CITRUS ADVANCED TECHNOLOGY PROGRAM

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

▶ INSTRUCTIONS

(●) *Quarterly Report* ( ) *Annual Report* ( ) *Final*

Required: What is the "headline" for this report (e.g. a one-sentence "newspaper headline" describing what you accomplished)

Controlling HLB spread by using RNAi to control psyllids

Proposal Title

RNAi Innocentive Project Extension (Phase II): In Planta

Today's Date	Sponsoring Organization (drop-down)	Category (drop down)
10/15/15	Citrus Research and Development Foundation	Vector Management

REPORT UPDATE (500 words; summarize your accomplishments)

This is a project to develop novel approaches to controlling psyllids . Effective techniques to reduce the rate of Huanglongbing (HLB) spread are key to slowing its incidence, especially for new citrus plantings. RNA-interference (RNAi) is a natural regulatory and anti-viral response in eukaryotes and can be manipulated to target mRNAs/gene expression, including to control insects. Our on-going collaboration has found that RNAi inducers, expressed in citrus trees using the Citrus tristeza virus (CTV) vector, reduce the survival of adult Diaphorina citri moving onto the trees, and greatly reduce their reproduction and acquisition of Candidatus Liberibacter asiaticus by progeny. Our goal is to further improve RNAi activity such that it will help to manage D. citri and HLB, allow reduction in pesticide use and lower grower costs for U.S. citrus.

Sequences of specific psyllid genes that are thought to be needed for the survival and reproduction of psyllids are cloned into the CTV vector. As the virus replicates in phloem cells, it produces large amounts of dsRNA intermediates that now also produces dsRNAs containing psyllid sequences. The normal plants RNAi defense mechanism processes the dsRNAs into small 21 nt siRNAs that target mRNA degradation. These siRNAs migrate from virus infected cells into the sieve element. As the psyllid feeds, it sucks up these siRNAs that now target the psyllid mRNAs and prevents the psyllid from making this protein. The lack of this protein has detrimental effects on the survival, reproduction, and CLas acquisition of psyllids.

So far, we have seen reduction in survival of adult psyllids placed on RNAi expressing plants, but the effects on reproduction of the new generation of psyllids has been much greater. This is likely because the nymphs are rapidly growing an need lots of new protein synthesis and because they uptake large amounts of phloem sap.

We had a technical glitch that slowed us down for a couple of months, but we are testing about twenty different anti-psyllid sequences to identify the most efficacious sequence.

We recently have found that the RNAi target sequences that are expressed from near the 3' terminus of the CTV vector appear to not be stable enough to be useful for controlling HLB spread in the field. We are recloning all of the target sequences between p13 and p20 or between CPm and CP looking for an optimal compromise between efficacy and stability.

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Organization UF, IFAS, CREC

Contract Number 00089625

Project Duration (years) 1 *Year of Project* 1

Total Funds (current year) $80,183.00

SUBMIT REPORT

CITRUS RESEARCH AND DEVELOPMENT FOUNDATION, INC.