



Draft Genome Sequence of "Candidatus Liberibacter asiaticus" from California

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We report here the draft genome sequence of "*Candidatus* Liberibacter asiaticus" strain HHCA, collected from a lemon tree in California. The HHCA strain has a genome size of 1,150,620 bp, 36.5% G+C content, 1,119 predicted open reading frames, and 51 RNA genes.

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"Candidatus Liberibacter asiaticus" is an unculturable, phloemlimited, alphaproteobacterium associated with citrus Huanglongbing (HLB), a devastating citrus disease worldwide (1, 2). Although HLB was observed in southern China in the 1890s (3), the disease was not found in Florida until 2005 (4). Since then, "Ca. L. asiaticus" has been reported in several southern states of the United States. In 2012, "Ca. L. asiaticus" was detected in a single lemon tree in Hacienda Heights of Los Angeles County, California (5). The infected tree was quickly removed, and no other "Ca. L. asiaticus"-infected citrus trees have been found in California. A recent study showed that the California strain was similar to strains in the Asiatic group (6).

For the benefit of HLB research and control, this study sequenced the whole genome of the California strain of "Ca. L. asiaticus," designated strain HHCA. The procedure of Zheng, Deng, and Chen (7) was followed, except for omitting the periwinkle enrichment step. DNA extract of strain HHCA was provided by Plant Pest Diagnostics Center, California Department of Food and Agriculture, with the estimate of 175 pg of "Ca. L. asiaticus" DNA per μ l. Bacterial DNA was enriched using the NEB-Next microbiome DNA enrichment kit (New England BioLabs, Inc., Ipswich, MA) and enlarged through multiple displacement amplification using the REPLI-g minikit (Qiagen, Inc., Valencia, CA). Genome sequencing was carried out by two runs of Illumina MiSeq (Illumina, Inc., San Diego, CA). The first run generated 4.4×10^7 reads (mean, 242 bp), and the second run generated 3.4×10^7 reads (mean, 251 bp). The whole-genome sequences of strains psy62 (8), gxpsy (9), and A4 (7) were used as references to extract "Ca. L. asiaticus" reads using standalone BLAST software (10) and Perl scripts. A total of 48,362 "Ca. L. asiaticus" reads (mean, 243 bp) were identified. With the help of Velvet 1.2.10 (https://www.ebi.ac.uk/~zerbino/velvet/) (11) and CLC Genomics Workbench 7.0 software, 239 contigs (512 bp to 39,757 bp, with $\sim 10 \times$ coverage) were obtained. The draft HHCA genome comprises 1,150,620 bp, with a G+C content of 36.5%. Annotation was performed by the RAST server (http://rast.nmpdr.org/) (12). The HHCA genome was predicted to have 1,119 open reading frames and 51 RNA genes, and similarities are 94% to the Psy62 genome (8), 91% to the gxpsy genome (9), and 95% to the A4 genome (7).

Low "*Ca.* L. asiaticus" titers in the infected plant presented a significant challenge for sequencing the strain HHCA genome. This study performed two Illumina runs to improve sequence coverage. The first run had a coverage of ~68%, and the second run reached ~93%. More runs will further increase coverage and yet increase the sequencing cost. The draft genome sequence presented here is sufficient for the characterization of strain HHCA and will serve as a reference for studying HLB in California.

Nucleotide sequence accession numbers. This whole-genome shotgun project has been deposited at DDBJ/EMBL/GenBank under the accession no. JMIL00000000. The version described in this paper is version JMIL02000000.

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The mention of trade names or commercial products in this publication is solely for the purpose of providing specific information and does not imply recommendation or endorsement by the U.S. Department of Agriculture.

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