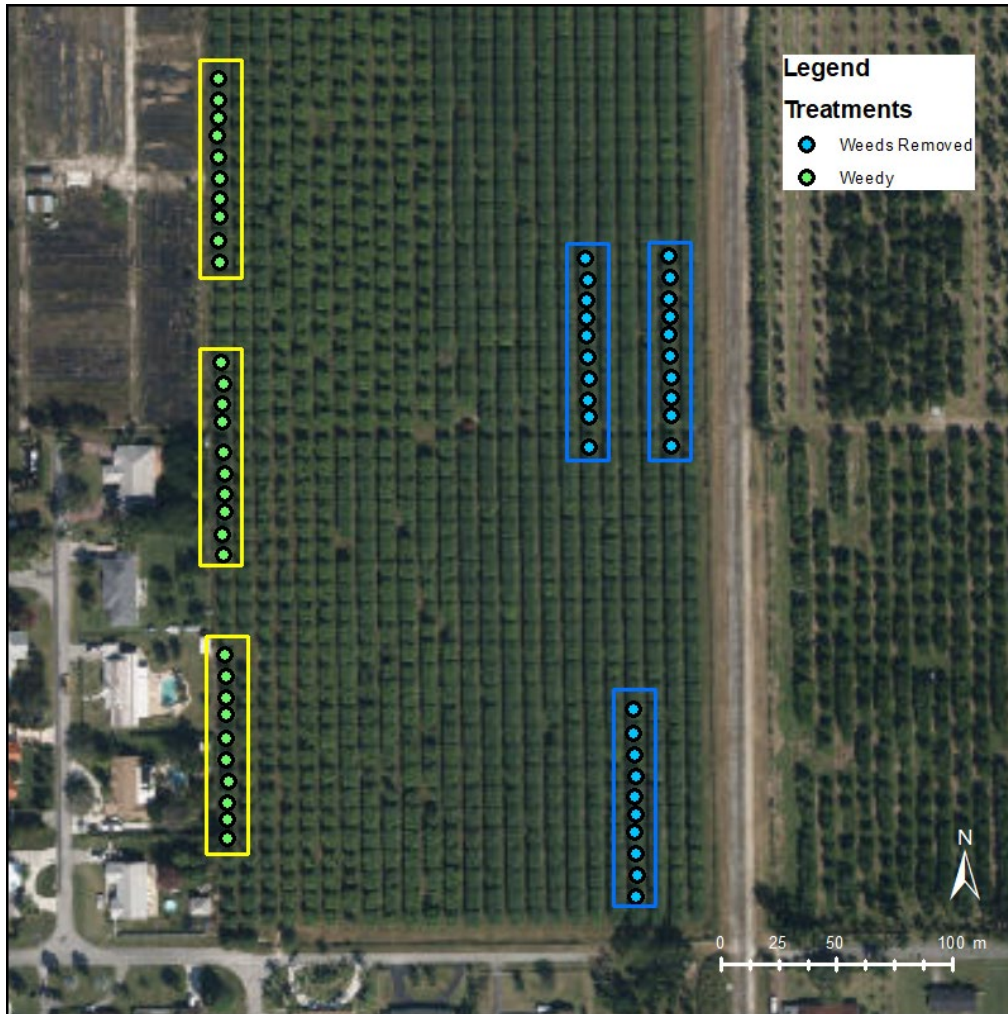


Figure 2 Farm Treatment Map of Weed and No-Weed Trees

3.2 Field Data Collection

Observations and collections of insects were from the mango trees and weeds. I recorded and collected insects interacting within each treatment, and on the crop, *M. indica*. I collected data from both treatments weekly from 11/20/2019- 05/08/2020, including the major 8-week blooming period of mango. I spent 5 minutes recording insect interactions, observing, and collecting specimens on the weeds around each of the 30 weedy mango trees, and 5 minutes per each mango tree in the weed treatment, totaling 7,500 minutes of observations across the 25 weeks. For the no-weed treatment, I spent 5 minutes recording insect interactions, observing, and

collecting specimens on each of the 30 no-weed mango trees, totaling 3,750 minutes of observations across the 25 weeks. Timed focal point observations were conducted weekly before, during and after the peak mango flowering season (i.e. from November until May) similar to methods outlined in Carvalheiro et al. 2012. In each 5- or 10-min observation period per tree I observed, recorded, and collected all insects, and all flower visitors were recorded and collected for identification.

Timing of observations per treatment was alternated each data collection day by alternating which trees were visited first to have a breadth of observations for each tree across the day. Insect specific Pentax binoculars were used to observe specimens in the upper canopy of the mango trees if needed. Each insect specimen was collected if novel or sight- ID wasn't possible. Additionally, specimens were collected if they displayed notable behavior such as pollinating flowers or feeding, using an insect aspirator, collection bag, or net. Specimens collected from the field were immediately placed into a plastic bag that was stored in a freezer (0°C) for further identification. There were two flushes of inflorescences on the mango trees during this season, allowing insects to be collected in mango flowers from 12/05/2019-05/08/2020.

3.2.1 Statistical Analyses

All statistical analyses were performed with SPSS software. Descriptive analyses will be conducted to describe the variables of interest overall and by treatment. These include means, standard deviations, frequencies, and proportions. To compare means between insects we will use two-sample t-test and to compare proportion we will use chi-square test to see if the observed distribution of insects differs from the expected distribution. Statistical analyses of effect of treatment on insects compared between the weedy and no-weed treatment, as well as these

effects on fruit yield, will be done using a general linear model by applying multivariate ANOVA (MANOVA) with insect/fruit yield measures as the dependent variable (multiple Y's) as a function of treatment while adjusting for tree age. Multivariate tests such as Pillai's Trace, Wilk's Lambda, Hotelling's Trace, and Roy's Largest Root will be examined for overall model significance, followed by further analyses using simplified F-test comparison adjusting for multiplicity.

Statistical analyses of soil will be done for pH using a paired t-test and univariate Analysis of Variance (ANOVA). Soil Carbon and Nitrogen will be analyzed using a paired t-test. Soil Phosphorus will be analyzed using a t-test, one-way ANOVA, and nonparametric tests. Leaf chlorophyll SPAD (Soil-Plant Analyses Development) results will be analyzed using a univariate Analysis of Variance (ANOVA). Comparison of insect proportions between weed species will be done using a chi-square test of independence. Analysis of proportion of plant species pollen carriers by several insect types collected in flowers will be done using a chi-square test. All differences or associations will be considered significant at the alpha level of 0.05 after the adjustment for multiplicity where appropriate.

3.2.2 Farm Maintenance

The farm is made up of 24 rows with 47 mature mango trees each and a mix of Tommy, Keitt, Kent, and Florida Red varieties. Three sections of 10 trees for each treatment were assigned to the Keitt mango trees. The weed treatment was placed in row 24, with buffer trees separating the sections, due to restrictions in field maintenance of separating the treatment across multiple rows. The 3 control sections were randomly assigned to 10 trees each in rows 2, 4, and 6.

The management practices used at the study site are ecologically oriented and minimal for a conventional farm compared to other mango farmers, and no insecticides are used by the grower to allow balanced insect biological control of pests. Mango farmers in the United States are limited in what chemicals they can apply to trees compared to foreign growers, for example in the foreign use of Topsin to treat mango malformation, outlawed in the U.S (Muhammad et al. 1999).

The major chemical applied to the trees were fungicides, including synthetics Bravo (chlorothalonil), biological fungicide Double Nickel 55 (*Bacillus amyloliquifaciens* strain D747), and organic OxiDate (hydrogen peroxide) to kill anthracnose, bacterial, and fungal pathogens. Spraying was based on weather once flowering of trees began till fruiting in July, about every 1-4 weeks depending on rain and humidity. When conditions are dry fungicide spray is not needed versus the rainy season when it was applied weekly. Before fruiting micronutrients are added to the trees such as Zinc, Manganese, and Iron. Sulfur, Potassium, and Magnesium is applied once a year spring, as well as cow manure instead of synthetic fertilizer.

Pruning of trees is an integral process for mango farmers and was done immediately after harvest using a TOL machine hedger and topper as well as by hand to thin out tree canopies, allow sunlight penetration, prevent fungus proliferation, and promote flower and fruit production (Davenport 2006). Mowing occurred in the farm as needed- outside of the weed treatment- with a sickle bar mower beneath the trees instead of herbicidal eradication of weeds. Control trees were maintained weed free using mechanical means of mowing/string trimming, as needed based on weed growth. The trees were not irrigated, as mango has low water needs and taproots which can access the water table (D. Lyons, pers. comm.).

3.2.3 Weed Species

Weed species for the weed treatment were vouchered and identified. 75 different species from 27 families were identified. 34 native, 38 non-native of which 6 are Category I invasive and 5 Category II invasive. Invasive exotic plants are termed Category I invasive when they are altering native plant communities by displacing native species, changing community structures or ecological functions, or hybridizing with natives (FLEPPC 2019). This definition does not rely on the economic severity or geographic range of the problem, but on the documented ecological damage caused. Category II invasive exotics have increased in abundance or frequency but have not yet altered Florida plant communities to the extent shown by Category I species. These species may become Category I if ecological damage is demonstrated.



Figure 3 Left: *Macropitium lathyroides*, "Phasey Bean" purple flowers, non-native.

Figure 4 Right: *Oxalis*, "Wood Sorrel", non-native.

3.3 Pollen Analysis

Insects feeding in flowers were collected and pollen samples taken to determine which plant specimens pollen they carry. The weed the insect was using as a resource was collected, vouchered, and identified. Pollen samples from weed and mango flowers were both taken, and cross referenced with insect specimen pollen. All weed and mango flower buds were vouchered and created into reference pollen slides in their pre-bloom stage to eliminate cross-pollination from insect visitors. Flower buds were collected in the field prior to them opening and transported to the lab. Stigmas were placed on microscope slides; anthers were dehisced, and pollen mounted on microscope slides in basic fuchsin gel to lightly stain pollen grains red (Kearns and Inouye 1993; Beattie 1971). Pollen grains were photographed under a light microscope at 40× magnification and created into a reference collection.

Insects feeding in pollen were analyzed under a dissecting microscope, and pollen scraped off the specimen's body and made into a slide for analysis. Pollen grains were taken from insect specimens, mounted onto microscopic slides in fuchsin gel, and photographed under a light microscope at 40× magnification. Pollen carried by insects was identified based on the reference collection. Photos of reference pollen and insect body pollen were analyzed to determine the plant from which the pollen on insects came from.

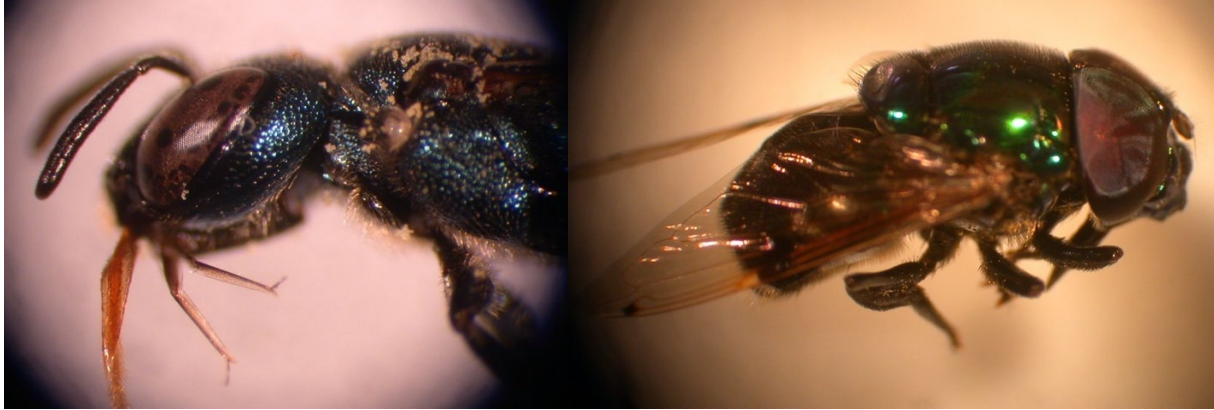


Figure 5 Left: *Ceratina* bee with Pollen

Figure 6 Right: *Ornidia obesa* Hoverfly with Pollen

3.4 Rearing

Nymphs and larval specimens were collected in the field and stored in individual containers in Dr. Suzanne Koptur's Plant Ecology lab at FIU and raised to assess possible emergence of parasitoids as well as for identification. Nymphs and larval specimens are difficult to identify, therefore they were raised by feeding them their host plant/prey and reared into their adult life stage to be identified.



Figure 7 Caterpillar Rearing from Weeds

Additionally, should a nymph or larval specimen have been parasitized prior to collection, the parasitoid which emerged was identified. Rearing was done concurrently during the field collection season as nymphs, larvae, and caterpillars were collected from the field. Rearing followed the procedure described in Koptur et al. (2015), using 1-gallon plastic bags per insect specimen to prevent contamination and assessment of parasitism. Feeding and monitoring was completed during specimen development until death, metamorphosis to adult stage, or emergence of a parasitoid. Parasitoid specimens were preserved in a 90% ethanol solution for further identification.

3.5 Fruit Yield

This study was completed within one year, from the end of one production season to the next. Sampling of fruit yield per tree was done visually as well as counted when harvested by the grower in concert with the producer's collection strategy at the end of the harvesting season. Visual fruit yield counts were done after bloom and all fruit had been pollinated and set. Fruit was counted per tree twice by two different observers 04/29/20 and 05/03/20 and averaged. A marker for the starting point of observation was placed on the ground, and an observer used a hand clicker counter and worked their way around the tree counting each individual mango fruit.

Data gathered on mango production included visual data on early fruit set (i.e. number of unripe fruits per tree, c. 6 weeks after the end of flowering ceased) (Carvalho et al. 2012). Early fruit set is only an indication of pollination efficiency but may not be a good indicator of pollination quality and, hence, of final crop production and economic value (Carvalho et al. 2012). Therefore, we also obtained information on final production as lbs of commercially suitable mangoes directly from the farmer. Fruit was harvested green for the Asian market early in the season on 05/04/2020, and again in July. Harvest of mango is done manually by pickers; therefore, a count was able to be taken as each tree was picked.

3.6 Soil Analysis

Soil samples were taken towards the beginning and at the end of data collection. A first sample was taken during the flowering stage of the mango trees on 02/23/2020, from two random trees per all 3 sections of both treatments for 12 total samples. A sample was then taken at the end of data collection from the same trees and after the harvest season concluded on 06/03/2020. Soil from the four corners of each sample tree were taken and mixed into a sample bag, and then analyzed for soil pH, carbon, nitrogen, and phosphorous to compare between treatments. Subsamples of collected soil were dried at 30 degrees Celsius, ground, and sieved through a 2 mm sieve, and analyzed at FIU's soils lab.

3.6.1 Total Carbon and Nitrogen (Soil)

Dried and ground (mortar and pestle) soil samples were analyzed for Total Carbon and Total Nitrogen by dry combustion with a LECO CN Analyzer (St. Joseph, Michigan).

3.6.2 Total Phosphorus (Soil)

Total Phosphorous in the soil samples were analyzed following the USEPA (1983) method colorimetrically with the SEAL Analytical AQ2 Discrete Auto Analyzer (Mequon, Wisconsin). For sample preparation oven dried samples were finely ground with a mortar and pestle. Between 0.017- 0.021 g of sample was added to 20 ml glass scintillation vial with Teflon Cap. 0.2 ml of 0.17 M MgSO₄ was added to each vile. The vials were then heated in the oven overnight at 70 °C uncapped. The vials were then removed from the oven and placed in the furnace at 500 °C for 4 hours. Once the samples had been ashed, 5 ml of 0.2N HCL was added to each vial and then capped tightly. The vials were then heated in the oven for 30 minutes at 80 °C. Ten ml of deionized water (DIW) was added to each vial. They were then capped tightly, vigorously shaken, and left overnight to settle.

Standard solution preparation of Phosphate working standard solutions were prepared starting with a phosphate stock standard solution of 4.394 g Potassium dihydrogen orthophosphate diluted to 1 l with DIW. From there, four working standard solutions were made with concentrations mg P/l of 0, 0.1, 0.4, 0.5, and 1. 27.

The AQ2 machine was turned on, and after 30 minutes, the daily startup was conducted, and water baselines were recorded. Reagents were added to the machine; wedge 1 was the phosphate color reagent, wedge 2 was the ascorbic acid, and wedge 3 was the CCV or 0.5 phosphate standard. Two hundred μ l of sample solution was added to its own sample cup and diluted with 1800 μ l of DIW for a 10x dilution. Duplicate samples were added in after every 10 samples and standards were added after every 20 samples. The sample tray locations were inputted into the computer scheduling sheet and the machine was run until all samples were tested.

3.6.3 Soil pH

Readings were taken to determine if there was a change in acidity caused by weeds which might affect growth. Soil slurries with a 3:1 water to soil ratio (4g distilled DI water to 2g wet soil) were made and pH readings taken using a Thermo Scientific Orion pH probe in the FIU Soil lab in which a glass electrode was placed in the mixture of soil and deionized water. The ideal pH range ideal for mango cultivation is between 4.5 and 7.5 pH, which is neutral to acidic soil. The mango tree does tolerate slightly alkaline soil (Carter, 2020).

3.7 Chlorophyll Analysis

To monitor and compare plant health between treatments, the average leaf chlorophyll concentration of each plant was measured using the Soil-Plant Analyses Development (SPAD) 502 Plus Chlorophyll Meter. A SPAD chlorophyll meter was used as a diagnostic tool to measure mango tree nitrogen status during the flowering stage on 02/23/2020. Two random trees

per section (12 total) in both treatments were chosen for the non-invasive SPAD analysis. Three new growth leaves, three regular, and three old leaves were analyzed per tree and averaged.

4. Results

4.1 Insects on Mango

There was a significant effect of treatment (weeds) on mango flower visitors ($F=31.109$, $df=1, 57$, $p<0.0001$) and parasitoids ($p<0.0001$), and nearly for predators ($F=29.525$, $df=1,57$, $p=0.076$) determined by Multivariate Generalized Linear Model statistics. There was no significant difference between herbivores on mango trees ($F=0.001$, $df=1, 57$, $p=0.972$). There was significantly greater counted ($F=181.317$, $df=1, 57$, $p<0.0001$) and harvested ($F=89.344$, $df=1,57$, $p<0.0001$) fruit in the weed treatment than weed-free. Age of mango trees (D. Lyons, pers. comm.) was considered as a covariate, however it was not significant.

Table 1 Weed Family and Species in Mango Farm			
Family	Species	Common Name	Native/Invasive
Anacardiaceae	<i>Ruellia blechum</i>	Green Shrimp Plant	Non-native FLEPPC II
	<i>Ruellia ciliatiflora</i>	Hairy Flower Wild Petunia	Non-native
	<i>Schinus terebinthifolia</i>	Brazilian Pepper tree	Non-native FLEPPC I
Arecaceae	<i>Arecales</i>	Palm	Native
Asteraceae	<i>Bidens alba</i>	Spanish Needles	Native
	<i>Conoclinium coelestinum</i>	Blue Mistflower	Native
	<i>Emilia fosbergii</i>	Florida Tasselflower	Non-native
	<i>Parthenium hysterophorus</i>	Santa Maria Feverfew	Non-native
	<i>Conyza canadensis</i>	Canadian Horseweed	Native
	<i>Ageratum houstonianum</i>	Floss Flower	Non-native
Brassicaceae	<i>Lepidium virginicum</i>	Virginia Pepperweed	Native
	<i>Lepidium densiflorum</i>	Common Pepperweed	Non-native

Burseraceae	<i>Bursera simaruba</i>	Gumbo Limbo	Native
Commelinaceae	<i>Commelina diffusa</i>	Climbing Day flower	Non-native
Convolvulaceae	<i>Ipomoea</i>	Morning Glory	Native
	<i>Ipomoea hederifolia</i>	Scarlet Morning Glory	Native
	<i>Ipomoea indica</i>	Blue Morning Glory	Native
Cucurbitaceae	<i>Melothria pendula</i>	Creeping Cucumber	Native
Cyperaceae	<i>Cyperus croceus</i>	Baldwin's Flatsedge	Native
Euphorbiaceae	<i>Acalypha ostryifolia</i>	Hophornbeam Copperleaf	Native
	<i>Acalypha</i>	Copperleaf	Non-native
	<i>Euphorbia heterophylla</i>	Mexican Fireplant	Native
	<i>Euphorbia hyssopifolia</i>	Hyssop Spurge	Native
	<i>Acalypha arvensis</i>	Field Copperleaf	Non-native
	<i>Poinsettia cyathophora</i>	Wild Poinsettia	Native
	<i>Euphorbia hirta</i>	Asthma Plant	Native
	<i>Rhynchosia minima</i>	Least Snout-Bean	Native
	<i>Crotalaria incana</i>	Shake-shake	Non-native
	<i>Leucaena leucocephala</i>	White Lead tree	Non-native
	<i>Desmanthus virgatus</i>	Wild Tantan	Non-native
	<i>Desmodium incanum</i>	Creeping Beggarweed	Non-native
	<i>Macroptilium lathyroides</i>	Phasey Bean	Non-native
	<i>Indigofera spicata</i>	Creeping Indigo	Non-native
Geraniaceae	<i>Geranium carolinia</i>	Carolina Geranium	Native
Lamiaceae	<i>Salvia occidentalis</i>	West Indian Sage	Native
Malvaceae	<i>Sida ulmifolia</i>	Common Wire Weed	Native
Moraceae	<i>Ficus aurea</i>	Strangler Fig	Native
Oleaceae	<i>Jasminum dichotomum</i>	Gold Coast Jasmine	Non-native FLEPPC I
	<i>Jasminum fluminense</i>	Brazilian Jasmine	Non-native FLEPPC I
Oxalidaceae	<i>Oxalis</i>	Wood Sorrel	Non-native
Papaveraceae	<i>Argemone mexicana</i>	Mexican Prickly Poppy	Native
	<i>Fumaria officinalis</i>	Common Fumitory	Non-native
Phyllanthaceae	<i>Phyllanthus amarus</i>	Carry-me Seed	Non-native
Poaceae	<i>Paspalum conjugatum</i>	Hilo grass	Native
	<i>Urochloa maxima</i>	Guinea grass	Non-native FLEPPC II

	<i>Sporobolus jacquemontii</i>	American Rat's Tail Grass	Non-native FLEPPC I
	<i>Digitaria ciliaris</i>	Southern Crabgrass	Native
	<i>Melinis repens</i>	Natal grass	Non-native FLEPPC I
	<i>Stenotaphrum secundatum</i>	St. Augustine grass	Native
	<i>Sorghum halepense</i>	Johnson grass	Non-native
	<i>Panicum maximum</i>	Guinea grass	Non-native
	<i>Cenchrus</i>	Sandbur	Native
	<i>Neyraudia reynaudiana</i>	Burma Reed	Non-native FLEPPC I
Psilotaceae	<i>Psilotum nudum</i>	Whisk Fern	Native
Rubiaceae	<i>Spermacoce remota</i>	Woodland False Buttonweed	Native
	<i>Spermacoce verticillata</i>	shrubby false buttonweed	Non-native FLEPPC II
	<i>Richardia brasiliensis</i>	Brazil Pusley	Non-native
	<i>Richardia scabra</i>	Florida Pusley	Non-native
	<i>Richardia grandiflora</i>	Large-flower Pusley	Non-native FLEPPC II
Sapindaceae	<i>Cardiospermum</i>	Soapberry	Native
Solanaceae	<i>Solanum americanum</i>	American Black Nightshade	Native
Urticaceae	<i>Pouzolzia</i>	Pouzolzia	Non-native
	<i>Laportea aestuans</i>	West Indian Wood-nettle	Non-native
	<i>Pilea microphylla</i>	Artillery plant	Native
Verbenaceae	<i>Lantana camara</i>	Common Lantana	Non-native
Vitaceae	<i>Cissus verticillata</i>	Possum Grape Vine	Native
	<i>Ampelopsis cordata</i>	Heartleaf Peppervine	Native
	<i>Parthenocissus quinquefolia</i>	Virginia Creeper	Non-native
Zamiaceae	<i>Zamia furfuracea</i>	Cardboard Palm	Non-native
Data from Florida Exotic Pest Plant Council: FLEPPC, and Atlas of Florida Plant Institute for Systematic Botany			

Many weed species (34) were found to be flowering herbaceous plants. Non-native species (38) were also identified and categorized as Category I and II invasive according the Florida Exotic Pest Plant Council (Table 1).

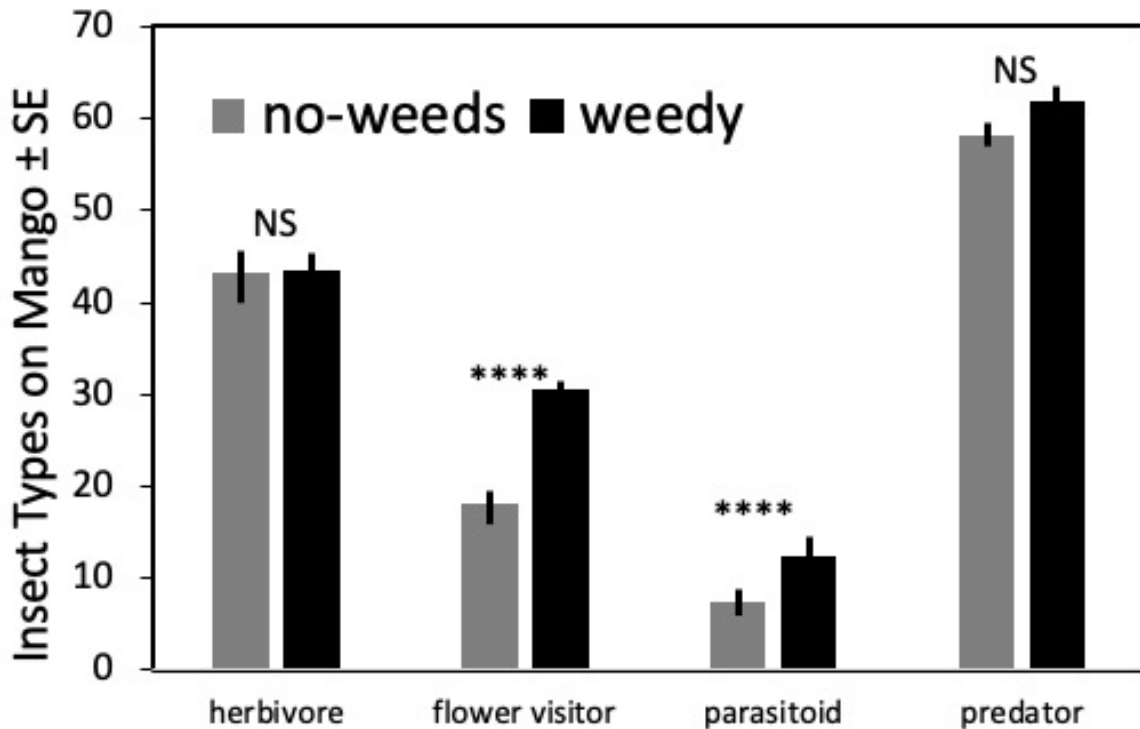
Table 2 Multivariate ANOVA Tests for effects of treatment and tree age on arthropod types.

Effect	Pillai's Trace	F	Hypothesis df	Error df	Sig
Intercept	0.93	135.1	5	53	<0.0001
Tree Age	0.13	1.62	5	53	0.17
Treatment	0.66	20.62	5	53	<0.0001

Table 3 Insect Types and Fruit Yield on Mango With or Without Weeds-significant differences have p-value in bold. GLM Multivariate ANOVA (MANOVA).

<i>Type</i>	<i>Mean</i>	<i>Std Deviation</i>	<i>N</i>	<i>Std Error</i>	<i>F</i>	<i>df</i>	<i>p-value</i>
Flower Visitor	30.47	1.93	30	0.35	31.109	1, 57	<0.0001
Weedy							
No-Weeds	18.10	3.24	30	0.59			
Predator	61.93	7.86	30	1.43	3.267	1, 57	0.076
Weedy							
No-Weeds	58.17	10.23	30	1.86			
Herbivore	43.50	3.69	30	0.67	0.001	1, 57	0.972
Weedy							
No-Weeds	43.23	6.12	30	7.86			
Parasitoid	12.33	4.05	30	0.74	29.525	1, 57	<0.0001
Weedy							
No-Weeds	7.33	2.83	30	0.51			
Harvested Fruit	179.37	65.34	30	11.88	181.317	1, 57	<0.0001
Weedy							
No-Weeds	37.67	14.63	30	2.66			

Counted Fruit	236.14	99.84	30	18.15	
Weedy					89.344 1, 57 <0.0001
No-Weeds	47.52	37.96	30	6.90	



Graph 1 Mean Number of Types of Insects on Mango by Treatment \pm SE. Significant Difference Indicated Above Each Type: NS $P > 0.05$, * $P \leq 0.05$, ** $P \leq 0.01$, *** $P \leq 0.001$, **** $P \leq 0.0001$.

Table 4: GLM- effects of treatment on counted fruit yield and main types of arthropods associated with mango trees.

Treatment	Dependent Variables	F (1,57)	P-Value
	Counted Fruit	89.3	<0.001
	Herbivores	0.001	0.97
	Flower Visitors	31.1	<0.001
	Parasitoids	29.5	<0.001
	Predators	3.3	0.76

Table 5 Multivariate ANOVA Tests for effects of tree age and treatment on arthropod orders

Effect	Pillai's Trace	F	Hypothesis df	Error df	P-Value.
Intercept	0.96	43.3	19	38	<0.0001
Tree Age	0.43	1.5	19	38	0.14
Treatment	0.90	18.7	19	38	<0.0001

Table 6 Insect Orders on Mango With or Without Weeds- significant differences have p-value in bold. GLM Multivariate ANOVA (MANOVA).

<i>ORDER</i>	<i>Mean</i>	<i>Std Dev</i>	<i>N</i>	<i>Std Error</i>	<i>F</i>	<i>df</i>	<i>p</i>
DIPTERA	65.77	10.59	30	1.93	3.792	1, 56	0.057
Weedy							
No-Weeds	59.34	14.94	29	2.77			
HYMENOPTERA	19.87	5.19	30	0.94	80.07	1, 56	<0.0001
Weedy							
No-Weeds	9.79	3.39	29	0.63			
LEPIDOPTERA	7.50	2.64	30	0.48	0.001	1, 56	0.970
Weedy							
No-Weeds	7.59	4.40	29	0.82			
HEMIPTERA	44.83	4.19	30	0.76	0.000	1, 56	0.992
Weedy							
No-Weeds	44.72	5.98	29	1.11			

ORTHOPTERA	0.27	0.52	30	0.09	0.281	1, 56	0.598
Weedy							
No-Weeds	0.21	0.41	29	0.08			
ODONATA	2.33	1.31	30	0.24	0.002	1, 56	0.968
Weedy							
No-Weeds	2.17	2.04	29	0.38			
COLEOPTERA	2.47	1.31	30	0.24	10.588	1, 56	0.002
Weedy							
No-Weeds	0.55	0.83	29	0.15			
NEUROPTERA	4.10	1.88	30	0.34	23.232	1, 56	<0.0001
Weedy							
No-Weeds	6.66	2.48	29	0.46			
ARANAE	22.77	3.69	30	0.67	0.777	1, 56	0.382
Weedy							
No-Weeds	21.93	4.09	29	0.76			
MITES	0.33	0.48	30	0.09	0.058	1, 56	0.664
Weedy							
No-Weeds	0.38	0.62	29	0.12			
THYSANOPTERA	6.10	2.19	30	0.40	8.208	1, 56	0.006
Weedy							
No-Weeds	7.66	2.24	29	0.42			
COLLEMBOLA	3.97	1.25	30	0.23	0.421	1, 56	0.519
Weedy							
No-Weeds	4.21	1.63	29	0.30			

4.1.1 Parasitoids

There were significantly more parasitoids on the weedy mango trees than the no- weed trees (Table 3, $F=29.525$, $df=1,57$, $p<0.0001$). This could be because parasitoid adults need floral nectar, and intercropping with flowering herbaceous plants increases parasitoid survivorship, fecundity, retention, and pest suppression in farms (Vattala et al. 2006; Patt et al. 1997). The added diversity provided by weeds can provide not only floral resources such as nectar to adults, but alternative larval sources for larvae through added diversity of hosts (Provost and Pedneault, 2016). These resources keep parasitoids within the farm and around the mango trees in between crop flowering- and allows them to spillover onto the mango trees. This can confer added biological control of mango pests, such as scales, moths, and hoppers. Parasitoids decrease significantly with distance from flowers (Lavandero et al. 2005); maintaining floral resources near crops may act selectively between trophic levels of hosts and parasitoids, boosting biological control of crop pests.

4.1.2 Lacewings

There were significantly more lacewing adults on the weed-free trees than the weedy mango trees as a result of a MANOVA statistical analyses (Table 6, $F=21.23$, $df=56$, $p<0.0001$). There were significantly more lacewing eggs on no-weed mango trees compared to the weedy trees (Table 7), with means of ≈ 6 and 3, respectively ($t = -6.9$, $df = 57$, $p < 0.0001$). The Chrysopidae (Neuropterans) are often released as biological control of crop pests, as their larval forms known as “Aphid Lions” are efficient natural predators of many pest species (Brooks and Barnard 1990). Their higher counts in the control trees could potentially be from an increase in their larval food source- soft bodied invertebrates such as aphids, thrips, mites, mealy bugs, whiteflies, and even small caterpillars.

Table 7 Lacewing (Neuroptera) Adults/Larvae /Eggs on Mango Trees With/Without Weeds T-Test

Type	Mean	Std Error	N	t	df	p-value
<i>Adults</i>	65.77	1.93	30	2.7	60	0.0009
Weedy						
No-Weeds	55.34	3.24	32			
<i>Larvae</i>	1.25	0.18	12	-0.18	20	0.86
Weedy						
No-Weeds	1.30	0.21	10			
<i>Eggs</i>	2.69	0.21	29	-6.9	57	<0.0001
Weedy						
No-Weeds	5.67	0.37	30			



Figure 8 Lacewing Eggs on Mango Inflorescence

4.1.3 Flower Visitors of Mango

There were significantly more flower visitors in the weed treatment than the no-weed treatments. This could be from the added floral diversity provided by weeds, enticing them to visit nearby less attractive co-flowering taxa. There were significantly more Hymenoptera on

weedy mangos (Table 5, $F = 80.07$, $df=56$, $p < 0.001$). This follows the suggestion that although bees tend to not be attracted to mango (Carvalho et al. 2010), if enticed to forage within mango fields they can spill-over and contribute to mango pollination. Added habitat diversity, nesting materials, and floral resources with weeds can create favorable habitats for hymenopterans.

Ants (Formicidae: Hymenoptera) have been found to be important in pollination of mango elsewhere (Free & Williams 1976; Anderson et al. 1982) and have been found to contribute 50% of the early fruit set and are not influenced by distance to natural habitat (Carvalho et al. 2012). Ants, however, are not very mobile and are more likely to contribute to self- than cross-pollination, potentially leading to fruit abortion.

Graph 2 Mean Number of Pollinator Insect Orders on Mango by Treatment \pm SE. Significant Difference Indicated Above Each Type: NS $P > 0.05$, * $P \leq 0.05$, ** $P \leq 0.01$, *** $P \leq 0.001$, **** $P \leq 0.0001$. Diptera *!: $P = 0.057$ weedy mango trees.

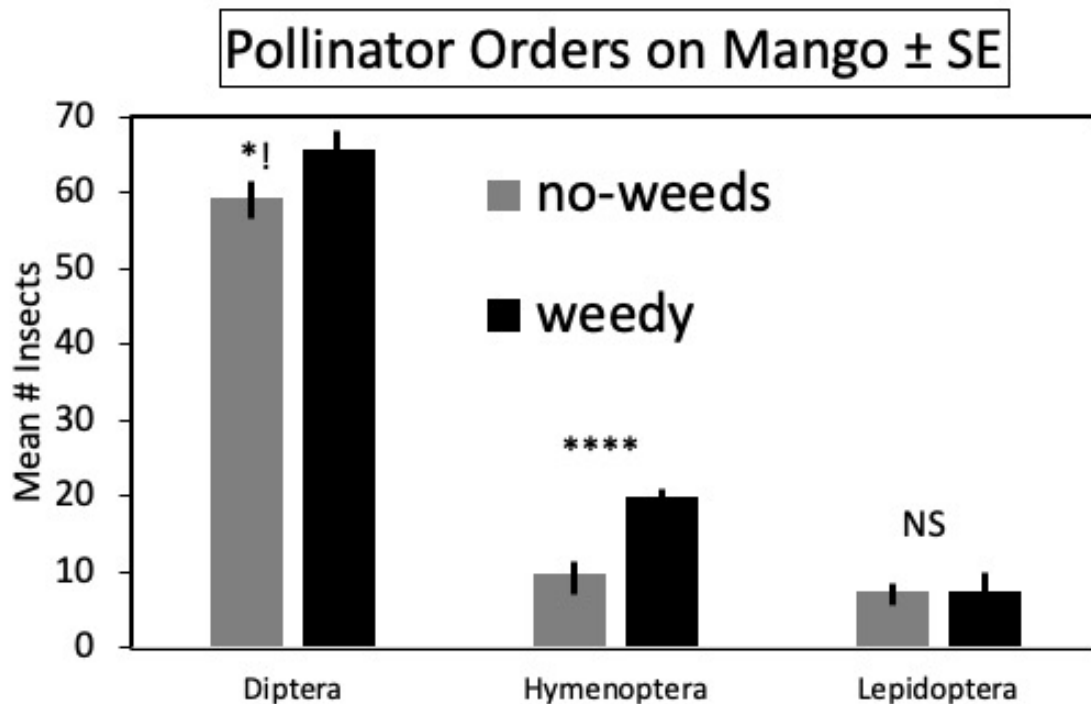


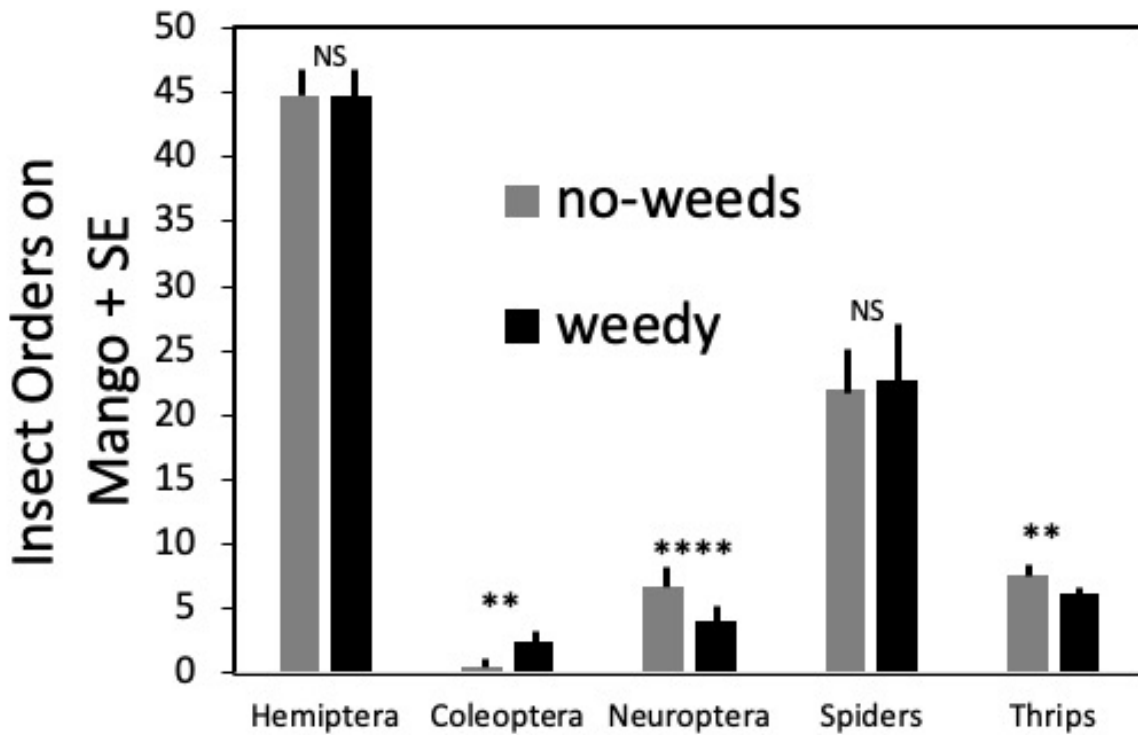


Figure 9 Top left Halictidae in *Bidens alba*, top right Skipper (Hesperiidae) in *Lantana camara*, bottom: Honey bees (*Apis mellifera*) in *Bidens alba*

Similarly, flies provided many flower visitors, and there were nearly significantly more on weedy mango trees than non-weedy (Graph 2, $F= 3.79$, $df=1,56$, $p = 0.057$). Diptera abundance and preference of mango flowers, as well as pollen carrying capacity makes them valuable pollinators of mango. Adding resources and diversity through weeds can be an effective strategy to bolster their populations, and in turn yield.



Figure 10 Left: Blow Fly (Calliphoridae), Right: Ants (Formicidae), in Mango Bloom



Graph 3 Mean Number of Insect Orders on Mango by Treatment + SE. Significant Difference Indicated Above Each Type: NS $P > 0.05$, * $P \leq 0.05$, ** $P \leq 0.01$, *** $P \leq 0.001$, **** $P \leq 0.0001$.

There were significantly more thrips (Table 6: Thysanoptera $F=8.208$, $df=1, 56$, $p=0.06$) on the weed-free mango trees than the weedy treatment trees (Graph 3). Thrips are an increasing

threat to the production of mango, as mango flower thrips feed on petals, anthers, pollen, and floral nectaries, resulting in discoloration, malformation, weakening of the inflorescence, and reduction of fruit set (Pena et al. 2002, Aliakbarpour and Rawi 2012). Thrips also can cause “bronzing” of the fruit surface due to the presence of air in emptied cell cavities and can make fruit unsuitable for fresh marketing (Nault et al. 2003). Flower thrips have a broad range of hosts, including weeds that provide refuge between mango flowering seasons. Previous research states that weeds act as a reservoir of thrips, enhancing populations in farms (Aliakbarpour & Che Salmah 2011). However, these results indicate the contrary: there are less thrips on the weedy mango trees, potentially indicating that the presence of weeds can act as a trap plant, pulling thrips away from the mango trees and inflorescences when they bloom. This interaction could also account for the significantly higher fruit yield in the weed treatment, as there were less flower thrips causing damage and reducing fruit set.

There was no significant difference in Lepidoptera (Table 6), a potential mango pollinator insect between weedy and non-weedy mango trees (Kumar et al. 2016). Other insect orders collected-Hemiptera, Orthoptera, Odonata, Ephemeroptera, Mites, and Collembola (Table 6) also did not differ significantly between treatments (the majority of which have pest or neutral tendencies). Therefore, their lack of difference, as well as the lack of significant difference between herbivores between treatment ($F=0.001$, $df=1, 57$, $p=0.972$) can confer that the presence of weeds does not push many insect orders onto mango trees, including sap-sucking herbivorous pests (Hemiptera: Pseudococcidae, Coccidae, Aphididae, Aleyrodidae, Pentatomidae, etc).

4.1.4 Predators

There was nearly a significant effect of treatment (weeds) on mango predators (Table 3: $F=29.525$, $df=1,57$, $p=0.076$), however there were more predators on weedy mango trees than

non-weedy (Table 2). This difference could be due to the enemies hypothesis which holds that predatory insects are more effective at controlling populations of herbivores in diverse systems of vegetation than in simple ones (Russell 1989; Ngatimin et al. 2014). This is in line with previous studies that weedy field strips increase populations of predatory *Orius spp.* (Hemiptera: Anthocoridae), significantly greater in plots with weedy margins than in weed-free plots. Weeds provide benefits to predators, such as nectar, pollen, shelter, and egg-laying sites as well as sources of insect prey (Atakan 2010).



Figure 11 Coccinellidae (Lady Beetle) eggs on *Bidens alba* with Aphididae Prey

There were significantly more spider webs ($F=38$, $df=8$, $p<0.0001$) and spiders in mango flowers hunting potential visitors (Table 8, $t=5.35$, $df=47$, $p=0.0001$) on the mango trees with weeds than without. There were slightly more spiders when weeds were present, however there wasn't a significant difference in the presence of spider (Aranae) adults between the treatments (Table 6: $F=0.7$, $df=56$, $p=0.38$). This could potentially mean that weeds serve as a reservoir for spiders with added microclimate and habitat diversity, increasing their movement to the mango

trees when in bloom with more webs. However, they may prefer to reside and hunt in the weeds than the mango trees, especially when not in bloom.

Table 8 Spiders on Mango Trees T-Test						
<i>Spider Webs</i>	<i>Mean</i>	<i>Std Error</i>	<i>N</i>	<i>t</i>	<i>df</i>	<i>p-value</i>
Weedy	4.00	0.34	30	5.64	44	<0.0001
No-Weeds	1.83	0.19	24			
<i>Spider Egg Sac</i>	1.94	0.30	18	0.32	40	0.75
Weedy						
No-Weeds	1.83	0.20	24			
<i>Spiders in flower hunting visitors</i>	3.93	0.30	30	5.35	47	<0.0001
Weedy						
No-Weeds	1.95	0.21	19			

4.1.5 Insect Behavior and Mango Diseases

There were significantly more instances of sooty mold found on weedy mango trees compared to non-weedy (Table 9, $F=8.8$, $df = 57$, $p = 0.004$). This could indicate increased presence of scale and hopper insects (Hemiptera) whose feeding activity excretes sweet sticky substances called honeydew which facilitates the development of sooty mold, a fungus. Sooty mold affects photosynthesis activity of leaf, negatively affecting plant health. Hopper is a serious pest of mango and in cases of severe infestation cause failure of good crop. Large number of nymphs and adults' puncture and suck the sap of tender parts of panicles, inflorescence, leaves and fruit. It results in weakening of inflorescences and affects fruit set/induces fruit drop (Gundappa & Shukla 2016).

Table 9 MANOVA Mango Diseases & Insect Behavior

	<i>Mean</i>	<i>Std Deviation</i>	<i>N</i>	<i>Std Error</i>	<i>F</i>	<i>df</i>	<i>p-value</i>
<i>Sooty Mold</i>	7.73	3.23	30	0.59	8.807	1, 56	0.004
Weedy							
No-Weeds	5.59	3.12	29	0.58			
<i>Spider Web</i>	4.00	1.84	30	0.33	38.081	1, 56	<0.0001
Weedy							
No-Weeds	1.48	1.12	29	0.21			
<i>Feeding in Flower</i>	25.90	8.93	30	1.62	45.934	1, 56	<0.0001
Weedy							
No-Weeds	10.62	8.46	29	1.57			
<i>Parasitized (mummy) Aphids</i>	0.47	0.78	30	0.14	0.530	1, 56	0.470
Weedy							
No-Weeds	0.34	0.72	29	0.13			
<i>Feeding on Prey</i>	0.83	1.21	29	0.22	0.113	1, 56	0.738
Weedy							
No-Weeds	0.72	1.07	30	0.19			



Figure 12 Hopper Nymph and Adult on Fruit Stem

Instances of mango anthracnose- an important fungal infection- were also recorded as is it a prominent field and post-harvest disease which causes direct loss of fruit and can blemish harvested fruit making it difficult to sell (Arauz 2000). There were more instances of anthracnose on the non-weedy trees, however this was not significantly different and sample sizes small ($T = -0.63$, $df = 8$, $p = 0.5$). Similarly, instances of insects with mouth on or scraping mango leaves were recorded, as well as feeding on honeydew of scales. Neither were statistically significant (Table 10), however there were more instances of feeding on honeydew in the weed treatment ($T=1.86$, $df=26$, $p=0.074$), again, possibly indicating an increased presence of sap feeding Hemiptera.

Table 10 T-test Mango Diseases & Insect Behavior							
ON MANGO:	<i>Mean</i>	<i>Std Deviation</i>	<i>N</i>	<i>Std Error</i>	<i>t</i>	<i>df</i>	<i>p-value</i>
<i>Scraping Leaf</i>	1.71	1.14	14	0.30	1.185	31	0.245
Weedy							
No-Weeds	1.37	0.50	19	0.11			

<i>Feeding on Honeydew</i>	2.56	1.98	18	0.46	1.86	26	0.074
Weedy							
No-Weeds	1.57	0.94	14	0.25			
<i>Feeding on Fruit</i>	3.86	2.94	28	0.56	0.714	49	0.479
Weedy							
No-Weeds	3.26	3.00	23	0.63			
<i>Anthracnose</i>	1.00	0.00	3	0	-0.632	8	0.5
Weedy							
No-Weeds	1.14	0.38	7	0.14			

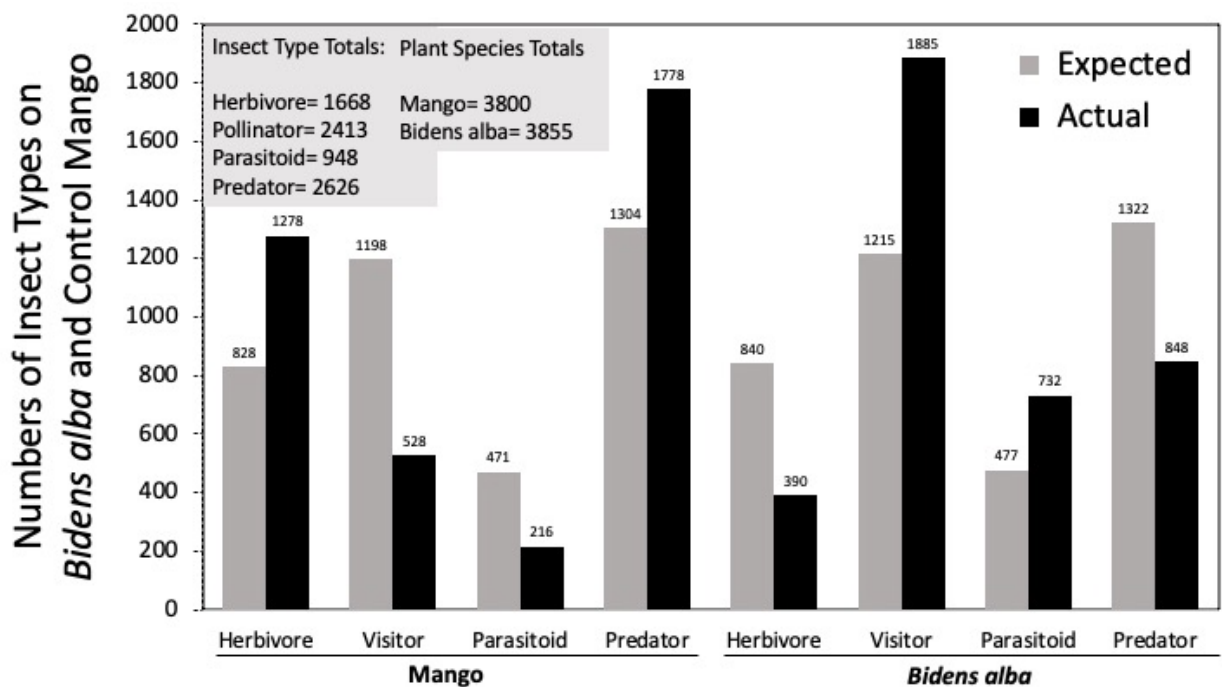
4.1.6 Insects on Weeds and Mango

A Chi-Squared Test analyzed the types of insects on the weed species in the weed treatment (Table 11). The Spanish Needle, or *Bidens alba* (Asteraceae), was the most common weed species encountered. It flowers all year long, allowing observations of insects feeding in its flower head. There were more herbivores, predators, prey, pupae, and parasitized insects found on mango trees than *Bidens alba*. However, there were more flower visitors, parasitoids, and caterpillars on *Bidens alba* than the mango trees (Graph 4, 5, and 6).

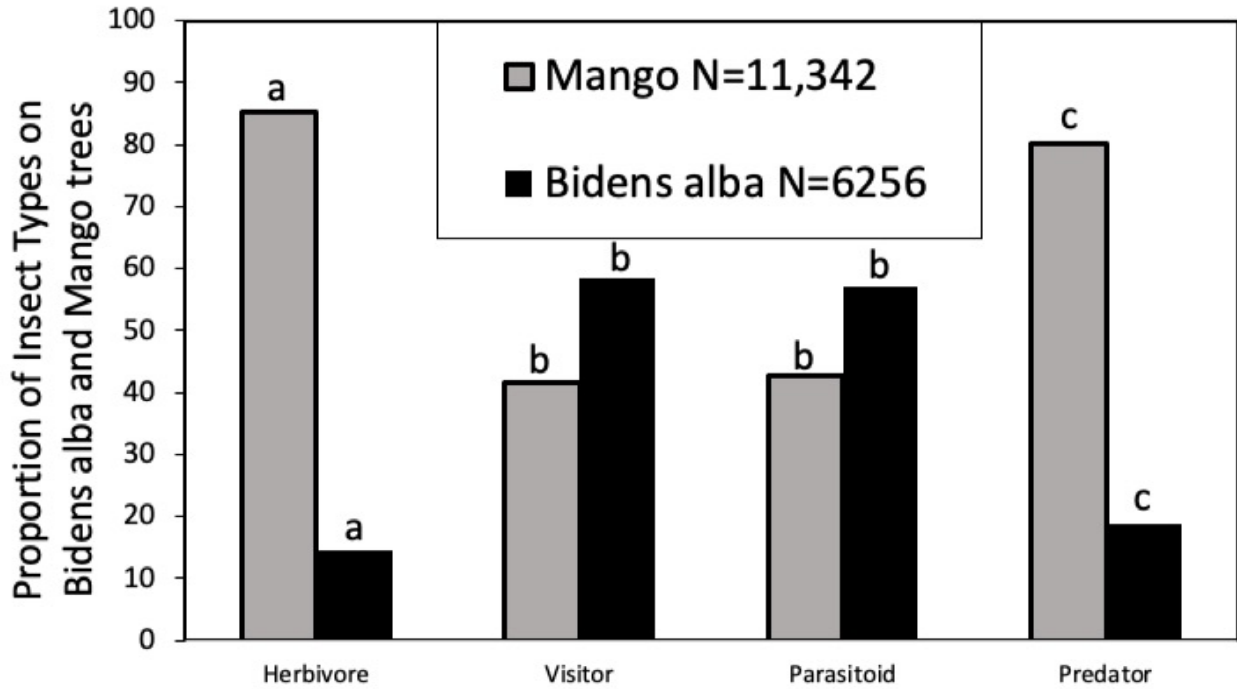
Table 11 Chi-Square Table Insect Types on 4 Common Weed Species.

	Herbivore	Visitor	Parasitoid	Predator	Total
<i>Ruellia blechum</i>	26	20	50	29	125
<i>Macroptilium lathyroides</i>	19	27	8	15	69
<i>Stenotaphrum secundatum</i>	46	50	35	60	191
<i>Sida ulmifolia</i>	16	32	19	29	96
<i>Total</i>	107	129	112	133	481

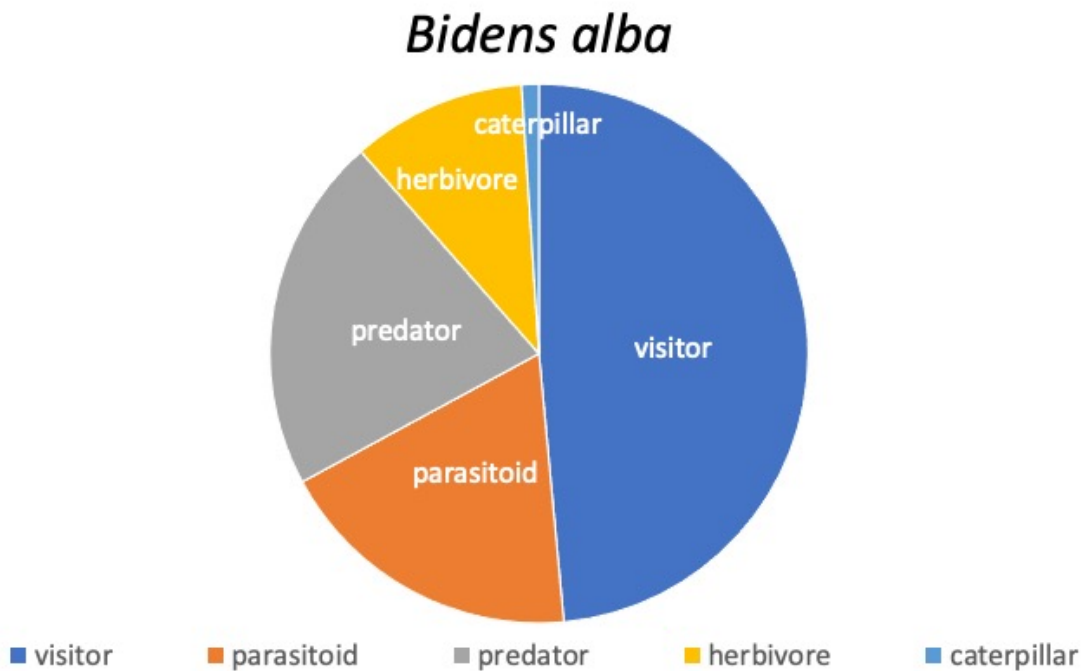
$X^2_9 = 38.065; P < 0.0001$

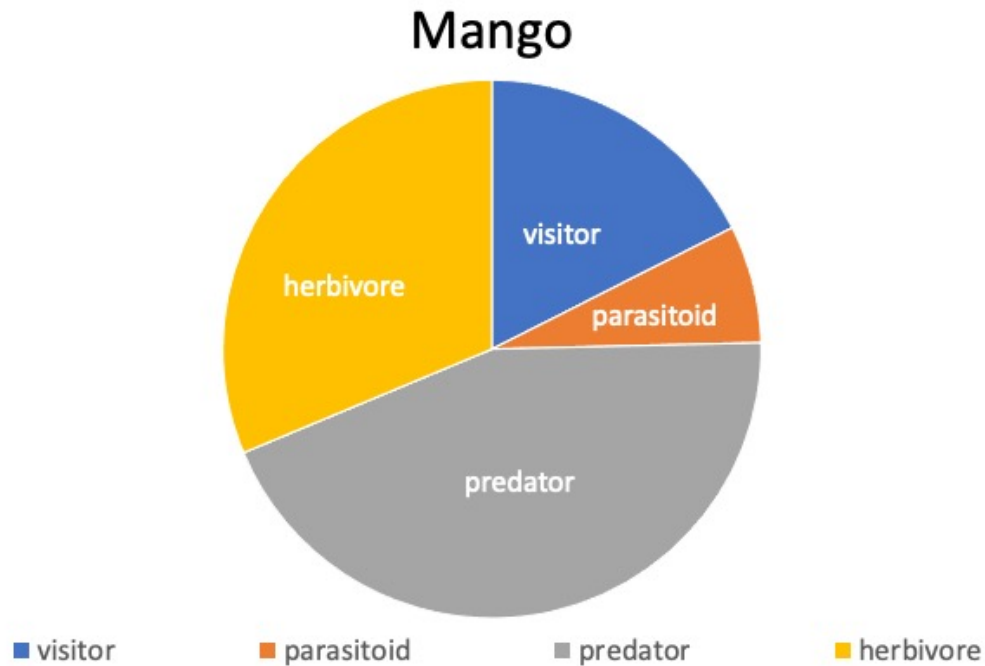


Graph 4 Expected and Actual Counts of Insect Types on the No-Weed Mango Trees and *Bidens alba* reported from a Chi-Square Test. $X^2_3 = 1845.806; P < 0.0001$.

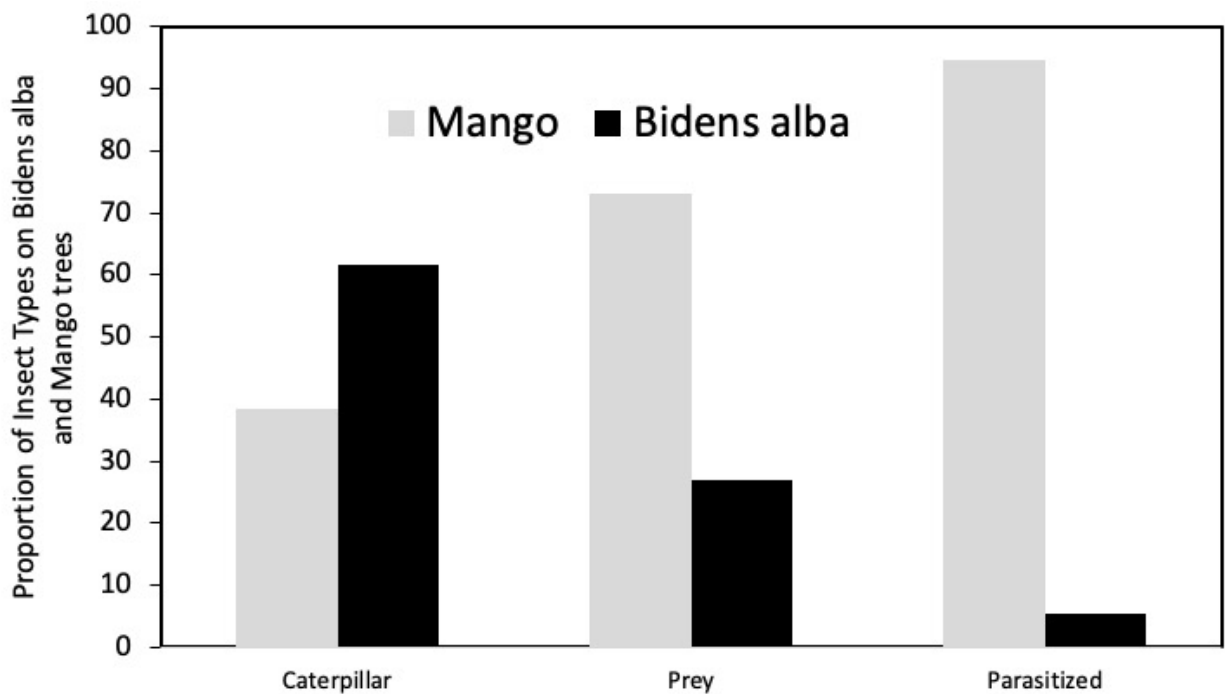


Graph 5 Proportions of Insect Types on all Mango Trees and *Bidens alba* reported from a Chi-Square Test. Different letters within plant species denotes significantly different numbers of those types. $\chi^2_3 = 2206.804$; $P < 0.0001$.



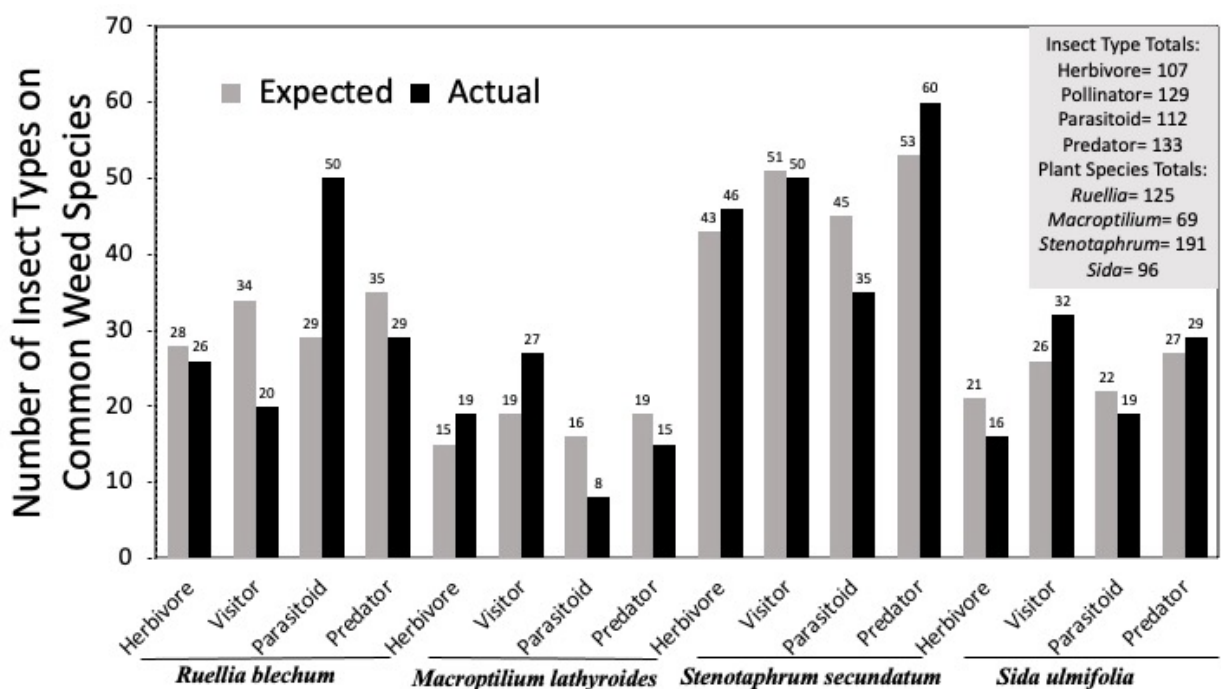


Graph 6 Percentage of Types of Insects on *Bidens alba* (Spanish needles) and Mango trees

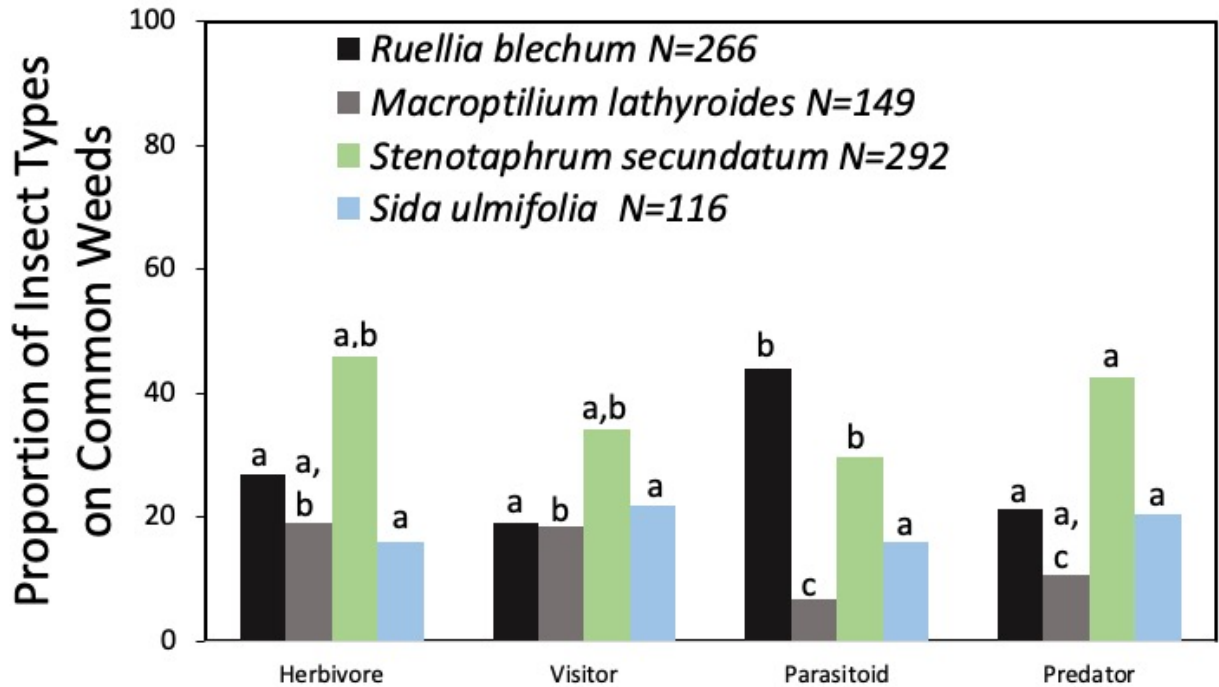


Graph 7 Proportion of Less Frequent Insect Types on Mango and *Bidens alba*. Number of arthropod counts per plant species were 6256 for *Bidens alba* and 11,342 for *Mangifera indica*.

When looking at insects on the other weed species, the majority of observations were on 7 weed species (Graph 8): *Ruellia blechum* (Shrimp weed), *Macroptilium lathyroides* (Phasey bean), *Stenotaphrum secundatum* (St. Augustine grass), and *Sida ulmifolia* (Common Wire Weed). There were the most herbivores, flower visitors, and predators on *Stenotaphrum secundatum*, and the most parasitoids and caterpillars on *Ruellia blechum*. All weed species hosted more beneficial insect types (parasitoid, predator, flower visitor) more so than pests/herbivores (Graph 9).

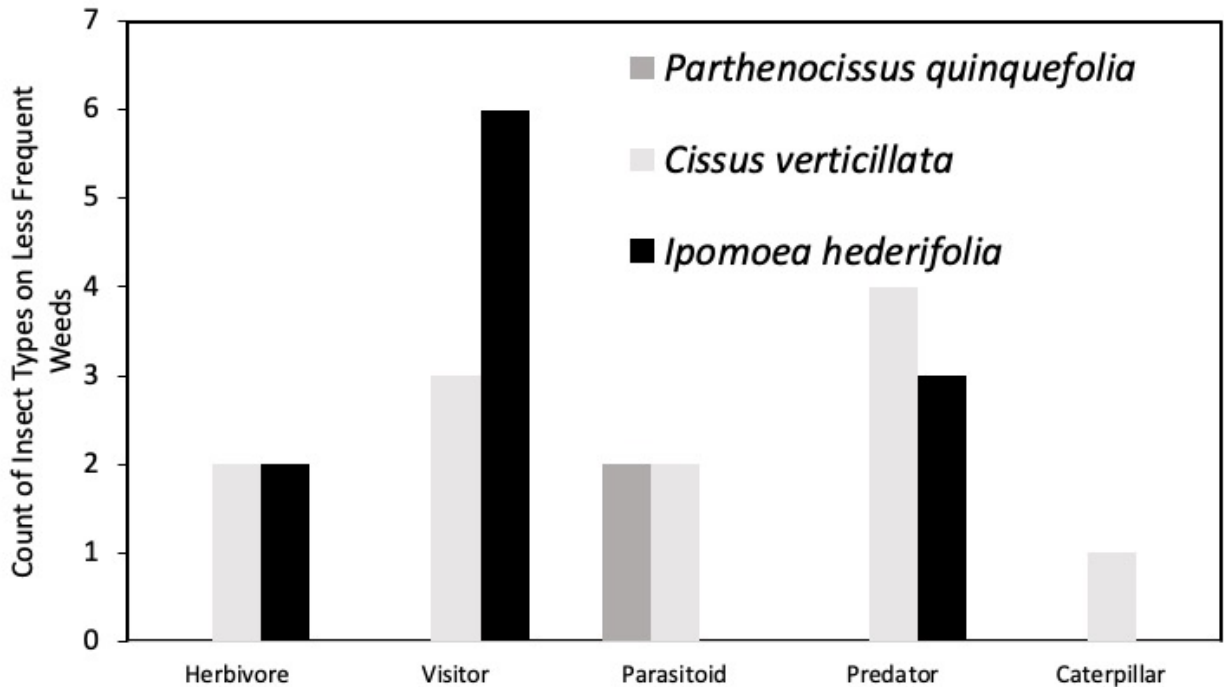


Graph 8 Expected and Actual Proportions of Insect Types on Common Weeds, reported from Chi-Square Test.



Graph 9 Proportions of Insect Types on Common Weeds. Different letters within plant species denotes significantly different numbers of those types reported from a Chi-Square Test. $X^2_9 = 38.065$; $P < 0.0001$.

Other less frequently encountered weed species were *Parthenocissus quinquefolia* (Virginia Creeper), *Cissus verticillata* (Possum Grape Vine), and *Ipomoea hederifolia* (Scarlet Morning Glory). There were more flower visitors in *Ipomoea hederifolia*, and more predators on *Cissus verticillata* (Graph 10).



Graph 10 Insect Types on Less Frequent Weeds *Parthenocissus quinquefolia* (Virginia Creeper), *Cissus verticillata* (Possum Grape Vine), and *Ipomoea hederifolia* (Scarlet Morning Glory). Since there were few observations, counts are reported rather than proportions.

Insect family observations were aggregated over all individual mango trees from both treatments, then the average of the two groups analyzed using a T-test. Of the total 126 insect families observed on the mango trees and weeds, 10 were significantly different on the mango trees when weeds were present (Table 12).

Many mango pollinating insect families were significantly greater on mango trees with weeds present: Apidae (bees) $T=4.08$, $df=55$, $p<0.001$, Calliphoridae (blowflies) $T=2.03$, $df=53$, $p=0.048$, Muscidae (housefly) $T=3.5$, $DF=57$, $P=0.001$, and Syrphidae (hoverfly) $T=3.34$, $df=44$. $P=0.002$.

Other beneficial families that were significantly greater on mango trees with weeds present were the parasitoid wasps Chalcididae $T=2.61$, $df=29$, $p=0.003$, and predatory wasps Vespidae $T=2.2$, $df=18$, $p=0.041$. Lycaenidae, the second largest family of butterflies including

the hairstreaks and blue butterflies were also significantly greater on mango when weeds were present, $T=2.18$, $df=48$, $p=0.031$.

The insect families significantly greater on the weed-free mango trees were Chironomidae (non-biting midges) $T=-2.5$, $df=58$, $p=0.016$, Coccidae (soft scales) $T=-13.2$, $df=58$, $p<0.0001$, and Drosophilidae (fruit/vinegar/pomace fly) $T=-2.68$, $df=33$, $p=0.011$.

Table 12 T-test Insect Families on Mango With/ Without Weeds							
ON MANGO:	<i>Mean</i>	<i>Std Deviation</i>	<i>N</i>	<i>Std Error Mean</i>	<i>t</i>	<i>df</i>	<i>p-value</i>
<i>Apidae</i>	4.17	2.12	30	0.39	4.08	55	0.000
Weedy							
No-Weeds	2.04	1.79	27	0.34			
<i>Calliphoridae</i>	4.07	2.14	28	0.41	2.03	53	0.048
Weedy							
No-Weeds	3.00	1.75	27	0.34			
<i>Chalcididae</i>	2.10	1.12	20	0.25	2.61	29	0.003
Weedy							
No-Weeds	1.18	0.41	11	0.12			
<i>Chironomidae</i>	4.53	1.65	30	0.30	-2.5	58	0.016
Weedy							
No-Weeds	6.00	2.75	30	0.50			
<i>Coccidae</i>	15.5	8.8	30	1.61	-13.2	58	0.000
Weedy							
No-Weeds	45.5	8.8	30	1.61			
<i>Drosophilidae</i>	1.28	0.58	18	0.14	-2.68	33	0.011
Weedy							
No-Weeds	2.06	1.09	17	0.26			

<i>Lycaenidae</i>	2.42	1.17	26	0.23	2.18	48	0.031
Weedy							
No-Weeds	1.83	0.64	24	0.13			
<i>Muscidae</i>	8.27	3.81	30	0.70	3.50	57	0.001
Weedy							
No-Weeds	5.10	3.09	29	0.57			
<i>Syrphidae</i>	6.78	2.93	27	0.56	3.34	44	0.002
Weedy							
No-Weeds	4.57	1.91	30	0.35			
<i>Vespidae</i>	1.94	1.39	16	0.35	2.2	18	0.041
Weedy							
No-Weeds	1.13	0.35	8	0.13			

4.2 Pollen Analysis

Pollen on insects was examined for 45 insects collected in flowers of weeds and mango trees to determine which species pollen they carry, including mango (Table 13). Pollen from each insect was mounted onto a slide, labeled for each insect and where it was collected from. Of these insects, 34 carried pollen: 20 Diptera, 12 Hymenoptera, and 3 Coleoptera.

Pollen from all flowering weed species was collected and voucher slides were made (38 total) and used to differentiate the pollen found on insects. Photos of vouchered weed pollen were grouped into families. A microscope stage calibration slide was used to calibrate the microscope camera for precise measurement, and the scale overlaid onto pollen images for measurements in micrometers.

The goal of this investigation was to see which insects visiting weed flowers also visit mango flowers. We also sought to see if mango visitors used resources from other flowers provided by the weeds. We found that insects collected feeding in mango flowers have more

weed species pollen on them than mango pollen, and that insects collected in weed flowers rarely carried mango pollen (Table 13).

Table 13 Pollen Carrying Insects in Mango and Weed Flowers				
Order	Insect Specimen	Treatment	Flower Collected From	# Different Pollen Types
HYMENOPTERA	Braconidae	weed	<i>Conoclinium coelestinum</i>	1
	Apidae Ceratina	weed	<i>Conoclinium coelestinum</i>	1
	Halictidae	weed	<i>Bidens alba</i> and mango	5
	Halictidae	weed	<i>Bidens alba</i>	1
	Halictidae	weed	<i>Bidens alba</i>	2
	Megachilidae	weed	<i>Bidens alba</i>	1
	Apidae <i>Apis mellifera</i>	control	Mango	1
	Apidae <i>Apis mellifera</i>	weed	<i>Bidens alba</i>	1
	Apidae <i>Apis mellifera</i>	weed	<i>Bidens alba</i>	1
DIPTERA	Muscidae	weed	Mango	2
	Sarcophagidae <i>Sarcophaga haemorrhoidalis</i>	weed	Mango	1
	Syrphidae <i>Ornidia obesa</i>	no-weed	Mango	2
	Syrphidae	weed	<i>Bidens alba</i> / <i>Conoclinium coelestinum</i>	2
	Syrphidae <i>Copestylum mexicanum</i>	weed	<i>Bidens alba</i> / <i>Conoclinium coelestinum</i>	1
	Calliphoridae	no-weed	Mango	1

	Calliphoridae	weed	<i>Bidens alba</i>	1
	Syrphidae <i>Copestylum mexicanum</i>	weed	Mango	1
	Syrphidae	weed	Mango	2



Figure 13 Left: Parasitoid Wasp Feeding in *Conoclinium coelestinum* with pollen, Middle: Syrphidae (*Copestylum mexicanum*) Feeding in *Bidens alba*/ *Conoclinium coelestinum*, Right: Syrphidae (*Copestylum mexicanum*) pollen on thorax

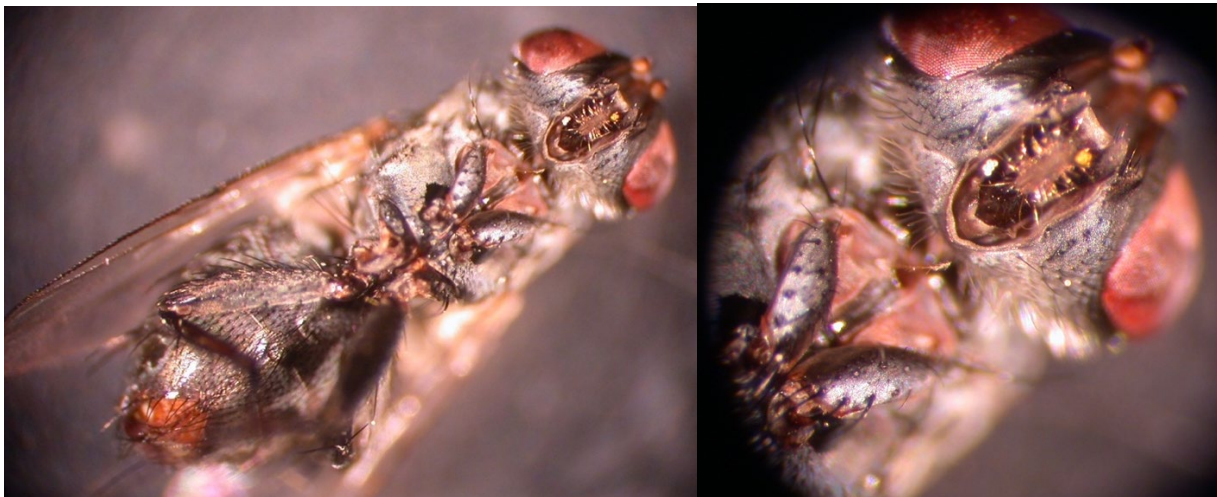


Figure 14 Left: *Sarcophaga haemorrhoidalis* Feeding in Mango Flower carrying multi-species pollen, Right: pollen on tongue

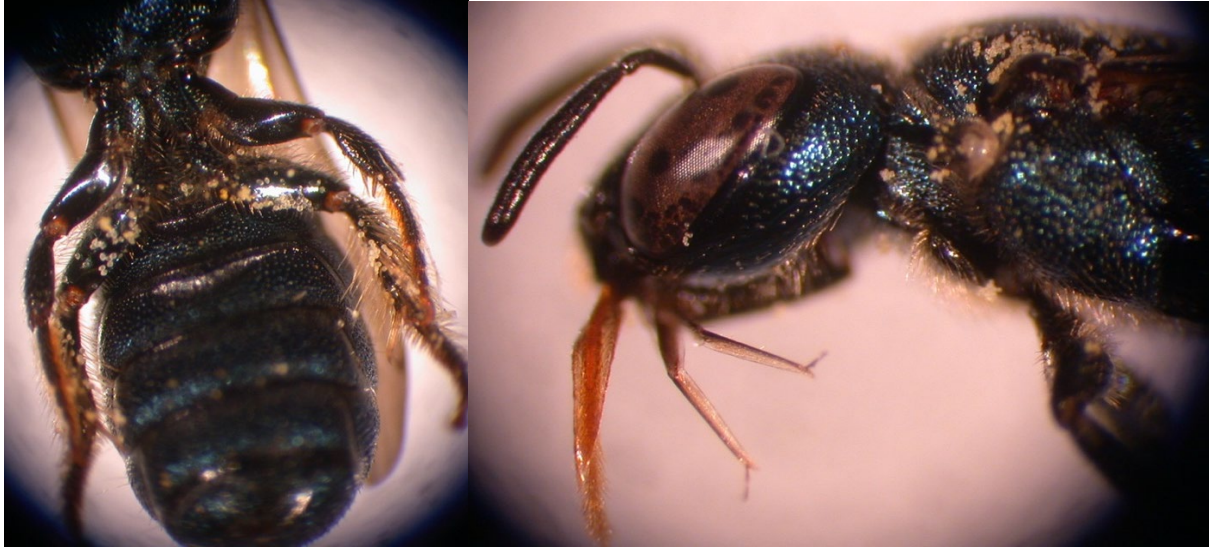


Figure 15 Left: Apidae (*Ceratina*) feeding in *Conoclinium coelestinum*, Right: Pollen on abdomen

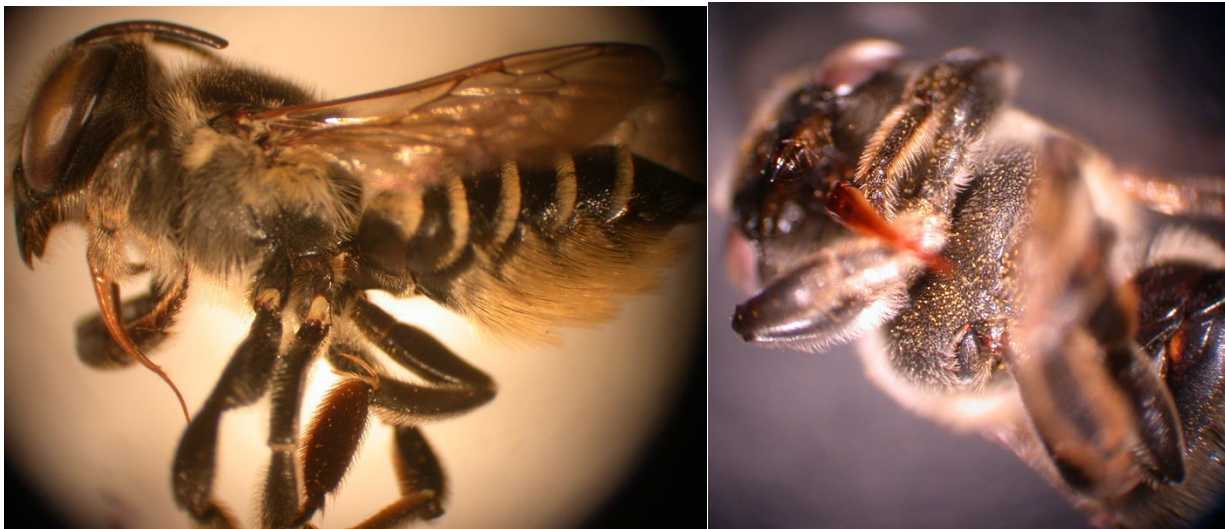


Figure 16 Left: Megachilidae feeding in *Bidens alba* flower, Right: Pollen on Body

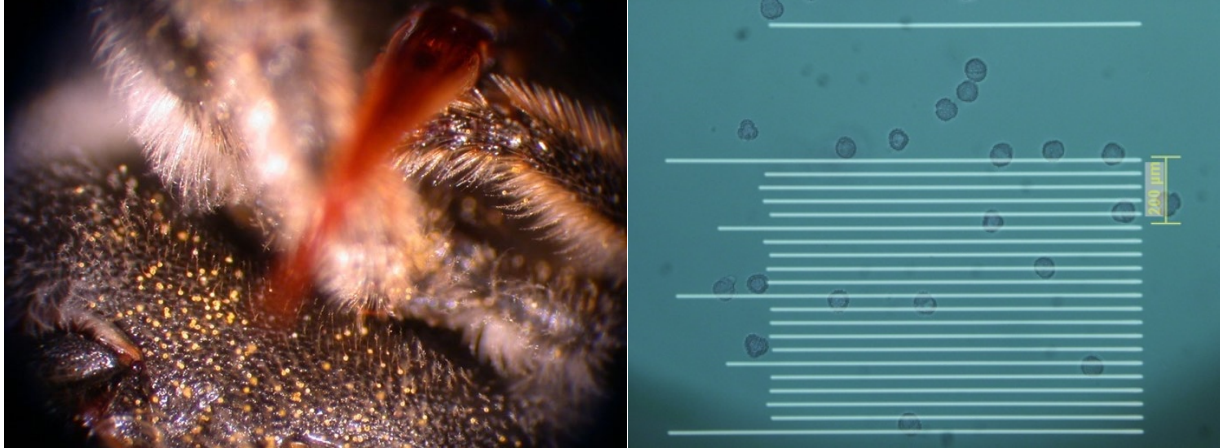


Figure 17 Honey bee *Apis Mellifera* feeding in Mango Flower, Right: Blue Mist Flower (*Conoclinium coelestinum*) Pollen

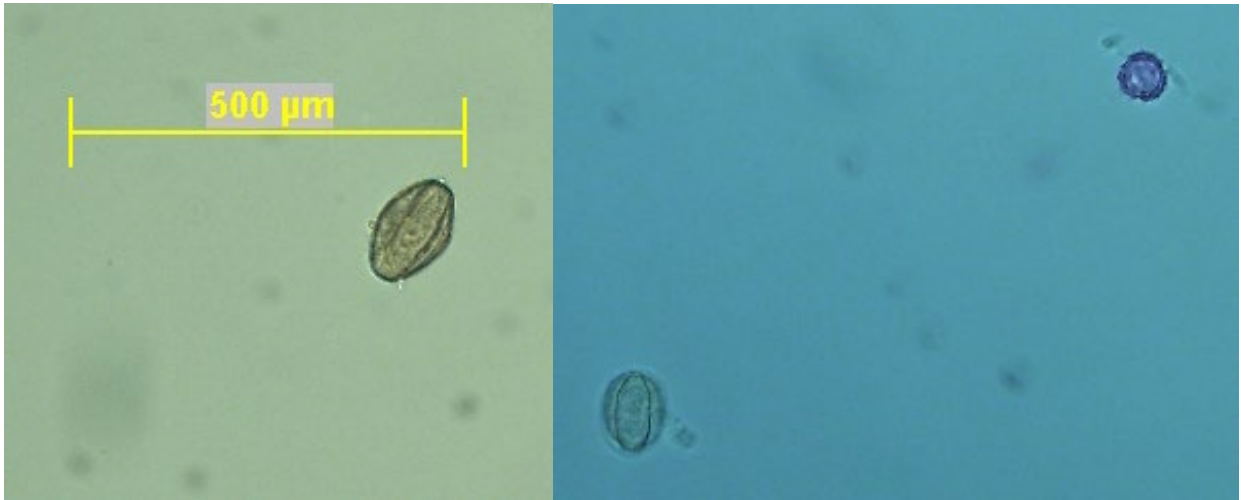


Figure 18 Left: Mango Pollen from Halictidae feeding in Mango Flower in Weed Treatment. Right: Pollen from Halictidae: Mango and *Bidens alba*.

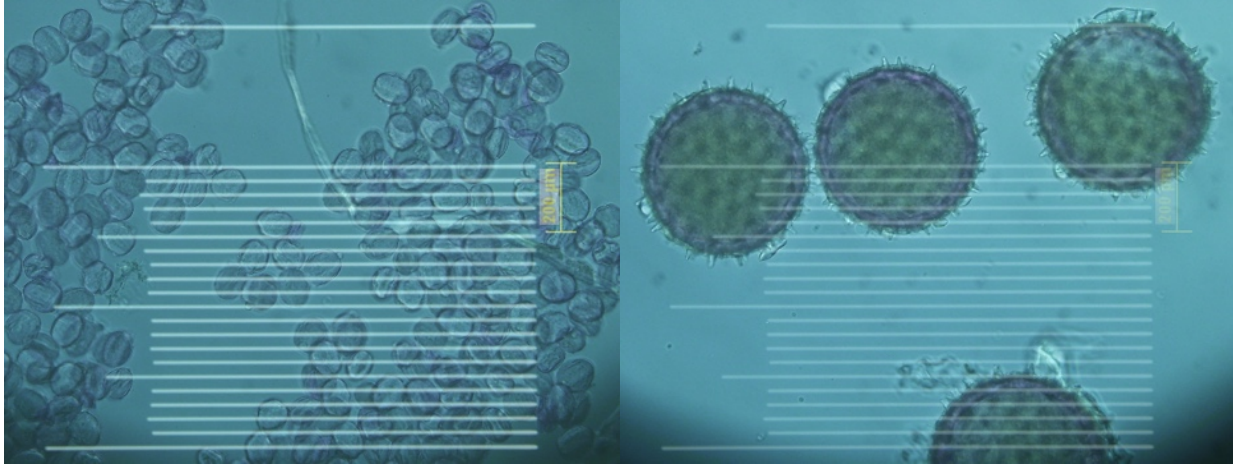


Figure 19 Left: *Crotalaria incana* Pollen. Right: Red Morning Glory (*Ipomoea hederifolia*) Pollen.

4.3 Rearing

In addition to insects that were recognizable as adults, we found many immature unknown stages (eggs, larvae, nymphs) which were reared. 228 insects were reared and identified: 35 Diptera, 42 Hymenoptera, 99 Lepidoptera, 25 Hemiptera, 5 Coleoptera, 4 Neuroptera, 6 Thysanoptera, 2 Collembola. Of the insects reared in the lab 57 had been parasitized: 4 Diptera, 1 Hymenoptera, 3 Lepidoptera, 23 Hemiptera, 1 Coleoptera, and 24 Aranae (spiders). Of the 228 reared specimens, 14 of the specimens were parasitized: 4 Diptera (including 3 Syrphidae), 1 Hymenoptera, 2 Lepidoptera (including 1 Geometridae), 4 Hemiptera (1 Aleyrodidae, 2 Aphididae, and 1 Coccidae), 1 Neuroptera, and 3 Aranae.



Figure 20 Scale (*Coccidae*) Parasitoids Reared from Mango Leaf Scale



Figure 21 Chalcididae Parasitoid Reared from Moth Caterpillar



Figure 22 Membracidae Nymph Reared



Figure 23 Hawkmoth (Sphingidae: *Erinnyis ello*) Caterpillar Reared from Euphorbiaceae.

4.4 Fruit Yield

Fruit count visual surveys by the two observers were averaged per tree, and the harvested count from the two harvest days added together per tree. There was significantly more fruit when weeds were present vs the no-weed treatments (Table 2). Both visual counts of fruit set and harvested counts were significantly higher in the weed treatment (Graph 11; Table 3: Harvested $F=181.317$, $df=1,57$, $p<0.0001$, Counted: $F=89.344$, $df=1,57$, $p<0.0001$). The mean harvested

fruit per tree in the weed treatment was 179 ± 65 , vs 38 ± 15 in the no-weed treatment. The mean counted fruit in the weed treatment was 236 ± 100 vs 48 ± 38 in the no-weed treatment. Each Keitt fruit weighs on average 1.5- 2.0 lbs (D. Lyons, pers. comm.). The average current price of a pound of Keitt mango across a spread of outlets, after deducting all marketing and packaging costs is 0.143/lb USD (Carvalho et al. 2012). As tree density was 30 trees in the weed treatment and implementation of weeds as insectary plants has no costs, it led to a gain of 908–1210 USD for all 30 trees, notwithstanding added gains from elimination of herbicidal applications. As farmers sell their product per lb and not per mango, weight is a good indicator of economic profit. Because production cost is mainly determined per hectare and is not influenced by volume of crop to be harvested, all increase in volume will have a positive impact on the economics of the crop (Carvalho et al. 2012).

Graph 11 Mean Number of Mango Fruit Harvested or Counted by Treatment \pm SE. Significant Difference Indicated Above Each Type: NS $P > 0.05$, * $P \leq 0.05$, ** $P \leq 0.01$, *** $P \leq 0.001$, **** $P \leq 0.0001$.

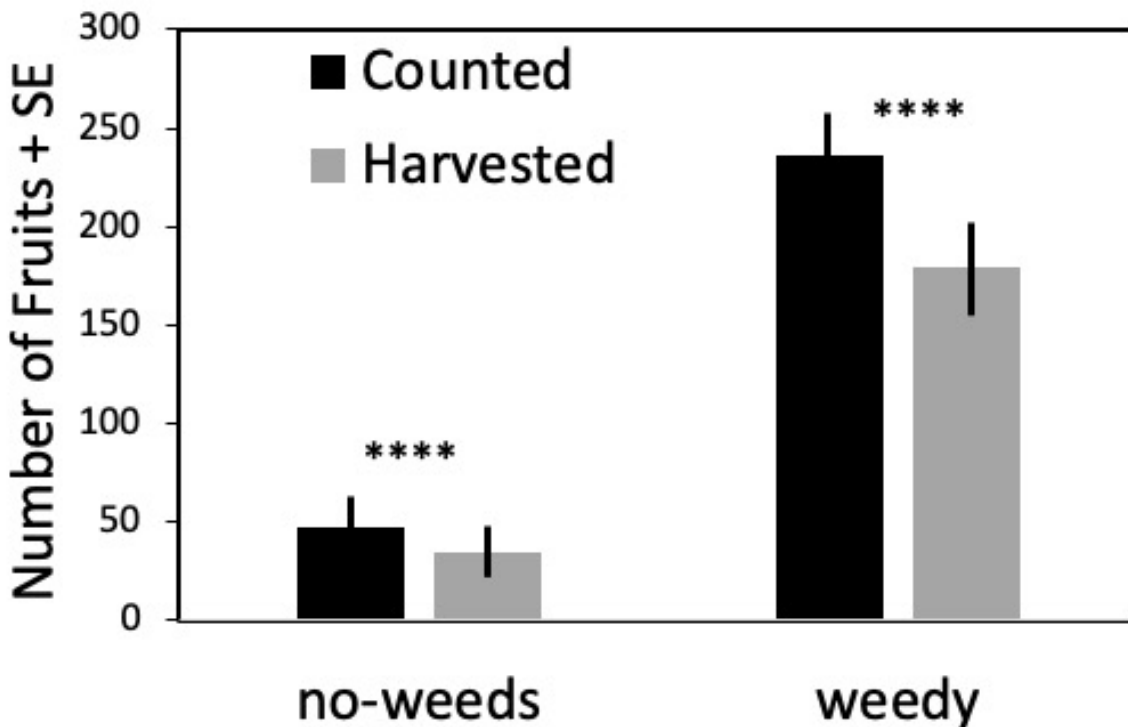




Figure 24 Mango “Keitt” Harvest

4.5 Soil Analysis

4.5.1 Total Carbon and Nitrogen

There was no significant difference between Carbon or Nitrogen in the weed and no-weed treatments for both sampling dates at the beginning and end of study (Graph 12; Table 14, T-Test). There was no statistically significant difference when comparing Carbon or Nitrogen in the no-weed treatment at the beginning and end of the study (Carbon: $T=-0.411$, $df=11$, $p=0.69$, Nitrogen: $T=-0.362$, $df=11$, $p=0.72$). There was no statistically significant difference when comparing Nitrogen in the weed treatment at the beginning and end of the study, however there was a significant difference in Carbon (Carbon: $T=-2.84$, $df=10$, $p=0.017$, Nitrogen: $T=-1.33$, $df=10$, $p=0.21$). Carbon increased significantly in the weed treatment from a mean of 11.13 to 12.77. This could potentially indicate that the presence of weeds benefits carbon availability in the soil by added organic matter, retention, biological cycling, and decreased runoff of nutrients.

Graph 12 Soil Carbon Mean by Treatment at the Beginning and End of Study.

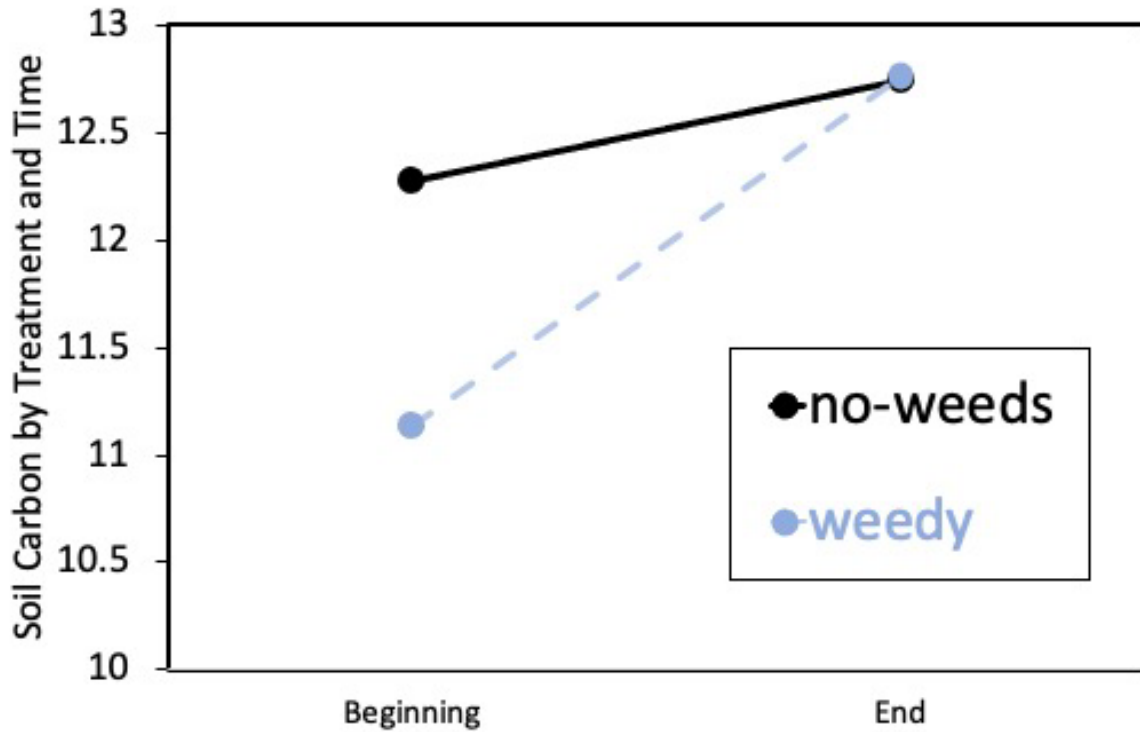


Table 14 Soil Carbon and Nitrogen Weedy or No-Weed Trees, T-Test.

<i>Begining</i>	<i>Mean</i>	<i>Std Deviation</i>	<i>N</i>	<i>Std Error</i>	<i>T</i>	<i>df</i>	<i>p-value</i>
<i>Nitrogen</i>	0.42	0.12	6	0.05	0.92	6.46	0.39
Weedy							
No-Weeds	0.55	0.32	6	0.13			
<i>End</i>	0.55	0.20	6	0.08	0.42	11	0.68
Weedy							
No-Weeds	0.62	0.94	7	0.13			
<i>Begining</i>	11.13	0.58	6	0.24	1.37	5.9	0.22
<i>Carbon</i>							
Weedy							
No-Weeds	12.27	1.95	6	0.79			

End	12.77	1.28	6	0.52	-0.023	11	0.98
<i>Carbon</i>							
Weedy							
No-Weeds	12.74	2.19	7	0.83			

4.5.2 Total Phosphorous

There was no significant difference in total Phosphorous in soil between treatments either at the beginning or end of the study, determined by a T-Test, one-way ANOVA, and nonparametric tests (beginning $T=0.68$, $df=10$, $p=0.51$; end $T=0.46$, $df=11$, $p=0.65$). Similarly, there was no difference in total Phosphorous when comparing all soil samples (beginning + end) between treatments (Table 15), or when combining treatments and comparing all soil samples across time (Table 16, $F=0.82$, $df=1, 23$, $p=0.375$). This could mean that the presence of weeds does not impact soil phosphorous either positively or negatively, as well as that this holds true for the continued presence of weeds.

Table 15 Total Phosphorous compared by treatment at the beginning and end of study, T-Test						
Beginning	<i>Mean</i>	<i>Std Error</i>	<i>N</i>	<i>t</i>	<i>df</i>	<i>p-value</i>
No-Weeds	3.42	0.88	6	0.682	1,10	0.511
Weedy	2.69	0.60	6			
End of Study	3.99	0.74	7	0.459	1, 11	0.655
No-Weeds						
Weedy	3.46	0.90	6			

Table 16 One-way Anova Comparing Total Soil Phosphorous Weeds vs. No-Weeds, Beginning and End Samples Combined					
	Sum of Squares	df	Mean Square	F	p-value
Between Groups	2.638	1	2.638	0.721	0.405
Within Groups	84.177	23	3.660		
Total	86.815	24			

4.5.3 Soil pH

There was no significant difference in soil pH between treatments (Table 17, Paired samples T-test and Univariate analysis of variance $F=0.001$, $df=1,25$, $p=0.975$). However, there was a significant effect between treatment (weeds) and time (before and after samples). For both treatments, pH became more neutral over time (Graph 11, $F=23.71$, $df=1, 25$, $p<0.0001$; Univariate Analysis of Variance). This could mean that as herbaceous vegetation grows and decomposes, adding organic matter to the soil, it becomes more acidic. The ideal pH range ideal for mango cultivation is between 4.5 and 7.5 pH, which is neutral to acidic soil, therefore the greater reduction of pH to 7.7 for the weed treatment could mean weeds can help mango trees achieve an ideal soil pH, and that the longer they are present could increase this effect.

Graph 13 Mean Soil pH by Treatment at the Beginning and End of Study.

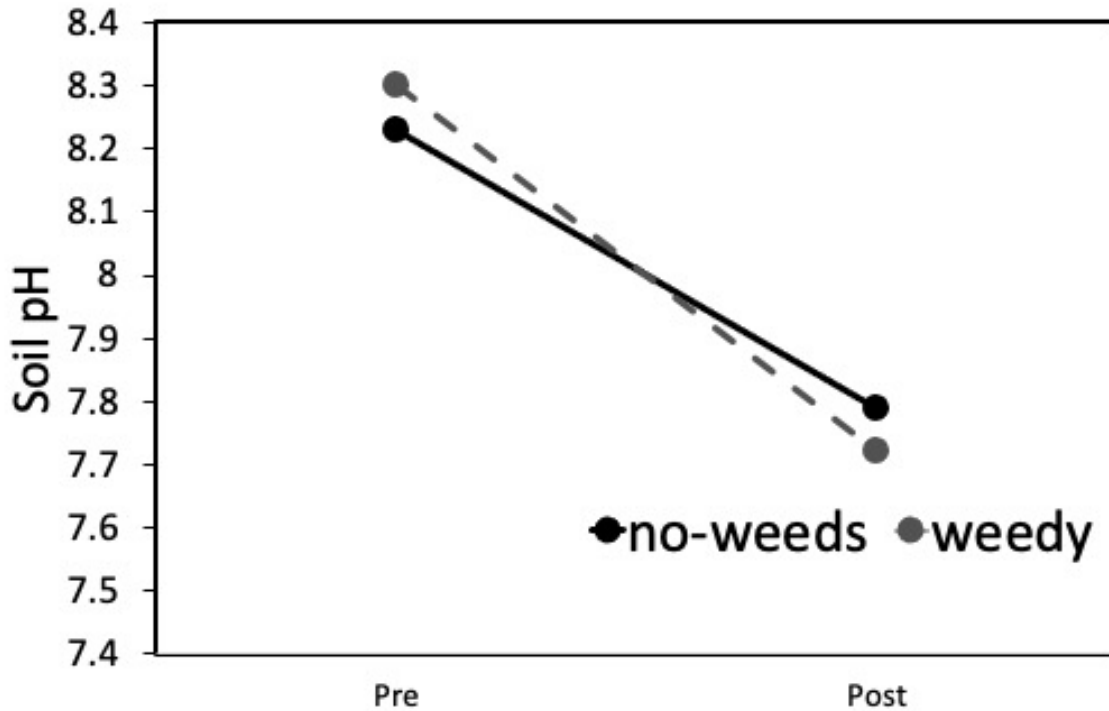


Table 17 Soil pH Between Treatments and Over Time, Univariate ANOVA

Treatment	Time	Mean	Std Error
Weed	Pre	8.3	0.111
	Post	7.72	0.096
No-Weeds	Pre	8.23	0.111
	Post	7.79	0.103

4.6 Chlorophyll Analysis

There is a significant interaction between treatment and leaf age (Table 18, $F=3.7$, $df=1, 2$, $p=0.03$) determined by a Univariate ANOVA. Mango trees in the no-weed treatment had higher SPAD readings, indicating greater chlorophyll content, most dramatically in the old and new leaves (Graph 14). The mature green leaves in both treatments contained similar amounts of chlorophyll. This could potentially mean that as the leaves are new or old, weeds influence leaf chlorophyll content. However, when the leaves are green, healthy, and most effective at photosynthesis, there is little effect of weeds on chlorophyll content.

Graph 14 Mean SPAD Chlorophyll Readings for Mango Leaves, Weeds vs. No Weeds, for New Leaves, Green Leaves, and Old Leaves.

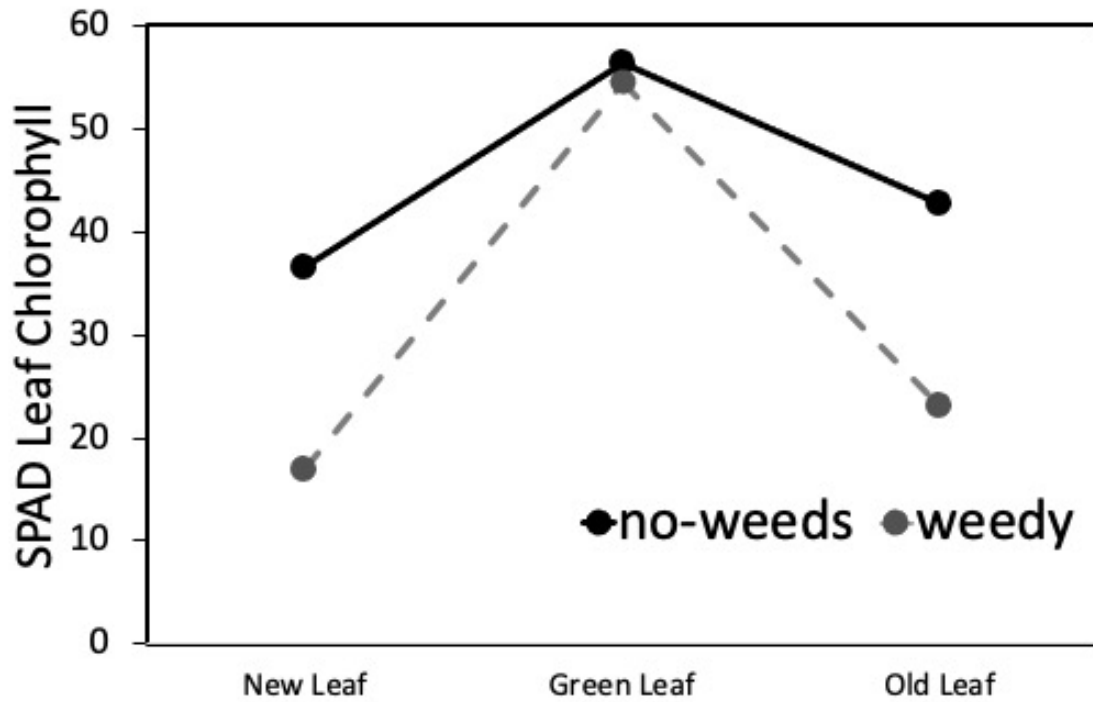


Table 18 SPAD Chlorophyll Readings for Mango Leaves, Weeds vs. No Weeds. Univariate ANOVA.

Weedy	Mean	Std Dev	Std Error	N
<i>New Leaf</i>	16.94	10.28	2.42	18
<i>Green Leaf</i>	54.44	8.05	1.89	18
<i>Old Leaf</i>	23.06	12.96	3.06	18
No-Weeds	36.64	23.75	5.06	22
<i>New Leaf</i>				
<i>Green Leaf</i>	56.44	12.26	3.06	16
<i>Old Leaf</i>	42.75	18.42	4.6	16

5. Discussion

The overall results indicate the successful implementation of weeds as insectary plants to increase beneficial insects (pollinators and parasitoids) and increase fruit yield. The added

diversity of 75 different weed species (many of which are native flowering herbaceous plants), increased the floral diversity for beneficial insects without negatively competing with the crop. Besides grasses, the majority of weeds were small native wildflowers, enhancing the microclimate for insects, birds, and other wildlife, without taking water or nutrients from the established mature mango trees in this study, and even adding Carbon and improving soil pH. Mango is one of the most cultivated tropical fruits worldwide and one of few drought-tolerant plants with minimal nutrition supplementation needs (Lipan et al. 2021). It's deep taproot and added fruit yield with maturity of the tree allow reduced negative impacts from intercropped herbaceous vegetation, and reliance on pollination means net gains from added floral diversity of weeds and pollinating insects.

Additionally, elimination of herbicidal applications in the weed treatment reduced the farmers production costs, chemical use in the adjacent suburban neighborhood, and potential runoff and negative effects of chemicals on surrounding ecosystems. It also led to an average gain of 908–1210 USD from added fruit yield in the weed treatment. However, given the constraints and application of the weed treatment within one row of mangos and not spread across the farm, potential confounding abiotic factors such as sunlight/adjacent landscapes could have also impacted fruit yield. Further studies should clarify how within and surrounding field weedy vegetation affects insect biodiversity and mango fruit yield, as well as soil abiotic and biotic functions and nutrition. Using other sources of insectary plants, such as flowers like sweet alyssum (Hogg et al., 2011), or native herbs such as the Bahama Senna (Salas, 2016) could be an alternative for farmers to garner beneficial insects. Additionally, this farm is surrounded by an increasingly urbanized landscape, and the relationship among urbanization, pollinator resource distribution, pollinator abundance, and pollination service provision are uncertain (Cusser et al.

2021). Further, pollen supplementation experiments comparing cross and self-pollination would help clarify the importance of cross-pollination in mango and insect's importance for fruit yield.

There was also an increased presence of rats and snakes in the weed treatment compared to the rest of the farm, as well as flies feeding in rat carrion. As pollinators require distinct and diverse resources throughout their life cycle, including larval habitat and adult food resources, the presence of rat carrion has been found to facilitate pollination services to plants by bolstering the pollinator community across an urban gradient. Rat carrion has been found to increase pollinator abundance by more than two-fold and plants received 11.2% greater pollination service across landscapes and higher viable seed set, especially in densely urban landscapes (Cusser et al. 2021). Mutualistic species with complex life histories can provide conduits between various levels of ecosystem processes. As blowflies/carrion flies (Calliphoridae) are one of the best pollinators of mango, weedy habitats can potentially serve dual purposes in supplying adult needs of floral resources as well as their larval needs of carrion by supporting rat populations.

6. Conclusion

Weeds can provide ground cover that can help to reduce soil erosion and N loss (Wortman 2016), increase soil Carbon much like a winter cover crop, and support insect natural enemies (Diehl et al. 2012), pollinators (Gibson et al. 2006, Bretagnolle and Gaba 2015, Blaix et al. 2018), birds (Thomas et al. 2001), and increase biodiversity at the field and landscape level (Marshall et al. 2003, Franke et al. 2009, Storkey et al. 2014). Conserving the biodiversity of plants and insects needs more focus now than ever, with increasing threats to farms, especially monocultures, in the face of climate change (Altieri et al., 2015). More biodiverse farms, hosting varied plants and insects, are more resilient and less vulnerable to stressors (Altieri et al., 2015).

Using native weeds, which have the potential to act as insectary plants when growing companion plants isn't possible and increasingly likely in changing climates, to add to the variety of diets for beneficial insects increases the conservation of plants and their plant-pollinator networks. Plant-feeding insects are an important cause of crop yield losses worldwide (Oerke 2006, Dhaliwal et al. 2015, Savary et al. 2019) and these losses are predicted to increase in response to climate change (Capinera 2005, Deutsch et al. 2018). Weeds, therefore, can provide resources to maintain balanced insect-ecological dynamics, and allow for less chemical use of pesticides and herbicides, and more environmentally sound agriculture across South Florida.

Understanding the ecological links among herbicides, pesticides, weeds, crops, and insects, can inform agriculturalists about potential benefits of biological complexity in agroecosystems. This will allow increased understanding of the confounding ecotoxicological effects of herbicides that impact insects worldwide. This study illuminates the interactions of invasive and herbicide resistant weeds with insects in a sub-tropical agricultural context, and any potential threats they pose to crops as insect hosts. This study promotes sustainable agriculture of an economically valuable exotic fruit that grows well in the United States and clarifies for growers how differing management practices can affect insect abundance and diversity, and in turn, fruit production.

Studies on weeds are needed for specific crop species, regions where they are grown, and varying combinations of weed species, to learn how these variables affect insect ecological dynamics. These, in turn, must be quantified economically. The measuring of fruit yield in this study quantifies the benefits of increased pollination and reduced pest damage and gained insight on the feasibility of implementing positive weed management across various crop monocultures, especially tropical fruit trees.

Farmers are important wells of knowledge, who deal hands-on with weeds and consider them a time and effort consuming blight, diminishing their profit margin. This study reduced the cost and time of eradicating every weed, and promotes the production of mango, a widely cultivated and highly valued crop of Homestead, Fl, worth \$2.1 million as of 2014 (Crane and Mossier, 2014). This research also supports the practice of some mango farmers who leave selected beneficial weeds, such as *Bidens spp*, around their trees (Heinrichs, 1988; Needham, 1948). Increasing societal worry regarding the health risks associated with herbicides are important (Stokstad, 2019); this research may encourage more farmers to avoid the use of herbicides in food crop cultivation. Weeds also benefit crop soil by fixing Nitrogen and bringing nutrients up into the topsoil (Gilmore, 2019), and this study provides evidence of their ability to increase soil Carbon and reduce soil pH. Additionally, weeds slow down the soil warming/drying process, which help farmers buffer against the risk of growing tropical fruit in increasing temperatures (Gilmore, 2019).

Overall, the use of weeds in increasing beneficial insects has shown promise (Araj et al. 2019; Provost and Pedneault 2016; Kremen et al. 2002; Pickett and Bugg 1998). However, a caveat to their use in agriculture is how to handle invasive weedy species. Selectively removing the noxious FLEPPC Category I invasive plants is recommended to slow their spread in South Florida, a major problem, and allow native flowering weeds to take over. While increasing plant diversity and resources to increase pollination may not always prove a success, studies on specific crops, regions, and weed species, can allow us to learn how these variables affect plant-insect ecological dynamics. Furthermore, with increased pollination and crop yield, economic valuation can allow us to gain insight on the feasibility of implementing this practice in agriculture.

This research has shown that studies are needed to understand not only the behavior of insects in floristically diverse vs. depauperate landscapes, but also how anthropogenic manipulation can affect ecological interactions among crops and pollinators. In attempting to increase pollinators by promoting the presence of weeds, it is inevitable that interactions between weeds and other species can occur, and should be investigated in various crops, as well as monitored in adjacent natural systems. The best management practices moving forward are to quantify the economic ramifications accompanying the increased habitat complexity provided by weeds, and in cases where there are benefits to this approach, to take advantage of the free services they may provide.

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