Female Grape Root Borer (Lepidoptera: Sesiidae) Mating Success Under Synthetic Sesiid Sex Pheromone Treatment[1]

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ABSTRACT Field observations of virgin female grape root borers (*Vitacea polistiformis* Harris) were made throughout daylight periods under different pheromone treatments and doses. The pheromone compounds used were: the grape root borer pheromone, a 99:1 blend of E,Z-2,13-octadecadien-1-ol acetate (EZ) and Z,Z-3,13-octadecadien-1-ol acetate (ZZ); EZ alone; and ZZ alone. Two different pheromone dispensers were used – rubber septa and Shin-Etzu twist-tie ropes. Treatments with 99:1 EZ: ZZ and 100% ZZ significantly reduced the ability of females to attract a mate. Shin-Etzu ropes were the most effective dispenser for prevention of mating.

KEY WORDS Insecta, sex pheromone, *Vitacea polistiformis* Harris

The grape root borer, *Vitacea polistiformis* (Harris), is one of the most important pests of grapes in North Carolina. The larvae of this native sesiid moth bore into the roots of vines, where they feed for two years before pupation and emergence. Feeding reduces vine vigor and cold tolerance, increases susceptibility to pathogens and drought, and hastens vine death. Because the insect is sheltered within the vine tissue for most of its life cycle, only adult moths and newly-hatched larvae are accessible for pesticide treatment to suppress populations. Although chlorpyrifos treatments have given successful control elsewhere (All et al. 1982), many factors (soil type, labor costs, and preharvest application interval) make this method unacceptable in North Carolina. Use of grape root borer pheromone for mating disruption is under consideration as an alternative control method.

The grape root borer pheromone has been identified as a 99:1 blend of E,Z-2,13-octadecadien-1-ol acetate (EZ) and Z,Z-3,13-octadecadien-1-ol acetate (ZZ) (Schwartz et al. 1983, Snow et al. 1987). Because of the grape root borer's two-year life cycle, efficacy of disruption treatments applied to one generation cannot

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be evaluated until two years later. For this reason, field observations of virgin female moths were made during mating disruption trials to evaluate immediately the efficacy of both grape root borer pheromone compounds at controlling grape root borer populations. Results of these field evaluations are reported in this paper. Observed behavioral changes of virgin females under pheromone treatments were described in Pearson (1992).

Materials and Methods

Observers recorded female calling and mating success under different pheromone treatments in 1988-1991 in a 0.53 ha vineyard of initially uninfested 2-yr-old vines located in Raleigh, NC. Female moths were collected from a heavily-infested vineyard 2 km from the treatment vineyard. The source vineyard was searched for newly-emerged females from 0900-1200 EDT each observation day. This source vineyard was treated with pheromones in 1988 and 1989 as part of a separate disruption experiment. In 1988, the source vineyard air was permeated with 99:1 blend EZ:ZZ (254 dispensers/hectare (dis/ha)). In 1989, it was treated with a 95:5 blend of EZ and E,Z-2,13-octadecadien-1-ol alcohol at the same rate. Use of this second compound was due to a manufacturer's formulation error, which was not discovered until after the 1989 season.

In 1990 and 1991, the source vineyard was untreated; however, the females collected in these years were the progeny of treated males and females in 1988 and 1989, respectively, as the grape root borer has a 2-yr life cycle. Examination of emergence patterns showed few new sites of oviposition (Pearson 1992). The effects of these pre-treatments should be negated within-years, because both control and treated females were drawn from this source vineyard.

Collection of virgin (0-d-old) female moths was ensured by collecting only emerging females with uninflated wings, or newly-emerged females near fresh pupal skins at the base of the vines. Non-virgin females were readily detected by their habit of flying when disturbed, or laying large numbers of eggs in the collection box. Virgin females usually sat motionless in the box. Observers placed females on grape leaves in plastic boxes 81 cm diam and 9 cm high, and sheltered under a shady vine to protect them from the sun. Lids were propped open with grape leaves, providing a 2 mm air-space between lid and box. Occasionally females were placed two per box.

Weather conditions at the source vineyard determined the actual time of female emergence. If the weather was cool, emergence began later in the day. Even though the search period ranged from 0900-1200 EDT, actual collection time averaged 1.5 h, so females generally spent less than 1.5 h in collection boxes.

The collection boxes containing females were transported to the observation vineyard once 10 females had been collected or 1200 EDT was reached, whichever came first. Between 1130 and 1200, a leaf with a female sitting on it was removed from each of the collection boxes and placed at the base of one of several designated observation vines. Field technicians pruned these vines to remove excess suckers and shoots, but retained enough foliage to keep the trunk in complete shade during the day. This allowed observers to follow the movements of the insects without exposing the females to direct sunlight (females exhibit a negative taxis to direct sunlight). Females were rotated daily within the vineyard so they were not observed on the same vine on consecutive days. Nearby vines not used for observations were pruned severely so a flying female could be easily located if she alighted on them. In this way the females were free to move about with an observer following their progress.

From this initial release point, each observer noted changes in position or behavior of one or more females every 5 min until 1700 or 1800 h, when females ceased calling, or called only sporadically. On average, each observer watched three insects simultaneously, with a maximum of five per observer. Virgin female grape root borer are relatively large bodied (2-3 cm) and adopt a distinct calling posture, in which the abdomen is lifted and pheromone gland/ovipositor extended. To minimize observer effects, an experienced observer (G.A.P.) trained field technicians in spotting this and other sterotypic behaviors. Records were compared until inter-observer agreement exceeded 95%.

Observers monitored female behavior in the presence or absence of air permeated by 100% EZ, 100% ZZ, or EZ:ZZ blended in 99:1 ratio. These synthetic pheromone treatments were released from rubber septa (1 mg/septum) or Shin-Etzu twist-tie ropes (80 mg/rope). Rubber septa dispensers were added because of the unreliability of the rope formulation chemistry. Each dispenser was hooked to double-headed nails on trellis posts (165 cm elevation) at densities of 254 or 508 dis/ha. Field technicians pruned back vine foliage near pheromone dispensers so leaves or vine shoots were no closer than 30 cm from the dispenser. Shin-Etzu ropes released pheromone at a nearly linear rate, averaging 0.21 mg/dis/d over an expected life of 120 to 150 d (Johnson et al. 1991). Output from rubber septa declines exponentially from initial values of 0.025 to 0.03 mg/dis/d over an expected life of 60 d (McDonough et al. 1989). This release decline should not be a factor in this study, because the rubber septa were used only for 2 to 7 d.

Different pheromone treatments were deployed each wk during the grape root borer flight season (mid-July through late-August). A 3 to 4 d period between treatments presumably allowed dispersal of pheromones. Control observations of females were made 1 d before placement of pheromone dispensers. This procedure established that most pheromone had dissipated from the vineyard and mating was not disrupted. Control observations were thus paired with a temporally adjacent treatment. Because the treatments were conducted in sequence throughout a summer, each treatment was temporally bracketed by control observations. Space and time considerations made simultaneous observation of treated and untreated females impossible.

Three pheromone treatments were used: the grape root borer pheromone (EZ: ZZ), and its individual components (ZZ alone and EZ alone). The experiment was conducted over 4 consecutive yrs (1988-1991) so that all available treatment combinations (3 pheromone blends X 2 dispenser types X 2 dispenser densities) could be tested and replicated. 100% EZ was not available in Shin-Etzu rope formulation, so the experimental design included only 10 of the 12 possible treatments. ZZ and EZ treatments at 254 dis/ha were not fully replicated and results are not reported here.

Field technicians observed a total of 122 female grape root borer under Shin-Etzu rope dispenser treatments; 129 females under rubber septa dispenser treatments; and 155 females under control conditions.

Data Analysis. Length of call before mating under different treatments was tested for association with treatments and daily weather parameters. These weather data were: maximum, minimum, and average temperature (°C), rainfall (cm), barometric pressure (cm), average wind speed (kph), maximum wind speed (kph), total min of sunshine, and percent of possible sunshine. The National Oceanic and Atmospheric Administration weather station at Raleigh-Durham International Airport, approximately 24 km from the observation vineyard, provided weather data. All variables were entered into the model, removing the least significant after each run, in a manual approximation of the stepwise regression procedure with the SAS procedure GLM (SAS Institute 1985).

Mating success under different treatments was compared with Chi-square tests, as these were discrete data (either mated or unmated).

Results

Disruption Success. All virgin females mated during control periods, usually within 10 minutes of call initiation (Tables 1 and 2). Female mating success under Shin-Etzu rope treatment periods is shown in Table 1. A significant level of mating disruption was achieved by all rope pheromone treatments (X^2 = 38.97; df =1; P = 0.0001). Significant year effects also were found (X^2 = 13.21; df =1; P = 0.0003).

Female mating success under rubber septa treatments ranged from 55 to 93% (Table 2). All septa pheromone treatments significantly reduced mating success (X^2 = 40.15; df = 1; P = 0.0001). Significant year effects were again seen (X^2 = 14.48; df = 1; P = 0.0001).

Length of Call Before Mating. Females which did mate during Shin-Etzu rope treatment periods called for a significantly longer time than control females before mating (Table 3). Pheromone treatments (F = 7.0; df =4,48; P = 0.0002), minimum temperature (F = 5.5; 1,48; P = 0.02), and average temperature (F = 4.4; df = 1,48; P = 0.04) affected the length of call before mating. No year effects were found.

Females which mated during rubber septa treatment periods also called significantly longer than controls (Table 3). Treatments (F = 7.9; df = 6,123; P = 0.0001) and average wind speed (F = 20.4; df = 1,123; P = 0.001) were associated with the length of call before mating. As wind speed increased, the length of call decreased. No year effects were found.

Discussion

The use of pheromones for control of the grape root borer appears to have potential in North Carolina. The mating suppression seen in the Shin-Etzu rope dispenser trial here matches results in earlier whole vineyard disruption trials in Arkansas with ZZ Shin-Etzu rope dispensers (Johnson et al. 1991).

However, in North Carolina vineyard trials with Shin-Etzu rope dispensers conducted concurrently with this study, populations declined only 54% under 99:1 EZ:ZZ Shin-Etzu rope treatments, and 11% under ZZ (254d/ha) Shin-Etzu rope treatments (Pearson 1992). The observations made in the collection vineyard, which was one of the vineyards under treatment in those trials, closely matched the reported mating success of the trials: 54% of females under observation mated (Table 1), and a 54% population reduction was observed in this vineyard.

Table 1. Mating disruption success for treatments with Shin-Etzu rope dispensers. Treatments are reported in the order they were run (dis/ha=dispensers per hectare).

Treatment	N observed	# Mated	% Mated	Mean time (min) from call initiation to mating (SE)	Range (min)
1988					
EZ:ZZ*	11	6	54	46.6 (19.9)	15-145
Control-1	12	12	100	8(3.8)	0-40
ZZ (508 dis/ha)	29	0	0	n/a	n/a
Control-2	10	10	100	11.9 (3.2)	0-32
EZ:ZZ	12	0	0	n/a	n/a
1989					
Control	5	5	100	26.4 (5.4)	25-45
EZ:ZZ	7	0	0	n/a	n/a
1990					
Control	4	4	100	21.7 (10.4)	6-52
EZ:ZZ	6	0	0	n/a	n/a
1991					
Control-1	6	6	100	9.7 (3.5)	2-36
EZ:ZZ	19	3	15.8	53.2 (32.3)	0-131
Control-2	9	9	100	21 (5.1)	8-46
ZZ (508 dis/ha)	12	2	16.7	107.5 (88.5)	19-196
EZ:ZZ	8	0	0	n/a	n/a
Control-3	7	7	100	10.4 (1.9)	6-21

* Observations were made in the 12-yr-old source vineyard, which was under treatment with 99:1 EZ: ZZ< 254 dis/ha. It is considered a separate treatment in this and subsequent analyses.

Table 2. Mating disruption success for treatments with rubber septa dispensers. Treatments are reported in the order they were run (dis/ha=dispensers per hectare).

Treatment	N observed	# Mated	% Mated	Mean time (min) from call initiation to mating (SE)	Range (min)
1989					
Control	5	5	100	26.4(5.4)	15-45
EZ:ZZ 254d/ha	10	4	40	97.6(34.2)	29-170
1990					
Control-1	7	7	100	12.6(3.4)	6-29
EZ:ZZ 254d/ha	16	15	93	24.8(4.5)	9-65
EZ:ZZ 508d/ha	11	10	91	38.1(11.5)	0-119
Control-2	7	7	100	3.4(2.1)	0-10
EZ 508d/ha	22	21	95	32.7(4.1)	7-71
Control-3	7	7	100	11.5(4.1)	0-27
ZZ 508d/ha	16	9	56	89.1(28.2)	10-201
Control-4	4	4	100	21.7(10.4)	6-52
1991					
Control-1	6	6	100	9.7(3.5)	2-26
EZ:ZZ 508d/ha	12	10	83	38.9(5.0)	16-63
Control-2	9	9	100	18.6(5.1)	8-46
ZZ (508 dis/ac)	9	5	55	79.6(18.9)	6-113
Control-3	7	7	100	10.4(1.9)	6-21
EZ 508d/ha	8	5	62	34.4(9.7)	15-68

Table 3. Means for length of call before mating (min), adjusted for temperature effects, under treatments with Shin-Etzu rope dispensers (dis/ha=dispensers per hectare).

Treatment	Mean	P: Trt = Control
Shin-Etzu rope dispensers		
Control	20.6	–
EZ:ZZ (254 dis/ha)*	60.0	0.005
EZ:ZZ (254 dis/ha)	43.1	n.s.
ZZ (508dis/ha)	138.6	0.0001
Rubber Septa Dispensers		
Control	27.2	–
EZ:ZZ (254 dis/ha)	29.4	n.s.
EZ:ZZ (508 dis/ha)	62.5	0.0001
ZZ (508 dis/ha)	61.8	0.008
EZ (508 dis/ha)	31.0	n.s.

* Observations were made in the 12-yr-old source vineyard, which was under treatment with 99:1 EZ: ZZ, 254 dis/ha. It is considered a separate treatment in this analysis.

Why was there is such a disparity between the observation vineyard mating success (0%) and the collection vineyard (54% mating success)? This could be explained by two factors. First, there was a very large grape root borer population in the collection vineyard. A larger number of males would increase a female's chance of being found; North Carolina historically has recorded the highest grape root borer population in the U.S. (Snow et al. 1991). Second, there was a much greater foliage mass in the collection vineyard, which was 10 years older than the observation vineyard. Dense foliage mass would inhibit uniform pheromone dispersal.

Septa dispensers were ineffective at suppressing mating; however, given the small amount of pheromone released by these dispensers, this is not surprising. Interestingly, at low doses, the individual pheromone components were more effective at disrupting mating than the complete blend.

Females often mated without any visible call (Tables 1 and 2). Perhaps these matings resulted from females who emitted small amounts of pheromone surreptitiously, or had pheromone adsorbed to their body or surrounding foliage. Some pheromone or other chemical cue seems to adhere to female pupal skins. These skins are attractive to males and initiate circular searching flight around the base of a vine (Pearson 1992).

Observations of males flying within 1 to 2 cm of clearly-exposed females but failing to find them suggest that male visual orientation has a minor role in mating behavior. Male refusal by females was seen 3 times in control tests, but never occurred in any treatment tests.

In conclusion, while mating disruption is possible in young North Carolina vineyards with a low grape root borer population, in larger, denser vineyards with an established population, it may not be as effective as reported elsewhere.

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