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MATING DISRUPTION OF PEA MOTH *Cydia nigricana* F. (LEPIDOPTERA: TORTRICIDAE) BY A REPELLENT BLEND OF SEX PHEROMONE AND ATTRACTION INHIBITORS

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Abstract—Synthetic sex pheromone of the pea moth *Cydia nigricana*, (*E*,*E*)-8,10-dodecadien-1-yl acetate (*E*8,*E*10–12:Ac), was applied in polyethylene dispensers at a rate of 30 g/ha and 600 dispensers/ha in a 0.6-ha pea field. The release rate of *E*8,*E*10–12:Ac was 140 mg/ha/day after six days, and 82 mg/ha/day after 20 days. Aerial concentrations of *E*8,*E*10–12:Ac, as measured by a portable EAG apparatus, ranged from 2 ± 2 to 7 ± 3 ng/m^3. The antennal signal was high and rather constant within pea canopy, but was lower and fluctuated strongly above canopy. Initially, >99% isomerically pure *E*8,*E*10–12:Ac was released, and male moths were attracted to dispensers. After nine days, isomeric blend composition had equilibrated to approx. 92% *E*8,*E*10–12:Ac and 8% of the inhibitory isomers *E*,*Z*-, *Z*,*E*-, and *Z*8,*Z*10–12:Ac. Males were then repelled from the pheromone-permeated field. Traps baited with 100 μg *E*8,*E*10–12:Ac caught 258 ± 133 *C. nigricana* males/trap in the control, but no males at all in the disruption field.

Key Words—Sex pheromone, attraction inhibitor, behavioral antagonist, mating disruption, air permeation, field EAG, *Cydia nigricana*, Tortricidae, Lepidoptera, pea moth, (*E*,*E*)-8,10-dodecadien-1-yl acetate.

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INTRODUCTION

Disruption of sexual communication and mating by atmospheric permeation with synthetic pheromones is employed worldwide as a specific, environmentally safe, and economical technique for the management of Lepidopteran pest insects (Jutsum and Gordon, 1989; Ridgway et al., 1990).

In the pea moth, *Cydia nigricana*, a number of elements should facilitate mating disruption. *C. nigricana* uses only one sex pheromone component, (*E*,*E*)-8,10-dodecadien-1-yl acetate (*E*8,*E*10–12:Ac) (Greenway, 1984; Witzgall et al., 1993). Its alcohol analog, *E*8,*E*10–12:OH is used for control of codling moth *C. pomonella*, and economic synthesis is already available (Yamamoto and Ogawa, 1989). *C. nigricana* has one flight period and green peas grown for canning or deep-freezing have to be protected only during three to four weeks; the crop is sprayed with insecticides until shortly before harvest. The canopy of pea fields is low and homogenous, and *C. nigricana* is day-active; both factors facilitate maintenance of pheromone concentrations and behavioral observations.

Complex pheromone blends comprising several or unstable compounds, crops susceptible to attack by more than one insect generation or species, and unhomogeneous or high crop canopies can be serious obstacles for the successful and efficient use of pheromonal methods.

Most of all, for the advancement of both new and established applications of the mating disruption technique, experimental field data on the behavior of moths and molecules is imperative (Arn, 1992; Kirsch, 1992). A portable electroantennogram apparatus, using the insect antenna as pheromone detector, has been developed by Sauer et al. (1992) to cover a most important methodological gap: the rapid and sensitive measurement of disruptant chemical under field conditions.

We have measured, during a first mating disruption trial against *C. nigricana:* (1) the amount and isomeric purity of sex pheromone *E*8,*E*10–12:Ac released from the dispenser material, (2) its aerial distribution and concentration in a pea field, and (3) visually observed male and female behaviors.

METHODS AND MATERIALS

Chemicals and Dispensers. (*E*,*E*)-8,10-Dodecadien-1-yl acetate (*E*8,*E*10–12:Ac) was formulated in polyethylene tube dispensers (Shin-Etsu Chemical Co., Tokyo) at ca. 50 mg *E*8,*E*10–12:Ac/dispenser. Isomeric purity of *E*8,*E*10–12:Ac before formulation was 99.1% (0.6% *E*,*Z*; 0.3% *Z*,*E*; <0.05% *Z*8,*Z*10–12Ac) and chemical purity was 99.7%, by capillary gas chromatography (GC).

Measurement of Release Rates. Dispensers collected from the field were suspended in stoppered, round-bottom glass flasks (250 ml) at 22°C for 2 hr

($N = 3$). The atmospheric concentration of *E*8,*E*10–12:Ac in these flasks was saturated after >5 hr (>50 μg recovered from the walls). Flasks were washed three times with 2 ml redistilled hexane, and 50 μg dodecyl acetate was added as internal standard to the first solvent portion. The combined extract was condensed under N_2 to 1 ml, and 3 μl was analyzed by GC. Data were corrected for 69% recovery of *E*8,*E*10–12:Ac from the glass surface.

GC analysis was done on a Hewlett Packard 5890 instrument with flame ionization detection (FID), on a nonpolar SE-54 column (splitless injection, 25 m, 0.32 mm ID, Kupper & Co., Bonaduz, Switzerland), programmed from 60°C (hold 2 min) at 10°/min to 100°C, and 1.5°/min to 230°C.

Field Facilities and Dispenser Placement. Dispensers (360) were placed in an unsprayed pea field (0.6 ha) near Höör (Skåne, Sweden) on June 23, 1992 (600 dispensers/ha; 30 g *E*8,*E*10–12:Ac/ha). This test field was partitioned into unit squares of 100 m^2, and their corners were flagged. On the corners, on the middle of the sides, and in the centers of these squares, dispensers were tied to top shoots of pea plants. Each square along the field border received four additional dispensers at 5 m around the center. Dispenser density was accordingly 9 and 13 per unit square; overall density, due to shared corners and sides was 3.3 and 10.2 dispensers/100 m^2, within the field and along the border. Dispensers in the centers of 10 squares were removed to install traps or the EAG apparatus.

The test field was surrounded by trees and bushes and was situated 50 m downwind from an unsprayed control pea field (8 ha). Within this control field, 300 m away from the pheromone-treated test field, three single test plots of 100 m^2, spaced at 30 m, received 4, 8, and 12 dispensers, respectively.

Field Trapping. Tetra traps (Arn et al., 1979) were baited with 100 μg *E*8,*E*10–12:Ac (99.8% isomeric purity by GC; Witzgall et al., 1993) on red rubber septa (Thomas Scientific, Illinois). Traps were placed ca. 10 cm below canopy, within the 0.6-ha test field (five traps), along its border (five traps), in the control field (10 traps), and in the centers of the three isolated 100-m^2 test plots in the control field (three traps). Traps in the pheromone-treated field were set in the centers of unit squares (see above), instead of a dispenser. Traps were checked and replaced every two to five days.

Larval Counts. After the annual flight period of *C. nigricana*, lots of 1 m^2 were chosen at random in the pheromone-treated ($N = 50$) and the control field ($N = 50$), and all pea pods were checked for *C. nigricana* larvae.

Behavioral Observations. Counts of adult female and male *C. nigricana* were made on 15 days, between June 22 and July 17, during their period of sexual activity, lasting 2 to 3 hr between 5 and 9 PM. At distances of <5 m, females and males are easily distinguished by their flight behavior; they were caught with a sweepnet in case of doubt. The observers were stationary in 100-m^2 squares for 15 min.

Field Electroantennogram Recordings. The field EAG device was developed by Sauer et al. (1992). It consists of an antenna holder, protected by a glass chamber from ambient air, a suction pump, a charcoal filter, a motor-driven syringe to deliver defined pulses of synthetic pheromone, and hardware/software peripherals to amplify and record the EAG signal. The antenna holder is a Plexiglas disk (20 × 3 mm), with two wells (3.5 mm diam.) adjacent to a scission (1.2 mm) across the center. The antenna bridges this slit, its cut ends are locked into the wells filled with Ringer solution (Beadle-Ephrussi).

Ambient air was pulled through the glass chamber and the slit in the antenna holder at 42 ml/sec. For measurement of the resting potential of the antenna, the airstream was filtered with active charcoal (100 g); for measurements of ambient pheromone, the charcoal filter was removed. Data were sampled at 18.2 Hz on a portable computer; peak heights were averaged off-line for the first 2 sec of each recording.

To compare recordings from different antennae, EAG amplitudes were calibrated to a standard stimulus, to obtain relative EAG amplitudes: a 2-ml pulse from a 20-ml syringe, holding a rubber septum loaded with 10 μg *E*8,*E*10–12:Ac (3–5 days old, stored at −20°C), was injected into the charcoal-filtered airstream within the holding chamber. Three such standard stimuli preceded and followed three measurements of ambient air, lasting up to 5 sec.

Field EAG recordings were done with antennae of *C. nigricana* males, during their diel flight period between 5 and 9 PM. Males were collected in the control field and stored at 8°C for one day; each antenna was used during ca. 30 min. Measurements were made at 16 selected locations in the pheromone-permeated test field and the control field, at ca. 20 cm above and below pea canopy. The apparatus was set in the center of 100-m^2 squares, where a monitoring trap had been placed instead of a dispenser (see above). The EAG apparatus thus had a maximal distance to dispensers; the trap was removed during recordings.

Calculation of Absolute Pheromone Concentrations. Pheromone concentrations were calculated from: (1) relative EAG amplitudes in response to ambient air from the test field, and an exponential correlation between (2) relative EAG amplitudes in response to stimuli from syringes containing filter papers loaded with different amounts of *E*8,*E*10–12:Ac and (3) the extrapolated concentrations of *E*8,*E*10–12:Ac in these syringes (Figure 1).

From 5-ml syringes, holding filter papers with 10:Ac (100, 300, 500, and 1000 μg) or *E*8,*E*10–12:Ac (500 and 1000 μg), 2 ml of the headspace volatiles was injected on the GC. Concentrations of *E*8,*E*10–12:Ac in these syringes, at filter paper loads used for EAG recordings (0.1–100 μg), were extrapolated under the assumptions that *E*8,*E*10–12:Ac is approx. 16 times less volatile than 10:Ac (a factor of 4 for each methylene group) and that the release rate is directly proportional to the filter paper load (see Bengtsson et al., 1990). From

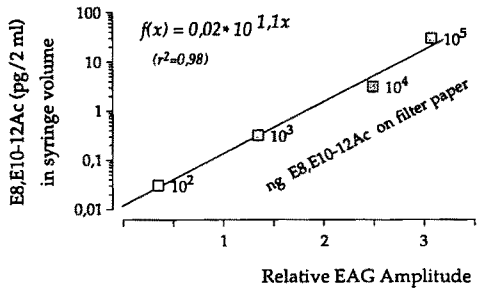

FIG. 1. Correlation between concentrations of *E*8,*E*10–12 : Ac (pg/2 ml) in 5-ml syringes holding filter papers loaded with 10^2–10^5 ng *E*8,*E*10–12 : Ac and relative EAG amplitudes in response to 2-ml pulses from these syringes ($N = 5$). EAG amplitudes were calibrated to the standard stimulus from a syringe holding a rubber septum loaded with 10 μg *E*8,*E*10–12 : Ac.

5-ml syringes, holding filter papers with 0.1–100 μg *E*8,*E*10–12 : Ac, 2 ml was injected into the holding chamber of the EAG apparatus within 0.5 sec. The relative EAG amplitudes to these stimuli were then correlated to the extrapolated *E*8,*E*10–12 : Ac concentrations in these syringes (Figure 1).

On injection into the EAG device, 2 ml of the syringe atmosphere was added within 0.5 sec to an air flow of 42 ml/sec and was therefore diluted by 1 : 23. Absolute concentrations of *E*8,*E*10–12 : Ac within the holding chamber of the EAG apparatus, from air sampled in the field, can thus be calculated from the exponential correlation shown in Figure 1.

All EAG recordings of ambient air in the field and of filter paper stimuli in the laboratory were calibrated to the standard stimulus (relative EAG amplitudes; see above). Both filter papers and rubber septa were kept in the syringes for 1 hr at ambient temperature before use. Error intervals were determined from the estimated experimental inaccuracies (air flow in EAG apparatus, *E*8,*E*10–12 : Ac concentrations in syringe, EAG amplitudes). Calibration of the EAG system is hampered by low sensitivity of the FID, compared to the male antenna: filter paper loads that allow concentration measurements by GC are beyond the response range of the antenna. This method, therefore, gives only an estimate of aerial pheromone concentrations.

RESULTS

The principal parameters of this first mating disruption trial against the pea moth *C. nigricana* are summarized in Table 1.

Release of Disruptant Chemicals. The mean release rate of *E*8,*E*10–12 : Ac

TABLE 1. PRINCIPAL PARAMETERS OF *C. nigricana* MATING DISRUPTION (HÖÖR, 1992)

Insect species	*Cydia nigricana* F. (Lep., Tortricidae)	
Crop treated	0.6-ha pea field (*Pisum sativum* L.)	
Sex pheromone	*E*8,*E*10–12:Ac	Greenway (1984)
Disruptant chemical	*E*8,*E*10–12:Ac	
Impurities		
After 6 days	3% *E*,*Z*-, 2% *Z*,*E*-, 1% *Z*8,*Z*10–12:Ac	Figure 2
After 20 days	4% *E*,*Z*-, 3% *Z*,*E*-, 2% *Z*8,*Z*10–12:Ac	
Activity	Attraction inhibitors (antagonists)	Witzgall et al. (1993)
Dispenser material	Polyethylene tube (Shin-Etsu)	
Dispenser placement	600 disp/ha	
Amount applied	50 mg/dispenser; 30 g/ha	
Release rate		
After 6 days	10 μg/disp/hr; 140 mg/ha/day	Figure 2
After 20 days	6 μg/disp/hr; 82 mg/ha/day	
Aerial concentration	~2 to ~7 ng/m^3 (within canopy)	Figures 1, 4
Communication disruption		
Traps (100 μg *E*8,*E*10–12:Ac)	100% reduction	Figure 5
Male behavior	Repelled from treated field	Table 2
Female behavior	Immigration of mated females	
Infestation		
Control field	67 larvae/m^2	
Treated field	52 larvae/m^2	

from the polyethylene tube dispensers, as measured by glass adsorption in a static atmosphere, was 7.6 ± 2.0 μg/hr between day 6 and day 20 (Figure 2). Isomeric purity of *E*8,*E*10–12:Ac was >99% before and after (day 0) formulation. Six and 20 days after dispenser placement, the nonpheromonal isomers *E*,*Z*; *Z*,*E*; and *Z*8,*Z*10–12:Ac (Witzgall et al., 1993) were released at 5.7% and 8.3% of *E*8,*E*10–12:Ac (Figure 2). This compares to isomerization of *E*8,*E*10–12:OH on polyethylene dispensers (Brown et al., 1992), but isomerization of *E*8,*E*10–12:Ac is faster on rubber septa (Davis et al., 1984; Witzgall et al., 1993).

Pheromone glands of calling *C. nigricana* females contain 0.8 ng *E*8,*E*10–12:Ac (Witzgall et al., 1993) and the release rate from the female gland is 1–5 ng/hr (Witzgall, unpublished). The dispensers used in this experiment thus emitted a 1000- to 10,000-fold amount of *E*8,*E*10–12:Ac.

Field EAG Measurements. A portable EAG apparatus was used to monitor the distribution and concentration of ambient pheromone under field conditions. Figure 3 shows recordings from *C. nigricana* antennae in the control pea field and the pheromone-treated field. The baseline is the resting potential of the antenna in filtered air; removal of the charcoal filter immediately changes the antennal potential, as ambient air is measured. Above the canopy, the antennal

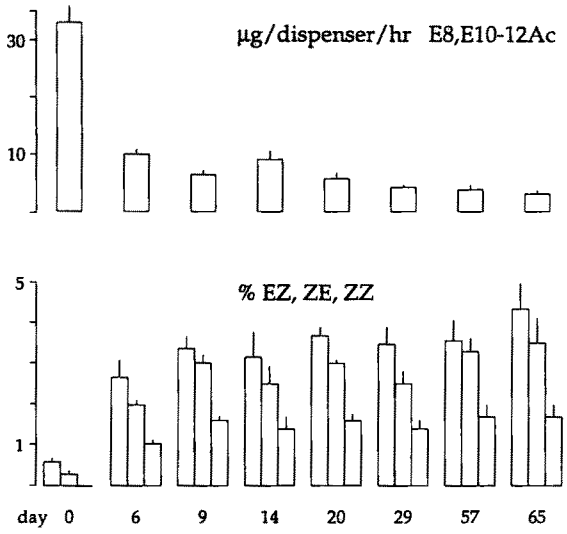

FIG. 2. Release of sex pheromone *E*8,*E*10–12:Ac (µg/hr) and antagonistic *E*,*Z*; *Z*,*E*; *Z*,*Z* isomers (relative to *E*,*E*) from polyethylene dispensers, 0–65 days after field application, as measured by glass adsorption in static atmosphere at 22°C (mean values ± SD; $N = 3$).

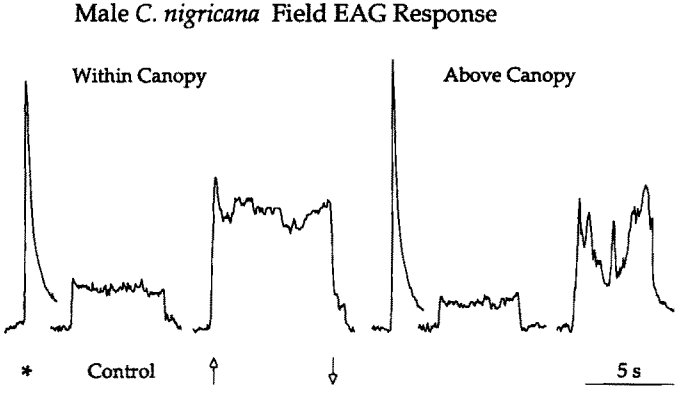

FIG. 3. Male *C. nigricana* field EAG response within the pea canopy and 20 cm above, nine days after dispenser application. Standard stimulus (asterisk: 10 µg *E*8,*E*10–12:Ac on rubber septum), control recording in untreated pea field, recording in test field (arrows: charcoal filter off/on).

signal fluctuated strongly, and overall response was always lower than within the canopy. Perception of plant volatiles probably accounted for the response in the control field.

Top pheromone concentrations built up along the border of the 0.6-ha test field (Figure 4), where 10.2 dispensers/100 m^2 had been placed, compared to 3.3 dispensers/100 m^2 in the middle. Trees protected the field border from wind and sun, peas were >60 cm high, with lush foliage; peas in the middle of the test field were <40 cm high, with sparse foliage.

In the 8-ha control pea field, upwind from the field permeated with pheromone, 4, 8, and 12 dispensers were placed in three isolated plots of 100 m^2. The average EAG response within the canopy of the 12-dispenser-plot was significantly higher than in the two other plots. Measurements above the canopy were not different (Figure 4; $N = 6$, Tukey test, $P = 0.05$).

Homogeneous distribution of pheromone within the pea canopy was probably due to less turbulent and slower air movement and desorption of pheromone from the pea foliage (Wall et al., 1981; Karg et al., 1990): 30 min after removal of the dispensers, relative EAG amplitudes within canopy of the 12-dispenser-plot were 0.35 ± 0.04 ($N = 3$), compared to 0.52 ± 0.08 immediately before removal.

Calculation of Pheromone Concentrations. Correlation of *E*8,*E*10–12:Ac concentrations in syringes and EAG responses to stimuli from these syringes was used to estimate absolute concentrations of ambient pheromone from the field EAG recordings (Figure 1). The average concentration within the pea

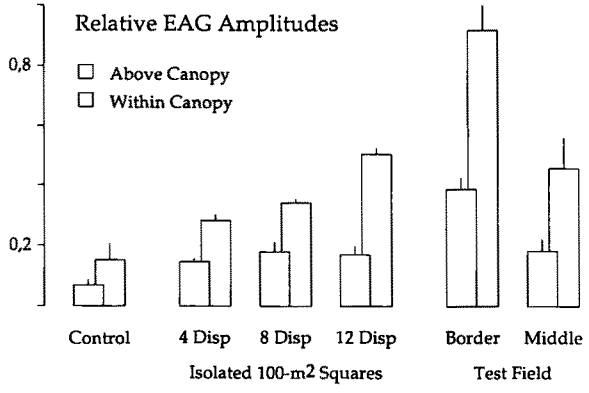

FIG. 4. Relative EAG amplitudes recorded in the centers of 100-m^2 squares, 6–25 days after dispenser application: in the pheromone-permeated test field, 5 m from the border (squares with 12 dispensers) and in the middle of this field (8 dispensers; $N = 18$), and in isolated squares within the control field (4, 8, 12 dispensers; $N = 6$). EAG amplitudes calibrated to standard stimulus (10 μg *E*8,*E*10–12:Ac on rubber septum).

canopy was 7 ± 3 ng *E*8,*E*10–12 : Ac/m^3 on the border, and 2 ± 2 ng *E*8,*E*10–12 : Ac/m^3 in the middle of the disruption field (data from Figure 4).

EAG Response to Nonpheromonal Isomers. The other isomers were tested in the laboratory at 3 to 100 ng on filter paper (half-decadic steps, $N = 4$). The EAG response to the *E*,*Z* isomer was 0.55 ± 0.09, to *Z*,*E* 0.27 ± 0.14 and to *Z*,*Z* 0.35 ± 0.08, relative to equal amounts of *E*8,*E*10–12 : Ac. The contribution of these isomers to the overall field response was estimated to ca. 2% on day 6 and ca. 3% on day 20, according to the proportions at which they were released from the dispensers (Figure 2); tests with binary blends showed no synergistic effect at the antennal level.

Field Trapping. Disruption of pheromonal communication in *C. nigricana* was monitored with 100 μg *E*8,*E*10–12 : Ac on red rubber septa. The dispensers were applied after 28 and 23 males had been trapped in the control and the test field. Trap catch on the border and within the pheromone-permeated field was thereafter completely suppressed throughout the whole flight period (Figure 5). Traps in the centers of the three isolated 100-m^2 plots within the control field, treated with 4, 8, and 12 dispensers, caught one, two, and one males on the first two days after dispensers were applied, but no males at all during the following 23 days.

Male Behavior. In untreated pea fields, male pea moths fly actively over the canopy in sunshine during late afternoons (Bradley et al., 1973; this study).

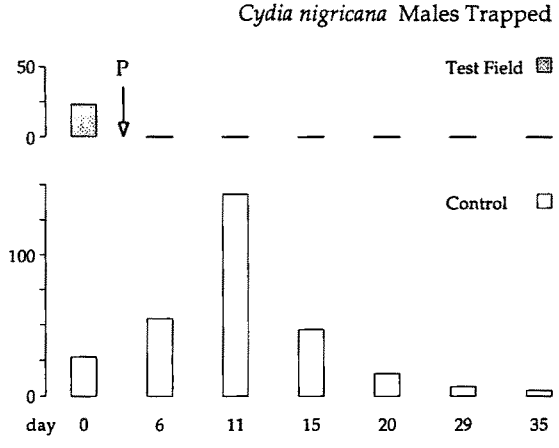

FIG. 5. Field attraction of *C. nigricana* males to traps baited with 100 μg of synthetic sex pheromone *E*8,*E*10–12 : Ac, in the control pea field ($N = 10$) and the pheromone-permeated field ($N = 10$), 0–35 days after dispenser application. Dispenser application (P).

They switch to slower, upwind-oriented flight as they encounter sources of pheromone.

On the first two days after dispenser placement, male moths were attracted to and even landed and wing-fanned on the dispensers or on surrounding pea plants (Table 2). From the untreated field, males flew upwind over several meters to fresh dispensers placed in three isolated 100-m^2 test plots; four males were also attracted to traps within these plots (see above).

In the permeated field (600 dispensers/ha), males were flying among pea plants, frequently alighting and wing-fanning on leaves, also in the vicinity of the dispensers (Table 2). However, directed orientation flights over more than 1 m towards dispensers were not observed. Males terminated searching by flying out of the plot or by resting on leaves. During this period, males may have found calling females, but traps in the test field were not attractive (Figure 5).

Male behavior was greatly changed a few days after dispenser placement. This was probably due to an increasing release of the inhibitory isomers *E,Z*-, *Z,E*-, and Z8,Z10–12:Ac, together with *E*8,*E*10–12:Ac (Figure 2) (Witzgall et al., 1993). Males were then no longer observed within the test field, and the few males arriving to the field border were obviously repelled and flew out rapidly, often 2 to 3 m above the ground. In the control field, *C. nigricana* males were observed throughout the whole flight period (Table 2).

Female Behavior. Mated females are attracted to peas, especially to the flowering parts, by upwind-oriented flight. They deposit single eggs on or near flowers and young pods (Bovey, 1972; this study). This behavior was unchanged in the presence of disruptant. Calling females were not seen in the control or test field.

Larval Counts. To facilitate behavioral observations, we chose field sites

TABLE 2. COUNTS OF ADULT *C. nigricana* IN MATING DISRUPTION AND CONTROL PEA FIELD

	Control field	4-dispenser plot	Test Field		
			Border	Border	Middle
Observation period[a]	6–17	0–1	0–2	6–17	6–17
Duration (hr)	9	5	6	11	5
Males/hour (±SD)	6.6 ± 3.5	14.4 ± 6.8	4.3 ± 4.0	0.8 ± 0.9	0
Males < 30 cm from dispenser	—	10.3 ± 8.2	3.0 ± 3.2	0	0
Females/hour	4.9 ± 2.7	[b]	[b]	7.2 ± 3.1	3.6 ± 2.1

[a] Days after dispenser placement.
[b] Not registered.

with high population densities and installed dispensers only one week after the onset of the flight period. The test field (0.6 ha) was 50 m downwind from the control pea field (8 ha), where 44 ± 14% of the pods were infested after the end of the flight period (67 larvae/m^2). Larval attack in the test field was not reduced significantly (34 ± 8%; 52 larvae/m^2) and we assume that this was due to immigration of gravid females.

DISCUSSION

The sexual communication of the pea moth, *C. nigricana*, was suppressed over the entire flight period by permeating a pea field with a blend of synthetic sex pheromone, *E*8,*E*10–12:Ac, and the antagonistic geometric isomers *E*,*Z*-, *Z*,*E*-, and *Z*8,*Z*10–12:Ac. Our results denote the potential of the mating disruption technique for the control of *C. nigricana*, as long as immigration of mated females is prevented, for example, in isolated fields.

A portable EAG system (Sauer et al., 1992), using the male antenna as detector, made it possible to monitor the fine-scale distribution and aerial concentration of disruptant chemical in the field. EAG recordings immediately "visualize dispersal of the disruptant chemical" (Arn, 1990), and greatly facilitate optimization of dispenser placement and interpretation of male behaviors. A disadvantage of the EAG method is the difficulty of calibrating the antennal response. Absolute concentrations of airborne disruptant may be more accurately determined by sampling several cubic meters of air over a few hours for subsequent chemical analysis (Caro et al., 1980, 1981; Wiesner et al., 1980).

Male *C. nigricana* behaviors changed as *E*8,*E*10–12:Ac isomerized within a few days in the field. Release of rather pure *E*,*E* isomer attracted males to dispensers and stimulated close-range search behavior, but release of *E*8,*E*10–12:Ac plus >5% of the inhibitory isomers *E*,*Z*-, *Z*,*E*-, and *Z*8,*Z*10–12:Ac (Figure 2) (Witzgall et al., 1993) repelled males from the treated plots. In *C. nigricana*, a blend of pheromone plus attraction inhibitors is thus a more efficient disruptant than pheromone alone.

A potent attraction inhibitor in the larch casebearer, *Coleophora laricella*, did not induce behavioral responses by itself, but acted as a strong repellent even to resting males when blended with sex attractant (Priesner and Witzgall, 1984). Upwind orientation of male *C. laricella*, *Trichoplusia ni*, and *Epiphyas postvittana* was suppressed by a blend and not by separate sources of attractant and inhibitor (Priesner and Witzgall, 1984; Witzgall and Priesner, 1991; Liu and Haynes, 1992; Rumbo et al., 1993). In the antennal lobes of the corn earworm, *Helicoverpa zea*, a distinct class of synergist neurons responded to a pheromone/inhibitor blend, but not to separate compounds (Christensen et al., 1991).

Control of lesser peachtree borer, *Synanthedon pictipes*, by mating disruption with sex pheromone *E*3,*Z*13–18:Ac was superior to insecticide treatment (Pfeiffer et al., 1991). The formulation contained 30% of *Z*3,*Z*13–18:Ac, a strong attraction inhibitor (Karandinos et al., 1977). Reliable control has been accomplished in the European grape berry moth, *Eupoecilia ambiguella* (Neumann et al., 1988; Neumann, 1990). Composition of the commercial formulation is not available from literature; a blend of sex pheromone *Z*9–12:Ac and at least 2% of inhibitory *E*9–12:Ac (Arn et al., 1986) was used by Rauscher and Arn (1979) and Vogt (1987). Racemic disparlure is being applied against the gypsy moth *Lymantria dispar* (Schwalbe and Mastro, 1988; Webb et al., 1988; Kolodny-Hirsch and Schwalbe, 1990); the (−)-enantiomer of disparlure is a weak behavioral antagonist (Miller and Roelofs, 1978; Preiss and Kramer, 1983). Male gypsy moths have been observed to terminate searching in arrays treated with racemic disparlure by rapidly flying high up into the tree canopy (Cardé et al., 1975).

In analogy to inhibitors, synergistic pheromone components do not initiate specific behaviors by themselves, but enhance male response when blended with the main component (Linn et al., 1986). In a number of lepidopteran species, including the Oriental fruit moth, *Grapholita molesta*, and codling moth, *C. pomonella*, suppression of mating is to date best achieved with a complete pheromone blend (e.g., Charlton and Cardé, 1981; Sanders, 1982; Audemard et al., 1989; Miller et al., 1990; Rice and Kirsch, 1990; Suckling and Clearwater, 1990; Tatsuki, 1990; Barnes et al., 1992; Howell et al., 1992; Pfeiffer et al., 1993).

There are also reports on the successful use of incomplete or imbalanced blends in *Vitacea polistiformis* (Johnson et al., 1991) and *Adoxophyes orana* (Charmillot and Pasquier, 1992). Male pink bollworms, *Pectinophora gossypiella*, intensified searching under permeation with two-component gossyplure, while they appeared inactive in response to the single components (Flint and Merkle, 1983). Similar observations were made for red-backed cutworm, *Euxoa ochrogaster* (Palaniswamy and Underhill, 1988). Application of the minor, but not of the major component, shifted the response of male *P. scutigera* to synthetic two-component blend ratios (Flint and Stone, 1985). Inhibitors alone were used against *Diparopsis castanea* (Marks, 1976) and *C. pomonella* (Hathaway et al., 1985).

Camouflage of natural plumes, competition between natural and synthetic plumes, imbalance of sensory input, and sensory overload have been proposed as the behavioral and physiological mechanisms of mating disruption (Bartell, 1982; Cardé, 1990). Sensory adaptation or central habituation alone cannot explain communication disruption (Charlton and Cardé, 1981; Novak and Roelofs, 1985; Miller et al., 1990), because the males may still use visual and

tactile stimuli (Richerson, 1977; Palaniswamy et al., 1986). False trail following or camouflage of calling females depends on assumptions that are hardly met in the field, i.e., sensorially largely unaffected insects, and unstructured synthetic plumes in the latter case (Bartell, 1982). Plume characteristics and stimulus concentrations are also expected to vary greatly within the crop. Quite obviously, the observed behavioral modifications cannot easily be attributed to distinct mechanisms; several mechanisms are assumed to synergize (Cardé, 1990).

The mechanisms of mating disruption derive from the disruptant chemicals—in relation to the pheromone composition of each species. Different chemicals, release rates, and dispenser densities result in different behaviors (e.g., Charlton and Cardé, 1981; Flint and Merkle, 1983; Palaniswamy et al., 1983; Flint and Stone, 1985; Palaniswamy and Underhill, 1988; Schwalbe and Mastro, 1988). Other variables are the insect's age and the duration of exposure to disruptant.

"If the proposition is accepted that a better understanding of the underlying mechanisms is desirable in order to design more robust systems of pest control through communication disruption, then it is clear that much work remains to be done at a fundamental level" (Bartell, 1982). This is still true in 1994. Such fundamental research depends most of all on the assessment and interpretation of behaviors. Field studies are difficult to achieve with night-active insects, and the natural milieu cannot be simulated in the laboratory; tools to identify the combined effects of adaptation, habituation, and disorientation are not yet available.

Pheromones are successfully applied against a number of pest species, and such mating disruption systems can be described by a few basic parameters. These must be assessed in order to interpret and optimize experiments, but also because they underlie the behavioral modifications to be studied. They can be measured with current techniques but are most often incompletely available from literature: (1) chemical composition of the disruptant and its behavioral effects at low doses, compared to natural pheromone; (2) dispenser placement, release rate, aerial concentration and distribution of disruptant; and (3) degree of communication disruption, as measured by pheromone-baited traps or live females, in relation to population density. Comparative analysis of these parameters from different applications and species may provide immediate input for further development.

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