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	7/15/09

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

Extirpation of 1000 psyllid alimentary canals from infective adult psyllids and initiation of cDNA library construction

TITLE and CONTACT INFORMATION

Proposal Title The citrus psyllid transcriptome and time course differential gene expression in Ca. Liberibacter-infected/free whole psyllids and organs

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Organization The University of Arizona; University of Florida	*Total Direct Funds (current year)* $164,999

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

The HLB pathogen is acquired by the Asian citrus psyllid during the feeding process, which indicates that the alimentary canal is the first organ of the transmission cycle.

Our first task in this collaboration has been completed by stockpiling this organ for mRNA analysis. 1000 adult, noninfective psyllids were submerged, one at a time, in 50% RNAse Later® (Ambion), a general protease that destroys transcript degrading enzymes.

The alimentary canals of each were extirpated in this solution and transferred with a pinpoint to 0.5ml of 100% TRIzol® (Invitrogen), a powerful phenolic that allows for deep freezing and long-term storage. The same low volume of phenolic and high number of guts has been shown with whiteflies to yield an outstanding 28 nanograms mRNA per microliter final concentration.

As an aside, alimentary canals were dissected and pooled from two adult psyllids confined to an infective orchard tree branch for three days, to see if a qPCR signal could be had in so small a sample. Results were a Ct value of 35, where 40 is no signal and 20 is a high-titer signal.

This first test run indicates that quantification of titer in individual guts is a highly feasible approach to quality control when stockpiling of organs from infective psyllids commences.

The laboratory rearing system is under construction and excellent progress has been made once psyllids began reproducing at high levels (June onward) and some adults and immatures, as well as samples from different portions of the plants on which colonies are being reared have been collected for qPCR analysis (in progress).