An *Ophiostoma* species and *Xyleborus glabratus* Threaten Red Bay (*Persea borbonia*) and Other Members of the Lauraceae in the Southeastern USA.

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BACKGROUND INFORMATION

Extensive mortality of red bay (*Persea borbonia* (L.) Spreng) has been observed in the coastal plains of South Carolina and Georgia since 2003. The disease has also been observed around Jacksonville, FL since the spring of 2005. The current range of the disease problem is presented in Figure 1.

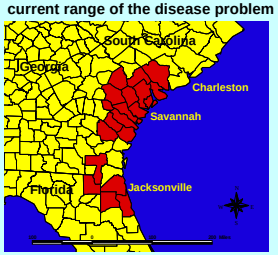

Figure 1. Range of red bay wilt, as of May 2006, in Georgia, South Carolina and Florida.

Dead and dying trees exhibit wilt-like symptoms (Figure 2). Trees often decline very rapidly with the wilt affecting the entire crown uniformly. In other cases, tree decline progresses more slowly affecting individual branches one at a time. The sapwood of the main stem and branches of affected trees exhibits discoloration (Figure 3). Small beetle entrance holes and tunnels are normally found in association with discolored areas of the sapwood. On some symptomatic trees the entrance holes have been rare and difficult to locate. On other trees, entrance holes are numerous.

Figure 2. Wilted red bay at Fort George Island near Jacksonville, FL (October, 2005)

Figure 3. Sapwood discoloration in a dying red bay tree near Colonels Island, GA (May, 2005).

An ambrosia beetle, *Xyleborus glabratus (*Eichhoff), has been routinely obtained from dead and dying trees. This non-native beetle was first trapped in the United States in 2002 at a port facility near Savannah GA. The recovery of the beetle from dead and dying red bay on Hilton Head Island (SC) in November, 2004 was the first indication that the beetle was established in forests of the southeastern USA. The beetle is native to Asia (e.g. India, Japan) where it is associated with plant species in the family Lauraceae (e. g. *Lindera latifolia, Litsea elongata).*

A fungus has been consistently isolated from the discolored xylem of symptomatic trees throughout the range of the problem. This fungus has been identified as an *Ophiostoma* sp. based on sequences of the ribosomal DNA and its tolerance of cycloheximide. The anamorph of the fungus is similar to species of ambrosia beetle symbionts in the genus *Raffaelea.*

During the spring and summer of 2005, sassafras mortality was also observed at several locations in coastal counties of Georgia. An examination of the trees found sapwood discoloration similar to that observed in red bay. *Xyleborus glabratus* and the *Ophiostoma* sp. previously isolated from wilted red bay were also associated with the wilted sassafras.

PATHOGENICITY TESTS

The pathogenicity of isolates of the *Ophiostoma* sp. to red bay was evaluated in growth chamber and field tests. The field test was conducted in March, 2005 near Bluffton, SC. Twenty-four red bay trees (heights, 1.8 to 4.9 m) were selected and paired. All trees were wounded (i.e. drilled holes). One tree of each pair was inoculated with an isolate of the *Ophiostoma* sp.; the other tree was treated as a control. Trees were evaluated 10 weeks after inoculation. All trees inoculated with the *Ophiostoma* sp. wilted and exhibited sapwood discoloration (Figure 4 A, B). Three of the 12 control trees also wilted but these trees had beetle entrance holes characteristic of *X. glabratus* as well as sapwood discoloration. The other nine control trees were healthy with no evidence of sapwood discoloration or entrance holes. Pieces of sapwood from all trees were placed on cyclohexamide-streptomycin malt agar (CSMA) medium, and the imperfect stage of the *Ophiostoma* sp. was isolated from discolored sapwood of all wilted control and fungus-inoculated trees.

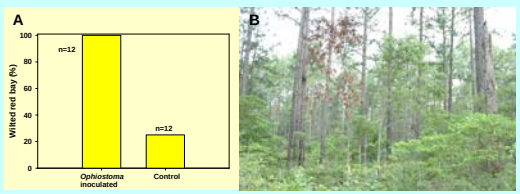

Figure 4. A) Percentage of wilted red bay trees following inoculation with isolates of an *Ophiostoma* sp.; B) Wilted, *Ophiostoma*-inoculated red bay (left) and healthy, control tree (right).

PATHOGENICITY TESTS, continued

In the growth chamber test, plants were wounded and inoculated with conidial suspensions of *Ophiostoma* isolates. Control plants were wounded and drops of sterile water were placed at the wound site. Plants were placed in a growth chamber (16 h photoperiod; temperatures, 25 to 28°C). Eight weeks after inoculation 19 of 20 plants inoculated with the *Ophiostoma* sp. exhibited symptoms of the wilt (Figs. 5 A, B). All wilted plants had sapwood discoloration, and the *Ophiostoma* sp. was isolated from all symptomatic plants. Control plants remained healthy and exhibited no symptoms of disease.

Figure 5. A) Percentage of wilted red bay plants following inoculation with isolates of the *Ophiostoma* sp.; B) Control plant (left) and wilted, *Ophiostoma*-inoculated red bay plant (right).

RED BAY PLANTS CHALLENGED WITH *XYLEBORUS GLABRATUS*

The objective of this experiment was to determine if *X. glabratus* beetles could attack healthy red bay seedlings. Beetles were obtained from infested red bay stems, placed in gel caps, and the caps were attached to the stems of the plants (Figure 6A). Most beetles bored into the seedlings within 48 hours (Figure 6B). The seedlings were observed over the course of the next eight weeks. Sixteen of the 22 seedlings, attacked by *X. glabratus,* wilted and exhibited sapwood discoloration (Figure 7A, B) The *Ophiostoma* sp. was isolated from all symptomatic plants as well as from five of the nonsymptomatic seedlings. The fungus was isolated only around beetle tunnels in the nonsymptomatic plants.

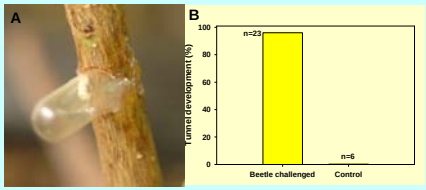

Figure 6. A) Gel cap with beetle attached to red bay plant (note wood boring dust in cap); B) Percentage of red bay plants with tunnel development after being challenged with *X. glabratus.*

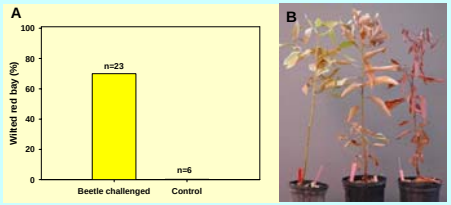

Figure 7. A) Percentage wilted red bay plants following colonization by *X. glabratus*; B) Symptomatic red bay plants attacked by *X. glabratus* and subsequently infected with the *Ophiostoma* sp.

SUSCEPTIBILITY OF OTHER SPECIES IN THE LAURACEAE

The susceptibility of sassafras (*Sassafras albidum*), spicebush (*Lindera benzoin*) and swamp red bay (*P. palustris*) to wilt caused by the *Ophiostoma* sp. was evaluated. Red bay plants were also inoculated for comparison, and all plants were grown in 1 or 3 gallon pots. Ten plants of each species were wounded and inoculated with conidial suspensions of isolates of the *Ophiostoma* sp. Five control plants of each species were also wounded and drops of sterile water were placed at wound sites. Plants were incubated in a growth chamber (16 h photoperiod; temperatures, 25 to 28°C).

Spicebush, sassafras, swamp red bay as well as red bay wilted following inoculation with isolates of the *Ophiostoma* sp. (Figures 8A, B). Wilted plants of all species had sapwood discoloration. In spicebush, leaf chlorosis and wilting was evident in some plants within 5 days after inoculation, and most plants were completely wilted within 3 weeks. In sassafras, leaves on inoculated plants turned red and readily abscised from stems. In red bay and swamp red bay, new shoots at branch terminals wilted. More mature leaves developed a reddish to purplish brown discoloration and dead leaves were persistent on stems. All red bay and swamp red bay plants died within 5 weeks after inoculation. The *Ophiostoma* sp. was isolated on CSMA medium from the discolored sapwood of all wilted plants.

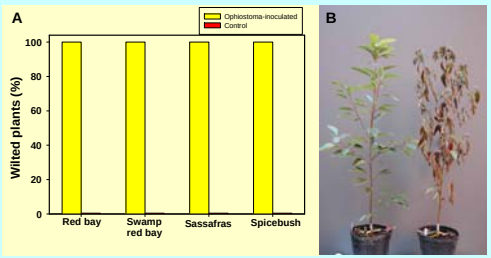

Figure 8. A) Percentage wilted red bay, swamp red bay, sassafras and spicebush plants following inoculation with isolates of an *Ophiostoma* sp.; B) Control (left) and *Ophiostoma*-inoculated (right) sassafras plants after five weeks.

SUMMARY AND CONCLUSIONS

Xyleborus glabratus and an *Ophiostoma* sp. are associated with extensive mortality of red bay in the southeastern USA. *Xyleborus glabratus* is capable of attacking healthy red bay plants, and the *Ophiostoma* sp. causes a rapid wilt in red bay as well as other members of the Lauraceae that are common in forests of the Eastern USA.

Based on the results of these studies, and the association of *X. glabratus* with species in the family Lauraceae, there is reason to be concerned that the wilt associated with red bay could affect other members of this family. Recent observations of the wilt affecting sassafras in Georgia provides support for this concern. Additional species in the Lauraceae in the Southeast include pondberry (*Lindera melissaefolium*) and pondspice (*Litsea aestivalis*) which are endangered or threatened plants. Various plant species in the Lauraceae are common components of forests in other regions of the United States and other countries in the Western Hemisphere.