CITRUS ADVANCED TECHNOLOGY PROGRAM

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

SELECT PERIOD March 2016

(●) *Quarterly Report* ( ) *Final*

Proposal Title

Metabolomic profiling to accelerate development of HLB tolerant rootstocks

Today's Date	Sponsoring Organization (drop-down)	Category (drop down)
4/15/2016	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.)

Good progress was made on work to identify metabolite profiles associated with tolerance to HLB and other stresses in advanced rootstock selections, and validate the effectiveness of these metabolite profiles for selection by comparison of existing rootstock selections within the USDA program. The work will focus on 12 rootstocks where previous studies have identified relative differences in tolerance to Las infection. Specific studies were continued to identify key metabolic compounds and collect the first stage of information to be used in the validation process.

In this quarter, leaf and root samples from one greenhouse test with different rootstocks and three field tests with different rootstocks were processed and shipped to the West Coast Metabolomic Center for metabolomic analysis by gas chromatography-time of flight (GC-TOF) mass spectrometry (MS). The GC-TOF-MS analysis for the first cycle of samples is expected to be completed in the next quarter and results available for detailed analysis by our research team in Ft. Pierce and Immokalee.

In preparation for the second cycle of sample collection in the next quarter (first year), clean seedling and budded trees were prepared in the greenhouse, and samples were collected and analyzed for PCR verification of Las and CTV infection status for trees in two different field trials. The experimental design of the first and second cycle of samples collected in the first year will allow comparison of metabolite profiles of the same genotypes in different seasons, providing critical information about how metabolite profiles change over the growing season.

In preparation for sample collection in the second year of the project, clean seedlings and budded trees were prepared for greenhouse studies, and clean seedlings and budded trees were prepared for field planting. At the field site, trees were removed and land prepared for planting of the new trees being used in the study. Material was prepared for controlled Las inoculation of trees to be studied in the greenhouse and the field.

The project was revised to include a new researcher at University of Florida in Immokalee, Dr. Ute Albrecht, who has experience and special expertise in this research field. Grant funds will be provided through a Non-Assistance Cooperative Agreement to support this work in Immokalee. The project changes actually reduce the overall cost of the project, but will significantly increase productivity and the opportunity for success.

In this quarter, a manuscript on a preliminary metabolomic study by our team was submitted for publication. That study used GC-TOF-MS analysis to compare metabolite profiles of six rootstock cultivars infected and not infected with Las. The study identified numerous chemically unknown compounds that appeared strongly associated with Las tolerance and offer good opportunities for further study and manipulation of the tolerance behavior. The study demonstrated striking metabolic differences between the HLB-sensitive rootstock Cleopatra and the HLB-tolerant rootstocks US-897 and US-942, both with and without Las infection, and will serve as a foundation for continuing work under this grant.

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