FINAL PROJECT REPORT

Project Title: Integrated biological control of woolly apple aphid

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Other funding sources

Agency Name: Washington State Commission on Pesticide Registration

Amount awarded: 14,338 (2010); 14,792 (2009); \$12,948 (2008) (total: \$42,078)

Notes: Two WSU-BioAg grants were successfully funded and completed in a supporting research area (attraction of cover crops to syrphids, important predators of woolly apple aphid), one in 2008 for \$10,934, and another in 2010 for \$13,539 (total \$23,933). The large plot field work was enhanced by the addition of funding from the SCRI grant "Enhancing biological control to stabilize western orchard IPM systems" (Jones et al. 2008; total \$2,244,274).

Total Project Funding: Year 1: \$38,238 Year 2: \$33,962 Year 3: \$36,435

Budget History:

Item	2008	2009	2010
Salaries	11,007	11,448	11,448
Benefits	941	954	955
Wages	19,432	15,560	20,180
Benefits	1,858	2,178	1,917
Equipment	0	0	0
Supplies	2,000	1,000	1,000
Travel	3,000	2,822	935
Miscellaneous	0	0	0
Total	\$38,238	\$33,962	\$36,435

Objectives

- 1. Identify and quantify the predators and parasitoids of woolly apple aphid in eastern Washington.
- 2. Determine the relative impact of the predators and parasitoids.
- 3. Determine the effect of orchard pesticides on the key natural enemies.

Significant Findings

- Woolly apple aphid (WAA) populations started to appear on the aerial parts of the apple plants in May, with two peak population periods (July/Aug and mid-Sept/mid-October). *Aphelinus mali* was the only parasitoid species observed attacking WAA. Syrphids were the most frequently encountered predators (62.7%), followed by lacewings (23.6%) and coccinellids (8.9%). Other predators (4.8%) included nabids, *Deraeocoris*, spiders, and earwigs.
- A. mali alone caused only a minor decrease in woolly apple aphid populations; when both predators and A. mali were present, woolly apple aphid populations were controlled.
- Delegate, Guthion, Lorsban, Entrust, and Sevin caused high levels of contact mortality of
 Aphelinus mali; Rimon, Warrior, Assail and Calypso were intermediate, while Altacor,
 cyazypyr, Kumulus, Kocide+Manzate, and Ultor were nontoxic. Rimon had no deleterious
 sublethal effects on A. mali.
- Both Rimon and Delegate appear to exacerbate woolly apple aphid populations in large-scale trials when used during the first codling moth generation; the effect is worsened when the two were used together. The effect occurred both in a block with a history of woolly apple aphid problems, and one with no prior history. The same pattern occurred where the same treatments were used two years in a row, except the mid-season outbreak was higher than the previous year. Intrepid/Altacor caused the fewest secondary pest outbreaks.

Results and Discussion

Obj. 1. Woolly apple aphid natural enemy survey. Aerial colonies of woolly apple aphid and its natural enemies (NE) were monitored in 20 orchards in 2008. The colonies appeared in May, with one or two population peaks in summer and fall, respectively (Fig. 1).

The only parasitoid found in the colony samples was *Aphelinus mali*. Parasitism fluctuated considerably throughout the season with no apparent pattern.

The predator complex consisted of lacewings, lady beetles, syrphids, predatory bugs, and spiders. Overall, syrphids were the most abundant predator observed in our samples, which corresponds to previous

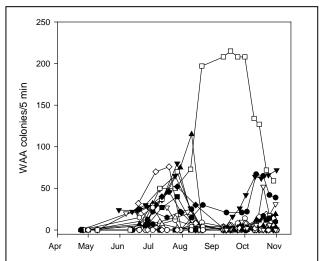


Fig. 1. Seasonal phenology of woolly apple aphid in eastern Washington, 2008.

studies in 2006 and 2007 (Fig. 2). All but one of the syrphid species identified from the 2008 study was *Eupeodes americanus* Wiedenmann; a single specimen of *Heringia calcarata* Loew was found at

an organic orchard in the Columbia Basin on July 1. The syrphid species, a specialist on woolly apple aphid, was not known to occur in Washington prior to this.

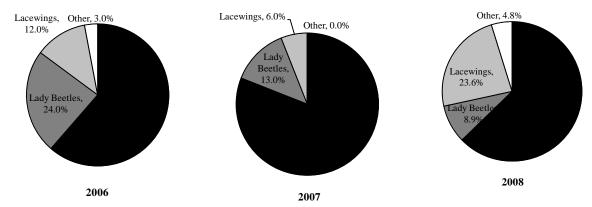


Fig. 2. Composition of woolly apple aphid predator complex, eastern Washington, 2008 (black sections are syrphids).

Obj. 2. Relative impact of the predators and parasitoids. The relative impact of predators and the parasitoid *A. mali* was demonstrated in exclusion cage studies in 2010. When both predators and parasitoids were excluded, woolly apple aphid populations continued to increase on the test trees (Fig. 3a, b). When parasitoids were allowed to attack the aphid colonies (but predators were excluded), the aphid populations increased, but at a slower rate than aphids alone. However, when both predators and parasitoids were allowed to attack the colonies, population either failed to increase (Fig. 3a) or increased slightly before declining (Fig. 3b).

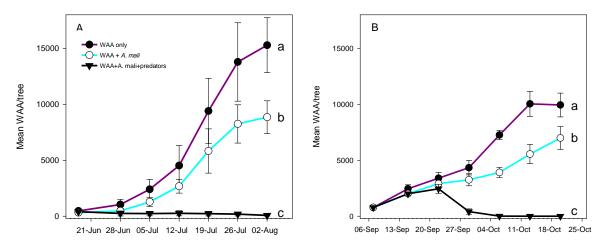


Fig. 3. Exclusion cage studies examining the effect of predators and parasitoids on population growth of woolly apple aphid. A - mid-summer; B - fall.

Obj. 3a. Laboratory tests of acute toxicity of orchard pesticides to *A. mali*. The organophosphate (Guthion, Lorsban) and carbamate (Sevin) insecticides were highly toxic to *A. mali*. Of the newer insecticides, Entrust and Delegate (spinosyns) were equally toxic (that is, close to 100% mortality at both the 1x and 0.1x rates). The neonicotinoids (Assail, Calypso) and the pyrethroid (Warrior II) were moderately to highly toxic (60-90% mortality at the 1x rate). Altacor and Cyazypyr (diamides) and Rimon (a benzoylurea chitin synthesis inhibitor) were moderately toxic (33-43% mortality at the 1x

rate). The fungicides (Kumulus, Kocide+Manzate) and Ultor (a tetramic acid derivative) were non-toxic to A. mali on contact.

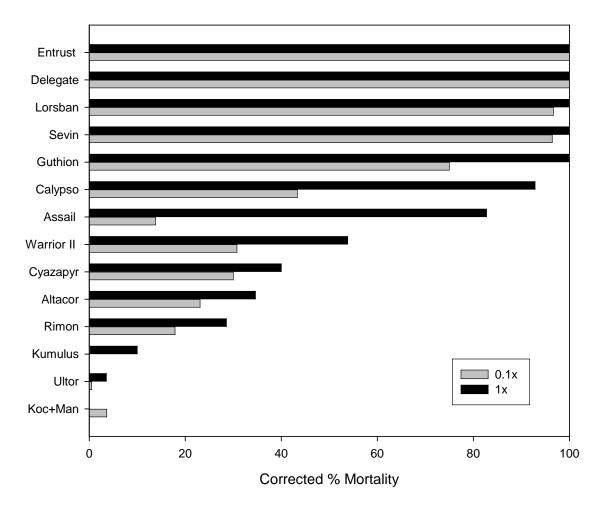


Fig. 4. Acute toxicity of the maximum label rate (1x) and 1/10 the maximum rate of various orchard pesticides on *A. mali* (Abbott's corrected mortality, 48 h).

Sublethal effects of pesticides are also under investigation; only the Rimon bioassay has been completed to date. Rimon reduced female A. mali fecundity and survivorship when compared to the control (Table 1). However, A. mali treated with Rimon produced F_1 offspring with a higher proportion of females (M:F, 0.40:0.60), which is likely why the estimate of the net reproductive rate estimate (R_0) was slightly higher for A. mali treated with Rimon. The higher number of females in the F_1 offspring may compensate for the reduced fecundity and survivorship of the Rimon-treated A. mali.

Table 1. Sublethal effects of novaluron (Rimon) on A. mali fecundity, sex ratio and net reproduction

Pesticide	Mean daily fecundity	% Mortality (Day 4)	Proportion females	R_0
Control	3.32	12.90	0.50	1.35
Rimon	2.90	24.24	0.60	1.55

Obj. 3b. Effect of seasonal program on populations of woolly apple aphid and its natural enemies. Large-scale tests of pesticide programs were conducted in two commercial orchards in central Washington, near Bridgeport ('Cameo') and Othello ('Delicious'). The blocks were 20-25 acres in size, with four treatments and four replicates of 1-1.5 acres in size. The treatments consisted of a petal fall (100 degree-days), delayed 1st and 2nd cover codling moth sprays (Table 1).

Table 2. Codling moth programs tested for effects on secondary pests and natural enemies

Trt.#	Treatment ¹ (PF, 1 st cover, 2 nd cover)	Rate/acre
1	Rimon-Delegate-Delegate	32 fl oz, 7 oz, 7 oz
2	Intrepid-Delegate-Delegate	16 fl oz, 7 oz, 7 oz
3	Rimon-Altacor-Altacor	32 fl oz, 4.5 oz, 4.5 oz
4	Intrepid-Altacor-Altacor	16 fl oz, 4.5 oz, 4.5 oz

¹Applications were made with an airblast sprayer calibrated to deliver 100 gpa.

Woolly apple aphid, green apple aphid, and spider mites were monitored from April through November, 2009, along with a broad range of natural enemies (lady beetles, syrphids, earwigs, spiders, and predatory mites). Samples were taken at 1-3 week intervals. Sample types included timed counts (both aphid species), leaf samples (mites and their predators), tap samples (various natural enemies), cardboard band traps (earwigs and spiders), and HIPV (herbivore-induced plant volatiles) attractant traps (lacewings, syrphids).

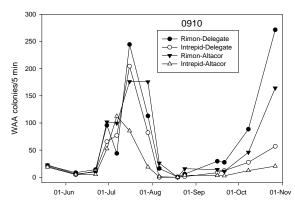


Fig. 5a. Woolly apple aphid populations, Bridgeport, 2009.

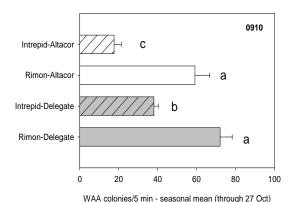


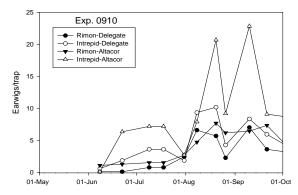
Fig. 5b. Seasonal means of woolly apple aphid timed colony counts, Bridgeport, 2009.

Bridgeport. Woolly apple aphid pressure was high in the Bridgeport plot, peaking in late July, and again in October (Fig. 5a). Significant treatment differences appeared by late June, with the Intrepid-Altacor treatment having consistently the lowest aphid densities. The highest densities occurred in the Rimon-Delegate plot, followed by the Rimon-Altacor treatment and the Intrepid-Delegate. Rimon appears to have the stronger disruptive effect on woolly apple aphid, despite the fact that it was only applied a single time at petal fall (PF). Delegate also appears to be disruptive, but somewhat less so than Rimon; when paired with Intrepid at PF, the seasonal mean aphid densities were significantly higher than when Intrepid was paired with Altacor (Fig. 5b).

Spider mite populations were moderately high in this block, peaking in late July-early August. While even the lowest treatment (Intrepid-Altacor) experienced some elevation of spider mites (ca. 11

mites/leaf), the maximum populations in the two treatments containing Rimon were significantly higher than the Intrepid-Altacor treatment on 28 July, with the Intrepid-Delegate treatment intermediate. However, this trend was reversed on the secondary peak in late August, although this may have been due to poorer leaf quality from prior mite damage. Because of this reversal, no significant differences were found in the seasonal cumulative mite days.

Earwigs were abundant in the Bridgeport plot, with densities caught in traps rising throughout the growing season, peaking in mid-September (Fig. 6a). There were clear differences among treatments by mid-June, and these differences persisted into fall. The Altacor-Intrepid treatment had the highest numbers of earwigs, while the Rimon-Delegate treatment had the fewest. Parallel to the woolly apple aphid results, Rimon and Delegate appear to be detrimental to earwigs, while Intrepid and Altacor are not (Fig. 6b). While these results are only correlations, they raise interesting questions about the role of earwigs as woolly apple aphid predators.



Rimon-Altacor

Rimon-Delegate

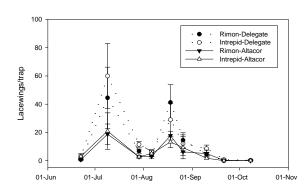
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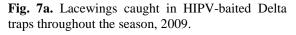
Earwigs/trap (seasonal sum) (8 Oct)

Fig. 6a. Earwig densities over time, Bridgeport, 2009.

Fig. 6b. Seasonal sums of earwigs caught in traps, Bridgeport, 2009.

The HIPV traps caught large numbers of lacewings (methyl salycilate+benzaldehyde) and syrphids (geraniol) throughout the season (data not shown). There were two distinct peaks in lacewing activity, one in early July, and the second in mid-August (Fig. 7a). There was also a treatment effect in lacewing captures, with an indication that Altacor may suppress lacewing populations (Fig. 7b). Although these insects are also important woolly apple aphid predators, there was no indication that this was the primary reason for of disruption.





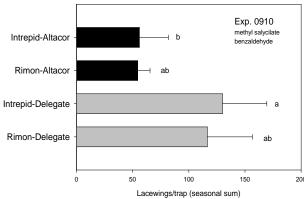
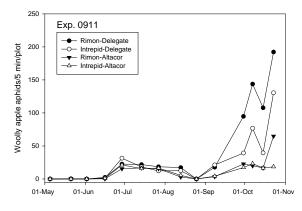


Fig. 7b. Seasonal sum of lacewing captures in Delta traps, Bridgeport, 2009.

Othello 2009. Woolly apple aphid densities were relatively low throughout the summer, but began increasing in early September, and peaking in October (Fig. 8a). Treatment effects were similar to the Bridgeport plot, in that the Rimon-Delegate treatment had significantly higher aphid densities than the other treatments. The Intrepid-Altacor treatment had the lowest levels, with the other two treatments intermediate. In this block, Delegate appeared to be slightly more disruptive than Rimon (based on the Rimon-Altacor treatment) (Fig. 8b).



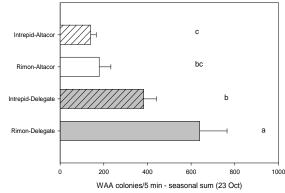


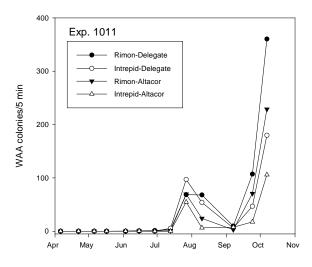
Fig. 8a. Woolly apple aphid densities, Othello, 2009.

Fig. 8b. Seasonal sums of woolly apple aphid colonies, Othello, 2009.

Earwigs and spider mite densities were very low in this plot throughout the season (data not shown); thus it is difficult to ascribe strong integrated mite control disruption to any of the products tested. Similarly, since the treatment effects on woolly apple aphid were the same in the absence of earwigs, it suggests that other mechanisms may be as much, if not more, important in the outbreak of woolly apple aphid. The other notable result from the Othello site was the trend in rust mite populations (data not shown). Both of the treatments containing Rimon had significantly depressed rust mite counts, which may contribute to disruption of integrated mite control.

Othello 2010. The pattern of WAA densities in 2010 was similar to 2009 (mid- and late-season peak) except that the mid-season peak was later than 2009 (late July instead of early July) (Fig. 9a); this may be due in part to the long, cold spring in 2010, with a substantial delay in insect and tree phenology. The most surprising aspect of this experiment was that the treatments behaved in a very similar fashion to 2009, with Rimon-Delegate having the highest populations, Intrepid-Altacor having the lowest, with the other two treatments (variations on the aforementioned materials) intermediate (Fig. 9b). Also like 2009, the treatment separation occurred in the late-season peak, months after the first generation CM sprays were applied.

Unlike 2009, there was a substantial mid-season peak of tetranychids in all treatments (Fig. 10a). In general, Intrepid-Altacor had the lowest mite levels (not exceeding five mites/leaf, although (surprisingly) there were no significant differences in the tetranychid CMDs, Fig. 10b). Predator populations were the same or higher than the pest mites, and the counts are among the highest I have ever recorded. Rust mites about the same as in 2009, with a typical late June peak.



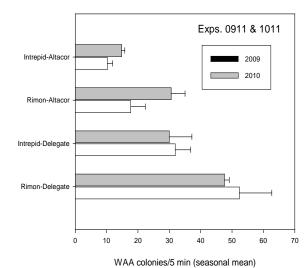
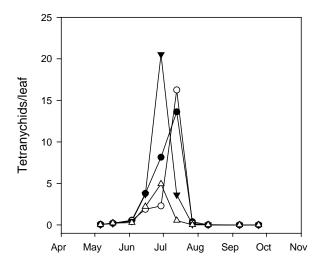
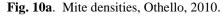


Fig. 9a. Woolly apple aphid densities, Othello, 2010.

Fig. 9b. Seasonal means of woolly apple aphid colonies, Othello, 2009 vs 2010.





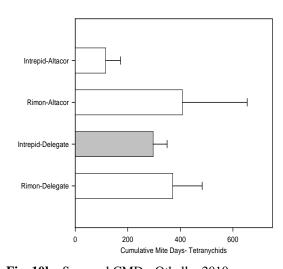


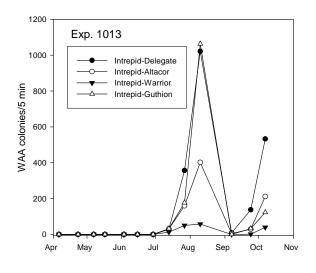
Fig. 10b. Seasonal CMDs, Othello, 2010.

Othello 2010 #2. This experiment was the first year for these treatments (Table 3). The plots were in the same block as 1011 (Othello, Bench Rd.), but 32 rows to the west. The reasoning for the treatments was to 1) take Rimon out of the PF program because it is too disruptive (replaced with all Intrepid), and re-test first generation Altacor and Delegate; 2) use Guthion as a "mite-neutral" comparison, which may also provide direct control of WAA (almost last chance before phase out!); 3) test Warrior as another possible OP replacement. All the treatments were applied first generation only; and were preceded by an oil/Lorsban in the delayed dormant. Guthion was sprayed over the entire plot second generation by the grower in response to codling moth pressure.

Table 3. Treatment schedule for large plot test, Othello #2, 2010

Trt #	Treatment (DD, PF, 1C, 2C)	Rate/acre
1	Oil+Lorsban, Intrepid, Delegate, Delegate	1.5%+4 pt, 1 pt, 7 oz, 7 oz
2	Oil+Lorsban, Intrepid, Altacor, Altacor	1.5%+4 pt, 1 pt, 4.5 oz, 4.5 oz
3	Oil+Lorsban, Intrepid, Warrior II, Warrior II	1.5%+4 pt, 1 pt, 2.56 fl oz, 2.56 fl oz
4	Oil+Lorsban, Intrepid, Guthion, Guthion	1.5%+4 pt, 1 pt, 2 lb, 2 lb

This plot produced one of the highest mid-season peaks I have yet recorded in a WAA plot, >1,000 colonies/5 min (Fig. 11a). The high populations occurred in the Delegate and Guthion plots, the Altacor was intermediate (Fig. 11b), and the lowest populations occurred in the Warrior plots. The latter comes as a surprise, since it argues that Warrior provides direct control of WAA. Another surprise is that Guthion appeared to be disruptive, and with apparently no direct toxicity; I had always assumed that all the OPs controlled/suppressed WAA, but perhaps it was really just Lorsban and Penncap). In addition to the mid-season peak, there was a secondary (fall) peak starting late September.



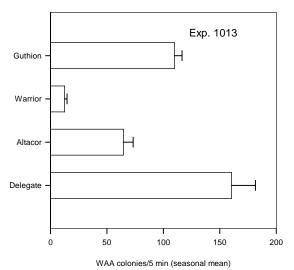


Fig. 11a. Woolly apple aphid populations, Othello #2, 2010.

Fig. 11b. Woolly apple aphid populations seasonal mean, Othello #2, 2010.

A mid-season mite outbreak occurred in all treatments, but to a lesser extent in the Intrepid-Altacor plot (data not shown). The mite population peak was tracked by the predators responding to the availability of prey. The highest predator levels occurred in the Delegate treatment, indicating that the first generation applications did not permanently prevent predator populations from developing. The most anomalous result was that the same peak occurred in the "mite-neutral" Guthion treatment, which normally causes few mite problems. For that matter, I did not necessarily expect the peak in the Warrior treatment; this material is somewhat miticidal. In addition, the rebound of predators in the Warrior treatment is a little surprising given how toxic it is in laboratory tests.

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Integrated Biological Control of Woolly Apple Aphid Executive Summary

This project provides an assessment of three key components of biological control of an important secondary pest. The first task was to discover and quantify the natural enemy complex of woolly apple aphid, which had not been done to any extent in the past. The second task was to determine the importance of the natural enemies, regardless of their abundance. Lastly, this information needed to be tested in the context of commercial orchards with routine pesticide applications, to see if pesticides were an important factor in disrupting biological control, and how that could be minimized. This systematic approach to biological control gives eastern Washington growers specific, science-based information to better manage this pest.

The first step was the quantification of natural enemies in our area – the basis for future decision-making as to which natural enemies were most critical to conserve for woolly apple aphid control. The 2008 survey was an extension of previous limited surveys done in 2006-2007, except that the number of orchards surveyed increased from 3 (in 2006-07) to 20 in 2008, making this information robust for eastern Washington. *Aphelinus mali* was the only parasitoid found in any year or orchard. Syrphids were the most commonly found predators in colonies, but lacewings and lady beetles were also found regularly. Coincident with the predator information, colony numbers were also assessed during the survey, establishing the seasonal phenology for this pest over a broad area. Most of the orchards sampled had either one or two peaks of populations, one in mid-summer, and one in fall.

The exclusion cage studies gave a clear indication that *A. mali* can reduce populations in mid-summer and fall to an extent, but cannot prevent them from increasing steadily. However, when predators were allowed to attack the colonies along with *A. mali*, the aphid populations increased only briefly, or not at all, before being driven to low levels. Where predators were allowed access, few parasitoids were recovered, probably because parasitized aphids were consumed indiscriminately along with non-parasitized ones. Thus, the choice of selective pesticides to minimize disruption should favor.

The acute toxicity bioassays of *A. mali* indicated that a number of materials, both older and newer compounds, caused high levels of adult mortality. These included Guthion, Lorsban, Sevin, Entrust and Delegate. A number of codling moth materials, Assail, Calypso, Warrior, Altacor, cyazypyr, and Rimon caused only moderate mortality, while Kumulus sulfur, Kocide+Manzate and Ultor were non-toxic in this type of exposure. Susceptibility of the lacewings and coccinellids is currently under investigation by a regional coalition of researchers working on a SCRI grant, and results should be available in about a year. Unfortunately, the key predator (syrphids) is not currently being tested, largely because they do not lend themselves to rearing; this is an information gap that needs to be addressed. In addition, the sublethal effects of pesticides on *A. mali* are in progress, with an estimated completion date of August 2011.

The large block tests are a report card on how these pesticides affect the natural enemy-pest system in the real world. In orchards with a history of woolly apple aphid pressure, Rimon and Delegate consistently caused an increase in woolly apple aphid populations, as well as tetranychid mites. Most surprising were the far-reaching effects from early season applications (petal fall through second cover). Another interesting insight is that the mechanism of disruption can vary depending on the orchard's underlying natural enemy complex; in an orchard where earwigs were abundant, the deleterious effect of Rimon was pronounced. This finding points out the strengths and weakness of different sampling methods; earwigs did not show up in predator surveys because they are primarily nocturnal feeders. Another interesting finding was the low levels of woolly apple aphid in the Warrior blocks indicating some level of direct control; this also merits further investigation. Based on multiple large-plot tests, it appears that Intrepid (PF) and Altacor (first generation CM) is the least disruptive combination to woolly apple aphid. This corresponds to a low toxicity rating in bioassays of acute and sublethal toxicity.