



## **Assay of Citrus Seed for Viable *Xanthomonas citri* pv. *citri* (Xcc) after Extraction from Fruit Affected by Citrus Canker**

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**ABSTRACT.** Trees for production of seed of rootstocks for citrus nursery propagations are typically located in blocks planted outdoors and are unprotected from pathogen and pest infestations. A seed-tree block in Dundee, FL, experienced an initial incursion of citrus canker in July 2009. By Sept. 2009, canker had spread in the block from south to north in the tops of Swingle citrumelo and Carrizo citrange trees. The disease distribution suggested that the Xcc inoculum was introduced in a single windblown rain event. This case illustrates the high risk for outdoor seed-tree blocks in Florida nurseries to become infected with canker. The purpose of this study was to provide evidence that the seed extraction and disinfection process for treating against fungi utilizing hot water, fungicide, and bleach treatments is effective for eliminating Xcc from seed harvested from canker-infected fruit. The Carrizo citrange fruit used for seed extraction in this experiment exhibited moderate to severe canker symptoms. Seed extracted from the fruit were subjected to the standard hot water/fungicide treatment and then were assayed for the presence of Xcc. To validate the assay process, seed were soaked in Xcc inoculum at 10<sup>4</sup> colony forming units per milliter; then they were washed, and the wash solution infiltrated into grapefruit leaves for detection of viable bacteria. The presence of canker lesions in the grapefruit confirmed that the recovery and bioassay methods were effective for detection of viable Xcc in seed washes. Treatments of seed extracted from canker-infected fruit with hot water at 125 °F for 10 min and 8-hydroxyquinoline sulfate, with or without 10% bleach in the wash step nullified recovery of viable Xcc. This was expected since each of the disinfectants has bactericidal activity. In conclusion, the protocol for extracting and disinfecting citrus seed was effective for Xcc disinfection of seed from canker-infected fruit.

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