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Large-plot field studies to assess impacts of newer insecticides on non-target arthropods in Western U.S. orchards

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Running head: P.W. Shearer et al. Testing impacts of reduced risk insecticide in orchards

Abstract

The non-target impacts of two reduced risk insecticides, chlorantraniliprole and spinetoram, were evaluated for two years in Oregon pear and California walnut orchards. Experiments were conducted in large replicated plots (approximately 0.25-0.4 ha) to assess the impact of these two insecticides on natural enemies of secondary pests when applied against codling moth, *Cydia pomonella*. Cumulative insect days (CID) of secondary pests and natural enemies were calculated from leaf samples, plant volatile traps, beat trays or cardboard trunk bands. Ratios of natural enemies and prey were also calculated. Results from these field studies demonstrate that applications of chlorantraniliprole can reduce abundance of predatory Neuroptera and that spinetoram negatively impacts parasitic Hymenoptera. However, these trends did not always occur each year. As a percentage among all trials within a crop, there were more treatment differences for natural enemy/prey ratios (50 and 33% for pears and walnut plots, respectively) than for natural enemy CIDs (25 and 13% for pears and walnut plots, respectively). It is likely that unseasonably cool weather during the two years of this study impacted both pest and natural enemy abundance. The intrinsic value of large-plot field studies is discussed.

Key words biological control; disruption; reduced risk insecticide; integrated pest management; natural enemy; secondary pest

1. Introduction

Integrated pest management (IPM) programs in orchards have evolved rapidly in the past 20 years in large part because the Food Quality Protection Act (FQPA) (US EPA, 1996) changed the suite of insecticides available to growers (Agnello et al., 2009; Jones et al, 2009, 2010). Of all the organophosphorus insecticides replaced by FQPA, the loss of azinphosmethyl was of greatest concern to IPM practitioners because it had been used regularly in tree fruit for control of codling moth, *Cydia pomonella* (L.) (Lep.: Tortricidae), since the 1950s (Whalon et al., 1999).

To fill the void left after the removal of azinphosmethyl, IPM practitioners began testing a suite of “reduced risk” pesticides as replacements (Viray and Hollingsworth, 2009). These new insecticides were often assumed to be less harmful to natural enemies because of results from previous field studies (Agnello et al., 2009; Atanassov et al., 2002; Roubos et al., 2014). However, laboratory studies have demonstrated detrimental effects of reduced risk insecticides on key natural enemies (Amarasekare and Shearer, 2013a,b; Kim et al., 2006), including studies presented in this issue (Amarasekare et al., this issue; Beers et al., this issue a,b; Mills et al., this issue a). Interestingly, the safety of these products to natural enemies is not a specific criterion used by the United States Environmental Protection Agency (EPA) to define this group of pesticides (National Research Council, 2000). Pesticides can be classified as reduced risk if they meet at least one of nine criteria including, but not limited to, reduced impact on human health, replacement of chemicals that pose health risks to workers, reduced effects on non-target organisms, or presumed compatibility with IPM.

While IPM has been practiced for more than 50 years (Stern et al., 1959), the foundation of tree fruit IPM was laid by Hoyt (1969), who developed a program that integrated chemical control of insects (primarily codling moth), with the use of conservation biological control to

73 suppress phytophagous mites. Hoyt demonstrated that proper choice of insecticides, rates, and
74 application methods, allowed *Galendromus* [= *Typhlodromus*] *occidentalis* (Nesbitt) (Acari:
75 Phytoseiidae) to survive and suppress populations of the McDaniel spider mite, *Tetranychus*
76 *mcdanieli* McGregor (Acari.: Tetranychidae), and the European red mite, *Panonychus ulmi*
77 (Koch) (Acari: Tetranychidae) in Washington apple orchards. The methods outlined by Hoyt
78 (1969) have been adapted to IPM programs on tree fruits around the world (Apple and Smith,
79 1976).

80 Orchard IPM programs include the use of pesticides and their selection should be based on
81 efficacy against target pests and safety towards natural enemies. Early examples of successful
82 orchard IPM programs involved applications of pesticides that did not significantly harm natural
83 enemies of phytophagous mite pests in apple (Croft and Soloman, 1981). Most of those early
84 examples were based upon physiological selectivity when selective miticides killed targeted
85 mites but not predatory mites. Later, several predator mite species evolved resistance to key
86 insecticides including azinphosmethyl; a result of its long history of seasonal use against primary
87 apple pests (Croft and Hoyt, 1978). Other natural enemies, including *Pnigalio flavipes*
88 (Ashmead) (Hym.: Eulophidae), a parasitoid of the leafminer, *Phyllonorycter elmaella* (Doganlar
89 and Mutuura) (Lep.: Gracillariidae) (Barrett and Brunner, 1990) and *Trioxys pallidus* (Haliday)
90 (Hym.: Braconidae), a parasitoid of walnut aphid, *Chromaphis juglandicola* (Kaltenbach) (Hem.:
91 Aphididae), have also developed resistance after repeated long-term exposure to azinphosmethyl
92 (Hoy and Cave, 1989). More recently, insecticide resistance management tactics have been
93 incorporated into many new insecticide labels to delay resistance development in primary pests.
94 These refined use patterns and resistance management tactics will likely prevent natural enemy
95 populations from developing natural field resistance (Jones et al., 2009).

Now that azinphosmethyl and several other organophosphorus insecticides have been removed from use and replaced by newer insecticides, growers and pest control advisors need more information on how to use them. Despite their efficacy against primary pests, some of these newer insecticides have been shown in the laboratory to have lethal and sub lethal effects on key natural enemies. In those studies, spinetoram was more toxic to parasitic Hymenoptera than chlorantraniliprole (Amarasekare et al., this issue; Beers et al., this issue a,b; Beers and Schmidt, 2014; Mills et al., this issue a), while chlorantraniliprole had a greater negative impact on predatory Neuroptera than did spinetoram (Amarasekare and Shearer, 2013 b; Amarasekare et al., this issue). While laboratory studies provide useful knowledge about potential impacts of pesticides on natural enemies, field studies are necessary to verify whether similar effects occur under orchard conditions. That is the purpose of this study.

This study, in addition to a similar study conducted in apple orchards (Beers et al., this issue), was part of a comprehensive USDA-NIFA Specialty Crops Research Initiative effort to enhance biological control in western orchards (Jones et al., this issue). Our study focused on whether applications of select reduced risk insecticides that were targeted for *C. pomonella* management caused outbreaks of one or more secondary pests by disrupting natural enemies in pear orchards in Oregon and walnut orchards in California.

2. Materials and methods

2.1. Site descriptions and experimental treatments

Studies to assess the impact of reduced-risk insecticides, applied against *C. pomonella*, on the abundance of key natural enemies and secondary pests found in pear and walnut orchards were conducted during 2010-11. Pear orchards were located in Hood River, OR while the walnut sites were located in Hamilton City, CA.

In OR, field trials were set up using a randomized complete block design in both years. The 2010 study was conducted in a 2.4 ha planting of mature ‘D’Anjou’ pear trees interplanted with ‘Bartlett’ pear pollinizers with a tree spacing of 4.6×7.9 m. This pear orchard had for the previous three years been treated with mating disruption for *C. pomonella*, but was not under mating disruption during the course of this study. The experiment was set up with three treatments and three replicate blocks with insecticide treatments applied to plots of approximately 0.27 ha in size in each block. In 2011, a second study was set up in a different orchard. This 2.2 ha planting of mature ‘D’Anjou’ pear trees with ‘Bartlett’ pear pollinizers, with a tree spacing of 3.0×6.0 m, was divided into 4 replicate blocks and each block contained two treatments. Individual plots were approximately 0.28 ha in size. This orchard was in its sixth year of using codling moth mating disruption during our study.

Insecticides used in the pear studies were timed for early egg hatch of first-generation *C. pomonella* (Brunner et al. 1987) and applied using a Rears Pack Tank research sprayer (Rears Manufacturing, Eugene, OR) delivering 935 L/ha. In 2010, the three experimental treatments (and application rate/ha) consisted of chlorantraniliprole (Altacor[®] 35 WDG, 315 g/ha, DuPont Crop Protection, Wilmington, Delaware, USA), spinetoram (Delegate[®] 25 WG, 490 g/ha, Dow Agro Sciences LLC, Indianapolis, Indiana, USA) and cyantraniliprole (Exirel[®] 100 g [AI] SE, 1.242 L/ha, DuPont Crop Protection, Wilmington, Delaware, USA) and were applied 16 Jun (217 DD after biofix) and 15 days later on 1 Jul 2010. One application of methoxyfenozide

(Intrepid 2F 877 ml/ha) was applied against second-generation codling moth on 3 Aug (990 DD) across all plots. In 2011, the second study, which was set up in a different 2.2 ha planting of mature 'D'Anjou' pear trees with 'Bartlett' pear pollinizers, was divided into 4 blocks and each block contained two treatments. Individual plots were approximately 0.28 ha. The experimental treatments consisted of chlorantraniliprole (315 g/ha) and spinetoram (490 g/ha) and were applied 17 Jun (231 DD) and 23 days later on 10 Jul 2011. Trees were planted on 3×6 m grid. Both pear orchards received standard disease and insecticide sprays from dormant through shortly after petal-fall.

In CA, three field trials were conducted as randomized complete block designs. In 2010, a single field trial was conducted in a 10.12 ha orchard of 'Vina' walnuts, with a tree spacing of 6.1 × 6.1m, to compare three experimental treatments; two insecticides targeting codling moth and a no-insecticide control. The three treatments were applied to 0.4 ha plots in each of four replicate blocks in one half of the orchard. In 2011, two trials were conducted, the first in the other half of the same 10.12 ha 'Vina' orchard used in 2010 (orchard A), and the second in part of a 20.23 ha orchard with a 7.6 × 7.6m tree spacing of 'Serr' walnuts (orchard B). In orchard A we compared three treatments (two insecticides plus a control) in 0.4 ha plots in four replicate blocks, and in orchard B we compared four treatments (three insecticide combinations plus a control) in 0.61 ha plots in three replicate blocks. All three orchards used codling moth mating disruption and applications of a combination of mancozeb plus copper hydroxide for control of walnut blight, *Xanthomonas campestris* pv. *juglandis*, early in the season.

Insecticide treatments were timed for egg deposition or larval hatch of codling moth in the first two generations and were applied using grower-operated speed sprayers delivering a volume of 935L/ha. In 2010, the two insecticides used were spinetoram (Delegate® 25 WG, 448

g/ha) applied at 650 DD after biofix of the first generation (1B flight) and 300 DD after biofix for the second generation (2A flight), and chlorantraniliprole (Altacor[®] 35 WDG, 280 g/ha) applied at 500 DD after biofix of the first generation (1B flight) and 150 DD after biofix for the second generation (2A flight). For orchard A in 2011, the two insecticides were chlorantraniliprole (Altacor[®] 35 WDG, 263 g/ha) and chlorantraniliprole combined with lambda-cyhalothrin (Voliam Xpress[®], 876 g/ha, Syngenta Crop Protection, LLC, Greensboro, NC), both applied at 500 DD after biofix of the first generation (1B flight) and 150 DD after biofix for the second generation (2A flight). For orchard B in 2011, four applications were made in total, timed at 200 DD and 550 DD after biofix for both generations. One insecticide combination consisted of half rates of spinetoram applied twice in the first generation and half rates of chlorantraniliprole applied twice in the second generation, and a second insecticide combination consisted of the reverse (chlorantraniliprole applications followed by spinetoram applications). A third insecticide combination, the grower standard, consisted of two applications of lambda-cyhalothrin (Warrior II[®], Syngenta Crop Protection, LLC, Greensboro, NC), 183 ml/ha) in the first generation followed by one application of chlorpyrifos (Warhawk[®], 4.7L/ha, Loveland Products, Greeley, CO) in the second generation (200DD).

2.2. Secondary pest monitoring

In pear, *Cacopsylla pyricola* (Förster) (Hemiptera: Psyllidae) nymphs were sampled weekly from ‘D’Anjou’ trees by collecting 180 or 240 leaves per treatment (3 leaves [basal, medial and distal] per terminal shoot × 2 terminal shoots per tree × 10 trees per plot × 3 or 4 plots/treatment). Leaves were brought back to the lab in coolers and *C. pyricola* nymphs were

counted using a stereo-microscope. Sampling started in early-mid June and ended in September both years.

In the walnut system, several secondary pests were monitored including walnut aphid, *C. juglandicola*, dusky-veined aphid, *Panaphis juglandis* (Goeze) (Hemiptera: Aphididae), twospotted spider mite, *Tetranychus urticae* Koch (Acar.: Tetranychidae) and European red mite *P. ulmi*. These were sampled every two weeks by collecting 45 or 60 leaves per treatment (three compound leaves per tree $\times$ 5 trees per plot $\times$ 3 or 4 plots/treatment). The two aphid species were first counted directly in the field before the leaves were placed in coolers and returned to the laboratory where they were brushed to give a single count per plot for the two mite species. Collections started in early May and continued until the end of September in both years.

2.3. Natural enemy monitoring

Natural enemy abundance was monitored several different ways. Natural enemies were counted and collected in pear orchards during weekly beat tray sampling and collections of corrugated cardboard bands placed around tree trunks. In walnuts, natural enemies were sampled from leaves collected every two weeks. Abundance of natural enemies were also estimated from sticky traps baited with one of several plant volatile lures (PV) deployed in both crops.

In pears, *Trechnites* spp. (Hym.: Encyrtidae; a parasitoid of pear psylla), predatory Heteroptera (including adult and immature *Campylomma verbasi* (Meyer), *Deraeocoris brevis piceatus* (Knight) (Hem.: Miridae), *Geocoris* spp. (Hem.: Lygaeidae), and spiders (Araneae)

were monitored weekly with 15 beat tray samples (Burts and Retan, 1973) per plot (45 trays per treatment in 2010; 60 trays per treatment in 2011).

Cardboard bands were deployed in pears to measure the abundance of European earwigs, *Forficula auricularia* Linnaeus (Derm.: Forficulidae). They were made from 7.6 cm wide corrugated wrap (Model S-11450, ULINE, Chicago, IL) cut into 3.8 cm wide strips. These bands, containing one smooth side and one corrugated side, were wrapped around pear tree trunks with the smooth side out and fastened to the bark with 1 cm deep staples. Fifteen cardboard bands per plot ($n = 45$ per treatment) were deployed in 2010 and 10 bands per plot were deployed in 2011 ($n = 40$ per treatment). Bands were replaced at weekly intervals. The cardboard bands were brought back to the laboratory in coolers and the bands from each plot were placed into a large ($33 \times 33 \times 53$ cm) plastic container where they were misted with water from a spray bottle. The water relaxed the glue so the two layers of cardboard could be pulled apart for inspection. The abundance of *F. auricularia* captured in the bands were pooled per plot and recorded.

In walnuts, *C. juglandicola* mummies parasitized by *T. pallidus* were counted directly in the field from the leaf samples collected for counts of aphids (see above). Phytoseiid mites were assessed at the same time as the tetranychid mites using the mite brushing machine as previously discussed.

Plant volatile (PV) traps were used to assess abundance of adult predatory Neuroptera (primarily *Chrysoperla* spp. and some Hemerobiidae), predatory Syrphidae (primarily Syrphinae) and parasitic Hymenoptera in the various treatments in both crops. Large white plastic delta traps with sticky liners (Suterra, Bend, OR) were used in 2010 and white or yellow sticky cards (Alpha Scents Inc., West Linn, OR) in 2011. The traps were constructed and

deployed as described by Jones et al. (this issue a). In 2010, the traps were baited with one of four synthetic plant volatile blends, 1) GMP [geraniol (Sigma-Aldrich Corp. St. Louis, MO), + methyl salicylate (Sigma-Aldrich Corp. St. Louis, MO) + 2-phenylethanol (Sigma-Aldrich Corp. St. Louis, MO)] with components in separate dispensers, 2) acetophenone (Fisher Scientific, Pittsburg PA), 3) squalene (Sigma–Aldrich, St. Louis, MO) or 4) phenylacetaldehyde (Fisher Scientific, Pittsburg PA). In 2011, two trap types were deployed, white sticky cards with the GMP lure and a blank yellow sticky card. In both crops and years, one of each type of trap was deployed in each replicate plot. The sticky cards were replaced weekly in pears and every two weeks in walnuts then covered with clear Saran™ wrap (S. C. Johnson & Son, Inc. Racine, WI) and frozen at -10°C until natural enemy taxa could be identified. Lures were changed monthly and traps were changed and rotated to adjacent trees when serviced. Numbers of captured target insects were pooled across trap types within each replicate plot.

2.4. Data analysis

The monitoring data from the orchards were summarized each year using cumulative insect-days (CID) to provide a season-long estimate of the potential for secondary pest damage and biological control by natural enemies (Jones and Parrella 1983, Ruppel 1983). CIDs were estimated as the average population density between two consecutive sampling dates multiplied by the number of intervening days and summed over the entire sampling period:

$$CID = \sum 0.5(P_a + P_b)D_{a-b}$$

where P_a is the population density (mean arthropods/per unit sampled) at time a , P_b is the population density at time b , and D_{a-b} is the number of days between times a and b . For

clarification, seasonal abundance of phytophagous and predatory mites were estimated using the same formula above and referred to as cumulative mite-days (CMD).

A season-long estimate of the percent parasitism of walnut aphids was obtained by dividing the cumulative number of *C. juglandicola* mummies by the combined cumulative numbers of *C. juglandicola* and mummies. Predator/prey ratios were calculated by dividing the CID (or CMD) of a particular natural enemy by the CID (or CMD) of a particular prey.

Cumulative insect-days, CMD, and predator/prey ratios were analyzed using the Statistical Analysis System (SAS 2014). PROC GLIMMIX was used to conduct generalized linear mixed effects models, using insecticide treatments as the fixed effect and replicate blocks as a random effect. CID and CMD data were log transformed to meet the assumptions of normality. Percent parasitism was analyzed similarly, but using a binomial distribution and non-transformed data. Treatment means were separated using pairwise comparisons of least-squares means ($P \leq 0.05$).

3. Results

3.1. Secondary pest monitoring

Densities of *C. pyricola* nymphs per pear leaf were extremely low in both pear orchards (maximum on any one sample date was < 0.4 and 0.25 nymphs per leaf in 2010 and 2011, respectively) and supplemental summer sprays were not required. There were no statistical differences in the average CID for *C. pyricola* nymphs between insecticide treatments in either year. In 2010, *C. pyricola* nymph CID levels were 17.7 ± 5.0 , 17.6 ± 0.8 , and 13.6 ± 1.7 for chlorantraniliprole, cyantraniliprole and spinetoram, respectively ($F = 0.22$, $df = 2, 4$, $P = 0.82$).

Similarly, in 2011 there were no statistical differences in *C. pyricola* nymph CIDs between the chlorantraniliprole and spinetoram treatments, which were 6.3 ± 1.5 and 5.3 ± 2.1 , respectively ($F = 0.98$, $df = 1, 3$, $P = 0.40$).

In the 2010 walnut study, *C. juglandicola* CIDs were higher in the spinetoram treatment than in the other two treatments (Table 1). There were no treatment differences in CIDs for *P. juglandis*, *T. urticae* or *P. ulmi*. For orchard A in 2011, both the lambda-cyhalothrin plus chlorantraniliprole and control treatments had higher secondary pest CIDs than the chlorantraniliprole treatment. For orchard B in 2011, the plots treated with spinetoram for first generation *C. pomonella* had the highest CID for *C. juglandicola*, compared with other treatments, while the grower standard plots had the lowest CID for *T. urticae*. *Panaphis juglandis* was not observed in any of the walnut plots in 2011.

3.2. Natural enemy monitoring

Beat tray sampling in the pear orchard in 2010 revealed higher CIDs for *Trechnites* spp. in the chlorantraniliprole and cyantraniliprole treatments than in the spinetoram treatment (Table 2). In 2011, the difference between the chlorantraniliprole and spinetoram treatments was similar to that observed in 2010 for the *Trechnites* spp. CIDs, but was not significant. There were no treatment differences in the beat tray CIDs for predatory Heteroptera or Araneae in either year. In 2010, CIDs for predatory Neuroptera captured on PV traps in pear was highest in plots treated with spinetoram and significantly lower in the plots treated with chlorantraniliprole and cyantraniliprole (Table 2). While the pattern was similar in 2011, the effect was not significant. There were no treatment differences in the CIDs for predatory Syrphidae in either year.

301 In both years, *F. auricularia* was abundant in the cardboard bands in the pear orchards. In
302 2010, CIDs for *F. auricularia* were highest in plots treated with cyantraniliprole, and
303 significantly lower in plots treated with chlorantraniliprole or spinetoram (Table 2). There were
304 no significant treatment differences in *F. auricularia* CIDs in 2011.

305 For the walnut orchards there were no differences in the CIDs for *C. juglandicola*
306 mummies between treatments in either year (Table 3). In 2010, the insecticide treatments had no
307 effect on phytoseiid mite CMDs, but in 2011, the CMDs were highest in the chlorantraniliprole
308 treated plots in orchard A and in both the control plots and those that were treated with
309 chlorantraniliprole first followed by spinetoram second in orchard B. In the walnut orchards,
310 there were no differences between treatments in the CIDs for the natural enemies captured on the
311 PV traps in either year.

312 313 3.3. Natural enemy/prey ratios 314

315 There were some statistical differences in natural enemy/prey ratios between treatments in
316 the pear orchards. In 2010, the ratio of *Trechmites* spp./*C. pyricola* CID's was highest in the
317 chlorantraniliprole treatment and lowest in the spinetoram treatment (Table 4). In contrast, the
318 ratio of predatory Neuroptera/*C. pyricola* CIDs were significantly higher in the spinetoram
319 treatment than the cyantraniliprole treatment in 2011. While the same trends between treatments
320 were observed in the other year of the trials for both of these ratios the effects were not
321 significant. There were no treatment differences in ratios of predatory Heteroptera/*C. pyricola*
322 CIDs in either year.

In the walnut orchards, there was a significant treatment effect on percent parasitism of *C. juglandicola* by *T. pallidus* in 2010, and plots treated with spinetoram had the lowest levels of parasitism (Table 5), but there were no treatment differences for percent parasitism in the two walnut orchards used in 2011. In contrast, there were no treatment differences in predator/prey ratios for phytoseiid and tetranychid mites in 2010, but there were in 2011. In 2011, CMD ratios for phytoseiid mites and either *T. urticae* or *P. ulmi* were higher in the chlorantraniliprole treated plots than in the control or lambda-cyhalothrin plus chlorantraniliprole treated plots for orchard A. However, there were no treatment differences between the CMD ratios for phytoseiid and tetranychid mites in orchard B.

4. Discussion

Results from these field studies did not show consistent negative effects of chlorantraniliprole or spinetoram on secondary pests and natural enemies when they were applied to manage *C. pomonella* in pear and walnut orchards. Overall, there was a general lack of year-to-year response to the insecticide treatments for most of the sampled taxa making it difficult to demonstrate consistent disruption of natural enemies and increases in secondary pest abundance. The inconsistency in these results is similar to that observed in some of the field studies in apple orchards described by Beers et al. (this issue, a,b). There were two key exceptions. In walnuts, higher levels of *C. juglandicola* abundance were observed in the lambda-cyhalothrin and spinetoram (either applied both generations or applied first generation followed by chlorantraniliprole for second generation) treatments and this was accompanied by lower levels of percent parasitism in the case of the spinetoram treatment in 2010. In pears, the

ratio of predatory Neuroptera adults (primarily *Chrysoperla* spp.) to *P. pyricola* nymphs was greater in the spinetoram versus the chlorantraniliprole treatments in both years, although the ratio was significant only in the first year. For all other taxa, if treatment differences occurred in one year, they didn't appear the other year. It is likely that two unseasonably cool summers in the western US was a significant factor that contributed to low levels of secondary pest and natural enemy abundance in both the pear and walnut orchards which may have prevented the detection of more consistent treatment effects.

Chlorantraniliprole and spinetoram were the two reduced-risk insecticides that were compared in pear and walnut orchards in both years. In related laboratory bioassays, Amarasekare et al. (this issue) determined that chlorantraniliprole was more detrimental to *Chrysoperla carnea* (Stephens) (Neur.: Chrysopidae) than spinetoram, but that spinetoram was more detrimental to *T. pallidus* than chlorantraniliprole. This was evident from a reduction in the extrapolated intrinsic rates of population increase to negative values in both cases. Although the field study results were not as clear as those from the laboratory bioassays for these two natural enemies, they do provide at least partial verification that strong laboratory effects do translate to the field.

However, in general, many of the effects seen from natural enemy exposure to insecticides in laboratory bioassays were not observed in our field studies, and this is thoroughly described elsewhere in this issue (Beers et al., this issue b). For example, both *D. brevis* and *G. occidentalis* were shown to be much more susceptible to spinetoram than to chlorantraniliprole in laboratory bioassays (Amarasekare and Shearer, 2013a; Beers and Schmidt, 2014). However, in our field study, there were no significant treatment differences in predatory Heteroptera CIDs collected with beat trays in pear in 2010 and only slightly lower numbers were observed in the

spinetoram treated plots the following year. Similarly, phytoseiid mite CIDs were only slightly lower in walnut plots treated with spinetoram during the first generation of codling moth in 2011 and not at all in 2010. Overall, as a percentage among all trials within a crop, there were more significant treatment effects for natural enemy/prey ratios (50% for pears, 33% for walnuts) than for natural enemy CIDs alone (25% for pears, 13% for walnut). A difference in natural enemy CIDs alone between insecticide treatments can be more difficult to interpret than a difference in natural enemy/prey ratios as it results from a combination of direct effects on the natural enemies themselves and indirect effects on the availability of prey as a resource for the natural enemies. In contrast, a difference in natural enemy/prey ratios represents a change in natural enemy abundance relative to prey abundance and thus estimates treatment effects on the natural enemy - prey interaction rather than on natural enemies alone. Factoring together the low densities of prey and natural enemies as ratios allowed subtle effects of the insecticide treatments to become more detectable in our field studies.

The use of PV-baited traps adds a new dimension to monitoring natural enemies (Jones et al., 2011, this issue a). They can be deployed in and around orchards to capture a wide variety of natural enemies and can be used to measure the impact of IPM programs on natural enemy populations, to measure the diversity of natural enemy communities (Mills et al., this issue b) and to gather information to create natural enemy phenology models (Jones et al., this issue b). One of the main benefits of PV traps is that they capture flying insects for an extended period of time compared with instantaneous collection of insects using beat tray sampling or in-situ visual examinations. PV traps are relatively new tools available for ecologists and IPM practitioners, and in our field studies, we used PV traps baited with several different lures and pooled the catches of natural enemies across lures because several of them have been shown to be cross

attractive (Jones et al., this issue a). Despite the high levels of abundance of several natural enemy taxa on these traps relative to the numbers found using other sampling methods, we did not detect differences between insecticide treatments other than for predatory Neuroptera in pears. It is unclear whether the lack of informative results from the PV traps in our field studies reflect the small size of our experimental plots (≤ 0.4 ha) and the strong flight ability of most of the natural enemies captured by the traps including the predatory Syrphidae and Neuroptera and parasitic Hymenoptera. Since the active space of these PV traps has not been determined it is also possible that they may also have attracted natural enemies from outside of the orchards, such as from adjacent refugia, which are known to be important for recruiting natural enemies into orchards (Miliczky and Horton, 2005). It is also possible that natural enemies were captured in the experimental plots as they were transiting through the plots.

Laboratory bioassays can provide meaningful data yet field trials provide the most realistic results (Prasifka et al., 2005). However, there are intrinsic difficulties with conducting on-farm research. Foremost is the difficulty for growers to provide untreated control plots that are used to provide background information about pest and natural enemy abundance. Horticultural crops are expensive to produce and most growers do not want to risk crop loss. In this study, only walnut growers provided plots with unsprayed trees. One solution is to budget for crop loss in grant applications that would compensate growers for losses they might incur.

Determining plot size is another aspect of on-farm trials. If investigators were only interested in sedentary or wingless natural enemies, plot size could be small, but larger sized plots are required if natural enemies are able to readily disperse between plots and surrounding habitat. However, large plots can constrain the number of treatment replications that can be

414 included. Larger plot sizes also increase costs in terms of labor, equipment and sampling time
415 (Prasifka et al., 2005).

416 The researchers associated with this current study have participated in large replicated field
417 studies and through experience, have concluded that it is best to conduct replicated studies within
418 an orchard versus blocking the study across multiple orchards. The main reason is that
419 variability is often greater between orchards than between treatments. This limits the ability to
420 determine significant treatment effects. Conducting a replicated study in one orchard minimizes
421 plot-to-plot variability because the orchard unit is relatively uniform which increases the
422 likelihood of treatment differences. Verification of results is then enhanced when the studies are
423 successfully reproduced elsewhere.

424 Another insight into on-farm research is how an investigator decides on which grower and
425 orchard to work with. Orchard management can be classified as a continuum ranging from
426 excellent progressive growers with well-managed orchards to t where growers are considerably
427 less progressive and their pest management is lacking. Researchers, including the authors, tend
428 to work with progressive growers because they are interested in new ideas, having research
429 conducted in their orchards, and are less apt to cause problems such as over-spraying or
430 harvesting the crop before notifying the researchers. The pear growers in these studies were
431 progressive and maintained well managed orchards and they did not need to treat for *C. pyricola*
432 during the summer. They likely conserved their natural enemies by using *C. pomonella* mating
433 disruption instead of insecticides and had extra-orchard habitat that was suitable as natural
434 enemy refugia. Zwick and Fields (1977) showed that the elimination of sprays against *C.*
435 *pomonella* in pears helps to conserve biological control and manage *P. pyricola*. Later, Riedl et
436 al. (2000) also demonstrated that pear growers who successfully implemented an integrated fruit

production program that encouraged biological control were able to reduce broad-spectrum insecticide use while maintaining good fruit quality at harvest. More recently, this concept has been expanded in the Hood River, OR pear district. Here growers have substituted *C. pomonella* mating disruption for insecticide sprays. This has allowed significant acreage to avoid treatments for *P. pyricola* during the summer season (Gallardo et al., in this issue; Warner 2012). It is possible that our failure to cause significant disruption of secondary pests in pears in this study was related to several years of low pest abundance in these progressive orchards. A similar situation occurred in the walnut studies that were conducted in well-managed orchards where populations of *C. juglandicola* were low.

Again, our studies were conducted during two unseasonably cool summers. Weather also plays a big part where high temperatures can result in more of the population being exposed to the toxicant (because they emerge and develop through more of the sensitive stages before the insecticide residue, which degrades on a calendar date basis, is gone) and cold temperatures have the opposite effect. Thus, year-to-year variation in weather patterns, which are inherent in any field study, contribute some level of serendipity to the success of large-plot field studies of insecticide effects on natural enemies.

Given the difficulties associated with conducting large-scale replicated research trials, properly designed laboratory bioassays are often a better alternative to rapidly screen a variety of insecticides. Laboratory bioassays are less expensive and time-consuming to conduct and are likely to yield results that are less variable than field studies. However, field studies conducted on several sites for several years are likely the best way to document the effects of insecticides on secondary pests and natural enemies. As the knowledgebase increases, growers and pest control consultants can then decide which insecticide to use based on efficacy against the target

460 pest and selectivity against natural enemies or they can choose to apply these products at a time
461 when natural enemies are in a less susceptible stage of their life cycle or seasonal phenology
462 (Jones et al., 2009).

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464

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Table 1. Mean ($\pm$ SE) cumulative insect (or mite)-days for secondary pests observed in the walnut orchard trials

Year	Orchard	Treatment	Cumulative insect (or mite)-days per walnut leaflet			
			<i>C. juglandicola</i> ¹	<i>C. juglandis</i>	<i>T. urticae</i>	<i>P. ulmi</i>
2010	A	Chlorantraniliprole	55.0 $\pm$ 6.3 b	32.5 $\pm$ 9.3	53.7 $\pm$ 12.8	45.6 $\pm$ 10.8
		Spinetoram	85.0 $\pm$ 18.9 a	25.3 $\pm$ 5.7	64.4 $\pm$ 30.1	71.2 $\pm$ 52.3
		Control	57.6 $\pm$ 8.1 b	34.4 $\pm$ 12.2	46.4 $\pm$ 24.6	36.4 $\pm$ 13.5
		<i>F</i>	6.02	0.11	1.25	0.27
		df	2, 6	2, 6	2, 6	2, 6
		<i>P</i>	0.04	0.90	0.35	0.77
2011	A	Chlorantraniliprole	4.5 $\pm$ 0.2 b	0.0 $\pm$ 0.0	0.2 $\pm$ 0.1 b	1.0 $\pm$ 0.4 b
		Lambda-cyhalothrin + chlorantraniliprole	9.9 $\pm$ 1.0 a	0.0 $\pm$ 0.0	1.4 $\pm$ 0.1 a	4.8 $\pm$ 1.4 a
		Control	6.6 $\pm$ 1.5 b	0.0 $\pm$ 0.0	2.0 $\pm$ 0.6 a	4.7 $\pm$ 0.9 a
		<i>F</i>	7.63		20.34	9.44
		df	2, 6		2, 6	2, 6
		<i>P</i>	0.02		0.002	0.01
	B	Chlorantraniliprole/Spinetoram ²	8.6 $\pm$ 1.3 b	0.0 $\pm$ 0.0	16.5 $\pm$ 5.0 a	14.6 $\pm$ 3.3
		Spinetoram/Chlorantraniliprole	20.8 $\pm$ 4.3 a	0.0 $\pm$ 0.0	10.2 $\pm$ 3.0 a	13.6 $\pm$ 3.9
		Grower standard	6.8 $\pm$ 2.2 b	0.0 $\pm$ 0.0	5.2 $\pm$ 3.6 b	7.2 $\pm$ 3.3
		Control	7.1 $\pm$ 1.2 b	0.0 $\pm$ 0.0	13.9 $\pm$ 2.0 a	11.2 $\pm$ 3.3
		<i>F</i>	6.20		6.31	1.41
		df	3, 6		3, 6	3, 6
		<i>P</i>	0.03		0.03	0.33

¹Means in a column for each year and orchard followed by different letters are significantly different ($P \leq 0.05$). Data natural log (X+1) transformed, actual means reported.

²Order of treatments refer to sprays applied to first then second generation codling moth.

Table 2. Mean ($\pm$ SE) cumulative insect-days for natural enemies observed in the pear orchard trials

Year	Orchard	Treatment	Cumulative insect-days per beat tray			Cumulative insect-days per trap		Cumulative insect-days per band
			<i>Trechnites</i> spp. ¹	Predatory Heteroptera	Araneae	Predatory Neuroptera ¹	Predatory Syrphidae	<i>Forficula auricularia</i>
2010	A	Chlorantraniliprole	11.9 $\pm$ 3.7 a	63.8 $\pm$ 8.4	17.9 $\pm$ 3.2	75.4 $\pm$ 12.1 b	576.9 $\pm$ 46.4	593.9 $\pm$ 233 b
		Cyantraniliprole	9.6 $\pm$ 3.2 a	76.1 $\pm$ 10.1	17.6 $\pm$ 0.7	46.7 $\pm$ 7.5 c	449.8 $\pm$ 5.7	910.2 $\pm$ 265 a
		Spinetoram	1.9 $\pm$ 0.9 b	63.2 $\pm$ 2.8	16.9 $\pm$ 3.2	131.2 $\pm$ 11.6 a	577.3 $\pm$ 20.1	262.5 $\pm$ 133 b
		<i>F</i>	27.05	2.13	0.04	12.67	6.28	9.73
		df	2, 4	2, 4	2, 4	2, 4	2, 4	2, 4
		<i>P</i>	0.005	0.23	0.96	0.02	0.06	0.03
2011	B	Chlorantraniliprole	8.8 $\pm$ 1.9	2.7 $\pm$ 0.4	3.9 $\pm$ 0.5	69.8 $\pm$ 35.3	32.8 $\pm$ 12.6	252.2 $\pm$ 96
		Spinetoram	4.1 $\pm$ 2.7	1.3 $\pm$ 0.3	3.9 $\pm$ 0.6	216.9 $\pm$ 33.0	70.1 $\pm$ 24.2	156.6 $\pm$ 43.4
		<i>F</i>	6.71	7.46	0.02	6.5	4.3	1.37
		df	1, 3	1, 3	1, 3	1, 3	1, 3	1, 3
		<i>P</i>	0.08	0.07	0.91	0.08	0.15	0.326

¹Means in a column for each year and orchard followed by different letters are significantly different ($P \leq 0.05$). Data natural log (X+1)

transformed, actual means reported.

Table 3. Mean ($\pm$ SE) cumulative insect (or mite)-days for natural enemies observed in the walnut orchard trials

Year	Orchard	Treatment	Cumulative insect (or mite)- days per walnut leaflet		Cumulative insect-days per trap		
			<i>Trioxys pallidus</i> mummies ¹	Phytoseiid mites	Predatory Neuroptera	Predatory Syrphidae	Parasitic Hymenoptera
2010	A	Chlorantraniliprole	26.8 $\pm$ 5.3	17.2 $\pm$ 5.8	827.0 $\pm$ 85.8	293.1 $\pm$ 12.5	948.0 $\pm$ 255.9
		Spinetoram	24.7 $\pm$ 4.7	35.2 $\pm$ 27.3	825.9 $\pm$ 89.1	515.1 $\pm$ 128.7	782.9 $\pm$ 178.1
		Control	34.1 $\pm$ 4.0	17.4 $\pm$ 2.6	753.1 $\pm$ 141.0	400.1 $\pm$ 60.5	709.1 $\pm$ 96.7
		<i>F</i>	1.97	11.32	0.26	1.36	0.29
		df	2, 6	2, 6	2, 6	2, 6	2, 6
		<i>P</i>	0.22	0.95	0.78	0.33	0.76
2011	A	Chlorantraniliprole	4.2 $\pm$ 0.5	28.4 $\pm$ 2.1 a	48.4 $\pm$ 24.1	171.8 $\pm$ 42.2	3258.4 $\pm$ 232.8
		Lambda-cyhalothrin + chlorantraniliprole	5.4 $\pm$ 0.4	19.7 $\pm$ 1.8 b	59.6 $\pm$ 17.6	121.9 $\pm$ 33.3	2983.9 $\pm$ 346.3
		Control	4.7 $\pm$ 1.0	19.9 $\pm$ 0.2 b	38.0 $\pm$ 14.2	155.6 $\pm$ 88.0	3738.8 $\pm$ 940.8
		<i>F</i>	1.12	11.32	0.47	0.31	0.19
		df	2, 6	2, 6	2, 6	2, 6	2, 6
		<i>P</i>	0.39	0.01	0.65	0.75	0.84
	B	Chlorantraniliprole/Spinetoram ²	5.3 $\pm$ 0.6	20.8 $\pm$ 1.0 a	74.0 $\pm$ 5.0	984.3 $\pm$ 0.6	1836.7 $\pm$ 433.3
		Spinetoram/Chlorantraniliprole	8.4 $\pm$ 1.1	15.1 $\pm$ 1.2 b	86.2 $\pm$ 24.7	959.0 $\pm$ 1.5	2011.8 $\pm$ 306.8
		Grower standard	6.1 $\pm$ 1.5	14.3 $\pm$ 1.0 b	101.7 $\pm$ 31.0	1065.7 $\pm$ 1.3	2352.8 $\pm$ 150.7
		Control	7.7 $\pm$ 1.3	19.1 $\pm$ 1.0 a	115.8 $\pm$ 9.3	459.5 $\pm$ 1.1	1167.8 $\pm$ 220.2
		<i>F</i>	1.41	8.78	0.62	0.75	3.15
		df	3, 6	3, 6	3, 6	3, 6	3, 6
		<i>P</i>	0.33	0.01	0.63	0.56	0.11

¹Means in a column for each year and orchard followed by different letters are significantly different ($P \leq 0.05$). Data natural log (X+1)

transformed, actual means reported.

²Order of treatments refer to sprays applied to first then second generation codling moth.

Table 4. Mean ($\pm$ SE) natural enemy/prey ratios based on cumulative insect days in the pear orchard trials

Year	Orchard	Treatment	Predator CID / prey CID ratios ¹		
			<i>Trechnites</i> spp. / <i>C. pyricola</i>	Predatory Heteroptera / <i>C. pyricola</i>	Predatory Neuroptera / <i>C. pyricola</i>
2010	A	Chlorantraniliprole	0.7 $\pm$ 0.1 a	4.1 $\pm$ 0.9	4.8 $\pm$ 0.9 ab
		Cyantraniliprole	0.5 $\pm$ 0.2 b	4.3 $\pm$ 0.5	2.7 $\pm$ 0.4 b
		Spinetoram	0.1 $\pm$ 0.1 c	4.7 $\pm$ 0.6	9.7 $\pm$ 0.2 a
		<i>F</i>	85.22	0.05	7.93
		df	2, 4	2, 4	2, 4
		<i>P</i>	0.001	0.95	0.04
2011	B	Chlorantraniliprole	1.5 $\pm$ 0.4	0.5 $\pm$ 0.1	9.8 $\pm$ 3.2 b
		Spinetoram	1.3 $\pm$ 0.4	0.6 $\pm$ 0.4	49.6 $\pm$ 10.8 a
		<i>F</i>	1.01	0.03	15.08
		df	1, 3	1, 3	1, 3
		<i>P</i>	0.39	0.87	0.03

¹Means in a column for each year and orchard followed by different letters are significantly different ($P \leq 0.05$). Data natural log (X+1)

transformed, actual means reported.

Table 5. Mean ($\pm$ SE) percent parasitism and predator/prey ratios (based on cumulative mite-days) in the walnut orchard trials

Year	Orchard	Treatment	Percent parasitism <i>C. juglandicola</i>	Predator CMD / prey CMD ratios ¹	
				Phytoseiid mite / Twospotted mite	Phytoseiid mite / European red mite
2010	A	Chlorantraniliprole	32.8 $\pm$ 3.8 a	0.32 $\pm$ 0.1	0.30 $\pm$ 0.0
		Spinetoram	23.8 $\pm$ 3.2 b	0.41 $\pm$ 0.2	0.44 $\pm$ 0.2
		Control	38.6 $\pm$ 5.1 a	0.69 $\pm$ 0.2	0.48 $\pm$ 0.1
		<i>F</i>	7.71	1.35	1.22
		df	2, 6	2, 6	2, 6
		<i>P</i>	0.02	0.33	0.36
2011	A	Chlorantraniliprole	46.8 $\pm$ 3.6	312.3 $\pm$ 109.8 a	205.3 $\pm$ 179.6 a
		Lambda-cyhalothrin + chlorantraniliprole	42.9 $\pm$ 8.6	14.1 $\pm$ 7.0 b	5.2 $\pm$ 2.6 b
		Control	33.7 $\pm$ 2.3	12.0 $\pm$ 2.5 b	4.5 $\pm$ 0.6 b
		<i>F</i>	2.35	34.63	6.82
		df	2, 6	2, 6	2, 6
		<i>P</i>	0.18	0.001	0.02
	B	Chlorantraniliprole/Spinetoram ²	39.2 $\pm$ 1.8	1.50 $\pm$ 0.4	1.55 $\pm$ 0.3
		Spinetoram/Chlorantraniliprole	29.8 $\pm$ 4.4	1.98 $\pm$ 0.9	1.50 $\pm$ 0.7
		Grower standard	48.3 $\pm$ 2.1	9.10 $\pm$ 10.5	2.98 $\pm$ 2.1
		Control	51.2 $\pm$ 3.4	1.45 $\pm$ 0.5	2.03 $\pm$ 1.0
		<i>F</i>	1.33	3.38	0.92
		df	2, 6	3, 6	3, 6
		<i>P</i>	0.35	0.10	0.48

¹Means in a column for each year and orchard followed by the different letters are significantly different ($P \leq 0.05$). Data natural log ($X+1$) transformed, actual means reported.

²Order of treatments refer to sprays applied to first then second generation codling moth.