

# Evaluation of permethrin for attracticide development against *Choristoneura rosaceana* and *Pandemis pyrusana* (Lepidoptera: Tortricidae) males

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## Abstract

The behavioural effects on *Choristoneura rosaceana* Harris and *Pandemis pyrusana* Kearfott male moths of permethrin added into a grease matrix containing the female sex pheromone for each species (i.e. an attracticide) were studied. Attracticides containing 0.16%, 1.6%, or 16% pheromone for each species were tested with or without insecticide (6% permethrin). Male moths responded similarly in indoor wind tunnel bioassays and field trials, as measured by source contacts and trap captures respectively, to formulations containing different concentrations (0.16%, 1.6%, or 16%) of pheromone regardless of the presence or absence of permethrin. *C. rosaceana* trap catches per trap/day increased with pheromone dose,  $0.025 \pm 0.025$  at 0.16% pheromone,  $0.75 \pm 0.13$  (1.6%) and  $2.60 \pm 0.64$  (16%), compared with  $0.025 \pm 0.025$  (0.16%);  $0.55 \pm 0.12$  (1.6%);  $2.18 \pm 0.46$  (16%) without permethrin. Similar results were obtained for *P. pyrusana*. Overall, the results indicated that the attracticides loaded with 6% permethrin had no behavioural effects on *C. rosaceana* and *P. pyrusana* male moths.

**Keywords:** Attracticides; *Choristoneura rosaceana*; *Pandemis pyrusana*; Permethrin; Repellency

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## 1. Introduction

Repellents are substances that prevent an insect's approach to a source (Metcalf and Metcalf, 1982), or elicit a behavioural response to move away from a source (Chou et al., 1997). True repellents are highly volatile compounds (Metcalf and Metcalf, 1982) detected by olfactory receptors which impede contact between the insect and a source (Hibbard and Bjostad, 1989), whereas compounds that stimulate an insect to leave a substrate after contact are called excitorepellents (Metcalf and Metcalf, 1982) or irritants (Hibbard and Bjostad, 1989). Baits (e.g. attracticides = sex pheromone + insecticide) that use insecticides which produce true repellency are undesirable since they might reduce control efficacy (Curkovic, 2004). In such

cases, it is necessary to enhance the formulation by either adding attractants that overcome the repellency (Rust and Reiersen, 1977) or select non-repellent insecticides (De Souza et al., 1992). The objective of this research was to assess in both indoor and field trials the possible repellent effects of permethrin on *Choristoneura rosaceana* Harris and *Pandemis pyrusana* Kearfott incorporated into an attracticide formulation.

## 2. Material and methods

### 2.1. Insects

*C. rosaceana* and *P. pyrusana* were obtained from colonies maintained at the Washington State University Tree Fruit Research and Extension Center, Wenatchee, WA, in growth chambers at  $23 \pm 2$  °C, 40–50% HR, and 16:8 h photoperiod. Male pupae were sexed and kept in the chambers. Upon emergence, males were placed in

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plastic cups and provided with honey solution via wicks: 2–4-day-old males were used in indoor experiments, since >97% survived under experimental conditions (Curkovic, 2004) and were sexually mature at that age (Delisle, 1995).

## 2.2. Wind tunnel

A wind tunnel, similar to that described by Carde and Hagaman (1979), maintained in an environmentally controlled room set at  $25 \pm 2^\circ\text{C}$ , was used. The airspeed was set at  $40 \pm 5\text{ cm/s}$  in the middle of the chamber,  $55 \pm 5\%$  RH, and light intensity at 2 lux during assays. Attracticide droplets inside hair rollers (6 cm length, 1.5 cm diameter, L&N Sales and Marketing, PA) were placed in the middle of the wind tunnel section, on a source platform held by a ring stand at approximately 10 cm from the upwind end. The attracticide droplets were caged to avoid direct contact with males, preventing later contamination inside the tunnel during experiments. Cages with males were placed on a release platform at about 70 cm from the source platform. The plume inside the wind tunnel was 1–3 cm width at the source, and 10–15 cm width at the release platform. The wind tunnel and materials used during bioassays were cleaned prior to each experiment.

## 2.3. Pheromones and attracticide sources

Pheromone components (all  $\geq 95\%$  purity) were provided by the manufacturer (Bedoukian Research Inc., Danbury, CT, USA). The attracticide matrix used in the experiments was a grease commercial formulation (Last Call™ IPM Inc. Technologies, Portland, OR, USA) that includes an ultraviolet stabilizer ( $\sim 70\%$ ), a thickener, and a sticker. Batches using the reported ratios for *C. rosaceana* (95.5 : 2 : 1.5 : 1, Z11-14Ac : E11-14Ac : Z11-14OH : Z11-14Al) and *P. pyrusana* (94 : 6, Z11-14Ac : Z9-14Ac) female sex pheromones (Roelofs et al., 1977; Vakenti et al., 1988) were prepared. The pheromone components were added directly to the matrix to prepare a 16% (by weight) mixture, then stirred while in a warm-bath, poured into a syringe, and stored at  $0^\circ\text{C}$ . Lower pheromone concentrations were obtained by diluting the original batch, adding pheromone-free grease. Technical permethrin (6% by weight) was added as described above into some batches. Insecticide-free batches were also used. Final mixtures were stored at  $0 \pm 1^\circ\text{C}$  and kept at  $22 \pm 1^\circ\text{C}$  for  $\sim 30$  min before bait preparation.

## 2.4. Handling and acclimation of moths for wind tunnel assays

Males were placed individually into mesh cylinder cages (4 cm diameter  $\times$  2 cm height), covered with a plastic lid, provided with honey solution via a cotton wick, and acclimated on a table next to the wind tunnel at least 2–3 h before experiments. Trials were run in the first 2–4 h of the scotophase, which corresponded to the sexual activity

period for both *C. rosaceana* (Evenden et al., 1999) and *P. pyrusana* (Knight and Turner, 1998). After setting the attracticide source (50 mg) inside the wind tunnel (upwind), each male cage was placed individually on the release platform (downwind); after 2 min the plastic lid was removed by pulling an attached string. One treatment, i.e., a particular concentration for either attracticide permethrin loaded or attracticide insecticide free, was run per night ( $n = 30$  males/treatment).

## 2.5. Observation of moth behavioural sequence

Visual observations in wind tunnel bioassays were made after acclimation of the observer for 10 min using a flashlight covered by a red filter (Kodak gelatin #29, Rochester, NY). Male responses were categorized as staying in or taking off from the platform, or making source contact with the cage containing the attracticide.

## 2.6. Attracticide baits, traps, and orchards

Individual attracticide formulation droplets (50 mg) were placed on aluminium foil in hairrollers and pinned inside the delta traps. Traps ( $n = 4$ /treatment) were placed on apple trees (1.5 m) in two central Washington orchards, separated by ca. 30 m, then serviced, and rotated clockwise after each examination to minimize the impact of location on captures. The orchards were not sprayed with insecticides during the trial period.

## 2.7. Data Analysis

$\chi^2$  and Tukey tests were used to identify multiple differences between proportions ( $p$ ) of males contacting sources in indoor bioassays. Responses were transformed by  $p = 1/2[\{\arcsine(\sqrt{x/(n+1)})\} + \{\arcsine(\sqrt{(x+1)/(n+1)})\}]$ , where  $x$  is the number of individuals contacting sources, and  $n$  the total of individuals released. A randomized block design was used to compare the proportions of cumulative trap captures per treatment. Data were transformed to arcsine square-root, analysed by ANOVA and treatment means compared by Tukey test when  $P < 0.05$  (Zar, 1996).

## 3. Results

### 3.1. Indoor bioassays

There were significant differences in *C. rosaceana* male moth responses (source contact) based on the concentration of pheromone in the attracticide formulation. Attracticides loaded with either 1.6% or 16% pheromone attracted similar proportions of males, but significantly more males than the source loaded with the lowest concentration (Fig. 1;  $\chi^2 = 51.66$ ,  $df = 10$ ,  $P < 0.001$ ). On the other hand, no significant differences were observed in the proportions of males contacting attracticide sources

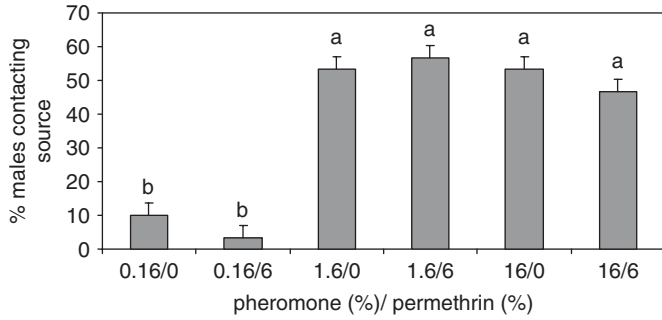


Fig. 1. Response (% source contact) of *Choristoneura rosaceana* males ( $n = 30$ /treatment) to 0.16%, 1.6%, and 16% pheromone concentrations in the attracticide source when loaded with 0% vs. 6% permethrin, in indoor wind tunnel trials.

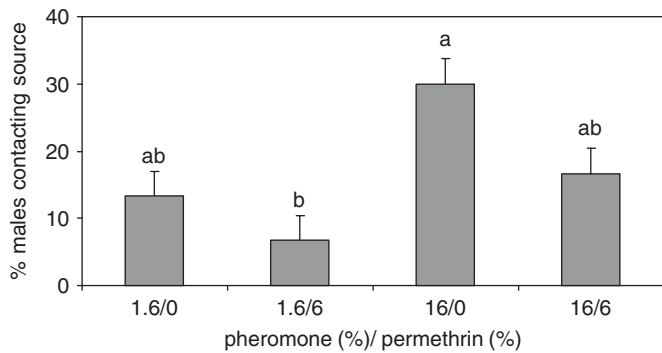


Fig. 2. Response (% source contact) of *Pandemis pyrusana* males ( $n = 30$ /treatment) to 1.6%, and 16% pheromone concentrations in the attracticide source when loaded with 0% vs. 6% permethrin, in indoor wind tunnel trials.

loaded with the same pheromone concentration, with (6%) or without (0%) permethrin.

Fewer *P. pyrusana* males were available for testing, so only attracticide formulations with the two highest pheromone concentrations (1.6% and 16%), with and without permethrin, were evaluated. Overall, the response of *P. pyrusana* males to the pheromone source was much lower than for *C. rosaceana* to the same pheromone concentrations. There were no statistically significant reductions in moths contacting the source between treatments containing the same pheromone concentration when either loaded or not with permethrin. However, unlike *C. rosaceana*, significant differences were found between the attracticide containing 16% pheromone and 0% permethrin, and the attracticide loaded with 1.6% pheromone and 6% permethrin (Fig. 2;  $\chi^2 = 42.28$ ,  $df = 6$ ,  $P < 0.001$ ).

### 3.2. Field trials

As with indoor bioassays, field trials showed no differences in the number of *C. rosaceana* males captured in traps baited with an attracticide containing the same pheromone concentration when either loaded or not with

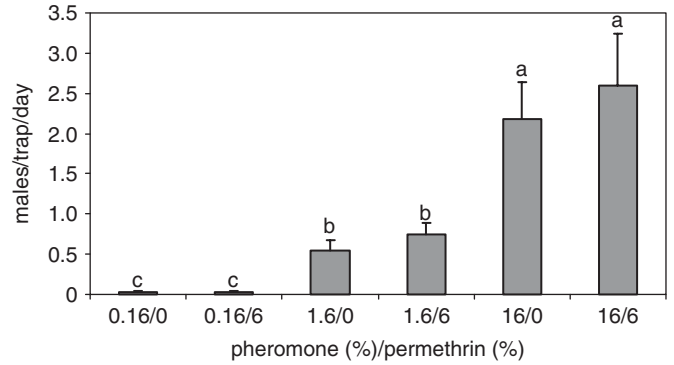


Fig. 3. Cumulative captures of *Choristoneura rosaceana* males in traps ( $n = 4$ /treatment) baited with an attracticide formulation loaded with 0.16%, 1.6%, or 16% plus 0% or 6% permethrin, Winchester, WA, 19th – 29th June, 2002.

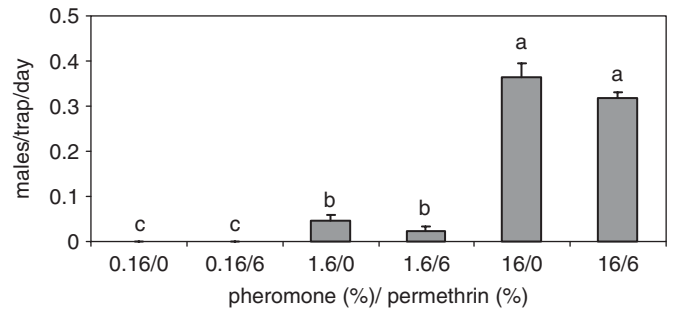


Fig. 4. Cumulative captures of *Pandemis pyrusana* males in traps ( $n = 4$ /treatment) baited with an attracticide formulation loaded with 0.16%, 1.6%, or 16% plus 0% or 6% permethrin, Winchester, WA, 19th – 29th June, 2002.

permethrin. However, significant differences in trap catches were observed when different pheromone concentrations at the source were compared irrespective of whether permethrin was present or not. The highest pheromone concentration tested (16%) attracted significantly more males than the 1.6% loading which was, in turn, significantly more attractive than the lowest concentration (0.16%) (Fig. 3,  $F = 45.23$ ,  $df = 5,12$ ,  $P < 0.001$ ).

Results for *P. pyrusana* demonstrated similar trends although lower captures were obtained to similar concentrations. No significant differences were observed comparing treatments containing the same pheromone concentration, either including or excluding permethrin. However, trap captures increased significantly with pheromone concentration at the source (Fig. 4,  $F = 25.70$ ,  $df = 5,12$ ,  $P < 0.001$ ). No male captures were observed when the lowest concentration, with or without permethrin, was tested. In another field trial (Wenatchee) designed to check the responses to the two lowest pheromone concentration, a few captures were obtained (not observed in Winchester at the lowest concentration), but again, no significant differences were observed when comparing attracticides loaded with the same pheromone

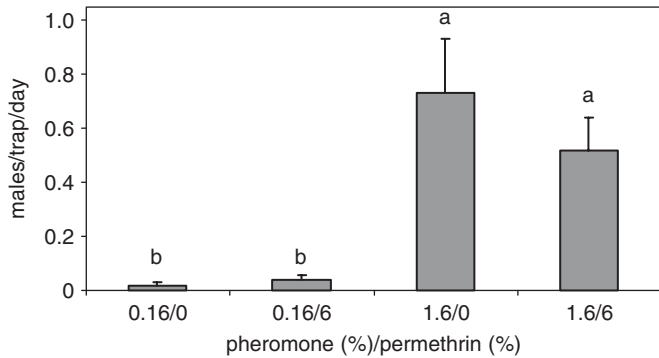


Fig. 5. Cumulative captures of *Pandemis pyrusana* males in traps ( $n = 4$ /treatment) baited with an attracticide formulation loaded with 0.16% or 1.6% plus 0% or 6% permethrin, Wenatchee, WA, 11th – 24th August, 2002.

concentration, regardless of whether the source was permethrin-loaded or not, and significantly greater captures were observed when the highest pheromone concentration was used (Fig. 5,  $F = 11.00$ ,  $df = 3,12$ ,  $P < 0.005$ ).

#### 4. Discussion

No true repellency resulted from the addition of permethrin into the attracticide formulations for both *C. rosaceana* and *P. pyrusana* males at any of the pheromone concentrations evaluated. Evaporation rate from sources is a key factor in eliciting true-repellency from target pest species (Schreck, 1977; Heitefuss, 1989). Thus, the behavioural responses observed in these experiments are probably due to the rate of evaporation of permethrin being below the concentration that is detectable to adult leafrollers. The moderate persistence and low volatility of permethrin (Hassall, 1990) support this hypothesis. True repellency to pyrethroids varies among insect species (Shemanchuck, 1981; Rieth and Levin, 1989), but it has not been widely reported in Lepidoptera (Haynes et al., 1986; De Souza et al., 1992). However, there is some evidence that pyrethroids may become repellent to some species when used above a behavioural threshold concentration (Schreck, 1977; Delabie et al., 1985; Phelan and Baker, 1987). These results indicate that, if such a threshold exists for permethrin to *C. rosaceana* or *P. pyrusana*, it was not exceeded by the 6% permethrin used in the attracticide formulation. Excitorepellency was not evaluated during bioassays because attracticides were caged and moth direct contact was excluded. However, previous reports (Curkovic and Brunner, 2005) indicated that both male *C. rosaceana* and *P. pyrusana* show excitorepellency immediately after direct contact with a permethrin-loaded attracticide. Despite the high pheromone doses used in some of the attracticide formulations, no repellent or inhibitory behavioural effects were observed over the range tested.

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