Draft Genome Sequence of Mn(II)-Oxidizing Bacterium

Oxalobacteraceae sp. Strain AB_14

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ABSTRACT Biological Mn(II) oxidation produces reactive manganese oxides that help to mitigate metal contamination in the environment. Here, we present the genome of Oxalobacteraceae sp. strain AB_14, a species of Mn(II)-oxidizing bacteria (MOB) that is notable for its ability to catalyze Mn oxidation at low pH (5.5).

Biological Mn oxidation produces highly reactive and abundant Mn oxide phases in the environment that can mitigate metal contamination (1). However, little is known about Mn oxidation in low-pH environments, where metal contamination is often a problem due to acid mine drainage (AMD). Here, we present the genome of Oxalobacteraceae sp. strain AB_14, isolated from AMD-affected Gessen Creek sediment located in the former Ronneburg uranium mining area in Germany (2). Notably, this bacterium catalyzes Mn oxidation at low pH (5.5). It is hypothesized that Mn(II)-oxidizing bacteria (MOB), such as strain AB_14, can reduce the aqueous metal load in heavy-metal-containing AMD environments (2, 3).

AB_14 was isolated on BM5.5 plates containing MnCO3 (modified Leptothrix medium according to Mulder and van Veen [4]), as described by Akob et al. (2), then maintained indefinitely on solid or liquid BM5.5 medium by transferring every 1 to 2 months. Sequencing of the 16S rRNA gene showed that the isolate was most closely related to type strain Duganella zoogloeoides IAM 12670 (97.8% identity). For genome sequencing, strain AB_14 was grown to high cell density in liquid BM5.5 media (modified Leptothrix medium according to Mulder and van Veen [4]), as described by Akob et al. (2). 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 (DOE) Joint Genome Institute (JGI) cetyltrimethylammonium bromide (CTAB) procedure for isolating high-molecular-weight genomic DNA (gDNA) (5). Isolated gDNA was analyzed using agarose gel electrophoresis to evaluate the quantity and quality, including molecular weight, of the extract according to the JGI guidelines (https://jgi.doe.gov/user-programs/pmo-overview/project-materials-submission-overview/).

Extracted DNA was sent to the JGI (Walnut Creek, CA) for whole-genome sequencing. An Illumina standard shotgun library and a long-insert mate pair library were constructed and sequenced using the Illumina HiSeq 2000 platform, resulting in 23,039,042 reads totaling 3,455.9 Mb and 30,522,994 reads totaling 2,777.6 Mb, respectively. Additionally, a PacBio SMRTbell library was constructed from the same gDNA extract and sequenced on
the PacBio RS platform (6); 82,778 raw PacBio reads yielded 129,789 adapter-trimmed and quality-filtered subreads totaling 282.8 Mb. All raw Illumina sequence data were passed through the DUK filtering program, which removes known Illumina sequencing and library preparation artifacts (7). Default parameters were used for all software unless otherwise specified. Filtered Illumina and PacBio reads were then assembled using ALLPATHS-LG version R37654 (PrepareAllpathsInputs: PHRED 64 = 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 3,438.6 Mb of illumina standard paired-end (PE), 2,777.4 Mb of illumina Cre-LoxP inverse PCR (CLIP) PE read, and 282.8 Mb of PacBio postfiltered data, which provided an average of 888.0× Illumina coverage and 40.4× PacBio coverage of the genome.

The final draft assembly contained 8 contigs \((N_{\text{contigs}}/L_{\text{contig}} 1/3.7 \text{Mb})\) in 6 scaffolds \((N_{\text{scaffolds}}/L_{\text{scaffold}} 1/3.8 \text{Mb})\), with a total genome size of 7.0 Mb and a G+C content of 64.32%. The AB_14 genome was annotated in the Integrated Microbial Genomes (IMG) database (9). One round of manual curation was performed using GenePRIMP (10). The Oxalobacteraceae sp. AB_14 genome contained 6,242 genes, with 6,125 of them coding for proteins. Annotation identified a total of 117 RNA genes, including 21 rRNA genes (7 copies each of the 5S rRNA, 16S rRNA, and 23S rRNA genes), 82 tRNA genes, and 14 genes for noncoding RNAs. The genome also contains one likely clustered regularly interspaced short palindromic repeat (CRISPR) array.

**Data availability.** The AB_14 draft genome is available from the IMG database under genome identifier 2522125015 and from the NCBI database under BioProject number PRJNA183351, BioSample number SAMN02440486, and Sequence Read Archive accession number SRP024932.

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We declare no competing financial interest.

**REFERENCES**