Complete Genome Sequence of *Pantoea stewartii* subsp. *indologenes* ZJ-FGZX1, a Lucky Bamboo Pathogen

Lei Ren, Song Zhang, Zhiying Xu, and Hanqiao Hu[†]

Department of Biotechnology, College of Coastal Agricultural Sciences of Guangdong Ocean University, Zhanjiang Guangdong, 524088, China

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Abstract

The phytopathogen *Pantoea stewartii* subsp. *indologenes* causes Stewart's wilt disease in lucky bamboo. Here, we report the complete genome of *P. stewartii* subsp. *indologenes* ZJ-FGZX1, which represents the first whole-genome sequence of an isolate from China. The assembled genome consisted of three contigs, with one circular chromosome of 4,550,072 bp and two circular plasmids of 326,337 and 106,454 bp. The complete genome will provide a valuable resource for further studies on bacterial blight worldwide.

Keywords

Dracaena sanderiana, genome, *P. stewartii* subsp. *indologenes*

Genome Announcement

Dracaena sanderiana, also called lucky bamboo, is one of the important export ornamental plants in southern China. Zhanjiang District, Guangdong Province, is one of the main areas to process and export lucky bamboo. *Pantoea stewartii* subsp. *indologenes* is a gram-negative gamma proteobacterium that causes Stewart's wilt disease in lucky bamboo, resulting in economic loss and quarantine of exports (Choi and Kim 2013; Zhang et al. 2020). *P. stewartii* subsp. *stewartii* is the causal agent of Stewart's wilt of sweet corn. Complete genome sequences have been generated for *P. stewartii* subsp. *stewartii* DC283 from a maize pathogen (Duong et al. 2017). There are two main virulence factors that contribute to Stewart's wilt disease caused by *P. stewartii* subsp. *stewartii*: the hypersensitive response and pathogenicity gene (*Hrp*) and exopolysaccharide (EPS) (Coplin et al. 1992a and b). A number of *hrp* genes in phytopathogenic bacteria have been characterized and most of them cluster on one chromosome (Huang et al. 2009). The complete genome of the *P. stewartii* subsp. *indologenes* strain ZJ-FGZX1 was sequenced and assembled using both Nanopore and Illumina reads to gain detailed insight into the genomic features of the strain. This genome information will certainly help researchers to further decipher the difference between *P. stewartii* subsp. *stewartii* and *P. stewartii* subsp. *indologenes* and will aid in exploring new management strategies for Stewart's wilt.

P. stewartii subsp. *indologenes* ZJ-FGZX1 was isolated from diseased lucky bamboo leaf obtained from Zhanjiang District, Guangdong Province, China, in 2017 (Zhang et al. 2020). A single colony of the strain was picked from a Luria Bertani (LB) plate and was cultured in liquid LB medium for genomic DNA extraction. The same colony was also picked as the template of direct colony PCR (Lu et al. 2012), and 16S ribosomal DNA was sequenced to identify the strain as *P. stewartia*. Genomic DNA of the strain was extracted using the Qiagen Genomic-tip. A ligation sequencing kit was used, with a randomly sheared 1 µg of DNA, to prepare the sequencing library for Nanopore sequencing. The library was loaded

[†]Corresponding author: H. Hu; huhanqiao@sina.com

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Table 1. Comparative accounts on genomic features of *Pantoea stewartii* subsp. *indologenes* ZJ-FGZX1 with *P. stewartii* subsp. *stewartii* DC283

Genomic features	<i>P. stewartii</i> subsp. <i>indologenes</i> ZJ-FGZX1	<i>P. stewartii</i> subsp. <i>stewartii</i> DC283
Accession number	NZ_CP049115.1	NZ_CP017581.1
Length (bp)	4,550,072	4,528,215
G+C content (%)	53.69	54.19
Total genes	4,699	5,417
Total protein-coding genes	4,586	5,318
Noncoding RNAs	12	6
Pseudogenes	78	437
Ribosomal RNA operon	7	7
Transfer RNA	79	72
Plasmids	2	11

onto a flow cell and was sequenced using a MinION device and the basecalling was accomplished via Guppy (v 3.2.13) integrated in MiniKNOW with default parameters. The Illumina library was prepared by fragmenting the genomic DNA of bacteria and ligating specialized adapters to both fragment ends. The genome of the bacteria was sequenced and assembled, using both Nanopore and Illumina reads (Madoui et al. 2015). A total of 617,083 reads were obtained after sequencing, with an N50 value of 9,427 bp and an average read length of 5,241 bp, consisting of 3,234,394,529 bases in total after Nanopore sequencing; 5,390,706 short reads consisting of 1,608,432,636 bp bases were obtained after Illumina sequencing. The reads of low quality and less than 2,000 bp were filtered out. A total of 2,889,464,526 bp (579.9-fold coverage), with N50 of 10,754 bp, high-quality reads were obtained after sequencing. Canu v1.8 was used to assemble the subreads after filtering and Pilon was used to correct the assembled sequences, using the second-generation sequence data (Koren et al. 2017; Walker et al. 2014). Chromosome and plasmids were circularized using Circlator 1.5.5 finally (Hunt et al. 2015). Automatic annotation for the *P. stewartii* ZJ-FGZX1 genome was performed using the National Center for Biotechnology Information Prokaryotic Genome Annotation Pipeline (Angiuoli et al. 2008; Tatusova et al. 2014). The genome of *P. stewartii* subsp. *indologenes* ZJ-FGZX1 was found to be composed of one unique circle chromosome and two unique plasmids. This is similar to the genome of *P. stewartii* subsp. *stewartii* DC283, which is composed of one circular chromosome and eleven plasmids. The whole genome consists of 4,982,863 bp and had a total of 53.54% GC content. There were 4,699 genes, 4,586 coding sequences, and 4,508 protein-coding genes, among the total 113 RNA-encoding genes, seven genes encoded for ribosomal RNAs, 79 encoded for transfer RNAs, 12 were identified as noncoding RNAs, and 78 were pseudo-genes. The chromosome is 4,550,072 bp, the sizes of the two plasmids are 326,337 and 106,454 bp. The EPS genes were in the chromosome, while *Hrp* genes were located in one plasmid. A comparative account of the genome features of *P. stewartii* subsp. *indologenes* ZJ-FGZX1 and *P. stewartii* subsp. *stewartii* DC283 was determined (Table 1).

The average nucleotide identity (ANI) between genomes was a gold standard classification of prokaryotes instead of DNA-DNA hybridization (Jain et al. 2018; Richter and Rosselló-Móra 2009). The reciprocal average nucleotide identity (ANI index) was calculated by ANI calculator (Goris et al. 2007). The ANI index between ZJ-FGZX1 and *P. stewartii* subsp. *indologenes* LMG 2632 is 99.21%, while ZJ-FGZX1 shares an ANI index of 99.03, 99.05, and 99.18%, respectively with three *P. stewartii* subspecies strains, DC283, CCUG, and MS1. ZJ-FGZX1 was identified as *P. stewartii* subsp. *indologenes*. To the best of our knowledge, this is the first announcement of the complete genome sequence of a *P. stewartii* subsp. *indologenes* strain. The annotated genome assembly of *P. stewartii* is available in GenBank under accession numbers CP049115, CP049116 and CP049117.

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