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)

Progress is being made on clean-up and transformation of mat

Proposal Title

Agrobacterium-mediated Genetic Transformation of Mature Citrus Tissue

Today's Date	Sponsoring Organization (drop-down)	Category (drop down)
1/19/11	Citrus Research and Development Foundation	Transgenic/Metagenomic Mediation of Resistance

REPORT UPDATE (500 words; summarize your accomplishments)

Researchers at the USDA Ft. Pierce: 1. Source of mature tissue. Four populations of adult phase trees were maintained in the greenhouse including Valencia sweet orange/Sun Chu Sha (73 trees), Ruby Red grapefruit/US812 (62 trees), US-942 citrange rootstock/Cleo (32 trees), Calamondin (31 trees), and Etrog Arizona 861-S1 citron (67 trees). In vitro bud emergence and growth manuscript accepted for publication. A manuscript was submitted to the journal Plant Cell, Tissue and Organ Culture that documents the system developed for producing in vitro adult phase shoots from cultured nodes of greenhouse trees. Shoot regeneration from mature tissue explants. A system was developed for the production of shoots from cultured internodes from greenhouse trees. The system results in shoot and bud formation in 70-90% of the explants. A manuscript is in preparation that documents this research. Agrobacterium-mediated transformation of mature tissue explants. Transformation of mature internode explants from greenhouse trees has been demonstrated in grapefruit (1 plant), US-942 (4 plants), and Etrog citron (4 plants) using the beta-glucuronidase reporter gene. Current efforts are now directed toward identifying the factors important for a system of sufficient efficiency for routine transgenic plant production. New tissue culture method of Agrobacterium-mediated transformation of tissue explants. Preliminary results using alternative culture methods suggest improved transformation efficiencies. These approaches will be further explored. At the CREC in the Gmitter lab, work is continuing on the use of Thin Cell Layers (TCLs) as explants for mature tissue transformation. Experiments have been done to induce regeneration in the TCLs by manipulating the amount of growth regulators, carbon source and also by pre-treating the TCLs with BA but regeneration is still problematic from these explants. In the Grosser lab, research confirmed the work of others showing that the flush used to generate transformation explants was critical. In the Machado laboratory in Brazil, research on the regeneration ability of various sweet oranges continues. These differences are present in both mature and juvenile tissues. In the Moore laboratory in Gainesville, experiments still are focused on using small peptides as vehicles to deliver cargos to plant tissues. If these techniques could be worked out they would have a number of applications for citrus transformation, perhaps even eventually allowing the transfer of genes or gene products to existing trees. A transient transformation expression system has also been developed using citrus leaves.

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Organization University of Florida

Contract Number NAS144

Project Duration (years) 3 *Year of Project* 2

Total Funds (current year) $224,000.00

SUBMIT REPORT

CITRUS RESEARCH AND DEVELOPMENT FOUNDATION, INC.