

# USDA-ARS Strategies to Address the Imminent Threat of Redbay Ambrosia Beetle and Laurel Wilt Disease to Florida Avocados

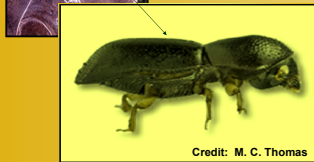
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The Avocado Industries in the U.S. and Mexico – as well as the USDA avocado germplasm collection in Miami – are in danger of collapse due to laurel wilt. This deadly disease of avocado and other trees in the Laurel family (*Lauraceae*) is caused by a mycopathogen (*Raffaelea lauricola*) vectored by an exotic wood-boring insect, the redbay ambrosia beetle (*Xyleborus glabratus* Eichhoff). Impact of this pathogen/pest complex is potentially devastating. Once infected with *Raffaelea*, tree mortality can occur in as little as 6 weeks. First detected in the U.S. near Savannah, GA in 2002, the beetle and its obligate fungal symbiont have spread along the coast into South Carolina and Florida. Recently, laurel wilt has been confirmed in Martin County, ~70 miles north of Florida's commercial avocado groves, estimated to be worth about \$13 million annually.

*Xyleborus glabratus* (Curculionidae: Scolytinae) is a cylindrical beetle about 2 mm long. Males are flightless and remain within the host tree. Mated females disperse and bore into new hosts, forming galleries where eggs are laid. The galleries are inoculated with *Raffaelea* spores (carried in mandibular mycangia), and the fungal growth ("ambrosia") provides food for larvae and adults. The fungus continues to spread throughout the host's vascular system, blocking water transport. This results in laurel wilt disease and ultimately tree death (Mayfield & Thomas 2006, Fraedrich et al. 2008, Harrington et al. 2008).

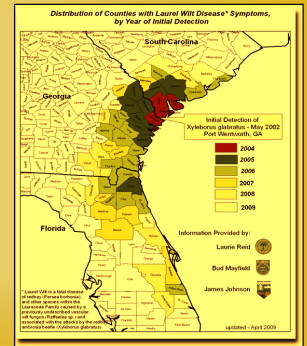


Credit: M. C. Thomas

Preliminary research provides no evidence of an aggregation pheromone nor attraction to the fungal pathogen (Hanula et al. 2008). Primary attractants appear to be the host tree volatiles. Identification of those semiochemicals is critical for the development of effective monitoring and management tools for this invasive pest.



**U.S. Hosts:** To date, laurel wilt pathogen has been isolated from redbay (*Persea borbonia*), avocado (*P. americana*), silkbay (*P. borbonia humilis*), swampbay (*P. palustris*), sassafras (*S. albidum*), pondberry (*Lindera melissifolia*), camphor tree (*Cinnamomum camphora*), and pondspice (*Litsea aestivalis*).



Also confirmed in Martin County - 30 June 2009

## Chemistry / Entomology Projects

### Surveys for Redbay Ambrosia Beetle & Laurel Wilt:

SHRS entomologists have participated in Florida county surveys coordinated by the USDA-APHIS-PPQ-CAPS and the FDACS-DPI-CAPS programs. This provided a working knowledge of standardized survey protocols and specimen collection techniques. The current monitoring system consists of Lindgren funnel traps baited with manuka oil lures hung in host trees. LowTox antifreeze is added to the collection cups to retain and preserve captured beetles.



### Evaluation of Current Attractant Systems:

Manuka oil and phoebe oil are the best attractive baits known for *X. glabratus* (Hanula and Sullivan 2008). These essential oil extracts were originally developed as attractants for the emerald ash borer, *Agrilus planipennis* (Buprestidae). Research is needed to evaluate efficacy for *X. glabratus*, and determine the release rates and longevity of commercial lure formulations.



### Development of Biologically-Based Management Systems:

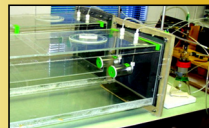
Research has been initiated to identify key semiochemicals from host trees (avocado, redbay) that can be exploited for development of specific field lures for *X. glabratus*. Effective synthetic lures are critically needed to improve pest detection and delimitation, and to develop biologically-based management approaches using attract-and-kill systems.

**Chemical Analysis.** To identify semiochemicals of interest, headspace volatiles from preparations of host tree leaves, bark, sapwood, and hardwood layers were collected using solid-phase microextractions (SPME) and injected into a Trace™ Gas Chromatograph and into a 5975B GC / Mass Spectrometer.



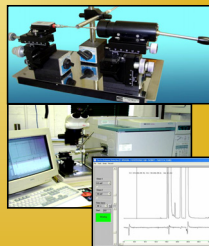
### Bioassays and Field Tests:

Once putative attractant chemicals have been identified, bioassays and field trapping tests will be initiated. Effective bioassay methods need to be developed, and various options will be considered, including laboratory two-choice olfactometer and flight tunnel tests, as well as field cage and field trapping tests. Since the beetle has not reached SHRS in Miami, experiments will be performed in collaboration with scientists at the USDA and University of Florida facilities in Gainesville, FL.



### Electrophysiology:

Electroantennographic (EAG) techniques will be explored to determine if antennal olfactory response can be quantified from *X. glabratus*. If a method can be developed, EAG will be used to evaluate putative attractants to determine dose ranges and potential combinations of attractants. In addition, EAG coupled with gas chromatography (GC-EAG) may be used to evaluate complex mixtures of host plant volatiles. With this approach, GC can separate the component compounds and EAG can identify the physiologically active peaks.



**Establishment of a Laboratory Colony:** University of Florida entomologists have taken steps to establish a colony of redbay ambrosia beetles in Gainesville, FL. This colony would greatly facilitate behavioral bioassays and electrophysiology work.

## Plant Science / Genetics Projects

### Protection of USDA Avocado Germplasm Collection at SHRS:

The Subtropical Horticulture Research Station (SHRS) in Miami, FL maintains ~400 avocado accessions. To ensure genetic diversity of avocado stock for future breeding, representative accessions will be treated by macro infusion with the fungicide Propiconazole (Alamo®). Injection into the tree's root flare in advance of the arrival of the *Raffaelea* pathogen will confer protection from laurel wilt disease. Treatment will be very costly, and longevity needs to be determined.



### Establishment of a Backup Germplasm Collection:

Before the arrival of laurel wilt in Miami-Dade County, the USDA avocado germplasm collection at SHRS will be backed-up at sites in Hawaii and California, in collaboration with university and ARS cooperators. All accessions will be grafted onto rootstock and maintained under quarantine conditions prior to transport, ensuring that plant material is free of pests and pathogens.

### Development of Resistant / Tolerant Genotypes:

University of Florida studies indicate that avocado cultivars may vary in susceptibility to the *Raffaelea* fungus. Phenotypic and molecular analysis will be conducted on the SHRS avocado collection to identify associations between molecular markers and tolerance/resistance to laurel wilt. Once markers have been identified, genotypes with potential for resistance will be propagated. These trees will then undergo further testing and screening using microsatellite and SNP markers.



### REFERENCES

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