

However, breeding pairs establish galleries at different times. The release rates of verbenone and aggregation pheromones of breeding pairs are not in synchrony. As a host tree becomes fully colonized, the total release rate of aggregation pheromones from all breeding pairs on the host decreases as the release rate of verbenone increases. Over time, proportionally more beetles deter from the initial host and attack adjacent trees. When a host is fully colonized, the total release rate of verbenone from all breeding pairs on the initial host far exceeds the release rate of aggregation pheromones. Beetle attacks on the initial host cease at that time, and all responding beetles are diverted to adjacent trees.

Beetles are exposed to various doses and ratios of aggregation and antiaggregation pheromones during an attack sequence. If the density regulation hypothesis is correct, then variations in these doses and ratios should be correlated with host quality as determined by densities of beetles on hosts. Selection should favor individuals that utilize this information by exhibiting a dose-dependent response to the antiaggregation pheromone verbenone. We hypothesized, therefore, that verbenone should interrupt the attraction of *D. ponderosae* to its aggregation pheromones in a dose-dependent fashion. Dose-dependent interruption of *D. brevicomis* LeConte to its pheromones by verbenone seems to occur in California (Paine & Haulon 1991).

Individuals of sympatric species may also show a dose-dependent response to verbenone. *Ips lactidens* (LeConte) and the pine engraver, *I. pini* (Say), are often associated with *D. ponderosae*, often breeding in the same hosts (Furriss & Carolin 1980). However, all three species tend to be found in single-species assemblages. Spatial separation of a host occurs, possibly to minimize interspecific
Table 1. Description of semiochemical-releasing devices

<table>
<thead>
<tr>
<th>Device</th>
<th>Chemical</th>
<th>Description</th>
<th>Release rate (mg/d)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Verbenone (+17-83)</td>
<td>Open polypropylene centrifuge tube (1.5 ml) containing one disposable pipette (2 µl)</td>
<td>0.03</td>
</tr>
<tr>
<td>2</td>
<td>Verbenone (+17-83)</td>
<td>Open polypropylene centrifuge tube (1.5 ml) containing one 2-ml long glass capillary tube (1.5 mm i.d.)</td>
<td>0.09</td>
</tr>
<tr>
<td>3</td>
<td>Verbenone (+17-83)</td>
<td>Polyethylene/syrax bubble cap</td>
<td>0.6</td>
</tr>
<tr>
<td>4</td>
<td>Verbenone (+17-83)</td>
<td>Open polypropylene centrifuge tube (1.5 ml) containing five 2-ml-long glass capillary tubes (1.5 mm i.d.)</td>
<td>0.04</td>
</tr>
<tr>
<td>5</td>
<td>Verbenone (+17-83)</td>
<td>Black polyethylene bubble cap</td>
<td>13.50</td>
</tr>
<tr>
<td>6</td>
<td>Verbenone (+17-83)</td>
<td>White polyethylene bubble cap</td>
<td>3.00</td>
</tr>
<tr>
<td>7</td>
<td>Verbenol (+17-83)</td>
<td>Polyethylene bubble cap</td>
<td>2.9</td>
</tr>
<tr>
<td>8</td>
<td>cis-Brevicomin (+50-50)</td>
<td>Laminar lure</td>
<td>0.50</td>
</tr>
<tr>
<td>9</td>
<td>Isopulegol (+50-50)</td>
<td>Polyvinyl bubble cap</td>
<td>0.2</td>
</tr>
<tr>
<td>10</td>
<td>Isopulegol (+50-50)</td>
<td>Polyvinyl bubble cap</td>
<td>0.2</td>
</tr>
<tr>
<td>11</td>
<td>β-Mycene</td>
<td>Polyethylene screw-cap bottle (15 ml)</td>
<td>281.0</td>
</tr>
</tbody>
</table>

Laminar lures supplied by Heron Environmental Company (Emigsville, PA).
* All chemical purities >99%.
* At 24-28°C.
* Each capillary tube and pipette was scaled at one end and filled with verbenone.
* 13.57 mixture of cis- and trans-verbenone.

competition among brood (Rankin & Borden 1994). Verbenone interrupts the attraction of I. pini to its pheromone, ipsdienol (2-methyl-6-methylene-2,7-octadien-4-ol) (Borden et al. 1992). We hypothesized that this phenomenon for L. pini should be dependent on the release rate of verbenone. We further hypothesized that verbenone should also interrupt the attraction of I. latidens to its pheromone, ipsenol (2-methyl-6-methylene-7-octadien-4-ol) (Miller & Borden 1990, Miller et al. 1991), and that interruption should be exhibited in a dose-dependent fashion.

Materials and Methods

Semiochemical-Releasing Devices. All devices used to release semiochemicals were supplied by Phero Tech (Delta, BC) (Table 1). Release rates for devices 8, 9, and 10 were determined by collection of volatiles on Porapak-Q and analysis by capillary gas chromatography. Release rates for all remaining devices were determined by weight loss analyses. Mycene is a host kairomone for D. ponderosa (Billings et al. 1978).

Experiments. We conducted three experiments, one for each hypothesis. Experiments 1, 2, and 3 were targeted for D. ponderosa, L. pini, and I. latidens, respectively. In each experiment, blocks of six multiple-funnel traps (Phero Tech) (Lindgren 1985) were set at least 100 m apart in stands of lodgepole pine near Princeton, BC. Traps were spaced 10–15 m apart in grids of 2 by 3 within each block. Twelve-unit traps were used in experiment 2; eight-unit traps were used in experiments 1 and 3. Each trap was at least 2 m from any tree and suspended such that the bottom of each trap was 0.2–0.5 m above ground level.

Experiment 1 consisted of four blocks of traps, set from 1 September to 30 October 1988. Experiment 2 consisted of eight blocks, set from 3 to 29 August 1990. Experiment 3 consisted of five blocks, set from 17 June to 20 July 1988. Populations of I. latidens and L. pini were endemic; those of D. ponderosa were post-epidemic.

In each experiment, treatments were assigned randomly to traps within each block as follows: attractive semiochemical(s) alone or with devices resulting in one of five verbenone release rates. The control treatment in each experiment was a trap baited only with attractive semiochemicals. In experiment 1, these semiochemicals were the aggregation pheromones exo-brevicomin and cis- and trans-verbenol, and the host kairomone mycene. In experiments 2 and 3, the semiochemicals were the pheromones ipsdienol and ipsenol, respectively.

In experiments 1 and 3, the release rates of verbenone were 0.03, 0.09, 0.62, 13.58, and 54.02 mg/d at 24-28°C. The four lowest rates were obtained by the use of devices 1, 2, 4, and 5, respectively (Table 1). The highest rate was obtained through the use of four device 5. In experiment 2, the release rates of verbenone were 0.01, 0.19, 1.82, 3.08, and 12.32 mg/d at 24-28°C. The two lowest rates were obtained with devices 1 and 2. The second highest rate was obtained with device 7. The third lowest rate was obtained with three devices 3 and the highest rate was obtained with four device 7.

Sexes of captured I. latidens and D. ponderosa were determined by dissection and internal examination of genitalia. Sexes of captured L. pini were determined using decidual characters (Lanier & Cameron 1999).

Statistical Analyses. The data were analyzed using the SYSTAT statistical package (SYSTAT 1990). Trap catches were transformed by log10(y + 1) to remove heteroscedasticity. Proportions of males in catches were transformed by arcsine(√y) to reduce
deviations from normality. Homoscedastic data were subjected to two-way analyses of variance (ANOVA), using block and treatment as model factors, and Tukey's multiple comparison tests set with an experimentwise F = 0.05. Catches of beetles, transformed by $\log_{10}(y+1)$, and proportions of male I. pini and D. ponderosa, transformed by arc sine($y$), in traps baited with vernonene devices, were regressed on the release rate of vernonene, transformed by $\log_{10}(x)$, using a general linear model.

**Results**

Vernonene significantly interrupted the attraction of I. latidens, I. pini, and D. ponderosa to pheromone-baited multiple-funnel traps (Fig. 1). Interruption was significantly dose dependent for all three species, with the highest dose having the greatest effect. Catches of I. latidens in traps releasing vernonene at the highest rate were significantly lower than those in traps without vernonene or with devices releasing vernonene at the two lowest rates (Tukey's test, P = 0.017, 0.010, and 0.017, respectively). Catches of I. pini and D. ponderosa in traps baited with vernonene devices releasing at the two highest rates were significantly lower than those in all other traps (Tukey's test, P < 0.05).

There were no differences in sex ratio of captured beetles among treatments for I. latidens (F = 1.38; df = 2, 4; P = 0.350) and I. pini (F = 1.94; df = 5, 35; P = 0.113). The mean (±SEM) proportions of male I. latidens and I. pini caught in traps were 0.17 ± 0.03 (n = 11) and 0.25 ± 0.01 (n = 48), respectively. Sex ratios of I. latidens in catches from traps baited with vernonene devices releasing at the three highest rates were not calculated because of low numbers.

There was a weakly significant difference in sex ratio of captured D. ponderosa among treatments (F = 3.34; df = 3, 8; P = 0.082). Control traps had a lower proportion of males than those baited with vernonene released at the third highest rate (Tukey's test, P = 0.006). The mean (±SEM) proportion of male D. ponderosa in control traps was 0.40 ± 0.07, those for traps with vernonene devices releasing at the three lowest rates were 0.48 ± 0.03, 0.47 ± 0.04, and 0.54 ± 0.03, respectively. None of the other comparisons was significant (Tukey's test, P > 0.25). Regressions of sex ratio on release rate of vernonene were not significant for either I. pini ($r^2 = 0.039$, $P = 0.523$) or D. ponderosa ($r^2 = 0.203$, $P = 0.165$). Sex ratios for catches of D. ponderosa and I. pini from traps with vernonene devices releasing at the two highest rates were not calculated because of low numbers.

**Discussion**

Over 200 million mature pines were killed by D. ponderosa in British Columbia between 1972 and 1988 (Van Sickle 1999). Vernonene has been tested as an alternative to harvesting as a tactic to minimize mortality of pines by D. ponderosa in attempts to protect both timber and non-timber values. The results of these attempts have been inconsistent (Amman 1994). Vernonene released from multiple points in stands of lodgepole pine significantly reduced the levels of infestations in comparison with controls in some trials (Amman et al. 1989, Lindgren et al. 1989, Gibson et al. 1991) but not in others (Gibson et al. 1991; B.S.L., unpublished data). In stands of ponderosa pine,
verbeneone did not reduce the levels of infestations (Bentz et al. 1989, Lister et al. 1990, Gibson et al. 1991). In two of these trials in ponderosa pine there appeared to be a negative correlation between levels of infestation and the release rate of verbeneone. High levels of variation between replicates may have reduced the power of multiple range tests associated with these trials.

In nature, the ratio of and by attacks on additional trees during epidemics. Dose-dependent responses to verbeneone over a broad range of release rates may be the mechanism facilitating navigation through an infested stand. The release of verbeneone from multiple points in a stand, as used in attempts to control D. ponderosae, should result in similar spatial variation in pheromone concentrations. It is reasonable to expect that beetles should be able to navigate through this type of pheromone landscape as well, ultimately reaching suitable hosts through responses to host kairomones and aggregation pheromones. The level of variation in responses of D. ponderosae tend to suggest that the use of verbeneone in mitigating the impacts of D. ponderosae may be quite limited. Successes to date tend to have occurred in stands with a low infestation level (<10/ft²) (B.S.L., unpublished data). One possible means of improving the efficacy and reliability of verbeneone against D. ponderosae is the addition of other semiochemicals, such as green leaf volatiles or pheromones of competing species, that help convey the message that trees in the stand are not suitable for attack. The trees may be perceived as resistant, nonhost species, or as trees already occupied by competing species (Faine & Hinton 1989). Borden et al. 1992, Dickens et al. 1992, Hobson 1995). However, the responses of beetles would have to preclude their ability to navigate through a pheromone landscape, possibly by causing beetles to leave the stand.

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