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GENOME SEQUENCES

Draft Genome Sequence of *Bordetella* sp. Strain FB-8, Isolated from a Former Uranium Mining Area in Germany

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ABSTRACT Here, we present the draft genome sequence of *Bordetella* sp. strain FB-8, a mixotrophic iron-oxidizing bacterium isolated from creek sediment in the former uranium-mining district of Ronneburg, Germany. To date, iron oxidation has not been reported in *Bordetella* species, indicating that FB-8 may be an environmentally important *Bordetella* sp.

The Gessen Creek, in the former uranium-mining district of Ronneburg (Thuringia, Germany), is contaminated with heavy metals and acid mine drainage (AMD) due to legacy acid leaching of low-grade black shale (1–3). However, heavy metals are naturally attenuated via coupled microbial iron cycling and metal precipitation (4, 5). To better understand the role of iron-oxidizing bacteria at the site, we isolated *Bordetella* sp. strain FB-8 from iron-rich, slightly acidic (pH 6.3) Gessen Creek sediments (site R3 [4]). Strain FB-8 was isolated as a microaerophilic, autotrophic iron oxidizer on $FeCO_3$ plates and screened for metabolic properties (Table 1) in medium described by Akob et al. (6).

For genome sequencing, FB-8 was grown to high cell density in lysogeny broth (LB) under oxic conditions. Biomass was harvested by centrifugation, frozen at −20°C, and shipped to the Deutsche Sammlung von Mikroorganismen und Zellkulturen (DSMZ; Braunschweig, Germany) for DNA extraction. DNA was extracted using the U.S. Department of Energy's (DOE) Joint Genome Institute (JGI) cetyltrimethylammonium bromide (CTAB) procedure for isolating high-molecular-weight genomic DNA (gDNA) (http://my.jgi.doe.gov/index.html). gDNA was assessed for quality control using agarose gel electrophoresis to evaluate the quality and quantity, including molecular weight, of the extract according to the JGI protocol "Genomic DNA QC Using Standard Gel Electrophoresis" (http://my.jgi.doe.gov/index.html).

DNA extracts were sent to the JGI for genome sequencing using Illumina technology. An Illumina standard shotgun library and long-insert mate pair library were constructed and sequenced using the Illumina HiSeq 2000 platform, which yielded 19,057,432 and 55,056,510 sequence reads, respectively. Raw sequence data were passed through DUK (7), which removes known Illumina sequencing and library preparation artifacts. Default parameters were used for all software unless otherwise specified. Filtered Illumina reads were assembled using AllpathsLG version R37654 (PrepareAllpathsInputs: PHRED 64=1 PLOIDY=1 FRAG COVERAGE=125 JUMP COVERAGE=25; RunAllpathsLG: THREADS=8 RUN=std pairs TARGETS=standard VAPI WARN ONLY=True OVERWRITE=True) (8). The final assembly was based on 2,858.2 Mb of Illumina standard paired-end (PE) and 4,789.5 Mb of Illumina Cre-LoxP inverse PCR (CLIP) PE postfiltered data, which provided an average of

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TABLE 1 Metabolic screening of *Bordetella* sp. strain FB-8

Type of growth	Compound[a]	Growth[b]
Heterotrophic[c]	Fructose (10 mM)	+
	Glucose (10 mM)	+
	Lactate (10 mM)	+
	Pyruvate (10 mM)	−
	Yeast extract (0.1%)	+
Autotrophic[d]	Fe(II) oxidation	+
	Thiosulfate (20 mM)	−
	Tetrathionate (10 mM)	−
	Sulphur (0.5%)	−

[a] pH range, 3 to 6. The pH optimum and minimum were determined in $FeCO_3$ liquid with the pH adjusted to values of 2 to 8 in steps of 1 pH value. Heavy metal tolerance (+) was determined in $FeCO_3$ liquid medium amended with a mixture of 10 mM Ni, 2.5 mM Cu, 2 mM Cd, 2 mM Co, and 22 mM Zn.

[b] +, Growth of the compound; −, lack of growth.

[c] Heterotrophic growth was assessed in medium containing, per liter, 20 ml of 50× basal salts solution (12) and 1 ml trace element solution (12), which was amended with an organic carbon compound.

[d] Autotrophic growth with reduced sulfur compounds was determined in pH 5.5 medium containing, per liter, 20 ml of 50× basal salts solution (12), 1 ml trace element solution (12), and either thiosulfate, tetrathionate, or sulfur. All reduced sulfur compounds were added from sterile, anoxic stocks; sulfur was autoclaved and then dispensed using aseptic techniques.

1,865.3× coverage. For postassembly quality control, we analyzed G+C histograms of contigs in conjunction with their taxonomic assignments as based on a BLASTP (9) search of the predicted genes. Further, we verified the 16S rRNA gene sequence in the FB-8 genome assembly.

The *Bordetella* sp. strain FB-8 genome contains 6 contigs (N_{50}, 2.4 Mbp) in 2 scaffolds (N_{50}, 4.1 Mbp) and constitutes a total of 4,079,718 bp with 63.35% G+C content. The FB-8 genome was annotated in the Integrated Microbial Genomes (IMG) database (10). One round of manual curation was performed using GenePRIMP (11). The genome of *Bordetella* sp. strain FB-8 contained 3,906 genes and 3,835 protein-coding genes. Functional annotation identified 3 copies of the 16S rRNA gene, 9 total rRNAs, 50 tRNAs, 12 other RNAs, and 138 pseudogenes.

The genus *Bordetella* contains mainly pathogens that infect different host organisms. *Bordetella* sp. strain FB-8 is unique because it is an environmental species that was isolated as an iron oxidizer. The genome of *Bordetella* sp. strain FB-8 may enable further analysis of the biomineralization in contaminated sites.

Data availability. *Bordetella* sp. strain FB-8 is available from the DSMZ under accession number DSM 24873. The genome sequence of strain FB-8 is available from IMG under the genome ID 2522125081 and the NCBI database under accession numbers PRJNA187096 (BioProject) and SRP053466 (Sequence Read Archive).

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We declare no competing financial interest.

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