Diaprepes in Florida citrus: Past, present and future

By Lukasz L. Stelinski, Lauren Diepenbrock and Larry Duncan

Given the focus on HLB in citrus management for more than a decade, important additional pests of citrus have sometimes not received the attention they may deserve. Trees with HLB are weakened and prone to succumb to the effects of the disease when challenged by secondary affliction(s), including infestation by diaprepes root weevil.

Diaprepes themselves cause root girdling and can promote infection of roots by *Phytophthora* spp. Prior to HLB, diaprepes and the sudden decline of groves caused by weevil damage to citrus root systems were primary concerns in Florida citrus production. This problem was never completely solved. However, it was not a pressing concern during the period between approximately 2008 and 2018 because of the intense spraying for psyllids during that time. This did not eradicate diaprepes but seemed to have maintained their populations at low levels.

More recently, spraying intensity targeting Asian citrus psyllid (ACP) has been somewhat less intense across Florida. While ACP and other pests still require and receive insecticide, application of 12 annual calendar sprays for ACP alone is becoming less common. Under these circumstances, diaprepes infestations appear to have increased in frequency. However, the situation is different now because trees are infected with the *Candidatus* Liberibacter asiaticus pathogen that causes HLB. HLB is a root disease and thus the root zone of infected trees is already weakened.

LIFECYCLE AND HABITS

Diaprepes attack and feed on a wide range of plants. This includes around 300 different species and many economically important crops in Florida and elsewhere such as citrus, sugarcane, vegetables, potatoes, strawberries, woody ornamentals, sweet potatoes, papaya, guava, mahogany, containerized ornamentals and non-cultivated wild plants. This complicates control and explains why the weevil is so well established in Florida.

Diaprepes adult damage to the vegetative portion of plants is most often seen as notching of young leaves (Figure 1, page 17). Adults mate and lay eggs between older leaves (Figure 2, page 18). However, the greatest damage is caused by larvae feeding below ground. Upon hatching, the larvae fall to the soil and make their way to the roots of plants where they feed and develop. This feeding can girdle major roots, greatly reduce the rhizosphere and cause damage that disables the plant from taking up water and nutrients, which can result in plant death.

Diaprepes damage facilitates secondary infections by *Phytophthora* fungus species. Young hosts can be killed by a single larva while several larvae can result in serious decline of older, established hosts. Because larvae develop below ground, it is difficult to detect them before decline of aboveground vegetation of the host is observed.

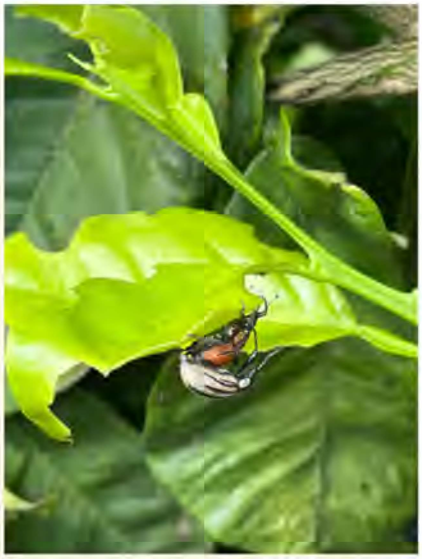

Figure 1. Typical leaf notching caused by diaprepes adult feeding

AVAILABLE MANAGEMENT TOOLS

Current chemical controls recommended for diaprepes are based on investigations that were conducted many years ago, but they are not outdated. Soil-applied insecticides like Brigade WSB and Capture 2EC can be applied to create a soil barrier that decreases larval entry. Foliar insecticides can reduce adult feeding, oviposition and viable egg production. Foliar chemical spray applications such as Danitol 2.4 EC, Imidan 70-WP, Kryocide 96 WP and Micromite 80WGS are most effective during peak seasonal diaprepes abundance.

A full list of insecticides recommended for diaprepes control can be found in the 2022–23 Florida Citrus Pest Management Guide. Many newly developed insecticide chemistries that are often used in Florida citrus have not yet been evaluated against diaprepes. Researchers are currently working on addressing this information gap.

The most effective method for

State of the CRISPR Tree

By Rick Dantzler, CRDF chief operating officer

The question growers most often ask me is: What is the state of the CRISPR tree? Since a breeding solution is the most likely way to put HLB behind us, it is a good question.

Clustered regularly interspaced short palindromic repeats (CRISPR) is based on a process that bacteria use to destroy other bacteria in which an enzyme — guided by RNA — can cut DNA in a particular area. For plants, this process can knock out a gene by removing one or a few nucleotides, making it nonfunctional. CRISPR mutations insert no foreign sequences into citrus and thus should be considered non-GMO.

The Citrus Research and Development Foundation (CRDF) funded initial work developing the CRISPR technology for citrus, and there are several labs now working on different approaches to creating trees resistant or tolerant to HLB. One lab has already created canker-resistant trees and is in the process of moving these trees into commercial production.

CRDF also funded the work that led to identification of potential "susceptibility" genes that allow *Candidatus* Liberibacter asiaticus to infect citrus that are the CRISPR targets of Soilcea, a biotech company commercially developing HLB-resistant citrus. This is a good example of how public sector research works. CRDF provided funding to the University of Florida Institute of Food and Agricultural Sciences to develop the technology and product, and the private sector acquired what had been developed and commercialized it. CRDF is discussing with Soilcea ways that we can assist in getting its trees into the field.

A problem with CRISPR and traditional citrus breeding is that plants initially have a juvenile phase that can last for years, which is why another CRDF project is the use of the citrus tristeza virus vector to quickly induce maturity. Regardless of the issues remaining with CRISPR, we can't let up. The next step is to get new creations into the field to determine if they will work in a commercial growing environment. There is a chance that a tree possessing enough tolerance or perhaps even resistance has already been developed. However, trees have the genes they do because they need them. So, until we get such trees in the field, we can't tell what will happen when certain genes are silenced.

CRDF is supporting several parallel approaches, too. One is a modification of production of transgenic citrus called "cisgenics." This is a process where all the sequences inserted into the tree are from citrus or citrus relatives that can be crossed into commercial citrus. The result is a tree that could have been produced by traditional crosses but produced in a shorter time, which may allow it to be considered non-GMO.

CRDF is also funding the production of transgenics that could provide resistance to HLB. Although these trees would be GMOs and require normal U.S. Department of Agriculture and Environmental Protection Agency regulations, until CRISPR or cisgenic trees are proven to be effective against HLB, we should pursue this strategy as a backup. Again, of paramount importance is to get these trees into the field for testing in commercial growing environments.

CRDF Grower Dollars at work for you

Column sponsored by the Citrus Research and Development Foundation controlling the more damaging diaprepes larvae found on roots is entomopathogenic nematodes (EPNs). These are roundworms from the genera *Heterorhabditis* or *Steinernema*. They are obligate parasites that kill their host with the aid of symbiotic bacteria. Native and introduced EPNs are infectious to all larval stages and possibly adults. Using EPNs can reduce larval populations of diaprepes. Growers have widely adopted the use of commercially formulated EPNs since they became available in 1990 to manage the soil stages of the weevil. Although EPNs are highly effective against diaprepes larvae, they are non-persistent, effective for about six weeks and too costly to apply frequently to the entire area under the tree canopy.

Figure 2. Mating pair of diaprepes adults

A problem that emerged from intense focus on ACP management is that companies, such as Becker Underwood, stopped producing and shipping EPN formulations for use in citrus. As the market declined because of psyllid sprays, the formulations became more difficult to find. Fortunately, as companies such as BASF have been made aware that diaprepes is emerging as an important problem again, they've begun growing these nematodes again and are preparing distribution of product.

The renewed interest by growers in using EPNs for larval control underscores the need for research to increase their efficacy by modifying the tactics by which EPNs are employed. For example, if EPNs were used to protect just the crown of the root system from weevil damage, fewer nematodes would be required, permitting frequent applications to keep them actively present in the most vulnerable part of the tree.

RESEARCH ROUNDUP

Research in Florida, Texas and California showed that certain landscape fabrics can be used as barriers to prevent the small, newly hatched diaprepes larvae from entering the soil, while also preventing young adult weevils from escaping soil and moving into the tree canopy.

A woven weed fabric product (Lumite woven groundcover), fitted over soil in pots, prevented soil entry by 99% of neonate weevil larvae in a trial conducted by the U.S. Department of Agriculture in Fort Pierce. The effectiveness of the fabric for weevil management was studied in several small plot experiments in Florida. Compared to trees in bare soil, fabric-mulched trees on the east coast grew 70% larger after three years. Mulched trees tolerated weevil damage better than trees in bare soil, but weevil feeding damage to the roots in each treatment did not differ.

In trials near Lake Alfred (newly planted trees) and Poinciana (20-year-old trees), fabric was installed with a width of 5 feet on each side of trees. Five years after fabric was installed at Lake Alfred, the trees in mulched plots were 31% larger than those in bare soil. As few as 1% as many diaprepes adults at the two sites were caught in ground traps installed beneath mulched trees compared to trees in bare soil.

In California, fabric mulch beneath lemon trees suppressed weevil emergence by 100%, except when the fabric was damaged by grove operations. Tree health after three years was higher for mulched than non-mulched trees.

Research in Texas confirmed the

benefit of fabric mulch for weevil population control and increased tree growth.

To date, there are no reports of whether fabric mulches affect the fruit yield of citrus trees.

FUTURE RESEARCH

In response to the recent resurgence of diaprepes, when most citrus trees are infected with HLB, we have begun work to develop the needed tools and management recommendations to limit the potential damage. The use of landscape fabric was shown to increase citrus tree growth rate and is thought to greatly reduce the population density of diaprepes; however, experimental validation is lacking. Cultural control could feature prominently for diaprepes management in the future, but these methods are entirely lacking currently.

We are hoping to establish a trial in which landscape fabric is installed in the tree rows mechanically. The effects of landscape fabric need to be investigated by determining effects on tree infestation by diaprepes, health and yield in future years as compared with no fabric controls in replicated field trials. For example, even if fabric reduces weevil damage, it is unknown whether trees debilitated by HLB can respond sufficiently to make fabric installation profitable.

In the coming years, we also plan to determine which currently available and new insecticides are useful against diaprepes and the required rates to update management recommendations. Finally, we still need to determine the common source of diaprepes infestations and accurately quantify the movement patterns of beetles between non-host habitats outside of groves, grove borders and grove interiors. Determining the source of diaprepes infestation and where the pest invades groves may allow us to target the source of these invasions to prevent or limit infestations before the need for whole-grove sprays.

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