

CALIFORNIA STATE SCIENCE FAIR 2017 PROJECT SUMMARY

Name(s)

Yusuf Ahmad

Project Number

S1601

Project Title

Citrus in Jeopardy: Phase I, Determination of Presence and Typology of CTV Using ELISA, DTBIA, PCR, and Real-Time PCR

Abstract

Objectives/Goals The purpose of this experiment is to ascertain the validity of the aforementioned lab techniques in assessing the presence as well as the typology of Citrus Tristeza Virus strains among suspect plants.

Methods/Materials

Plant tissue gathered from 8 suspect samples along with a healthy control were homogenized and used throughout the experiments. ELISA used antibodies to capture viral antigen particles and substrate used for measurement of virus concentrations in a spectrophotometer to determine virus presence. The nitrocellulose membrane in DTBIA was treated with antibody and presence of virus was determined through a light microscope. PCR used the virus' coat protein cDNA (generated through a thermocycler machine) in gel electrophoresis with banding patterns used to indicate the presence of the virus. Real-time PCR consisted of the use of various probes to detect the presence of virus as well as to characterize the strain of the virus using the BioRad Lightcycler software to actively measure PCR.

Results

ELISA, DTBIA, PCR, and Real-Time PCR all showed matching results between plants tested positive for CTV in those infected with Real-Time PCR also characterizing different strains of the virus.

Conclusions/Discussion

ELISA, DTBIA, PCR, and Real-Time PCR were indicated to reliably test for CTV in infected plants with Real-Time PCR additionally characterizing the strain of the virus. Mild virus strains which were detected to be present in certain suspect plants using ELISA, DTBIA, and PCR were differentiated from harmful strains of the CTV virus using Real-Time PCR.

Summary Statement

ELISA, DTBIA, PCR, and Real-Time PCR all detected presence of the CTV virus with Real-Time PCR also differentiating between harmful and harmless strains of the virus.

Help Received

The procedure for all the aforementioned techniques were given by the USDA laboratory with lab equipment at the facility also being used. All the experiments were conducted under the supervision of Dr. Yokomi, Dr. Seleveraj, and Dr. Maheshwari.