



Complete Genome Sequence of a *Pseudomonas simiae* Strain with Biocontrol Potential against *Aphanomyces* Root Rot

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ABSTRACT *Aphanomyces euteiches* is a soilborne plant pathogen. It causes severe root rot in leguminous crop species. We report the complete genome sequence of a biocontrol strain, *Pseudomonas simiae* K-Hf-L9. The strain inhibited *Aphanomyces euteiches* mycelia and zoospores and suppressed root rot in field peas grown under controlled growth chamber conditions.

The genus *Pseudomonas* includes species that demonstrate a great deal of metabolic diversity and are found in many habitats (1). Strains of *Pseudomonas simiae* are reported to exhibit biocontrol activity against fungal and oomycete pathogens via siderophore-mediated competition for iron and induction of systemic resistance in the host plant (2, 3). *Pseudomonas simiae* strain K-Hf-L9 was isolated from the soil of a pea field crop in Saskatchewan, Canada (4).

Pseudomonas simiae strain K-Hf-L9 was inoculated from frozen stock (50% glycerol, in -80°C storage) on half-strength Trypticase soy agar plates (half-strength Difco Trypticase soy broth plus 1.5% agar) and grown for 24 h at 28°C . Genomic DNA was extracted using the FastDNA Spin kit (MP Biomedicals) according to the manufacturer's instructions. Genomic DNA library preparation and whole-genome sequencing were performed at the University of Regina (Regina, Canada). Genomic DNA was sheared using a g-TUBE (Covaris, Woburn, MA, USA) and a centrifugation protocol yielding an average DNA fragment size of approximately 8,000 bases (Eppendorf 5424 centrifuge and two 30-s centrifugations at $2,000 \times g$). A library was prepared for Nanopore sequencing using the ligation sequencing kit (SQK-LSK109) together with the native barcoding expansion kit (EXP-NBD103; Oxford Nanopore Technologies). Sequencing was performed using a MinION Mk1B device and a FLO-MIN106D (R9.4.1) flow cell. The fast5 files generated by the run were base called and demultiplexed after sequencing using Guppy version 4.4.1 and the high-accuracy (HAC) model.

MinION DNA sequencing data were evaluated using NanoPlot version 1.27.0 (5). Sequencing of *P. simiae* strain K-Hf-L9 yielded a total of 39,604 reads. FilTlong version 0.2.0 (<https://github.com/rswick/FilTlong>) was used to generate two sets of filtered reads. Filtered set 1 included 35,496 reads that were greater than 1,000 bp long, representing the top 95% of sequences in the data set. Filtered set 2 included all reads (39,159 reads) that were greater than 1,000 bp. Trycycler version 0.3.3 (<https://github.com/rswick/Trycycler>) was used to semiautomate the generation of a circular consensus genome assembly produced by the assembler programs Flye version 2.8.1-b1676 (6), miniasm version 0.3-r179 (7) with minipolish version 0.1.2 (8), Raven version 1.2.2 (9), and Redbean version 2.2.5 (10). Filtered set 1 was used to generate the assemblies, while filtered set 2 was used in all postassembly steps and during the

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TABLE 1 Summary of the *Pseudomonas simiae* K-Hf-L9 genome characteristics

Characteristic	Value(s)
No. of contigs	1
Genome size (bp)	6,199,521
G+C content (%)	60.28
Total no. of genes	5,679
Total no. of CDSs ^a	5,587
No. of CDSs with protein	5,425
No. of RNA genes	92
No. of complete rRNAs (5S, 16S, 23S)	7, 6, 6
No. of tRNAs	69
No. of noncoding RNAs	4
Total no. of pseudogenes	162

^aCDSs, coding sequences.

final polishing step, which was performed using Medaka version 1.0.3 (<https://github.com/nanoporetech/medaka>) and its r941_min_high_g360 model. Then, the assembly was evaluated using QUAST version 5.0.2 (11), minimap2 version 2.17 (7), SAMtools version 1.9 (12), and mosdepth version 0.3.1 (13) reported 59.87-fold average sequencing coverage of the final genome assembly. The *P. simiae* K-Hf-L9 genome was annotated using the National Center for Biotechnology Information (NCBI) Prokaryotic Genome Annotation Pipeline (PGAP) version 5.1 (14). The genome has a consensus length of 6,199,521 bp and a G+C content of 60.28% (Table 1). Average nucleotide identity (ANI) analysis using JSpeciesWS (15) was used to compare the K-Hf-L9 genome to other sequenced genomes of *Pseudomonas* strains. Strain K-Hf-L9 has an ANI value of 99.3% with respect to the *P. simiae* type strain CCUG 50988 (16).

Data availability. This whole-genome shotgun project has been deposited in GenBank under accession number [CP066169.2](https://www.ncbi.nlm.nih.gov/nuccore/CP066169.2), BioProject number [PRJNA662093](https://www.ncbi.nlm.nih.gov/bioproject/PRJNA662093), and BioSample number [SAMN16076439](https://www.ncbi.nlm.nih.gov/biosample/SAMN16076439). The raw reads are published in the SRA database under the accession number [PRJNA662093](https://www.ncbi.nlm.nih.gov/sra/PRJNA662093).

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We declare no conflicts of interest.

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