# FINAL PROJECT REPORT
**WTFRC Project Number: CP-07-708**

**Project Title:** Improving apple IPM by maximizing opportunities for biological control


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**Other funding Sources**

**Agency Name: N/A**
**Amount awarded:**
**Notes:**


**Total Project Funding: (one year project only)**

**Budget History:**

| Item | Year 1: |
|---|---|
| **Salaries** | $82,489 |
| **Benefits** | $20,445 |
| **Wages** | $3,000 |
| **Benefits** | $345 |
| **Equipment** | $0 |
| **Supplies** | $4,500 |
| **Travel** | $2,021 |
| **Total** | **$112,800** |

**Objectives**

1. Determine the effects of thinning sprays on natural enemy abundance, diversity, and phenology.
2. Directly estimate predation intensity on codling moth and leafroller survival in different sprayed environments created in Objective 1 and determine which predators are the key sources of LR and CM biological control.
3. Evaluate the biological capacities of two tachinid species, which are the dominant parasitoids of leafrollers and the influence of habitats, season, and insecticides on their behavior and performance.
4. Develop phenology models for key natural enemies and integrate those data and the ranking of natural enemy importance and seasonality into WSU-DAS management recommendations.

**Significant Findings**

OBJECTIVE 1: EFFECTS OF THINNING SPRAYS ON NATURAL ENEMIES

- Thinning sprays had apparently only modest effects on tree-dwelling predators, and no noticeable effects on ground-dwelling predators

OBJECTIVE 2: ESTIMATE PREDATION INTENSITY ON CODLING MOTH AND LEAFROLLER

- There were no significant differences observed in predation induced mortality of cocooned codling moth larvae between the two orchard management treatments.
- Several carabid, spider and one "daddy long legs" species had codling moth remains in guts in late-summer and fall at sites having high densities of codling moth. Gut content analyses of mid-summer populations are ongoing and will continue through the winter.
- Three carabid beetle species, two spiders, and one "daddy long legs" species showed no interest in CM larvae in cocoons but they aggressively attacked and ate active larvae in lab studies.
- Free moving leafrollers proved unreliable for estimating predation rates, thus the effort to use sentinel leafrollers in evaluating predation was dropped early in the season.

OBJECTIVE 3: EVALUATE TACHNID SPECIES

- *Nilea erecta and Nemorilla pyste* have a different way of maturing eggs that may dictate their sensitivity to IGR pesticides in the adult stage
- Preliminary studies show damaged leaves + larvae + silk + frass have a greater attraction to *N. pyste* than diet + larvae + silk + frass
- Benzaldehyde added to sticky cards significantly increases trap catch opening the door for monitoring phenology and abundance as well as helping determine movement patterns of both Tachinid species.
- Intrepid and Esteem did not appear to be highly detrimental to immature tachinids, but results reported last year showed that females emerging from treated larvae were sterile. More work is needed to confirm results.

OBJECTIVE 4: DEVELOP PHENOLOGY MODELS FOR KEY NATURAL ENEMIES

- Timing of first appearance in orchards differed substantially among species of predators
- Trees were banded at 13 orchards to collect overwintering predators for emergence study (ongoing)

**Results and Discussion**

OBJECTIVE 1: EFFECTS OF THINNING SPRAYS

Thinning sprays were applied to 1 acre plots of non-bearing Red Delicious apples located at an orchard in west Yakima. We had four control plots and four treatment plots. Thinning sprays were made to treatment plots on April 28 and May 7 (2% lime sulfur + 2% rocker fish oil), and on May 23 (Sevin 4F; 3 pints per acre with 3 oz. NAA Fruit fix 200 K-salt in 100 ga.). Predatory arthropods were sampled using beat trays (45 per plot) for tree-dwelling predators, and pitfall traps (10 per plot) for ground-dwellers. Samples were taken at scheduled intervals beginning two weeks preceding the first lime sulfur spray, and extending to the last sample in late July. In October, 15 trees per plot were banded to assess overwintering densities of natural enemies.

Tray counts are shown for ladybird beetles, true bug predators, spiders, and all predators (Fig. 1B). The final sample taken on July 27 is not shown, to allow all bars to fit in the figure. There is a weak suggestion for true bugs and all predators that densities declined in the treatment plots (black bars), whereas the opposite was true for the ladybird beetles. If we plot cumulative counts of total predators through time (Fig. 2A), this slight decline in the treatment plots shows up more readily, and appears to coincide with the onset of the thinning sprays. Pitfall samples are shown for spiders, daddy long-legs, ground beetles, and all predators (Fig. 1B). There is no suggestion whatsoever that the thinning sprays affected numbers of these ground-dwelling predators (Fig. 2B). Overwintering predators in bands were dominated by spiders (Fig. 3); early season thinning sprays had no effect on eventual overwintering densities.

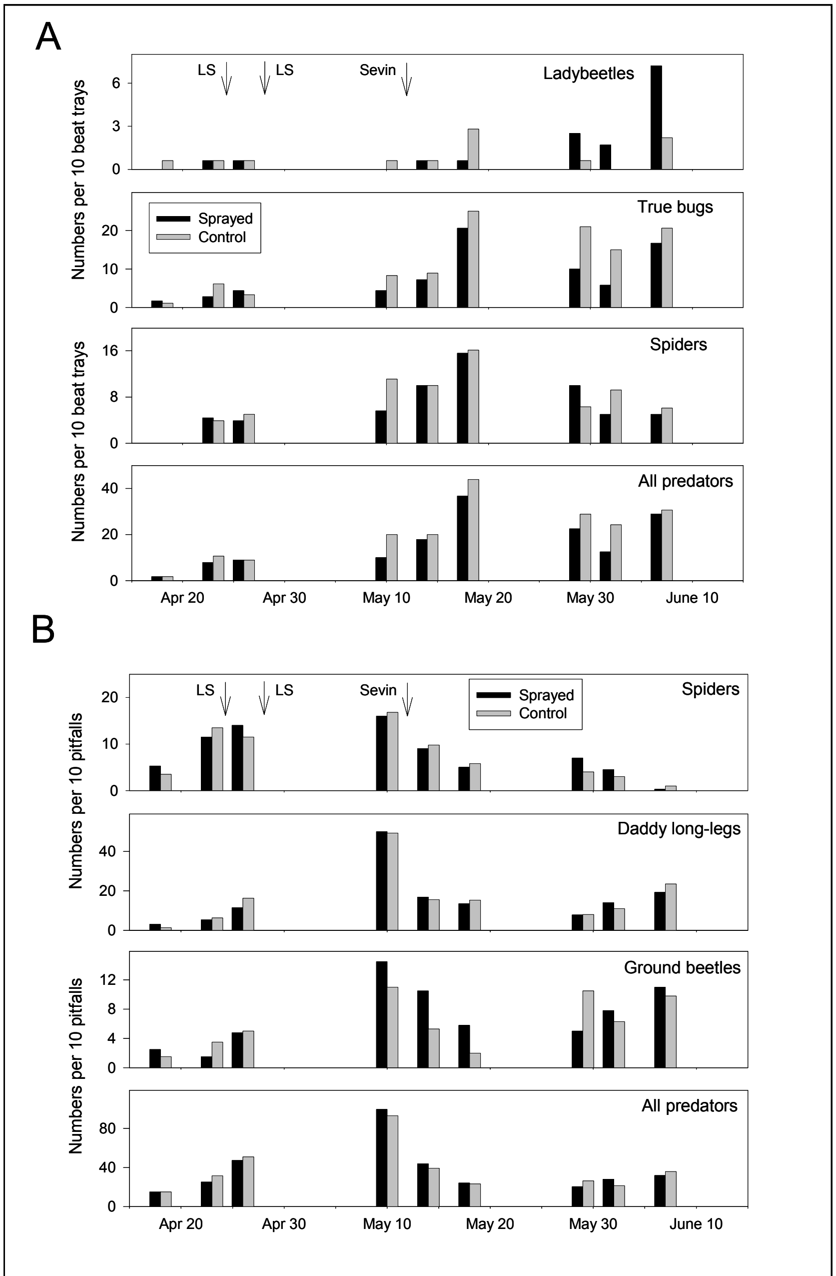



**Fig. 1**. Effects of lime-sulfur (LS) and Sevin thinning sprays on densities of tree-dwelling (A) or ground-dwelling predators (B).

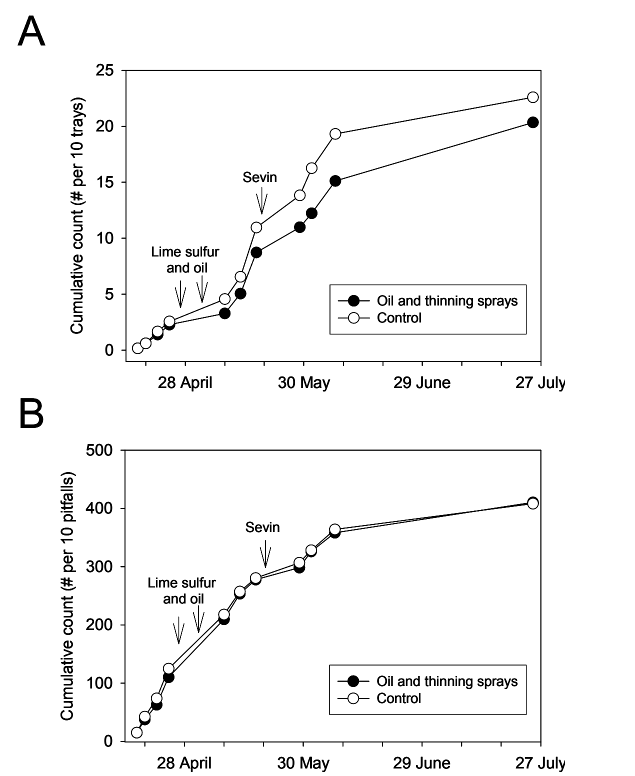



**Fig 2**. Effects of thinning sprays on cumulative numbers of tree-dwelling (A) and ground-dwelling (B) predators

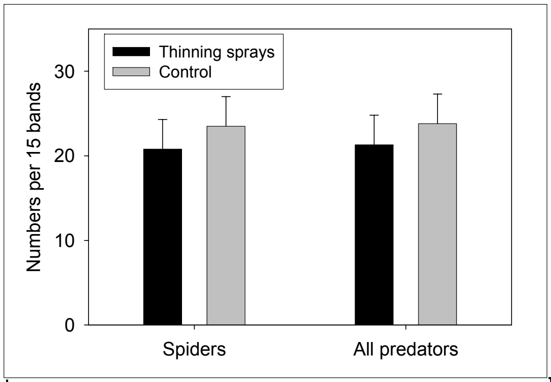



**Fig. 3**. Effects of thinning sprays on overwintering densities of predators

OBJECTIVE 2: ESTIMATE PREDATION INTENSITY ON CODLING MOTH AND LEAFROLLER

Diapausing male codling moth larvae that had cocooned in small wooden blocks were deployed in each of the eight plots used in the thinning spray experiment, with 18 blocks per plot, at five intervals: mid April until the first thinning spray, mid May, mid June, mid July and mid August, following a spray of Assail (in the thinning plots only) at the end of July. Predation was identified from larval remains showing signs of insect feeding, and holes cut in cocoons that are not characteristic of that caused by codling moth when abandoning the cocoon. Fig. 4 shows clearly that flower and fruit thinning sprays had no effect on codling moth predation. Similarly there was no effect of the late-July Assail spray on predation rates. The most noticeable pattern seen with the sentinel exposures was the appearance and dramatic increase of the category of codling moth disappearance we call 'removed' which was characterized by very clean-cut holes in the cocoons and codling moth larvae missing. Currently, we hypothesize that this removal was done by an as yet unknown spider or by social wasps. We came to this conclusion by providing sentinel blocks to those species of carabids and spiders that were readily collected in the pitfalls, and found that none of these species produced this signature of predation on cocoons. The overall effect of this removal category was to increase mortality from 20% early in the season to some 60-85% later (Fig. 5). Again there was no evidence for an effect of thinning sprays or a single Assail spray on mortality levels. Gut content analyses will be presented at the research review.

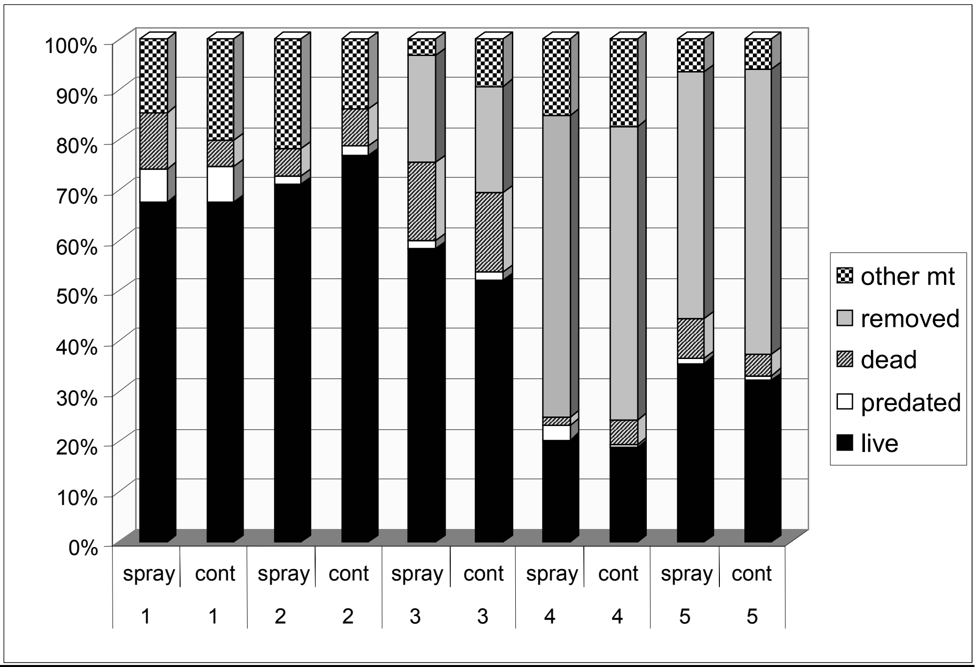



**Fig. 4.** Effects of lime sulfur spray (between interval 1 and 2), Sevin spray (between interval 2 and 3), and a single spray of Assail (between interval 3 and 4) on the survival and predation of cocooned codling moth larvae deployed in sentinel wooden blocks. The black bars are live larvae, white are dead larvae showing clear predation marks, dashed bars are larvae dead from unknown causes, gray bars are "removed" larvae accompanied by a cleanly cut hole in cocoon; other "mt" appears to be codling moth that left cocoon naturally.

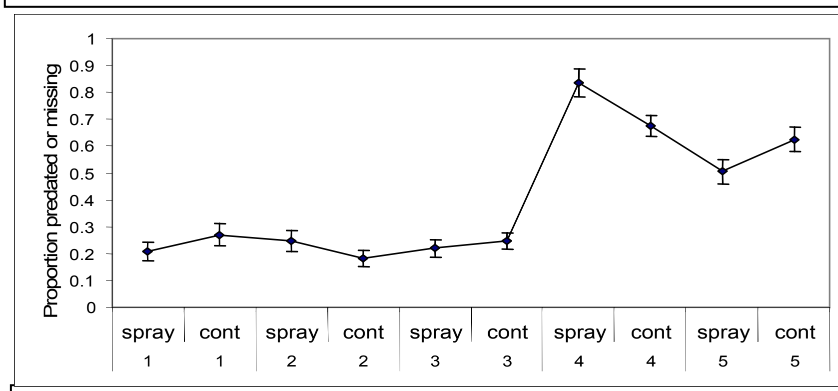



**Fig. 5**. Pattern of mortality of cocooned codling moth in sprayed and unsprayed plots during the season. Removal of codling moth through a characteristic hole in the coocons accounts for the upswing in mortality in period 4 and 5 (Jul, Aug).

OBJECTIVE 3: EVALUATE TACHNID SPECIES

*Potential to limit OBLR population growth.* We examined the development and incubation of eggs in the adult parasitoid because the rate at which parasitoids can produce eggs has implications to the amount of time needed for a parasitoid population to respond to an increasing or decreasing pest population. We found that *N. erecta* females emerge with approximately 500 eggs in each ovary (Fig. 6), but, *N. pyste* ovaries contain only 3-5 eggs at emergence (Fig. 7). Initial tests of reproductive rates with *N. pyste* have measured up to 16 eggs deposited in a 24 hr period, so the maturation of an egg from the basal cell must be relatively rapid. The differential ways that eggs mature are also potentially a key factor in susceptibility to IGR insecticides. For example, it is possible that *N. pyste* adults exposed to IGR's will not be able to mature eggs until residues decline. Conversely, it is also possible that *N. erecta* emerging from an intoxicated leafroller may all be sterile. Further work to answer these questions is required.

In parasitoids with similar modes of egg development, adult feeding is critical to egg production. Tachinid flies are primarily nectar feeders, so it is possible that relative distance to nectar sources could affect parasitism by this species in orchards.

Experiments are underway to determine how parasitoid egg production is affected by size and age of flies, and exposure to different host densities. This will provide the information necessary to understand the value of these parasitoids as mortality factors of OBLR in orchards in terms of parasitism rates, total reproductive potential, and ability to respond to OBLR outbreaks.

We also have begun to examine the modes of attraction of the flies into orchards. In general, flies are more visually oriented than other parasitoids, but their olfactory capabilities are also important. Among the key question we hope to answer is how the flies find their hosts in the environment. One possibility is that the flies key in on volatile compounds given off by larvae, larval products such as silk or frass, or damaged foliage.

Our approach is to begin evaluating the suite of potentially important odors simultaneously, and then remove individual components to try to isolate particular sources of attraction. These studies are performed in Y-tubes under wind current. One side of the tube has an odor of possible interest at the source, and the other side has a control odor. Initial trials were performed to try to work out reasonable timeframes for response and sources of flies that respond reliably. Responses can be affected by the intrinsic state of the insect, so age, mating status, and number of eggs can affect the results.

**Fig. 6.** The ovary of a 3-day-old virgin female *N. erecta* contains ~ 500 eggs, each measuring approx. 0.2 mm. One ovary is pictured.

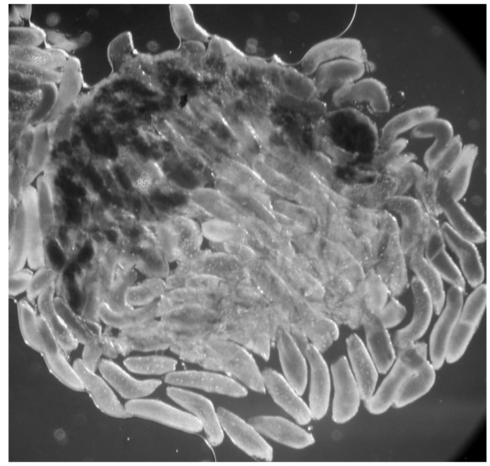


**Fig. 7.** The ovary of a 3-day-old virgin female *N. pyste* contains 3-5 eggs (arrow), each measuring approx. 0.7 mm. Both ovaries are pictured.

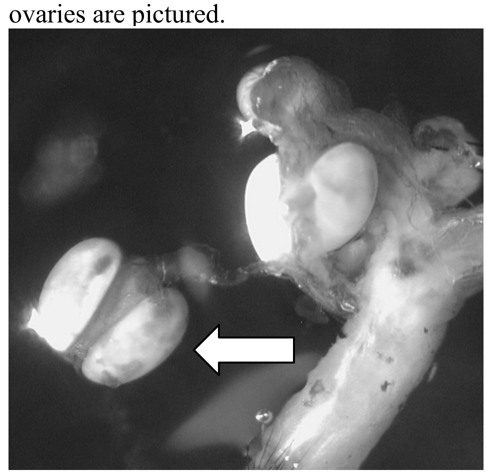
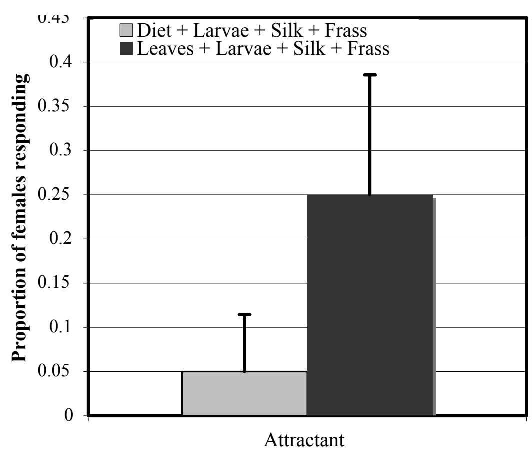


**Fig. 8.** Results of preliminary trials evaluating the attraction to hosts, host products and their associated odors to *N. pyste* females.



Our initial trials suggest that despite being reared from OBLR on artificial diet, *N. pyste* females are more responsive to the odors produced from the more natural association of damaged leaves, larvae, silk, and frass, compared to larvae, silk, and frass on artificial diet (Fig. 8). Response levels were relatively low, which raises the possibility that some aspects of these trials are suboptimal. However, *N. erecta* showed a response rate of 42 % to damaged leaves, frass, silk and larvae. This apparent disparity in the response rate of the two species is congruent with differences in the mode of attack of the two species. Because *N. erecta* oviposits on foliage near hosts, one might expect that this species is more adapted to detect host-associated odors. By contrast, *N. erecta* attacks hosts directly, and may be more visually attracted to hosts. Future work will address these issues.

*Detecting Adults in the Field.* The ability to detect parasitoids with a trap would provide an easier way to estimate their population levels, phenology, and potentially a way to detect parasitoids in non-orchard habitats to help determine which habitats are utilized. The literature provides several examples of potential plant volatiles that are released when insects feed on foliage. We tested squalene, which occurs in golden delicious foliage that has been damaged by leafminer, but does not occur when foliage is damaged mechanically. This suggests that squalene may act as a kairomone to attract natural enemies. Another compound, which has been reported by David James at WSU-Prosser to specifically attract tachinids in a different crop system, is benzaldehyde. This is a common aromatic compound that is associated with fruit and flowers.

Traps consisted of 2 ml of squalene, benzaldehyde. or water (for the controls) in glass vials attached to sticky cards which were hung from foliage. There were five traps representing each compound in a block with known tachinid population. Traps were refilled frequently and were rotated to avoid spatial bias. Benzaldehyde proved more attractive than either squalene or the control traps (Fig. 9), capturing specimens of both *N. pyste* and *N. erecta*. Squalene did attract significant numbers of green lacewings in this test, suggesting that it may be useful in monitoring lacewing populations.

**Fig. 9.** The number of tachinids attracted to sticky cards baited with benzaldehyde, squalene, and water.

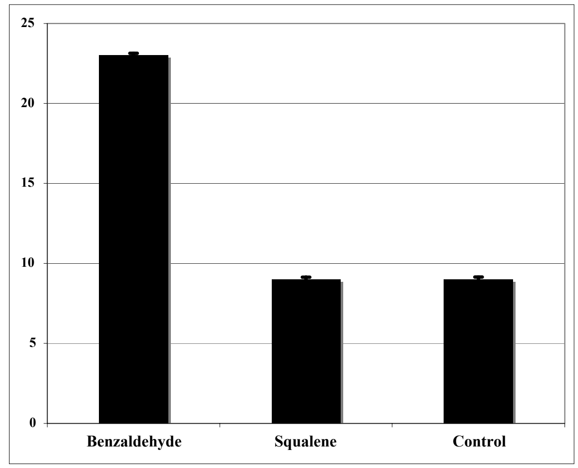



**Fig. 10.** Parasitism success of *N. pyste* attacking larvae in the three treatments.

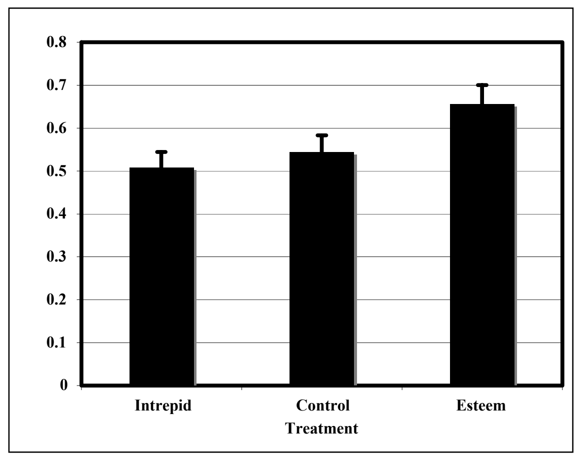
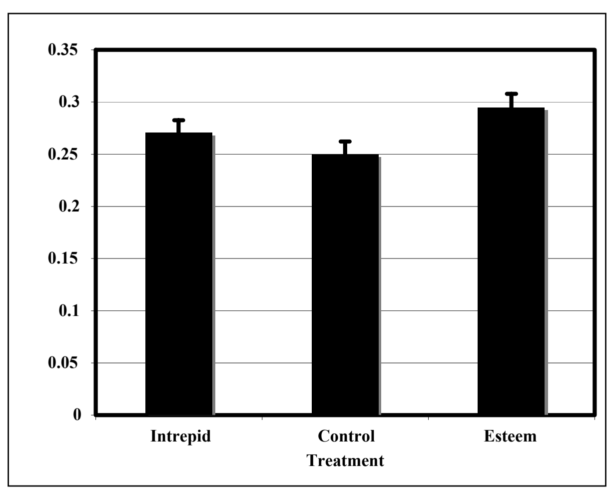
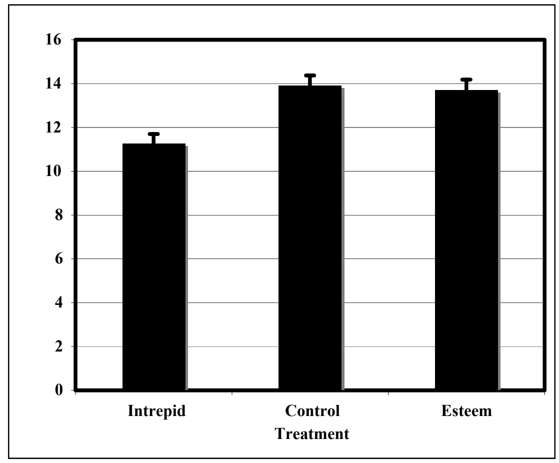



**Fig. 11.** Mass of *N. pyste* pupae reared from treated and control host larvae.



*Effects of IGR insecticides on Tachinid Parasitism.* Late applications of IGR insecticides for leafroller coincide with the time when tachinids are most common. To provide an estimate of the impact, OBLR larvae from the WSU colony were force-fed on leaf discs treated with low doses of Esteem, Intrepid, or water. These larvae were then exposed to adult *N. pyste* for parasitism and their development tracked until the emergence of the adult fly. Last year we reported primarily on the effects of Esteem; this year we are able to provide data for Intrepid, and thus a more comprehensive comparison.

**Fig. 12.** The rate of development of the endoparasitic stages of *N. pyste* in treated hosts.




In terms of parasitism success, the maggots inhabiting OBLR that had been treated with a sublethal dose of Esteem were more likely to survive to the adult stage than were their counterparts in control or Intrepid-treated larvae (Fig. 10). This suggests that treatment of host larvae with Esteem actually made these larvae more favorable hosts for the flies. One potential explanation is that the host was weakened by intoxication, compromising the immunological response against the parasitoid, or the flies may have reacted favorably to the hormone analog itself. The flies reared from Esteem and Intrepid treated hosts were also significantly larger than control flies (Fig. 11). In terms of development time within the host (endoparasitic stages), the maggots that were in hosts treated with Intrepid took longer than those in control or Esteem-treated hosts to reach the pupal stage (Fig. 12), suggesting that this compound had a negative effect on their development. The development of adult structures took significantly longer when parasitoids were reared from Esteem-treated OBLR, but flies reared from Intrepid-treated hosts took roughly the same amount of time to mature as flies reared from untreated hosts.

While our experiments demonstrate that exposure to low doses of Esteem and Intrepid in the host are not very harmful to the immature stages of *N. pyste*, the adults may be very susceptible. Hormones are important regulators of the development of sexual function in insects, so it is not surprising that exposure to compounds such as Esteem and Intrepid, which are synthetic hormone analogues, might interfere with sexual development. Fertility effects of insect growth regulators have been demonstrated in a variety of insects, and *our research report last year suggested that exposure to Esteem in the host had a negative impact on the fertility of adult flies*. Effects of Intrepid exposure on adult fertility are currently being run.

OBJECTIVE 4: DEVELOP PHENOLOGY MODELS FOR KEY NATURAL ENEMIES

Two methods are being used to obtain quantitative information on early-season predator phenology, suitable for inclusion in the WSU decision aid system: (1) weekly tray samples in orchards; (2) monitoring emergence of overwintering predators. From the tray samples, there were distinct species' differences in timing of first appearance in orchards (Fig. 13). For instance, early arriving species included some ladybird beetles (*Microwesia*, *Stethorus*, *Hippodamia*), true bugs (*Deraeocoris*), and spiders (*Philodromus*, *Sassacus*). Later-arriving species included some ladybird beetles (*Coccinella septempunctata, Harmonia axyridis*, *Coccinella transversoguttata*), several true bugs (*Anthocoris* spp., *Orius tristicolor*, *Campylomma verbasci*), and *Chrysoperla plorabunda* (a green lacewing).

| | Mar 23 | Mar 30 | Apr 6 | Apr 12 | Apr 19 | Apr 25 | May 3 | May 10 | May 17 | May 23 | May 31 | Jun 4 | Jun 8 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| LADYBIRD BEETLES | | | | | | | | | | | | | |
| *Microwesia* | ■ | ■ | ■ | ■ | ■ | ■ | ■ | ■ | ■ | ■ | ■ | ■ | |
| *Stethorus* | ■ | | | ■ | | ■ | ■ | ■ | ■ | ■ | ■ | | |
| *Hippodamia* | ■ | ■ | | | | ■ | ■ | ■ | ■ | | | | |
| *Cocc. septem.* | | | | | | ■ | | ■ | ■ | ■ | ■ | | ■ |
| *Harmonia* | | | | | | ■ | | ■ | | | | | |
| *Cocc. transv.* | | | | | | ■ | ■ | ■ | | | ■ | | ■ |
| TRUE BUG PREDATORS | | | | | | | | | | | | | |
| *Deraeocoris* | ■ | ■ | | ■ | ■ | ■ | ■ | ■ | ■ | ■ | ■ | ■ | ■ |
| *Anthocoris* | | | | | ■ | ■ | ■ | ■ | | ■ | | | |
| *Orius* | | | | | | | ■ | ■ | ■ | ■ | ■ | | |
| *Campylomma* | | | | | | | ■ | ■ | ■ | ■ | ■ | ■ | ■ |
| GREEN LACEWINGS | | | | | | | | | | | | | |
| *Chrysoperla* | | | | | | | ■ | ■ | ■ | ■ | ■ | ■ | ■ |

| | Mar 23 | Mar 30 | Apr 6 | Apr 12 | Apr 19 | Apr 25 | May 3 | May 10 | May 17 | May 23 | May 31 | Jun 4 | Jun 8 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| SPIDERS | | | | | | | | | | | | | |
| *Philodromus* | ■ | | | ■ | ■ | ■ | ■ | ■ | ■ | | ■ | | ■ |
| *Sassacus* | ■ | | ■ | | | | ■ | ■ | ■ | ■ | | | ■ |
| *Pelegrina* | | | ■ | ■ | ■ | ■ | | | ■ | ■ | ■ | | ■ |
| *Meioneta* | | | | ■ | | ■ | ■ | ■ | ■ | ■ | ■ | ■ | |
| *Xysticus* | | | | | | | | | | ■ | ■ | ■ | ■ |

**Fig. 13**. Presence (indicated by filled squares) of predators in two apple orchards as determined by beating tray samples.

For the emergence study, we have banded trees at orchards in the Wenatchee area and the Yakima area. At the Wenatchee sites, we have set up eight different sites with 40 bands per site:

1. TFREC 5 (untreated)
2. TFREC 24 (untreated areas banded)
3. Columbia View 18 (mostly commercial sprays)
4. WSU-Sunrise (Organic Reds)
5. Wenatchee Valley College orchard (organic w/virus)

6. Brewster reds (organic w/virus)
7. Orondo Goldens (Commercial)
8. Mattawa Organic Galas (Organic w/virus)

At the Yakima sites, we have banded trees at 5 orchards, with 50-100 bands per site:

1. Mike Young goldens (organic)
2. Borton reds (soft, non-bearing)
3. Moxee mix of varieties (untreated)
4. Leach golden/red (organic)
5. Brooke reds (organic)

We will collect these bands before first snowfall and hold them in screened outdoor lean-to's. In late January, we will evaluate natural enemy emergence from the bands by checking them every 2-3 days. We have a data logger recording temperature in the containers so that we can determine emergence on a degree-day scale. The beating tray data (to be continued in 2008) will be used to confirm degree day models estimated from the emergence data.

At the Wenatchee sites, we also banded 25 trees in three orchards from late-June to the end of October at weekly intervals. We recorded the number of codling moth larvae, earwigs, lacewings, spiders, ants, and other insects. Besides codling moth, earwigs were the most common in all three plots and averaged just shy of 3 earwigs per codling moth collected over all three plots throughout the season. Spiders were the next most common, but were found at roughly 1/3 the number of earwigs and lacewings were roughly 18 times less common than earwigs. These data and others suggest that earwigs need to be more seriously evaluated for their role in BC. The amount of predation on CM may be high simply because there are so many earwigs that are found in similar locations to where CM larvae spin-up, but the numbers may be inflated because earwigs have an aggregation pheromone. This would tend to make the ratio of codling moth to earwigs a misleading statistic that would overestimate the importance of earwigs.