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A Blend of Ethanol and (−)-α-Pinene were Highly Attractive to Native Siricid Woodwasps (Siricidae, Siricinae) Infesting Conifers of the Sierra Nevada and the Allegheny Mountains

Nadir Erbilgin[1] · Jack D. Stein[2] · Robert E. Acciavatti[2] · Nancy E. Gillette[3] · Sylvia R. Mori[3] · Kristi Bischel[4] · Jonathan A. Cale[1] · Carline R. Carvalho[3] · David L. Wood[5]

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Abstract Woodwasps in *Sirex* and related genera are well-represented in North American conifer forests, but the chemical ecology of native woodwasps is limited to a few studies demonstrating their attraction to volatile host tree compounds, primarily monoterpene hydrocarbons and monoterpene alcohols. Thus, we systematically investigated woodwasp-host chemical interactions in California's Sierra Nevada and West Virginia's Allegheny Mountains. We first tested common conifer monoterpene hydrocarbons and found that (−)-α-pinene, (+)-3-carene, and (−)-β-pinene were the three most attractive compounds. Based on these results and those of earlier studies, we further tested three monoterpene hydrocarbons and four monoterpene alcohols along with ethanol in California: monoterpene hydrocarbons caught 72.3% of all woodwasps. Among monoterpene hydrocarbons, (+)-3-carene was the most attractive followed by (−)-β-pinene and (−)-α-pinene. Among alcohols, ethanol was the most attractive, catching 41.4% of woodwasps trapped. Subsequent tests were done with fewer selected compounds, including ethanol, 3-carene, and ethanol plus (−)-α-pinene in both Sierra Nevada and Allegheny Mountains. In both locations, ethanol plus (−)-α-pinene caught more woodwasps than other treatments. We discussed the implications of these results for understanding the chemical ecology of native woodwasps and invasive *Sirex noctilio* in North America. In California, 749 woodwasps were caught, representing five species: *Sirex areolatus* Cresson, *Sirex behrensii* Cresson, *Sirex cyaneus* Fabricius, *Sirex longicauda* Middlekauff, and *Urocerus californicus* Norton. In West Virginia 411 woodwasps were caught representing four species: *Sirex edwardsii* Brullé, *Tremex columba* Linnaeus, *Sirex nigricornis* F., and *Urocerus cressoni* Norton.

Keywords Monoterpenes · Ethanol · Host stress compounds · *Sirex areolatus* · *Sirex behrensii* · *Sirex edwardsii*

Introduction

Woodwasps in the family Siricidae (Hymenoptera) are well represented in North America, and all members of the subfamily Siricinae exclusively attack conifers (Middlekauff 1960; Schiff et al. 2006; Slippers et al. 2015). Three aspects of woodwasp biology are particularly relevant to their interaction with host trees and the development of potential monitoring and management tactics: their life cycles, their use of host volatiles in host location, and their exploitation of additional compounds from stressed host trees to optimize progeny survival. First, these subcortical feeders complete their life cycles almost entirely within host trees (Schiff et al. 2006; Slippers et al. 2012). When an adult female woodwasp emerges from her pupal case beneath the bark in summer, she mates and lays

✉ Nadir Erbilgin
erbilgin@ualberta.ca

[1] Department of Renewable Resources, University of Alberta, 4-42 Earth Science Building, Edmonton, Canada

[2] Emeritus. USDA-FS, Forest Health Technology Enterprise Team, Morgantown, WV, USA

[3] Emeritus. USDA-FS, Pacific Southwest Research Station, Berkeley, CA, USA

[4] AECOM, Environmental Services, San Francisco, CA, USA

[5] Division of Organisms and Environment, University of California, Berkeley, CA, USA

several eggs nearly two centimeters into the wood of a weakened or dying tree. Within three to four weeks, larvae hatch from the eggs and tunnel into the wood and begin eating the sapwood just under the bark; they then move into the heartwood deeper in the stem, and then later return to the outer sapwood to complete feeding. Larval feeding on decaying wood persists through the development of four or five immature stages, taking at least one year or as many as five years to complete. Pupation takes place at the end of the larval gallery in the outer sapwood. After five or six weeks as a pupa, the adult emerges by chewing through about two centimeters of bark and wood, leaving a round exit hole about five to ten millimeters in diameter.

Second, woodwasps are attracted to the principal volatile chemicals emitted by their host plants (Simpson 1976; Simpson and McQuilkin 1976; Böröczky et al. 2012; Crook et al. 2012). A number of studies in the eastern and Midwestern US have reported native woodwasps are attracted to host pine monoterpene hydrocarbons (MTH hereafter) such as α-pinene and 3-carene (Costello et al. 2008; Coyle et al. 2012). The same MTH similarly are attractive to the exotic European woodwasp (*Sirex noctilio* Fabricius) in its native and introduced range (Ayres et al. 2014; Corley et al. 2007; Crook et al. 2012; Hurley et al. 2015). Finally, woodwasp attraction is facilitated by so-called host stress compounds such as ethanol (Böröczky et al. 2012; Zylstra et al. 2010). These compounds usually are associated with recently dead or dying trees, especially those severely injured or killed by fire or drought (Ayres et al. 2014; Dodds et al. 2014).

Basic knowledge of the chemical ecology of native woodwasps in North America is limited (Costello et al. 2008; Coyle et al. 2012) because, unlike *S. noctilio*, they are not considered economically significant pests. Traps baited with ethanol and α-pinene alone or in combination with other plant volatiles and/or bark beetle pheromones have been shown to attract several woodwasp species. Only Coyle et al. (2012), however, have focused exclusively on the chemical ecology of native woodwasps and tested the attractiveness of several chemical compounds in Minnesota (USA). Others have used general kairomone attractants to sample woodboring insects in various orders, including long-horned beetles (Coleoptera: Cerambycidae), metallic wood-borers (Col: Buprestidae), and woodwasps (e.g., Costello et al. 2008).

In North America, a 70/30 blend of α/β-pinene is used commonly to attract *S. noctilio* to traps (Coyle et al. 2012), which generally catch only females (Crook et al. 2012; Dodds and de Groot 2012). Likewise, bioassays of putative pheromones of *S. noctilio* have produced mixed results, especially at low woodwasp densities (Cooperband et al. 2012; Dodds and de Groot 2012; Dodds et al. 2014; Hurley et al. 2015). It may be that additional host volatiles are involved in signaling, because girdled and/or herbicide-treated trap trees nearly always outperform traps baited with the recommended blend (Crook et al. 2012). Thus, we undertook the current investigation to determine the extent of native woodwasp-host chemical interactions in the eastern and western U.S.A., in which we tested two classes of chemicals associated with host trees, including MTH and alcohols. Understanding the chemical basis of woodwasp host-finding could be used to aid in development of lures for native and exotic woodwasps.

Methods and Materials

We investigated the chemical ecology of native woodwasps occurring in western (northern California) and eastern (West Virginia) conifer forests of North America by testing the attractiveness of several host tree chemicals to native woodwasps from 2004 to 2006. Release devices emitting attractants were attached to flight intercept traps (IPM Techn., Portland, OR, USA). Chemicals and release devices were provided by Synergy Semiochemicals Corp. (Burnaby, BC, CAN). We did not include 70/30 blend of α/β-pinene in our experiments because the attractiveness of this blend was not known at the time of our investigation. For all experiments, we placed traps 20 m apart and organized them into blocks spaced at least 500 m apart. During each collection period, traps were relocated to a different position within their block. This was accomplished by rotating the traps one position (e.g., first trap moved from position 1 into position 2, second trap moved from position 2 into position 3, etc.). This rotation procedure was used for subsequent collections, conducted at 2 wk. intervals. The date of collection, treatment, trap number, and position number were recorded for each trapped woodwasp. Specimens were stored in a refrigerator until being identified to species using Middlekauff (1960). Identifications of specimens were confirmed by Dr. W.W. Middlekauff at the Division of Insect Biology, University of California, Berkeley. Sample specimens were deposited into the University of California, Berkeley Essig Museum of Entomology.

Experiment 1 We initiated our field experiment using common conifer monoterpenes that have been shown to be attractive to various woodboring insects including woodwasps (Byers 1995; Erbilgin et al. 2001; Gijzen et al. 1993; Kelsey and Joseph 2003; Simpson and McQuilkin 1976). The following six treatments were tested in California: (1) (−)-α-pinene (97% pure), (2) (+)-α-pinene (96% pure), (3) (−)-β-pinene (98%), (4) (+)-3-carene (96%), (5) Red Turpentine Lure (a blend of 1:1:1 ratio of (+)-α-pinene, (−)-β-pinene, and (+)-3-carene), and (6) blank control. The red turpentine lure was included because it was commonly used to trap woodboring beetles at the time of our field experiments. The approximate release rate of α-pinene was 2500 mg/d and 30–50 mg/d for the remaining MTH. The release rate of α-pinene was higher than the others because it usually constitutes a large portion of

many conifer species (Gijzen et al. 1993). The experiment was conducted in seven blocks in mixed-evergreen coniferous Blodgett Experimental Forests (38°53′11″N, 120°38′52″W; 397 m above sea level) near Georgetown in northern California from September 20 to October 6 2004. Each treatment was replicated once in each block (total 42 traps). Traps were emptied and rotated at 4 d intervals (total 4 collections).

Experiment 2 Based on the results of our 2004 experiment (Table 1) and electroantennography studies conducted by Simpson (1976), we tested the attractiveness of ethanol (99%), and two chemical classes of terpenes including three MTH [(−)-α-pinene, (−)-β-pinene, (+)-3-carene] and four monoterpene alcohols [(−)-*cis*-verbenol (97%), (+)-*cis*-verbenol (97%), (−)-*trans*-verbenol (96%), (+)-*trans*-verbenol (96%)]. (−)-α-Pinene, (+)-3-carene, and (−)-β-pinene were selected based on the results of our 2004 experiments, which revealed that they were the three most attractive compounds that caught 85% of total woodwasps. In addition, Simpson (1976) investigated electroantennography responses of *S. noctilio* and reported that these woodwasps reacted positively to *cis*-verbenol and *trans*-verbenol present in *Pinus radiata*. *Cis*-Verbenol and *trans*-verbenol, typical oxygenation products of α-pinene, also are significant components in the chemical ecology of North American bark beetle species including mountain pine beetle (*Dendroctonus ponderosae*) and several pine engraver beetles (*Ips* spp.) (Allison et al. 2012; Pureswaran et al. 2000). To improve the spectrum of woodboring insects attracted, we tested both enantiomers of *cis*-verbenol and *trans*-verbenol in our experiments.

The approximate release rates were 400 mg/d for ethanol, 2500 mg/d for α-pinene, and 30–50 mg/d for the remaining chemicals. We established four mixed-evergreen coniferous forest sites in Blodgett Experimental Forests and assigned one site for one of the two chemical classes. Each stand had 3 blocks, and each treatment was replicated once in each block. The number of traps in each block varied depending on the number of compounds tested (plus blank control) in each class (total 30 traps). All traps were set up on July 25, 2005, and were emptied and rotated every 2 wk. until October 20, 2005 (total 6 collections per treatment per class).

Experiment 3 Based on the results obtained in 2005 (Table 2), we baited traps with fewer compounds in 2006. The following four treatments were tested in both California and West Virginia: (1) ethanol, (2) (+)-3-carene, (3) ethanol plus (−)-α-pinene, and (4) blank control. In California, the experiment was conducted in three blocks, while we used four blocks in West Virginia. All sites in West Virginia were located in mixed-evergreen coniferous forests in Monongahela National forests, near the town of Rimel (38°07′24″N 79°57′05″W; 748 m above sea level). In both locations, the experiment was conducted from June 30 to September 26, and each treatment was replicated once in each block. Traps were emptied and rotated at 2 wk. intervals (total 6 collections).

Statistical Analyses Mean number of insects per trap per sampling interval was analyzed using a mixed effects Poisson regression model for over-dispersed distributions of count data to address potential overdispersion arising from the repeated measurements.

$$\text{Expected}\left[\text{count}_{i,j,\text{site}} \middle| C_{\text{block*site}}, \varepsilon_{\text{site}}\right] = e^{\text{Treat*week}_{ij}+\lambda_{\text{block*site}}+\varepsilon_{\text{site}}}$$

where i is the number of treatments and j sampling periods, and λ and ε are the overdispersion errors due to remeasurement in the same block in site during the sampling weeks (Julian days).

A Generalized Estimating Equations (Liang and Zeger 1986) method was used to estimate the model's parameters with SAS (v. 9.1.3; PROC GENMOD), and the Wald *chi-square* test with the Bonferroni adjustment was used for multiple comparisons for an experiment-wise error rate of 0.05. The data per trap were temporally autocorrelated because the same site was remeasured six times (sampling periods). This autocorrelation was accounted for by including site*block effect and Site as random effects.

Results

In three years of trapping in California, we caught 749 woodwasps in five species. *Sirex areolatus* Cresson made up 31.91% of the total catch, followed by *Sirex behrensii* Cresson (22.03%), *Sirex cyaneus* Fabricius (18.29%), *Sirex longicauda* Middlekauff (14.42%), and *Urocerus californicus* Norton (13.35%). We caught 411 woodwasps in four species in West Virginia. *Sirex edwardsii* Brullé was the most abundant with 34.55% of total catch, followed by *Tremex columba* Linnaeus (26.03%), *Sirex nigricornis* F. (25.55%), and *Urocerus cressoni* Norton (13.87%). Traps exclusively caught female wasps except for three males of *S. behrensii* in the (+)-3-carene treatment.

For the 2004 sampling, total woodwasp captures significantly differed among lures. Traps baited with (−)-α-pinene were most attractive to woodwasps in general, catching 42% of total woodwasps, followed by (+)-3-carene (19.57%), (−)-β-pinene (17.39%), the red turpentine lure (13.04%), and (+)-α-pinene (7.25%) (Table 1). The attractiveness of treatments also varied among species, with all baited traps generally capturing more individuals than unbaited controls. (−)-α-Pinene was the most attractive lure to *S. areolatus* and *S. longicauda*, whereas all lures except (+)-α-pinene were equally and most attractive to *S. behrensii* and

Table 1 Trap catch of several species of woodwasps (hymenoptera: siricidae) in traps baited with common conifer monoterpenes in Northern California, September 20-October 6, 2004

Treatments	Mean No ± SE of insects/trap/4-day					
	Sirex areolatus	*Sirex behrensii*	*Sirex longicauda*	*Urocerus californicus*	*Sirex cyaneus*	Total Siricids
(−)-α-Pinene	1.08 ± 0.11 a	0.32 ± 0.04 a	0.36 ± 0.04 a	0.18 ± 0.02 a	0.18 ± 0.02 a	2.11 ± 0.05 a
(+)-α-Pinene	0.11 ± 0.02 c	0.04 ± 0.01 b	0.04 ± 0.01 c	0.04 ± 0.01 b	0.14 ± 0.04 a	0.36 ± 0.03 c
(−)-β-Pinene	0.18 ± 0.02 c	0.29 ± 0.03 a	0.11 ± 0.02 b	0.14 ± 0.02 a	0.14 ± 0.02 a	0.86 ± 0.3 b
(+)-3-Carene	0.32 ± 0.5 b	0.22 ± 0.3 a	0.11 ± 0.2 b	0.14 ± 0.02 a	0.18 ± 0.3 a	0.96 ± 0.3 b
RTB Lure*	0.14 ± 0.03 c	0.21 ± 0.04 a	0.00 ± 0.00 c	0.11 ± 0.02 a	0.18 ± 0.03 a	0.64 ± 0.0.4 bc
Unbaited Control	0.00 ± 0.00 d	0.00 ± 0.00 b	0.00 ± 0.00 c	0.00 ± 0.00 b	0.00 ± 0.00 b	0.00 ± 0.00 d
F-value $_{(5, 162)}$	3.21	2.87	2.75	2.38	2.41	3.87
P-value	0.009	0.016	0.021	0.041	0.039	0.002

Means with different letters in a column in each chemical class indicate significant differences among treatments at $\alpha = 0.05$ using the Bonferroni approach

*RTB lure is a blend of 1:1:1 ratio of (+)-α-pinene, (−)-β-pinene, and (+)-3-carene

U. californicus (Table 1). All lures were equally attractive to *S. cyaneus* (Table 1).

Among the two classes of chemicals tested in 2005, MTH caught 72.3% of all woodwasps (Table 2). For the MTH class, (+)-3-carene (62.27% of total catch) was overall the most attractive chemical to woodwasps followed by (−)-β-pinene (27.04%), and (−)-α-pinene (10.69) (Table 2). Although species-specific attraction varied among MTH, (+)-3-carene was the most attractive to all species, except for *S. areolatus* where (+)-3-carene and (−)-β-pinene were equally attractive. For all species except *S. areolatus*, the number of woodwasps captured in (−)-α-pinene-baited traps was not different from that of the control. However, the total catch of woodwasps responding to (−)-α-pinene was significantly higher than the control.

Among the five alcohols tested, ethanol was the most attractive, catching 41.4% of total woodwasps trapped (Table 2). (−)-*cis*-Verbenol and (−)-*trans*-verbenol were the second and third most attractive, respectively. Only the results from *S. areolatus* were statistically significant, and ethanol, (−)-*cis*-verbenol and (−)-*trans*-verbenol caught significantly more of this species than the control. Other species of woodwasps did not show any preference among alcohols tested.

In experiments conducted in California in 2006, traps baited with ethanol plus (−)-α-pinene caught more woodwasps (53.71%) than any other treatments, followed by (+)-3-carene (25.32%), ethanol (20.20%), and control (0.77%) (Table 3). Ethanol plus (−)-α-pinene caught the highest number of woodwasps. Ethanol and (+)-3-carene had the lowest catch among baited traps, followed by the control. For individual species, ethanol plus (−)-α-pinene was the most attractive blend for *S. areolatus*, *S. behrensii*, *S. longicauda*, and *U. californicus*. Both ethanol plus (−)-α-pinene and (+)-3-carene caught equal numbers of *S. cyaneus*.

In West Virginia, ethanol plus (−)-α-pinene was again the most attractive blend and caught 57.42% of all woodwasps trapped (Table 4). The second most attractive compound was (+)-3-carene, followed by ethanol, which was the least attractive. Ethanol plus (−)-α-pinene was the most attractive blend for all individual species, which caught 51.41% of *S. edwardsii*, 60% of *S. nigriconis*, 51.4% of *T. Columba*, and 78.95% of *U. cressoni*. (+)-3-Carene was the second most attractive compound. Attraction of *S. nigricornis* and *T. columba* did not differ between ethanol and (+)-3-carene. The number of trapped *U. cressoni* was similar between ethanol and control.

Discussion

Native woodwasps were attracted to 3-carene, β-pinene, α-pinene, *cis*-verbenol, and *trans*-verbenol. Likewise, these two classes also elicited antennal responses from *S. noctilio* in Europe (Simpson 1976). These results demonstrate that tree volatiles are important in woodwasp host-finding in coniferous forests (Böröczky et al. 2012; Sarvary et al. 2015). The most attractive compounds were those that are most abundant in pines including ethanol, (+)-3-carene, (−)-β-pinene, and (−)-α-pinene (Böröczky et al. 2012; Gijzen et al. 1993), supporting the hypothesis of a semiochemical-based co-evolutionary relationship between woodwasps and their host trees. It is noteworthy to state that the communities of woodwasps trapped in California and West Virginia were non-overlapping, suggesting the importance of host chemicals to geographically distinct native woodwasp species across North America (Coyle et al. 2012). Because our study included only widely separated woodwasp species found in

Table 2 Trap catch of several species of woodwasps (hymenoptera: siricidae) in traps baited with one of two classes of chemical compounds in Northern California, July 25-October 20, 2005

Treatments/ Chemical Class	Mean No ± SE of insects/trap per week					
	Sirex areolatus	*Sirex behrensii*	*Sirex longicauda*	*Urocerus californicus*	*Sirex cyaneus*	Total Siricids
Monoterpenes						
(−)-α-Pinene	0.39 ± 0.12 b	0.23 ± 0.10 d	0.17 ± 0.09 c	0.06 ± 0.06 b	0.12 ± 0.08 b	0.95 ± 0.22 c
(+)-3-Carene	1.50 ± 0.15 a	1.17 ± 0.19 a	1.17 ± 0.15 a	0.84 ± 0.15 a	0.84 ± 0.15 a	5.50 ± 0.62 a
(−)-β-Pinene	1.23 ± 0.15 a	0.67 ± 0.11 b	0.34 ± 0.14 b	0.12 ± 0.08 b	0.06 ± 0.06 b	2.39 ± 0.31 b
Unbaited Control	0.00 ± 0.00 c	0.00 ± 0.00 d	0.00 ± 0.00 c	0.00 ± 0.00 b	0.00 ± 0.00 b	0.00 ± 0.00 d
F-value $_{(3, 68)}$	5.24	6.01	4.38	3.78	2.99	7.45
P-value	0.003	0.001	0.007	0.014	0.037	<0.0001
Alcohols						
Ethanol	0.65 ± 0.14 a	0.17 ± 0.09	0.23 ± 0.10	0.17 ± 0.09	0.23 ± 0.10	1.45 ± 0.35 a
(−)-*cis*-Verbenol	0.37 ± 0.11 b	0.18 ± 0.11	0.07 ± 0.09	0.07 ± 0.09	0.12 ± 0.08	0.81 ± 0.19 b
(+)-c*is*-Verbenol	0.17 ± 0.09 bc	0.06 ± 0.06	0.12 ± 0.08	0.06 ± 0.06	0.06 ± 0.06	0.45 ± 0.25 c
(−)-*trans*-Verbenol	0.27 ± 0.14 b	0.07 ± 0.0	0.13 ± 0.10	0.07 ± 0.09	0.13 ± 0.10	0.67 ± 0.25 bc
(+)-*trans*-Verbenol	0.06 ± 0.06 c	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.06 ± 0.06	0.12 ± 0.012 d
Unbaited Control	0.00 ± 0.00 c	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00 d
F-value $_{(5, 102)}$	3.11	1.07	0.98	0.75	0.85	2.87
P-value	0.012	0.381	0.437	0.588	0.518	0.018

Chemical compounds were selected based on electroantennography studies conducted by Simpson (1976) and Simpson and McQuilkin (1976). Means with different letters in a column in each chemical class indicate significant differences among treatments at $\alpha = 0.05$ using the Bonferroni approach

California and West Virginia, further sampling is required to validate this conjecture.

We found that synergism between two of the most common host compounds (ethanol and α-pinene) provided the strongest attractant to native woodwasps in both West Virginia and California. Synergism between ethanol and various MTH is commonly reported for other bark and woodboring insects (Chénier and Philogène 1989; Lindgren et al. 2012; Miller and Rabaglia 2009; Wood 1982). For example, ethanol plus α-pinene are used commonly to monitor the activities of several conifer-infesting woodboring beetles (Chénier and Philogène 1989; Costello et al. 2008; Miller 2006).

Ethanol is a product of anaerobic plant and microbial metabolism (Moeck 1970), and is present in varying levels in all pine species (Kimmerer and Kozlowski 1982). Increased production of ethanol along with other volatile compounds, such as ethylene, is indicative of host stress (Kelsey et al. 2014; Kimmerer and Kozlowski 1982; Manter and Kelsey 2008). Thus, it is not surprising that insect species that rely on stressed host trees—including a wide variety of bark and woodboring insects—have co-evolved to associate these signals with susceptible hosts (Joseph et al. 2001; Kelsey and Joseph 2003; Kelsey et al. 2013; Miller and Rabaglia 2009). α-Pinene is another highly abundant volatile host compound to which many species of woodboring insects are attracted (e.g., Byers 1995; Erbilgin and Raffa 2000). Based on our results and results reported in earlier studies, we hypothesize that peak woodwasp activity likely coincides with peak emission of ethanol and host MTH, e.g., when they are severely injured by fire, and the synchrony of insect behavior with host plant phenology may reflect a co-evolved response to emission patterns (Rieske and Raffa 1991).

Because a standard blend (70/30 α/β-pinene) to trap woodwasps had not been developed by the time of our experiments, its attractiveness was not tested relative to our most attractive lure (ethanol plus α-pinene). In Minnesota (USA), this blend caught more native woodwasps than ethanol plus α-pinene (Coyle et al. 2012). Ethanol plus α-pinene caught a total of 67 siricid woodwasps (0.01 per trap per day, Coyle et al. 2012) compared to 210 (0.83 per trap per day) and 236 (0.70 per trap per day) individuals in California and West Virginia, respectively, in the current study. Differences between this and the earlier study may simply reflect differences in the amount of chemicals released and/or regional differences in responses among woodwasp populations. Since Coyle et al. (2012) did not provide the release rates of chemicals tested, we cannot speculate on the possible effect of differences in release rates. In terms of composition of woodwasps between regions, the native woodwasp community in Minnesota is similar to that in West Virginia, as four species are common in both regions, albeit at much lower densities than in the earlier study.

Table 3 Trap catch of several species of woodwasps (hymenoptera: siricidae) to traps baited with one of various host chemicals in Northern California, June 30-September 26, 2006

Treatments	Mean No ± SE of insects/trap/week					
	Sirex areolatus	*Sirex Behrensii*	*Sirex longicauda*	*Urocerus californicus*	*Sirex cyaneus*	Total Siricids
Ethanol	0.95 ± 0.13 b	1.17 ± 0.20 b	0.61 ± 0.14 b	0.78 ± 0.19 b	0.89 ± 0.21 b	4.39 ± 0.46 b
(+)-3-Carene	1.28 ± 0.31 b	0.89 ± 0.20 b	0.78 ± 0.21 b	0.73 ± 0.19 b	1.84 ± 0.31 a	5.5 ± 0.36 b
Ethanol plus (−)-α-pinene	3.5 ± 0.26 a	2.89 ± 0.29 a	1.5 ± 0.27 a	1.62 ± 0.27 a	2.17 ± 0.22 a	11.67 ± 0.79 a
Unbaited Control	0.06 ± 0.06 c	0.00 ± 0.00 c	0.00 ± 0.00 c	0.11 ± 0.07 c	0.00 ± 0.00 c	0.17 ± 0.09 c
F-value $_{(3, 68)}$	5.38	4.97	2.99	3.19	4.13	5.88
P-value	0.002	0.004	0.037	0.029	0.01	0.001

Chemical compounds were selected based on the results of studies conducted in 2005. Means with different letters in a column indicate significant differences among treatments at $\alpha = 0.05$ using the Bonferroni approach

The results have implications for understanding the chemical ecology of native woodwasps and *S. noctilio* in North America. First, woodwasp attraction to host trees depends on chemical signals that are either not present or are released in relatively small amounts from host trees under normal conditions, but are produced in larger amounts in response to damage or stress. The release of ethanol from foliage into the atmosphere is often minimal (Kreuzwieser et al. 2001), occurring primarily through stomata (Schade and Goldstein 2002). Although some ethanol may also evaporate from fungal-infected pine stems (Gara et al. 1993), the amounts are relatively small compared to that from wounded or stressed stems (Manter and Kelsey 2008). Given that physiologically stressed trees are more attractive and susceptible to attacking *S. noctilio* females (Ayres et al. 2014; Dodds et al. 2014), ethanol production from stressed hosts could serve as a signal facilitating host location and oviposition by *S. noctilio*.

Second, the emission of abundant host volatiles, such as MTH, has strong impact on woodwasp attraction. Although healthy trees emit MTH both from foliage and stems (Schade and Goldstein 2002), emission rates are small relative to those from stressed trees (Gijzen et al. 1993). For example, α-pinene emission changes in response to physiological damage or stress in trees (Lusebrink et al. 2016). Female *S. noctilio* are frequently attracted to trees recently felled or injured by fire, windthrow, or lightning (Ciesla 2003; Zylstra et al. 2010). For example, in the northeastern USA, girdled trees sustained the highest *S. noctilio* trap captures within four weeks of insect flight (Zylstra et al. 2010). These results suggest that attraction is based primarily on MTH and possibly other volatiles released from injured tree stems (Salvary et al. 2015). Identifying these compounds could be a critical step to increase the efficacy of lures in monitoring and surveying activities for *S. noctilio*. Furthermore, Fernández Ajó et al. (2015) recently reported that volatiles released from fungal symbionts of *S. noctilio* seem to provide a better attractant than host pine compounds in Y-tube olfactometer, supported by walk-in flight tunnel studies by Sarvary et al. (2016).

Finally, the identification of high risk areas (i.e., stands subject to prolonged drought or having experienced a fire) and tests of trap-tree approaches should be incorporated with the deployment of attractive chemicals in long-term monitoring programs (Ciesla 2003; Dodds et al. 2014). Given the clustered distribution of attacked trees during the early stages

Table 4 Trap catch of several species of woodwasps (hymenoptera: siricidae) in traps baited with one of various host chemicals in West Virginia, June 30-September 26, 2006

Treatment	Mean No ± SE of insects/trap/week				
	Sirex edwardsii	*Sirex nigricornis*	*Tremex columba*	*Urocerus cressoni*	Total Siricids
Ethanol	0.88 ± 0.18 c	0.84 ± 0.18 b	1.04 ± 0.24 b	0.00 ± 0.00 c	2.75 ± 0.36 c
(+)-3-Carene	2.0 ± 0.32 b	0.92 ± 0.25 b	1.13 ± 0.27 b	0.5 ± 0.16 b	4.54 ± 0.68 b
Ethanol plus (−)-α-pinene	3.04 ± 0.29 a	2.63 ± 0.24 a	2.29 ± 0.21 a	1.88 ± 0.26 a	9.83 ± 0.51 a
Unbaited Control	0.00 ± 0.00 d	0.00 ± 0.00 c	0.00 ± 0.00 c	0.00 ± 0.00 c	0.00 ± 0.00 d
F-value $_{(3, 92)}$	4.78	4.02	3.99	3.89	8.96
P-value	0.004	0.01	0.01	0.012	<0.0001

Chemical compounds were selected based on the results of studies conducted in 2005. Means with different letters in a column indicate significant differences among treatments at $\alpha = 0.05$ using the Bonferroni approach

of woodwasp colonization in pine forests (Corley et al. 2007), it is important to use attractants to detect early stages of forest colonization by woodwasps so that infested trees can be promptly detected and treated.

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