Final Report for Ceres Trust

Project Title: Evaluating Biological Control of Brassica Pests in Urban Land Repurposed for Farming

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Abstract:

Urban farms bring economic and ecological productivity to underdeveloped spaces in Midwestern (USA) cities. As planted row and vegetable crops are added, there is increased susceptibility to plant damage by herbivorous pests. We surveyed urban farm managers on perceived risks of crop damage from insect pests. Based on the survey results, we investigated the effects of surrounding landscape and production type on natural enemy and pest populations. We investigated natural enemy and pest populations in urban agriculture and the potential of insect predators and parasitoids to control cabbageworms and aphids in community gardens and urban farms. In a laboratory assay, we measured the efficacy of natural enemies at suppressing cabbage looper. In another experiment, we investigated the impact of supplemental floral resources on natural enemies and brassica production. Urban agriculture contains a diverse community of natural enemies, dominated by parasitoid wasps and several predator groups. Cabbageworms and aphids were present at low, but similar, densities at community and urban farms. Brassica defoliation was less than 20%, on average, across sites. The lab study identified lady beetles as the most effective predator of cabbageworm eggs. In gardens, we found greater mortality in cabbage loooper eggs compared to larvae. Natural enemy abundance did not affect mortality rates, and variation in cabbage looper mortality was poorly explained by within-garden or landscape factors. The diverse natural enemy community in urban farms provides a positive outlook for growers relying on biological control as a tool in pest management. Our results confirm the roles of generalist predators in suppressing pest populations in urban agriculture.

Introduction:

Urban consumers are increasingly interested in local, organic food production (Grewal and Grewal 2012). This has coincided with a conversion of underutilized vacant space to food production in Midwestern USA cities. Chicago, IL, has greater than 4,000 urban agriculture sites (Taylor and Lovell 2012), although only 10% are urban farms or community gardens. Starting an urban farm requires investments to prepare the land for food production and to protect crops from pests and pathogens, yet few data-driven resources are available to the scale of the urban farmer.

The addition of row and vegetable crops increases the risk for damage by herbivorous pests that locate new forage. In particular, plants in the cucurbit and brassica families are susceptible to damage from insect pests (Appendix 1). Brassica are primarily damaged by three caterpillar pests - imported cabbageworm (*Pieris rapae*), cabbage looper (*Trichoplusia ni*), and diamondback moth (*Plutella xylostella*) – commonly called cabbage worms. Several species of specialist parasitic wasps (Godin and Boivin 1998) and insect predators (Schmaedick and Shelton 1999) attack cabbage worms. However, it is unknown if urban areas host sufficient natural enemy populations for effective cabbageworm control. To enhance populations of insect predators and parasitoid wasps within larger, rural farms, field edges can be supplemented with flowers (White et al. 1995, Blaauw et al. 2012). However, the small size of many urban farms reduces the amount of space that can be allocated to non-crop plants. As a result, the local area surrounding urban farms could influence natural enemies and the rate of biological control within urban food production.

The smaller scale of urban agriculture may affect the dynamics of natural enemy and pest populations. Farms offer a relatively stable source of plant resources and insect hosts for natural enemies relative to other habitats in urban neighborhoods. As a result, biocontrol and natural enemy populations could be greater at sites with increasing agricultural intensity. There is a need to study patterns of natural enemies and crop pests within cities. We investigate if natural enemies and biocontrol differ between two scales of urban agriculture, urban farms and community gardens. We classify urban farms as sites managed as a single unit for commercial production. Community gardens have multiple plots that are managed independently, and produce is often grown for personal consumption or donation to low-income individuals. Extensive crop damage by insect pests would devastate small-scale urban growers but would have an acute economic effect on urban farms. Additional information about natural enemies and biocontrol would assist growers in developing pest management strategies in brassica

Objectives:

1) Assess cabbage worm populations, natural enemy populations, and the rate of biocontrol at different production scales in urban agriculture

2) Evaluate if natural enemy groups vary in their effectiveness at controlling cabbage looper

3) Investigate drivers of natural enemy populations and biological control of cabbage looper

4) Identify if the addition of native resources in brassica rows reduces defoliation

Objective 1:

Methods

We included 17 food production sites from two scales of agricultural production: community gardens (N=9) and urban farms (N=8) in Chicago, IL. We focused on two important groups of brassica pests. The first group, cabbageworms, includes a complex of 3 specialist herbivore species: cabbage looper (*Trichoplusa ni*), diamondback moth (*Plutella xylostella*), and imported cabbageworm (*Pieris rapae*). The second group, sap-feeding aphids, primarily includes green peach aphid (*Myzus persicae*) and cabbage aphid (*Brevicoryne brassica*). At each site, we randomly selected five *Brassica oleracea* plants (i.e. cabbage, broccoli, kale) and inspected five leaves on each plant for the presence of aphids and cabbageworm eggs, larvae, and pupae. In addition to counting pests, we estimated defoliation on the five selected plants on a 0-100% scale at 10% intervals. As we walked through crop rows, we also recorded the presence of adult cabbageworms in-flight. Cabbageworms were identified to the species level but aphids were not. We sampled for cabbageworms and aphids at six intervals between 16 June and 26 August 2014, counting pests and estimating defoliation every other week.

We set up four 18x14 cm yellow sticky cards (Alpha Scents: West Linn, OR) within brassica crops at each farm or garden. Cards remained in the field for two-week intervals and were replaced when we visited sites to record pests. In total, we had 5 two-week intervals of natural enemy sampling. After each sampling period, we brought yellow sticky cards to the lab to identify predators and parasitoids on the card. We identified parasitoid wasps to the family level. Since parasitoids vary in their selection of host insects, we also noted the 18 families of parasitoid wasps known to parasitize the egg, larval, or pupal stages of Lepidoptera, the insect Order that includes cabbage worms (Appendix 2). We also noted parasitoids in the Aphidiinae subfamily of Braconidae, which are specialist parasitoids of several aphid species. From here on, we refer to these 19 families as 'specialist parasitoids'. Additionally, we recorded the abundance of common insect predators that may attack aphids and the immature life stages of cabbageworms.

We used Generalized Linear Models (GLMs) to test for the effects of scale of agriculture, sample date, and natural enemies on insect pests. Our response variables included abundance of cabbageworm larvae, adults, aphids, and defoliation. We used poisson distribution for pests and binomial distribution for defoliation. For post-hoc comparisons, we used generalized linear hypothesis tests (glht) with Tukey's pairwise comparisons in the multcomp package (Hothorn et al. 2008). We hypothesized that pest abundance would be greater at farms, since they offer more brassica resources.

Results

On average, 4.1±0.6 eggs, 1.7±0.3 larvae, and 0.3±0.1 pupae of cabbageworms were found on five brassica plants per site. Over 70% of cabbage worms on brassica were imported cabbage worm (ICW). Cabbage looper was the next most common pest. First generation ICW peaked in late June, and second generation ICW peaked in mid-August (Fig. 1). Natural enemy populations were highest in August during the occurrence of second generation cabbage worms (Fig. 2). Damage to brassica plants averaged 10%, with the highest defoliation in mid-late June, which coincided with first generation ICW and the period with the fewest natural enemies. GLMs revealed a significant effects of specialist parasitoid wasps and total parasitoid wasp abundance on cabbageworm larvae and aphids (Table 1). Parasitoid wasps were the most common natural enemy followed by long-legged flies (Fig. 3). Only specialist parasitoid wasps of cabbageworms were significantly more abundant in farms than community gardens (F=19.19, P<0.001)

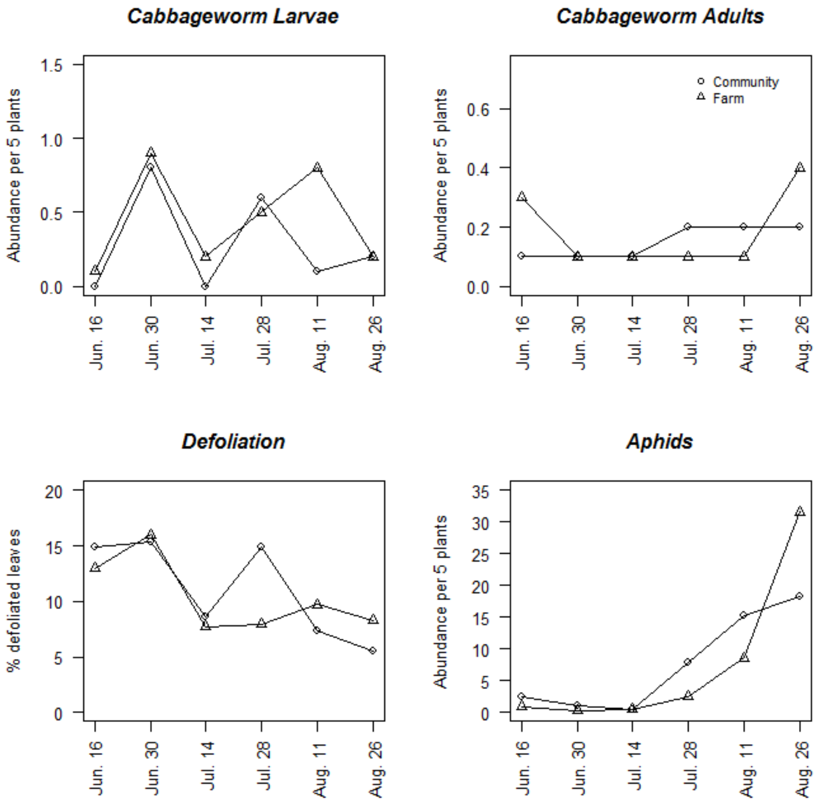

Figure 1. Mean cabbageworm larva, adult, and aphid abundance, and defoliation, across the six sample dates.

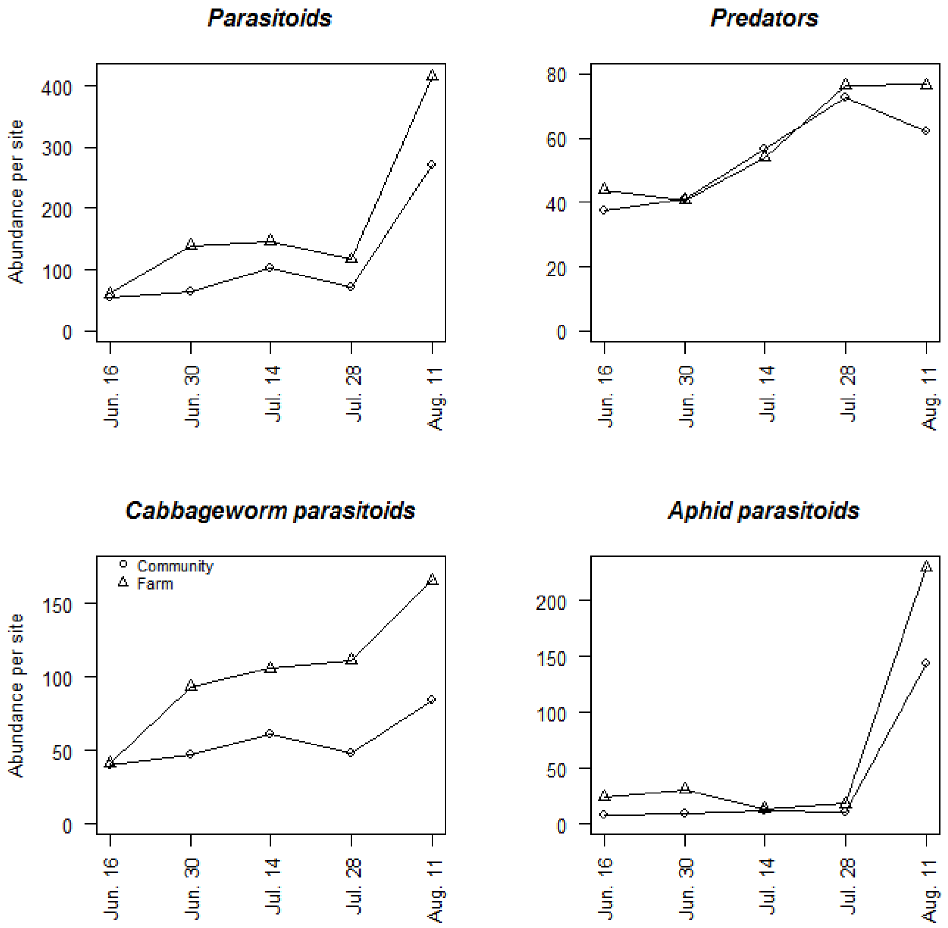

Figure 2. Mean natural enemy abundance on yellow sticky cards at the three scales of agriculture. Sample date is the start of 2-week period when sticky cards were left in each garden. Cabbageworm parasitoids include 18 families known to attack Lepidoptera. Aphid parasitoids are specialists from the Aphidiinae subfamily.

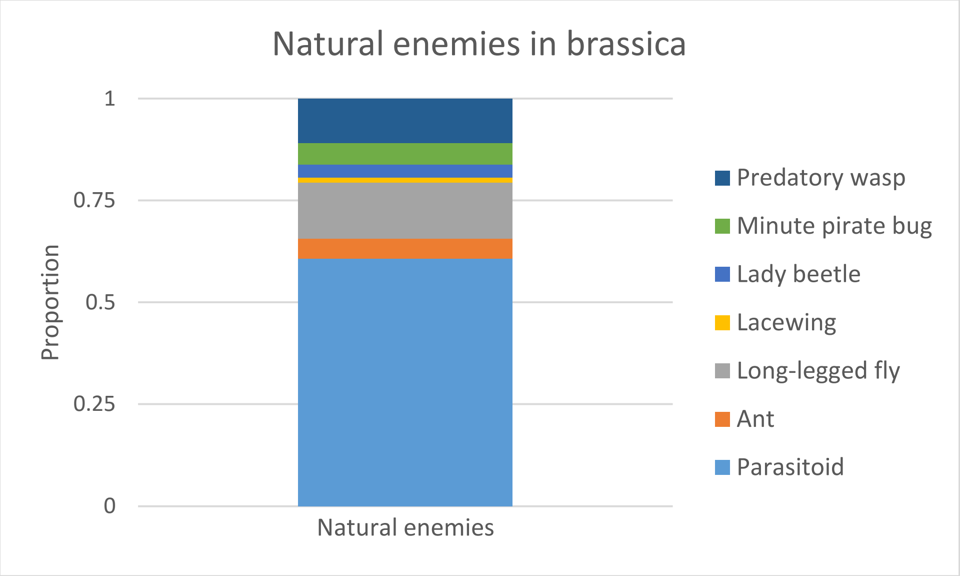

Figure 3. Stacked barplot of natural enemies collected on yellow sticky cards in brassica. Family-level identifications of parasitoids is present in Appendix 2.

Table 1. Results from GLM to determine effects of scale of agriculture, date, and natural enemies on brassica pests and defoliation. Each cell contains the F value for the predictor variable in the column heading. Significant effects have the following notation: #P≤0.10, *P≤0.05, **P≤0.01. Pseudo R^2 [1 – (Residual/Null deviance)] is presented for the reduced model that only contains the significant terms.

Response variable	Scale	Date	Scale x Date	Parasitoids	Specialist parasitoids	Predators	Pseudo R^2
CW Larva	1.50	2.94*	0.57	3.31#	7.71**	0.53	0.36
CW Adult	0.69	1.11	2.13	0.54	1.16	0.17	0
Aphid	0.98	0.96	0.26	9.74**	3.45#	0.06	0.24
Defoliation	0.38	1.50	0.29	0.79	0.36	0.10	0

Objective 2:

Methods

We investigated the effectiveness of prey suppression by several predator taxa recorded in urban agriculture. Due to difficulty in collecting certain taxa, we had an uneven number of replicates for each natural enemy. Larval consumption assays had 14-30 replicates per natural enemy. Egg consumption assays had at least 20 replicates except for ground beetles (Carabidae) (N=6), harvestman, (N=13), and lady beetle larvae (N=10). We collected *Coccinella septumpunctata* adults and larvae, ground beetles, harvestmen, ants (Formicidae), and spiders (Aranae) from the field. We used Linyphiidae and Lycosidae spiders, as neither family differed in egg consumption rates (F=0.60, P=0.45). We purchased minute pirate bug (*Orius insidious*), and lacewings (Chrysopidae) from an insectary. We also supplemented field-collected lady beetles with purchased *Harmonia axyridis* adults to reach a sufficient number of replicates in larval consumption assays.

We set up assays in which a natural enemy, starved for 24 hours, was presented with an egg mass of 23-30 cabbage looper (CL) eggs in a petri dish with a moistened cotton roll. We used a similar design, presenting a third instar CL to a single natural enemy item. An untreated control group with a single egg mass or a larva was set up to confirm that CL damage was caused by insects rather than handling damage or natural mortality. After 24 hours, we recorded the proportion of damaged or entirely consumed eggs and the number of damaged larvae. To evaluate egg consumption rates by natural enemy taxa, we used ANOVA with percentage of eggs consumed or damaged as biocontrol rate. Biocontrol rate was arcsin transformed in all analyses. We analyzed the frequency of larvae predation by natural enemies using a chi square test with a Bonferroni correction for post-hoc comparisons.

Results

Natural enemies varied in the proportion of cabbage looper eggs (F=20.8, P<0.01; Fig. 4) and larvae consumed (Chi=37.09, P<0.01; Fig. 5). Lady beetle adults and larvae were the most efficient egg predators. Lady beetles, pirate bugs, and spiders attacked cabbage looper larvae most frequently.

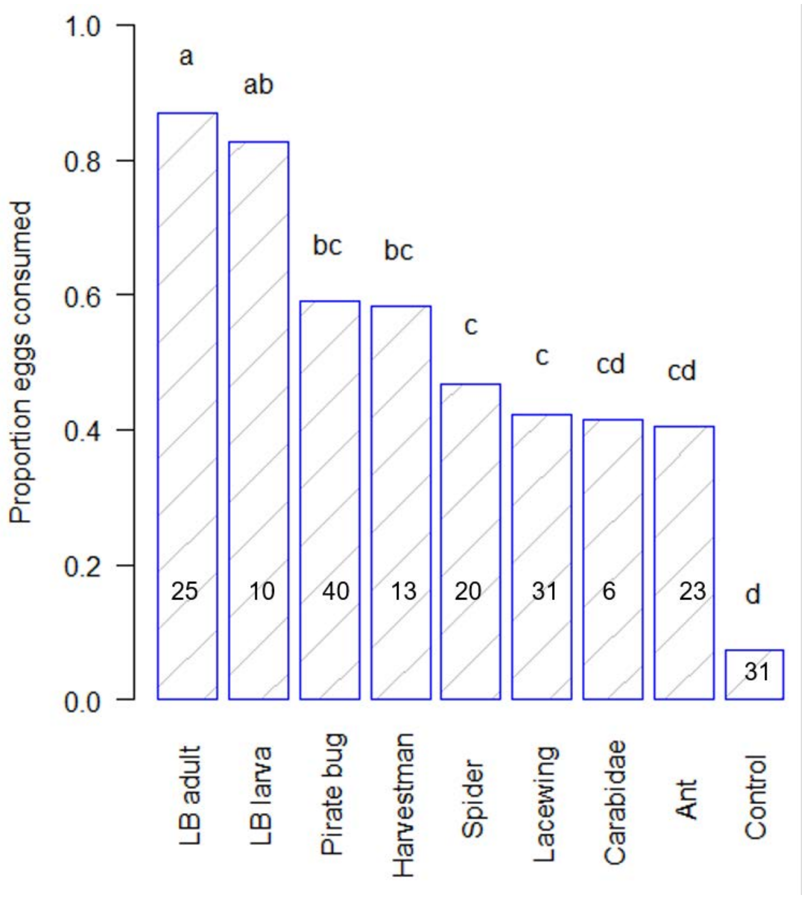

Figure 4. Mean egg consumption by natural enemies in assays measuring predator effectiveness. Number of replicates for each predator is listed within the bar. Letters above bars indicate significant differences as calculated using Tukey's HSD. LB= Lady beetle

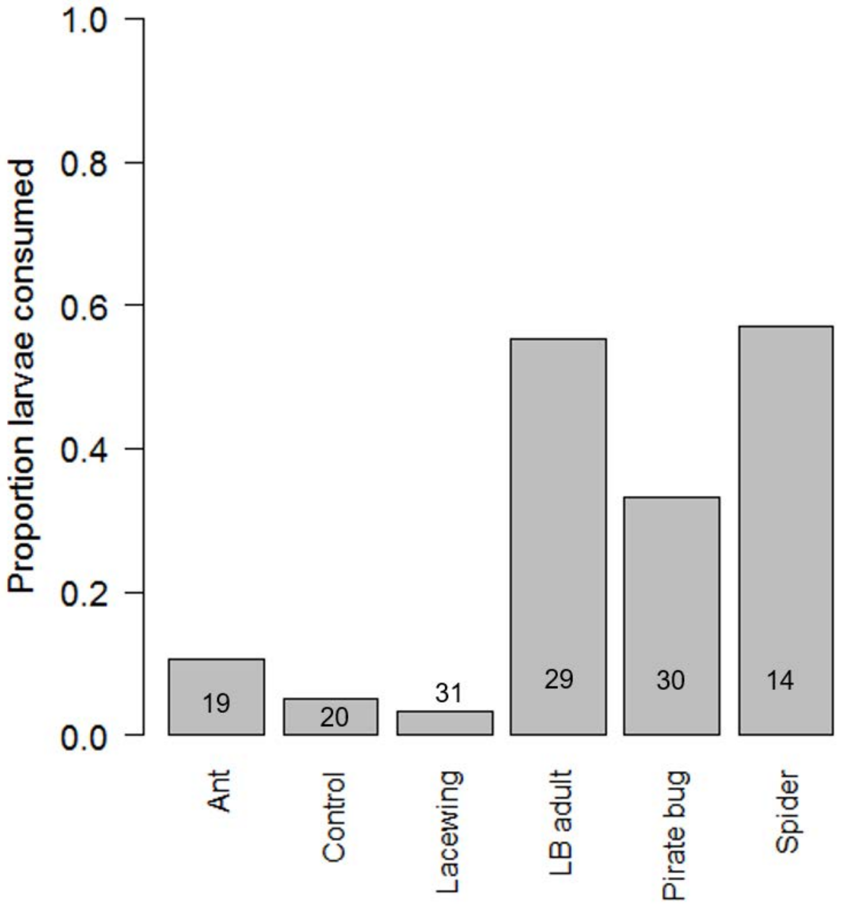

Figure 5. Mean larval consumption by natural enemies in assays measuring predator effectiveness. Number of replicates for each predator is listed within or above the bar.

Objective 3:

To evaluate biocontrol rates in urban agriculture, we obtained cabbage looper from an insectary (Frontier Scientific) and used them as sentinel items. Egg masses were oviposited onto paper towel and standardized into 23-28 eggs by cutting the paper towel into approximately 2 x 2 cm sections. We pinned 3 egg masses, singly, beneath 0.5 m elevated pieces of green cardstock and left the cardstock in rows of cultivated brassica at each site. An egg mass covered by a petri dish that excluded natural enemies served as a control. We also deployed 3 larvae, covered with a 2 mm mesh bag to prevent escape, beneath each platform. In addition, a larva covered by a petri dish, which excluded predators, was placed beneath a platform at each site as a control. Egg suppression was measured at each site on Aug. 1 and Aug. 14, 2014; larvae suppression was measured on July 13, and Aug. 14. Sentinel items remained in the field for 72 hours, after which remaining eggs were counted, and each larva was inspected for evidence of predation. All undamaged eggs and larvae were returned to the lab and preserved in a 26º C growth chamber for 1 week to check for the emergence of parasitoids. The number of damaged items was pooled over both sampling dates, and we calculated separate mortality/disappearance indices for eggs and larvae. This index was calculated at each site as the number of sentinel items damaged by natural enemies reduced by the number of damaged sentinel items in the control assays protected from predators.

To confirm that sentinel items were damaged by insect natural enemies, we set up time lapse cameras (Brinno TLC200 Pro) for 24 hr periods at a subset of sites and a total of 168 hours of footage. We investigated the effects of peedator and parasitoid abundance, taken from yellow sticky cards, within-garden (local factors) and landscape factors (tree and grass cover) taken from a Quikbird satellite image, on the rate of biological control using model selection in the MuMIn package in R 3.1. We constrained the results of model selection by only considering a subset of models with ΔAICc < 2 as the most predictive models of cabbage looper mortality.

Results:

On average (± SE), 54.5 ± 2.8% of eggs and 25 ± 4.0% of larvae were damaged or attacked at each site. From time lapse cameras and site observations, we recorded ants, spiders (Lycosidae), lady beetle adults and larvae from two species (*Coleomegilla maculata, C. septumpunctata)*, and parasitic wasps were observed foraging on or attacking eggs and larvae. Several specialist parasitoids attack imported cabbageworm (ICW) and CL, but we only observed two instances of ICW larvae parasitized by a specialist wasp, *Cotesia glomerata* (Fig. 6). No parasitoids emerged from reared sentinel eggs or larvae. Egg biocontrol rate did not differ by scale of agriculture (F=1.65, P=0.21). Similarly, larval biocontrol rate did not differ by scale of agriculture (F=0.37, P=0.54) but was significantly greater at the first sample date (F=8.62, P<0.01).

Model selection did not identify any significant effect of predator or parasitoid abundance on egg or larval mortality. The top performing model for egg biocontrol only included a marginally negative effect of defoliation (Table 2), while the top performing model for larval biocontrol included a weak negative effect of grass cover (Table 3).

Table 2. Summary of top-performing models that explain egg suppression and have ΔAICc < 2. Higher Akaike weights indicate increased model fit.

Model	Coefficients (Beta)	AICc	Akaike weight	Maximum likelihood Pseudo R^2
Defoliation	B1= -0.02	277.1	0.305	0.11
Defoliation + Percent brassica	B1= -0.03 B2= 0.37	277.5	0.255	0.18
Intercept		278.0	0.193	
Defoliation + Percent brassica + Trees	B1= -0.02 B2= 0.44 B3= 0.01	278.7	0.134	0.23
Parasitoid abundance	B1= 0.34	279.1	0.114	0.05

Table 3. Summary of top-performing models that explain larval suppression and have ΔAICc < 2. Higher Akaike weights indicate increased model fit.

Model	Coefficients (Beta)	AICc	Akaike weight	Maximum likelihood Pseudo R^2
Grass	B1= -0.04	85.1	0.251	0.11
Grass + Trees	B1= -0.05 B2= 0.02	85.3	0.228	0.17
Intercept		86.1	0.159	
Trees	B1= 0.01	86.5	0.125	0.06
Grass + Floral richness	B1= -0.05 B2= 0.05	86.6	0.123	0.14
Grass + Floral abundance	B1= -0.05 B2 = 0.001	86.7	0.114	0.13

Figure 6. Imported cabbage worm larva parasitized by *Cotesia glomerata*, a specialist parasitoid wasp that attacks cabbage worms. The white objects surrounding the caterpillar are the emerging *Cotesia* pupae. Photo taken at urban farm.

Objective 4:

Methods

We examined if the addition of potted purple coneflower (*Echinacea purpurea* var. 'Magnus') and blazing star (*Liatris spicata* var. 'Kobold') would attract natural enemies to brassica sites and consequently reduce plant damage. On June 23, 2014, we added potted plants to 14 separate rows of brassica at two community gardens. As a control, we included 14 additional brassica rows without supplemental resources. All plots were separated by at least 10 m. Yellow sticky cards were set up in each row to assess natural enemy abundance and were replaced weekly until 1 week after plants stopped blooming (July 25, 2014). Defoliation and cabbageworm abundance were also estimated weekly in each plot. Repeated measures ANOVAs were used to analyze the effect of added plant resources on natural enemy abundance, cabbageworm abundance, and brassica defoliation.

Results

There was no significant difference in natural enemy abundance between the control and rows supplemented with potted flower (F=0.472, P=0.49). Additionally, supplemental flowers did not reduce cabbageworm larvae populations (F=0.09, P=0.76) or defoliation (F=1.57, P=0.22).

Conclusions:

Urban agriculture supports a natural enemy community dominated by parasitoids. While farms had the highest abundance of specialist parasitoids of cabbageworms, this did not result in the greatest rate of biocontrol against cabbage worms. We observed scant evidence of parasitism in field trials and noted the efficiency of lady beetles as a biocontrol agent in a lab assay. Further work is needed to evaluate if lady beetles consume cabbage worm eggs in a field environment.

Growers who seek to control cabbage worms should consider the time of outbreak when designing a pest management plan. Few natural enemies were recorded during the first generation of imported cabbageworm. Cultural controls and BT applications would be prudent to control cabbageworms during the early growing season. Our findings of a significant association between parasitoids and brassica pests suggests that second generation ICW as well as cabbage looper are likely controlled by existing natural enemies. We found that flowers adjacent to crops did not affect natural enemies or reduce herbivory. Nonetheless, sowing ornamental plants within farm boundaries offers nectar as a resource to natural enemies, which may attack pests other than cabbageworms and aphids.

The results of this study offer a positive outlook for crop production in large metropolitan areas. Farms that are integrated in densely populated residential neighborhoods already host predators and parasitoids of cabbage worms. We identified that a diverse natural enemy community is present across different food production intensities in urban agriculture. Further work is needed to evaluate how the availability of alternate prey and intraguild predation influence biocontrol of cabbage looper in urban agriculture.

Outreach:

I presented the findings of this project at an Advocates for Urban Agriculture-Research Panel in November 2014 and at the Organic Agriculture Research Symposium in February 2015. A poster about natural enemies in urban agriculture was presented at the Entomological Society of America meeting in November 2015. I produced a written pamphlet on organic control of cabbageworms and distributed this material to project participants as well as >25 other community garden and urban farm groups in Chicago, IL. I recently submitted an article for peer-review to *Biological Control* and plan to submit a second peer-reviewed article to *Environmental Entomology*. I led a module on identifying natural enemies in brassica and designing beneficial habitats at an urban farm field day in June 2015. Finally, I hired and trained an undergraduate student in insect identification, data collection, and outreach with growers.

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Appendices.

Appendix 1. Scale of damage from insect pests to vegetable plants reported by urban community garden and farm managers in Chicago, IL (N=13 garden managers). Cucurbits and brassica experience greatest damage from insect pests.

1= No noticeable damage	4= 51-75% damage
2= Up to 25% damage	5= 76-100% damage
3= 26-50% damage	

Plant	**Percentage of gardens growing plant**	**Average damage ± SD**
Bean	92%	1.42±0.64
Broccoli/Cauliflower/Sprouts	84%	2.09±1.08
Cabbage	84%	1.90±1.16
Carrot	84%	1.18±0.48
Cucumber	100%	2.23±1.24
Mint	84%	1.0
Onion	84%	1.18±0.57
Pea	77%	1.20±0.40
Pepper	100%	1.23±0.42
Potato	69%	1.44±0.68
Squash	92%	2.67±1.24
Tomato	100%	1.62±0.62

IRB approval # 2013-1142 was received for the data appearing in Appendix 1.

Appendix 2. Abundance of parasitoid wasps recorded on yellow sticky cards. Families are listed in order of total abundance. Percentages are calculated separately for parasitoids and insect predators.

Natural enemies	Total abundance	Percent of total abundance
Parasitoid wasps	20550	
Braconidae specializing on aphids (Aphidiinae)	5844	28.44
Ichneumonidae*	4805	23.38
Pteromalidae*	2739	13.33
Eucoilidae	2269	11.04
Braconidae not specializing on aphids*	1669	8.12
Diapriidae*	834	4.06
Platygastridae	531	2.58
Eulophidae*	337	1.64
Mymaridae*	333	1.62
Ceraphronidae	324	1.58
Encyrtidae*	208	1.01
Trichogrammatidae*	86	0.42
Chrysididae	83	0.40
Cynipidae	73	0.36
Pompilidae	69	0.34
Perilampidae*	61	0.30
Scelionidae*	55	0.27
Eupelmidae*	52	0.25
Aphelinidae	42	0.20
Megaspilidae	39	0.19
Eurytomidae*	32	0.16
Proctotrupidae	28	0.14
Eucharitidae	11	0.05
Tiphiidae	9	0.04
Tanaostigmatidae*	4	0.02
Torymidae	4	0.02
Signiphoridae*	3	0.01
Bethylidae*	2	0.01
Chalcididae*	2	0.01
Figitidae	2	0.01

* Indicates that wasp family is classified as a 'specialist' capable of parasitizing insects in the Order Lepidoptera.