

Draft Genome Sequences of Four Hospital-Associated *Pseudomonas putida* Isolates

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We present here the draft genome sequences of four *Pseudomonas putida* isolates belonging to a single clone suspected for nosocomial transmission between patients and a bronchoscope in a tertiary hospital. The four genome sequences belong to a single lineage but contain differences in their mobile genetic elements.

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Pseudomonas putida is a Gram-negative bacterium belonging to the *fluorescens* group of *Pseudomonas* species and is most commonly found in the soil (1). *P. putida* occasionally causes opportunistic infections of the skin and soft tissue, bacteremia, and may cause serious infections among immunocompromised patients (1–3). A number of hospital-associated cases and small outbreaks of *P. putida* have been described (1, 3–5). *P. putida* is a metabolically versatile pathogen that can withstand a number of physical and chemical stressors, enabling it to survive in a wide range of environmental niches (6–8).

In 2014 to 2015, during the course of routine infection prevention surveillance, four isolates of *P. putida* were identified. One isolate (PSP1) was obtained from a bronchoscope, while the remaining three isolates (PSP2, PSP3, and PSP4) were obtained from clinical cultures from three different patients, raising the possibility of hospital-associated transmission (9). Conventional molecular typing by pulsed-field gel electrophoresis (PFGE) showed 86% band similarity among all four isolates. All isolates had intermediate or full resistance to trimethoprim-sulfamethoxazole, ceftriaxone, and aztreonam.

An isolated colony of each strain was grown overnight on sheep blood agar. Genomic DNA was extracted using the Qiagen DNeasy extraction kit. Illumina Nextera genomic libraries prepared according to the method of Baym et al. (10) were sequenced using the Illumina MiSeq version 2 (500-cycle) kit. The sequencing generated a coverage depth of 43 to 80×. Reads were assembled using SPAdes version 3.5 (11) and annotated using Prokka version 11 (12). Single nucleotide polymorphisms (SNPs) were assessed by mapping PSP2 to PSP4 reads against the PSP1 reference using bwa-mem, GATK tools, and custom python scripts (13, 14). Pairwise SNP distances were calculated from reads using PSP1 as a reference genome.

The four draft genome sequences were assembled into 68 to 155 contiguous nucleotide sequences with 5,332 to 5,396 predicted genes. Core genome phylogenetic analysis showed that all

four strains belonged to a distinct phylogenetic cluster that was not closely related to available *P. putida* reference strains; all four genomes were within 61 to 163 pairwise SNP distance. A number of gene differences related to mobile genetic elements were identified. PSP1, PSP3, and PSP4 contained a novel 28-kb prophage-related chromosomal island. Likewise, PSP1, PSP2, and PSP3 contained a putative 50-kb plasmid that had less than 50% similarity with any known plasmid sequence.

These findings are in keeping with the transmission and microevolution of a unique clone of *P. putida* containing previously uncharacterized mobile genetic elements within the hospital. Continued surveillance and monitoring of patients and the hospital environment are required to monitor the evolution of *P. putida* and other strains associated with hospital transmission. Whole-genome sequence (WGS) analysis is rapidly becoming the gold standard for hospital infection surveillance and for the investigation of suspected health care-associated transmission of infectious pathogens (15). The availability of comparison strains, such as those presented in this work, is essential for comparative genomic analyses of nosocomial pathogens.

Accession number(s). This whole-genome shotgun project has been deposited at GenBank under the accession numbers [MCBG00000000](https://ncbi.nlm.nih.gov/nucl/NC_020000000), [MCBH00000000](https://ncbi.nlm.nih.gov/nucl/NC_020000001), [MCBI00000000](https://ncbi.nlm.nih.gov/nucl/NC_020000002), and [MCBJ00000000](https://ncbi.nlm.nih.gov/nucl/NC_020000003). The version described here is the first version, with [MCBG01000000](https://ncbi.nlm.nih.gov/nucl/NC_020000004), [MCBH01000000](https://ncbi.nlm.nih.gov/nucl/NC_020000005), [MCBI01000000](https://ncbi.nlm.nih.gov/nucl/NC_020000006), and [MCBJ01000000](https://ncbi.nlm.nih.gov/nucl/NC_020000007), respectively.

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