CAPTURE OF *XYLOSANDRUS CRASSIUSCULUS* AND OTHER SCOLYTINAE (COLEOPTERA: CURCULIONIDAE) IN RESPONSE TO VISUAL AND VOLATILE CUES

Austin M. Gorzlancyk[1,*], David W. Held[1], Dong-Joo Kim[2] and Christopher M. Ranger[3]
[1]Auburn University Department of Entomology and Plant Pathology, 301 Funchess Hall, Auburn, AL 36849, USA

[2]Auburn University Dept. of Materials Engineering, 276 Wilmore Laboratory, Auburn, AL 36849, USA

[3]USDA-Agricultural Research Service, Horticultural Insects Research Laboratory, 1680 Madison Ave., Wooster, OH 44691, USA

*Corresponding author; E-mail: amg0043@auburn.edu

Abstract

In Jun and Jul 2011 traps were deployed in Tuskegee National Forest, Macon County, Alabama to test the influence of chemical and visual cues on the capture of bark and ambrosia beetles (Coleoptera: Curculionidae: Scolytinae). The first experiment investigated the attractiveness of traps baited with different ratios of ethanol to methanol (0:100, 25:75, 50:50, 75:25, 100:0). No differences occurred in total Scolytinae trap captures for any of the various ratios of ethanol to methanol. The second experiment tested the attractiveness of ethanol-baited traps supplemented with light emitting diodes (LED) of various wavelengths to Scolytinae. Ethanol-baited traps supplemented with UV (395 nm) and green (525 nm) LEDs were more attractive than traps baited only with ethanol, but they were not more attractive than ethanol-baited traps supplemented with blue (470 nm) and red (625 nm) LEDs. This study indicates ethanol-baited traps supplemented with UV or green LEDs would be useful for detecting various Scolytinae.

Key Words: Alabama, attractants, LED, ethanol, methanol, UV, Scolytinae, traps

Resumen

En junio y julio del 2011, se utilizaron trampas en el Bosque Nacional de Tuskegee, en el condado de Macon, Alabama para probar la influencia de señales visuales y químicas sobre la captura de escarabajos ambrosia y de la corteza (Coleoptera: Curculionidae: Scolytinae). El primer experimento investigó el atractivo de las trampas cebadas con diferentes proporciones de etanol a metanol (0:100, 25:75, 50:50, 75:25, 100:0). No hubo una diferencia en el total de los escarabajos Scolytinae capturados en las trampas para todas las proporciones diferentes de etanol a metanol. El segundo experimento probó la atracción de los Scolytinae hacia las trampas cebadas con etanol y suplementadas con diodos emisores de luz (DEL) de diferentes longitudes de onda. Las trampas cebadas con etanol y suplementadas con UV (395 nm) y de DELs verdes (525 nm) fueron más atractivas que las trampas cebadas sólo con etanol, pero no fueron más atractivas que las trampas cebadas con etanol y suplementadas con DELs azules (470 nm) y rojos (625 nm). Este estudio indica que las trampas cebadas con el etanol y suplementadas con UV o DELs verdes serían útiles para la detección de varios Scolytinae.

Palabras Clave: Alabama, atrayentes, DEL, etanol, metanol, UV, Scolytinae, trampas

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Se utilizaron trampas para determinar el atractivo de diferentes proporciones de etanol a metanol, y los diodos emisores de luz suplementarios (LED) de diferentes longitudes de onda a los coleópteros ambrosia y de la corteza (Coleoptera: Curculionidae: Scolytinae). El total de los Scolytinae capturados no fueron diferentes en cualquiera de las proporciones de etanol a metanol, sin embargo las trampas con UV suplementario y los LED verdes eran más atractivos que las trampas cebadas sólo con el etanol. Este estudio indica que las trampas cebadas con el etanol y suplementadas con UV o LEDs verdes serían útiles para la detección de varios Scolytinae.

Palabras Clave: Alabama, atrayentes, LED, etanol, metanol, UV, trampas

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The granulate ambrosia beetle, *Xylosandrus crassiusculus* (Motschulsky) (Coleoptera: Curculionidae: Scolytinae), is an ambrosia beetle of Asian origin that was first detected in the continental United States near Charleston, South Carolina (Anderson 1974). While *X. crassiusculus* is considered only a nuisance pest in some areas, this insect has caused significant losses (in excess of US $5,000 per nursery) in several nurseries across the southeastern United States and in Texas (Ree & Hunter 1995). The damage potential of this pest is further amplified by *X. crassiusculus* having in excess of 120 potential tree host species, ranging from hardwoods to pine in both the nursery setting and forest (Weber & McPherson 1983; Solomon 1995; Hudson & Mizell 1999; Oliver & Mannion 2001). Scolytinae pests also include bark beetles, which are the most significant pest of coniferous forests, but can also attack deciduous trees.

Adult ambrosia beetles cause mechanical damage to trees as they tunnel into the heartwood of their hosts. Spores of a symbiotic fungus, which serves as a source of nutrition for the larvae and adult ambrosia beetles, are found within a pouch-like structure called a mycangium. As ambrosia beetles tunnel, the tunnel walls are coated with spores of the symbiotic fungus giving rise to colonial proliferation (Stone et al. 2007). The symbiotic fungus can block parenchyma cells within the vascular tissue of the trees, but Dute et al. (2002) found ambrosia beetle fungi were not particularly pathogenic. However, secondary pathogens such as *Fusarium* spp. can also be introduced to the host during the process of tunneling, or due to the open wounds created in trees. Secondary pathogens are suspected of playing a role in the eventual dieback and the death of infested hosts (Reding et al. 2010).

The physiological condition of a potential host tree plays an important role in the selection behavior of *X. crassiusculus* and other ambrosia beetles (Ranger et al. 2012a). When a tree is stressed, it can produce and emit acetaldehyde, acetic acid, acetone, ethane, ethanol, ethylene, and/or methanol (Kimmerer & Kozlowski 1982; Kimmerer & MacDonald 1987; Holzinger et al. 2000; Ranger et al. 2012a). Water damage, improper planting, drought stress, pollutants, low temperature, impaired root function, and diseases are some of the factors that can lead to the emission of ethanol and other stress-related volatile cues (Kelsey 2001; Ranger et al. 2012). This gaseous ethanol is important for attracting ethanol-responsive ambrosia beetles to invade host trees (Ranger et al. 2010; Ranger et al. 2012a,b). Ethanol is an attractant for a number of ambrosia beetles (Ranger et al. 2010, 2011, 2012b). Mixtures of ethanol and methanol have also increased trap captures of the bark beetle *Hypothenemus hampei* (Ferrari) compared to ethanol alone (Da Silva et al. 2006), indicating there is potential for synergistic relationships among volatile compounds.

In addition to olfactory cues, visual cues can also play a role during host selection by the Scolytinae. For instance, Da Silva et al. (2006) found green plastic bottle traps increased captures of *H. hampei* and other Scolytinae species compared to clear traps or transparent red traps. Similarly, Strom et al. (1999) demonstrated that both the southern pine beetle *Dendroctonus frontalis* (Zimmerman) and *Thanasimus dubius* (Fabricius) are affected by altering visual silhouettes on traps, finding that white paint and white trap paneling caught significantly fewer southern pine beetles than black or transparent coloration. In order to improve the detection and monitoring of various bark and ambrosia beetles, olfactory and visual cues were examined for their ability to enhance the attractiveness of ethanol-baited traps. Current management of ambrosia beetles in nurseries relies primarily on preventive treatments of insecticides applied to the trunks of trees, which can be improved with a greater knowledge of their seasonal activity and ecological interactions (Reding et al. 2010; Hudson & Mizell 1999).

Materials and Methods

Experiment 1: Influence of Olfactory Cues

The ability of methanol to enhance the attractiveness of ethanol to various Scolytinae was examined using Baker traps (Bambara et al. 2002; Oliver et al. 2004). Traps consisted of 2 L transparent bottles cut with 3 windows above the bottom and hung from a shepherd's hook, resulting in the traps being ≈1.2 m above the ground. A mixture of water and liquid dish detergent (100:1; v:v) was added to the bottom of the container to subdue and kill the entering beetles. An 8 dram (29.5 ml) clear glass vial with a cotton wick (8 × 2 cm, Cotton® American Fiber and Finishing Inc., St. Albemarie, North Carolina) containing the volatile attractants (28 mL) held in place with parafilm was hung inside each individual bottle trap. Release rates for the volatiles were held constant at 3.8 g per day across all treatments by filling the uniform bottle (diameter 25 mm) completely with a cotton wick. Trap treatments included the following ratios of 95% ethanol to 99.8% methanol (EMD chemicals Inc. Darmstadt, Germany) (v:v): 0:100, 25:75, 50:50, 75:25, 100:0. Attractant test solutions were replenished every 3 days over the course of the experiment.

Traps were deployed in a randomized complete block design in Tuskegee National Forest, Macon County, Alabama (N 32° 29' 19" W 85° 35' 39") from 12 Jun to 6 Jul 2011. Traps within each block were 6 m apart and were located in separate parts of Tuskegee National Forest. The trap sites were densely populated with a mixture of pine and hardwood trees and were partially shaded. Traps were returned to the laboratory for specimen identification. *Xylosandrus crassiusculus* was identified to species, but the remaining specimens were grouped as total Scolytinae.

Experiment 2: Influence of Visual Cues

A second trapping experiment was conducted to examine the attractiveness of different wavelengths to *X. crassiusculus* and other Scolytinae. Embedded into the top of the aforementioned traps and spaced equally along the top rim of the bottle were four 1.5 watt light-emitting diode (LED) bulbs (Boesch Built LLC, Waterford Township, Michigan) powered by a 6 volt battery (McMaster-Carr® Elmhurst, Illinois) that was fastened directly underneath the traps. The LED color wavelengths included 395 nm (UV), 470 nm (blue), 525 nm (green), 625 nm (red), and a blank control. The LED lights were powered continuously throughout the entire duration of the experiment. Each trap was also baited with the aforementioned 95% ethanol lure. Traps were arranged in randomized complete blocks in Tuskegee National Forest. The field test was conducted from 12 Jun to 6 Jul 2011. There were 7 replicates per treatment.

Statistics

A one-way analysis of variance (ANOVA) was performed on $\log_{10}(x + 1)$ transformed data to compare the attractiveness of varying ratios of ethanol to methanol on Scolytinae trap captures ($\alpha = 0.05$; PROC GLM; SAS Institute 2003). Differences between means were separated using Tukey's honestly significant difference (HSD) test ($P < 0.05$). A one-way ANOVA ($\alpha = 0.05$; PROC GLM; SAS 2003) on $\log_{10}(x + 1)$ transformed data and Tukey's HSD test ($P < 0.05$) was also used to compare Scolytinae trap captures associated with the various LED colors.

Results

Experiment 1: Influence of Olfactory Cues

No significant difference was detected in captures of all Scolytinae for traps baited with varying ratios of ethanol to methanol (100:0; 75:25; 50:50; 25:75; 0:100) ($F_{4,30} = 0.99$, $P = 0.43$) (Fig. 1). No significant differences in mean (± SE) captures of *X. crassiusculus* were detected for traps baited with ethanol alone (0.57 ± 0.30), methanol alone (1.0 ± 0.38), or varying ratios of ethanol to methanol at 75:25 (1.0 ± 0.53), 50:50 (1.57 ± 0.53), 25:75 (0.86 ± 0.59) ($F_{4,30} = 0.54$, $P = 0.71$).

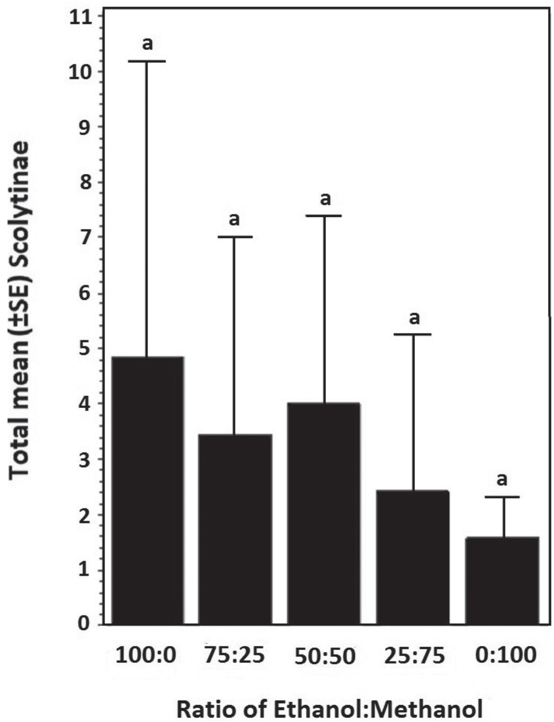

Fig. 1. Number (mean ± SE) of Scolytinae adults collected with bottle traps baited with various ratios of ethanol to methanol (100:0, 75:25, 50:50, 25:75, 0:100) deployed from Jun to Jul 2011 in Tuskegee National Forest, Alabama, USA. Means with different letters indicate significant differences (one-way ANOVA; $P < 0.05$).

Experiment 2: Influence of Visual Cues

Ethanol-baited traps supplemented with green (525 nm) and UV (395 nm) LED lights attracted significantly more Scolytinae than traps baited with ethanol alone ($F_{4,30} = 4.34$, $P = 0.007$) (Fig. 2). However, no significant difference was detected in total *X. crassiusculus* trap captures for ethanol-baited traps supplemented with UV (395 nm) (0.71 ± 0.29), blue (470 nm) (0.57 ± 0.43), green (525 nm) (1.0 ± 0.44), and red (625 nm) (0.14 ± 0.14) LEDs ($F_{4,30} = .99$, $P = 0.43$).

Discussion

Montgomery & Wargo (1983) did not find a mixture of 50% ethanol, 5% methanol, and 5% acetaldehyde to be more attractive than ethanol alone to various Scolytinae. Similarly, Ranger et al. (2010) found methanol-baited traps were more attractive to *Xylosandrus germanus* (Blandford) than acetaldehyde- and acetone-baited traps, but less attractive than ethanol-baited traps. Injecting sweetbay magnolia, *Magnolia virginiana* L.

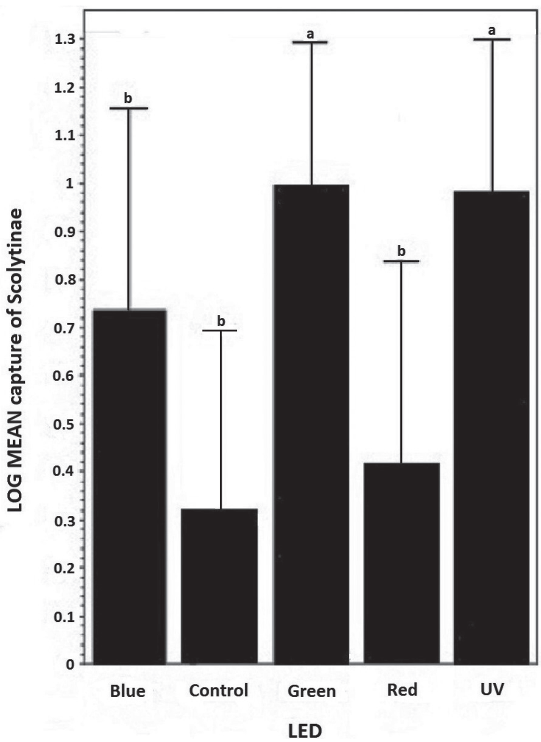

Fig. 2. Number (mean ± SE) of Scolytinae adults collected with ethanol-baited traps supplemented with various LED lights during Jun to Jul 2011 in Tuskegee National Forest, Alabama, USA. Means with different letters indicate significant differences (one-way ANOVA; $P < 0.05$).

(Magnoliales: Magnoliaceae), with methanol did not induce more ambrosia beetle attacks than a non-injected control, but over 120 attacks per plant were induced following ethanol-injection (Ranger et al. 2010). We found methanol alone was just as attractive as ethanol alone (Fig. 1), which differs from findings of other studies involving Scolytinae (Montgomery & Wargo 1983; Ranger et al. 2010). The lack of significant differences among any of the treatments involving various ratios of ethanol to methanol could be due to low trap counts from deploying traps after peak flight, which typically takes place in Mar to Apr in Macon County, Alabama (Gorzlancyk, pers. obs.) The relatively high release rate of ethanol and methanol from our lure apparatus may also have resulted in low trap counts. A positive concentration response to ethanol has been demonstrated for a few ambrosia beetles (Klimetzek et al. 1986; Ranger et al. 2011, 2012b), but too high of a release rate could have a repellent effect on ambrosia beetle orientation. Montgomery & Wargo (1983) found ethanol released at 2g/day attracted more Scolytinae than higher release rates. Thus, a mixture of ethanol and methanol cannot be recommended as an alternative or replacement of ethanol alone as an attractant for *X. crassiusculus* and other Scolytinae.

UV and household floodlights are currently used in general insect traps (Syms & Goodman 1987; Ferreira et al. 2012). Results from our current study demonstrated Scolytinae showed a higher attraction to ethanol-baited traps supplemented with green (525 nm) and UV (395 nm) LED lights compared to ethanol-baited traps alone. Attraction to green wavelengths has also been noted to occur in other members of the Curculionidae. For instance, Nakamoto & Kuba (2004) found evidence that the weevil *Euscepes postfasciatus* (Fairmaire) (Coleoptera: Curculionidae) preferred green LEDs to the UV light traps.

Our results suggest ethanol-baited traps supplemented with green or UV LEDs would be useful for detecting various Scolytinae in a nursery or forest setting. The integration of specific LED wavelengths into other trap configurations could potentially yield species specific responses. A more detailed examination is therefore warranted to elucidate the physiological mechanisms underlying the role of visual cues in behavioral responses by *X. crassiusculus* and other Scolytinae.

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