Biology and Host Associations of Redbay Ambrosia Beetle (Coleoptera: Curculionidae: Scolytinae), Exotic Vector of Laurel Wilt Killing Redbay Trees in the Southeastern United States

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ABSTRACT The redbay ambrosia beetle, *Xyleborus glabratus* Eichhoff (Coleoptera: Curculionidae: Scolytinae), and its fungal symbiont, *Raffaelea* sp., are new introductions to the southeastern United States responsible for the wilt of mature redbay, *Persea borbonia* (L.) Spreng., trees. In 2006 and 2007, we investigated the seasonal flight activity of *X. glabratus*, its host associations, and population levels at eight locations in South Carolina and Georgia where infestations ranged from very recent to at least several years old. Adults were active throughout the year with peak activity in early September. Brood development seems to take 50–60 d. Wood infested with beetles and infected with the *Raffaelea* sp. was similar in attraction to uninfested redbay wood, whereas both were more attractive than a nonhost species. Sassafras, *Sassafras albidium* (Nutt.) Nees, another species of Lauraceae, was not attractive to *X. glabratus* and very few beetle entrance holes were found in sassafras wood compared with redbay. Conversely, avocado, *Persea americana* Mill., was as attractive to *X. glabratus* as swampbay, *P. palustris* (Raf.) Sarg., and both were more attractive than the nonhost red maple, *Acer rubrum* L. However, avocado had relatively few entrance holes in the wood. In 2007, we compared *X. glabratus* populations in areas where all mature redbay have died to areas where infestations were very active and more recent. Trap catches of *X. glabratus* and numbers of entrance holes in trap bolts of redbay were correlated with the number of dead trees with leaves attached. Older infestations where mature host trees had been eliminated by the wilt had low numbers of beetles resulting in trap catches ranging from 0.04 to 0.12 beetles per trap per d compared with 4–7 beetles per trap per d in areas with numerous recently dead trees. Our results indicate beetle populations drop dramatically after suitable host material is gone and provide hope that management strategies can be developed to restore redbay trees. The lack of attraction of *X. glabratus* to sassafras suggests that spread of *X. glabratus* may slow once it is outside the range of redbay.

KEY WORDS invasive species, exotic species, Lauraceae, laurel wilt

The redbay ambrosia beetle, *Xyleborus glabratus* Eichhoff (Coleoptera: Curculionidae: Scolytinae), was first discovered in the United States in 2002 at Port Wentworth near Savannah, GA (Rabaglia et al. 2006). The country of origin is unknown but the beetle is native to India, Bangladesh, Japan, Burma, and Taiwan (Rabaglia et al. 2006). Within 2–3 yr of the initial detection of *X. glabratus,* reports of dead and dying redbay, *Persea borbonia* (L.), trees in the Savannah area led to the discovery that a fungal symbiont of *X. glabratus* was responsible for a wilt disease in redbay trees (Fraedrich et al. 2008). Since then, the beetle and its associated fungus have spread rapidly. The rapid development of this epidemic through natural spread, plus suspected introductions through human-aided transport, have resulted in very high mortality rates of redbay trees along the Atlantic coast from just south of Charleston, SC, to south of Jacksonville, FL, and inland as much as 110 km. Within 2 yr of invading a new site, almost all redbay trees >2.5 cm in diameter are killed (Fraedrich et al. 2008). The fungus also can infect and kill other members of the Lauraceae, including sassafras, *Sassafras albidum* (Nutt.), and avocado, *Persea americana* Mill. (Fraedrich et al. 2008).

Redbay are evergreen, aromatic trees commonly found in forests of the southeastern Coastal Plain. Although generally relatively small (3–10 m in height), they can grow much larger on good sites (Brendemuehl 1990). Swampbay, *Persea palustris* (Raf) Sarg., is another *Persea* sp. sometimes recognized as a separate species (Sargent 1922, Dunkin and Dunkin 1988) or a variety of redbay [*P. borbonia* var. *pubescens* (Pursh) Little] (Brown and Kirkman 1990). Here, we refer to redbay as *Persea borbonia* sensu lato without attempting to discern species or varietal dif-

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ferences except in one experiment in Jennings State Forest, FL, where a botanist confirmed the trees tested were swampbay.

Information about *X. glabratus* within its native range is scant. Likewise, nothing was known about the biology of the beetle and its attraction to various host trees in the United States. Understanding the biology and host associations of *X. glabratus* are important steps in developing management strategies for this exotic beetle. Therefore, we investigated the seasonal abundance of *X. glabratus* adults on a barrier island in South Carolina, various factors contributing to its host finding success, and its attraction to other potential host tree species.

Materials and Methods

The studies were conducted from March 2006 to September 2007. Hunting Island State Park (Beaufort County, SC) located on a small barrier island in the southeastern corner of the state was the primary study site. The forest between the dunes and coastal marsh consisted of a dominant overstory of loblolly pine, *Pinus taeda* L.; a midstory of redbay; live oak, *Quercus viginiana* Nutt.; and cabbage palm, *Sabal palmetto* (Walt.) Lodd. ex J.A. & J.H. Schultes, trees; a shrub layer of waxmyrtle, *Morella cerifera* (L.) Small, and redbay; and an understory dominated by cabbage palm seedlings.

Seasonal Flight. In February 2006, several trees wounded by machinery at Edisto Beach, SC, were attacked by *X. glabratus* and the attacks were focused on wound sites, suggesting wounding increased attraction of the beetle. Therefore, to monitor adult flight of *X. glabratus,* we wounded four healthy and four recently infected redbay trees (i.e., wilt symptoms but no evidence of beetle attacks) on Hunting Island on 24 March 2006. Trees, 10–15 cm in diameter at 1.4 m above ground, were wounded by scraping the bark down to the xylem from an area ≈15 by 15 cm and ≈1.5 m above the ground because numerous beetle attacks were observed at this height on other attacked trees. A piece of heavy gauge steel wire was wrapped around each tree and shaped into a hook so traps could be hung on the trees directly over the wound sites. Traps were identical to the flight intercept traps used by Ulyshen and Hanula (2007), which consisted of two pieces of Plexiglas (20 by 30 cm) with a saw cut halfway through the middle in the longest direction. The two Plexiglas pieces were then slid together to form a +-shaped barrier to beetles attracted to the tree. The Plexiglas barrier was attached with wire to a white, 2-liter capacity plastic bucket hung directly below the barrier. Beetles attracted to the trees bumped into the barrier and fell into the bucket containing a solution of nontoxic RV antifreeze (propylene glycol). A single trap was hung on each tree. Traps were not oriented in a particular direction but they were hung directly over the initial wound site. Traps were checked at ≈2-wk intervals through 12 March 2007. To ensure trees remained attractive, we continued to wound them every 2 wk, as close to the traps as possible, by scraping a small amount (≈6 by 6 cm) of bark from the tree.

Attraction of Diseased, Healthy, and Wounded Redbay Trees. The fungus responsible for laurel wilt is a fungal symbiont of *X. glabratus*, found in mycangia at the base of each mandible, that is readily transmitted to redbay and other susceptible plants (Fraedrich et al. 2008). To determine whether this symbiotic fungus also produced attractants enabling beetles to locate suitable trees for brood development, we conducted an experiment consisting of four treatments: 1) healthy, nonwounded redbay trees; 2) healthy, wounded redbay trees; 3) diseased (infected with laurel wilt), wounded redbay trees; and 4) nonhost live oak (*Quercus virginiana* Mill.) trees. This experiment was run concurrently with the adult flight study so the wounded trees from that study also were used for this experiment to minimize damage to trees. Trees were selected in groups so they were ≈10 m apart within groups and groups of trees were at least 50 m apart. Groups consisted of the four treatments, and no trees had evidence of beetle attacks at the start of the study. Traps were placed on trees 24 March 2006 and checked every 2 wk until 3 June 2006. Traps were hung from wire wrapped around the tree so nonwounded trees had no known wound sites.

Attraction of *X. glabratus* Infested and Uninfested Redbay Bolts. Four bolts ≈0.6 m long were cut from two trees heavily infested with *X. glabratus* and infected with the fungus, and similar sized bolts from two healthy trees to determine whether *X. glabratus* produced pheromones or if beetles boring into trees increased their attraction to host-seeking beetles. Four bolts also were cut from a nonhost pine tree for comparison. Bolts were enclosed in black, noseeum mesh cloth sleeves sealed at both ends with wire ties to prevent new beetle attacks. Before encasing each in cloth mesh, a screw eye was inserted in one end and attached to a wire so the bolt could be hung from a rope tied between two nonhost trees. Bolts were suspended above the ground ≈1.5 m, and they were hung in four groups with each group containing the three treatments. Groups were spaced 50–100 m apart and bolts within groups were spaced 10 m apart. Traps consisting of a single Plexiglas panel (20 by 30 cm) attached to a white bucket partially filled with RV antifreeze were used to catch *X. glabratus.* The traps were hung from a wire hook wrapped around the screw eye so that traps were directly in front of the log bolts. Traps were checked every 2 wk from 12 July to 3 September 2006.

Attraction of Swampbay and Avocado. *X. glabratus* and its associated fungus have the potential to be serious pests in avocado orchards in Florida. We compared attraction of uninfested swampbay, avocado, and red maple, *Acer rubrum* L., bolts at Jennings State Forest, Clay County, FL. Bolts were hung as before (10 m apart within groups and 50 m or more between groups) and Plexiglas flight intercept traps were used to capture beetles attracted to host odors. Bolts and traps were deployed on 6 June and removed on 23 August 2007 and checked approximately every 2 wk. After trapping was completed two 100 cm^2 areas of bark were removed from each bolt to count the number of gallery entrance holes.

Afterward, all bolts were placed in black plastic rearing containers, constructed from file boxes, to determine whether the beetles successfully completed development in avocado wood.

Attraction of Redbay and Sassafras Bolts. Sassafras, *Sassafras albidum* (Nutt.) Nees, is another member of the family Lauraceae that can be infected by the laurel wilt (Fraedrich et al. 2008). The range of sassafras is much larger than redbay extending over much of the eastern half of the United States (Griggs 1990), so concern about whether the beetle will be able to attack sassafras in high numbers, complete brood development, and continue to spread through its range prompted a test of its attractiveness to *X. glabratus*.

In September 2006, four 0.5-m-long bolts were cut from redbay and sassafras trees ≈8–12-cm-diameter at breast height (dbh) on the Savannah River Site in Barnwell County, SC. A screw eye was inserted in one end of each bolt, and the bolts were enclosed in mesh cloth as described previously. A similar-sized sweetgum, *Liquidambar styraciflua* L., tree was selected as a nonhost tree for comparison and four bolts were cut from it and treated in the same manner. Bolts were hung in groups consisting of the three tree species so that each bolt in a group was suspended from a rope tied between two nonhost trees. Bolts within groups were at least 10 m apart and groups were spaced ≈30 m apart. A single panel Plexiglas flight intercept trap was attached to each bolt, and samples were collected every 2 wk from 21 September to 28 October 2006.

In March 2007, sticky traps were tested as an alternative to flight intercept traps and found to be equally effective for catching *X. glabratus*, so they were used in subsequent studies. Sticky traps were white, wing style trap bottoms (Scentry Biologicals, Inc., Billings, MT) attached to a Plexiglas panel with two medium size (3.2 cm) binder clips. A wire loop created by attaching a piece of picture hanging wire to holes in the top corners of the Plexiglas was hooked over a nail in the bolts to suspend traps vertically directly in front of the bolts.

In July 2007, sassafras bolts were cut from a single tree in Clarke County, GA, and compared with similar sized bolts cut from a healthy redbay tree on Hunting Island State Park. Bolts cut from a healthy live oak served as a nonhost control. Bolts were hung from ropes between nonhost trees in groups in the same manner as 2006. However, in 2007 two sticky traps were hung on each bolt to capture beetles attracted to them and the bolts were not enclosed in cloth screen to determine whether beetles used sassafras or oak for brood production. The experiment was replicated six times with three replicates on Hunting Island and the other three at Lake Warren State Park near Hampton, SC. Traps were checked every 2 wk from 16 July to 27 August. At each trap check a small portion of bark was scraped from each bolt to increase the release of volatile attractants.

Bolts were collected at the end of the study, the bark was removed in the laboratory, and *X. glabratus* gallery entrance holes were counted. The entrance holes are about the same diameter as paper clip wire (size no. 1, 0.8 mm in diameter), so the wire was used to ensure the larger entrance holes of *Xylosandrus crassiusculus* (Motschulsky), which commonly attacks redbay wood, were not counted. Afterward, the bolts were placed in rearing containers constructed from black plastic file boxes. A plastic jar lid was attached to the underside of the container at one end, and a 12-cm-diameter hole cut through the lid and the container. A clear plastic jar was screwed on to the lid where beetles attracted to the light were collected.

Beetle Populations at Varying Tree Densities. The fate of beetle populations after the death and deterioration of redbay host material is unknown. Beetle populations should decline and possibly disappear in areas where redbay suitable for breeding have been eliminated if no other wood is used for brood production. Eight sites were selected along the coast of South Carolina and Georgia representing a range of infestation histories from no known infestation to the longest infested areas where most redbay trees had been killed several years before. These sites were 1) I'on Swamp near Mt. Pleasant, SC, on the Francis Marion National Forest, an area with no history of infestation; 2) Edisto Beach State Park, SC, an area near the leading edge of the infestation; 3) Hunting Island State Park, SC, a heavily infested area with many dead and dying redbay; 4) Newhall Audubon Preserve on Hilton Head Island, SC, one of the oldest infested areas with no standing redbay with leaves attached; 5) the North Ridge Tract on Hilton Head also an old infested area; 6) Colonels Island, GA, an older infested area with very few redbay left; 7) a portion of the Richmond Hills Wildlife Management Area near Eulonia, GA, a heavily infested area; and 8) Jekyll Island State Park, GA, a recently infested area.

Three freshly cut uninfested redbay bolts were hung as described previously at each area. Bolts were spaced at least 75 m apart and, to increase attraction, two strips of bark were removed (≈5 cm in width and the length of the bolt) on opposite sides of each bolt. A sticky trap was stapled to one side of each bolt and a vial containing 5 ml of manuka oil was attached so it was near the center of the trap. Manuka oil was found to be attractive to *X. glabratus* just before the start of this experiment (J.L.H., unpublished data) and was added to increase the likelihood of catching beetles in low population areas. Manuka oil (Coast Biologicals Ltd., South Aukland, New Zealand) was released at a rate of ≈17 mg/d from a glass vial with a 4-cm-long cotton string wick extending above the plastic cap in which a 4-mm-diameter hole was drilled. Redbay bolts and traps were placed in the forests on 8–9 August 2007 and removed 4–5 September 2007. In September, all live and recently dead (some brown leaves present) redbay trees were counted and measured in an 0.08-ha plot centered at the point where each bolt was located (three plots/location). Only redbay 2.5 cm in diameter and larger were measured because smaller sizes are rarely attacked (Fraedrich et al. 2008). Observations were also made on the presence or absence of dead redbay trees visible beyond the plot boundaries.

Statistical Analyses. Each trial was analyzed as a two-way analysis of variance (ANOVA) with treatment and

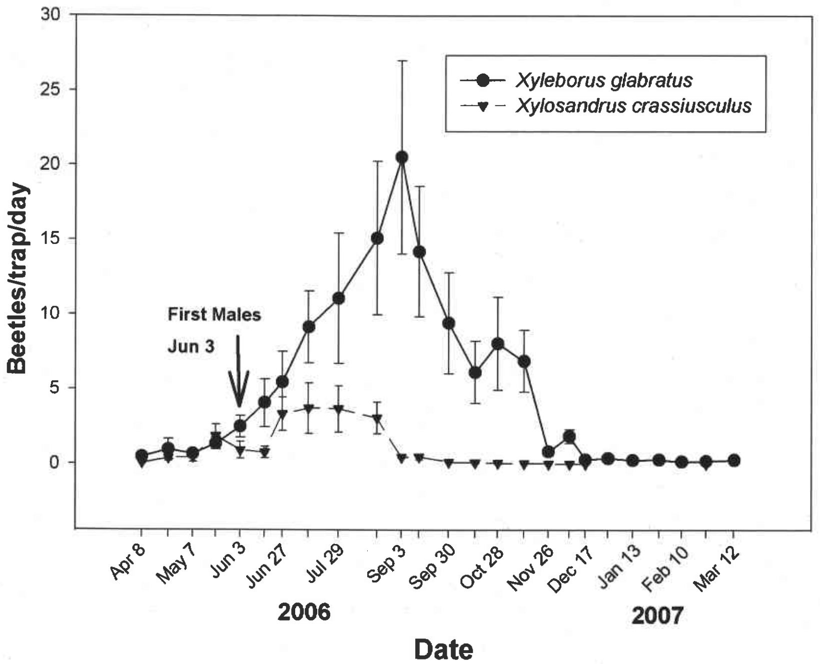

Fig. 1. Mean number (±SE) of *X. glabratus* and *X. crassiusculus* beetles captured on flight intercept traps hanging on wounded redbay trees in Hunting Island State Park, South Carolina.

replicate (groups) as the independent variables, and beetle catch or number of entrance holes as the dependent variables by using Proc GLM (SAS Institute 1985). Means were separated using the Ryan–Einot–Gabriel–Welsch (REGWQ) multiple comparison test (Day and Quinn 1989). Data were transformed using a log transformation when the Shapiro–Wilk test for normality (Proc Univariate, SAS Institute 1985) indicated the data were not normally distributed. In most cases, trap catches were expressed as the number of beetles captured per trap per day. Relationships between *X. glabratus* entrance holes and traps catches, and trap catches or entrance hole densities and total numbers of dead redbay trees per plot were analyzed by simple linear regression by using Proc GLM.

Results

Seasonal Flight. Trap catches of *X. glabratus* ranged from 0.25 to 0.50 beetles per trap per d during April and early May (Fig. 1). In late May, catches began to increase and continued to do so through the summer to a high of 21 beetles per trap per d on 3 September 2006. Beetle catches steadily declined through the fall months and numbers were very low during the winter, although a few were caught in almost every collection period.

X. crassiusculus, another Asian ambrosia beetle that frequently breeds in dead and dying redbay trees, was also regularly caught in beetle traps although numbers tended to peak earlier than those for *X. glabratus*, and occurred at significantly lower levels.

Attraction of Diseased, Healthy, and Wounded Redbay Trees. Wounded diseased host trees were not more attractive to redbay ambrosia beetles than wounded healthy host trees (Fig. 2). Both wounded healthy and wounded diseased host trees caught more beetles than nonhost oak trees. Traps on wounded host trees caught almost twice as many beetles as traps on healthy nonwounded host trees, but the results were not statistically significant. Likewise, traps on healthy nonwounded host trees did not capture significantly more beetles than traps on nonhost oak trees, although traps on nonwounded redbay trees caught approximately three times as many beetles as traps on oak trees.

Attraction of *X. glabratus* Infested and Uninfested Redbay Bolts. Bolts cut from trees heavily attacked by *X. glabratus* were not more attractive than bolts from healthy trees showing that beetles excavating galleries in host trees do not produce additional attractants (Fig. 3). In addition, bolts cut from healthy redbay trees remained attractive to *X. glabratus* for at least 2 mo (Fig. 4).

Attraction of Swampbay and Avocado. Avocado wood was as attractive to *X. glabratus* as wood from healthy swampbay trees (Fig. 5), and both species were more attractive than red maple. However, although entrance hole densities were not significantly different among the three species, avocado and red maple wood had less than two entrance holes per 100 cm^2 compared with six holes per 100 cm^2 in swampbay wood (Fig. 5).

Attraction of Redbay and Sassafras Bolts. In 2006, the numbers of *X. glabratus* captured in traps on

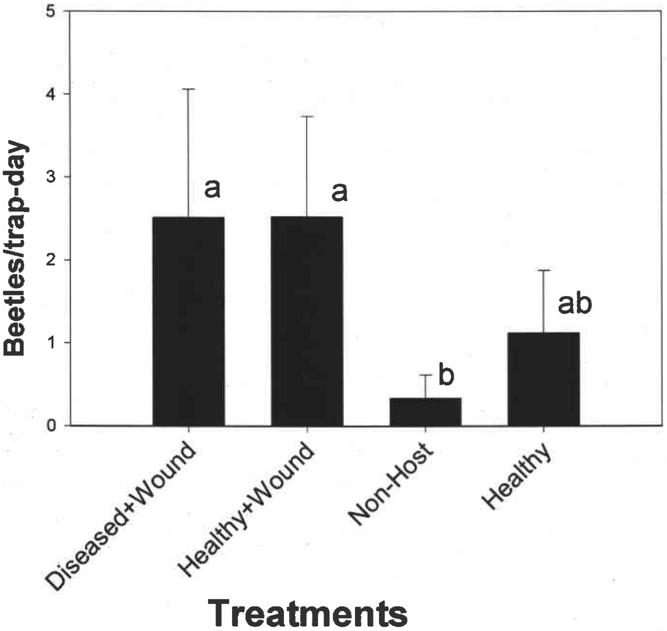

Fig. 2. Mean number (±SE) of *X. glabratus* females/trap/d in traps on redbay or nonhost (*Quercus virginana* Mill.) trees in Hunting Island State Park, South Carolina. Traps were operated from 24 March to 3 June 2006. Columns with the same letter above them are not significantly different at $P < 0.05$ (REGWQ, SAS Institute 1985).

bolts cut from healthy redbay, sassafras, or sweetgum were not significantly different ($P < 0.05$; Fig. 6). The numbers of beetles captured in traps on redbay bolts in this experiment were very low compared with similar experiments earlier in the year (e.g., Fig. 4). However, traps near redbay captured more than twice as many beetles as traps near sassafras or sweetgum, suggesting sassafras was not as attractive as redbay to *X. glabratus.*

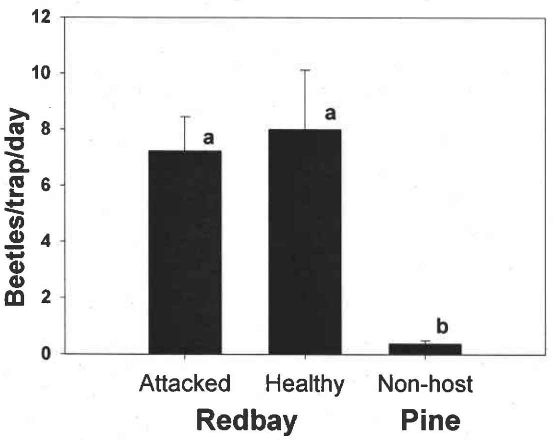

Fig. 3. Mean number (±SE) of *X. glabratus* females per trap per d in traps hanging next to bolts cut from redbay or nonhost pine trees at Hunting Island State Park, South Carolina. Traps were deployed from 12 July to 3 September 2006. Columns with the same letter above them are not significantly different at $P < 0.05$ (REGWQ, SAS Institute 1985).

In contrast, the number of *X. glabratus* caught on redbay bolts in 2007 was 40 times greater than the number caught on sassafras or live oak bolts (Fig. 7). Sassafras and oak were not significantly different. Entrance hole densities averaged 21.5 holes per 100 cm^2 on redbay compared with 0.6 holes per 100 cm^2 on sassafras and 0.04 holes per 100 cm^2 on oak (Fig. 7).

Beetle Populations at Varying Tree Densities. The number of beetles captured on sticky traps attached to redbay bolts and the density of entrance holes in the bolts were directly related to the number of dead redbay trees within 0.08-ha plots centered on the trap locations (Fig. 8). Likewise, the density of gallery entrance holes exhibited a positive linear relationship to numbers of beetles captured ($y = 2.3 + 4.7x$, $P < 0.0001$; $r^2 = 0.62$).

No *X. glabratus* were captured outside of the infested area in I'on Swamp (Table 1), but six entrance holes were found in the three redbay bolts at that location. Beetles extracted from two of the galleries were species other than *X. glabratus*, and no beetles were found in the other galleries. At two locations on Hilton Head Island, standing dead redbay did not occur on the plots (Table 1) or within visible distance of them, although these areas all had numerous redbay at one time based on the large numbers of stumps and logs of redbay on the ground. At both locations, *X. glabratus* were caught at very low numbers (Table 1). Colonels Island had a total of two dead trees on three survey plots, both <3 cm in diameter, and only 10 beetles were caught. Conversely, at Hunting Island, plots had an average

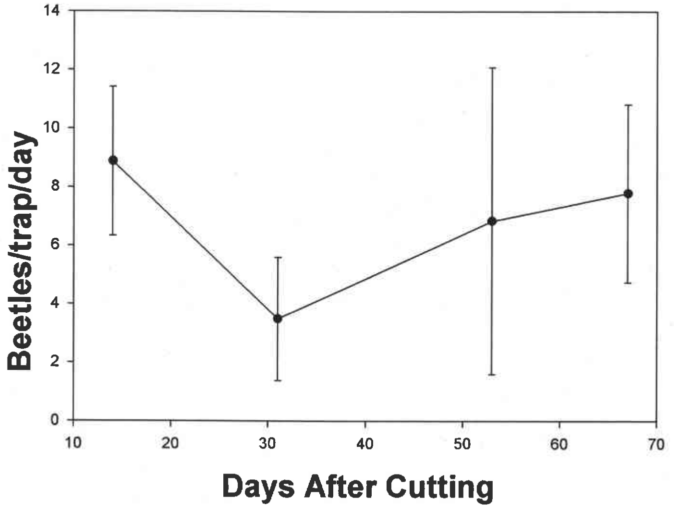

Fig. 4. Mean number (±SE) of *X. glabratus* females/trap/d in traps hanging next to bolts cut from healthy redbay trees in Hunting Island State Park, South Carolina. Traps were deployed from 12 July to 3 September 2006.

of 17 dead trees and a total of 577 beetles were caught on three traps.

Discussion

The number of adult *X. glabratus* increased over the summer, with no indications of distinct generations. This suggests the beetles go through either a single generation per year or, more likely, multiple overlapping generations. Chang (1993) reported a similar pattern of seasonal flight for *Xyleborus* spp. in Hawaii, although with fewer beetles captured in the summer, possibly because of an "orchard cleaning" when dead wood was removed. Conversely, *X. celsus* Eichoff has two attack or flight periods per year in Missouri (Gagne and Kearby 1979), and *X. dispar* (F.) has two in the Pacific Northwest (Chamberlin 1939).

The first male *X. glabratus* was captured on 3 June. After that date, males were recorded in every collection (not from every tree), but they were not included in beetle counts because they were not attracted to the trees. Males are flightless so they most likely fell into traps as they moved about trees. The emergence of the first male was noteworthy because it provides information on the time necessary for *X. glabratus* to complete one generation. Thus far, attempts to obtain this information from log inoculations and beetle rearing under laboratory conditions have not been successful (J.L.H., unpublished data). Trees used in the experiment had no indications of beetle attacks before 24 March when trapping began. If the male beetle was the progeny of one of the first females to arrive at the tree then a maximum developmental time would be ≈56 d. This is consistent with the reported develop time of a similar species, *Xyleborus dispar* (F.), which takes 10–11 wk to develop in Washington state. However, in the laboratory increased temperatures shortened developmental time by as much as 50% (Bhagwantin 1992). Gagne and Kearby (1979) reported *X. celsus* required 35 d to develop from egg to adult. However, the time from first *X. celsus* adult attack on a tree to emergence of their progeny was ≈7–8 wk. This is comparable with the ≈8-wk observed for *X. glabratus* to complete development during spring in coastal South Carolina, although their development is likely to be shorter during the warmer summer.

Wounded diseased and healthy trees were examined on 30 May, and *X. glabratus* attacks were noted. At that time only three trees had evidence of extensive attacks and one had a few attacks. On the remaining four trees little evidence of *X. glabratus* activity was found. Very few beetles were caught in April and May despite warm temperatures along the coast of South Carolina during those months. Large numbers of *X. glabratus* were caught only after the first male was caught, even though large numbers of dead trees with evidence of beetle activity were present throughout the island. The low numbers of attacks evident on trees in late May and the relatively few beetles captured from March through May, suggest that few adult beetles were active during that period. If *X. glabratus* overwintered in all stages, or as adults like *X. dispar* (Bhagwantin 1992) and *X. celsus* (Gagne and Kearby 1979), we would have expected more beetles earlier in the year.

Throughout much of the late spring and summer, trap buckets on standing trees had large amounts of frass and boring dust from *X. glabratus* and *X. crassiusculus*. Boring dust was first noted 3 June and continued until 30 September. On the latter date, all traps were relatively free of boring dust, indicating beetles had stopped attacking the trees. This may have resulted from beetles no longer finding the host material attractive or acceptable for brood production, or beetles emerging late in the year began to disperse. *X. celsus* was able to use hickory (*Carya* spp.) wood for >2 yr for brood production in Missouri (Gagne and Kearby 1979). Because redbay wood is characterized

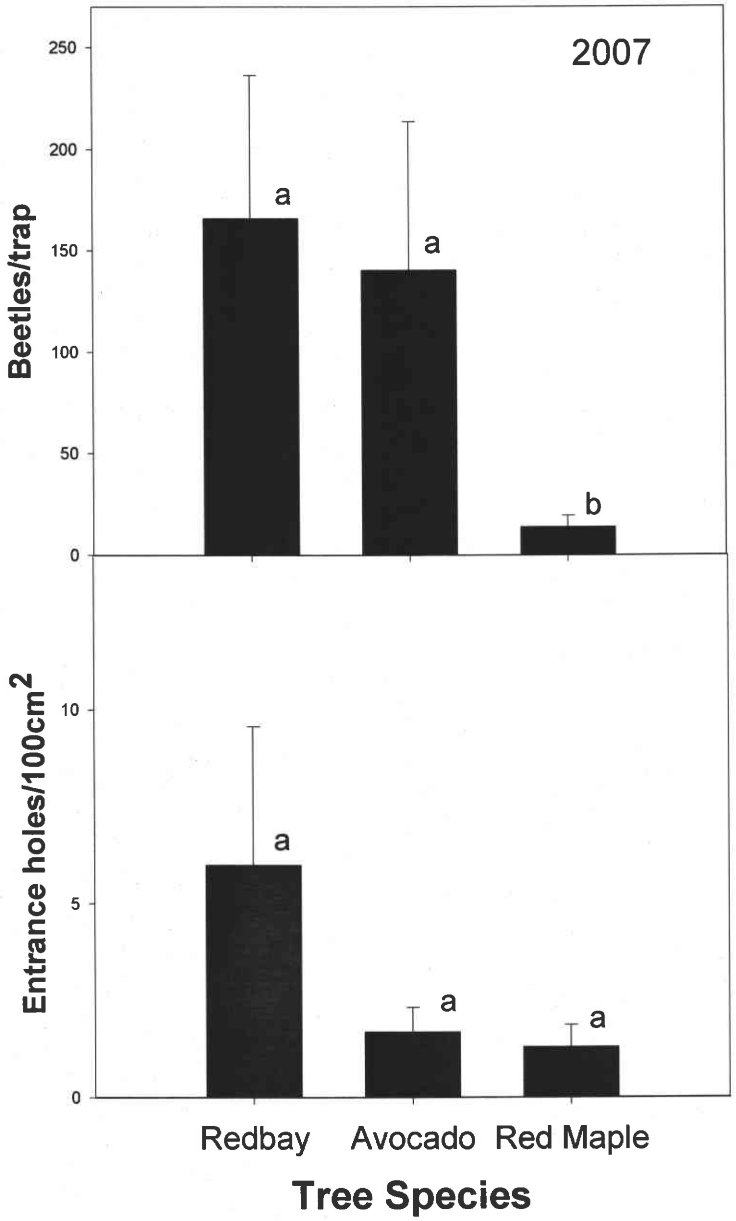

Fig. 5. Mean number (±SE) of *X. glabratus* females per trap captured in traps hanging on bolts cut from of healthy redbay, avocado, or red maple trees and the mean number of beetle entrance holes counted at the end of the study. Traps were deployed at Jennings State Forest, Clay County, FL, from 6 June to 23 August 2007. Columns with the same letter above are not significantly different according to the REGWQ multiple comparison test ($P < 0.05$).

as heavy, hard, and strong (Brendemuehl 1990) it might remain useable for a similar period of time. However, Gagne and Kearby (1979) reported an average of only 61–235 attacks per tree per yr compared with 20–50 attacks per 100 cm^2 in 1 mo for *X. glabratus* in areas of high beetle activity. Therefore, it is possible that the high attack densities for *X. glabratus* in combination with attacks by *X. crassiusculus* resulted in more rapid deterioration of the host wood.

The almost identical numbers of beetles captured on wounded stems of both healthy and diseased redbay trees indicates the fungal pathogen responsible for laurel wilt was not important in beetle host finding success. In addition, although wounding a healthy tree did not significantly increase beetle captures compared with nonwounded redbay trees, the much higher numbers of beetles captured at wounded trees suggests wounding increased attraction to some degree. Likewise, nonwounded healthy redbay trees were not significantly more attractive than nonwounded oak trees. Because at least some beetles were caught throughout our studies in non-

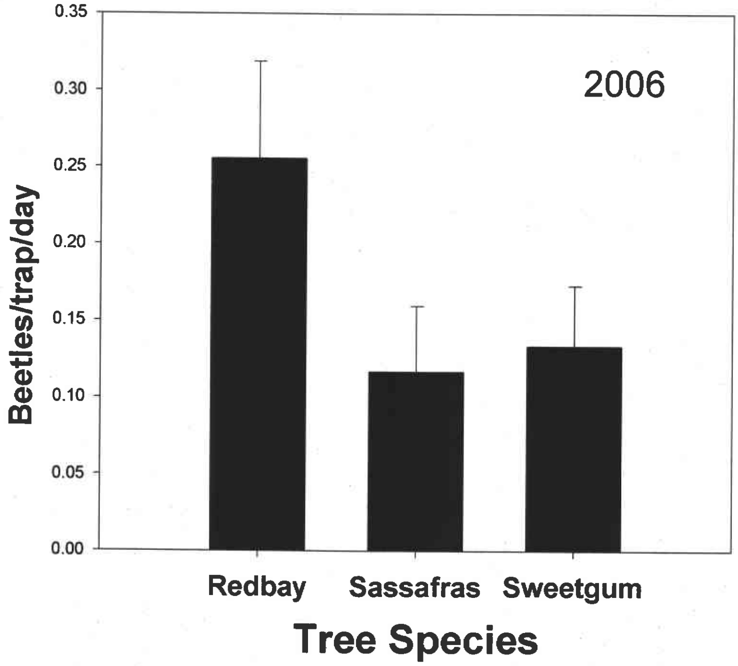

Fig. 6. Mean number (±SE) of *X. glabratus* females per trap per d in traps hanging next to bolts cut from healthy redbay, sassafras, or sweetgum trees. Traps were deployed from 21 September to 28 October 2006 at Hunting Island State Park, South Carolina. Means were not significantly different at $P < 0.05$.

host baited traps regardless of the tree species used to bait them, it is likely that *X. glabratus* randomly land on tree surfaces as they move through a forest. Nonwounded host trees may produce some odor cues resulting in slightly more beetles landing on them but not the amount produced by wounding.

Redbay wood with numerous *X. glabratus* attacks were not more attractive to flying beetles than redbay wood cut from uninfested trees and protected from subsequent attack. These data suggest that *X. glabratus* does not produce an aggregation pheromone and that the extensive frass produced by beetle gallery construction does not contribute to attraction either. It is possible that beetles produce an aggregation pheromone during the early stages of attack when only a few beetles are present. However, no one has reported pheromones in other *Xyleborus* spp. despite being common in species in other ambrosia beetle genera (El-Sayed 2007). In addition, these results confirm our previous results that the fungus causing the wilt disease does not contribute to host attraction. Infested bolts in this experiment were cut from trees with extensive staining in the sapwood and showed symptoms of the wilt disease as well as having large numbers of beetle attacks. However, the number of *X. glabratus* captured at diseased, infested bolts was nearly the same as the number captured at bolts from healthy, uninfected trees. The experiment was conducted during the hottest part of the summer but redbay wood remained attractive for at least 68 d after cutting and the number of beetles capture were similar to trap captures at 14 d after cutting. These results demonstrate that the aromatic compounds in redbay are stable and remain attractive to *X. glabratus* for long periods.

Avocado and swampbay were similar in attraction to *X. glabratus*, and Fraedrich et al. (2008) demonstrated that avocado seedlings were susceptible to the laurel wilt fungus. A mature avocado landscape tree in Jacksonville, FL, also has succumbed to laurel wilt (Mayfield et al. 2008). In 2005, Florida had ≈2,600 ha in avocado production primarily in Dade County (Economic Research Service 2006) located at the southern tip of Florida. Although this area is roughly 100 km from the southernmost *X. glabratus* infestation, redbay is distributed throughout the state (Brendemuehl 1990) making it likely *X. glabratus* and laurel wilt will eventually reach the avocado-producing areas. Because a single beetle attack can transmit the disease pathogen and cause tree mortality, these results combined with those of Fraedrich et al. (2008) indicate *X. glabratus* and laurel wilt may be a threat to avocado orchards in Florida. However, if avocado wood is less suitable for brood development, then the threat may not be as serious. Furthermore, although *X. glabratus* can readily bore in and transmit the laurel wilt fungus when caged onto young avocado plants, susceptibility to the disease seems to vary depending on host cultivar (A.E.M., unpublished data).

Freshly cut sassafras was not attractive to *X. glabratus*, suggesting that the spread of laurel wilt may slow once it reaches the edges of the range of redbay. In addition to the apparent lack of host odor cues, sassafras also may be more difficult for *X. glabratus* to locate in the landscape because it is more widely scattered. However, we recently observed several fencerows in Bullock Co., GA, with sassafras exhibiting signs of laurel wilt and ambrosia beetle attacks. In

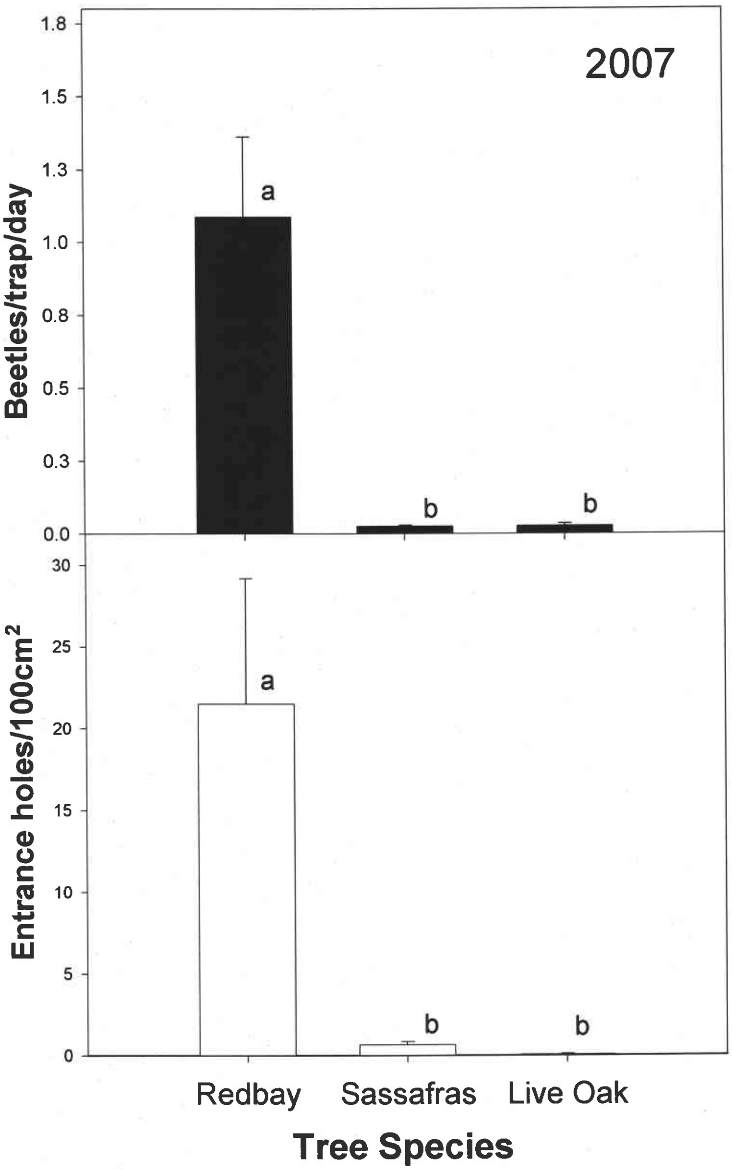

Fig. 7. Mean number (±SE) of *X. glabratus* females per trap per d captured in traps hanging on bolts cut from of healthy redbay, sassafras or live oak trees and the mean number of beetle entrance holes at the end of the study. Traps were deployed at Hunting Island (three replicates) and Lake Warren State Parks (three replicates) in South Carolina from 17 July to 27 August, 2007. Columns with the same letter above are not significantly different according to the REGWQ multiple comparison test ($P < 0.05$).

all cases, we found numerous entrance holes (25–50 holes per 100 cm^2) and live and dead *X. glabratus* adults in the wood. These sassafras populations were in the range of redbay and numerous dead or dying redbay were in the surrounding area, so it is difficult to know whether the large numbers of beetles emerging from redbay resulted in chance encounters with sassafras or whether the beetles were attracted in some way. However, it was clear *X. glabratus* successfully bred in sassafras wood. How *X. glabratus* would behave if accidentally transported into areas where sassafras is more abundant (e.g., the central Appalachians) is not known, but the lack of attractiveness demonstrated in this study is encouraging.

Entrance holes in sassafras and oak were the appropriate size for X. *glabratus*, but other ambrosia beetles occurred in the area and we cannot rule out the possibility the entrance holes were caused by a species other than *X. glabratus*. No attempt was made to extract beetles from the wood directly but bolts were placed into rearing containers. Only one *X. glabratus* and two *Xyleborinus saxeseni* (Ratzeburg) were collected from emergence containers with sassafras. Also, sassafras was not more attractive than live oak (a

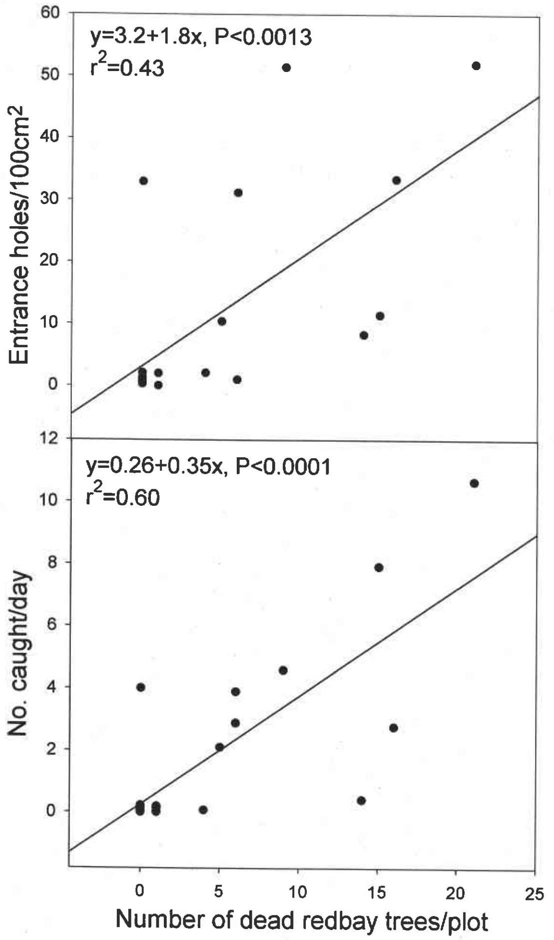

Fig. 8. Linear regression of the number of *X. glabratus* caught in traps and the number of entrance holes in bolts of redbay wood with the number of recently dead redbay trees at eight locations along the coast of South Carolina and Georgia. Three traps were deployed at each location for 27 d from 8 August to 4 September 2007.

nonhost species), and it does not seem to be more attractive than sweetgum (another nonhost).

The reason for the low numbers of *X. glabratus* captured on redbay bolts late in the year in 2006 in the initial sassafras trial is unclear. However, bolts of healthy redbay captured eight beetles per trap per d in July and August (Fig. 3) and similar bolts caught only 0.25 beetles per trap per d in late September and October (Fig. 6). In addition, seasonal trapping on wounded, infested redbay trees caught five to seven beetles per trap per d throughout the same period of the 2006 sassafras and redbay comparison (Fig. 1), so adult beetles were active during the study period. It is possible the *Persea* sp. from the Savannah River site differed from coastal redbay. As mentioned above, swampbay is recognized as a separate species by some authors (e.g., Sargent 1922, Dunkin and Dunkin 1988), but the characters can be difficult to distinguish from *P. borbonia*. However, in the trial comparing swampbay to avocado wood, traps baited with swampbay caught an average of 2.6 beetles per trap per d or 10 times as many as traps on similar-sized bolts of redbay captured late in 2006. Therefore, it seems unlikely differences in species attractiveness caused the low trap captures.

Another possibility is *X. glabratus* undergo a change in behavior late in the season and respond less to host odors. Seasonal changes in ambrosia beetle response to chemical cues are not unprecedented. For example, Stone and Nebeker (2007) caught high numbers of *Xylosandrus mutilatus* (Blandford) in ethanol-baited traps in the spring when attacks on saplings were low (Stone et al. 2007), and very low numbers were caught when attacks were high in the fall. If *X. glabratus* responds differently to host odors at certain times of the year, it could affect efforts to monitor and control this pest.

Redbay was at one time common to Hilton Head Island and Colonels Island, most likely in large numbers similar to Hunting Island; so, it is clear the beetle populations declined greatly at those locations after the redbay trees were killed. However, at least a few beetles are still present on these islands where redbay populations have been decimated. Colonels Island had two small dead trees suitable for brood production on the plots and several other dead trees within visual range of the plots, so some beetles were expected. However, that was not the case at Hilton Head Island where no recently dead redbay trees, and no trees >2.5 cm in diameter were found on the plots or within visible range. This raises questions about the long-term survival of *X. glabratus* in

Table 1. Mean numbers of living and dead redbay trees over 2.5-cm diameter on 0.08-ha plots located along the east coast of the United States from north of Charleston, SC, to the southern border of Georgia

Location	Stage of *X. glabratus* infestation	Mean no. of redbay trees		*X. glabratus*	
		Live	Dead	Mean no./trap/d	RAB size entrance holes/100 cm²
I'on Swamp	Uninfested	37.0 (5.0)	0	0	0.4 (0.45)
Edisto Beach SP	Early	18 (8.4)	2.5 (1.5)	0.1 (0.02)	1.6 (0.46)
Jekyll Island SP	Early	23.0 (11.59)	8.3 (2.85)	1.8 (0.73)	6.6 (2.84)
Hunting Island SP	Heavy	3.5 (0.5)	17.3 (1.86)	7.1 (2.31)	32.3 (11.72)
Richmond Hills	Heavy	1.0 (0.47)	5.0 (2.65)	4.1 (0.22)	38.6 (6.48)
New Hall Preserve, Hilton Head Island	Late	0.7 (0.27)	0	0.04 (0.02)	0.6 (0.33)
North Ridge Tract, Hilton Head Island	Late	0.3 (0.27)	0	0.1 (0.06)	1.1 (0.53)
Colonels Island	Late	0.3 (0.27)	0.67 (0.33)	0.1 (0.04)	0.4 (0.45)

Plots varied in age of *X. glabratus* infestation. *X. glabratus* (RAB) populations were measured by number of beetles caught per day on sticky traps attached to redbay bolts from 8 August to 4 September 2007 and by the number of *X. glabratus*-sized entrance holes in the bolts at the end of the study.

the absence of redbay trees. Hilton Head had numerous dead and dying trees in 2004 and 2005 and probably into 2006, so it is likely that some of the wood either in stumps or lying on the ground was sufficiently sound to support brood production and/or the beetles were attracted from relatively long distances.

X. glabratus and laurel wilt are a serious threat to redbay trees throughout their range but results of the current study provide some hope for redbay and other members of the Lauraceae in the eastern United States. First, *X. glabratus* was not attracted to sassafras in these studies. Although sassafras have been killed by laurel wilt in areas where there also were high populations of redbay for beetles to breed in, once the beetle has reached the edge of the range of redbay its spread may slow if beetle host finding success is limited by lack of attractants in sassafras. Second, once mature redbay trees in an area have been killed, beetle populations dropped to very low levels, suggesting they were unable to breed effectively in other types of wood. Thus, with the availability of new lures (J.L.H., unpublished data), it may be possible to use trapping or attract and kill techniques to suppress the already low beetle populations and allow redbay seedlings and stump sprouts to reach maturity. This may be particularly useful on barrier islands that are somewhat isolated from the mainland. Although *X. glabratus* and laurel wilt may not spread quickly through the range of sassafras, they will likely be a threat to avocado trees in Florida, at least when infestations reach avocado-producing areas. However, as the mature redbay trees die and decompose in such areas, beetle populations should decline. If control measures such as intensive sanitation and insecticide treatments are feasible in production fruit orchards, *X. glabratus* and laurel wilt may not be a serious threat to them over the long term.

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References Cited

Bhagwandin, H. O., Jr. 1992. The shothole borer: an ambrosia beetle of concern for chestnut orcharding in the Pacific Northwest, pp. 168–177. 93rd Annual Report of the Northern Nut Growers' Association. (www.wcga-.net/shb.htm).

Brendemuehl, R. H. 1990. *Persea borbonia* (L.) Spreng. Redbay, pp. 503–510. *In* R. M. Burns and B. H. Honkala [tech. coord.], Silvics of North America, vol. 2, Hardwoods. Agric. Handb. 654. U.S. Dep. Agric.–Forest Service, Washington, DC.

Brown, C. L., and L. K. Kirkman. 1990. Trees of Georgia and adjacent states. Timber Press, Portland, OR.

Chamberlin, W. J. 1939. The bark and timber beetles of North America. Oregon State Coop Association, Corvallis, OR.

Chang, V.C.S. 1993. Macadamia quick decline and *Xyleborus* beetles (Coleoptera: Scolytidae). Int. J. Pest Manag. 39: 144–148.

Day, R. W., and G. P. Quinn. 1989. Comparison of treatments after an analysis of variance in ecology. Ecol. Monogr. 59: 433–463.

Dunkin, W. H., and M. B. Dunkin. 1988. Trees of the southeastern United States. The University of Georgia Press, Athens, GA.

Economic Research Service. 2006. Commodity highlight: avocados. *In* Fruit and tree nuts outlook. FTS-321. U.S. Dep. Agric.–Economic Research Service, Washington, DC.

El-Sayed, A. M. 2007. The pherobase: database of insect pheromones and semiochemicals. (http://www.pherobase.net).

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 USA. Plant Dis. 92: 215–224.

Gagne, J. A., and W. H. Kearby. 1979. Life history, development, and insect-host relationships of *Xleborus celsus* (Coleoptera: Scolytidae). Can. Entomol. 111: 295–304.

Griggs, M. M. 1990. *Sassafras albidium* (Nutt.) Nees, Sassafras, pp. 773–777. *In* R. M. Burns and B. H. Honkala [tech. coord.], Silvics of North America, vol. 2, Hardwoods. Agric. Handb. 654. U.S. Dep. Agric.–Forest Service, Washington, DC.

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

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.

Sargent, C. S. 1922. Manual of the Trees of North America. Dover Publications, Inc., New York.

SAS Institute. 1985. SAS guide for personal computers, version 6th ed. SAS Institute, Cary, NC.

Stone, W. D., and T. E. Nebeker. 2007. Distribution and seasonal abundance of *Xylosandrus mutilatus* (Coleoptera: Curculionidae). J. Entomol. Sci. 42: 409–412.

Stone, W. D., T. E. Nebeker, and P. D. Gerard. 2007. Host plants of *Xylosandrus mutilatus* in Mississippi. Fla. Entomol. 90: 191–195.

Ulyshen, M. D., and J. L. Hanula. 2007. A comparison of the beetle (Coleoptera) Fauna captured at two heights above the ground in a North American temperate deciduous forest. Am. Midl. Nat. 158: 260–278.

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