The Redbay Ambrosia Beetle (Coleoptera: Curculionidae: Scolytinae) Uses Stem Silhouette Diameter as a Visual Host-Finding Cue

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ABSTRACT The redbay ambrosia beetle (*Xyleborus glabratus* Eichhoff) is an invasive pest and vector of the pathogen that causes laurel wilt disease in Lauraceous tree species in the eastern United States. This insect uses olfactory cues during host finding, but use of visual cues by *X. glabratus* has not been previously investigated and may help explain diameter-related patterns in host tree mortality. The objective of this study was to determine whether *X. glabratus* females visually detect silhouettes of tree stems during host finding and are more likely to land on large diameter stems than smaller ones. Three field experiments were conducted in which stem silhouettes (black cylinders or standing nonhost pines) of varying diameters and identical capture surface areas were baited with essential oil lures. The Log_{10}-transformed number of *X. glabratus* trapped per week increased as a function of silhouette diameter in 2011 and 2012, using artificial silhouette diameters ranging 2–18 and 3–41 cm, respectively. When lures and capture surfaces were attached to standing pines ranging 4–37 cm in diameter, a positive relationship between Log_{10}(*X. glabratus* trap catch) and stem diameter was modeled using nonlinear quadratic plateau regression and indicated a diameter above which visual attraction was not enhanced; however, there was not a maximum diameter for enhanced *X. glabratus* attraction that was generally consistent across all experiments. These results 1) indicate that *X. glabratus* incorporates visual information during host finding, 2) help explain diameter-related patterns of redbay (*Persea borbonia* (L.) Sprengel) mortality observed during laurel wilt epidemics, and 3) are applicable to the management of this forest pest.

KEY WORDS laurel wilt, invasive species, stem diameter, visual orientation, *Xyleborus glabratus*

Before an insect can successfully use its host, it must first find, recognize, and accept that host as being suitable for oviposition or consumption (Kogan 1994). The finding of host plants by phytophagous insects is often a complex process involving behavioral responses to chemical, visual, and other stimuli (Prokopy and Owens 1983, Harris and Foster 1995), and while much research attention has been given to olfaction and other forms of chemoreception, visual host-finding cues are commonly used and integrated with other cues in many insect systems (Reeves 2011). In bark beetles (Coleoptera: Curculionidae: Scolytinae), for example, the importance of both olfaction and vision in host location is well recognized, and manipulation of visual stimuli (including stem or trap shape, orientation, and color) has been used successfully in bark beetle monitoring and management tactics (Payne and Coulson 1985, Strom and Goyer 2001, Strom et al. 2001). Although phytophagous insect responses to visual cues commonly occur only when appropriate chemical cues are present (Bernays and Chapman 1994), visual stimuli may serve as the dominant host-finding cue for some insects (Machial et al. 2012).

The redbay ambrosia beetle (*Xyleborus glabratus* Eichhoff) is native to Asia and was first detected in the United States near Port Wentworth, GA, in 2002 (Rabaglia et al. 2006). It is the principal vector of *Raffaelea lauricola* T.C. Harrington, Fraedrich & Aghayeva, the fungus that causes laurel wilt disease and one of several ambrosial symbionts present in the beetle's mycangia (Harrington et al. 2008, 2010). Laurel wilt is a deadly vascular disease of several tree species in the family Lauraceae and has caused widespread mortality of redbay (*Persea borbonia* L. Sprengel) in the Atlantic and Gulf Coastal Plain regions of the southeastern United States (Fraedrich et al. 2008). Laurel wilt also kills sassafras (*Sassafras albidum* (Nuttall) Nees) and avocado (*Persea americana* Miller) and the pathogen has been recovered from wilted specimens of the rare and threatened native plants pondspice (*Litsea aestivalis* (L.) Fernald) and pondberry (*Lindera melissifolia* (Walter) Blume (Fraedrich et al. 2008, Smith et al. 2009, Mayfield et al. 2008, Hughes et al. 2011). Laurel wilt occurs when female *X. glabratus* fly, locate, and bore into the stems or branches of live host trees and inoculate the xylem with *R. lauricola* spores. The fungus moves systemically through the plant and causes the foliage to discolor and wilt within

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a few weeks or months. *X. glabratus* has spread rapidly from its initial point of detection through an area extending from eastern North Carolina south to Florida and west to Mississippi (Bates et al. 2012). Concern exists that Lauraceae in other parts of the Americas may be at risk of damage from *X. glabratus* and laurel wilt disease.

To date, research on the host-finding behavior of *X. glabratus* has focused largely on beetle responses to volatile chemical cues. Several studies have demonstrated the attraction of flying *X. glabratus* females to host-produced volatiles emanating from cut bolts or essential oil lures, with significant effects of host species and lure type on beetle capture (Hanula et al. 2008, 2011; Kendra et al. 2011; Mayfield and Hanula 2012). Hanula et al. (2008) found no difference in *X. glabratus* attraction to infested versus uninfested bolts, suggesting that there is no aggregation pheromone or long-range attraction to volatiles emanating from beetle frass or fungal symbionts during host finding. Analyses of the volatile chemical composition of attractive substances, including redbay wood, manuka oil (extract of *Leptospermum scoparium* Forst. and Forst., family Myrtaceae), and phoebe oil (extract of *Phoebe porosa* Mez., family Lauraceae) suggest α-copaene is a primary, host-produced attractant (Hanula and Sullivan 2008), although other plant-derived compounds are likely important in host location as well (Kendra et al. 2011, Niogret et al. 2011). As for responses to visual stimuli, Hanula et al. (2011) evaluated *X. glabratus* catch on various colors of cross vane and sticky traps baited with manuka oil, but found no color to be consistently more attractive than others.

At least two previous studies have reported diameter-related patterns in redbay mortality during laurel wilt epidemics in forest stands (Fraedrich et al. 2008, Shields et al. 2011). In these studies, percent mortality of redbay because of laurel wilt was highest in the larger diameter classes, whereas seedlings and small diameter saplings were much less affected. Possible explanations for this pattern include 1) the susceptibility of redbay to the laurel wilt pathogen increases with stem diameter, or 2) the probability of attack by *X. glabratus* increases with stem diameter. The first explanation seems unlikely because artificial inoculation with the laurel wilt pathogen causes rapid wilting and mortality in small diameter redbay seedlings and saplings (Fraedrich et al. 2008) as well as in mature redbay trees (Mayfield et al. 2008a). If the second explanation (probability of *X. glabratus* attack increases with stem diameter) is correct, there are several mechanistic hypotheses that may explain or support it. One is that flying *X. glabratus* females use vision when finding hosts and land on larger diameter hosts preferentially. Alternatively, larger diameter trees may produce more or stronger volatile attractants, or are more likely to be encountered simply because of larger surface areas. In addition, host recognition or acceptance may be affected by physical or chemical bark surface properties (assessed by the insect after landing) that are more likely to promote entry of *X. glabratus* relative to smaller diameter stems.

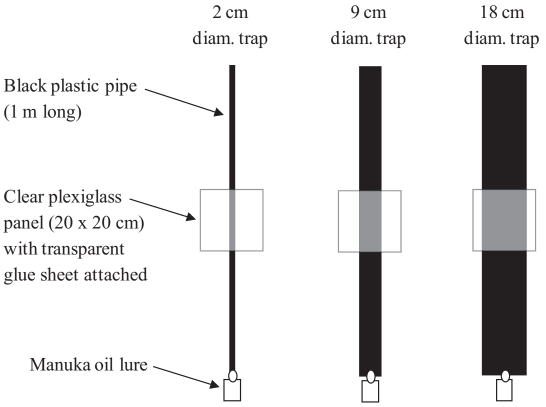

Fig. 1. Schematic of artificial stem traps designed to mimic tree boles of three different diameters, used to capture *Xyleborus glabratus* in July 2011 near Cordesville, SC.

The objective of this research was to test the hypothesis that flying *X. glabratus* females visually detect potential host stems during host finding and are more likely to land on large diameter silhouettes than smaller ones. To test this hypothesis, field trapping bioassays were implemented in which silhouette diameter varied, but the surface area on which beetles could be captured and the volatile attractants released from each trap were held constant.

Materials and Methods

Artificial Stem Experiments. *Experiment 1.* Experiments were conducted in 2011 and 2012 using artificial stem traps that were designed to visually mimic the boles of trees. In 2011, traps were constructed from sections of plastic pipe, 1 m long and of three different outside diameters: 2, 9, and 18 cm (Fig. 1). The body of the 2 and 9 cm diameter traps was constructed of schedule 40 poly vinyl chloride (PVC) plastic pipe (Charlotte Pipe, Charlotte, NC), whereas the body of the 18 cm trap was constructed of high density polyethylene (HDPE) plastic drain pipe (Hancor, Inc. Findlay, OH) because of unavailability of PVC pipe at the desired diameter. Each pipe was painted with flat black spray paint (Valspar Corp., Chicago, IL) for uniform color and texture. Two 400 cm^2 (20 by 20 cm) transparent Plexiglas panels were attached with wire at the midpoint of each trap on opposing sides. A clear photocopier transparency sheet (18 by 18 cm) printed with a 2 by 2 cm grid and covered with a thin layer of clear Tanglefoot Tangle-Trap insect glue (Contech Enterprises Inc., Victoria, BC, Canada) was attached to each Plexiglas panel with binder clips. A half-size lure of manuka oil (Synergy Semiochemical Corp., Burnaby, BC, Canada), an *X. glabratus* olfactory attractant (Hanula and Sullivan 2008), was attached to the bottom rim of each pipe trap, positioned 38 cm below one of the sticky sheets. The lure consisted of an internal cellulose matrix soaked with 2 ml of manuka oil housed in a plastic pouch. Field release rates of this lure are variable and in a recent study decreased from 170 mg/d to <5 mg/d over 14 d in Georgia (Hanula et al. 2013).

On 13 July 2011, traps were deployed on the border between a longleaf (*Pinus palustris* Mill.) and loblolly (*Pinus taeda* L.) pine stand and a floodplain stand of mixed hardwood or pine species that paralleled the headwaters of Alligator Creek on the Francis Marion National Forest near Cordesville, SC (33.16707 N, −79.87013 W). Traps were deployed during the expected peak flight period for *X. glabratus* in this region (late July through early September; Hanula et al. 2008). Common tree species in the floodplain stand included swamp tupelo (*Nyssa sylvatica* variety *biflora* (Walter) Sargent), swampbay, red maple (*Acer rubrum* L.), sweetgum (*Liquidambar styraciflua* L.), baldcypress (*Taxodium distichum* (L.) Richard), laurel oak (*Quercus laurifolia* Michaux), loblolly pine, and longleaf pine. Nearly all swampbays >7 cm in diameter were dead or dying from laurel wilt disease, and evidence of ambrosia beetle activity on such trees (sawdust, entrance holes) was widespread. Traps were hung on ropes suspended between two non-swampbay trees ≈2 m above ground. No trap was placed within 3 m of a swampbay tree. Traps were arranged as eight randomized complete blocks with three traps per block. Each block contained one trap of each of the three diameter treatments (2, 9, and 18 cm). Traps were spaced at least 12 m apart and blocks were spaced at least 15 m apart. After 2 wk, traps were removed and sticky sheets with trapped insects were returned to the laboratory. The number of *X. glabratus* trapped within the 18 by 18 cm grid of each sticky sheet was counted and expressed as beetles per trap per week.

Experiment 2. In August 2012, a similar experiment at a different location was conducted using different trap materials, a different lure, and a wider range of trap diameters. Traps were constructed by forming a cylinder with 14-gauge welded fence wire (5 by 10 cm mesh size) (Keystone Steel & Wire Co., Peoria, IL), wrapping the outer surface of the cylinder with black landscaping fabric (Greenscapes, Inc., Chatsworth, GA), and coating the outer surface with flat black spray paint (Valspar Corp.). Cylinders were 1 m long and of the following diameters: 3, 10, 20, 30, and 41 cm. Plexiglas panels with transparent sticky sheets were attached to each trap as described above. Each trap was baited with one cubeb oil lure (Synergy Semiochemical Corp.), another essential oil lure with properties attractive to *X. glabratus* (Hanula et al. 2013). The lure contained 2 ml of distilled cubeb oil in a 29-mm diameter bubble releaser; summer field release rates of this lure were 45 mg/d initially and decreased to 5–10 mg/d for several weeks thereafter (Hanula et al. 2013). Each lure was attached ≈38 cm below one of the sticky sheets. Traps were deployed in a mature loblolly pine plantation near Kelly, NC (34.47875 N, −78.29576 W) with dead and dying swampbay trees in the understory. Traps were arranged in a rectangular grid using five replicate rows (blocks) with each of the five trap diameters represented once per row. Each trap was hung from a rope suspended between two pine trees ≈2 m above ground, and no trap was placed within 3 m of a swampbay tree. Traps were spaced at least 15 m apart within rows and rows were spaced at least 20 m apart. After 12 d, sticky sheets were collected and replaced, and traps were systematically rotated two positions within each row to help minimize positional effects on beetle capture. Final trap collections were made 17 d later and the number of *X. glabratus* trapped during each period was expressed as beetles per trap per week. One or more sticky sheets (or their associated lures) were damaged or pulled down from four stem silhouettes during 1–13 August (affecting 2, 1, and 1 traps of the 30, 20, and 10 cm diameter treatments, respectively) and three stem silhouettes during 13–30 August (affecting 2 and 1 traps of the 3 and 30 diameter treatments, respectively). These traps were presumably damaged by black bears (*Ursus americanus* Pallas, as evidenced by footprints and black hairs in the sticky cards) and data associated with these traps were discarded.

Standing Pine Experiment. *Experiment 3.* On 27 July 2011, an experiment was initiated in the same location as the 2011 artificial stem experiment (experiment 1) described above, this time using standing live pine trees for the trap silhouettes. Standing pines stems were chosen as silhouettes in this experiment because 1) they potentially presented a more natural-looking host stem profile compared with suspended cylinders, 2) pines of a diversity of diameters were present at the site where *X. glabratus* was abundant, and 3) cut pine bolts (8–12 cm diameter and 60 cm long, not baited with lures) were unattractive to the redbay ambrosia beetle in a previous study (Hanula et al. 2008), and thus the standing pines were not expected to be a source of olfactory attractants. Twenty-four pine trees (longleaf or loblolly) were used, four from each of the following diameter classes: <5, 5 to <10, 10 to <15, 15 to <20, 20 to <25, and ≥25 cm. From a starting location at one end of the floodplain and pine forest boundary, one of the six diameter classes was randomly chosen (without replacement), and the nearest pine tree in that diameter class was selected and its diameter at 1.5 m above ground (diameter at breast height, DBH) was recorded. The next pine was selected by walking 12 m along the forest edge, randomly choosing a diameter class, finding the nearest tree in that class, and so on, until all diameter classes were represented four times. A transparent 20 by 20 cm Plexiglas panel was attached with an aluminum nail to each selected pine on the main stem 1.5 m above the ground. Gridded transparent sticky sheets were attached to each panel as described previously. A half-size manuka oil lure was stapled to the tree 15 cm below the bottom of the sticky sheet. After 7 d, sticky sheets were removed and the number of *X. glabratus* per trap was counted. Voucher specimens of *X. glabratus* were preserved in 70% alcohol and stored at the U.S. Department of Agriculture Forest Service Southern Research Station in Asheville, NC.

Data Analysis. For the 2011 artificial stem experiment (experiment 1), analysis of variance (ANOVA) appropriate for a randomized complete block design,

Table 1. Mean (and SE) no. of *X. glabratus* captured per week on artificial silhouette traps of various diameters and baited with manuka oil (experiment 1, Cordesville, SC, 2011) and cubeb oil (experiment 2, Kelly, NC, 2012)

Experiment (year)	Silhouette diameter (cm)	*X. glabratus*/trap/wk	
		Mean	SE
1 (2011)	2	12.8	1.5
	9	99.5	10.7
	18	166.7	15.5
2 (2012)	3	1.1	0.4
	10	4.6	0.9
	20	6.6	1.7
	30	14.7	3.9
	41	16.9	4.4

Note. In both experiments, data were Log_{10}-transformed for analysis of variance but untransformed means and SE (SEM) are shown here.

with factors block and treatment, was performed on Log_{10}-transformed catch per trap per week. The treatment effect with two degrees of freedom was partitioned using orthogonal polynomial contrasts into linear and quadratic (i.e., lack-of-fit to linear) components, and an F test was computed for each (Steel et al. 1997, p. 386–395). Analysis of data from the 2012 artificial stem experiment (experiment 2) was carried out on Log_{10}-transformed catch from both periods to reduce variance heterogeneity. The mixed model ANOVA accounted for blocks and period as sources of replication and included a fixed effect for treatment (with five levels) and random effects for block, treatment*block, period and treatment*period. The treatment effect (with 4 df) was partitioned into linear, quadratic and lack-of-fit to quadratic components (with 1, 1, and 2 df, respectively). PROC MIXED (version 9.2, SAS Institute Inc., Cary, NC) was used to carry out the ANOVA, and adjusted treatment means were obtained using the LSMEANS statement to account for the missing observations. Results for both experiments 1 and 2 are represented graphically with Log_{10}(*X. glabratus* trapped per week) plotted against silhouette diameter. Simple means and standard errors of *X. glabratus* trapped per week in experiment 1 and 2, calculated using the untransformed data and ignoring the experimental design, are presented in Table 1.

For the standing pines experiment (experiment 3), data were again Log_{10}-transformed to reduce variance heterogeneity. The graph of Log_{10}(X. *glabratus* trapped per week) versus silhouette diameter suggested a nonlinear relationship with Log_{10}(trap catch) increasing to a plateau as tree DBH increased. Several 2- and 3-parameter models were fit and assessed visually and via R^2 values, and based on these assessments, a quadratic-plateau segmented regression model provided the best description of the data. This model assumes a quadratic relationship between log-transformed trap catch (Log_{10}Y) and DBH (X) up to an unknown DBH value (D0), and a horizontal line relationship above that value. The D0 parameter represents the diameter above which there is no increase in trap catch and the join at D0 between the two segments is forced to be smooth, reducing the number of unknown parameters to 3. The 3-parameter model is written as:

$\text{Log}_{10}\text{Y} = \text{A} + \text{B}(\text{X}) + \text{C}(\text{X}^2)$ when $\text{X} < \text{D0}$;

$\text{Log}_{10}\text{Y} = \text{A} + \text{B}(\text{D0}) + \text{C}(\text{D0}^2)$ when $\text{X} \geq \text{D0}$

with the smoothness constraint $\text{C} = -\text{B}/(2 \text{ x } \text{D0})$.

The iterative Gauss–Newton method of optimization implemented in PROC NLIN (version 9.2, SAS Institute Inc., Cary, NC) was used to estimate the parameters of this nonlinear model (Rawlings et al. 1998, chapter 15).

To facilitate visual comparisons between the three experiments, quadratic-plateau models were also fit to the Log_{10}-transformed data from experiments 1 and 2 and graphed. Only three distinct silhouette diameters were used in experiment 1, so the fitted quadratic-plateau passed exactly through the treatment means and model assessment was not possible.

Results

Artificial Stem Experiments. *Experiment 1.* For the artificial stem experiments (experiments 1 and 2), simple means and standard errors for the untransformed *X. glabratus* trap catch by silhouette diameter indicate that both mean catch and variance increased with diameter (Table 1). In 2011, when using an artificial stem silhouette baited with an olfactory attractant (manuka oil), the effect of silhouette diameter on Log_{10}(*X. glabratus* trapped per week) was highly significant ($F = 177.28$; df = 2,14; $P < 0.0001$). Log-transformed catch in 2011 increased (at a decreasing rate) as silhouette diameter of manuka-oil baited traps increased from 2 to 18 cm (Fig. 2A; $F = 301.1$, df = 1, 14, $P < 0.0001$ and $F = 53.42$, df = 1, 14, $P < 0.0001$ for the linear and lack-of-fit to linear components of the treatment effect, respectively).

Experiment 2. In 2012 there was a significant effect of artificial silhouette diameter on Log_{10}-transformed catch of *X. glabratus* ($F = 6.76$; df = 4,7; $P = 0.017$). The relationship between Log_{10}(trap catch) and silhouette diameter of cubeb-oil baited traps (ranging 3–41 cm diameter) was mainly linear (Fig. 2A; $F = 22.72$, df = 1, 6, $P = 0.006$; $F = 2.32$, df = 1, 7, $P = 0.175$; and $F = 0.69$, df = 2, 7, $P = 0.535$ for the linear, quadratic, and lack-of-fit to quadratic components of the treatment effect, respectively), although graphical presentation of the data indicated a leveling-off of Log_{10}-transformed catch at the larger diameters (Fig. 2A).

Standing Pine Experiment. *Experiment 3.* When baiting standing pine trees of various diameters, there was a significant positive relationship between tree DBH and Log_{10}(*X. glabratus* trapped per week) ($F = 11.61$; df = 2, 21; $P \approx 0.0004$). Stem DBH explained 53% of the variability in log-transformed trap catch using a quadratic-plateau regression model (Table 2; Fig. 2B). The diameter at which *X. glabratus* trap catch was estimated to plateau (parameter D0) was 9.5 cm, with an approximate 95% confidence range of 5.5–13.4 cm (Table 2).

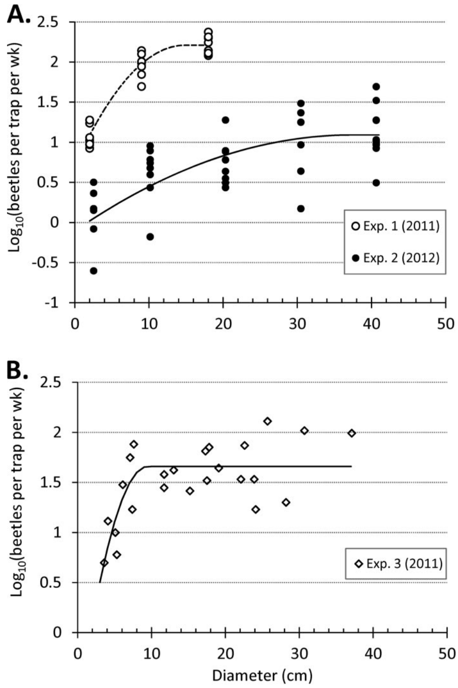

Fig. 2. Scatterplots of the relationship between silhouette diameter (centimeter) and the log-transformed number of *X. glabratus* captured per trap per week on baited silhouette traps in three different experiments. Panel A: Artificial silhouette traps baited with manuka oil (experiment 1, Cordesville, SC, July 2011) and cubeb oil (experiment 2, Kelly, NC, August 2012) lures. Panel B: Standing pine trees baited with manuka oil (experiment 3, Cordesville, SC, July–August 2011). In Panel B, the relationship is modeled using nonlinear quadratic plateau-segmented regression, where $\text{Log}_{10}\text{Y} = \text{A} + \text{B(X)} + \text{C}(\text{X}^2)$ when $\text{X} < \text{D0}$, and $\text{Log}_{10}\text{Y} = \text{A} + \text{B(D0)} + \text{C}(\text{D0}^2)$ when $\text{X} \geq \text{D0}$. D0 is an estimated parameter representing the diameter above which trap catch does not increase. Regression statistics and parameters for panel B are reported in Table 2. Quadratic plateau regression curves are also fitted to the data in panel A for the purposes of comparison with panel B. The regression curve for experiment 1 is shown as dashed because there are only three distinct silhouette diameters, so the fitted quadratic-plateau passes exactly through the treatment means and model assessment is not possible.

Discussion

Stem silhouette diameter had a significant effect on host finding by female *X. glabratus* in all three experiments of this study. When using the same type of lure and identical capture surface on dark silhouettes of various diameters, catch of *X. glabratus* increased with diameter. This effect was demonstrated at two different sites, using three different types of silhouette materials (black plastic pipe, black wire-and-fabric cylinders, and standing pines) and two different kinds of attractant lures (manuka oil and cubeb oil). Very few to no *X. glabratus* are captured in flight traps that lack volatile attractants, even when the trap provides a vertically oriented stem section or stem-like silhouette (Hanula and Sullivan 2008, Hanula et al. 2011, Mayfield and Hanula 2012). Thus, host finding by *X. glabratus* appears to integrate the use of both chemical and visual cues, although the relative importance or temporal sequence in which these two types of cues are used is still uncertain. Presumably, wider silhouettes are easier for the beetles to see, and larger diameters increase the probability that *X. glabratus* will find and land on the stem. This is consistent with several other species of stem-boring insects that integrate visual detection of vertical stem silhouettes with olfactory cues during host finding (Hunt and Raffa 1991; Strom et al. 1999; DeGroot and Nott 2001; Campbell and Borden 2006a, b). For example, visual discrimination of host stems by the ambrosia beetle *Trypodendron lineatum* Wood & Bright is synergized by the presence of host and nonhost volatiles (Vité and Bakke 1979, Campbell and Borden 2009).

Diameter-dependent host selection has been observed in a number of stem-colonizing insects. A visual preference for larger diameter branches by the weevil *Pissodes strobi* Peck is thought to be related to avoidance of host resistance mechanisms (resin ducts) (VanderSar and Borden 1977). Certain pine bark beetle species, such as *Dendroctonus ponderosae* Hopkins and *Dendroctonus valens* LeConte, preferentially colonize large diameter hosts, a pattern that is at least partially explained by enhanced beetle brood production afforded by thicker phloem (Amman 1972, Mitchell and Preisler 1991, Liu et al. 2011). Although our study was not designed to investigate the possible ecological or evolutionary mechanisms that underlie enhanced visual attraction of *X. glabratus* to larger diameters, this behavior may be of adaptive significance to this insect as well. As host trees die from infection with the laurel wilt pathogen, *X. glabratus* constructs egg galleries in the xylem in which it cul-

Table 2. Statistics for a nonlinear plateau regression model describing the relationship between the stem diam (centimeter) of manuka-oil baited pine trees (X variable) and the log-transformed no. of *X. glabratus* beetles captured per week (Y variable) near Cordesville, SC, Aug. 2011

Model parameters					Regression statistics					
			Approx. 95% CI						Approx.	
Param.[a]	Estimate	SE	Lower	Upper	Source	DF	MS	F	*P*	R^2
A	−0.8	1.0	−2.8	1.2	Model	2	0.85	11.6	<0.001	0.53
B	0.5	0.3	−0.1	1.1	Error	21	0.07			
D0	9.5	1.9	5.5	13.4	Total	23				

[a] $\text{Log}_{10}(\text{Y}) = \text{A} + \text{B(X)} + \text{C}(\text{X}^2)$ when $\text{X} < \text{D0}$; $\text{Log}_{10}(\text{Y}) = \text{A} + \text{B(D0)} + \text{C}(\text{D0}^2)$ when $\text{X} \geq \text{D0}$. D0 is an estimated parameter representing the diam above which trap catch does not increase.

tivates symbiont fungi as a food source for adults and larvae (Harrington et al. 2010). One adaptive hypothesis explaining a visual preference for larger diameters is that a greater volume of xylem allows for construction of longer egg galleries and more brood production relative to smaller diameter stems. Similarly, thicker stems may allow for decreased competition with conspecifics or other wood borers for the xylem resource, or may provide more optimal moisture content for fungal cultivation or larval development compared with smaller stems. These are testable hypotheses that could be evaluated to better understand *X. glabratus* host selection behavior.

The results of the current study help explain diameter-related patterns in redbay mortality that have been observed during laurel wilt disease epidemics. Fraedrich et al. (2008) monitored a stand infested with *X. glabratus* in northern Florida in which mortality of redbay trees ≥2.5 cm DBH increased from 10 to 92% over 18 mo. Redbays in the largest diameter classes succumbed to laurel wilt first, and by the end of the study, redbay mortality was <1, 79, 92, and 100% in the <2.5, 2.5–5, 5–10, and >10 cm DBH classes, respectively (Fraedrich et al. 2008). Similarly, Shields et al. (2011) observed 2, 30 and 100% redbay mortality in the seedling (<1.2 m tall), sapling (<10 cm DBH, ≥1.2 m tall) and overstory (≥10 cm DBH) strata, respectively, during a 2 yr period in another Florida stand. The current study suggests that larger diameter redbay stems are more attractive to *X. glabratus* than smaller stems, increasing their probability of being successfully attacked by females and inoculated with the laurel wilt pathogen.

There may be other characteristics of larger diameter host trees that act in concert with visual cues to increase their apparency or attractiveness to *X. glabratus* relative to smaller trees. Although these experiments were designed to hold olfactory cues constant and thereby isolate the visual effect of silhouette diameter on beetle capture, the strength of olfactory stimuli (e.g., host-produced volatile compounds) released from live redbay trees may vary with stem diameter. Furthermore, this study did not evaluate cues that *X. glabratus* may use to recognize or accept the host after landing on the surface, which might include additional visual, olfactory, tactile, or gustatory stimuli present at or beneath the bark surface. Such cues would determine whether *X. glabratus* bores into and inoculates the sapwood with *L. lauricola;* if these cues also varied with stem diameter, they could further explain diameter-related patterns in laurel wilt-caused mortality (Fraedrich et al. 2008, Shields et al. 2011).

The results presented here suggest that there may be stem silhouette diameters above which visual attraction of *X. glabratus* does not increase. In experiment 3, the quadratic-plateau model indicated that Log_{10}(trap catch) began to plateau at ≈9.5 cm DBH (Table 2; Fig. 2B). In the artificial stem experiments, although there was a strong, significant linear component of the diameter treatment effect, scatterplots of the data indicated that Log_{10}(trap catch) increased with silhouette diameter at a decreasing rate, and nonlinear plateau models were also consistent with these data (Fig. 2A). However, the tendency for Log_{10}(trap catch) to plateau appeared to occur at greater diameters in the artificial silhouette experiments compared with traps on standing pines (Fig. 2), indicating that there was not a generally consistent maximum diameter for enhanced *X. glabratus* attraction. These differences between experiments could have been a result of differences in visual properties of the silhouette material (pine bark vs. black cylinders), type of lure (manuka oil vs. cubeb oil), beetle abundance (high in 2011 vs. low in 2012), or environmental factors. Furthermore, it is possible that standing pines have properties that are repellent to *X. glabratus,* and that this repellency (perhaps chemically mediated) increased with increasing diameter. Additional studies are needed to determine whether the strength or shape of the positive relationship between stem diameter and *X. glabratus* visual attraction varies with host tree species, *X. glabratus* population densities, forest structure (e.g., stem density, basal area, and species composition), or other environmental variables (e.g., light). Additionally, this study did not evaluate the nature of this relationship at very small diameters (<2–3 cm).

The results of this study are applicable to the management of *X. glabratus* and laurel wilt and suggest areas of additional research. In detection or monitoring surveys for *X. glabratus,* a baited trap with a stem-like silhouette at least 10 cm in diameter should catch more beetles per unit capture surface area than a trap with a narrower or no distinct stem silhouette. Increasing trap diameter above this value may further enhance beetle capture. A standard, black, Lindgren multi-funnel trap (Lindgren 1983), which is commercially available and widely used for detection and monitoring of Scolytinae and other stem-boring beetles (Brockerhoff et al. 2006), has a silhouette diameter of ≈20 cm and is an effective trap for *X. glabratus* when baited with manuka oil (Hanula et al. 2011). Other funnel or stem silhouette-based traps of wider diameter could be compared with the standard 20-cm multi-funnel trap to see if *X. glabratus* capture can be improved. This study also suggests a behavioral mechanism by which small diameter trees survive during laurel wilt epidemics, a mechanism that could perhaps be exploited for management purposes, such as by altering the appearance of larger diameter hosts to visually disrupt host finding by *X. glabratus* (Strom et al. 1999). Furthermore, silvicultural manipulation of redbay diameter distributions (in favor of smaller diameter stems) may be worthy of evaluation as a management tool to help reduce stand-level impacts of *X. glabratus* and laurel wilt.

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References Cited

Amman, G. D. 1972. Mountain pine beetle brood production in relation to thickness of lodgepole pine phloem. J. Econ. Entomol. 65: 138–140.

Bates, C., L. Reid, R. Trickel, J. Eickwort, J. J. Riggins, and D. Stone. 2012. Distribution of Counties with Laurel Wilt Disease by Year of Initial Detection. U.S. Dep. Agric. Forest Service, Forest Health Protection, Southern Region. (http://www.fs.fed.us/r8/foresthealth/laurelwilt/dist_map.shtml).

Bernays, E. A., and R. F. Chapman. 1994. Host-plant selection by phytophagous insects. Chapman & Hall, New York, NY.

Brockerhoff, E. G., D. C. Jones, M. O. Kimberley, D. M. Suckling, and T. Donaldson. 2006. Nationwide survey for invasive wood-boring and bark beetles (Coleoptera) using traps baited with pheromones and kairomones. Forest Ecol. Manag. 228: 234–240.

Campbell, S. A., and J. H. Borden. 2006a. Close-range, in-flight integration of olfactory and visual information by a host-seeking bark beetle. Entomol. Exp. Appl. 120: 91–98.

Campbell, S. A., and J. H. Borden. 2006b. Integration of visual and olfactory cues of hosts and non-hosts by three bark beetles (Coleoptera: Scolytidae). Ecol. Entomol. 31: 437–449.

Campbell, S. A., and J. H. Borden. 2009. Additive and synergistic integration of multimodal cues of both hosts and non-hosts during host selection by woodboring insects. Oikos 118: 553–563.

De Groot, P., and R. Nott. 2001. Evaluation of traps of six different designs to capture pine sawyer beetles (Coleoptera: Cerambycidae). Agric. For. Entomol. 3: 107–111.

Fraedrich, S. W., T. C. Harrington, R. J. Rabaglia, M. D. Ulyshen, A. E. Mayfield III, J. L. Hanula, J. M. Eickwort, and D. R. Miller. 2008. A fungal symbiont of the redbay ambrosia beetle causes a lethal wilt in redbay and other Lauraceae in the southeastern United States. Plant Dis. 92: 215–224.

Hanula, J. L., and B. Sullivan. 2008. Manuka oil and phoebe oil are attractive baits for *Xyleborus glabratus* (Coleoptera: Scolytinae), the vector of laurel wilt. Environ. Entomol. 37: 1403–1409.

Hanula, J. L., A. E. Mayfield III, S. W. Fraedrich, and R. J. Rabaglia. 2008. Biology and host associations of the redbay ambrosia beetle, (Coleoptera: Curculionidae: Scolytinae), exotic vector of laurel wilt killing redbay trees in the southeastern United States. J. Econ. Entomol. 101: 1276–1286.

Hanula, J. L., M. D. Ulyshen, and S. Horn. 2011. Effect of trap type, trap position, time of year, and beetle density on captures of the redbay ambrosia beetle (Coleoptera: Curculionidae: Scolytinae). J. Econ. Entomol. 104: 501–508.

Hanula, J. L, B. T. Sullivan, and D. Wakarchuk. 2013. Variation in manuka oil lure efficacy for capturing *Xyleborus glabratus* (Coleoptera: Curculionidae: Scolytinae), and cubeb oil as an alternative attractant. Environ. Entomol. (in press).

Harrington, T. C., S. W. Fraedrich, and D. N. Aghayeva. 2008. *Raffaelea lauricola*, a new ambrosia beetle symbiont and pathogen on the Lauraceae. Mycotaxon 104: 399–404.

Harrington, T. C., D. N. Aghayeva, and S. W. Fraedrich. 2010. New combinations in *Raffaelea, Ambrosiella,* and *Hyalorhinocladiella*, and four new species from the redbay ambrosia beetle, *Xyleborus glabratus.* Mycotaxon 111: 337–361.

Harris, M. O., and S. P. Foster. 1995. Behavior and integration, pp. 3–46. *In* R.T. Carde and W.J. Bell (eds.), Chemical Ecology of Insects 2, Chapman & Hall, New York, NY.

Hughes, M., J. A. Smith, A. E. Mayfield III, M. C. Minno, and K. Shin. 2011. First report of laurel wilt disease caused by *Raffaelea lauricola* on pondspice in Florida. Plant Dis. 95: 1588.

Hunt, D.W.A., and K. F. Raffa. 1991. Orientation of *Hylobius pales* and *Pachylobius picivorus* (Coleoptera: Curculionidae) to visual cues. Great Lakes Entomol. 24: 225–229.

Kendra, P. E., W. S. Montgomery, J. Niogret, J. E. Peña, J. L. Capinera, G. Brar, N. D. Epsky, and R. R. Heath. 2011. Attraction of the redbay ambrosia beetle, *Xyleborus glabratus,* to avocado, lychee, and essential oil lures. J. Chem. Ecol. 37: 932–942.

Kogan, M. 1994. Plant resistance in pest management, pp. 73–128. *In* R. L. Metcalf and W. H. Luckman (eds.), Introduction to Insect Pest Management. Wiley, New York, NY.

Lindgren, B. S. 1983. A multiple funnel trap for scolytid beetles (Coleoptera). Can. Entomol. 115: 299–302.

Liu, Z., B. Wang, B. Xu, and J. Sun. 2011. Monoterpene variation mediated attack preference evolution of the bark beetle *Dendroctonus valens.* PLoS ONE 6: e22005. (doi:10.1371/journal.pone.0022005).

Machial, L. A., B. S. Lindgren, and B. H. Aukema. 2012. The role of vision in the host orientation behaviour of *Hylobius warreni.* Agric. For. Entomol. 14: 286–294.

Mayfield, A. E. III, and J. L. Hanula. 2012. Effect of tree species and end seal on attractiveness and utility of cut bolts to the redbay ambrosia beetle and granulate ambrosia beetle (Coleoptera: Curculionidae). J. Econ. Entomol. 105: 461–470.

Mayfield, A. E. III, E. L. Barnard, J. A. Smith, S. C. Bernick, J. M. Eickwort, and T. J. Dreaden. 2008a. Effect of propiconazole on laurel wilt disease development in redbay trees and on the pathogen in vitro. Arboric. Urban For. 35: 317–324.

Mayfield, A. E. III, J. A. Smith, M. Hughes, and T. J. Dreaden. 2008. First report of laurel wilt disease caused by *Raffaelea lauricola* on avocado in Florida. Plant Dis. 92: 976–976.

Mitchell, R. G., and H. K. Preisler. 1991. Analysis of spatial patterns of lodgepole pine attacked by outbreak populations of the mountain pine beetle. For. Sci. 37: 1390–1408.

Niogret, J., P. E. Kendra, N. D. Epsky, and R. R. Heath. 2011. Comparative analysis of terpenoid emissions from Florida host trees of the redbay ambrosia beetle, *Xyleborus glabratus* (Coleoptera: Curculionidae: Scolytinae). Fla. Entomol. 94: 1010–1017.

Payne, T. L., and R. N. Coulson. 1985. Role of visual and olfactory stimuli in host selection and aggregation behavior by *Dendrocontus frontalis*, pp. 73–82. *In* L. Safranyik (ed.), The Role of the Host in the Population

Dynamics of Forest Insects. Forestry Canada, Pacific Forestry Centre, Victoria, BC, Canada.

Prokopy, R. J., and E. D. Owens. 1983. Visual detection of plants by herbivorous insects. Annu. Rev. Entomol. 28: 337–364.

Rabaglia, R. J., S. A. Dole, and A. I. Cognato. 2006. Review of American Xyleborina (Coleoptera: Curculionidae: Scolytinae) occurring north of Mexico, with an illustrated key. Ann. Entomol. Soc. Am. 99: 1034–1056.

Rawlings, J. O., S. G. Pantula, and D. D. Dickey. 1998. Applied regression analysis: a research tool, 2nd ed. Springer, New York, NY.

Reeves, J. L. 2011. Vision should not be overlooked as an important sensory modality for finding host plants. Environ. Entomol. 40: 855–863.

Shields, J., S. Jose, J. Freeman, M. Bunyan, G. Celis, D. Hagan, M. Morgan, E. C. Pieterson, and J. Zak. 2011. Short-term impacts of laurel wilt on redbay (*Persea borbonia* [L.] Spreng.) in a mixed evergreen-deciduous forest in northern Florida. J. For. 109: 82–88.

Smith, J. A., T. J. Dreaden, A. E. Mayfield III, A. Boone, S. W. Fraedrich, and C. Bates. 2009. First report of laurel wilt disease caused by *Raffaelea lauricola* on sassafras in Florida and South Carolina. Plant Dis. 93: 1079.

Steel, R.G.D., J. H. Torrie, and D. A. Dickey. 1997. Principles and procedures of statistics: a biometrical approach. McGraw–Hill, New York, NY.

Strom, B. L., and R. A. Goyer. 2001. Effect of silhouette color on trap catches of *Dendroctonus frontalis* (Coleoptera: Scolytidae). Ann. Entomol. Soc. Am. 94: 948–953.

Strom, B. L., L. M. Roton, R. A. Goyer, and J. R. Meeker. 1999. Visual and semiochemical disruption of host finding in the southern pine beetle. Ecol. Appl. 9: 1028–1038.

Strom, B. L., R. A. Goyer, and P. J. Shea. 2001. Visual and olfactory disruption of orientation by the western pine beetle to attractant-baited traps. Entomol. Exp. Appl. 100: 63–67.

VanderSar, T.J.D., and J. H. Borden. 1977. Visual orientation of *Pissodes strobi* Peck (Coleoptera: Curculionidae) in relation to host selection behaviour. Can. J. Zool. 55: 2042–2049.

Vité, J. P., and A. Bakke. 1979. Synergism between chemical and physical stimuli in host colonization by an ambrosia beetle. Naturwissenschaften 66: 528–529.

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