

# SYNTHETIC PHEROMONES DISRUPT MALE *DIORYCTRIA* SPP. MOTHS IN A LOBLOLLY PINE SEED ORCHARD

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## Abstract

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Synthetic sex pheromones released in a loblolly pine, *Pinus taeda* L. (Pinaceae), seed orchard interfered with the ability of male coneworm moths, *Dioryctria* Zeller spp. (Lepidoptera: Pyralidae), to locate traps baited with sex pheromones or live females. Pherocon 1C<sup>®</sup> traps baited with synthetic pheromones or live conspecific females were hung near the center of two 1.2-ha circular plots during emergence of *Dioryctria amatella* (Hulst), *Dioryctria disclusa* (Heinrich), and *Dioryctria merkei* (Mutuura and Munroe). In a paired design, trap catches for the mating-disruption treatment with synthetic pheromone dispensers consisting of three polyvinyl chloride rods placed in every tree were compared with the control treatment. Treatments were alternated at intervals of 2–3 d. Trap catches of *D. amatella* were reduced by 91% when plots were treated with 2.5 g/ha of Z-11-hexadecenyl acetate. Catches were reduced by 99.5% for *D. disclusa* and by 97% for *D. merkei* when plots were treated with 12.5 g/ha of Z-9-tetradecenyl acetate, whereas catches of *D. amatella* were unaffected by this mating-disruption treatment. Daily disappearance of Z-9-tetradecenyl acetate from the dispensers averaged 0.46 g/ha or less. Manually placing dispensers on nylon lines in the tops of trees was an effective method for releasing synthetic *Dioryctria* pheromones in the orchard. These data suggest it may be feasible to prevent mating of *Dioryctria* spp. in pine seed orchards by using synthetic pheromones for mating disruption, but large-scale tests will be required to demonstrate cone protection.

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## Résumé

La libération de phéromones sexuelles synthétiques dans une plantation à semence de pins taeda, *Pinus taeda* L. (Pinaceae), entrave la capacité des mâles de *Dioryctria* Zeller spp. (Lepidoptera : Pyralidae) de localiser des pièges garnis de phéromones sexuelles ou de femelles vivantes. Des pièges Pherocon 1C<sup>®</sup> garnis de phéromones synthétiques ou de femelles vivantes conspécifiques ont été suspendus près du centre de deux enceintes circulaires de 1,2 ha pendant l'émergence de *Dioryctria amatella* (Hulst), *Dioryctria disclusa* (Heinrich) et *Dioryctria merkei*

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(Mutuura et Munroe). Dans un montage pair, des pièges conçus pour perturber les accouplements ont été munis d'appareils dispensateurs de phéromone synthétique constitués de trois bâtonnets de chlorure de polyvinyl et placés dans chaque arbre pour fins de comparaison avec les témoins. Les traitements ont été administrés en alternance à intervalles de 2-3 jours. Les captures de *D. amatella* ont été réduites de 91% après administration de 2,5 g/ha d'acétate de Z-11-hexadécényle. Les captures de *D. disclusa* ont été réduites de 99,5% et celles de *D. merkei* de 97% après traitement à 2,5 g/ha d'acétate de Z-9-décényle, enfin les captures de *D. amatella* sont demeurées inchangées après le traitement perturbateur des accouplements. La diminution d'acétate de Z-9-décényle dans les dispensateurs était en moyenne de 0,46 g/ha par jour ou moins. L'installation manuelle de dispensateurs le long de fils de nylon à la cime des arbres s'est avérée une méthode efficace de diffusion des phéromones synthétiques de *Dioryctria* spp. dans la plantation. Ces résultats indiquent qu'il est possible de perturber la reproduction de *Dioryctria* spp. dans les plantations de pins à semence en utilisant des phéromones synthétiques capables d'empêcher les accouplements, mais il faudra procéder à des tests sur une grande échelle pour démontrer que ce traitement protège les cônes.

[Traduit par la Rédaction]

### Introduction

Coneworms in the genus *Dioryctria* (Lepidoptera: Pyralidae) damage and kill cones of many conifer species throughout North America (Hedlin *et al.* 1980). Four sympatric species of *Dioryctria* infest the cones of loblolly pine, *Pinus taeda* L. (Pinaceae), in the southern United States: *Dioryctria amatella* (Hulst), *Dioryctria clarioralis* (Walker), *Dioryctria disclusa* Heinrich, and *Dioryctria merkei* Mutuura and Munroe (Ebel *et al.* 1980). Coneworm infestations threaten the production of genetically improved seed in southern pine seed orchards.

Males of three species of *Dioryctria* found in loblolly pine seed orchards are attracted to Z-9-tetradecenyl acetate (Z-9-14:Ac) (Hanula *et al.* 1984a); it is the only attractive compound identified from female *D. disclusa* (Meyer *et al.* 1982) and the major component of the sex pheromone of *D. clarioralis* (Meyer *et al.* 1984). The sex pheromone produced by *D. merkei* has not been identified, but this species is also attracted to Z-9-14:Ac (Cameron 1981; Hanula *et al.* 1984a; Meyer *et al.* 1984). The major component of the *D. amatella* pheromone is Z-11-hexadecenyl acetate (Z-11-16:Ac) (Meyer *et al.* 1986). Only about 5 ng of pheromone or less is present in a female moth (Meyer *et al.* 1986), but male *Dioryctria* spp. respond to very small amounts of sex pheromones (Meyer *et al.* 1982, 1984, 1986; Grant *et al.* 1987).

Synthetic pheromones are used to detect and monitor *Dioryctria* populations in pine seed orchards across the southern United States (DeBarr *et al.* 1982; Weatherby *et al.* 1985). For survey purposes traps are baited with 100 µg of Z-9-14:Ac for *D. disclusa*, *D. clarioralis*, and *D. merkei* and 100 µg of Z-11-16:Ac for *D. amatella* (Weatherby *et al.* 1985).

Pheromone-mediated mating disruption reduces damage in pine plantations caused by the ponderosa pine tip moth, *Rhyacionia zozana* (Lepidoptera: Tortricidae) Kearfott (Niwa *et al.* 1988), and the western pine shoot borer, *Eucosma sonomana* (Lepidoptera: Tortricidae) Kearfott (Overhulser *et al.* 1980; Sartwell *et al.* 1980). *Dioryctria* spp. are good candidates for suppression by mating disruption because this control method is most effective for forest insect pests that cause extensive damage at relatively low population levels (Daterman *et al.* 1980). This could be an attractive alternative to chemical insecticides that now are repeatedly applied in seed orchards. Seed orchards are ideal sites for insect control with pheromones or other semiochemicals

because they are usually less than 40 ha in size and the high value of their genetically improved seed crop makes the economics favorable.

Our objective was to determine if a high density of synthetic pheromone dispensers would affect the chemical communication behavior of male *D. amatella*, *D. clarioralis*, *D. disculsa*, and *D. merkei* moths in a loblolly pine seed orchard. Treatments were designed to demonstrate the potential of mating disruption as a technique for protecting cone crops from attacks by these *Dioryctria* spp.

### Materials and Methods

Three mating-disruption field tests were conducted in the Briarpatch loblolly pine seed orchard (33°48'N, 83°16'W), Putnam County, Georgia, United States. There were about 86 trees/ha, and two 1.2-ha circular plots were used for each test. The trees were 15 years old and 10.7–13.7 m tall for the test with *D. amatella*, and 20 years old and 13.7–17 m tall for the tests with *D. disculsa* and *D. merkei*, respectively.

**Pheromone Deployment.** Synthetic pheromone containing less than 2% impurities was incorporated into polyvinyl chloride (PVC) rods at about 5% active ingredient by weight, as described by Daterman (1974). Z-11-16:Ac (Albany International Corporation, Needham, Massachusetts) was used for *D. amatella*, and Z-9-14:Ac (AgriSense Inc., Fresno, California) for *D. disculsa* and *D. merkei*. Each pheromone dispenser consisted of a piece of fiberglass screen folded and stapled to form a 2.5 × 7 cm envelope containing three 5-cm-long pieces of 3-mm-diameter PVC rod. Two dispensers were manually deployed per tree in all tests as follows. Two 1-cm wire eyes, spaced about 0.5 m apart, were twisted onto a primary branch located within 2 m of the top of each tree within the plots. A large loop of nylon cord was threaded through the eyes and tied off on the bole of each tree at 1.5 m above the ground. During mating-disruption periods, two dispensers were attached to small loops in the nylon cord with safety pins and raised so they were located in the upper and midcrown positions. During control periods the dispensers were detached from the lines, removed from the plot, and stored in a freezer.

**Assessment of Mating Disruption.** Male moth response was measured using male catches in Pherocon 1C® (Zoecon Corp., Palo Alto, California) traps baited with synthetic pheromones or live conspecific female moths (*D. amatella* test only) and placed in the upper crowns of 10 trees in each test plot as described by DeBarr *et al.* 1982. Traps baited with synthetic pheromone are as attractive as those baited with feral *D. amatella* (Meyer *et al.* 1986), *D. clarioralis* (Meyer *et al.* 1984), or *D. disculsa* females (Meyer *et al.* 1982). Although not identified from the females, Z-9-14:Ac is a strong attractant (Cameron 1981; Hanula *et al.* 1984a) and most likely the major component of the *D. merkei* pheromone. We assumed that a significant reduction in male moth catch during mating-disruption periods indicated that normal pheromone communication between males and females had been disrupted. Mean numbers of moths caught per trapping period (weighted for number of days in the *D. amatella* test) were ranked and treatment means were compared using the Wilcoxon *T* distribution approximation for the rank-sum test in SAS NPAR1WAY (SAS Institute Inc. 1990).

***Dioryctria amatella.*** Using the paired design of Hendricks *et al.* (1982), we applied the mating-disruption treatment to both plots and continued to alternate it with the control at 2- to 3-d intervals from 10 September to 29 October 1984. A rate of 2.5 g/ha of Z-11-16:Ac was used during mating-disruption periods. Plot 1 had 110 trees and plot 2, located about 0.2 km to the east, had 106 trees.

Six traps were baited with red rubber septa (Thomas Scientific Co., Philadelphia, Pennsylvania) impregnated with 300 µg of Z-11-16:Ac (Meyer *et al.* 1984). Four additional traps were baited with female *D. amatella* moths from a laboratory colony (Fedde 1982) and we assumed that these females were competitive with feral females. Two virgin females held in separate compartments of a small cylindrical wire cage were suspended in the center of each trap. Females were replaced at death or at 7-d intervals, whichever came first. A synthetic pheromone baited trap was hung at the center of the plot and traps with septa or females were alternated in a circular pattern around it. Female-baited traps were located about 26 m from the center of each plot, whereas those baited with synthetic pheromone were about 20 m from the center.

***Dioryctria disclusa.*** Methods and procedures were the same as those for the *D. amatella* test, with the following exceptions. Plot 1 had 107 trees, and plot 2, about 0.4 km to the north of plot 1, had 103 trees. We randomly assigned the mating-disruption treatment to one plot and the control to the other plot, then repeated this procedure at 2-d intervals from 24 May to 12 June 1989. A rate of 12.5 g/ha of Z-9-14:Ac was used during mating-disruption periods.

Traps were baited with red rubber septa loaded with 100 µg of Z-9-14:Ac (Meyer *et al.* 1982) and hung in the upper crowns of the 10 trees nearest to the center of each test plot. Distances from the plot center to the traps varied from 7 to 20 m. Release of synthetic pheromone over the trapping period in this test was estimated by comparing the amount of Z-9-14:Ac in 10 PVC rods held in the laboratory at -30°C with the amount found in PVC rods from 10 dispensers that were randomly selected from those used in the orchard. A piece weighing about 150 mg was cut from the center of each PVC rod and extracted in 3 mL of hexane for 24 h. The quantity of pheromone was then determined by injecting measured hexane fractions into a Hewlett Packard 5880A gas chromatograph (GC) (Hewlett Packard, McMinnville, Oregon) equipped with a flame ionization detector. Analyses were made on a 30 m × 0.25 mm i.d. (internal diameter) capillary column (DB-1, J&W Scientific Co., Folsom, California) with Z-6 heneicosen-11-one as an internal standard.

***Dioryctria merkei.*** This test was conducted from 21 August to 21 October 1989. Plots and procedures were the same as those for the *D. disclusa* test, but new dispensers, trap bottoms, and rubber septa impregnated with 100 µg of Z-9-14:Ac were used (Hanula *et al.* 1984a; Meyer *et al.* 1984). On 27 September 1989, new trap bottoms and red rubber septa impregnated with 100 µg of Z-11-16:Ac (Meyer *et al.* 1986) were installed to determine if the Z-9-14:Ac dispensers affected the mate-locating ability of *D. amatella* males.

## Results

***Dioryctria amatella.*** Trap catches of *D. amatella* were low during the test with Z-11-16:Ac (Fig. 1A). Twenty-eight males were caught in traps baited with laboratory-reared females and 20 were caught in traps baited with synthetic pheromone, but only four were caught during mating-disruption periods. Catch of *D. amatella* males was reduced by 91% during the mating-disruption periods and was lower than the catch during control periods ( $P > |T| = 0.0002$ ).

***Dioryctria disclusa.*** Trap catch of *D. disclusa* was reduced by 99.5% during the mating-disruption periods with Z-9-14:Ac. Male catch totaled 1507 in the control periods compared with 8 during the mating-disruption periods (Fig. 1B) ( $P > |T| = 0.0006$ ). GC

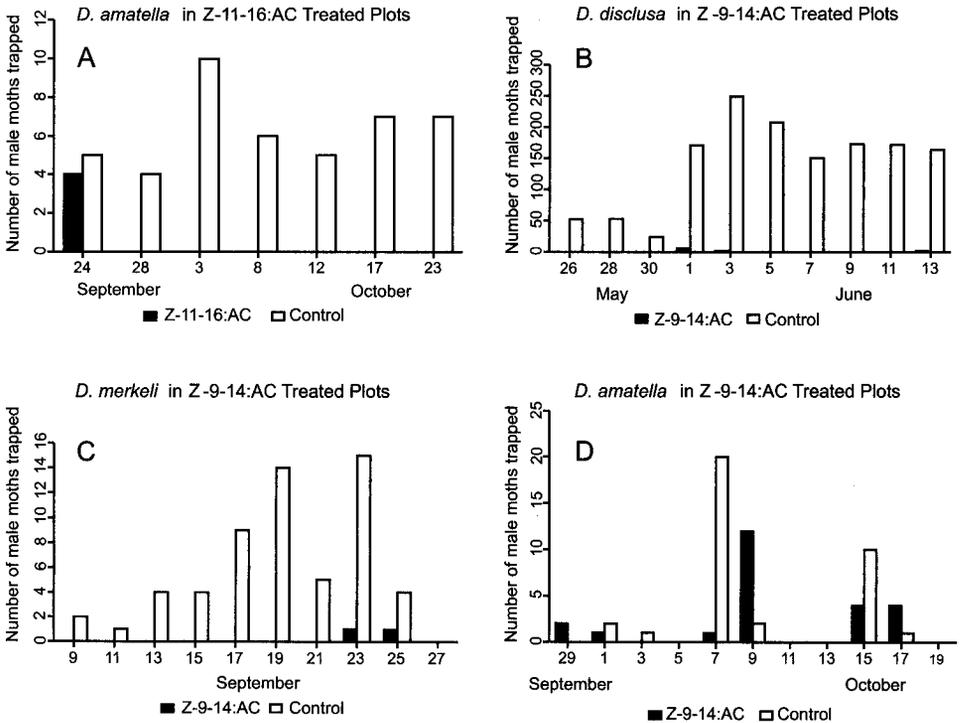


FIGURE 1. Numbers of male *Dioryctria* spp. moths caught in Pherocon 1C<sup>®</sup> traps baited with synthetic pheromone or live females during mating disruption (Z-9-14:AC or Z-11-16:AC released) or control (no pheromone released) in a *Pinus taeda* seed orchard, Putnam County, Georgia. Note the difference in scale of the y axes.

analyses of the PVC rods containing Z-9-14:Ac revealed that 74% of the pheromone disappeared from the dispensers. Assuming no loss to degradation and 100% recovery, the daily release rate was 0.458 g/ha, with a total release of 9.16 g/ha during the 20-d exposure period.

***Dioryctria merkeli*.** Of a total of 60 *D. merkeli* males trapped from 21 August to 27 September (Fig. 1C), all but two were caught during control periods. Trap catches differed between the mating-disruption and control periods ( $P > |T| = 0.0031$ ). Catches of *D. amatella* males totaled 60 from 27 September to 21 October (Fig. 1D), but did not differ between mating-disruption and control periods ( $P > |T| = 0.9756$ ). Hanula *et al.* (1984a) found that the addition of Z-9-14:Ac to traps baited with Z-11-16:Ac reduced the catch of *D. amatella* by about 50%.

## Discussion

Two dispensers placed in the upper tree crowns in our mating-disruption treatments significantly reduced the ability of male moths to locate pheromone or female-baited traps in a loblolly pine seed orchard at rates of 2.5 g/ha of Z-11-16:Ac for *D. amatella* and 12.5 g/ha of Z-9-14:Ac for *D. disclusa* and *D. merkeli*. The response of *D. clariorailis* males could not be evaluated because too few male moths were trapped.

Preemptive, area-wide pest management tactics, such as mating disruption, require relatively large plot sizes to be effective, thereby limiting opportunities for spatial

or temporal replication (Schneider 1989). Our 1.2-ha orchard plots were too small to prevent encroachment by mated gravid female moths from surrounding untreated areas which can reduce or negate the effectiveness of a mating-disruption strategy. We used small plots for these tests because they are useful first steps in assessing mating disruption before undertaking large field trials (Roelofs and Novak 1981). Opportunities for testing the efficacy of mating disruption for coneworm control in seed orchards have been limited, but now that advanced generations of loblolly pine orchards produce most of the seed crops, older orchards may be available for mating-disruption tests. The potential success of mating disruption is greater in seed orchards which are relatively isolated, limiting immigration by gravid females.

Aerial applications of pheromone dispensers have been highly successful for controlling forest insects infesting large areas of pine plantations (Overhulser *et al.* 1980), but in pine seed orchards manual distribution of pheromone dispensers would also be practical. Permanent release points could be established throughout an orchard and pheromone dispensers could be raised on cords to the tops of trees during emergence periods for *Dioryctria* spp. or other species of cone and seed insect pests. Previous research has shown that traps must be located in the tops of orchard trees to be effective (DeBarr and Berisford 1981; Hanula *et al.* 1984b; Grant *et al.* 1987). If dispensers for mating disruption of *Dioryctria* spp. must also be in the tops of the trees, manual deployment may be a better method for permeating seed orchards with semiochemicals than aerial applications, which are likely to distribute much of the pheromone in the lower crowns or on the ground.

These tests are the first to suggest that mating disruption to control *Dioryctria* may be feasible in pine seed orchards. The inability of male moths to locate synthetic baits or females in traps in an orchard with a high density of synthetic pheromone dispensers is an indirect measure of mating success, but the measured effect is a prerequisite to successful control of *Dioryctria* by mating disruption. Our positive results suggest that it is worthwhile to pursue mating disruption as a pest management alternative to insecticides in seed orchards.

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