CITRUS ADVANCED TECHNOLOGY PROGRAM

QUARTERLY & FINAL REPORTS: Control of Citrus Greening, Canker & Emerging Diseases of Citrus

SELECT PERIOD September 2014

(●) *Quarterly Report* ( ) *Final*

Proposal Title

Development of Promising New Scions for Florida Citrus: Exploiting HLB Resistance and Tolerance

Today's Date	Sponsoring Organization (drop-down)	Category (drop down)
10/14/14	Citrus Research and Development Foundation	Conventional Citrus Breeding

REPORT UPDATE (500 words-It is not necessary in this public report to disclose your institution's proprietary information or intellectual property.)

Evaluation of existing standard and non-standard cultivars ('Hamlin', 'Temple', 'Fallglo', 'Sugar Belle', 'Tango', and 'Ruby Red') for HLB resistance/tolerance is complete. In August 2010, the plants were established at Pico's farm in Ft. Pierce Fl. Data on the growth rate, disease severity, and Candidatus Liberibacter asiaticus (CLas) titer levels have been collected since April 2012. All trees exhibited symptoms of HLB and tested positive for CLas. During the 4-year period, there were significant differences in disease severity, stem diameter, and CLas levels among the varieties. 'Fallglo' had the lowest incidence of HLB symptoms, whereas 'Ruby Red' had the highest incidence. 'Ruby Red' also appears to be in significant decline. The highest CLas titer levels were observed in November, December, and January with 'Sugar Belle' and 'Tango' had the highest titer levels while 'Fallglo' and 'Temple' had the lowest. Despite the high titer levels found in 'SugarBelle', it had the greatest overall increase in diameter and was the healthiest in overall appearance. These results indicate that compared to 'Hamlin', 'Fallglo' and 'Temple' appear to display field resistance to HLB while 'SugarBelle' appears to have significant tolerance. Imidacloprid was applied quarterly to a subset of trees and significantly increased stem diameter compared to the non-treated trees but did not have a significant effect on tree height, disease severity, or CLas titer levels.

Progress has been made on the antibiotic treatment of HLB infected bud-wood. Bud-wood of nine HLB symptomatic varieties, 3 fairly resistant ('Temple', GnarlyGlo', and 'Nova') 3 tolerant ('Jackson', FF-5-51-2, and Ftp 6-17-48), and 3 susceptible ('Flame', 'Valencia', and 'Murcott'). In November 2013 and May 2014, HLB positive bud-wood was treated with various concentrations of penicillin and streptomycin and grafted on sour orange rootstock. In August 2014, standard growth measurements (stem diameter and height), disease severity were evaluated and leaves were sampled for qPCR analysis. Evaluations and sampling will continue on quarterly basis.

Development of periclinal chimeras with resistant vascular tissue from Poncirus and remaining layers from sweet orange is currently underway. One hundred and fifty etiolated seedlings of the trifoliate 'Rubidoux' and the sweet orange 'Hamlin' have been approach grafted together. Generation of new chimeras has been difficult. Several adventitious buds have emerged from the treated graft region, and one appears to be a chimera. The newly emerged plants will be tested using LC/MS to determine the origin of the three layers. To increase the success rate, additional plants will be grafted over the next twelve months.

In October 2013, 34 unique genotypes (USDA hybrids) some of which appear to have tolerance to HLB, and 16 standard commercial varieties were exposed to an ACP no-choice feeding trial and have been transferred to the field at Ft. Pierce Fl. Standard growth measurements and disease ratings were initiated in July 2014 and will continue on a monthly basis. In September 2014 there were significant differences in trunk diameter ($p<0.0001$). At this time there were no significant differences in disease symptoms. However, it is still too early to assess for HLB resistance/tolerance. Three leaves were randomly samples and CLas titer levels will be quantified using qPCR.

LC-MS assessment of potential HLB resistant biomarkers in Citrus and Citrus relatives is being explored.

A method for the rapid identification of potential sources of HLB resistance is also being developed. This project involves the screening of citrus seedlings at the 3 to 5 leaf stage, or very small micrografted trees, that are exposed to HLB infect ACP feeding. CLas titer levels, using real time PCR, are evaluated at 3, 6, and 9 weeks Seedlings of Hamlin and Dancy show early CLas proliferation and systemic movement. Only very low levels of CLas have been observed in Carrizo.

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