

New discovery speeds scientists' push for Huanglongbing-tolerant citrus

February 28 2024, by Jan Suszkiw



A genetic discovery by ARS and University of Florida scientists could speed the search for hybrid citrus trees that tolerate citrus greening disease and produce orange-like fruit ideal for making juice. (Photo by David Bartels, USDA

It's one thing for a hybrid citrus tree to tolerate citrus greening disease (a.k.a. Huanglongbing) and quite another if it also produces orange-like



fruit—especially if the juice makes for a delicious breakfast beverage. Now, that holy grail of traits could be closer at hand, thanks to the chemical and genetic sleuthing of a team of Agricultural Research Service (ARS) and University of Florida Institute of Food and Agricultural Sciences (UF/IFAS) scientists.

Huanglongbing (HLB) was first detected in Florida's Miami-Dade County in 2005. The disease has since spread throughout Florida, threatening a citrus industry there that contributes nearly \$7 billion to the state's economy and employs more than 32,000 individuals. The disease also poses a threat to other U.S. citrus-growing areas, including California, Louisiana and Texas.

In Florida, sweet oranges like Valencia, Hamlin and Midsweet are the main varieties used to make <u>orange juice</u>. However, sweet orange (Citrus sinensis) is highly susceptible to HLB, which is caused by the bacterium Candidatus Liberibacter asiaticus and transmitted by insects called Asian citrus psyllids.

The disease causes <u>citrus trees</u> to become unproductive and fruit quality to decline. In sweet oranges, for example, the fruit of diseased trees tend to stay green and produce bitter-tasting juice, which diminishes their marketability but poses no consumer danger. Infected trees cannot currently be cured.

Scientists are investigating countermeasures on multiple fronts in hopes of providing the Sunshine State—which went from producing nearly 80% of U.S. non-tangerine citrus fruit to less than 42%—with a ray of hope. ARS efforts indirectly took root in the 1960s, when agency scientists created citrus hybrids using a relative named Poncirus trifoliata (the cold-hardy trifoliate orange) to shore up the trees' cold tolerance.

The start of the HLB epidemic more than four decades later revealed



something else: The hybrids also appeared to tolerate the new disease, prompting intensive research by ARS and the UF/IFAS to understand why and how this related to fruit quality.

Initial field tests and flavor evaluations showed that some of the Poncirus-derived hybrids—with the notable exception of US Sundragon—tended to produce juice with an undesirable off-flavor but aroma profiles similar to sweet orange.

So, the scientists re-assessed their approach. They decided that in addition to using data from analyses of juice-aroma compounds, they needed to get a better handle on the individual chemicals that give orange juice its characteristic flavor. And they did just that, identifying 26 total flavor compounds and seven chemicals called esters deemed essential to the desired flavor profile of orange juice.

That advance, in turn, enabled the team to pinpoint the esters' master gene, CsAAT1, and make what's known as a DNA marker for it—a tool that can be used to quickly check for the genetic presence of a desirable trait in germinated seeds versus observing its physical expression in 10-or 15-year-old mature plants.

"Breeders can use this DNA marker to screen seedlings for desired flavor profiles at an early stage," explain Anne Plotto and Jinhe Bai, plant physiologists with the ARS Citrus and Other Subtropical Products Research Unit in Fort Pierce, Florida.

"By incorporating this gene into the genetic makeup of HLB-tolerant hybrids derived from Poncirus trifoliata and mandarin, or from many other possible crosses with the same objective, breeders can ensure that these new hybrids not only possess HLB tolerance but also maintain the characteristic sweet orange flavor."



A full description of the team's approach was <u>published</u> February 28, 2024 in the journal *Science Advances*.

Plotto and Bai, the principal investigators, co-authored the paper together with 11 other collaborators from ARS' U.S. Horticultural Research Laboratory in Fort Pierce, Florida, and Daniel K. Inouye U.S. Pacific Basin Agricultural Research Center in Hilo, Hawaii, and UF/IFAS' Citrus Research and Education Center in Lake Alfred, Florida, and Gulf Coast Research and Education Center in Balm, Florida.

The researchers caution that even with the use of high-tech tools like machine learning, the first commercial releases of orange-like hybrids with HLB tolerance will be contingent on several more years of testing and refinement.

Nonetheless, "this research represents a significant step in citrus breeding, combining traditional techniques with modern genetic tools," said Plotto and Bai. "The approach could also serve as a model for other crop improvement programs."

More information: Zhen Fan et al, Chemical and genetic basis of orange flavor, *Science Advances* (2024). DOI: 10.1126/sciadv.adk2051

Provided by Agricultural Research Service

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