Pheromone-based trapping of larval codling moth, *Cydia pomonella*, in apple orchards

Zaid Jumean\(^1\), Jean-Pierre Lafontaine\(^2\), Charlene Wood\(^1\), Gary J. R. Judd\(^3\) & Gerhard Gries\(^1\)*

\(^1\)Department of Biological Sciences, Simon Fraser University, Burnaby, BC V5A 1S6, Canada, \(^2\)Phero Tech Int., Delta, BC V4G 1E9, Canada, \(^3\)Agriculture and Agri-Food Canada, Pacific Agri-Food Research Centre Summerland, BC V0H 1Z0, Canada

Accepted: 14 July 2006

**Key words:** pheromone, larval aggregation, tree banding, integrated pest management, Lepidoptera, Tortricidae

**Introduction**

In apple orchards throughout the world, larvae of the codling moth, *Cydia pomonella* L. (Lepidoptera: Tortricidae), are the primary cause of insect damage to fruit (Landolt et al., 1999). Tactics available as part of integrated pest management (IPM) programs for *C. pomonella* include pheromone-based mating disruption (Judd et al., 1997) or attract-and-kill of adult moths (Charmillot & Hofer, 1997), sterile insect technology (Judd & Gardiner, 2005), biological or conventional insecticide targeting neonate larvae (Knight et al., 1994; Lacey et al., 2004), release of parasitic wasps, like *Ascosyrphus quadridendrata* Wesmael (Clausen, 1978) and *Mastrus ridibundus* Gravenhorst (Unruh, 1997) which parasitize egg and larval stages, respectively, and use of cultural controls like tree bands placed on the lower bole to trap fifth-instar larvae seeking pupation sites (Judd et al., 1997; Judd & Gardiner, 2005).

Cocoon-spinning *C. pomonella* larvae release an aggregation pheromone that attracts both the larval parasitoid *M. ridibundus* (Jumean et al., 2005b) and conspecific larvae (Duthie et al., 2003; Jumean et al., 2004, 2005a). This pheromone\(^1\) is comprised of eight components: 3-carene, octanal, nonanal, decanal, \((E)-2\)-octenal, \((E)-2\)-nonenal, sulcatone, and geranylacetone (Jumean et al., 2005b). We tested whether a synthetic blend of the larval aggregation pheromone could be used to increase the efficiency of cardboard bands for trapping *C. pomonella* larvae in apple orchards.

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1. A Patent Cooperation Treaty patent application for use of this pheromone was filed on 6 April 2005.
2. Correspondence: Gerhard Gries, Department of Biological Sciences, Simon Fraser University, Burnaby, BC V5A 1S6, Canada. E-mail: gries@sfu.ca

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**Materials and methods**

**Preparation of pheromone-treated cardboard bands**

A polyurethane matrix was prepared by mixing a 1,3-polybutadiene holopolymer with diphenylmethane diisocyanate (proprietary materials). The matrix was made flexible by inclusion of the plasticizer acetyl tri-n-butyl citrate (Uniplex Chemical Corporation, Greensboro, NC, USA), and was stabilized by Ethanox 702 (Ethyl Corporation, Baton Rouge, LA, USA). Under continuous stirring of the mixture, the synthetic blend of *C. pomonella* larval aggregation pheromone was added at doses of 0, 10 000, 100 000, or 1 000 000 larval hour equivalents (LHE) per 30 cm of band. Ten LHE represented 22.8 ng of pheromone [sulcatone (0.81 ng), octanal (0.94 ng), 3-carene (0.95 ng), \((E)-2\)-octenal (4.10 ng), nonanal (4.10 ng), \((E)-2\)-nonenal (10 ng), decanal (1.40 ng), geranylacetone (0.50 ng)], equivalent to that produced by 10 cocoon-spinning larvae in 1 h except that \((E)-2\)-octenal and \((E)-2\)-nonenal were enhanced \(\times 10\) (Jumean et al., 2005a). All pheromone components were purchased from Aldrich (Milwaukee, WI, USA) or Bedoukian (Danbury, CT, USA), and were >95% chemically pure. A custom-built machine (30 \(\times\) 10 \(\times\) 10 cm) moved cardboard bands at a rate of 4.5 m min\(^{-1}\) while applying a thin stream of the polyurethane/pheromone mixture at a rate of 9 g min\(^{-1}\) to the band’s centre. Treated bands were covered with wax paper, cured overnight at room temperature, and then rolled and stored in Mylar® bags (West Coast Food Pak, Vancouver, BC, USA) at –20 °C.

**Experimental sites**

Experiments were conducted in two 2.5-ha commercial apple orchards [Orchard 1 (49° 55.988′N, 119° 21.636′W), Orchard 2 (49° 53.649′N, 119° 20.891′W)] in Kelowna,
British Columbia, Canada. These orchards were 30–45 years old and planted at a density of 200–240 trees ha\(^{-1}\), with tree × row spacings of 3 × 5 m (Orchard 1) and 5 × 5 m (Orchard 2), respectively. Apple varieties in Orchard 1 were Spartan, MacIntosh, Gala, Delicious, Granny Smith, and Golden Delicious, and in Orchard 2 MacIntosh and Red Delicious. Orchard 2 was certified organic, managed under guidelines of the Certified Organic Association of British Columbia (http://www.certifiedorganic.bc.ca/), but no insecticides were sprayed in either orchard during the study.

Experimental set-up

On each tree in each orchard, either the trunk or a main scaffolding limb that was at least 30 cm in circumference was wrapped with a single corrugated cardboard band (10 cm wide × 30–46 cm, Shippers Supply, Richmond, BC, Canada) treated either with a strip of pheromone-impregnated polyurethane (treatment) or untreated polyurethane (control). A main scaffolding limb was banded when the trunk circumference was >60 cm (<10%), because such large trunks would have required longer bands with increased pheromone release. All bands were attached at a height of 50–150 cm above ground. Treatment and control bands were systematically assigned to alternate trees in each row, leaving perimeter rows untreated to avoid possible edge effects. In each orchard, the total area of treated trees was ca. 1 ha. Experiments were initiated between 23 and 30 August 2004, and terminated between 9 and 11 October 2004, at which time bands were removed and numbers of cocooning second-generation larvae destined for overwintering diapause were recorded.

In Experiment 1 conducted in Orchard 1, we compared the efficiency of bands treated with 0 or 100 000 LHE of aggregation pheromone for trapping *Cydia pomonella* fifth-instar larvae. The relatively high pheromone dose in Experiment 1 was based on considerations that (i) cardboard bands would remain in the field for a long time; (ii) pheromone release rates would likely decline over time; and (iii) the active space over which foraging larvae are recruited would increase. In Experiment 2 conducted in Orchard 2, we tested the effect of larval pheromone dose (0, 10 000, 100 000, or 1 000 000 LHE per 30 cm of band) on capture efficiency of bands.

Measurement of pheromone release rates

Three corrugated cardboard bands (each 10 × 120 cm) were loaded with equivalent amounts of polyurethane-containing pheromone at 1 000 000 LHE per 30 cm (see above), cured overnight, rolled, and then placed in three separate glass chambers (15.5 in diameter × 20 cm high) with corrugations running parallel to air flow. Air was drawn at 24 °C through the chambers at ca. 1.5 l min\(^{-1}\) and then through a glass column (14 × 1 cm in diameter) containing Porapak
Figure 2  Release rates of pheromone components (mean ± SE) from a pheromone-impregnated polyurethane strip on cardboard bands (n = 3). Porapak Q traps were replaced every day for 8 days, and then after days 11, 14, 17, and 27. Bands were impregnated with 1,000,000 LHE (see text) of pheromone.
Q (50–80 mesh, Waters, Milford, MA, USA). Porapak Q traps were replaced every day for 8 days, and after days 11, 14, 17, and 27. Volatiles were eluted from each Porapak Q trap with a pentane rinse (3 ml). With tetradecane as an internal standard, pheromone components in each sample were quantified (mg per 30 cm day\(^{-1}\)) by gas chromatography, employing a Hewlett Packard (HP) 5890 Series II gas chromatograph equipped with a GC column (30 m × 0.32 mm in diameter) coated with DB-5 (J and W Scientific, Folsom, CA, USA) [temperature program: 50 °C (1 min), 10 °C min\(^{-1}\) to 200 °C (held for 5 min)]. Release rates of all pheromone components were quantified.

Statistical analyses

In Experiment 1, mean numbers of *C. pomonella* larvae coocooning in treatment (n = 91) and control bands (n = 97) were compared with a two-sample Student’s t-test. Experiment 2 was analyzed using a one-way analysis of variance (ANOVA) followed by Tukey’s multiple comparison procedure to compare means of 0 (n = 44), 10 000 (n = 50), 100 000 (n = 46), and 1 000 000 (n = 51) LHE treatments. The experimental error rate in both experiments was set at α = 0.05 (Zar, 1999).

Results and discussion

In Experiment 1, bands treated with pheromone at 100 000 LHE were more effective in capturing *C. pomonella* larvae than untreated bands (t = 2.37, d.f. = 186, P = 0.018) (Figure 1). In Experiment 2, bands receiving the 1 000 000 LHE treatment captured more than twice as many larvae as untreated control bands (F = 4.53, d.f. = 3,187, P = 0.0043) (Figure 1). Bands receiving the 1 000 000 or 100 000 LHE treatments both captured more larvae than bands with the 10 000 LHE treatment, which did not capture significantly more larvae than control bands. Our data provide proof-of-concept that synthetic larval aggregation pheromone can be used to enhance captures of fifth-instar *C. pomonella* larvae in trapping devices. This is the first such demonstration for a larval aggregation pheromone.

Polyurethane appears suboptimal as a pheromone-dispensing matrix and should be replaced with a more suitable dispenser. 3-Carene as one of the critical pheromone components (Jumean et al., 2005a) was detectable in the effluvium of cardboard bands for fewer than 3 days after initiation of release rate studies, and most other compounds were released at very high rates during the first 3 days, with rapidly declining release rates thereafter (Figure 2).

Larvae exited apples about 1 week after bands were placed on trees (Gavin Young, Field Manager, Okanagan-Kootenay Sterile Insect Release Program, pers. comm.), suggesting that they responded to pheromone release rates that had already declined to biologically more ‘appropriate’ levels. Although pheromone release rates after day 4 (Figure 2) were still up to 1000 times greater than the dose (228 ng day\(^{-1}\)) that attracted larvae in laboratory bioassays (Jumean et al., 2005a), these release rates were apparently effective in covering the greater active space over which ‘field lures’ would be expected to attract foraging larvae. To ensure continuous optimal efficiency of the larval trapping devices, dispensers with constant pheromone release rates ought to be developed.

Use of tree bands to capture and reduce populations of overwintering larvae is an effective supplemental tactic within IPM programs for *C. pomonella* (Judd et al., 1997; Judd & Gardiner, 2005); our data suggest that synthetic larval pheromone can be used to enhance the efficiency of this control tactic.

Acknowledgements

We thank Gavin Young for assistance in locating field sites, Dennis and Kathleen Behnke and Surjit Nagra for allowing access to their orchards, and John Borden for reviewing the manuscript. Financial support was provided by a BP Beirne Prize in Pest Management to ZJ, a Natural Sciences and Engineering Research Council of Canada – Industrial Research Chair to GG, with Phero Tech Int., SC Johnson Canada, and Global Forest Science (6F-18-2004-216; 6F-18-2004-217) as industrial sponsors, and by a grant from the Washington Tree Fruit Research Commission to GJRJ and GG.

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