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*	HLB Pathology	2/12/10

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

Understand the HLB disease development by studying the gene expression of different citrus varieties at different conditions

TITLE and CONTACT INFORMATION

Proposal Title Characterization of the virulence mechanism of the citrus Huanglongbing pathogen Candidatus Liberibacter asiaticus

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Organization Citrus Research and Education Center, UF	*Total Direct Funds (current year)* 160000

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

The goal of the proposed research is to understand how Candidatus Liberibacter asiaticus causes Huanglongbing (HLB) disease and how tolerant plant resists Ca. L. asiaticus infection. Citrus HLB is the most devastating disease on citrus. There are very few options for management of the disease due to the lack of understanding of the pathogen and citrus interaction. Understanding the citrus and citrus HLB pathogen interaction is needed in order to provide knowledge to develop sustainable and economically viable control measures.

Major achievements: 1. One refereed journal article was published. Kim et al. 2009 Response of sweet orange (Citrus sinensis) to 'Candidatus Liberibacter asiaticus' infection: microscopy and microarray analyses. Phytopathology. 99:50-7. Investigation of the host response was examined with citrus microarray. The microarray analysis indicated that HLB infection significantly affected expression of 624 genes related to plant pathogenesis/stress, anthocyanin biosynthesis, cell wall metabolism, cell division, detoxification, lipid metabolism, metabolite transport, metal transport, nucleotide metabolism,phenylpropanoid / flavonoid / terpenoid metabolism, phytohormones, protein kinase, protein metabolism, protein-protein interaction, signal transduction, sugar metabolism, transcription/translation factors and unknown/hypothetical genes. The anatomical analyses indicated that HLB bacterium infection caused phloem disruption, sucrose accumulation, and plugged sieve pores. The up-regulation of three key starch biosynthetic genes including ADP-glucose pyrophosphorylase, starch synthase, granule-bound starch synthase and starch debranching enzyme likely contributed to accumulation of starch in HLB affected leaves. The HLB-associated phloem blockage resulted from the plugged sieve pores rather than the HLB bacterial aggregates since 'Ca. Liberibacter asiaticus' does not form aggregate in citrus. The upregulation of pp2 gene is related to callose deposition to plug the sieve pores in HLB-affected plants. The cDNA sequence of PP2 has been requested by four different research groups to use as the target to suppress the HLB symptom development. 2. Leaf samples from different varieties including grapefruit, Murcott, and Hamlin were collected from Florida citrus groves. Both healthy and infected trees were sampled. It is expected gene expression of those trees in the citrus grove will reveal more information when compare with the gene expression profile in greenhouse. Those samples will be used for gene expression analysis using microarray or SSH approaches. 3. Gene expression of Valencia leaf samples (healthy vs infected) in citrus grove are being conducted. Currently, two biological replicates were included in the preliminary test. Further microarray analysis is needed. 4. To further expand our current understanding of Ca. L. asiaticus-host interaction, we are currently comparing two susceptible and two resistant/tolerant cultivars grown in greenhouse. The plants have been inoculated in greenhouse. Finding key genes involved in HLB symptom development will reveal potential management strategy and lead to innovative research to control HLB.

Research plan 1.1 Host response in greenhouse condition. Both Carrizo citrange and Poncirus trifoliata exhibited tolerance in the initial test. However, the inoculation has been inconsistent. The tolerance of Poncirus trifoliata to HLB is inconclusive. Thus host response of those varieties to Ca. L. asiaticus infection could not be done. Instead, we have adjusted our plan based on the most recent data on pathogenicity assays of different varieties to Ca. L. asiaticus. Both Persian lime and Eureka lemon are tolerant to HLB. 1.2 Host response in field condition. Due to the differences in greenhouse and field conditions, the disease development in greenhouse and field is quite different. We would to focus on sweet orange. we will use the following samples: leaf, stem, and root.