Florida Citrus Advanced Technology Program

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

Instructions Complete the fields based on your project specs. When finished, save the form to your local disk using a unique name. Then, go to **http://research.fcprac.com**, and log in with your user name and password using Researcher Login in the lower left. Find this project title and click on **Submit a Report**. Update your profile information if needed, then upload this report as directed.

2009-2010 REPORT	CATEGORY (drop-down)	TODAY'S DATE (m/d/yr)
○ *Quarterly Report* ◉ *Annual Report* ○ *Final*	Genetics	3/19/10

WHAT IS THE "HEADLINE" FOR THIS REPORT (e.g. a one-sentence "newspaper headline" describing what you accomplished)

Disease Resistance Vigorously Pursued, While Improved New Varieties Are Released to Florida Growers

TITLE and CONTACT INFORMATION

Proposal Title Surviving HLB and canker: genetic strategies for improved scion and rootstock varieties

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Phone (863) 956-1151	*Project Duration (years)* 3 *Year of Project* 1
Organization UF-CREC	*Total Direct Funds (current year)* 840,045.60

REPORT UPDATE (650 words; provide details about your headline)

Transgenic citrus trees (900 containing 15 potential HLB/canker resistant constructs) were planted at 2 locations with severe pressure. At least 1500 transformations with various new constructs and promoters, mostly using sweet orange selections, have been produced for greenhouse disease tests; many of these will be planted in fall 2010. Our transgenic field trial in Gainesville, testing disease resistance transgenes, survived the brutal winter. We have verified 4 tissue-specific promoters that target gene expression only to phloem. Testing of previously produced transgenics indicates several thus far with delayed, reduced or no symptoms following HLB-inoculation. Transgenic Carrizo plants were produced with insecticidal genes; preliminary feeding tests showed one gene tested killed aphids and psyllids. Several transgenic and hybrid plants have been identified with tolerance to canker. New disease resistance genes were cloned from grape and tested in tobacco for efficacy; preliminary results identified two potential candidates for citrus. A newly developed grape-derived anthocyanin marker allows visual selection of transformants, and will be tested in citrus, an alternative to non- plant markers used in most transformations. Experiments on transient expression of gene constructs are underway; when optimized, this will allow more rapid testing of promising disease resistance genes. Two microarray platforms (our Agilent chip containing disease resistant genes and the Affymetrix Citrus GeneChip) are being used to analyze citrus gene expression over time in response to canker and HLB. Comparisons of sugar and starch metabolism in HLB-infected and healthy plants revealed that starch, sucrose, and glucose accumulated in infected leaves, maltose decreased and fructose levels were unchanged; studies are underway comparing activity of cell-wall bound invertase, a critical enzyme involved in sucrose metabolism and plant defenses. Using the iTRAQ technique, proteomic analysis was used to compare healthy and HLB-diseased mature leaves of sweet orange; 19 proteins were differentially expressed, out of which 9 proteins involved in stress and defense responses were highly up-regulated. Previously, microarray experiments highlighted canker defensive genes in kumquat; real-time PCR confirmed roles of one kumquat R-gene, two kinases, and one transcription factor, new targets for transformation. Cybrid grapefruit plants were produced with kumquat, and preliminary results show that some behave like kumquat in challenges. New DNA samples from a genetic population are being genotyped to contribute to the ICGC sequencing project. Hybrid plants have been produced for rootstock improvement from the previous season, and new crosses made 2009 are growing off. Preliminary results of an experiment to study experimental rootstock effects on the growth of HLB-infected budwood revealed significant differences in disease expression and growth. Previous work to develop rootstocks against other maladies (CTV, blight, Phytophthora, Diaprepes, etc.) continues, as we collected data from replicated trials and plantings. 15,000 trees were propagated from > 100 advanced UF-CREC rootstock selections, for several planting density/open hydroponic system trials located throughout Florida, to evaluate their potential in closely-spaced groves under intensive cultural management. > 200 candidate rootstock hybrid seedlings were selected following a screen for Phytophthora tolerance /hi-pH soil adaptation. New pummelo-grapefruit seedless hybrids were created, and others available were found to be more tolerant of canker than grapefruit. New grapefruit-type candidates have been found this season, with selection based on fruit quality attributes. Sugar Belle, UF's 1st fresh release, was licensed to NVDMC, and the first crop was successfully marketed. Six new seedling orange selections were released, as unpatented and unlicensed varieties; 5 are early maturing with exceptional quality, and one with reported canker tolerance. Our first UF-bred orange cultivars, Valquarius (SF14W-62) and Valenfresh (N7-3), are being licensed, to provide 6-8 week earlier and later maturity.