FINAL PROJECT REPORT

Project Title: Tests of a sprayable pheromone formulation against winterform psylla

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Other funding sources

Agency Name: Western Region IPM Grants Program Amount awarded: \$23,844

Total Project Funding: \$19,000

Budget History:

Item	2014	2015
Salaries	\$11,250	\$1,500
Benefits	\$ 3,750	\$ 500
Plot Fees ¹	\$ 1,000	\$1,000
Total	\$16,000	\$3,000
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¹ Pruning, herbicide, horticultural oil

OBJECTIVE:

Develop and test a sprayable formulation of pheromone

- 1. Confirm pheromone retains activity in oil
- 2. Disruption trials
 - a. Large cage study
 - b. Large plot study

SIGNIFICANT FINDINGS:

- Trapping studies indicated that the pheromone applied as a sprayable in 1% horticultural oil retained its attractiveness to male winterform psylla.
- Large cage study showed that treatment of potted pear trees with sprayable formulation led to *higher* mating rates by females rather than a hoped-for lowered mating rate.
- Large replicated field plot trials in two years confirmed the large cage study that the sprayable pheromone formulation led to enhanced mating rates.
 - Hatch rates of field-collected eggs were similar in pheromone plots and control plots, as expected given absence of mating disruption.

RESULTS AND DISCUSSION:

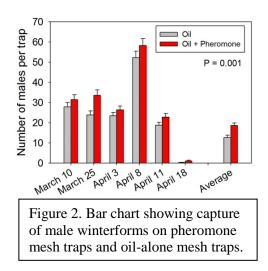
1. Confirm pheromone retains activity in oil

Methods. Paired limbs (each 2-3 foot in length) were drenched with oil (1% in water) + pheromone or 1% oil, and then enclosed in a clear sticky mesh to trap male psylla (Fig. 1). Solutions were applied to drip using a salad spritzer. I prevented overspray from contaminating non-target parts of the tree by collecting overspray with a towel held behind target limbs. I estimate each oil + pheromone limb to have received about 50 female equivalents of spray (some of which was lost to overspray). Traps were collected after 3 days in the field. The trial was done on 6 dates during the winterform generation.

Results. Male winterforms preferentially accumulated on the oil + pheromone traps compared to the oil-alone traps (Fig. 2). These results indicate that the pheromone retained its modest volatility and attractiveness to males even when applied in 1% horticultural oil + water.



Figure 1. Mesh traps encircling oil + pheromone shoots or oil-alone shoots.



2a. Large cage study to examine effects of sprayable pheromone on mating

Methods. Cage studies were conducted out-of-doors with lab-reared (virgin) summerforms, using two large ($6 \times 6 \times 6$) ventilated cages (Fig. 3: photographs). Two fully leaved potted trees (approximately 4 foot in height) were set in the center of each cage and misted with either 1% oil in water, or with the pheromone solution (1% oil in water + pheromone; volume of solution approximately equal to 200 female equivalents). To both cages were added 100 females, followed 24 hours later with 100 males. After 48 hours, 50 females from both cages were collected, and then dissected to determine number of matings.

Results. Results showed that probability of having been mated was actually higher in the pheromonetreatment than the control treatment (Fig. 3: bar graph). Thus, there is evidence that contact with the pheromone in 1% oil actually led to enhanced male success at locating and mating female psylla.



Figure 3. Cage study showing pear trees (2 per cage) and psyllid release vial. Bar chart shows percentage of females that were mated in oil alone trees (gray bars) and oil + pheromone trees (dark bars).

2b. Large plot trials to look for disruption of winterform mating

Methods. Twelve plots (each 16 trees in size) were set out at the Moxee research farm in February 2014 and 2015; six plots were designated oil + pheromone, and six were control (oil alone) plots (see Fig. 4 for design). Psylla were collected at approximately weekly intervals beginning in early February and dissected to determine onset of ovarian development. Once the first mature eggs were seen in dissected females and before mating had begun, plots were sprayed with the two solutions (early March both years). Each plot received approximately 4 gallons of solution applied through a 25 gallon weed sprayer (Scorpion Sprayer) attached to a 4 wheeler (Fig. 5). Tray samples were taken 1 week following application to determine adult densities and sex ratios. At 1 week following application, 40-50 females were collected from each plot and dissected to determine number of matings. Once eggs were seen in the field, spurs (enough to provide data on at least 100 eggs) were collected from each plot for monitoring egg fertility. Eggs were counted on each spur, cut ends of spurs were placed in water, and spurs were re-examined 1 week later to determine hatch rates (Fig. 6).

Results. Probability of being mated was higher both years in pheromone plots than control plots, a result consistent with results using the sprayable formulation in my large cage trials (Fig. 7). At this

time, I cannot explain the increased mating rates in the pheromone plots. Sex ratios were the same in control and pheromone plots, thus there was no evidence that enhanced mating in the pheromone plots compared to control plots was due to a higher ratio of males to females in the pheromone plots. One possible explanation for the enhanced rates of mating in pheromone plots is that presence of pheromone stimulated males to increase their search efforts on treated trees, leading to increased rates of contact with females. However, controlled behavioral trials will be needed to examine this hypothesis. Given the absence of mating disruption, I unsurprisingly found no hoped-for reductions of hatch rates of eggs in pheromone plots (Fig. 8).

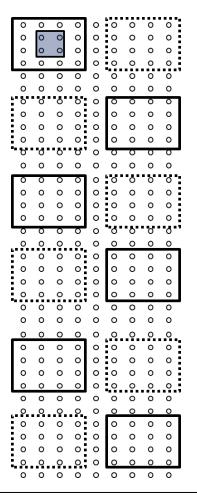


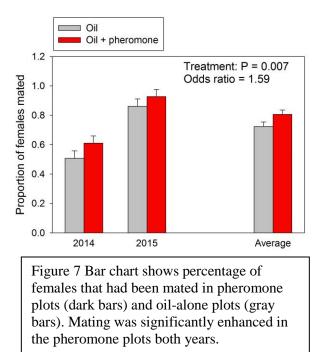
Figure 4. Design of large plot trial showing f oil-alone plots (solid line rectangles) and oil + pheromone plots (dashed line rectangles). Small square in one plot shows the 4 trees from which females were collected for dissection.

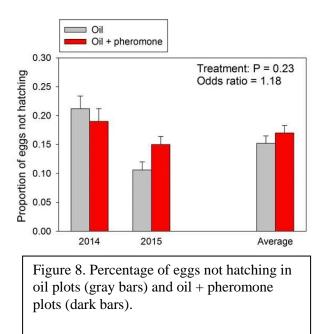


Figure 5. Applying oil + pheromone solution.



Figure 6. Field-collected spurs being monitored for hatch of psylla eggs.





EXECUTIVE SUMMARY

Management of pear psylla requires some level of control of the post-wintering winterform generation. Pear psylla females overwinter in an unmated condition, and strategies that could be used to delay mating, leading to delays in egglaying combined with the production of early-season infertile eggs, would be useful. The sex pheromone of pear psylla is unfortunately highly non-volatile, making it logistically infeasible to saturate pear orchards by dispensing the compound through traditional dispensers. However, the compound is a hydrocarbon and is thus fully soluble in the horticultural oils used during the delayed dormant period in commercial orchards. This study examined whether using a sprayable, to saturate orchard with pheromone, would interfere with the male's ability to find unmated winterform females during the early post-winter generation.

A trapping study showed that the sprayable formulation is attractive to mate-seeking male winterforms, indicating that the pheromone retains its attractiveness when mixed with horticultural oil. However, cage studies and large field-plot trials failed to demonstrate that mating success declined on trees or in plots fully saturated with the sprayable formulation. Indeed, I observed the opposite effect in both types of trials: the sprayable formulation, when applied to whole-trees or to multi-tree plots, led to *higher* rates of mating than observed in control (oil alone) plots. The reasons for this response are not clear. One hypothesis is that the compound stimulated increased mate-searching activities of the male psyllids, possibly including an increase in acoustic signaling by males, which we now know (from a 2015 publication) is used by males of a very closely related pear psyllid to locate females for mating. This hypothesis merits attention, as disruption of mating through saturation of the environment with synthetically produced acoustic signals has been shown to reduce mating in acoustic-signally leafhoppers and in another pest psyllid (Asian citrus psyllid).