Yellow Shoot, Green Fruit: Citrus Greening Disease

from interviews with Tim Gast and Tim Watkins, summarized by Stacy Reader

ECHO's Technical Response Unit recently received a few requests from areas in the Caribbean, for information about what might be causing citrus death. One request came from ECHO network member Jean Eloi, founder of the Hope for Haiti Foundation (*www.hopeforhaitifoundation.com* (http://www.hopeforhaitifoundation.com/)), who noticed citrus decline in Haiti:

> Recently I was introduced to a priest whose community has encountered a problem with their citrus trees. These trees are dying and they have not been able to find out the root cause of their death. Do you have teams working with farmers in that area? If so have they encountered this problem? The citrus trees (grapefruits, oranges and tangerines) are all having similar issues and they were hoping that there is a scientific solution that can be found hence for the problems to be able to be prevented.

With multiple requests for information about widespread citrus decline, we decided to learn more about its potential cause and practical management tools. We interviewed Tim Gast, Citrus Production Manager at the University of Florida's Southwest Florida Research and Education Center, and Tim Watkins, Head of Agricultural Operations at ECHO Florida's Global Farm. All information in this article was taken from the interviews, unless otherwise cited.

This article focuses on citrus greening disease, otherwise known as Huanglongbing, which has spread to numerous countries. An assortment of other disease and pest pressures can negatively affect citrus trees--citrus leafminers, canker, root rot, and much more. For information and diagnostic help for these other citrus problems, contact your local extension officer or agricultural technician, visit University of California Davis's problem diagnosis page (http://ucce.ucdavis.edu/files/datastore/530-15.pdf), or visit University of Florida's Citrus Extension Plant Pathology page (http://www.crec.ifas.ufl.edu/extension/plant_pathology/) or Identification guide (http://citrusagents.ifas.ufl.edu/agents/futch/PDF/SP176_English.pdf).

The Cause

Bacteria in the genus *Candidatus* Liberibacter have caused a decline in citrus trees around the globe. The bacteria clog the sugar transport system (phloem) of a tree, effectively destroying the tree's ability to send synthesized starches from the leaves to the roots. The roots die without access to the simple starches that are their food. The compromised root system is then unable to supply leaves with sufficient water and nutrients. Once a tree is infected, there is no cure for the disease. However, we now have more hope that we can help trees recover from and grow out of the disease than we used to.

Two species of *Candidatus* Liberibacter negatively affect citrus trees: *Ca.* L. asiaticus (native to South Asia) and *Ca.* L. africanus (native to South Africa). Originally, the name 'Huanglongbing' was associated with symptoms caused by *Ca.* L. asiaticus, while the name 'greening' was associated with symptoms caused by *Ca.* L. africanus.

'Huanglongbing' ('yellow shoot disease' in Mandarin) originated in Guangdong province in South China, and was first identified in India in the late 1700s to early 1800s. Many translate directly from its written characters to mean 'yellow dragon disease,' but culturally 'long' is a slang word to mean 'plant shoot.' Yellow coloration of new shoots is one sign of infection in trees.

'Citrus greening,' discovered independently in the 1940s and 1950s in South Africa, was also named for a symptom of infection. Infected trees bear fruit that stays small and green or that fails to mature evenly and ends up misshapen.

For simplicity, we will talk about the disease as 'greening' for the remainder of the article.

The Vectors

A vector is an organism that transfers a disease or pathogen. Two species of small, sucking insects called psyllids are vectors of greening: the Asian citrus psyllid (*Diaphorina citri*: Figure 1) and the African citrus psyllid (*Trioza erytreae*). African citrus psyllids are heat sensitive, which limits their range. However, the vector's recent detection in the EU (https://www.freshfruitportal.com/news/2018/01/11/citrus-greening-insect-advances-quickly-portugal-sparking-concerns-spain/) is causing concern (the disease has not yet been detected there). Asian citrus psyllids are present throughout southern Asia and the Arabian Peninsula and in parts of the Americas (CABI Invasive species consortium (https://www.cabi.org/isc/datasheet/18615)). Both psyllids are able to carry either of the bacteria that cause greening. The Arabian Peninsula is one of the few parts of the world where both vectors (psyllid species) and both pathogens (Ca. L species) are present. The ranges of the disease and of the psyllids must overlap for psyllids to become vectors.

Adult citrus psyllids feed on plant stems and on both new and mature leaves, but they prefer young leaves. When an uninfected adult psyllid lands on an infected tree and feeds on new leaves, it incubates the *Liberibacter* bacterium in its gut and becomes a vector of the disease. Later, when it feeds on another tree, it transfers the bacterium and passes on the infection. The most efficient vector is an adult female psyllid who has acquired the bacterium, which incubates in her for 1 to 2 weeks. She later feeds on a shoot (passing on the infection), then lays eggs on the shoot. When nymphs (juveniles) emerge, they feed on the now-infected shoot and ingest the bacteria, which multiply in their guts. As they grow, nymphs continue to eat and infect the same shoot. This constant reinfection weakens the tree.

Figure 1. Adult Asian citrus psyllid (*Diaphorina citri*). *Source: Tim Motis*

To properly monitor potential infection and to understand appropriate treatment, you will first need to identify whether or not psyllids are present on your trees. Adult psyllids are most likely to feed on new growth or shoot tips. They are 2 to 4 mm long (about the size of common aphids). Their brown bodies tilt at an angle as they feed, making them look like thorns (Figure 1). If psyllids have been feeding on a new shoot, you will notice a pinching-like distortion of the leaf edge (Figure 2). Nymphs and eggs are difficult to see without a magnifying hand lens such as those sold by NASCO.

(https://www.enasco.com/product/S07924M)With magnification, nymphs appear orange and secrete white tubules; they are typically found on new stem tissue. Eggs are yellow and are most often deposited on new leaves. For more information about monitoring for pests, see *EDN*

Figure 2. Citrus leaf distortion caused by Asian citrus psyllid feeding. *Source: Tim Motis*

(https://www.echocommunity.org/resources/78ba129d-56a3-43b6-abd9-dc963495f235)136
(https://www.echocommunity.org/resources/78ba129d-56a3-43b6-abd9-dc963495f235).

Psyllids are the predominant vectors of greening, but humans can also unwittingly cause transmission of the bacteria by propagating with or transporting infected plant material. Use of infected budwood for grafting or budding will spread the disease to the newly grafted trees. Infected trees that are then sold and carried throughout the surrounding region provide more infected hosts for psyllids.

Symptoms

Monitoring for psyllids is extremely important, because there is no simple method for early detection of greening, and significant root death occurs below ground before any signs are apparent above ground. If you live in a region affected by greening, and you see psyllids on your trees, the trees are most likely infected regardless of whether or not you see other symptoms.

Leaves

Figure 3. Leaf with blotchy mottled greening symptom (left) and a healthy leaf (right). *Source: Tim Motis*

As the Asian name 'yellow shoot disease' suggests, yellow newly emerged shoots in a citrus tree canopy are a sign of infection . This yellowing is not caused by a nutrient deficiency, but by the buildup of starches synthesized in the leaves; bacteria clog the vascular tissue so that the starches cannot travel down the phloem of the tree to the roots.

Nutrient deficiencies can also cause yellowing, but it is possible to distinguish between them and greening symptoms. A leaf yellowed because of greening will have an asymmetrical blotchy mottled pattern, with small islands of green (Figure 3). By contrast, leaves that are yellowed due to nutrient deficiencies will have symmetrical patterns on either side of the leaf midvein. [Nutrient deficiencies are common on trees infected with greening; zinc deficiency is especially common. Corky or raised mid-veins are also common on leaves of infected trees, but alone do not diagnose greening.]

Infected trees may drop their leaves, and their new leaves may be pointed and look like 'rabbit ears.' Mottled leaves along with leaf drop and/or pointy new growth are strong indicators of citrus greening.

Figure 4. Citrus fruit with greening symptoms (left) and a healthy fruit (right). *Source: Tim Motis*

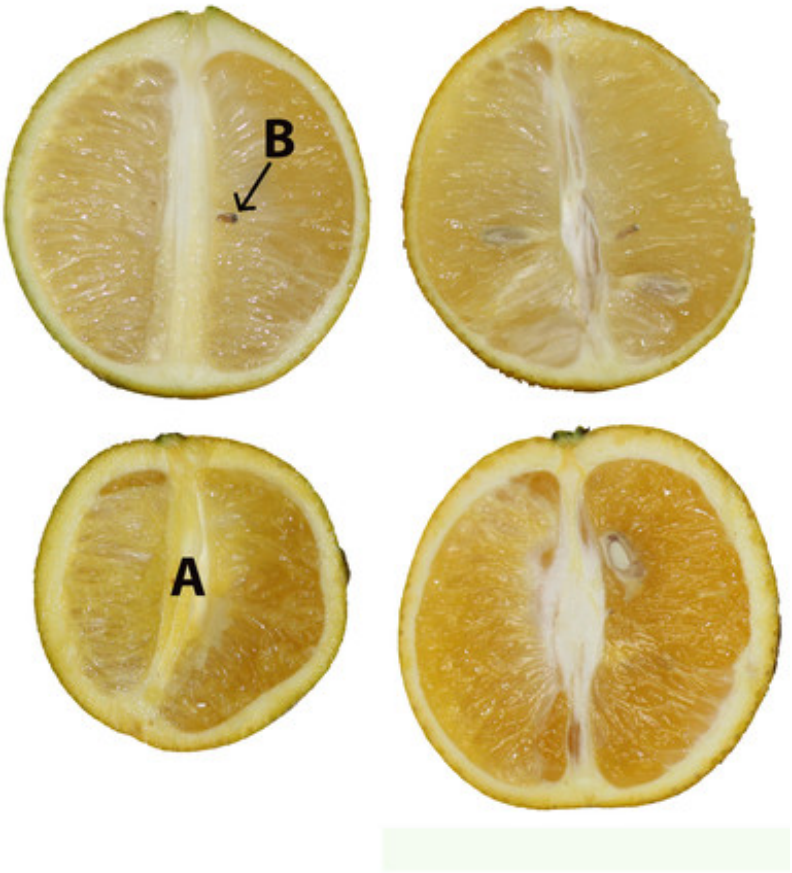

Figure 5. Cross section of citrus fruit with greening symptoms (left) showing a curved central core (A) and immature, brown seeds (B). Healthy fruit (right). *Source: Tim Motis*

Fruit

Fruit from infected trees may appear lopsided or misshapen; it also might stay green even when mature (Figure 4). Additional symptoms may been seen when fruit are cut in half; there may be a yellow stain under the calyx button (where the fruit attached to the tree), a curved central core, and aborted seeds which are hard and brown (Figure 5). Fruit from infected trees may taste salty or bitter.

Tree

General tree symptoms can give some indication of infection; these include twig dieback (resulting in a less dense tree canopy), stunting, off-season flowering, and overall tree decline. However, these general symptoms may result from a different disease or from stress, so look for a combination of leaf, fruit, and tree symptoms when monitoring for greening.

University of Florida/IFAS Citrus Extension Plant Pathology has sites where you can view photos of common greening symptoms (http://www.crec.ifas.ufl.edu/extension/greening/symptoms.shtml) or compare greening symptoms with nutrient deficiency symptoms (http://www.crec.ifas.ufl.edu/extension/greening/ndccg.shtml) and also provide sampling instructions (http://www.crec.ifas.ufl.edu/extension/greening/PDF/ACP_sampling_english.pdf) for in-field monitoring.

Protection Strategies

Several strategies can be used to protect against citrus greening. Here, we focus on strategies for farmers with limited access or availability of resources. A short list of some recent, more technical approaches is included at the end of this article under "Additional Resources." A range of organic and inorganic options are presented below. In selecting and applying any of these, consider integrated pest management principles.

Protect in the Nursery

Nurseries that propagate citrus plants or other plants that host citrus psyllids must use preventive measures, to stop the geographic spread of any diseased or vector hosting material and to make sure that citrus propagation material is clean (uninfected). Curry leaf tree (*Murraya koenigii*) and jasmine orange (*Murraya paniculata*), relatives of citrus that are sold as ornamentals, are hosts of Asian citrus psyllids and must be included in preventive measures.

Be sure to obtain clean budwood for grafting and budding. Research institutions, government-led programs and other entities actively protect and maintain budwood to preserve healthy stocks of grafting material. Ask your local extension agent or field technician for sources of locally available material. Fruitmentor™ (http://www.fruitmentor.com/citrus-budwood-programs) offers an incomplete list of international sources of budwood. When importing plant material, various international and national laws, such as a requirement for phytosanitary documentation, may apply.

Screens on greenhouses can help exclude psyllids, especially if combined with positive pressure ventilation, in which greenhouse air pressure is kept higher than outside air pressure. When a door is opened, air rushes out at a rate faster than an insect can fly, excluding insect pests (Mears and Both 2002 (https://www.researchgate.net/publication/279979529_A_positive_pressure_ventilation_system_with_insect_screening_for_tropical_and_subtropical_greenhouse_fa Where positive pressure ventilation is not feasible, screens with a hole size of 530 x 530 microns (32 x 32 treads/inch) will exclude psyllids (Stansly (http://www.crec.ifas.ufl.edu/extension/trade_journals/2006/March%202006%20managing%20psyllids.pdf)and Rogers 2006 (http://www.crec.ifas.ufl.edu/extension/trade_journals/2006/March%202006%20managing%20psyllids.pdf)).

Experts recommend that you treat nursery trees every six months with a systemic, soil-drench neonicotinoid, such as those with active ingredients thiamethoxam or imidacloprid. Neonicotinoids are broad-spectrum insecticides that quickly spread to every part of the tree and deter insects from feeding. Some formulations can be applied to the foliage, but a soil drench is said to be most effective (Rogers (http://edis.ifas.ufl.edu/in686)*et al.* (http://edis.ifas.ufl.edu/in686) 2016 (http://edis.ifas.ufl.edu/in686)). These insecticides may not be widely available or affordable to small-scale farmers. Farmers may also wish to use insecticides less likely to impact beneficial insects such as bees. For such cases, see content in the next section for foliar-applied psyllid control options.

Monitor your nursery regularly, looking for infection symptoms and for the presence of psyllids. If you identify tree seedlings with greening, remove them immediately and burn the diseased plant tissue.

Tree removal

Regionally, if greening is detected early, eradication of affected trees may be necessary to protect the local industry. If you discover greening in an area that has an infection rate less than 2%, keep pulling out trees. Both Texas and California citrus industries in the United States still practice eradication programs in an effort to remove diseased material and slow down disease transmission. However, past a certain point, eradication ceases to be an effective protection strategy. According to Tim Gast, economic models and studies show that in areas with more than 4 to 5% infected trees, you will not get ahead of the disease through tree removal.

Management Strategies

"We used to say, 'It's dead as soon as the tree gets it, it's done. In a couple of years, it's gonna die.' But that's not the truth," said Tim Gast as we were talking about citrus management. He gave us at ECHO hope that, with proper management strategies, we can help trees grow out of citrus greening disease and be productive again. He informed us that every two years, citrus trees replace all of their leaves; the trees are also constantly putting on new phloem. As we give citrus trees targeted support, chances are that they will grow out of the disease. "I've seen thousands of trees grow out of it," Tim encouraged us. Infected trees must be properly cared for and managed to minimize stresses such as overwatering, insufficient watering, over fertilization, under fertilization, sunburn, frost burn and excessive pest pressure.

Psyllid Control

Young trees

Young trees tend to flush more frequently than mature trees, attracting psyllids and making young trees more susceptible to infection and re-infection.

Shoots and leaves can be treated two times per flush, once as the new leaves emerge and again just after they harden off. Because temperatures and rainfall fluctuate in the tropics, the timing of flushes may be unpredictable, so be prepared to treat trees at any time. There are other commonly used foliar applications that either kill psyllids or deter them from eating. Oil and soap dilutions sprayed on tree leaves kill psyllids while diatomaceous earth, wood ash, and kaolin clay applications deter insects from feeding on leaves.

Tim Motis, co-editor of *EDN*, shared a simple recipe to control psyllids:

> After observing a number of citrus greening psyllids on my lemon tree at home, I decided to spray a combination of liquid dish soap and vegetable oil. I added 2 teaspoons (10 ml) of dish soap and 1 teaspoon (5 ml) of vegetable oil to a gallon (3.8 l) of water, in a one-gallon sprayer. After shaking the sprayer to mix the ingredients, my 7 and 11 year old boys took turns spraying the leaves. The tree was short enough for them to reach most of the canopy, and I helped reach leaves at the very top. When I inspected the leaves a day or so later, every psyllid I found was dead. This simple recipe, used in conjunction with a hand-operated sprayer, is very doable for one or two trees in my yard.

On a larger scale, many citrus growers treat young trees every six months with a systemic, soil-drench neonicotinoid to deter psyllids. The soil drench is applied during drier periods, to prevent the treatment from leaching beyond the root zone. In place of soil drenches, neonicotinoid foliar sprays are also an option.

Mature trees

During cold and/or dry seasons, the growth rates of both trees and psyllids typically slow down. Psyllids become lethargic during a prolonged dry or cold period, providing the opportunity to manage them as a population--for example, by spraying trees with soap or oil sprays as described above.

Biological controls

Several common generalist insect/arthropod predators consume Asian citrus psyllid nymphs. Lacewings, spiders, and especially ladybugs all feed on psyllid nymphs. An introduced wasp, *Tamarixia radiata*, is a highly effective predator, killing up to 95% of nymphs (Michaud 2004 (http://www.imok.ufl.edu/hlb/database/pdf/00000535.pdf)). Female wasps lay eggs on the bodies of psyllid nymphs; after hatching, wasp larvae consume the body fluid of the psyllids, killing them.

Supply Nutrient and Water Requirements

Blockage of the phloem in an infected tree causes it to lose between 50 and 70% of its feeder roots. As a result, roots have limited ability to send water and nutrients to the leaves. In order to facilitate healthier conditions for growth, we must supply nutrients and water to an infected tree's disjointed parts. (Note that the disease affects the tree vertically but does not spread rapidly throughout the tree horizontally except when psyllids reinfect different shoots of the same tree.)

Root Fertilization

Feed roots by supplying macro and micronutrients, to support the root system while you wait for the tree to put on new phloem. Aged manure, compost and synthetic fertilizers are all options. What you choose to use will depend on availability and access. Fertigation--micronutrient supplementation through an irrigation system--is very effective but may not be readily available.

The amount of fertilizer and frequency of application will depend on the type of fertilizer you choose, the climate of your region, and the sizes of trees. Contact your local extension officer or field technician for guidance.

Irrigation

Infected trees need more frequent irrigation than healthy trees, because greening compromises the root system, reducing water supply to the leaves. Irrigate trees as the soil dries out, but be careful not to overwater. Wet soil or poor drainage can cause root rot, which can easily kill an already weakened tree.

Shoot Fertilization

Shoot health can be supported by applying organic or inorganic foliar nutrient sprays. If using synthetic foliar sprays, refer to product instructions. Homemade foliar sprays require labor inputs but can utilize local resources and can be ready to use in as little as two weeks. *West Africa Note* (https://www.echocommunity.org/resources/849b5186-0779-47eb-8235-125a6bb31e0d) 1 (https://www.echocommunity.org/resources/849b5186-0779-47eb-8235-125a6bb31e0d) includes a recipe for organic liquid fertilizer made from manure, green matter, soil and water. Fermented fish can be used to make a foliar feed high in nitrogen. Natural Farming (https://www.echocommunity.org/en/resources/45715fab-1156-43b3-8c5a-1363487b43af) techniques include instructions for making foliar sprays. If you make a foliar nutrient spray for your crops, please share your experience and insight with the ECHO Community (https://conversations.echocommunity.org/t/foliar-feed-sprays/332?u=stacy_reader).

Figure 6. Mandarin 'Orah' variety from Israel cultivated in China. Trees are kept short for ease of maintenance. White material on the leaves is lime, which is applied to help prevent sun burn on the fruit. *Source: Tim Gast*

Keep Trees short

At a grove in China, Tim Gast observed that the grower kept trees short for ease of maintenance, which was entirely done by hand (Figure 6). There, laborers apply agricultural chemicals with backpack sprayers that are able to reach the short trees. Greening is endemic to the region he visited, but he did not see greening symptoms in the grove. They also utilize geese for weed management, but must supplement the birds' diets with feed during winter months (Figure 7). Here at ECHO Florida, we have found that chickens and sheep effectively control weeds under a number of tree species.

Figure 7. Geese removing weeds at a grove in China. Grove manager says that they do have to supplement geese feed in winter months when weeds are not vigorous. *Source: Tim Gast*

Variety Selection

Certain citrus varieties, including Nova Tangelo, Dancy Tangerine and Sugar Bell Tangerine, are more tolerant of greening, showing fewer symptoms of the disease than other varieties. You may create resiliency in your orchard by choosing tolerant varieties. If your region has greening and psyllids, but you do not know which local citrus varieties are tolerant, observe trees to see which are asymptomatic. Harvest a few fruits from those trees (with permission) and evaluate the taste, shape and evenness. Share your observations with your community and with researchers or extension workers in your region.

Conclusion

Citrus trees are discouragingly susceptible to a myriad of diseases, including citrus greening. But farmers and researchers are making hopeful new observations and discoveries. ECHO has been encouraged by recent conversations with local fruit growers. We hope that understanding how best to steward your citrus trees helps them to survive and, one day, thrive.

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Additional Resources

General Citrus Problem Information

University of California Davis has a comprehensive citrus disease diagnostic chart (http://ucce.ucdavis.edu/files/datastore/530-15.pdf) which lists citrus symptoms, likely causes, and recommended control methods.

Arizona Cooperative Extension has a home diagnostic sheet (https://extension.arizona.edu/sites/extension.arizona.edu/files/pubs/az1492.pdf) that includes helpful images of symptoms related to citrus problems.

Pathogen and Vector Information

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The California Department of Food and Agriculture Plant Health & Pest Prevention Services provides a condensed overview (https://www.cdfa.ca.gov/plant/acp/docs/factsheets/psyllidbrochureaug05.pdf) of the asian citrus psyllid history, distribution, life cycle and role as a vector and also gives suggested control methods. The Department also provides a list of plants that are hosts (https://www.cdfa.ca.gov/plant/pdep/target_pest_disease_profiles/hostlists/AsianCitrusPsyllid-HostList.pdf) to the Asian citrus psyllid.

Florida Department of Agriculture and Customer Services offers options on biological control (https://www.freshfromflorida.com/Divisions-Offices/Plant-Industry/Bureaus-and-Services/Bureau-Of-Methods-Development-Biological-Control/Biological-Control/Asian-Citrus-Psyllid-Biological-Control/Biological-Control-of-Asian-Citrus-Psyllid-in-Dooryard-Citrus-and-Ornamentals) of Asian citrus psyllids and, depending on levels of stock, will ship *Tamarixia radiata* (a species of parasitic wasp) to eligible applicants who file a release (https://www.freshfromflorida.com/Divisions-Offices/Plant-Industry/Bureaus-and-Services/Bureau-Of-Methods-Development-Biological-Control/Biological-Control/Asian-Citrus-Psyllid-Biological-Control/Biological-Control-of-Asian-Citrus-Psyllid-in-Dooryard-Citrus-and-Ornamentals/Tamarixia-Release-Application) application (https://www.freshfromflorida.com/Divisions-Offices/Plant-Industry/Bureaus-and-Services/Bureau-Of-Methods-Development-Biological-Control/Biological-Control/Asian-Citrus-Psyllid-Biological-Control/Biological-Control-of-Asian-Citrus-Psyllid-in-Dooryard-Citrus-and-Ornamentals/Tamarixia-Release-Application).

University of Florida IFAS offers an Integrated Pest Management page on Asian Citrus Psyllid and Citrus Greening Disease (http://ipm.ifas.ufl.edu/Agricultural_IPM/asian.shtml) as well as a species highlight on *Tamarixia radiata* (http://entnemdept.ufl.edu/creatures/beneficial/wasps/tamarixia_radiata.htm#top). They also have an updated 2017-2018 Citrus Production Guide (http://www.crec.ifas.ufl.edu/extension/pest/PDF/2017/Huanglongbing.pdf); Huanglongbing (Citrus Greening) with recommended management practices.

CABI (Centre for Agriculture and Biosciences International) has an Invasive Species Compendium with datasheets on citrus huan (https://www.cabi.org/isc/datasheet/16567)g (https://www.cabi.org/isc/datasheet/16567)longbing (greening) disease (https://www.cabi.org/isc/datasheet/16567), Asian citrus psyllid (https://www.cabi.org/isc/datasheet/18615), and (https://www.cabi.org/isc/datasheet/54914)A (https://www.cabi.org/isc/datasheet/54914)frican citrus psyllid (https://www.cabi.org/isc/datasheet/54914). These sheets include useful distribution maps of the disease and vectors, which can help you discern the likelihood of the vector and/or disease reaching your area.

Recent and Upcoming Technical Advancements

A recent article from Growing Produce (http://www.growingproduce.com/citrus/first-time-california-no-1-citrus/) shared that University of California Davis citrus experts are working with farmers to develop detection methods that can chemically profile leaves early in the infection process.

Many researchers around the world are breeding, grafting and culturing citrus plant tissue in an effort to identify and/or develop tolerant or resistant citrus tissues. There has been some success; for example, the preferred rootstock for many nurseries in the United States: 'US-942,' is 'HLB tolerant'. The University of Florida's '#4' rootstock is considered resistant to greening, but is not available yet. A recent article in Science Daily (https://www.sciencedaily.com/releases/2017/12/171218092421.htm) summarizes the hope of imparting greening tolerance with use of new citrus rootstock varieties.

The UF/IFAS Citrus Research and Education Center is the site of much research on citrus and greening. UF/IFAS Extension is investigating the interactions of soil microbial populations (http://www.imok.ufl.edu/programs/soil-microbiology/citrus-soil-amendment/), applied both directly and indirectly, to understand if changes in the soil microbe population can benefit citrus trees.

United States Department of Agriculture has evaluated varieties for tolerance and resistance (http://citrusagents.ifas.ufl.edu/events/GrowersInstitute2016/pdf/HLB%20Tolerence%20and%20Resistance%20Florida%20Citrus%20Growers'%204.16.pdf) to HLB and is also testing a new trap to trick psyllids with sound (https://agresearchmag.ars.usda.gov/2016/may/psyllids/).