Rootstocks and HLB tolerance — another perspective

By Ute Albrecht

Tolerance to a disease is generally defined as the ability to be productive in the presence of disease-causing organisms. This is contrary to resistance, which is defined as the ability to completely evade a pathogen due to specific resistance mechanisms.

The question as to what defines an HLB-"tolerant" rootstock was posed in an article by Bill Castle, Jude Grosser, Ed Stover and Kim Bowman in the June 2015 issue of Citrus Industry magazine. The article describes observations from several rootstock field trials conducted in collaboration with commercial growers. Independent surveys of these trials conducted by researchers and growers on tree appearance and crop ratings indicated that no one rootstock appears to be tolerant 100 percent of the time. Rather, tree performance seemed erratic and subject to influences by other factors such as psyllid incidence, cultural practices and soil environment.

Even though observations on commercially grown citrus trees show tremendous variability, it has clearly been demonstrated that some rootstock varieties, when grown as seedlings, are highly tolerant to HLB under greenhouse and field conditions and remain unaffected by the disease (Figure 1 and Figure 2). However, whatever enables these rootstock seedlings to tolerate the HLB pathogen (*Candidatus* Liberibacter asiaticus, CLas) without succumbing to disease does not appear to be sufficiently transferred to a grafted tree to induce the desired degree of tolerance.

MECHANISMS ASSOCIATED WITH HLB TOLERANCE

What exactly makes these rootstocks HLB tolerant and what are the mechanisms? And how does this help us in the fight against HLB? Among the mechanisms that may be associated with HLB tolerance are the ability to produce compounds independent to or in response to infection that are harmful to the pathogen, the ability to produce defense regulators which enhance the tree's natural immune response against the pathogen, or the absence of specific nutrients or other substances that are important for pathogen growth and survival.

Studies on rootstock seedlings responding differently to HLB are in progress to decipher the biochemical composition of rootstocks. Initial results demonstrate clear differences based on rootstock type. More studies are underway to identify compounds that may be related to HLB tolerance and other rootstock traits.

Unfortunately, thus far no rootstock was shown to completely eliminate HLB-induced damage in a grafted tree. This indicates that potential antibacterial compounds or defense regulators either do not move in sufficient quantities past the graft union, or are only produced in

Figure 1. Cleopatra (a) and US-897 (b) rootstock seedlings. The red ovals indicate CLas-infected plants confirmed by PCR. US-897 seedlings are not affected by HLB, whereas Cleopatra seedlings are stunted and chlorotic. Healthy control plants are on the left in each photo for comparison.

Figure 2. Row of US-897 and US-942 seedling trees at the U.S. Horticultural Research Laboratory farm in Fort Pierce, Florida. Trees remained healthy four years after becoming infected with CLas.

the photosynthesizing portion of the rootstock, which is removed upon grafting. Continued studies on different germplasm will allow identification of potentially important compounds or defense regulators that can then be developed for use in engineering both rootstocks and scions with HLB tolerance, or even resistance using gene editing and other new technologies.

The ability to uptake nutrients and water more efficiently is strongly associated with the root structure of a particular rootstock.

Besides antibacterial compounds and defense regulators, additional mechanisms inherent to specific rootstocks are likely responsible for the better performance of some trees under HLB pressure. These include the ability to more efficiently uptake nutrients and water, the ability to faster regenerate roots lost to HLB, the capacity to induce higher vigor or a combination of different mechanisms.

The ability to uptake nutrients and water more efficiently is strongly associated with the root structure of a particular rootstock. Preliminary studies comparing root systems of greenhouse-grown rootstock liners clearly show differences in important root traits between different cultivars, such as the specific root length (the ratio of root length to root mass), which is generally considered to be an indicator of nutrient- and water-uptake efficiency. Among the commercially available rootstocks investigated thus far, US-942 was shown to have the highest ratio.

Researchers demonstrated that HLB induces fibrous root loss soon after infection with *C*Las. It is therefore expected that higher vigor and the ability of a rootstock to regenerate roots faster is of considerable importance for tree resilience.

Field trials conducted in Saint Lucie County during the early years

A Glimpse at CRDF Programs and Planning

By Harold Browning

As we start the new fiscal year at the Citrus Research and Development Foundation (CRDF), there are a number of priority activities in place to focus effort and resources on important tools for managing citrus in the presence of HLB. Here are some highlights.

Currently, CRDF is sponsoring 86 projects that combine the traditional research efforts overseen by the Research Management Committee of citrus growers and the Commercial Product Delivery Committee (CPDC), a diverse group devoted to moving research and development results to the field. The current portfolio of projects represent a portion of the more than 400 projects that have been conducted through CRDF oversight and funding since the organization began.

Project managers and the committees mentioned above are working to reduce the focus to the most critical projects that can contribute to short-term management tools for growers in the 2- to 3-year timeframe, while narrowing the longer-term research to just a few key topics, such as HLB-resistant plants. This is made possible by the commitment of federal funds to support longer-term research that can provide novel approaches to HLB management.

We encourage you to browse the CRDF website (citrusrdf.org), and in particular the "CPDC Project Managers Quarterly Reports," on the lower left of the home page under "CRDF Reports." Here you can learn more about the progress being made on topics such as bactericides, thermal therapy, HLB resistance, Asian citrus psyllid management and more. The next quarterly report will be posted following the August board meeting. Individual research project progress reports remain available through the search function highlighted under the "Growers" tab on the website.

External review of the CRDF and other U.S. citrus research progress is being conducted by the National Academy of Sciences review panel. Feedback from this intensive review will provide additional direction and priorities.

CRDF is working to improve the coordination of the many projects being conducted by a wide range of scientists, and our efforts include how to improve communication to growers and other interested parties on progress with this difficult disease challenge. Combined technical reports and a grower panel at Florida Citrus Mutual's annual industry conference in June offered an update on progress on some topics. The Citrus Expo coming in mid-August will again highlight progress in the battle against HLB, with a number of update presentations highlighting work sponsored by CRDF. Research will be presented by University of Florida/Institute of Food and Agricultural Sciences as a means to share with growers the best available information.

Finally, CRDF continues to receive and respond to a wide range of solver communications that offer varying levels of evidence for technologies or products that have been developed or are in testing. Through our project managers and then through the CPDC, CRDF is responding to these inquiries to ensure that all possible solutions are given consideration.

We invite you to attend our committee and board meetings to learn more. They are public meetings, and the schedule is posted on our webpage.

Harold Browning is Chief Operations Officer of CRDF. The foundation is charged with funding citrus research and getting the results of that research to use in the grove.

CRDF Grower Dollars at work for you

Column sponsored by the Citrus Research and Development Foundation

Table 1. Trunk cross sectional area (TCA), canopy height and canopy volume of Valencia trees were measured in 2015 on 17 rootstocks in a Polk County trial. Total soluble solids (TSS) and cumulative yield were measured from 2012 to 2015. Different letters indicate statistically significant ($p < 0.05$) differences within columns.

Rootstock	Scion TCA 2015 (cm^2)	Canopy height 2015 (m)	Canopy volume 2015 (m^3)	TSS 2012–15 (%)	Cumulative yield 2012–15 (kg/tree)
US-942	80.6 b-d	2.67 a-d	6.23 a-e	9.73 a-d	227 a
US-1516	86.7 a-c	2.63 a-e	6.28 a-d	9.41 b-e	211 ab
US-896	64.0 e-g	2.39 e	5.09 d-g	9.97 ab	205 a-c
US-1503	89.2 ab	2.71 a-c	6.47 ab	9.60 a-e	197 a-c
US-1524	80.8 b-d	2.66 a-d	6.08 b-e	9.59 a-e	193 a-c
Swingle	6.56 d-g	2.42 de	4.95 e-g	9.19 de	180 a-d
US-802	97.3 a	2.84 a	7.44 a	9.55 a-e	176 b-d
Carrizo	79.3 b-d	2.46 c-e	5.35 b-f	9.74 a-d	168 b-d
US-852	76.9 b-e	2.49 c-e	5.00 d-g	9.72 a-d	167 b-d
Kuharske	89.2 ab	2.56 b-e	5.35 b-f	9.69 a-e	166 b-d
US-809	60.2 fg	2.40 ef	4.47 fg	9.61 a-e	164 b-d
US-812	81.0 b-d	2.67 a-d	6.41 a-c	9.35 c-e	162 b-d
Cleopatra	98.6 a	2.44 de	5.53 b-f	9.15 e	158 cd
US-827	77.4 b-e	2.75 ab	6.20 b-e	9.50 a-e	141 d
US-897	53.7 g	2.17 f	3.92 g	9.81 a-c	137 d
Kinkoji	86.7 a-c	2.49 c-e	5.13 c-g	9.64 a-e	130 d
US-801	71.8 c-f	2.53 b-e	5.10 d-g	9.97 ab	128 d

after discovery of HLB in Florida showed that sweet orange trees on the high vigor-inducing rootstock US-802 exhibited less canopy damage and were among the most productive trees. Trees on Volkamer lemon, which also induces large tree size, experienced the least canopy damage in the youngest plantings. This suggests that rootstocks with high vigor and the ability to maintain a healthy root system can better cope with the damaging effects of the disease. However, the potential positive impact of high vigor-inducing rootstocks must be carefully considered if other factors such as fruit quality are of importance.

ROOTSTOCKS WITH BETTER PERFORMANCE

Despite inconsistent observations on rootstock performance in some field trials, other field experiments have clearly shown that trees on some rootstock varieties perform better than trees on other rootstocks. A recent U.S. Department of Agriculture (USDA) field trial conducted in Polk County in collaboration with Wheeler Farms showed considerable differences in productivity of Valencia trees on different rootstocks during the first four years of production (2012–2015). The trial included 17 different rootstocks, most of them commercially available, and was planted in 2008.

Although no significant differences were observed in bacterial titer levels of trees on the different rootstocks, notable differences were found among rootstocks in tree size, fruit quality and yield. As shown in Table 1, trees on US-942 and US-1516 were significantly more productive than trees on common commercial rootstocks such as Carrizo, Kuharske and Cleopatra. US-1516, a hybrid of African pummelo and Flying Dragon trifoliate orange, was released in 2015 by USDA and is now commercially available.

Another collaborative rootstock trial with Barron Collier in Collier County, planted in 2002, found US-802 and US-942 among the most productive rootstocks. Other rootstocks included in this trial were Cleopatra, Carrizo, Swingle, Kinkoji, US-812 and US-897.

Interestingly, fruit drop counts conducted during the 2015–16 and 2016–2017 seasons in the same two trials suggest that the degree of HLB-induced fruit drop may also be influenced by rootstock. Trees grown on US-1516 and US-942 (Polk County trial) and on US-802 (Collier County trial) exhibited considerably less fruit drop compared with trees on other rootstocks, specifically Swingle.

Several new rootstock varieties with good commercial performance under high HLB pressure have been released by USDA in 2014, and many of the new University of Florida (UF) rootstocks are currently being evaluated via the Fast Track/New Varieties Development & Management Corporation program. Many other field trials combining new rootstock selections from both the UF and the USDA breeding programs are in the early stages and will be closely monitored in the upcoming years in a collaborative effort of researchers from

both programs. For more information on these trials, see the article by Jude Grosser and Fred Gmitter on page 14.

CONCLUSION

Until new rootstocks are identified that induce the desired level of HLB tolerance to a grafted commercial tree, rootstock tolerance should be defined as the ability to allow a tree to remain commercially productive despite high HLB pressure, independent of its mechanism. When planting new groves, rootstocks should always be chosen very wisely as their performances are strongly influenced by factors other than HLB, especially soil type, soil pH and soil-borne pests and diseases.

Equally important are practices directed at maintaining a healthy root system. These include proper irrigation, nutrition and other root-health improvement strategies. Even a rootstock that is able to induce superior levels of tolerance to HLB will not thrive in the presence of unfavorable conditions unrelated to the disease.

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For further rootstock reading:

- http://swfrec.ifas.ufl.edu/hlb/database/pdf/2_Castle_15.pdf
- http://swfrec.ifas.ufl.edu/hlb/database/pdf/23_Tolerance Trifoliate_11.pdf
- http://swfrec.ifas.ufl.edu/hlb/database/pdf/00003019.pdf
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