

E-MYRCENOL: A NEW PHEROMONE FOR THE PINE ENGRAVER, *ZPS PZNZ* (SAY)
(COLEOPTERA: SCOLYTIDAE)

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Abstract

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E-Myrcenol reduced catches of the pine engraver, *Ips pini* (Say), to ipsdienol-baited, multiple-funnel traps in a dose-dependent fashion. The sex ratio was unaffected by E-myrcenol treatments. Lures containing E-myrcenol in ethanol solution failed to protect freshly cut logs of lodgepole pine from attack by *I. pini*. Rather, *I. pini* preferentially attacked logs treated with devices releasing E-myrcenol and ethanol, over nontreated, control logs. Our results demonstrate that E-myrcenol is a new pheromone for *I. pini*, and emphasize the importance of understanding basic pheromone biology before utilisation of a semiochemical in forest pest management.

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Résumé

Selon la dose utilisée, le E-myrcenol a réduit les captures du scolyte du pin, *Zps pini* (Say), dans des pièges à entonnoirs multiples appâtés avec l'ipsdienol. Le rapport des sexes n'était pas affecté par les traitements avec le E-myrcenol. Les appâts contenant le E-myrcenol dans une solution d'éthanol n'ont pas réussi à protéger des billes du pin lodgepole fraîchement coupées de l'attaque du *I. pini*. L'attaque du *I. pini* s'est faite plutôt sur les billes libérant le E-myrcenol et l'éthanol, comparativement aux billes témoins non-traitées. Nos résultats démontrent que le E-myrcenol est une nouvelle phéromone pour *I. pini* et soulignent l'importance d'une compréhension de la biologie fondamentale des phéromones avant d'utiliser un composé semiochimique dans la lutte intégrée contre les ravageurs forestiers.

Introduction

The pine engraver, *Ips pini* (Say) (Coleoptera: Scolytidae), aggregates rapidly and in large numbers to suitable host pines (Anderson 1948). Both sexes are attracted to males producing the pheromone ipsdienol (2-methyl-6-methylene-2,7-octadien-4-ol) (Vité *et al.* 1972; Stewart 1975). No other pheromone component has been reported.

Ipsdienol is an oxidation product of the monoterpene myrcene (Hughes 1974; Renwick *et al.* 1976; Hendry *et al.* 1980). Several other oxidation products of myrcene are associated with the pheromone biology of *I. pini* (Fig. 1). Ipsenol (2-methyl-6-methylene-7-octen-4-ol) is produced by other species of *Ips*, such as *I. paraconfusus* Lanier, and repels *I. pini* (Birch and Wood 1975; Birch and Light 1977; Birch *et al.* 1977). Linalool (3,7-dimethyl-1,6-octadien-3-ol) has been identified in frass of male *I. pini* (Young *et al.* 1973; Stewart 1975) and elicits response from antenna1 receptors of *I. pini* (Angst and Lanier 1979; Mustaparta *et al.* 1979). Linalool may (Birch and Wood 1975) or may not (Birch and Light 1977) inhibit the response of other *Ips* spp., such as *I. paraconfusus*, to sources of their own pheromones. E-Myrcenol (2-methyl-6-methylene-2,7-octadien-3-ol) has been found in volatiles of *I. pini* (Gries *et al.* 1988), *I. schmutzenhoferi* Holzschuh, and *I. sexdentatus* (Francke *et al.* 1988). E-Myrcenol is also produced by other species such as *Dendroctonus ponderosae* Hopkins (Conn 1981; Hunt *et al.* 1986; Pierce *et al.* 1987). E-Myrcenol was attractive to *D. ponderosae* in laboratory assays (Conn 1981), but not in the field (Conn *et al.* 1983).

E-Myrcenol is structurally similar to compounds active for *I. pini*, such as ipsdienol, ipsenol, and linalool (Fig. 1), and is produced in comparable quantities (Gries *et al.* 1988). Therefore, we hypothesized that E-myrcenol is a pheromone for *I. pini*. If true, then it

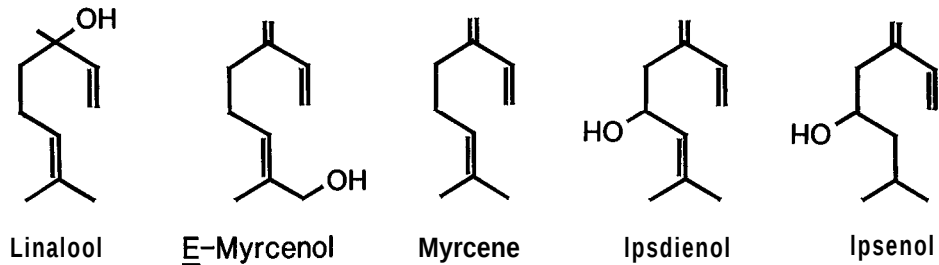


FIG. 1. Myrcene and its principal oxidation products.

should affect the behaviour of *I. pini* to sources of the known pheromone ipsdienol, and/or to host material. We tested the effect of E-myrcenol with ipsdienol-baited, multiple-funnel traps and with freshly felled logs of lodgepole pine, *Pinus contorta* var. *latifolia* Engelmann.

Materials and Methods

Chemicals and Release Devices. E-Myrcenol (>99% *E*; chemical purity, 83%) was obtained from H.D. Pierce, Jr. (Dept. of Chemistry, Simon Fraser University, Burnaby, B.C. V5A 1S6). Racemic ipsdienol (chemical purity, 98%) was obtained from Phero Tech Inc. (1140 Clark Dr., Vancouver, B.C. V5L 3K3). E-Myrcenol and ipsdienol were released from separate devices. Each device consisted of a length of C-flex@ tubing (ID = 1.6 mm; OD = 2.4 mm) (Concept Inc., 12707 U.S. 19 South, Clearwater, FL 33546-7295), filled with an ethanol solution of either myrcenol or ipsdienol (80 mg/mL), or plain ethanol (99%) for controls, and heat-pressure sealed at both ends. Different release rates, obtained by varying the length of C-flex@ tubing, were estimated at a constant 24°C.

Trapping Experiments. On 3 July 1987, 80 eight-unit multiple-funnel traps (Lindgren 1983) (Phero Tech Inc., 1140 Clark Dr., Vancouver, B.C. V5L 3K3) were set in a mature forest of lodgepole pine near Princeton, B.C., in 10 replicates of eight traps each. Replicates were spaced at least 100 m apart, and traps were spaced 10-15 m apart in a 2 x 4 grid. Each trap was baited and suspended from a metal pole such that the top funnel of each trap was 1.3-1.5 m above ground.

Two ranges of E-myrcenol release rates were tested in two experiments. In both experiments, E-myrcenol was tested at three different release rates, singly and in combination with racemic ipsdienol. Ipsdienol and an ethanol control were the remaining two treatments. The release rate for ipsdienol was approximately 0.6 mg per day. Release rates for E-myrcenol were approximately 0.06, 0.18, and 0.60 mg per day in the low-range experiment, and 0.6, 3.0, and 6.0 mg per day in the high-range experiment. The low range of release rates was tested from 16 July to 9 August 1987, and the high range was tested from 3 to 16 July 1987.

Experiment with Logs. On 7 August 1988, 20 lodgepole pines (mean diameter at breast height? SE = 20.0 ± 0.47 cm) were felled near Gang Ranch, B.C., and sawed into logs, 10 m in length. Each log was marked into 10 equal segments of 1 m each. The logs were grouped into four replicates of five treatments each. Replicate plots were spaced 1-2 km apart, and the logs were 25-50 m apart within replicates. The treatments, set in a randomised block design, were as follows: (1) nontreated control; (2) one E-myrcenol release device at the centre of the log; (3) three devices, spaced 5 m apart; (4) five devices spaced 2.5 m apart; and (5) nine devices spaced 1.25 m apart. Each device was placed on the uppermost surface of each log and protected from damage with wire mesh. The release rate of E-myrcenol from each device was approximately 0.6 mg per day (determined by

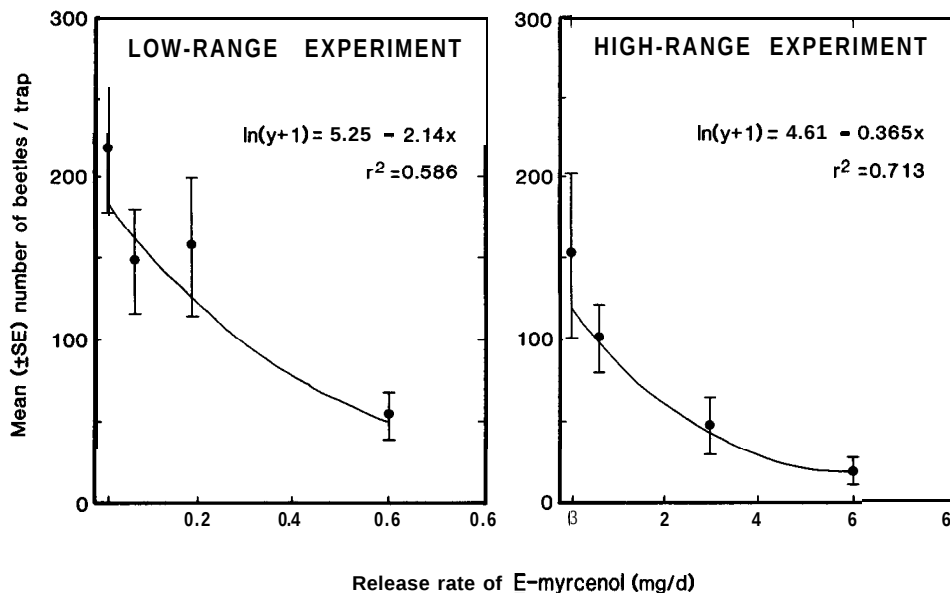


FIG. 2. Effects of low (left) and high (right) release rates of E-myrcenol on the total catch of *Ips pini* attracted to ipsdienol-baited multiple-funnel traps near Princeton, B.C., from 16 July to 9 August 1987 and 3 to 16 July 1987, respectively. Catches in control traps, and in traps baited with E-myrcenol alone, never exceeded five beetles.

collection of volatiles on Porapak-Q). Attacks by *I. pini*, on the upper half of each log, were assessed on 31 August 1988.

Statistical Analyses. The data were analysed using the SPSSX statistical package (SPSS Inc., Suite 3000, 444 N. Michigan Ave., Chicago, IL 60611). Trap catch data were transformed by $\ln(Y + 1)$ to remove heteroscedasticity (Bartlett-Box F, $P = 0.996$ and $P = 0.753$ for the low- and high-range experiments, respectively), and subjected to regression analyses and two-way ANOVA. Data from the experiment with logs were transformed by $\ln(Y + 0.1)$ to remove heteroscedasticity (Bartlett-Box F, $P = 0.275$ and $P = 0.694$ for the number of attacks per log and the number of l-m-long segments attacked per log, respectively), and subjected to one-way ANOVA and Duncan's multiple range test.

Results

Trapping Experiments. E-Myrcenol inhibited the response of *I. pini* to traps baited with racemic ipsdienol (Fig. 2). Inhibition was weakly significant in the low-range experiment [ANOVA, $F(3,25)$, $P = 0.082$] and strongly significant in the high-range experiment [ANOVA, $F(3,30)$, $P = 0.004$]. The presence of ipsdienol significantly increased trap catches in both tests [ANOVA, $F(1,25)$, $P < 0.001$, and $F(1,30)$, $P < 0.001$ for the low- and high-range experiments, respectively], compared with the catches in traps baited with E-myrcenol alone.

In both experiments, the numbers of *I. pini* responding to traps baited with ipsdienol decreased exponentially as the release rate of E-myrcenol increased (Fig. 2). The slope was steeper in the low-range experiment than in the high-range experiment (t-test, $df = 16$, $P < 0.01$). Heteroscedasticity in the data sets precluded fitting the data directly to exponential equations.

E-Myrcenol had no significant effect on the sex ratio of *I. pini* responding to traps baited with ipsdienol (Kruskal-Wallis test, $P = 0.762$, $df = 17$, and $P = 0.888$, $df = 17$, for

Table 1. Effect of E-myrcenol on the attraction of *Ips pini* to l-m-long logs of lodgepole pine near Gang Ranch, B.C., 7-31 August 1988 (*n* = 4)

Number of devices releasing E-myrcenol	Mean* (± SE) number of attacks per log	Mean* (± SE) number of l-m segments attacked per log
0	6.3 ± 6.25a	0.3 ± 0.25a
1	5.5 ± 5.17ab	1.0 ± 0.71ab
3	1.3 ± 0.63ab	1.0 ± 0.41abc
5	15.3 ± 4.89bc	3.0 ± 0.91bc
9	82.3 ± 46.64c	5.5 ± 2.10c

*Means within a column followed by different letters are significantly different at $\alpha = 0.05$ [Duncan's multiple range test on data transformed by $\ln(Y + 0.1)$].

the low- and high-range experiments, respectively). However, the mean proportion (± SE) of males in traps was higher (t-test, $P < 0.01$, *df* = 16) in the high-range experiment (0.28 ± 0.013) than in the low-range experiment (0.19 ± 0.015). This is probably a consequence of differential activity of males and/or females toward pheromone sources during the season rather than an effect of the treatments.

Experiment with Logs. Both the mean number of attacks per log and the mean number of l-m-long segments attacked increased as the number of E-myrcenol release devices increased (Table 1) [ANOVA, $F(4,15)$, $P = 0.02$ and $P = 0.017$, respectively]. There was no significant effect due to the diameter of the trees at breast height [ANOVA, $F(4,15)$, $P = 0.66$].

Discussion

Our results support the hypothesis that E-myrcenol is a pheromone for *I. pini*; it is produced by *I. pini* and influences their behaviour toward hosts and sources of ipsdienol. The decreased trap catches due to the presence of E-myrcenol in ipsdienol-baited traps (Fig. 2) suggest that E-myrcenol is an anti-aggregation pheromone. It may facilitate spacing of galleries on host material and switching to new hosts when a current host is saturated.

However, when we attempted to protect lodgepole pine logs from *I. pini*, we found that lures containing E-myrcenol and ethanol induced attack. Ethanol alone is not attractive to *I. pini* (unpublished results). However it is possible that ethanol interacted with host odours to induce an attraction that was not countered by the presence of E-myrcenol. Alternatively, E-myrcenol might have interacted with either host odours, or both ethanol and host odours, in attracting *I. pini*. It is possible that E-myrcenol could function as a multifunctional pheromone, attracting beetles to a log undergoing attack but acting as an epideictic pheromone (Prokopy 1981) when beetles are in close proximity to established galleries.

E-Myrcenol may also act as a kairomone, as it is produced by other species, such as *D. ponder-owe* (Conn 1981; Hunt *et al.* 1986; Pierce *et al.* 1987). In some instances, brood of *I. pini* can outcompete brood of *D. ponderosae* (Rankin 1988), and *I. pini* could benefit by attacking hosts soon after attack by *D. ponderosue*. E-Myrcenol may help to locate such hosts. Multifunctionalities of semiochemicals are known for other scolytid species. For example, at most release rates frontalin inhibits the response of *D. ponderosae* to semiochemical-baited traps (Borden *et al.* 1987) but induces attacks on standing trees (Chatelain and Schenk 1984; Borden *et al.* 1987; unpublished data). More detailed work is obviously required to understand fully the functional value of semiochemicals such as E-myrcenol.

The importance of understanding basic pheromone biology cannot be overemphasized. Our experience with E-myrcenol shows that repellency with a pheromone-baited trap should not be used as a sole criterion to select anti-aggregation pheromones for operational

protection of logs or trees. Premature utilisation of such semiochemicals may cause high losses and impede development of a valuable technology.

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References

- Anderson, R.F. 1948. Host selection by the pine engraver. *J. econ. Ent.* **41**: 596-602.
- Angst, M.E., and G.N. Lanier. 1979. Electroantennogram responses of two populations of *Ips pini* (Coleoptera: Scolytidae) to insect-produced and host tree compounds. *J. Chem. Ecol.* **5**: 131-140.
- Birch, M.C., and D.M. Light. 1977. Inhibition of the attractant pheromone response in *Ips pini* and *I. paraconfusus* (Coleoptera: Scolytidae): field attraction of ipsenol and linalool. *J. Chem. Ecol.* **3**: 257-267.
- Birch, M.C., D.M. Light, and K. Mori. 1977. Selective inhibition of response of *Ips pini* to its pheromone by the *S*-(-)-enantiomer of ipsenol. *Nature* **270**: 738-739.
- Birch, M.C., and D.L. Wood. 1975. Mutual inhibition of the attractant pheromone response by two species of *Ips* (Coleoptera: Scolytidae). *J. Chem. Ecol.* **1**: 101-113.
- Borden, J.H., L.C. Ryker, L.J. Chong, H.D. Pierce, Jr., B.D. Johnston, and A.C. Oehlschlager. 1987. Response of the mountain pine beetle, *Dendroctonus ponderosae* Hopkins (Coleoptera: Scolytidae), to five semiochemicals in British Columbia lodgepole pine forests. *Can. J. For. Res.* **17**: 118-128.
- Chatelain, M.P., and J.A. Schenk. 1984. Evaluation of *frontalin* and *exo-brevicomin* as kairomones to control mountain pine beetle in lodgepole pine. *Environ. Ent.* **13**: 16661674.
- Conn, J.E. 1981. Pheromone production and control mechanisms in *Dendroctonus ponderosae* Hopkins. M.Sc. thesis, Simon Fraser University, Burnaby, B.C. 66 pp.
- Conn, J.E., J.H. Borden, B.E. Scott, L.M. Friskie, H.D. Pierce, Jr., and A.C. Oehlschlager. 1983. Semiochemicals for the mountain pine beetle, *Dendroctonus ponderosae* (Coleoptera: Scolytidae) in British Columbia: field trapping studies. *Can. J. For. Res.* **13**: 320-324.
- Francke, W., J. Bartels, H. Schmutzenhofer, U. Kohnle, and J.P. Vité. 1988. The odor-bouquet of *Ips schmutzenhoferi* Holzschuh (Col.: Scol.). *Z. Naturforsch.* **43**: 958-960.
- Gries, G., H.D. Pierce, Jr., B.S. Lindgren, and J.H. Borden. 1988. New techniques for capturing and analyzing semiochemicals for scolytid beetles (Coleoptera: Scolytidae). *J. econ. Ent.* **81**: 1715-1720.
- Hendry, L.B., B. Piatek, L.E. Browne, D.L. Wood, J.A. Byers, R.H. Fish, and R.A. Hicks. 1980. In vivo conversion of a labelled host plant chemical to pheromones of the bark beetle *Ips paraconfusus*. *Nature* **284**: 48.5.
- Hughes, P.R. 1974. Myrcene: a precursor of pheromones in *Ips* beetles. *J. Insect Physiol.* **20**: 1271-1275.
- Hunt, D.W.A., J.H. Borden, H.D. Pierce, Jr., K.N. Slessor, G.G.S. King, and E.K. Czyżewska. 1986. Sex-specific production of ipsdienol and myrcenol by *Dendroctonus ponderosae* (Coleoptera: Scolytidae) exposed to myrcene vapors. *J. Chem. Ecol.* **12**: 1579-1586.
- Lindgren, B.S. 1983. A multiple-funnel trap for scolytid beetles. *Can. Ent.* **11.5**: 299-302.
- Mustaparta, H., M.E. Angst, and G.N. Lanier. 1979. Specialization of olfactory cells to insect- and host-produced volatiles in the bark beetle *Ips pini* (Say). *J. Chem. Ecol.* **5**: 109-123.
- Pierce, H.D., Jr., J.E. Conn, A.C. Oehlschlager, and J.H. Borden. 1987. Monoterpene in female mountain pine beetles, *Dendroctonus ponderosae* Hopkins, attacking ponderosa pine. *J. Chem. Ecol.* **13**: 1455-1480.
- Prokopy, R.J. 1981. Epideictic pheromones that influence spacing patterns of phytophagous insects. pp. 181-235 in Nordlund, D.A., R.L. Jones, and W.J. Lewis (Eds.), *Semiochemicals. Their Role in Pest Control*. John Wiley and Sons, New York, NY.
- Rankin, L.J. 1988. Competitive interactions between mountain pine beetle and the pine engraver in lodgepole pine. MPM Professional Paper, Simon Fraser University, Burnaby, B.C. 33 pp.
- Renwick, J.A.A., P.R. Hughes, G.B. Pitman, and J.P. Vité. 1976. Oxidation products of terpenes identified from *Dendroctonus* and *Ips* bark beetles. *J. Insect Physiol.* **22**: 725-727.
- Stewart, T.E. 1975. Volatiles isolated from *Ips pini*: isolation, identification, enantiomeric composition, biological activity. M.Sc. thesis, State University of New York, Syracuse, NY.
- Vité, J.P., A. Bakke, and J.A.A. Renwick. 1972. Pheromones in *Ips* (Coleoptera: Scolytidae): occurrence and production. *Can. Ent.* **104**: 1967-1975.

Young, J.C., R.G. Brownlee, J.O. Rodin, D.N. Hildebrand, R.M. Silverstein, D.L. Wood, M.C. Birch, and L.E. Brown. 1973. Identification of linalool produced by two species of bark beetles of the genus *Ips*. *J. Insect Physiol.* 19: 1615-1622.

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