4.3 Microbiome analysis of HLB pathogen infected citrus using Phylochips and 16S rDNA clone library sequencing

Sagaram U.S.1*, DeAngelis K.M.*, Trivedi P.1, Kim J-S.1, Andersen G.L., and Wang N.2

Citrus Research and Education Center, University of Florida, 700 Experiment Station Road, Lake Alfred, FL 33850, USA.
2 Lawrence Berkeley National Lab Earth Sciences Division, One Cyclotron Road, MS-90R1116, Berkeley, CA 94720 510-486-5246
* Those two authors contribute equally to this work.

*Candidatus* Liberibacter asiaticus is an endophytic bacterium of citrus that is phloem limited and is believed to cause citrus Huanglongbing (HLB) or citrus greening. Disease control has been a big challenge since little is known about the ecology of the pathogen and all the attempts to culture the pathogen remain unsuccessful. In this study we describe the bacterial diversity associated with citrus leaf midribs infected with *Candidatus* Liberibacter asiaticus. We employed a combination of high density phylogenic 16S rDNA microarray and 16S rDNA clone library to determine if differences exist in microbial community composition between symptomatic and asymptomatic citrus midribs. Analysis of 16S rRNA gene amplicons using phylochip arrays indicated that nine taxa were more abundant in symptomatic midribs compared to asymptomatic midribs. The taxa otu_7603, representing Liberibacter species, was detected at a very low level in asymptomatic plants, but over 200 times more abundant in symptomatic plants. Comparison of microarray with clone libraries disclosed successful detection and classification of most of the clone groups. The correspondence between phylochip analysis and 16S rDNA library suggests that the bacterial community data presented here is representative of predominant bacterial groups which account for a major portion of bacterial population of citrus. In addition, this study has shown that a comprehensive assessment of bacterial population of plants can be obtained using a combinatorial approach of phylochip and 16S rDNA clones analysis.