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GENOME SEQUENCES

Draft Genome Sequences of Two *Bacillus* sp. Strains and Four *Cellulomonas* sp. Strains Isolated from Heavy-Metal-Contaminated Soil

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ABSTRACT We present the draft genome sequence for *Bacillus* sp. strain PF3, *Bacillus* sp. strain K6W, *Cellulomonas* sp. strain B12, *Cellulomonas* sp. strain K38, *Cellulomonas* sp. strain K39, and *Cellulomonas* sp. strain K42B. These bacteria were isolated from contaminated soils, and their genomes contain genes related to chromate transport and reduction.

Previously, bacteria were isolated from soil at a Department of Transportation site (Seymore, IN) that was contaminated with chromium (1, 2). Chromium is a heavy metal and has two naturally occurring oxidation states, Cr(III) and Cr(VI), the latter being more soluble and toxic than the other (3, 4). To gain a better understanding of bacterial tolerance (resistance and/or reduction) to Cr(VI), bacteria from chromium-contaminated soils were isolated on 50% tryptic soy agar (TSA) amended with 0.25 mM Cr(VI) (K_2CrO_4). Here, we present the draft genome sequences of multiple Cr(VI)-tolerant bacteria (*Bacillus* sp. strain PF3, *Bacillus* sp. strain K6W, *Cellulomonas* sp. strain B12, *Cellulomonas* sp. strain K38, *Cellulomonas* sp. strain K39, and *Cellulomonas* sp. strain K42B) and examine their genomic potential to tolerate chromate.

Genomic DNA was extracted using the FastDNA spin kit (MP Biomedical, Santa Ana, CA). The DNA was sequenced using a whole-genome shotgun sequencing method utilizing the Illumina HiSeq 2000 platform at the Cincinnati Children's Hospital Medical Center's Genetic Variation and Gene Discovery Core facility. Adapters and primers on the raw reads were trimmed using Trimmomatic 0.33 (5), and the quality was checked using FastQC 0.11.3 (https://www.bioinformatics.babraham.ac.uk/projects/fastqc/). R2 reads had low quality, so the reads were cut (length of 70 bp) and then passed through a quality filter (-Q, 33; -q, 30; -p, 50) with the FastX toolkit (http://hannonlab.cshl.edu/fastx_toolkit/). Reads were then subsampled to 4 million reads (more reads did not increase assembly quality) with seqtk (https://github.com/lh3/seqtk) and then assembled with SPAdes 3.11.1 (6, 7). The resulting assemblies were quality assessed with QUAST 3.0 (8) and CheckM (9). For the *Bacillus* strains, the total length of each assembly ranged from 5.1 to 5.2 Mb with a GC content of 35% and N_{50} values of 130,611 bp (K6W) and 53,321 bp (PF5) (Table 1). The *Cellulomonas* strains had total lengths ranging from 3.6 to 4.1 Mb, GC content of 74%, and N_{50} values ranging from 2,844 to 5,917 bp (Table 1). CheckM estimated that all 6 environmental isolates had >95% completion and <5% contamination (Table 1).

The assembled genomes were annotated by the Department of Energy's Joint Genome Institute Integrated Microbial Genomes (IMG) system (10). Annotated gene counts ranged from 3,467 to 5,386, and Pfam gene counts ranged from 2,623 to 4,440 (Table 1). Only *Cellulomonas* sp. strains K38 and B12 had annotated chromate reductases that are NAD(P)H-dependent flavin mononucleotide (FMN) reductases. The Cr(VI)-reducing bacterium *Pseudomonas putida* KT2440 uses a NAD(P)H reductase, *chrR*, to enzymatically reduce Cr(VI) (11). Both *Bacillus* sp. strains had annotated chromate

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TABLE 1 Assembly and annotation quality control and assessment data[a]

Strain	Total length (bp)	GC (%)	No. of contigs	Largest contig (bp)	N_{50} (bp)	Comp. (%)[b]	Contam. (%)[c]	Annotated gene count	Annotated Pfam count	WGS accession number[d]
Bacillus sp. K6W	5,254,820	35	124	421,542	130,611	99	0	5,386	4,440	QMGC00000000
Bacillus sp. PF5	5,148,681	35	184	296,030	53,321	99	0	5,260	4,298	QMGB00000000
Cellulomonas sp. B12	3,640,453	74	1,788	43,552	2,844	97	4	3,467	2,636	QMGD00000000
Cellulomonas sp. K38	3,883,871	74	1,811	39,943	3,096	95	2	3,827	2,820	QMGE00000000
Cellulomonas sp. K39	4,007,628	74	1,277	31,101	5,078	98	1	4,113	2,991	QMGF00000000
Cellulomonas sp. K42B	4,103,616	74	1,158	36,569	5,917	99	2	4,194	3,078	QMGG00000000

[a]Strain heterogeneity was 0 for each strain listed.
[b]Comp., completion.
[c]Contam., contamination.
[d]DDBJ/ENA/GenBank whole-genome shotgun project.

transporters. *Pseudomonas aeruginosa* uses a chromate transporter, *chrA*, to provide resistance to Cr(VI) (12). All environmental isolates had annotated genes coding cobalt-zinc-cadmium efflux system proteins, which have been linked to heavy metal tolerance in other bacteria (13, 14). Therefore, the genomes provide evidence of how these environmental isolates tolerate chromate and suggest tolerance to other heavy metals.

Data availability. The raw sequencing reads have been deposited in the Sequence Read Archive (SRA) under the accession number SRP120551. This whole-genome shotgun project has been deposited at DDBJ/ENA/GenBank under the accession numbers QMGB00000000 to QMGG00000000, as listed in Table 1. Integrated Microbial Genomes (IMG) annotations have the Genomes Online Database (GOLD) study identification (ID) number Gs0130379.

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