Asian citrus psyllid and huanglongbing disease: A threat to California citrus

In 2008, the tiny, aphid-sized Asian citrus psyllid (ACP) was first identified in California. ACP injects a toxin when it feeds on citrus leaves or stems, causing shoot deformation and plant stunting. But this damage isn't the growers' greatest concern. ACP is a vector of the bacterium associated with huanglongbing disease (HLB), the most serious citrus disease in the world. HLB causes leaves to yellow and fruit to become small, misshapen, and develop a bitter taste.

There is no cure for the disease, and trees infected with the HLB pathogen eventually die, sometimes in as little as three years. In March 2012, HLB was detected in a residential citrus tree in Los Angeles County. That tree was destroyed, but it is likely there are more infected trees in California. The disease is also spreading northward from Mexico toward California.

ACP and HLB together present a grave threat to California's $2.1 billion citrus industry, the livelihood of citrus farmers and thousands of farmworkers, and the fragile economies in California's rural citrus belt. The presence of ACP and HLB prevents exports to countries that do not have this pest and disease. Loss of citrus trees in urban areas of California will change the face of the landscape and reduce local fruit availability. UC is working with the citrus industry to wage an all-out battle against both the pest and disease. Much of the research is conducted with funding from the citrus industry through the Citrus Research Board. Other funding sources are UC Agriculture and Natural Resources, CDFA (Specialty Crops Block Grants), and the USDA-NIFA (National Institute of Food and Agriculture).

UC is approaching the ACP-HLB threat from five angles:

1. Ensure that citrus trees start out HLB-free.
2. Reduce ACP populations.
3. Detect HLB-infected trees so they can be removed as quickly as possible.
4. Find a long-term cure.
5. Engage the public and enlist their help in fighting ACP and HLB.

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ACP and ants on newly expanded citrus leaves.

1 – *Ensuring clean plant material*

The state of California has strict regulations and methods in place to ensure that citrus trees are tested for pathogens to verify that they are free of disease before they are sold. However, the general public does not always understand the importance of these regulations, and people sometimes unknowingly bring diseased plant material (citrus and other hosts of ACP-HLB) into California and graft their own trees.

Citrus Clonal Protection Program

The UC Citrus Clonal Protection Program (CCPP), housed at UC Riverside, is the gatekeeper of California citrus. CCPP is one of the three programs in the nation authorized to import citrus budwood from overseas and is charged by the state to conduct disease diagnosis, pathogen elimination, and the distribution of true-to-type, clean citrus propagative material of fruit and rootstock varieties to nurseries and private individuals. The CCPP is directed by Georgios Vidalakis, UC Cooperative Extension specialist in the Department of Plant Pathology and Microbiology at UC Riverside.

"Unforbidden" fruits

Scientists at UC Riverside are developing a legal source of plant material for popular non-citrus ACP hosts, such as bael tree, a native food plant of India also used for traditional medicine, and Indian curry leaf, a food flavoring. The program—run by Tracy Kahn, principal museum scientist, and David Karp, associate in the Agricultural Experiment Station, in the UC Riverside Department of Botany and Plant Sciences—will provide clients with clean plants, reducing the incentive for smuggling plants and plant material into California that potentially harbor ACP or HLB. Kahn has also joined forces with the U.S. Department of Agriculture to assemble the only collection in California that includes all the major host plants of ACP and HLB—all varieties of citrus and closely related plants in the family Rutaceae.

2 – *Managing the psyllid*

ACP is currently found only in Southern California. The majority of commercial citrus is grown in Central California. If ACP can be prevented from spreading, it minimizes quarantine and export issues and reduces the threat to Central Valley citrus production. If psyllid populations are kept low wherever they are found, their chances of picking up the HLB pathogen are reduced and disease spread is slowed.

Citrus database for rapid pest response

The geographical information systems team at the UC Kearney Agricultural Research and Extension Center in Parlier, in collaboration with the Citrus Pest and Disease Prevention Program, is developing a geographic database that will provide treatment coordinators with information to ensure quick action when psyllids or HLB are found. The database is enriched with details about the citrus groves including ownership—types of trees, conventional or organic management, and who is packing the fruit. In addition, it will identify factors that could influence the direction and speed of ACP spread, such as weather patterns and traffic corridors. "If there is an ACP or HLB find, we can use the database to assess the risk of spread into urban areas and commercial citrus," said Kris Lynn-Patterson, the GIS coordinator who is leading the project.

Building a better ACP trap

Anandasankar Ray, professor in the Department of Entomology at UC Riverside, is testing olfactory neurons in the antennae of ACP to screen hundreds of chemicals as possible attractants and/or repellents. Attractants added to the standard yellow sticky trap would make a more efficient trap for improved detection and monitoring of psyllids and could eventually lead to an "attract and kill" product. Repellents could also be used as a spray to protect citrus trees.

Treatments for organic citrus farms

Treatment options for homeowners and farmers who do not use synthetic pesticides on their citrus are being explored by scientists with UC Cooperative Extension. The current recommendation for organic growers is to spray a low rate of oil on trees at 14-day intervals. Beth Grafton-Cardwell, UCCE specialist in the Department of Entomology at UC Riverside, is evaluating the effect of this treatment on citrus health, productivity, and fruit quality for San Joaquin Valley navel oranges. Jim Bethke, UCCE advisor in San Diego County, is screening additional organic insecticides on a greenhouse colony of ACP to find products that may have greater persistence and efficacy against ACP.

Introducing natural enemies for biological control of ACP

Mark Hoddle, UCCE specialist in the Department of Entomology at UC Riverside, collected two natural enemies of ACP in Pakistan. These two tiny wasps, *Tamarixia radiata* and *Diaphorencyrtus aligarhensis*, lay eggs underneath or inside ACP nymphs; the hatching larvae eat the nymphs, killing them. Hoddle released *T. radiata* in Southern California in 2012 and is monitoring its establishment and distribution. *D. aligarhensi*s releases are expected in mid 2013. The natural enemies won't eradicate ACP, but scientists predict they will reduce densities of the pest in urban areas, giving other control practices a better chance of working. Richard Stouthamer, professor in the Department of Entomology at UC Riverside, is developing optimal methods for mass-rearing ACP parasitoids, determining the relationship between the numbers of parasitoids released and the rates of establishment, and the impact of parasitoids on ACP populations.

Protecting trees in California plant nurseries

In Florida, ACP was not controlled and it quickly spread on nursery shipments of citrus and orange jasmine that were planted in homeowner backyards. Officials are working proactively to prevent a similar scenario in California. Production nurseries regularly ship young citrus trees in small containers to retail nurseries and outlets. Frank Byrne, associate research entomologist, and Joe Morse, professor, both in the Department of Entomology at UC Riverside, are studying the efficacy of the systemic pesticide imidacloprid. "The treatments can protect the young trees for up to three months," Byrne said. Because retail outlets do not apply pesticides in their facilities, plant protection using chemical pesticides must take place before they leave the production nursery. "Many of the treatments are providing good protection," Byrne said. "One concern is that if trees are kept in stores for extended periods of time, the level of protection starts to diminish."

3 – *Detecting HLB-infected trees*

Finding HLB-infected trees and eliminating them before ACP picks up the disease and spreads it to neighboring trees is a major challenge. The pathogen in the tree cannot be detected by leaf testing for three to nine months after infection, and the symptoms don't show up in the tree for a year or more after infection. Meanwhile, the disease can be spread by ACP. Research is under way to develop early HLB detection so that infected trees can be rapidly removed.

Electronic "sniffer" for determining HLB

Scientists at UC Davis are refining a mobile chemical sensor that can detect diseased citrus trees by sniffing their volatile organic compounds (VOCs). VOCs are emitted by all types of plants and contribute to their distinctive odors—such as the perfume of orange blossoms and pungent scent of garlic in the air. VOCs must exist at very high levels for humans to smell them, and there are some VOCs people cannot smell at all. Cristina Davis, professor in the UC Davis Department of Mechanical and Aerospace Engineering, and Abhaya Dandekar, professor in the UC Davis Department of Plant Sciences, collected samples of VOCs emitted from HLB-infected trees in Florida every month for a year in order to "train" the mobile sensor to recognize the "smell" of HLB. "The idea is to extract a group of compounds that create the signature for the presence of HLB," Dandekar said. A software program develops an algorithm that lets the machine know it is detecting HLB. Davis is working with Applied Nanotech, Inc., in Texas to commercialize this artificial nose.

Metabolic changes can signal presence of disease

When pathogens infect plants or people, changes begin almost immediately to the metabolism of the host. Carolyn Slupsky, UC Davis assistant professor with a split appointment in the Department of Nutrition and Department of Food Science and Technology, is looking at the metabolism of citrus trees infected with the pathogen that causes HLB. "We want to see what chemical or metabolic processes change when the tree is infected," Slupsky said. "Once we understand the metabolic changes induced by the pathogen, the information may help with development of earlier detection methods and new treatments."

The ACP-HLB situation is complex because it involves a tree, an insect, and a pathogen. Homeowners need to understand the impact of HLB on their citrus trees and take part in the management program.

Small RNAs could be early diagnosis markers of HLB

Bacteria, such as the one that is associated with HLB, can cause plants to produce unique molecules called small RNAs, which repress gene expression in plants, animals, and many fungi. These small RNAs signal the plant to produce host-response proteins to protect itself from pathogens. Hailing Jin, associate professor in the Department of Plant Pathology and Microbiology at UC Riverside, is working to identify the HLB-induced small RNAs that will indicate whether a citrus tree is infected with the disease long before the plant expresses symptoms.

Protein secretions may hold the key to early HLB identification

Because the pathogen that is associated with HLB doesn't spread throughout infected citrus trees right away, selecting a branch to test is a shot in the dark. Wenbo Ma, associate professor in the Department of Plant Pathology and Microbiology at UC Riverside, believes that the proteins secreted by the bacterium that is associated with HLB are moved rapidly throughout the tree in the phloem, the food-conducting tissue of the plant. Pathogen-specific proteins in the phloem could be a more reliable disease detection tool than the pathogen itself. Ma and her research associates developed a simple and fast method to sample the phloem. "Choose a couple of branches, cut them off and blot the cut ends on filter paper," she said. "Back in the lab, antibodies are used to detect proteins on the membrane." Ma has used this disease detection technique successfully to detect citrus stubborn disease.

4 – *Finding long-term solutions*

Managing psyllids with insecticides and biological control doesn't eliminate the entire population, and it is difficult to remove HLB-infected trees fast enough to stay ahead of the disease spread. Long-term solutions are needed to develop a citrus tree that can resist or withstand the bacterium and produce good-tasting, abundant fruit or confound the psyllid so that it cannot transmit the disease.

The first HLB-positive citrus tree in California.

Gene fusion could boost citrus trees immunity to HLB

UC Davis scientists are using bioengineering to develop rootstocks that are resistant to HLB and other diseases. Abhaya Dandekar, professor in the Department of Plant Sciences at UC Davis, and his colleagues will use gene fusion, which fuses two immunosuppressive genes that attack HLB in different ways to make the plant more effective at fighting the disease. "Many disease-causing microbes can evade one defensive action of a host plant, but we believe that most microbes would have difficulty overcoming a combination of two immune-system defenses," Dandekar said. Gene fusion is common in nature and is part of what sparks evolution. In a sense, Dandekar and his colleagues are helping citrus evolve into a plant that is immune to HLB.

Gene sequencing for developing resistant citrus

In Florida, researchers have found trifoliate orange rootstock to have some natural resistance to HLB. They have enlisted their long-time collaborators in California to help determine the mechanism of this partial resistance and, eventually, transfer it to edible citrus varieties. Mikeal Roose, professor in the Department of Botany and Plant Sciences at UC Riverside, and his colleagues are assisting in the genetic analysis of about 200 hybrid crosses between sweet orange and trifoliate orange, a species used as a rootstock. "In Florida, they are studying each plant to see how resistant it is," Roose said. "My lab is sequencing a large number of genome fragments to find particular fragments that are associated with resistance. That will help us identify what gene is involved so we can work on transferring that gene." In addition, the Roose lab is trying to make plants resistant to diseases like HLB by using chemical genomics. The objective is to find, in a library of hundreds of thousands of chemicals maintained at UC Riverside, one that mimics a genetic mutation, making the plant more resistant to disease.

"UC ANR research is vital to defeating this major threat to California's $2.1 billion citrus industry. We simply cannot afford to lose this fight."

—Earl Rutz
San Diego County citrus grower

5 – *Engaging and educating the public*

The ACP-HLB situation is complex because it involves a tree, an insect, and a pathogen. Homeowners need to understand the impact of HLB on their trees and participate in the management program.

Communicating with farmers and the public

Grafton-Cardwell has pulled together a team to develop large-scale extension activities and aggressive management programs to stave off devastating losses from ACP and HLB in California. The project is funded with a five-year grant from UC Agriculture and Natural Resources. Project team members will conduct statewide education of commercial growers and the general public. Free PowerPoint presentations (*http://ucanr.edu/acp-hlbpowerpoint*) in English and Spanish can be downloaded and presented by farm advisors, Master Gardeners, and state and federal officials at community meetings, garden clubs, and schools. An online webinar, now in development, will enable growers, homeowners, and landscapers to view detailed information on ACP and HLB from their homes and offices. Grafton-Cardwell and Mary Louise Flint, UCCE specialist and associate director of the UC Integrated Pest Management Program, updated an online 3,000-word Pest Note on ACP and HLB (*http://ucanr.edu/acp-hlbpestnote*), a concise overview of pest identification, disease symptoms, control options, and ways homeowners can help combat the problem. In county Cooperative Extension offices, an ACP-HLB Quick Tip (*http://ucanr.org/acp-hlbquicktip*) in English or Spanish provides the basics in a bookmark-sized format.

UC gathers details on ACP and HLB hosts

A team of USDA and UC scientists are producing a palm-sized flipbook to give CDFA inspectors ready access to photographs and identifying features of 25 plants that host ACP, and, in many cases, can carry HLB. "ACP are found on the Rutaceae family of plants, which includes citrus and many plants that do not look like they have any relationship to citrus at all," said Kahn of UC Riverside, the project leader. Each entry in the flipbook contains a color photo of the full-sized plant, plus close-ups of leaves, blossoms, and fruit. Each plant's characteristics, uses, and risk to California citrus are described. The flipbook will be published by the Citrus Research Board.

Training retail nurseries and Master Gardeners

Matt Daugherty, Cooperative Extension specialist in the Department of Entomology at UC Riverside, will be researching the plant management practices used in retail nurseries and garden centers, such as irrigation frequency, soil type, and pot size. As part of this program, funded with a USDA grant, he will train the nursery industry on best management practices for minimizing the establishment of ACP. Pam Geisel, UCCE academic coordinator for the UC Master Gardener program, is working with Daugherty on a statewide effort to engage UCCE's 5,500 volunteer Master Gardeners in an ACP-HLB education program. Scientists will train Master Gardeners so they can convey information about the pest and its management to residents they serve. The USDA grant will provide funding to evaluate the educational effort, documenting best management practices for extending information to the public via a vast network of volunteers.

Economic models help with management decisions

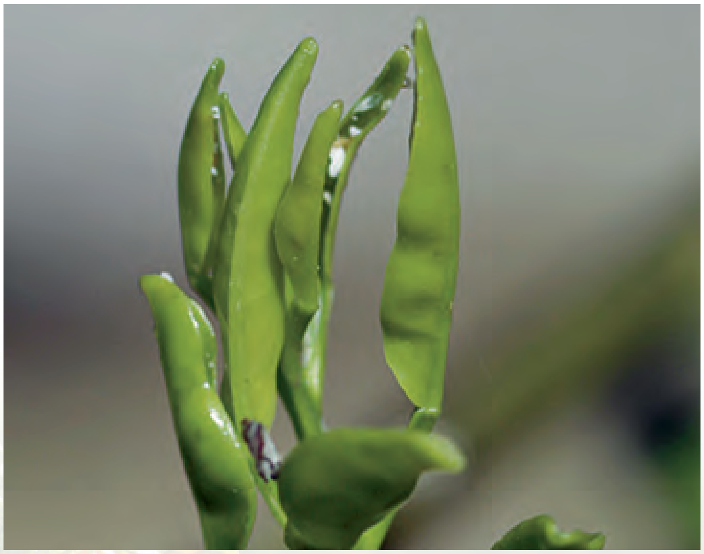
As ACP spreads through California, and when HLB becomes established in California, homeowners, growers, and pest control advisers must make tough pest and disease management decisions. Karen Jetter, associate research economist with the UC Agricultural Issues Center in Davis, is developing economic models to estimate the costs of ACP and HLB management in backyard citrus and commercial orchards and linking the information to a geospatial database. The economic models will take into consideration the tree's age (i.e., its remaining productive life), how ACP management affects other pest management needs, the costs of ACP management programs, and tree removal. "The tool will include all the information necessary for a homeowner, grower, or pest control adviser to determine the most effective and affordable ACP management for his or her situation," Jetter said.

Asian Citrus Psyllid

Adult Asian citrus psyllid feeding.

ACP nymphs excrete distinctive waxy tubules.

Leaves showing symptoms of HLB infection in Florida.



Twisted tips of citrus foliage caused by psyllid feeding.