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AQ1–AQ5

Traps and Attractants for Monitoring Navel Orangeworm (Lepidoptera: Pyralidae) in the Presence of Mating Disruption

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Abstract

The use of aerosol mating disruption for management of the navel orangeworm *Amyelois transitella* (Walker) in California tree nuts has increased markedly. This treatment suppresses pheromone monitoring traps in neighboring orchards as well as in the orchard under treatment. The current study, therefore, addresses the interrelated topics of which attractant is most effective, how the number of adults captured is affected by trap design, and what sex is captured. Under most circumstances, wing traps baited with phenyl propionate (PPO) captured more adults than those baited with a 5-compound kairomone blend. Adding a pheromone lure along with the dispenser for the experimental attractant increased the number of adults captured with PPO both in and near-mating disruption. In contrast, addition of a pheromone lure along with the kairomone blend only increased the number of adults captured in orchards near but not under mating disruption. Delta traps, which are preferred by the industry, captured fewer adults than wing traps. Improvements in the number of adults captured with PPO in delta traps from addition of a pheromone lure and from design modifications were additive. Both sexes were captured by all attractants and trap designs tested, and the sex ratio was highly variable. Open traps like the wing trap captured a slightly higher proportion of males than closed traps. These findings are discussed in the context of management of the navel orangeworm.

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AQ7 **Key words:** *Amyelois transitella*, almond *Prunus dulcis*, pistachios *Pistacia vera*, phenyl propionate, kairomone

Semiochemical tools are an increasingly important part of insect pest management, particularly for lepidopteran orchard pests (Witzgall et al. 2010, Miller and Gut 2015, Abd El-Ghany 2019). Lepidopteran sex pheromones are particularly useful for monitoring and timing insecticide applications because they are able to detect even low-density populations (Witzgall et al. 2010). The use of sex pheromones to decrease both the abundance and damage of lepidopteran pests offers distinct advantages due to low toxicity and low impact on humans and wildlife, including beneficial arthropods (Witzgall et al. 2010). However, use of pheromone mating disruption can greatly reduce the usefulness of lepidopteran sex pheromones as a monitoring tool where mating disruption is being applied (Knight et al. 2005). Other semiochemicals are used to address this need for an alternative monitoring method, such as (2E,4Z)-decadionoate (pear ester) for codling moth *Cydia pomonella* L. (Light et al. 2001,

Knight et al. 2019), and (4-methyl-1-propan-2-yl-1cyclohex-2-enyl) acetate terpinyl acetate (2-(methyl-3-cyclohexenyl)isopropyl acetate) for oriental fruit moth *Grapholita molesta* (Busck) (Rice and Kirsch 1990, Kovanci and Walgenbach 2005).

In California, the navel orangeworm *Amyelois transitella* (Walker) is a key pest of pistachio *Pistachia vera* L. and almond *Prunus dulcis* (Mill.) D. A. Webb and situationally important in walnut *Juglans regia* L. (Adaskaveg et al. 2019, Beede et al. 2019, Grant et al. 2019). For almond and pistachio, fundamental practices for managing navel orangeworm include sanitation of unharvested nuts to remove overwintering harborages and populations, and timely harvest to minimize the opportunity for damage (Adaskaveg et al. 2019, Beede et al. 2019). Timely and judicious insecticide treatment at key times is also frequently necessary to hold navel orangeworm damage to acceptable limits (Siegel et al. 2019a,b). In addition, mating disruption

is increasingly used for navel orangeworm pest management in these crops (Higbee and Burks 2008, Higbee et al. 2017, Burks and Thomson 2019). Currently, it is estimated that up to 200,000 ha (Higbee, unpublished data) of the California almond and pistachio crops (totaling >600,000 ha, USDA-NASS 2019) are under mating disruption for control of navel orangeworm.

Current monitoring practice for navel orangeworm in almond and pistachio includes pheromone and egg traps (Adaskaveg et al. 2019, Beede et al. 2019). Egg traps provide an artificial oviposition substrate for navel orangeworm females (Rice et al. 1976, Van Steenwyk and Barnett 1985, Burks et al. 2011, Higbee and Burks 2011). There is no evidence that the oviposition of gravid females entering mating disruption blocks is affected by this treatment. Pheromone traps provide more reliable, sensitive, and labor-efficient detection compared with egg traps (Higbee et al. 2014, Burks and Higbee 2015). However, captures of males in navel orangeworm pheromone traps almond are almost completely eliminated by mating disruption, and substantially reduced in orchards up to several km from the orchard under treatment (Burks 2017). Trap design is also a factor in the effectiveness of pheromone traps in the absence of mating disruption. Wing traps baited with pheromone capture navel orangeworm more efficiently than delta traps with pheromone (Burks and Higbee 2015, Kuenen and Siegel 2016), but delta traps nonetheless remain more widely used because pest control advisors find them easier to work with and more trouble-free due to a more protected glue liner. Ovipositional baits have also been used to trap females instead of eggs (Burks et al. 2011, Nay et al. 2012). There is evidence that monitoring based on gravid females might have a higher association with subsequent damage compared to pheromone traps (Rosenheim et al. 2017), although the data were from an area heavily affected by mating disruption.

Two nonpheromone attractants have been developed with the potential to improve detection of navel orangeworm in the presence of mating disruption. One of these is a synthetic blend that consists of five components: a straight-chain alkenol, a spiroketal, and three benzoinoids (Beck et al. 2009, 2012) (hereafter referred to simply as the kairomone blend). The other attractant is the single component, phenyl propionate (PPO) (Burks et al. 2016, Burks 2017). Both of these attractants capture a mixture of males and females when used as lures in traps (Beck et al. 2012, Burks et al. 2016).

Efficacy of the kairomone blend as an attractant in traps has been tested in experiments in almonds and pistachios, including in almonds under mating disruption (Beck et al. 2009, 2012). Traps baited with the kairomone blend capture navel orangeworm adults in the presence of mating disruption (Beck and Higbee 2015). As with the kairomone blend, traps baited with PPO alone capture navel

orangeworm adults in mating disruption, whereas capture of navel orangeworm in traps baited with only pheromone is completely suppressed. Moreover, in orchards under mating disruption, traps baited with both a pheromone lure and a PPO dispenser (PPO-combo traps) capture more adults as PPO-only lures, just as observed in orchards not under mating disruption (Burks et al. 2016). This capacity of PPO-combo lures to detect well and capture similarly in disrupted, partially disrupted (e.g., downwind), and nondisrupted orchards has practical importance (Burks 2017). Both the kairomone blend and PPO lures capture a combination of males and females (Burks et al. 2009, Beck et al. 2012), and the females captured are predominantly mated (Beck et al. 2012, Burks et al. 2016).

While there are data characterizing both the kairomone blend and PPO as navel orangeworm lures, the two attractants have not been compared directly in the same experiment. Here, we present a study comparing the performance of traps with these two types of attractants in almond and pistachio orchards under mating disruption, and in orchards near those treated with mating disruption where effectiveness of the pheromone traps currently used is compromised. In addition, we 1) examined the effect of a pheromone lure presented along with PPO or kairomone; 2) examined the effect of different trap designs on the effectiveness of sex pheromone presented with PPO to augment the number of adults captured; and 3) compared the effect of these factors (mating disruption status, crop, attractant, pheromone co-attractant, and trap design) on the sex ratio of adults captured. These related objectives are intended to provide practical options for improving monitoring for navel orangeworm in area affected by mating disruption.

Materials and Methods

A series of experiments conducted between 2017 and 2019 compared navel orangeworm adults captured in traps with PPO or kairomone blend with or without a pheromone lure, and examined the impact of the factors examined in these experiments on the ratio of males to females captured in traps (Table 1). The data from 2017 have previously been partially presented in a conference proceeding (Burks et al. 2018).

Comparison of PPO and the Kairomone Blend In or Near-Mating Disruption

A season-long experiment was conducted in almond and pistachio orchards near Kettleman City (nonmating disruption) and Coalinga (mating disruption), in California's Western San Joaquin Valley. Horticultural details of these sites are provided in Burks et al. (Burks et al. 2018). The orchards near Kettleman City were not directly

Table 1. Experiments with PPO and kairomone blend for monitoring *Amyelois transitella* by crop, mating disruption status, and date

Experiment	Crop	Under mating disruption?	Number of treatments	Number of replicates	Dates
Comparison of PPO and kairomone blend	Almond	No	5	8	7 June to 12 Sept. 2017
	Almond	Yes	5	8	
	Pistachio	No	5	8	
	Pistachio	Yes	5	8	
Early-season comparison of PPO and kairomone blend	Almond	No	5	8	6 Apr. to 12 June 2018
	Almond	No	5	8	
	Pistachio	Yes	5	8	
Impact of trap design: wing, bucket, and delta	Almond	Yes	5	8	21 June to 12 July 2018
Impact of trap design: wing, bucket, and modified delta	Almond	Yes	5	8	12 July to 28 Sept. 2018
Impact of trap design: co-attractant and delta modification	Almond	Yes	7	8	17 May to 10 July 2019

under mating disruption treatment, but proved to be influenced by mating disruption in orchards within 2 km.

A series of randomized complete block experiments were used to compare navel orangeworm adults captured between: 1) a commercial pheromone trap; 2) PPO alone; 3) PPO with a pheromone lure; 4) the kairomone blend alone; and, 5) the kairomone blend with a pheromone lure. These tests were conducted with wing traps (Fig. 1) to mimic previous studies with PPO (Burks et al. 2016, 2018; Burks 2017). Orchard rows served as replicate blocks. Traps containing attractants were placed in orchards under mating disruption midway between mating disruption dispensers. At the mating disruption sites there were therefore ~45 m between treatments within replicate blocks, and 90 m between replicate blocks. At the nonmating disruption sites the distances were ~50 m between trap treatments within block, and 100 m between replicate blocks. In the nonmating disruption almonds two groups of four replicate blocks were located 100 m apart across an orchard road within the management block, whereas in all other sites all replicate blocks were in a straight line. The pheromone lure used was NOW Biolure (Suterra LLC, Bend, OR), which was adhered to the inside roof of the wing trap. Phenyl propionate was dispensed from a membrane pouch (as previously described, Burks et al. 2016). The kairomone blend components were placed in dispensers in the ratio and amounts as previously described (Beck et al. 2012). Briefly, the components 1-octen-3-ol, ethyl benzoate, methyl salicylate, acetophenone, and conophthorin in a 12:4:4:1:1 ratio, respectively, were diluted at 200 parts per thousand

in ethyl acetate, which was placed on cotton plugs in 7-ml Nalgene bottles. Holes (3 mm in diameter) were drilled into the lids when these dispenser bottles were placed in the field. Phenyl propionate dispensers and the kairomone blend vials were suspended from a wire in the center of the trap. Traps were placed in the field on 6–8 June 2017, and data were collected and traps serviced weekly until 12 September 2017. Lures were replaced at 6-wk intervals. Trap liners for the treatments other than pheromone were brought back to the laboratory to identify the sex of the moths captured.

In spring 2018, this comparison was extended in both of the Kettleman City (nonmating disruption) sites, and in the pistachio orchard under mating disruption near Coalinga. The other Coalinga site was not used for the lure trial because of difficulties with early-season site access, although this site was used for subsequent tests on impact of trap design. The initial monitoring interval, from 9 April to 30 April 2018, was removed from the Coalinga pistachio data set because mating disruption was not in use at that site for much of this period.

Mating disruption in the 2017 was provided by the cooperator using a commercial application of Suterra Puffer NOW (Suterra LLC), In 2018 and 2019, this cooperator used ISOMATE NOW Mist (Pacific Biocontrol, Vancouver, WA). Differences between these two systems are described in Burks and Thomson (2019). Briefly, the Puffer system emits around 90 mg active ingredient per ha per night using dispensers placed at a density of 5/ha and emitting at a frequency of four times an hour for 12 h (17:00 to 05:00), whereas

Fig. 1. Trap types used. Wing trap (upper left), delta trap (upper right), bucket trap (lower left), and modified delta trap (lower right).

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ISOMATE emits around 10% more active ingredient using dispensers placed at a density of 2.5/ha, emitting nine times an hour, from 20:00 to 06:00.

Impact of Trap Design on Navel Orangeworm Adults Captured With PPO

In June of 2018, we initiated an experiment examining the impact of trap design on navel orangeworm adults captured (Table 1, Fig. 1). Based on results from the previous experiments, these trials were limited to PPO with or without pheromone.

The first of these trials compared: 1) a pheromone lure only in a wing trap, 2) a PPO dispenser with a pheromone lure in a wing trap; 3) a PPO dispenser with a pheromone lure in a standard commercial delta trap; 4) a PPO dispenser alone in a bucket trap; and 5) a PPO dispenser along with a pheromone lure in a bucket trap. The first two treatments were considered as, respectively, a negative control (under mating disruption) and the most effective treatment. The bucket trap was examined because capturing adults without a glue liner offered advantages for research projects, and because this type of trap is favored by a few pest control advisors. The delta trap was examined because this is the most widely used trap by commercial pest control advisors. Traps used in this study included the following: wing trap (Suterra LLC); delta traps (PHEROCON VI, Trece Inc., Adair, OK); and bucket trap (universal moth traps or Unitraps, Semiochemicals Corp., Burnaby, BC, Canada). The wing traps were modified slightly to make it easier to change the liners (Kuenen et al. 2005; Fig. 1). For the delta trap, as for the wing trap, the pheromone lure was adhered to the side of the trap near the upper middle, and the PPO dispenser was suspended from a wire hook in the center of the trap. For the bucket trap, PPO was suspended from a hook placed through a small hole upper portion of the trap. The pheromone lure was adhered inside. The killing agent in the bucket traps was dichlorvos, provided by cutting Hot Shot No Pest Strips (United Industries Corp., St. Louis, MO) into 25 mm^2.

Based on initial observations, a subsequent trial in 2018 (Table 1) replaced the regular commercial delta traps. The traps were modified by cutting an opening in each side of the trap of 17.8 × 6.4 cm,

centered on the side of the trap and with the bottom 1.3 cm from the bottom corner (Fig. 1). The attractant types and positions used in this second trapping experiment were the same as the first; the only change was the modification to the delta trap.

A third experiment in 2019 further examined more specifically the impact of features of wing and delta traps on navel orangeworm adults captured with PPO alone, or with a pheromone lure (Table 1). Treatments included the following: 1) a wing trap with a pheromone lure; 2) a wing trap with a PPO dispenser alone; 3) a wing trap with both a PPO dispenser and a pheromone lure; 4) a delta trap with PPO alone; 5) a delta trap with PPO and a pheromone lure; 6) a modified delta trap with PPO alone; and 7) a modified delta trap with both a PPO dispenser and a pheromone lure. Trap types, placement of attractant, and modifications were as described for the previous experiments.

Data Analysis

Experiments at each site were analyzed separately, and counts were summed over the length of the experiment. Traps with missing data in any monitoring interval were excluded from analysis. Most of the experiments were analyzed using a generalized linear mixed model (GLMM) with an appropriate error distribution (Table 2), and with treatments (different attractant or trap types) and replicates respectively modeled as fixed and random effects. For the GLMM analyses, the Tukey's multiple range test was used to examine differences between treatment means. In two cases, the nonparametric Welch analysis of variance (ANOVA) was used because of failure of convergence when trying to specify a GLMM model (Zar 1999, McDonald et al. 2014, Mangiafico 2015). The Games–Howell posthoc test was used for multiple comparisons following a significant Welch ANOVA (Mangiafico 2015, Peters 2018).

The sex ratio was explored by comparing the proportion of males captured in traps by the various factors examined (mating disruption stats, crop, attractant, co-attractant, trap type, and seasonal factors). Spearman rank correlation was used as a nonparametric examination of seasonal trends in the 2018 attractant trial, and a GLMM with a binomial error distribution was used to compare males as a

Table 2. Statistical tests, parameters, and *P*-values

Experiment	Crop	Dates	Procedure	Statistic, df, and *P*-value
Comparison of PPO and kairomone blend	Almond, disrupted	7 June to 12 Sept. 2017	Welch ANOVA	$F = 59.84$, df = 4, 35; $P < 0.001$
	Almond, not disrupted		GLMM, negative binomial	$F = 66.3$, df = 4, 28; $P < 0.001$
	Pistachio, disrupted		GLMM, negative binomial	$F = 109.96$, df = 4, 28; $P < 0.001$
	Pistachio, not disrupted		GLMM, negative binomial	$F = 29.95$, df = 4, 28; $P < 0.001$
Early season comparison of PPO and kairomone blend	Almond, not disrupted	6 Apr. to 12 June 2018	GLMM, Gaussian	$F = 20.11$, df = 4, 27; $P < 0.001$
	Pistachio, disrupted		GLMM, Gaussian	$F = 7.89$, df = 4, 25; $P < 0.001$
	Pistachio, not disrupted		GLMM, Gaussian	$F = 16.01$, df = 4,27; $P < 0.001$
Impact of trap design: wing, bucket, and delta	Almond	21 June to 12 July 2018	GLMM, negative binomial	$F = 45.11$, df = 4, 28; $P < 0.001$
Impact of trap design: wing, bucket, and modified delta	Almond	12 July to 28 Sept. 2018	Welch ANOVA	$F = 36.72$, df = 3, 28; $P < 0.001$
Impact of trap design: co-attractant and delta modification	Almond	17 May to 10 July 2019	GLMM, negative binomial	$F = 34.87$, df = 6,42; $P < 0.001$
Sex ratio: seasonal trend	Almond	6 Apr. to 12 June 2018	Spearman correlation	$N = 5$, rho = –0.9, $P = 0.037$
Sex ratio: differences between trap types	Almond		GLMM, binomial	$F = 6.43$, df = 3, 19; $P = 0.003$

proportion of total navel orangeworm adults capture for trap types in June 2018 (Table 2). The correlation proportion males with dates was based on all pooled males and adults for all replicates, and the comparison of proportions of males between traps used pooled data for all monitoring intervals.

Data were summarized and processed using R 3.6.0 (R Core Team 2019). GLMMs with binomial and negative binomial error distribution were implemented in the GLIMMIX procedure of the SAS System (SAS Institute Inc. 2016).

Results

The analysis used and the values for the associated test statistic, df, and *P*-value are listed in Table 2.

Comparison of PPO and the Kairomone Blend In or Near-Mating Disruption

There were significant differences among the treatments in each of the four sites in 2018 (Table 2, Fig. 2). Several patterns emerged. First, in both of the orchards not treated with mating disruption, wing traps baited with PPO captured more adults than traps baited with a pheromone lure alone. Second, in all cases, the wing traps baited with PPO alone captured more adults than the wing traps

baited with the kairomone blend alone. Third, a pheromone lure as a co-attractant significantly increased the number of adults captured in wing trap for PPO in both orchards directly under mating disruption and those effected by nearby mating disruption. For the kairomone blend, however, this effect was only significant in orchards indirectly effected by nearby mating disruption. Fourth, under the conditions of this test, the kairomone blend trap without a pheromone lure as co-attractant did not capture significantly more adults than the pheromone lure alone in orchards under treatment for mating disruption.

Trends from the lure experiment in spring of 2018 were similar to those from 2017 (Table 3). Wing traps baited with PPO alone captured significantly more adults than wing traps baited with kairomone alone in the nondisrupted almonds, and nominally more at the other two sites (i.e., adults in traps baited with PPO and pheromone were in different ranges in the Tukey's multiple range test). Likewise, more adults captured in wing traps baited with either PPO or kairomone and a pheromone lure co-attractant compared with these attractants alone. Wing traps containing the kairomone alone did not capture significantly more adults than pheromone alone in the pistachios under mating disruption. In the nondisrupted pistachio orchard, pheromone-baited wing traps captured fewer adults than PPO-baited wing traps (as observed in both nondisrupted orchards
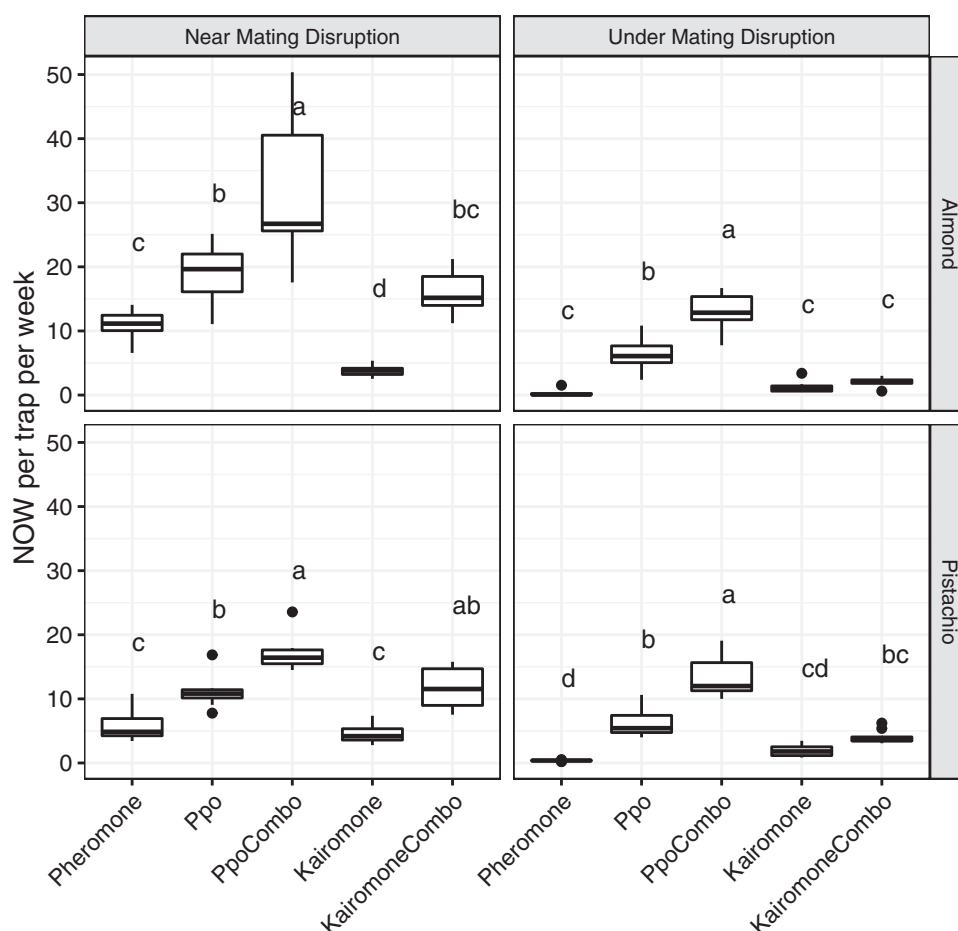

Fig. 2. Box plot showing the average weekly rate of *Amyelois transitella* adults captured in wing traps in almond or pistachio sites from June to September 2017. 'Under Mating Disruption' indicates orchards directly treated with mating disruption, and 'Near Mating Disruption' indicates orchards not treated with mating disruption but affected by mating disruption treatments in the region. Attractants were a commercial pheromone lure, Ppo, and a kairomone blend. The suffix 'Combo' indicates that Ppo or kairomone were presented with a commercial pheromone lure as a co-attractant. Treatments within the same crop-MD combination with different letter superscripts are significant different (experiment-wise *P* < 0.05).

Table 3. Effect of attractant on *Amyelois transitella* adults per trap per week (*n*, mean ± SE) for wing traps in three sites in spring 2018, by mating disruption (MD) status

Attractant	Almond, no MD	Pistachio, no MD	Pistachio under MD
Pheromone	33 ± 2.0b	7 ± 1.0bc	1.3 ± 0.22c
PPO	26 ± 2.2bc	10 ± 1.2ab	6.6 ± 0.92ab
PPO and pheromone	46 ± 3.5a	14 ± 1.4a	9.5 ± 1.9a
Kairomone	22 ± 2.3c	4 ± 0.7c	3.1 ± 0.3bc
Kairomone and pheromone	39 ± 2.1ab	8 ± 0.9ab	6.0 ± 0.68ab

April to June 2018. *n* = 8 except for kairomone treatment at the no MD sites, in which *n* = 7. Means followed by different letters are significant different (experiment-wise *P* < 0.05).

Table 4. *Amyelois transitella* adults captured per week (mean ± SE, *n* = 8) under mating disruption in bucket or sticky traps baited with AQ10 PPO with or without pheromone as a co-attractant

Trap	Lure	June	Aug.
Wing	Pheromone	0.1 ± 0.1c	0.0 ± 0.0
Wing	PPO with pheromone	57 ± 4.8a	43 ± 4.1a
Delta	PPO with pheromone	14 ± 2.5b	26 ± 4.1b
Bucket	PPO	9 ± 1.7b	5 ± 1.6c
Bucket	PPO with pheromone	14 ± 1.5b	9 ± 1.7c

*For June to July 2018 the delta traps used were stock; whereas for July to Sept. 2018 modified delta traps were used. Means followed by different letters are significant different (experiment-wise *P* < 0.05).

in 2017), whereas the converse was true in the nondisrupted almond orchard in 2018.

Impact of Trap Design on Navel Orangeworm Adults Captured With PPO

Wing traps baited with both PPO and a pheromone lure captured significantly more adults than other trap types, with or without a pheromone lure co-attractant (Table 4). There was no significant difference in the number of adults captured between bucket traps baited with PPO, with or without a pheromone lure, and the stock delta trap baited with both PPO and a pheromone lure. After the stock delta traps were replaced with delta traps with openings in the sides, these customized delta traps baited with PPO and a pheromone lure captured significantly more adults than bucket traps baited with PPO in either the presence or absence of a pheromone lure, but significantly fewer adults than wing traps baited with PPO and a pheromone lure (Table 4).

The 2019 experiment examining the impact of delta trap modifications in more detail revealed an impact of both the modification and the use of a pheromone lure as a co-attractant (Fig. 3). Stock delta traps baited only with PPO captured significantly more adults than wing traps baited with a pheromone lure, and all versions of the delta trap baited with PPO captured nominally or significantly fewer adults than a wing trap baited with PPO alone, and significantly fewer adults than a wing trap baited with both PPO and a pheromone lure. Among delta trap variations, addition of a pheromone lure significantly increased the number of adults captured, and the open-sided modification had the same effect. Adding a pheromone lure to PPO presented in the open-sided modified version of a delta trap brought captures to within the same range in the multiple range test as the wing trap with PPO alone. Wing traps with PPO captured
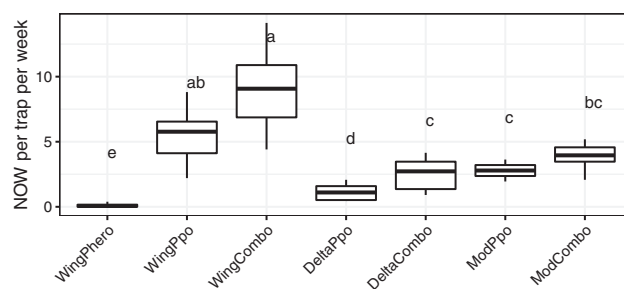

Fig. 3. Box plot showing the average weekly rate of *Amyelois transitella* adults captured in sticky traps in an almond site under mating disruption, from May to July 2019. Traps used were standard wing traps, standard delta traps, or modified delta traps (ModPpo and ModCombo). Attractants used were a commercial pheromone lure (Phero) and Ppo. The suffix 'Combo' indicates that Ppo was presented with a commercial pheromone lure as a co-attractant. Treatments within the same crop-MD combination with different letter superscripts are significant different (experiment-wise *P* < 0.05).

significantly more adults than stock delta traps baited with PPO, whether or not accompanied by a pheromone lure (Fig. 3).

Effect of Attractant, Trap, and Environmental Factors on Sex Ratio in Traps

Across all factors examined in the attractant comparison in 2017 (crop, mating disruption, attractant, and augmentation with a pheromone lure, 970 observations), there were no differences in the ratio of males to females. The median proportion of males was 0.75 and the first and third quartiles were, respectively, 0.625 and 0.875, and the minimum and maximum proportions were 0 and 1. There was no evidence of seasonally based differences in the sex ratio in 2017.

In early 2018, however, there was evidence of a change in sex ratio in early season (Fig. 4). In the single almond site examined there was a trend of a decrease in the proportion of males capture over the period from mid-April to late May. This trend was not evident in either of the pistachio sites examined at the same time (one with mating disruption, one not disrupted). In the subsequent experiment, there was also evidence that trap design influenced sex ratio of adults captured with PPO. Bucket traps baited with PPO and a pheromone lure captured a significantly lower proportion of males than a wing trap baited with PPO and a pheromone lure, whereas capture was in an intermediate range for delta traps baited with PPO and a lure and for bucket traps baited only with PPO (Fig. 5).

Discussion

Comparison of the data from the current study to data from previous studies suggest that the effectiveness of attractants in traps was influenced by mating disruption used in nearby orchards. In earlier experiments conducted far from mating disruption (Burks et al. 2016), wing traps baited with PPO captured around 20% of the adults compared with traps baited with pheromone lures. A subsequent study was found, after it began, to have been conducted near commercial mating disruption (Burks 2017). In this study data were compared between wing traps baited with either pheromone lure only or both PPO and a pheromone lure. These traps were placed in almond block not treated with mating disruption but located 1–4.5 km from a 65-ha almond orchard under mating disruption. Unlike the previous study (Burks et al. 2016), in which wing traps baited only with pheromone captured nominally (not significantly) more adults than traps baited with both PPO and a pheromone lure,
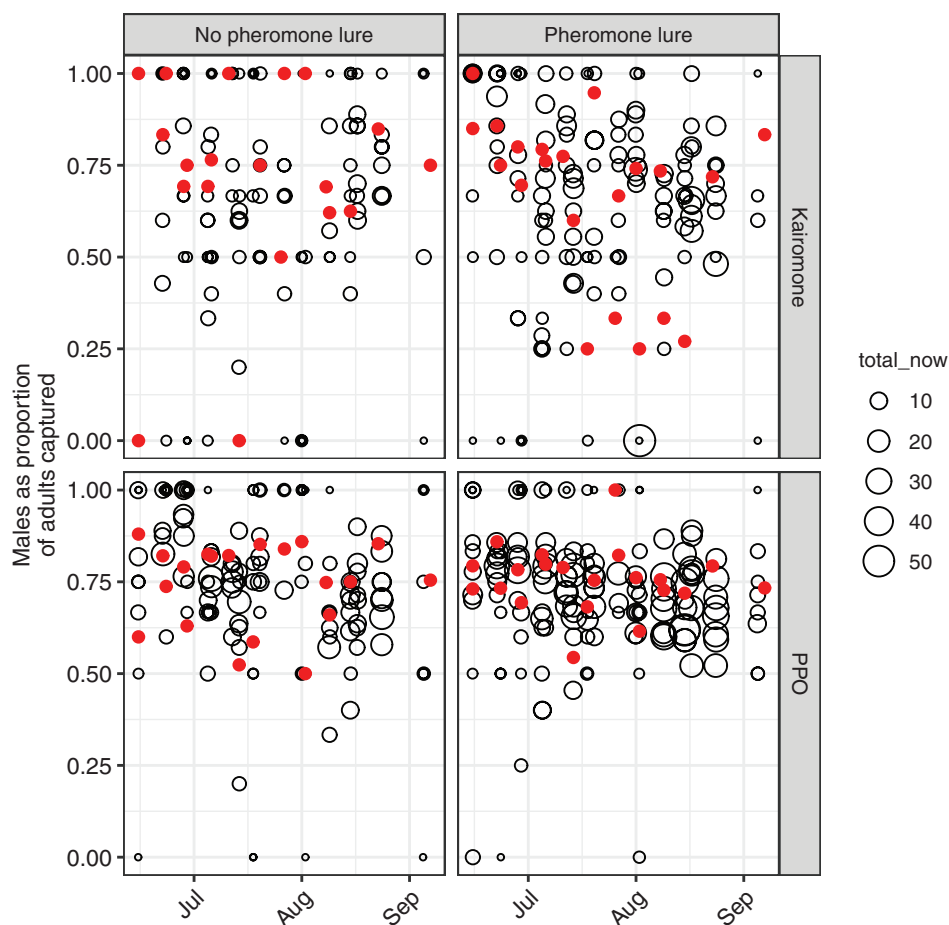
Fig. 4. *Amyelois transitella* males as a proportion of adults captured in wing traps in March through May 2018 in an almond orchard that was not treated with mating disruption, but influenced by mating disruption in the region. Open circles represent individual traps, and filled circles are sex ratios for the sums of males and females for a given trapping ratio. There was a significant negative correlation (Spearman, $P < 0.05$) between date and the proportion of males captured in traps in this time and site.
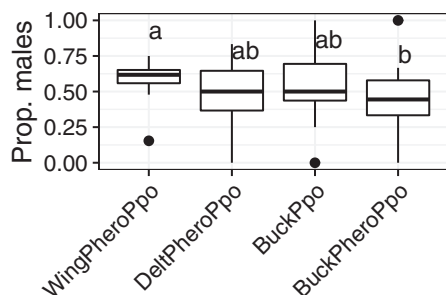

Fig. 5. Comparison of proportion of males captured with wing, stock delta, or bucket traps in an orchard treated with mating disruption. The suffix 'Ppo' indicates that phenyl propionate was the only attractant used, and 'PheroPpo' indicates that Ppo was presented with a commercial pheromone lure as a co-attractant treatments with different letter superscripts are significant different (experiment-wise $P < 0.05$).

the converse was observed in the latter study (Burks 2017). In this latter study, traps baited with both PPO and pheromone captured nominally more adults than traps baited with pheromone alone in a location 4.5 km and cross-wind from the identified almond block treated with commercial mating disruption, and the combination traps in nondisrupted almond orchards nearer to almond orchard under mating disruption captured significantly more adults than traps baited with adults only pheromone. Based on these previous observations, trends after the current test began suggested that the nonmating disruption site was affected by mating disruption in the vicinity. Further inquiries confirmed the use of commercial navel orangeworm mating disruption in an almond orchard 1.5 km to the east of the Kettleman City nonmating disruption almond site and suggested that it was also being used to the north of the nonmating disruption pistachios at Kettleman City. Figure 2 and Table 1 in this current study should, therefore, be seen as a contrast between the performance of these attractants in an orchard directly under mating disruption versus a nonmating disruption orchard influenced by mating disruption in the vicinity. That is, however, probably a more likely situation for practical use of one of these attractants.

The data from this study demonstrated more robust performance for PPO than kairomone as a monitoring tool under the conditions examined. Traps baited with PPO alone captured more adults than traps baited with pheromone alone in both the orchards under mating disruption treatment, and the nonmating disruption orchards effected by neighbors under mating disruption (Fig. 2). Kairomone did not capture significantly more adults than pheromone lures in the presence of mating disruption. Another way of expressing these data is that, in the orchards directly treated with mating disruption, PPO-baited traps had zero capture on 20–23% of the times they were checked, whereas kairomone traps were

empty 45–62% of the time (n = 101 to 104). It is also notable that PPO was significantly augmented by the addition of a pheromone lure in both the mating disruption-influenced orchards and those directly treated with mating disruption, whereas this was only true of kairomone in the orchards influenced by mating disruption and not those under mating disruption treatment. These observations indicate that the kairomone formulation tested here is a valid navel orangeworm attractant, but not the better of the two attractants examined here for monitoring navel orangeworm in the presence of mating disruption.

Trap design can also have profound effect on the effectiveness of monitoring traps (Burks and Higbee 2015) and can also involve practical trade-offs for practitioners. In areas not impacted by mating disruption, pheromone-baited wing traps capture more males than pheromone-baited delta traps (Burks and Higbee 2015, Kuenen and Siegel 2016). Delta traps nonetheless remain in more widespread use, indicating that California pest control advisors have concluded that reduced trap numbers are an acceptable trade-off for the ease of use of their preferred trap. In the first of the trap design experiments in 2017, the principal objective was to examine bucket traps as a research tool, to obtain navel orangeworm specimens not contaminated with the glue from sticky traps. The delta trap was included to confirm our hypothesis that the reduction in performance of the PPO-pheromone lure combination would be no worse than that observed with pheromone lures in the absence of mating disruption. The result from the first experiment, in June, suggested that the difference between the performance of the PPO-pheromone lure combination in the delta versus the wing trap was more profound that that described previously for the pheromone lure in these traps in nondisrupted orchards. We hypothesized that having air flow on all sides was necessary for effectiveness of the PPO-pheromone lure combination, and the subsequent experiments in 2018 and 2019 confirmed this hypothesis. In the 2019 trial, the wing trap baited only with pheromone was empty in 90% of the weekly trials (n = 48), and for the wing traps baited with PPO (either alone or with a pheromone lure), this figure was 8%. For the four delta trap configurations shown in Fig. 3, these figures are, respectively, 40, 27, 25, and 19%. The combination of the modified delta trap and augmentation with a pheromone lure therefore offers reasonably robust performance while avoid use of a wing trap.

The sex ratio was of interest because a previous study indicated that counts from traps baited with lures attractive to gravid females predicted damage at harvest better than other lures (Rosenheim et al. 2017). These baits (e.g., meal or egg traps) nonetheless have other weaknesses. Compared with black light traps, or pheromone traps in nondisrupted orchards, they seem to be more affected by changes in the orchard volatile environment accompanying phenological changes (Rice et al. 1976). They also present the problem of a long-tailed frequency distribution, with a frequent median of 0 moths captured (Burks et al. 2011, Higbee and Burks 2011). This low capture causes worry concerning false negatives and means these types of traps inherently require intensive effort to obtain specific information. If, therefore, one of the attractants examined proved clearly more attractive to females, this might be a consideration. The data, however, indicated that there were no such large differences. The higher proportion of males captured in early 2018 with both PPO and kairomone may be because of a lower number of mated females in this early season, since both attractants attract mated more than unmated females (Beck et al. 2012, Burks et al. 2016). One hypothesis concerning the greater association of female than male traps with navel orangeworm damage is that the female traps have a smaller trapping radius and therefore sample more locally. It is possible that PPO also has a more smaller trapping radius in an orchard under mating disruption and would, therefore, be more closely associated with damage in that situation. Further research is needed to examine this possibility.

In summary, compared with the kairomone blend, PPO provided more robust and similar captures of navel orangeworm both in almond and pistachio orchards under mating disruption and also in orchards not treated with mating disruption but affected by mating disruption treatments in the vicinity. Trap type has a large impact on the effectiveness of PPO. It works better with open-sided traps than with the closed-sided delta traps favored by the California tree nut industry, but modification of delta traps and/or use of a pheromone lure as a co-attractant offers a range of trade-offs between trap effectiveness and user preference. Both PPO and the kairomone capture both sexes in variable ratios that usually favor males. Obtaining an attractant that consistently captures predominantly females (particularly unmated females) could improve options for monitoring and mass trapping, but remains an elusive goal.

Acknowledgments

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