Integrating Biological Control for the Key Pests Diamondback Moth (*Plutella xylostella)* and Cabbage Aphid (*Brevicoryne brassicae)* in Organic Brussels Sprouts

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1. Project Summary

Diamondback moth (*Plutella xylostella*) and cabbage aphid (*Brevicoryne brassicae*) are primary pests of Brussels sprouts on the California Central Coast. Given the damage potential of these pests and the lack of effective, organically compliant pest control materials, Brussels sprouts production is extremely challenging for organic growers. We investigated the efficacy and diversity of naturally-occurring parasitoids and predators in organic Brussels sprouts. Seasonal DBM parasitism by the native Ichneumonid parasitoid *Diadegma insulare* averaged 55% and 27% in 2010 and 2011, respectively. *D. insulare* was effective during both sample years in preventing DBM-related yield reductions. Syrphid larvae demonstrated high aphid predation rates, particularly during the second-half of the growing season. Unfortunately, this predation was often not sufficient in preventing aphids from feeding on the potentially-harvestable sprouts. An integrated aphid management approach that 1) utilizes and encourages early season predation by the six different species of syrphid larvae that were recorded during this study and 2) incorporates aphid-directed OMRI-approved insecticides in the second half of the season, may improve the economic viability of organically-produced Brussels sprouts.

2. Introduction to Topic

California leads the nation in Brussels sprouts production, currently producing 90% of the nationally marketed product. However, organic growers have been left out of any research efforts to rationalize pest management of the key pests of Brussels sprouts: cabbage aphid and diamondback moth (DBM). This is in part due to the fact that conventional production has been largely reliant on organophosphate insecticides. This lack of attention has largely precluded any organic pest control innovation and has created a knowledge gap for improving organic production techniques. The lack of organically compliant pest management tools (both physical and knowledge-based) is exacerbated by the ability of specialist herbivores (cabbage aphid and DBM) to take advantage of Brussels sprouts' nine-month production cycle, often causing outbreaks that dramatically reduce organic yields. Very low economic thresholds established by the processors further reduce yields. As a result of these formidable challenges, most growers are reluctant to grow Brussels sprouts organically.

We worked closely with organic farmers who are keenly interested in improving and implementing a biologically-based whole systems approach to the production of organic Brussels sprouts. The farmers involved in this project have been very proactive in seeking assistance and encouraged the preliminary documentation of parasitoids and predators in cultivated fields. A scientifically rigorous approach to their pest problems would help provide the tools needed for improved organic yields and economic viability.

3. Objectives Statement

In 2010 and 2011, we investigated three research-oriented objectives regarding biological control in organic Brussels sprouts:

1) Document the degree of parasitism by *Diadegma insulare* in organic Brussels sprouts

D. insulare (Fig. 1) has been recognized as an effective biological control agent of DBM in other regions of the country (Sarfraz et al. 2005). However, how this parasitic wasp functions in organically-managed systems in Northern California had not been documented. Our aim was to provide a quantitative assessment of DBM parasitism in organic Brussels sprouts, and ultimately, determine the efficacy of this parasitoid in preventing DBM-related yield reductions.

2) Describe the diversity and relative abundance of syrphid larvae in Brussels sprouts

Members of the predacious fly family Syrphidae are the most economically important biological control agents of the cabbage aphid (*Brevicoryne brassicae*) in broccoli on the California Central Coast (Nieto *et al*. 2006). Similar to broccoli, syrphid larvae play a vital role in suppressing cabbage aphid populations in Brussels sprouts. However, there have been very few descriptions of syrphid ecology in California Brussels sprouts. Given that the action threshold for aphids inside fresh market Brussels sprouts is ≥1, it is critically important to understand and utilize syrphid dynamics for organic cabbage aphid control. Also, understanding how the extended growing season that is unique to Brussels sprouts (among cole crops) affects these predator-prey dynamics is also useful for aphid management.

These key components address larger issues in organic cole crop production: biological control of cabbage aphids via syrphid predation, although often effective, is considered inconsistent. One possible explanation of these inconsistencies is a poor understanding of syrphid larvae dynamics at a species level. Once individual syrphid species can be identified and recognized by growers and pest control advisors, what are now considered "inconsistencies" can be adjusted to recognize behavioral species-specific differences that can help make organic cole crop systems more predictable and thereby more productive. For example, different syrphid larvae species demonstrate varying aphid-consumption rates (Hooper et al. 2011), which could help explain inconsistencies in aphid reduction.

3) Determine percent parasitism and predation rates when providing nectar-based floral resources in organic Brussels sprouts

Predator and parasitoid performance is occasionally improved by the integration of insectary habitats: nectar-providing flowers that improve a beneficial's ability to reduce pest populations. The effects of sweet alyssum (*Lobularia maritima*) were tested on the spatial distribution of aphids, syrphid larvae, DBM and *D. insulare* in 2010 and 2011. *D. insulare* longevity is greatly improved in the presence of sweet alyssum (Johanowicz and Mitchell 2000). Sweet alyssum also increases fecundity of adult syrphid flies, thereby improving aphid predation rates (Hogg et al. 2011). By comparing predation and parasitism at varying distances from insectary habitats during the relatively long Brussels sprouts growing season, we will assess the value of these floral resources for improving DBM and cabbage aphid management. It should be noted that annual buckwheat (*Fagopyrum esculentum)* was removed from this experiment due to a short flowering period (relative to the long sprouts' season) and a low resiliency to weed pressure.

4. Materials and Methods

Experiments were conducted on three organically certified farms in Santa Cruz County in 2010 and 2011. Data were collected weekly from Rodoni Farms in Davenport, Jacobs Farms at Wilder Ranch in Santa Cruz (sprouts plants donated by Rodoni Farms; Fig. 2) and the Center for Agroecology and Sustainable Food Systems (CASFS) at the University of California, Santa Cruz (sprouts plants donated by Rodoni Farms; Fig. 3). At these sites, sprouts were planted on beds approximately 36 inches apart and with a planting density of 30-46 cm. Beds were 37m, 61m and 122m long at Wilder Ranch, CASFS and Davenport, respectively. Jacobs Farm/Del Cabo has maintained organic certification with Oregon Tilth Certified Organic, while Rodoni Farms and CASFS have maintained certification with California Certified Organic Farmers.

1) Document the degree of parasitism of DBM by *Diadegma insulare* in organic Brussels sprouts

To document parasitism of DBM by *D. insulare*, late-stage DBM larvae and pupae were collected from Brussels sprouts fields in Davenport and Santa Cruz. Specimens were placed in vials and brought (within 2-3 hours) to the CASFS laboratory for rearing. Once in the lab, each DBM larva or pupa was placed in a Petri dish; larvae were fed Brussels sprout leaves (grown at CASFS) and monitored daily until pupation. Dates of collection, pupation and emergence (moths vs. wasps) were recorded. Ten locally-collected specimens of *D. insulare* were initially verified by Dr. David Wahl at the American Entomological Institute in Gainesville, FL in 2009.

2) Describe the diversity and relative abundance of syrphid larvae in organic Brussels sprouts

To record syrphid species and their relative abundances, syrphid larvae were collected from Brussels sprouts plants at each experimental site. Syrphid larvae were collected and transported to CASFS in a similar manner as described above. Reared syrphid larvae were provided with cabbage aphids collected from sprouts grown at CASFS. Emerged adult syrphids were identified to species and recorded. Photographs of collected syrphid larvae were also taken to confirm the physical characteristics of various syrphid species to improve the accuracy of future larval field identifications.

3) Test percent parasitism and predation rates by providing nectar-based floral resources in Brussels sprouts

To test the effects of sweet alyssum on the abundance of predators (syrphid larvae) and the parasitoid *D. insulare*, a replicated experiment on 0.1 ha was conducted at Wilder Ranch in 2010 and 2011. Single rows of beneficial insectaries bordered 20-row plantings of Brussels sprouts. Flowers were direct-seeded approximately two weeks before Brussels sprout seedlings were planted. Rows of alyssum and sprouts were watered with overhead sprinklers during the growing season according to the growers' normal irrigation schedule. No insecticides or other insect management techniques were implemented during these trials. Four Brussels sprouts replicates were used to collect insect and harvest data. In 2010, replicates were 167m^2 in size and separated by 3m. In 2011, replicates were 58m^2 large and separated by 2m. Insects were counted and collected from plants in each row away from alyssum (12 rows in 2010 and 7 rows in 2011). A randomized complete-block design ANOVA was used to identify potential row differences regarding pest and/or natural enemy abundance.

5. Results

Diamondback Moth

At Wilder Ranch, diamondback moth densities were high in 2010 (0.99 ± 0.05 DBM/plant) and low (0.14 ± 0.01 DBM/plant) in 2011. Notably in 2010, pest densities peaked at almost four DBM (larvae plus pupae) per plant (Fig. 4). However, due to robust parasitism by *D. insulare*, DBM densities were reduced to <1/plant by season's end. In 2010, parasitism averaged 0.55 ± 0.04% and peaked at 0.92 ± 0.20% on 2 September. With low DBM host densities in 2011, parasitism was more modest, averaging 0.27 ± 0.07% and peaking at 0.67 ± 0.33% on 14 September. No hyperparasitism was recorded from the 370 total specimens collected during this study.

The distribution (based on row # relative to sweet alyssum) of DBM abundance ($F = 0.12$, $P = 0.757$) and percent parasitism ($F = 5.04$, $P = 0.108$) was not significantly different in 2010. Similarly in 2011, neither DBM ($F = 0.84$, $P = 0.552$) nor parasitism ($F = 0.66$, $P = 0.690$) distribution differed due to sweet alyssum field-placement. Harvested organic Brussels spouts were almost completely lacking in DBM (Table 1). Over two seasons, 1,162 sprouts were dissected in search of a DBM larva or pupa; only one was encountered.

Cabbage Aphid

At Wilder Ranch, pest densities averaged 39.6 ± 2.7 and 54.8 ± 2.8 aphids/plant in 2010 and 2011, respectively. At CASFS, seasonal density averaged 158.1 ± 8.0 aphids/plant in 2011. There was a substantial syrphid larvae presence in 2011, with single-date means peaking at 2.7 ± 0.3 and 6.2 ± 0.7 larvae/plant at Wilder and CASFS, respectively (Fig. 5). At season's end, mean aphid/plant densities (not including the sprouts) were 50.0 in 2010 and < 20.0 at both field sites in 2011.

Despite substantial aphid field reductions in October and November, harvest rates were nonetheless negatively affected by the previous infestations (Table 2). In these unsprayed fields, cabbage aphids contaminated between 1/6 and 1/3 of inspected sprouts. Syrphid larvae were also found in up to 10% of inspected sprouts (Table 2).

In 2010, the establishment of sweet alyssum was influential in shaping the row distribution of cabbage aphids (F = 21.09, P = 0.019), but not syrphid larvae (F = 8.25, P = 0.062) in organic Brussels sprouts. Aphid abundance was greatest in rows of sprouts closest to sweet alyssum (Fig. 6). In 2011, neither cabbage aphids (F = 1.48, P = 0.240) nor syrphid larvae (F = 0.78, P = 0.710) demonstrated a spatial pattern relative to sweet alyssum placement.

Field-collected larvae reared at UCSC yielded six different syrphid species, including *Allograpta obliqua*, which was most commonly encountered during both 2010 and 2011 (Figs. 7-8). Larval mortality during the rearing process was considerable: 44.2% in 2010 and 65.3% in 2011. Relative to successfully reared syrphid flies, parasitism of syrphid larva by the Ichneumonid *Diplazon* sp. was 10.8% and 2.0% in 2010 and 2011, respectively.

6. Conclusions and Discussion

Diamondback Moth

DBM is commonly cited as a primary pest of cole crops (Sarfraz et al. 2005). This study confirmed high mid-season DBM densities on organic Brussels sprouts. However, *D. insulare* provided excellent biological control during this study and largely prevented reductions in harvestable sprouts. *D. insulare* is one of the most efficacious DBM paraistoids in North America and often parasitizes 70-90% of DBM larvae (Sarfraz et al. 2005). Despite this prominence, *D. insulare* had only been documented in Southern California (Oatman and Platner 1969) and at CASFS-UCSC (Fox et al. 1990); no published accounts described this parasitoid in commercial agriculture in Northern California. In the absence of a disturbance (ecological, chemical, climatic, etc.), *D. insulare* appears capable of negating the pest potential of DBM in organic Brussels sprouts.

There are two caveats to this DBM biological control: First, minor foliar damage/defoliation on the outer leaves of a Brussels sprout was not included in our harvest assessments. This type of damage, while not as serious as larval contamination, nonetheless reduces product value. Without having observed the defoliator, it was not feasible to attribute damage to a specific pest (e.g. DBM, cabbage looper, imported cabbage worm, etc.). DBM damage estimates, which we calculated to be almost zero, may therefore be oversimplified or underestimated due to this exclusion.

The second caveat to our reported DBM biological control is that parasitism will likely be diminished while managing other pests (namely cabbage aphids). While DBM is very well-suited to developing chemical resistance to insecticides, including some OMRI-approved materials (Sarfraz et al. 2005), *D. insulare* is very susceptible to insecticide applications (Xu et al. 2001). Thus, imbalances between DBM and *D. insulare* are created that lead to yield reductions (Idris and Grafius 1993).

Generally speaking, this study was tremendously useful in identifying *D. insulare* as an effective DBM biological control agent in Northern California. Tracking DBM parasitism is actually quite feasible for growers: collected DBM pupae can be stored in a transparent container at room-temperature. No more than ten days later (in most cases), either a diamondback moth or *D. insulare* wasp will emerge. Sampling every 1-2 weeks will establish parasitism trends so that growers can make informed management decisions.

Future work should focus on the extent of DBM parasitism in related cropping systems. For instance, is *D. insulare* active in other cole crops, such as broccoli, and if so, is it effective in controlling this pest during a much shorter growing season? Additionally, what is the extent of DBM parasitism in conventional Brussels sprouts, where DBM is still considered a legitimate pest with considerable damage potential? If parasitism is diminished, are there ways to mitigate the effects of insecticide use on this parasitic wasp?

Cabbage Aphid

While DBM is sufficiently managed by naturally-occurring biological control in organic Brussels sprouts, cabbage aphids are not adequately controlled by predators or parasitoids and in fact present a serious challenge to the viability of this crop. Cabbage aphids are specialists on cole crops and utilize the long growing season of Brussels sprouts to arrive and increase colony size. Based on this study's findings, only after sprouts are well-developed do syrphid larvae diminish aphid densities. By that time, aphids have often colonized the sprouts themselves. Their feeding causes foliar damage, either directly through wilting or indirectly through honeydew-induced mold. Consequently, even if predation removes cabbage aphid from these sprouts, significant damage to the harvestable portion of this plant has often already occurred. While this type of damage was not included in aphid-related harvest calculations, the majority of sprouts, many of which were free of insects, had clearly been previously fed upon by aphids, and as such, had suffered substantial cosmetic damage.

Insect contamination in Brussels sprouts is also a valid concern. Aphids were found deep inside sprouts, making their removal difficult. Once inside, aphids were reduced or removed from a sprout only by syrphid larvae (maggots), which are themselves contaminants. Thus, the biological solution becomes a commercial problem.

While difficult, there are growers on the central coast who are producing organically-managed Brussels sprouts with some success. An integrated pest management approach, which balances biological control with well-timed insecticide applications may provide a framework for organic cabbage aphid control. Prior to the development of lateral buds (sprouts), growers could potentially utilize syrphid larvae predation to maintain aphid populations to reasonable levels. However, since predator densities during these first two months are often inadequate, future research should focus on investigating sources of alternate prey to accelerate the ascension of larval abundance in Brussels sprouts fields. For example, bell beans and barley were identified as possible candidates to support alternate aphid species that would boost syrphid populations for the eventual management of the lettuce aphid in organic lettuce (Daane et al. 2008). If such an intercrop was successfully established, cabbage aphid could conceivably be managed by naturally-occurring predators for almost half of the growing season.

During the second half of the growing season, however, when sprouts have formed and are vulnerable to aphid feeding, insecticide applications are warranted. The insecticidal soap M-Pede ® appeared to be efficacious in reducing aphid densities. Scouting a field thoroughly and consistently would be critical for proper aphid management during this particularly vulnerable period of the growing season.

7. Outreach

On April 18th, 2012, the results of this project were presented at the San Mateo County Farm Bureau to roughly two dozen participants (mostly Brussels sprouts growers). Joji Muramoto also presented research findings from the OFRF project entitled "Integrated soil-borne disease and weed management for organic strawberries using anaerobic soil disinfestation, broccoli residue incorporation and mustard cake application". The findings of this OFRF project were also presented at Agriculture and Land-Based Training Association (ALBA) on January 16, 2013.

8. Financial accounting

Please provide a list of the expenditures made to conduct this project. OFRF requests that any unspent funds be returned to OFRF.

9. Leveraged resources

As a product of the Brussels sprouts extension meeting, we were able to establish a cohort of interested grower-collaborators who would later assist in our successfully-funded California Department of Pesticide Regulation grant proposal "Integrated pesticide reduction strategies for insect and disease management in cole crops". Among other goals, this project strives to reduce insecticide applications targeted at the cabbage maggot fly in Brussels sprouts and broccoli.

10. References

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11. Photos and other addenda

Figure 1. The diamondback moth parasitioid, *Diadegma insulare*.

Figure 2. Organic Brussels sprouts study site at Jacob's Farm at Wilder Ranch in Santa Cruz, CA. At right, UCSC student-intern Kevin Landaw searches a Brussels sprout plant for pests and their associated natural enemies in 2011.

Figure 3. Organic Brussels sprouts study site in 2011 at the Center for Agroecology and Sustainable Food Systems (UCSC) in Santa Cruz, CA.

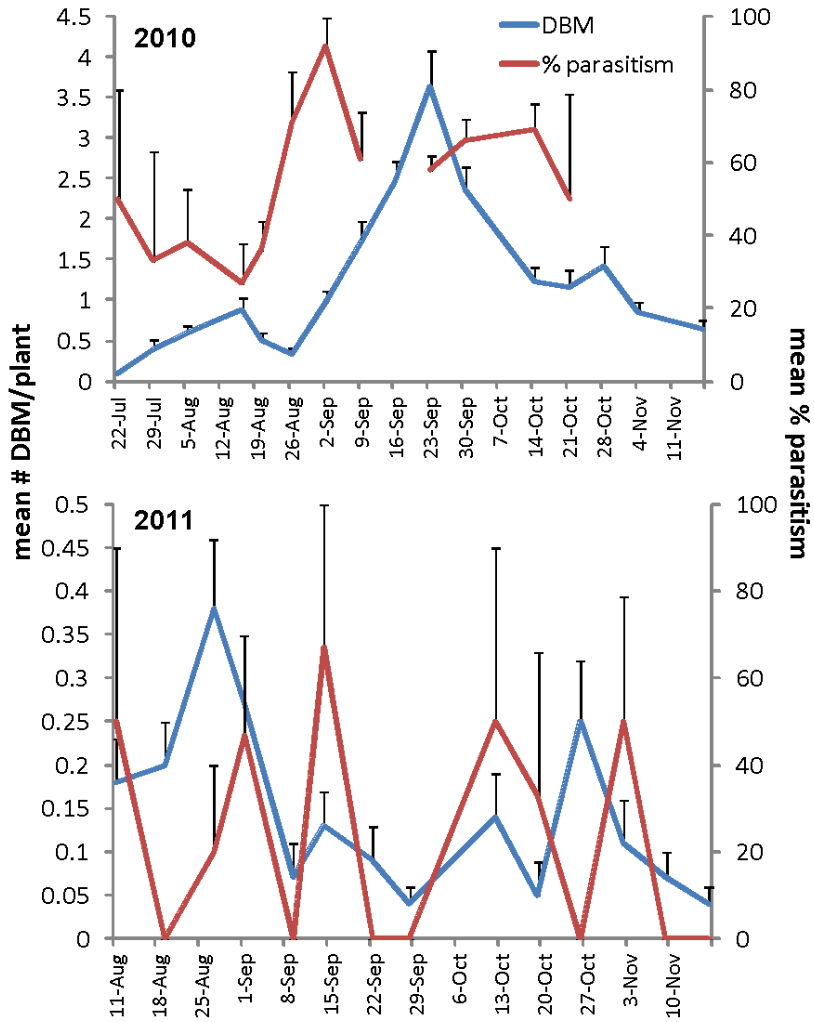

Figure 4. Mean diamondback moth (DBM) abundance (larvae + pupae) per Brussels sprouts plant and percent parasitism of DBM by *Diadegma insulare*. Data collected at Wilder Ranch in Santa Cruz, CA.

Table 1. Rates of diamondback moth (DBM) contamination in harvested organic Brussels sprouts. Sprouts collected from Wilder Ranch and the Center for Agroecology and Sustainable Food Systems (UCSC) in Santa Cruz, CA.

Study Site	Year	N	#DBM	% Sprouts Infested
Wilder	2010	320	0	0
Wilder	2011	712	0	0
CASFS-UCSC	2011	130	1	0.80%
TOTAL		1162	1	0.0009%

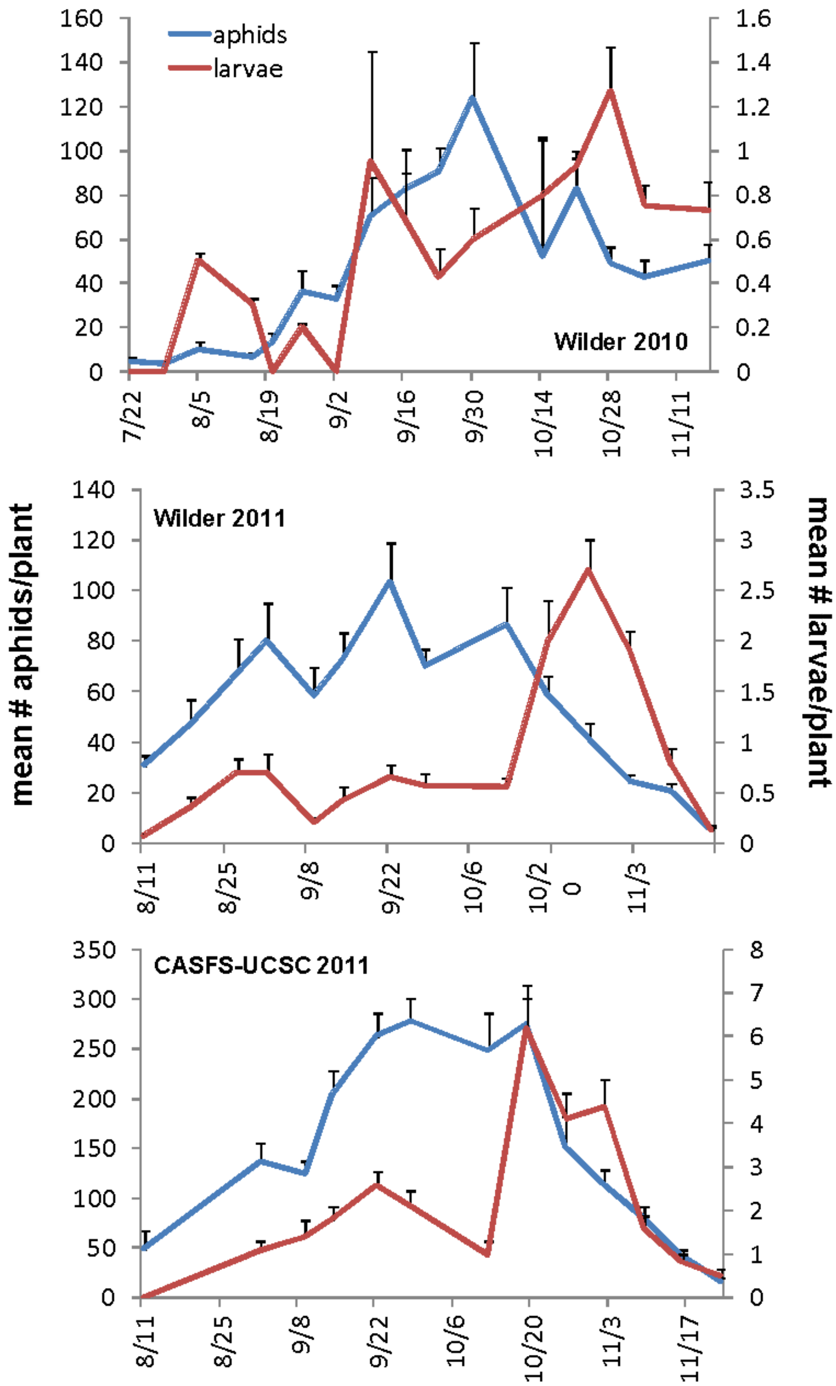

Figure 5. Mean cabbage aphid and syrphid larvae abundance per Brussels sprouts plant. Data collected from Wilder Ranch and the Center for Agroecology and Sustainable Food Systems in Santa Cruz, CA.

Table 2. Rates of cabbage aphid and syrphid larvaae contamination in harvested organic Brussels sprouts. Sprouts collected from Wilder Ranch and the Center for Agroecology and Sustainable Food Systems (UCSC) in Santa Cruz, CA.

	study site	year	N	mean	max/sprout	% sprouts infested
Cabbage Aphids	Wilder	2010	320	0.36 ± 0.05	5	19.7
	Wilder	2011	357	0.57 ± 0.13	30	16.2
	CASFS	2011	130	1.96 ± 0.35	20	33.0
Syrphid Larvae	Wilder	2010	320	0.12 ± 0.20	2	10.6
	Wilder	2011	357	0.09 ± 0.02	5	7.3
	CASFS	2011	130	0.08 ± 0.03	3	6.0

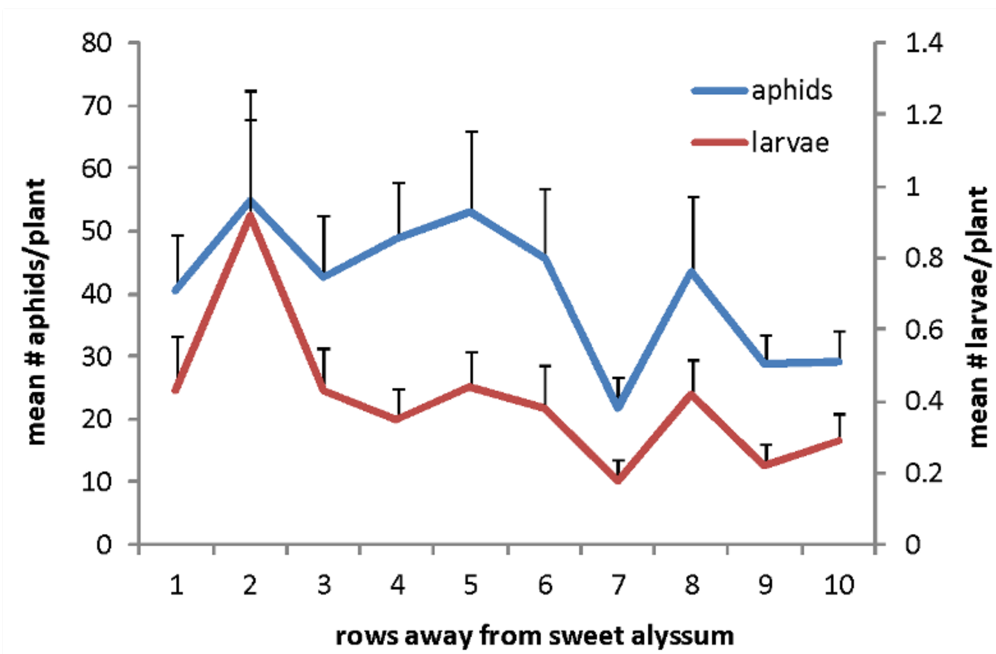

Figure 6. Seasonal mean cabbage aphid and syrphid larvae abundance per Brussels sprout plant measured in rows away from sweet alyssum. Data collected in 2010 from Wilder Ranch in Santa Cruz, CA.

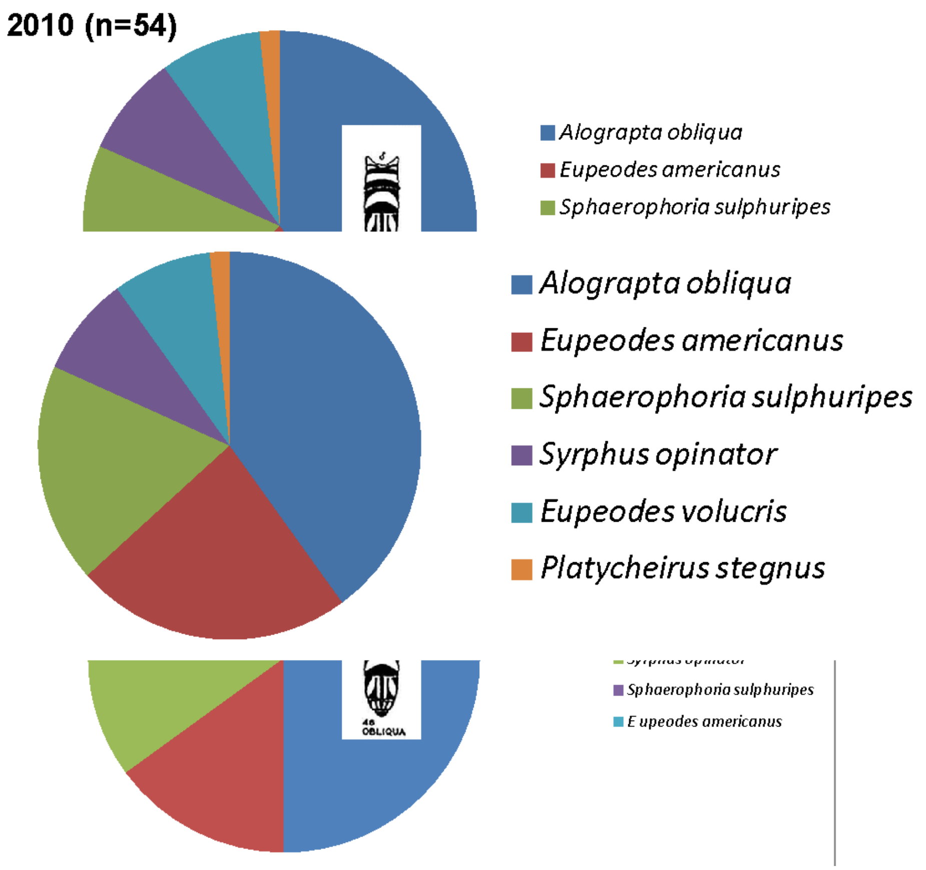

Figure 7. Syrphid species composition of successfully reared adults collected from organic Brussels sprouts. Data from Wilder Ranch and the Center for Agroecology and Sustainable Food Systems (UCSC) in Santa Cruz, CA.

Figure 8. Syrphid larvae collected in organic Brussels sprouts: A - *Sphaerophoria sulphuripes*, B - *Syrphus opinator*, C - *Allograpta obliqua*, D - *Platycheirus stegnus*. Photos taken at the University of California, Santa Cruz.