Seeking Long-term Solutions to HLB

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This research team uses a multiapproach to seek long-term solutions to the devastating citrus disease HLB. Our strategies are based on the positive or negative regulations of the citrus immune system that controls the level of resistance, tolerance, or susceptibility to HLB. We have generated transgenic citrus trees that over-express positive immune regulators and provide robust tolerance to HLB. These transgenic trees are already in field tests as a potential management option for HLB. We also attempted to produce transgene-free HLB-tolerant plants by gene editing using



A negative regulator of the citrus immune system has been silenced using CTV-RNAi, a non-transgenic approach, in the 'Pineapple' sweet orange plants in the right panel. All plants have been infected and contain similar levels of the HLBcausing bacterial pathogen.

CRISPR/Cas9. To this end, we use CTV-delivered RNA interference (CTV-RNAi) to screen negative regulators of the citrus immune system for gene editing targets. Candidate genes that provide HLB tolerance when silenced by RNAi have been identified. Since CTV-RNAi is a non-transgenic approach, silencing of these genes can be used as an interim treatment of already infected mature trees and in resets. We are currently applying for permits for field testing of this strategy. Furthermore, we have built a highly efficient intragenic vector using citrus DNA sequences, which allows production of intragenic plants that do not contain "foreign DNA" sequences. Compared with transgenic plants, intragenic plants are expected to have less stringent regulatory requirements. We use this vector to produce intragenic citrus trees that are tolerant to HLB. Finally, we continue to educate the citrus communities about the CTV-RNAi, CRISPR/Cas9, and intragenesis technologies and their benefits to the citrus industry and consumers to help establish market acceptance for modern biotechnology-based citrus products.

Funding



USDA National Institute of Food and Agriculture U.S. DEPARTMENT OF AGRICULTURE

