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Visualization of ‘*Candidatus Liberibacter asiaticus*’ Cells in Citrus Seed Coats with Fluorescence In Situ Hybridization and Transmission Electron Microscopy

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‘*Candidatus Liberibacter asiaticus*’ is bacterium implicated as the causal agent of the economically damaging disease of citrus called huanglongbing (HLB). The bacterium is spread by movement of infected citrus propagation material and by the Asian citrus psyllid, *Diaphorina citri* Kuwayama. Seed transmission is a possible additional route of dissemination for the pathogen. Some published studies on seed transmission found abundant bacterial DNA in seed coats but no infections of germinated seedlings. No direct observations of bacteria in seed coats were made in these studies. In vascular bundles from citrus seed coats, we confirmed the presence of bacterial cells with transmission electron microscopy (TEM) and Fluorescence In Situ Hybridization (FISH). The physical measurements and the morphology of individual bacterial cells revealed by TEM and FISH were consistent with those described in the literature for ‘*Ca. Liberibacter asiaticus*’. No bacterial cells were observed in seed coats from non-infected trees. A library of clones of prokaryote 16S rRNA gene sequences amplified from a non-infected tree contained no ‘*Ca. Liberibacter asiaticus*’ sequences, whereas 95% of the sequences in a similar library prepared from an infected tree were ‘*Ca. Liberibacter asiaticus*’, providing additional data that the bacterial cells observed by TEM and FISH in seed coats from infected trees were ‘*Ca. Liberibacter asiaticus*’.