Synergism of Turpentine and Ethanol as Attractants for Certain Pine-Infesting Beetles (Coleoptera)

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Environ. Entomol. 17(3): 456–462 (1988)

ABSTRACT Responses of seven species of pine-infesting beetles to traps baited with either turpentine, ethanol, turpentine and ethanol released from separate dispensers, or a 1:1 solution of turpentine and ethanol released from one dispenser were assessed in three field experiments. The weevil species, *Pachylobius picivorus* (Germar), and the cerambycid pine sawyer, *Monochamus carolinensis* (Olivier), were attracted to turpentine and were unaffected by the addition of ethanol. The ambrosia beetle, *Xyleborus affinis* Eichhoff, responded to ethanol alone but was not attracted to turpentine, nor did the presence of turpentine significantly affect its response to ethanol. The remaining four species displayed responses to turpentine that were enhanced by the addition of ethanol, but in different ways according to the method of deployment. *Hylobius pales* (Herbst) weevils and *M. titillator* (F.) sawyers displayed greatest attraction to turpentine and ethanol whether they were released from side-by-side dispensers or as a solution from one dispenser. The black turpentine beetle, *Dendroctonus terebrans* (Olivier), displayed the highest response to turpentine and ethanol in solution. The ambrosia beetle, *X. pubescens* Zimmermann, responded in low numbers to turpentine or ethanol deployed singly, but displayed an enhanced response (20-fold increase) to turpentine and ethanol deployed side-by-side and an even greater response (60-fold increase) to a solution of turpentine and ethanol. Reasons for increased responses by some species to a solution of turpentine and ethanol over the two released separately are not clear; they may lie in different dosages or evaporation rates of volatiles in the field. Laboratory analyses of trapped headspace volatiles from dispensers containing only turpentine and those containing a solution of turpentine and ethanol revealed no differences in the amounts of four principal monoterpene hydrocarbons (α-pinene, camphene, β-pinene, and limonene) released over time. The synergistic effect of turpentine and ethanol for some species and not others may point to ecological differences between species with regard to the condition of preferred host material.

KEY WORDS Insecta, pine beetles, attractants, terpenes

VARIOUS SPECIES of Coleoptera that infest moribund conifers are known to be attracted by odors emanating from potential host trees. Monoterpene hydrocarbons present in conifer oleoresin have been suspected of being primary attractants for certain tree-killing scolytids (e.g., Heikkenen & Hrutfiord 1965) and have been confirmed as attractants for many nonaggressive scolytids and other Coleoptera (Thomas & Hertel 1969, Selander et al. 1974, Byers et al. 1985, Fatzinger 1985). Ethanol is produced by stressed or cut conifers (Moeck 1970, Kimmerer & Kozlowski 1982) and is attractive to many scolytids and cerambycids (Cade et al. 1970, Moeck 1970, Roling & Kearby 1975, Montgomery & Wargo 1983, Dunn et al. 1986). In addition to the attractive nature of tree-produced odors, individual terpene hydrocarbons and ethanol are known to enhance or synergize the activity of pheromones produced by certain conifer-infesting beetles (e.g., Bedard et al. 1969, Borden et al. 1980; see review by Borden 1982).

Commercial gum turpentine is a mixture of terpenes distilled from the oleoresin of pine trees (*Pinus* spp.) and as such, represents an easily obtainable solution of host terpenes that is potentially attractive to many generalist pine beetles. Hopkins (1909) documented flight responses of the black turpentine beetle, *Dendroctonus terebrans* (Olivier), and the red turpentine beetle, *D. valens* Le Conte, to odors of turpentine. Fatzinger (1985) quantified the seasonal responses of *D. terebrans* to turpentine-baited traps and studied responses by the weevils *Hylobius pales* (Herbst) and *Pachylobius picivorus* (Germar) and by the cerambycid pine sawyers *Monochamus carolinensis* (Oliver) and *M. titillator* (F.). Billings (1985) found that the rapid release of turpentine (1,800–3,600 mg/d) greatly enhanced the responses of *D. frontalis* Zimmermann and *Ips grandicollis* (Eichhoff) to their species-specific pheromones but reduced the response of *Ips avulsus* (Eichhoff) to its pheromone. Tilles et al. (1986) reported that ethanol acted synergistically with a mixture of terpenes occurring in *Pinus sylvestris* L. to be highly attractive to the large pine weevil, *H. abietis* L. Similarly, Vité et al. (1986) showed that ethanol acts synergistically

with host monoterpenes to attract the scolytid *Tomicus piniperda* L. Ethanol alone attracted several species of ambrosia beetles (Scolytidae), and a low level of ethanol enhanced the response of *Leperisinus varius* (F.) to its pheromone in studies by Klimetzek et al. (1986).

Fatzinger (1985) reported that ethanol synergized the response of *D. terebrans* to turpentine when they were in solution together, but he did not test their effect by releasing them from separate elution devices in the same traps. A later study examined responses to turpentine and ethanol by the pine weevil species, *H. pales* and *P. picivorus*, and the pine sawyer species, *M. titillator* and *M. carolinensis*, but turpentine and ethanol were presented only as a solution when tested together in one treatment (Fatzinger et al. 1987).

Siegfried et al. (1986) and Siegfried (1987) assessed the responses of black turpentine beetles and pine weevils, respectively, to individual monoterpenes, but all compounds were diluted in ethanol and any possible enhancement or synergism was not investigated. Because ethanol was known as an enhancer or synergist for beetle attractants, apprehension concerning its use as a solvent for semiochemicals has been noted (e.g., Pitman et al. 1975, Rudinsky & Ryker 1980). We assume from Raoult's Law that a solution of compounds with differing vapor pressures can yield a mixture of vapors in its headspace with a composition different from that in the solution. This phenomenon is of great concern to researchers of pheromone applications who must devise controlled release formulations for multiple compounds (e.g., Heath et al. 1986), as it should be for those initially investigating the activity of multiple semiochemicals.

We were curious to know whether the reported synergism of turpentine and ethanol as an attractant blend for the black turpentine beetle (Fatzinger 1985) was synergism in the sense known for multiple semiochemicals of other insects, or perhaps a result of an evaporation change in the turpentine or its components while in solution with ethanol. The term synergism, as we (and others) use it in relation to multiple semiochemicals, means that the response of the insects to the combination of semiochemicals is greater than the sum of the responses to the individual semiochemicals. We initiated studies to examine the response of *D. terebrans* to turpentine and ethanol when released separately or in solution. Subsequently, we expanded our studies to examine responses of several species of Coleoptera that occur in the southeastern United States and are reported to display turpentine–ethanol synergism or are known to be attracted by pine odors. This paper reports the results of field experiments that examined beetle responses to the same four treatments: turpentine only, ethanol only, turpentine and ethanol released from separate dispensers, and turpentine and ethanol in solution released from one dispenser. We also investigated the evaporation rates of turpentine, ethanol, and the constituent monoterpene hydrocarbons of turpentine from our dispensers using gravimetric and chemical analyses.

Materials and Methods

Three separate field experiments were conducted during the summer and fall of 1986. Our field experiments compared the activity of the same four attractant bait types and were intended to assess the responses of different species or groups of species at the different sites. Experiments 1 and 3 were conducted in a recently clearcut area (approximately 6 mo after harvest) of a commercial slash pine, *Pinus elliottii* Engelm. var. *elliottii*, forest maintained by the Owens-Illinois Corporation north of Gainesville in Alachua County, Fla. Beetles trapped at this site were presumed to have bred in the logging slash, residual damaged trees, and numerous fresh stumps that were available. Experiment 2 was conducted on the grounds of a Georgia Pacific Corporation plywood mill in Putnam County, just east of Hawthorne, Fla. Preliminary trapping studies at the plywood mill revealed large numbers of pine beetles belonging to various species. Experiments 1 and 2 used stovepipe traps (Clements & Williams 1981, Fatzinger 1985) that consisted of a section of blackened air-conditioning duct (23 cm diameter by 120 cm long) vertically centered in a plastic wading pool (120 cm diameter by 25 cm deep) by three guy wires tethered to the pool rim. The wading pool was filled with about 20 liters of soapy water, and attractant baits were suspended from the top outside edge of the stovepipe. Responding insects either flew directly into the pool of water or contacted the stovepipe and fell into the water. Experiment 3 used 16-unit (16 funnel) Lindgren funnel traps (Lindgren 1983) (20 cm diameter by 138 cm tall). Traps were suspended from PVC pipe standards so that the collection jars were no more than 10 cm above the ground. Attractant baits were suspended on the pipe standards at a height of 80 cm and were located about 30 cm from the center of the funnel trap.

Bait dispensers were 250-ml Nalgene screw-top bottles, each with 5 cm of a 15-cm cotton dental wick protruding from a 1-cm hole cut in the center of the top. Bottles were completely filled with bait material; the dental wicks served as the substrates from which the attractive liquids evaporated. The four treatments were whole gum turpentine, 95% ethanol, a dispenser of whole gum turpentine and a dispenser of 95% ethanol placed side by side (wicks no more than 10 cm apart), and a dispenser with a 1:1 solution of turpentine and 95% ethanol. The turpentine was distilled from mixed oleoresins of slash and longleaf pines, *P. palustris* Mill. (obtained from Shelton Naval Stores Processing, Valdosta, Ga.). Preliminary gas chromatographic (GC) analysis of our turpentine (using the methods described below) revealed the composition of the six principal monoterpenes to be α-pinene, 66.46%;

β-pinene, 29.41%; camphene, 1.67%; limonene, 1.67%; β-phellandrene, 0.09%; myrcene, 0.01%.

All three experiments were deployed as completely randomized block designs. Experiment 1 assessed the response of black turpentine beetles to baited stovepipe traps placed at the corners of a square with sides of approximately 40 m. The four bait treatments were randomly assigned to the traps for 24 h, after which the responding beetles were collected from the traps and the baits were randomly reassigned to the traps for another 24 h; a total of four 24-h trapping periods (blocks) were deployed. Experiment 2 used stovepipe traps spaced 40–90 m apart in a line at the inside edge of a peripheral wooded area (mixed slash pine and turkey oak, *Quercus laevis* Walt.) bordering the log storage area of the plywood mill. Responses by the weevils *H. pales* and *P. picivorus* and the pine sawyers *M. carolinensis* and *M. titillator* were evaluated in a total of eight randomized blocks represented by eight 24-h trapping periods. Funnel traps in experiment 3 were deployed at the clearcut site in a straight line with 40 m spacing between traps. We scored the responses of the scolytid ambrosia beetles, *Xyleborus pubescens* Zimmermann and *X. affinis* Eichhoff, to the four treatments in four completely randomized 24-h blocks. Beetles in all experiments were counted and separated by sex; the *Xyleborus* spp. collected were assumed to be females because males are flightless (Bright 1976). Raw data (for the sexes separately and combined) from each of the experiments were subjected to square root transformation ($\sqrt{x + 0.5}$) and analysis of variance followed by means comparisons using Fisher's least-significant difference (LSD) test; sex ratios of responding beetles were examined with χ^2 tests for departures from unity.

Relative evaporation rates of whole gum turpentine, 95% ethanol, and a 1:1 mixture of turpentine and ethanol from our dispensers were assessed gravimetrically. Five dispensers of each of the three solutions were prepared, weighed, and placed in a fume hood in the laboratory at a constant wind speed of about 3.6 km/h, an average temperature of 24°C and an average RH of 60%. Dispensers were kept in the fume hood for 4 d (the time period they would have been deployed in the field before being replaced) and weighed every 24 h. The average weight loss from each bait type for each of the four 24-h periods was determined.

Evaporation rates of the six principal monoterpenes from whole gum turpentine dispensers and dispensers with a 1:1 solution of turpentine and 95% ethanol were determined by collection of volatiles and analysis by GC. One dispenser each of turpentine and the turpentine and ethanol mixture was prepared daily for five consecutive days and placed on a bench in a ventilated laboratory at 23°C and 60% RH. After 24 and 96 h, each dispenser was placed under a conical (15 cm diameter base, 7 cm diameter top, 35 cm tall) silanized glass bell jar, raised 1 cm above the table surface to provide a vent, and equipped with a hole and nipple at the top (small) end to which suction from a water pump could be applied. A glass column (1.6 × 11.0 cm) filled with 6.0 g of the absorbant Porapak-Q (Waters Assoc., Framingham, Mass.) was connected to the outlet hole of the bell jar and air was pulled through the column at a flow rate of 0.5 liters/min. Volatiles from the bait dispensers were collected on the Porapak-Q for 15 min, after which the Porapak-Q was extracted by slowly dripping 30 ml of spectragrade pentane through the column; the Porapak-Q columns were subsequently cleaned by further extraction with 30 ml each of diethyl ether and pentane prior to drying and reuse. Each 30-ml pentane extract was spiked with 1.5287 mg of *p*-cymene and 0.6435 mg of decane as internal GC standards, mixed, and then slowly concentrated to a volume of about 1.0 ml under a constant stream of N_2.

Concentrated samples were subjected to quantitative GC analysis in a Varian 2400 series instrument equipped with a flame ionization detector and a Hewlett Packard 3390A recording integrator. We employed a copper column (6.09 m by 3.17 mm OD) packed with 20% Carbowax 20M on 80/100 mesh Chromosorb W-HP operated under the following conditions: column temperature, 120°C isothermal; injector, 170°C; detector, 290°C; N_2 carrier gas, 20 ml/min. Under these conditions our internal standards and monoterpene standards had the following approximate retention times in minutes: decane, 8.6; α-pinene, 11.2; camphene, 14.2; β-pinene, 17.3; myrcene, 19.6; limonene, 25.5; β-phellandrene, 27.3; *p*-cymene, 36.2. Hourly release rates of the six monoterpenes were calculated for each dispenser and compared between bait types within sample times (24 h and 96 h), and between sample times within bait types using *t* tests. Percent composition of the monoterpenes in the trapped headspace volatiles was also determined for each bait, subjected to the angular transformation (arcsine$\sqrt{x}$), and similarly compared between bait types and sample times.

Results

In the first field experiment we found that the solution of turpentine and ethanol was significantly more attractive to *D. terebrans* than turpentine alone, and ethanol alone was not attractive (Table 1). Male *D. terebrans* responded in significantly higher numbers ($P < 0.05$) to the turpentine–ethanol solution than to the undiluted turpentine or the turpentine and the ethanol deployed side by side. Response of female *D. terebrans* to the turpentine–ethanol solution differed from response to the side-by-side treatment at the $P = 0.078$ level and from response to undiluted turpentine at the $P = 0.065$ level. Response of male and female *D. terebrans* combined to the turpenetine–ethanol solution differed from that to the side-by-side treatment at the $P = 0.076$ level. In Experiment 2 (Table

Table 1. Response of *D. terebrans* in Experiment 1 to traps baited with turpentine and ethanol either singly, in combination, or in solution

Treatment	Mean no. captured (SE) per trap		
	♂♂	♀♀	Total
Turpentine	2.25a (0.74)	1.25a (0.95)	3.50a (1.66)
Ethanol	0b	0b	0b
Turpentine and ethanol	2.75a (1.11)	2.25a (0.48)	5.00ac (1.41)
Solution of turpentine and ethanol	5.00c (1.08)	5.50a (2.06)	10.50c (2.02)

Means in a column followed by different letters are significantly different ($P < 0.05$, Fisher's LSD test); n, 4.

2) we found that the responses of *H. pales* males and females to turpentine were greatly enhanced by the presence of ethanol when it was released either separately or in solution. Conversely, *P. picivorus* males and females were attracted to turpentine, but the addition of ethanol had no apparent effect on these responses. The sex ratio of *H. pales* caught in any trap containing turpentine in Experiment 2 was strongly biased toward females (Table 2). As with *P. picivorus*, the responses of *M. carolinensis* to turpentine were unaffected by the presence of ethanol (Table 3). Female *M. titillator*, however, displayed significantly greater responses to turpentine with ethanol, whether deployed separately or in solution. Ethanol alone displayed the lowest activity for all beetles caught in Experiment 2. In Experiment 3 (Table 4), the responses of *X. pubescens* to turpentine alone and ethanol alone were similarly low. Greater numbers of *X. pubescens* were caught when turpentine and ethanol were released side by side from the same traps, but this response was more than tripled when turpentine and ethanol were released as a solution (Table 4). In the same experiment, *X. affinis* was attracted to ethanol alone, and the addition of turpentine in any form did not significantly affect this; turpentine alone attracted very few *X. affinis*.

The average weight loss of whole-gum turpentine from bait dispensers over a 4-d period in the laboratory was 4.70 ± 1.04 SE$_{\bar{x}}$ g/d, the turpentine–ethanol solution lost 10.14 ± 1.69 SE$_{\bar{x}}$ g/d, and 95% ethanol lost 23.64 ± 4.45 SE$_{\bar{x}}$ g/d. The turpentine–ethanol solution released about twice as much material as undiluted whole-gum turpentine. However, quantitative analyses of volatiles collected from the two bait types revealed that turpentine components were released from undiluted turpentine and the turpentine–ethanol solution at the same rates (Table 5). Myrcene or β-phellandrene were not detected in the headspace volatiles collected from any bait. The very low concentration of these components in our turpentine (0.01% and 0.09%, respectively) may have resulted in levels too low for us to detect in the headspace volatiles. Evaporation rates of the remaining four principal monoterpenes did not differ significantly between the two types of baits within either sample time. Evaporation rates of the four monoterpenes from dispensers with undiluted turpentine were significantly reduced after 96 h compared with those from the same dispensers after 24 h (t test, $P < 0.05$). There were no qualitative differences in headspace volatiles between dispensers with pure turpentine and those with the turpentine–ethanol solution. GC analysis of the liquid turpentine and of the turpentine–ethanol solution also did not reveal any qualitative differences upon a peak-for-peak comparison of chromatograms.

Analysis of variance for all three field experiments detected no significant block (day-to-day) effects. Therefore, we do not believe that temporal variants, such as the reduction in monoterpene evaporation from undiluted turpentine noted above, had significant effects on the responses of beetles.

Discussion

At least four general response categories can be identified for behavior of the pine beetle species captured during this study. First, some species were attracted simply to turpentine, and their responses

Table 2. Response of *H. pales* and *P. picivorus* in Experiment 2 to traps baited with turpentine and ethanol either singly, in combination, or in solution

Treatment	Mean no. captured (SE) per trap						
	H. pales				*P. picivorus*		
	♂♂[a]		♀♀	Total	♂♂	♀♀	Total
Turpentine	10.75a (3.76)	**	23.50a (9.10)	34.25a (12.40)	2.38a (0.87)	1.49a (0.53)	4.13a (1.22)
Ethanol	1.00b (0.46)		1.38b (0.38)	2.38b (0.75)	0.00b (0.00)	0.25b (0.16)	0.25b (0.16)
Turpentine and ethanol	51.75c (12.10)	**	83.88c (22.57)	135.63c (34.08)	2.13a (0.55)	1.00a (0.33)	3.13a (0.58)
Solution of turpentine and ethanol	36.63c (9.22)	**	73.63c (22.57)	110.25c (30.71)	2.63a (0.68)	1.88a (0.72)	4.50a (1.25)

Means in a column followed by different letters are significantly different ($P < 0.05$, Fisher's LSD test); n, 8.
[a] **, sex ratio of weevils responding to a treatment departed significantly from unity ($P < 0.01$, χ^2 test).

Table 3. Response of *M. carolinensis* and *M. titillator* in Experiment 2 to traps baited with turpentine and ethanol either singly, in combination, or in solution

Treatment	Mean no. captured (SE) per trap					
	M. carolinensis			*M. titillator*		
	♂♂	♀♀	Total	♂♂	♀♀	Total
Turpentine	2.13a (0.83)	3.38a (0.86)	5.38a (1.59)	0.75a (0.31)	0.88a (0.35)	1.50a (0.63)
Ethanol	0.38b (0.26)	0.75b (0.31)	1.13b (0.44)	0.00b (0.00)	0.13b (0.13)	0.13b (0.13)
Turpentine and ethanol	1.88a (0.44)	4.88a (1.39)	6.75a (1.70)	1.13a (0.30)	3.00c (0.85)	4.13c (1.06)
Solution of turpentine and ethanol	2.13a (0.61)	4.38a (1.38)	6.50a (1.69)	1.88a (0.64)	2.13c (0.69)	4.00c (0.94)

Means in a column followed by different letters are significantly different ($P < 0.05$, Fisher's LSD test); n, 8.

were not affected by the addition of ethanol; this was exemplified by *P. picivorus* and *M. carolinensis* (Tables 2 and 3). Second, one species, *X. affinis*, was attracted only to ethanol and was relatively unaffected by the presence of turpentine (Table 4). Third, three species, *H. pales*, *M. titillator*, and *X. pubescens*, displayed enhanced responses to turpentine by the synergistic effect of ethanol being released from a separate dispenser (Tables 2–4). Fourth, an additional form of turpentine–ethanol synergism was displayed by *D. terebrans* and *X. pubescens* (Tables 1 and 4), for which responses to turpentine and ethanol in solution were greater than to turpentine only or to turpentine and ethanol released side by side. Our results on the responses of turpentine beetles, weevils, and sawyer beetles to a solution of turpentine and ethanol generally corroborate those reported by Fatzinger et al. (1987).

The different responses to turpentine and ethanol among the species we studied may be caused by differences in odor cues these beetles would perceive from different types of hosts or from host material in various stages of stress or decomposition. Stressed plants as different as pines and hardwoods produce ethanol (Kimmerer & Kozlowski 1982). *X. affinis* is a host generalist and is reported to breed in decomposing xylem tissue of conifers and hardwoods (Bright 1976, Wood 1982). Hence, ethanol may be a general host-finding cue for *X. affinis*, as it probably is for several other species of polyphagous ambrosia beetles (e.g., Roling & Kearby 1975). Moeck (1970) found that ethanol was released from logs of *Pseudotsuga menziesii* (Mirb.) Franco that had aged in the forest for several months or that had been subjected to anaerobic incubation, but he found very little ethanol was released from freshly cut logs. Beetles included in our third and fourth response categories, ethanol-synergized response to turpentine, may be naturally inclined to orient to pine host material that is in some stage of decomposition in which ethanol is produced at a level relative to the stage of decomposition. Beetles in our first category, response to turpentine only, may orient to "fresher" host material (e.g., recently injured or traumatized) with lower ethanol levels but with the release of terpene volatiles from oleoresin. If our hypothesized ecological reasons for different responses by beetles are true, they may help to explain how these different species belonging to a common pine-feeding guild could partition the available resource in time or space.

Black turpentine beetles responded more to a solution of turpentine and ethanol than to the components released separately (Table 1). This phenomenon is seen more dramatically in the very large response of *X. pubescens* to the solution (Table 4), even though turpentine and ethanol were synergistic when released side by side in separate dispensers. The reasons for this enhanced synergism of turpentine and ethanol in solution are not readily apparent. At the onset of our study, we presumed that synergism between two or more semiochemicals, if it were to occur for the response of a particular insect, would be equally evidenced whether the components were evaporated separately or from a solution, barring any significant differences in amounts of components released. Our laboratory study of monoterpene evaporation rates (Table 5) indicated no differences in amounts of the four major turpentine volatiles released from either whole turpentine or a turpentine–ethanol

Table 4. Response of *X. pubescens* and *X. affinis* in Experiment 3 to traps baited with turpentine and ethanol either singly, in combination, or in solution

Treatment	Mean no. captured (SE) per trap	
	X. pubescens	*X. affinis*
Turpentine	2.25a (1.03)	2.00a (1.15)
Ethanol	2.00a (1.08)	40.25b (13.90)
Turpentine and ethanol	44.50b (22.40)	15.75ab (10.30)
Solution of turpentine and ethanol	144.25c (16.42)	33.25b (7.53)

Means in a column followed by different letters are significantly different ($P < 0.05$, Fisher's LSD test); n, 4.

Table 5. Mean rate of evaporation, mean percentage composition, and standard errors of means (in parentheses) of volatile monoterpene hydrocarbons from trap baits of turpentine or a solution of turpentine and ethanol (1:1)

Sample time	Monoterpene	Undiluted turpentine		1:1 solution of turpentine and ethanol	
		Mean rate (mg/h)	Mean % composition	Mean rate (mg/h)	Mean % composition
24 h	α-pinene	5.8680 (1.0493)	68.61 (0.65)	4.3172 (0.9854)	69.43 (0.56)
	Camphene	0.1302 (0.0224)	1.54 (0.06)	0.0962 (0.0342)	1.36 (0.22)
	β-pinene	2.4507 (0.4393)	28.50 (0.58)	1.7654 (0.4366)	27.90 (0.35)
	Limonene	0.1104 (0.0168)	1.33 (0.08)	0.0819 (0.0201)	1.31 (0.02)
96 h	α-pinene	1.5212 (0.3897)	69.43 (1.16)	2.3594 (0.9879)	68.65 (1.24)
	Camphene	0.0505 (0.0097)	2.72 (0.72)	0.0748 (0.0307)	2.31 (0.64)
	β-pinene	0.5909 (0.1566)	26.62 (0.83)	1.0224 (0.5008)	27.34 (1.18)
	Limonene	0.0290 (0.0093)	1.22 (0.32)	0.0583 (0.0276)	1.67 (0.16)

No significant difference ($P > 0.05$) found between bait types for evaporation rate of any monoterpene.

solution, and we detected no qualitative chemical differences between the two bait types. Environmental conditions of our field experiments varied greatly, particularly those of wind speed, temperature, and amount of solar radiation. It is possible that bait dispensers in the field did not release ethanol and turpentine components at relative rates similar to those of our laboratory study, and that different beetle responses could be caused by different dosages of attractants. The release rate of ethanol (which we did not quantify chemically) could have affected responses to different treatments. Our gravimetric study revealed that dispensers containing only 95% ethanol lost about twice as much weight over time as those containing a 1:1 solution of turpentine and ethanol. Therefore, traps baited with side-by-side dispensers of turpentine and ethanol probably were deploying at least twice as much ethanol as traps baited with the turpentine–ethanol solution.

Maximal responses of *D. terebrans* and *X. pubescens* to the turpentine–ethanol solution could have been caused by the lower dosage of ethanol compared with the side-by-side treatment. Klimetzek et al. (1986) reported certain dosage–response cases for bark beetles in Germany, in which low levels of ethanol optimally synergized attractants compared to high levels, but their overall release rates were much lower than ours. We are investigating the possibility of dosage-dependent response to ethanol in the beetles we studied.

It is obvious that substantial differences in response can occur between treatments in which multiple semiochemicals are deployed individually and those in which they are deployed in solutions. Pitman et al. (1975) reported that the addition of ethanol to two different pheromone–terpene mixtures, presumably as a solvent for the pheromones or terpenes or both, synergistically enhanced the response of *D. pseudotsugae* Hopkins to its pheromones and speculated that ethanol may have affected responses to pheromones in earlier work. Rudinsky & Ryker (1980) later refuted this speculation by using a behaviorally inactive solvent for the *D. pseudotsugae* pheromone 3,2-MCH. As in other studies, our results also point to the potential for misleading conclusions that can be drawn from studies in which semiochemicals are diluted in ethanol or ethanol is used to clean bioassay devices (as cited in Tilles et al. 1986). It is common practice in tests of insect semiochemicals to release individual compounds from separate release devices, and this is well justified to standardize methods and control dosages. However, it is important to remember that volatiles from natural sources (plants or insects) are often evaporated from complex mixtures. Our results indicate that mixtures of semiochemicals can, in some cases, elicit maximal behavioral responses by insects.

Acknowledgment

We appreciate the reviews of earlier drafts of this manuscript by R. F. Billings, C. W. Fatzinger, R. L. Hedden, B. S. Lindgren, and M. P. Popp. Carl Fatzinger kindly provided the chromatography column used for the terpene analyses. This research was supported by grant no. 85-CRCR-1-1690 from the USDA. This paper represents Florida Agricultural Experiment Station Journal Series No. 8341.

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Received for publication 20 August 1987; accepted 28 December 1987.