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# Helping Citrus Growers Deal With a Nasty Invader

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# Helping Citrus Growers Deal With a Nasty Invader

Growers battling a devastating citrus disease may soon benefit from a one-two punch from Florida researchers. Two Agricultural Research Service scientists at the U.S. Horticultural Research Laboratory in Fort Pierce are using two very different strategies to help citrus growers and processors deal with Huanglongbing (HLB), a disease that poses a major threat to the survival of the citrus industry and is costing it millions of dollars each year.

Elizabeth Baldwin, research leader of the Citrus and Subtropical Products Research Unit, is leading a team developing technology that orange juice processors can use to determine whether their product has the right taste. David Hall, research leader of the Subtropical Insects Research Unit, has found a way to better exploit a fungus that naturally controls HLB.

HLB, or citrus greening, was first detected in Florida in 2005 and is now established in all citrus-growing counties in the state. The disease was confirmed soon afterwards in backyard citrus trees in Georgia, Louisiana, and South Carolina. Citrus greening was recently confirmed in a commercial grove in Texas and in a single residential tree in California in 2012. A 2012 study by the University of Florida Institute of Food and Agricultural Sciences estimates that HLB has cost the state's citrus industry $3.6 billion since 2006. The disease also occurs in tropical and subtropical Asia, India, Africa, and South America.

STEPHEN AUSMUS (D2703-18)

In an orange grove heavily infected with citrus greening disease, entomologists Matt Hentz (left) and David Hall inspect trees for Asian citrus psyllids that have been killed by the beneficial fungus *Hirsutella citriformis*.

The presumed cause of HLB is the bacterium *Candidatus* Liberibacter asiaticus, and it is carried from plant to plant by the Asian citrus psyllid *(Diaphorina citri),* a tiny, winged insect that feeds on all citrus relatives. When a tree is infected, the bacterium moves into its phloem tissues, blocking the passage of nutrients through the vascular system until it makes the plant unproductive. Infected trees usually decline in productivity and die within 5 to 10 years of infection. Fruit on infected trees often falls to the ground before harvest, and those that remain on trees may become misshapen, sometimes only partially ripening in color. Juice from symptomatic fruit usually has an acidic, bitter taste.

The best strategy to control HLB is to remove infected trees (the source of the bacterium for the psyllid) and control the psyllid vector. But many Florida growers are reluctant to pull trees if they are still productive. To help citrus growers and juice processors, Baldwin and her team are investigating the effects of HLB on the taste of orange juice produced from diseased trees. Their goal is to maintain the high-quality product that consumers want and expect while scientists search

STEPHEN AUSMUS (D2705-10)

Horticulturalist Elizabeth Baldwin (left) and chemist Jinhe Bai review data from the “electronic tongue,” a device that can detect chemical differences related to taste to see whether it can distinguish between juice from HLB-infected oranges and from noninfected oranges.

for a permanent solution to the threat posed by HLB.

As the disease continues to spread, the quality of juice from diseased trees is becoming more of a concern to growers, particularly in Florida, where 90 percent of the oranges raised are used primarily for juice. In general, if fruit from a diseased tree is asymptomatic and looks normal, the juice will taste normal, but if it is symptomatic (smaller, greener, and lopsided), the juice flavor may be affected, depending on the number of symptomatic fruit in a batch of juice.

“There is no specific point where fruit becomes so symptomatic that it affects the taste. It’s more of a spectrum or range, so the juice processors need an objective way of analyzing fruit for taste so they can make an informed decision about what they’re buying,” Baldwin says.

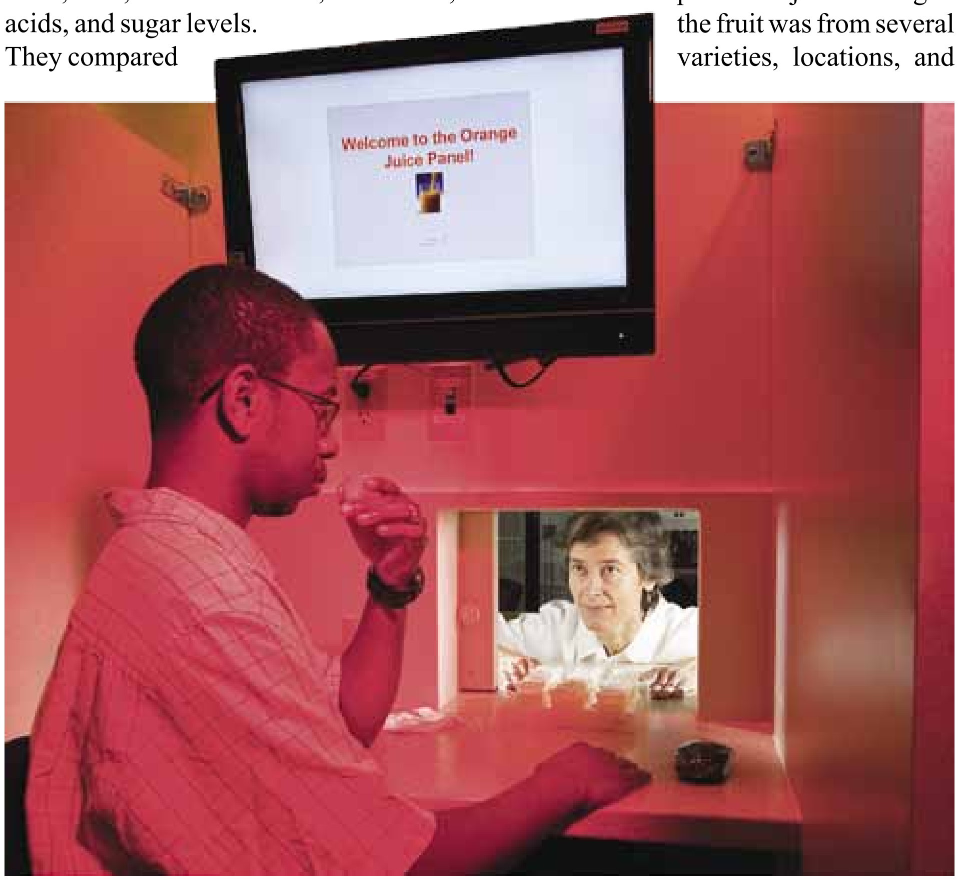
Baldwin and her team are working on two fronts: They are investigating the effects of the disease on the taste and quality of juice and developing new technology that will help processors objectively determine the taste qualities in their juice.

**Studying Disease Effects on Taste**

The researchers evaluated both asymptomatic and symptomatic fruit produced over two growing seasons for several fruit and juice characteristics, including color, size, aroma volatiles, limonoids, acids, and sugar levels. They compared Midsweet, Hamlin, and Valencia oranges, the three principal varieties harvested for processing, and used gas and liquid chromatography to analyze compounds in fruit that went into the juice.

They found that juice from symptomatic fruits was more often higher in limonin and nomilin, compounds that can give orange juice a bitter taste, but that the compounds were generally detected at amounts below threshold levels in taste panels. Results were published in the *Journal of Agricultural and Food Chemistry* in 2010.

In another study, the researchers investigated how HLB infection affects juice quality with respect to cultivar, maturity, and processing method. They compared the same three varieties of infected and disease-free fruit. The results showed tremendous variability, depending on the harvest date and variety of orange. In general, the researchers found more of a problem with off-flavored juice from diseased Hamlin oranges than with diseased Valencia and Midsweet varieties, and off-flavors were more prevalent earlier in the harvest season. But they concluded that use of some symptomatic fruit would not adversely affect the flavor of commercially processed juice as long as the fruit was from several varieties, locations, and




STEPHEN AUSMUS (D2706-5)

To see whether off-flavors are perceived in juice from HLB-infected oranges, plant physiologist Anne Plotto hands juice samples to taste-tester Keith Williamson. The red lighting in the tasting booth helps him focus only on taste and smell.

DAVID HALL (D2713-1)

Adult Asian citrus psyllid killed by the fungus *Hirsutella citriformis*. A psyllid cadaver, seen here, can remain on a citrus leaf for extended periods and continue to spread the biocontrol fungus to other psyllids.

seasons and the mixture contained juice from healthy fruit.

Results were published in the *Journal of Food Science* in 2010.

Baldwin has been awarded a 2012 cooperative research and development agreement with Southern Gardens Citrus (a subsidiary of the U.S. Sugar Corporation), which grows oranges and processes them into juice, to develop an instrument that processors can use to measure flavor compounds and detect off-flavors in juice. The technology could be designed so that juice is tested in bulk or on an assembly line at the processing plant. From the way the disease is progressing in Florida, Baldwin estimates that the technology will be needed within the next few years to ensure a consistent, delicious, and high-quality product.

### Tapping the Effects of a Beneficial Fungus

Many citrus growers apply chemicals to control plant pests (insects and mites) and plant pathogens. Hall has found that some of these inhibit proliferation of a beneficial fungus, *Hirsutella citriformis,* which has the ability to naturally control the Asian citrus psyllid.

Soon after HLB was discovered in Florida in 2005, *H. citriformis* was seen as having a kind of double-barreled impact. It killed Asian citrus psyllids, and the dead psyllids remained on the citrus leaves for extended periods, spreading the fungus to other psyllids.

But field observations in Florida's citrus groves turned up a puzzling pattern, says Hall. "We were seeing fewer cadavers one year than in the next year, and we asked ourselves what could be causing that. Were there environmental factors, such as the humidity levels, affecting these fungal populations? Or were the sprays and chemical treatments applied to citrus groves reducing the numbers?"

Hall and his colleagues methodically counted psyllid cadavers and live adults on mature leaves of 30 trees, randomly selected each week, for 118 weeks from January 2006 to April 2008 in an orange grove near Vero Beach. Hall's partners included Drion Boucias from the University of Florida and Jason Meyer from Purdue University. As is routine in many commercial operations, the grove was periodically sprayed with petroleum oil and copper hydroxide to control greasy spot, a fungal disease that defoliates citrus. The researchers tagged some leaves where they found psyllid cadavers so they could assess how long they remained on leaves. They also measured rainfall, air temperatures, and relative humidity to see if weather patterns played a role in cadaver and psyllid numbers.

The results showed that the cadavers continued to spread the beneficial fungus on citrus leaves for an average of 68 days and that sometimes there were more cadavers than live adults on leaves. They also found that cadavers were most abundant in the fall and winter months and that cadaver numbers were higher in 2006, a year when no copper hydroxide was sprayed.

The researchers then conducted laboratory experiments to evaluate the effects on *H. citriformis* of six chemical compounds commonly sprayed to control greasy spot and other citrus diseases in Florida citrus groves. In collaboration with insect pathologists at the University of Florida, the team successfully cultured the fungus on media in petri dishes. Five-week-old cultures were sprayed with the compounds. Three of them—copper hydroxide, petroleum oil, and elemental sulfur—reduced the fungus's ability to infect the Asian citrus psyllid. The other three—copper sulfate pentahydrate, aluminum tris, and alpha-keto/humic acid—did not slow it down.

The study, published in March 2012 in *BioControl,* is the first investigation into the field dynamics of a fungal pathogen that attacks the Asian citrus psyllid, Hall says. It opens the door to more widespread use of *H. citriformis* in HLB management programs.

"Knowing that *H. citriformis* is naturally occurring and that it confers this benefit may encourage growers to capitalize on it by using alternatives to some of their current treatments," Hall says.

Because the researchers detailed how they cultured *H. citriformis* in the paper published in *BioControl,* others can expand on the work by developing strategies that use the fungus to control the psyllid in commercial citrus operations. For example, citrus could be sprayed with a water solution containing fungal spores to supplement existing populations. The fungus may be of particular interest to organic producers, who need alternatives to traditional insecticides, and to growers in developing countries less likely to spend money on traditional sprays.

"Now that we know how to culture this beneficial fungus, it becomes a potential weapon that could be more widely deployed in our war with this nasty citrus pest," Hall says.—By **Dennis O'Brien,** ARS.

*This research is part of Quality and Utilization of Agricultural Products (#306) and Crop Protection and Quarantine (#304), two ARS national programs described at www.nps.ars.usda.gov.*

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