





Draft Genome Sequence of *Polaromonas eurypsychrophila* AER18D-145, Isolated from a Uranium Tailings Management Facility in Northern Saskatchewan, Canada

Alexander A. Grigoryan,^{a,b} Viorica F. Bondici,^{a,c}  Yuriy Kryachko,^a Nurul H. Khan,^{a,d} John R. Lawrence,^e Gideon M. Wolfaardt,^{f,g} Niradha Withana Gamage,^a Deeksha Shetty,^a  Darren R. Korber^a

^aFood and Bioproduct Sciences, University of Saskatchewan, Saskatoon, SK, Canada

^bSaudi Aramco, Dhahran, Saudi Arabia

^cCanadian Light Source, Saskatoon, SK, Canada

^dLallemand Plant Care, Saskatoon, SK, Canada

^eEnvironment Canada, Saskatoon, SK, Canada

^fDepartment of Chemistry and Biology, Ryerson University, Toronto, ON, Canada

^gDepartment of Microbiology, Stellenbosch University, Stellenbosch, South Africa

ABSTRACT The 4.8-Mbp draft genome sequence of *Polaromonas eurypsychrophila* AER18D-145, isolated from a uranium tailings management facility, is reported. The sequence may provide insights into the mechanisms of the hypertolerance of this strain to extreme conditions and help determine its potential for bioremediation applications.

Polaromonas spp. have been reported to be among the most abundant microorganisms in glacial and seasonally cold nonglacial environments (1–5). These bacteria were shown to be capable of oxidizing molecular hydrogen (6), arsenite (7), and various recalcitrant organic compounds (8, 9). Several *Polaromonas* species were demonstrated to be capable of nitrate reduction (10, 11). Sun et al. (12) suggested that some *Polaromonas* spp. might also be capable of vanadate reduction. Despite the fact that several microorganisms belonging to this genus have been previously isolated and their metabolic capabilities investigated, few studies have been dedicated to the determination of genome sequences of *Polaromonas* spp. inhabiting uranium-rich environments.

Here, we report the draft genome sequence of *Polaromonas eurypsychrophila* AER18D-145 from a uranium tailings management facility in Key Lake, Northern Saskatchewan, Canada (57°13'N, 105°38'W). The strain was isolated from a tailings sample collected at an 18-m depth below the tailings-water interface (13). To isolate the microorganism, 0.2 g of the sample was suspended in 1 mL of sterile Tris-EDTA buffer, pH 8, plated on Reasoner's 2A (R2A) agar, and incubated aerobically at 5°C for 3 weeks. Following isolation, colonies were subcultured three times. The pure culture was stored at –80°C in 15% glycerol/5% tryptic soy broth. A DNA extraction kit (Qiagen, Maryland, USA) was used to extract DNA from glycerol-stock cells, which were regrown on R2A agar.

Genomic DNA was extracted using the DNeasy blood and tissue kit (Qiagen) according to the manufacturer's recommendations. Libraries were prepared using the Nextera XT library preparation kit (Illumina) with a MiSeq reagent 300-cycle V2 kit (Illumina), and sequencing was performed on an Illumina MiSeq instrument, resulting in 725,002 paired reads (209.75 Mbp). The A5-miseq assembly pipeline version 20140604 (14, 15) was used for error correction, quality trimming, contig assembly, misassembly corrections, and scaffolding. The genome consists of 135 contigs (N_{50} , 77,773 bp) and is 4,822,403 bp long; no gaps were identified. The genome coverage is 42×, and the G+C content is 63.1%. Annotation of the genome was done using the NCBI Prokaryotic Genome Annotation Pipeline (PGAP) version 5.1 (16, 17). As a result

Editor J. Cameron Thrash, University of Southern California

Copyright © 2022 Grigoryan et al. This is an open-access article distributed under the terms of the [Creative Commons Attribution 4.0 International license](https://creativecommons.org/licenses/by/4.0/).

Address correspondence to Darren R. Korber, darren.korber@usask.ca.

The authors declare no conflict of interest.

Received 7 January 2022

Accepted 25 February 2022

Published 9 March 2022

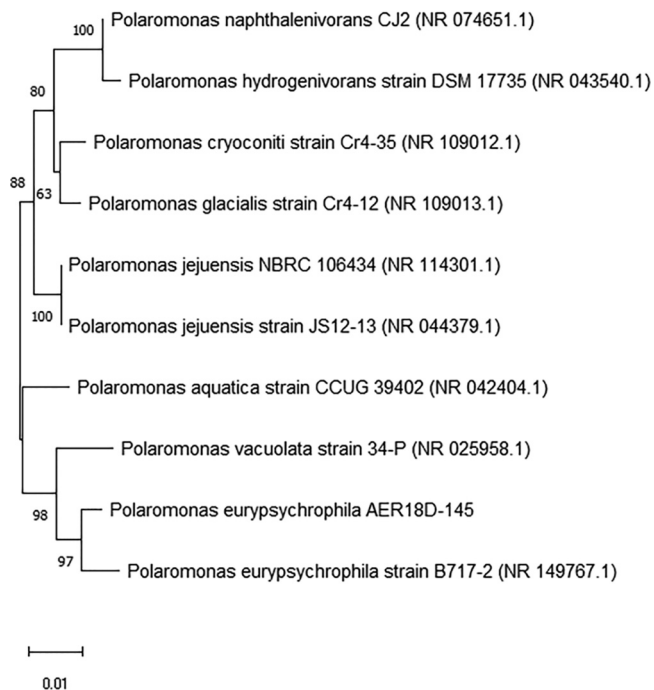


FIG 1 Neighbor-joining tree showing type strains having the highest percent identities to *P. eurypsychrophila* AER18D-145, as determined through the comparative analysis of 16S rRNA gene sequences. GenBank accession numbers of the type strains are shown in parentheses. Bootstrap values are indicated next to the tree. The scale bar indicates the number of nucleotide substitutions per site. The tree was generated using MEGA version 11 (22); the MUSCLE sequence alignment method (23), 1,000 bootstrap replications, and the maximum composite likelihood nucleotide substitution model/method were selected; otherwise, MEGA default settings were applied for phylogeny reconstruction.

of annotation, 4,490 protein-coding sequences, as well as 49 RNA-coding sequences, were identified in the genome.

Comparison of the 16S rRNA gene sequence of *P. eurypsychrophila* AER18D-145 to the RefSeq database sequences (18) using the BLASTN algorithm (19) showed that its 1,425-bp fragment was 100% identical to that of *P. eurypsychrophila* strain D3M1 (GenBank accession number [MW647764](https://www.ncbi.nlm.nih.gov/nuccore/MW647764)). Comparison to the 16S rRNA gene sequences of type strains indicated the highest percent identity of 98.8% to *P. eurypsychrophila* strain B717-2 (11) (Fig. 1), confirming the species identity of *P. eurypsychrophila* AER18D-145.

Some genes indicating the potential utility of this bacterium in bioremediation applications were identified through the genome analysis using RAST (Rapid Annotations using Subsystems Technology) version 2.0 (20, 21). In particular, *merA*, *merP*, and *merT*, responsible for mercury resistance, *chrA* and *chrF*, responsible for resistance to chromium-containing compounds, and *dedA* and *cysA*, which may play roles in selenium oxyanion uptake, were among the identified genes.

Data availability. This whole-genome shotgun project was deposited in DDBJ/ENA/GenBank under the accession number [NZ_NBZV00000000](https://www.ncbi.nlm.nih.gov/nuccore/NZ_NBZV00000000). The raw data were deposited in the SRA under the accession number [SRR16891862](https://www.ncbi.nlm.nih.gov/sra/SRR16891862) (BioProject number [PRJNA381359](https://www.ncbi.nlm.nih.gov/bioproject/PRJNA381359)).

ACKNOWLEDGMENTS

This research was supported by Cameco Corporation (Saskatoon, Canada) and the Natural Sciences and Engineering Research Council of Canada–Collaborative Research and Development (NSERC–CRD).

REFERENCES

- Foght J, Aislabie J, Turner S, Brown CE, Ryburn J, Saul DJ, Lawson W. 2004. Culturable bacteria in subglacial sediments and ice from two Southern hemisphere glaciers. *Microb Ecol* 47:329–340. <https://doi.org/10.1007/s00248-003-1036-5>.
- Darcy JL, Lynch RC, King AJ, Robeson MS, Schmidt SK. 2011. Global distribution of *Polaromonas* phylotypes—evidence for a highly successful dispersal capacity. *PLoS One* 6:e23742. <https://doi.org/10.1371/journal.pone.0023742>.

3. Franzetti A, Tatangelo V, Gandolfi I, Bertolini V, Bestetti G, Diolaiuti G, D'Agata C, Mihalcea C, Smiraglia C, Ambrosini R. 2013. Bacterial community structure on two alpine debris-covered glaciers and biogeography of *Polaromonas* phylotypes. *ISME J* 7:1483–1492. <https://doi.org/10.1038/ismej.2013.48>.
4. Gawor J, Grzesiak J, Sasin-Kurowska J, Borsuk P, Gromadka R, Górniak D, Świątecki A, Aleksandrak-Piekarczyk T, Zdanowski MK. 2016. Evidence of adaptation, niche separation and microevolution within the genus *Polaromonas* on Arctic and Antarctic glacial surfaces. *Extremophiles* 20:403–413. <https://doi.org/10.1007/s00792-016-0831-0>.
5. Ciok A, Budzik K, Zdanowski MK, Gawor J, Grzesiak J, Decewicz P, Gromadka R, Bartosik D, Dziewit L. 2018. Plasmids of psychrotolerant *Polaromonas* spp. isolated from Arctic and Antarctic glaciers: diversity and role in adaptation to polar environments. *Front Microbiol* 9:1285. <https://doi.org/10.3389/fmicb.2018.01285>.
6. Sizova M, Panikov N. 2007. *Polaromonas hydrogenivorans* sp. nov., a psychrotolerant hydrogen-oxidizing bacterium from Alaskan soil. *Int J Syst Evol Microbiol* 57:616–619. <https://doi.org/10.1099/ijs.0.64350-0>.
7. Osborne TH, Heath MD, Martin ACR, Pankowski JA, Hudson-Edwards KA, Santini JM. 2013. Cold-adapted arsenite oxidase from a psychrotolerant *Polaromonas* species. *Metallomics* 5:318–324. <https://doi.org/10.1039/c2mt20180a>.
8. Mattes TE, Alexander AK, Richardson PM, Munk AC, Han CS, Stothard P, Coleman NV. 2008. The genome of *Polaromonas* sp. strain JS666: insights into the evolution of a hydrocarbon- and xenobiotic-degrading bacterium, and features of relevance to biotechnology. *Appl Environ Microbiol* 74:6405–6416. <https://doi.org/10.1128/AEM.00197-08>.
9. Yagi JM, Sims D, Brettin T, Bruce D, Madsen EL. 2009. The genome of *Polaromonas naphthalenivorans* strain CJ2, isolated from coal tar-contaminated sediment, reveals physiological and metabolic versatility and evolution through extensive horizontal gene transfer. *Environ Microbiol* 11:2253–2270. <https://doi.org/10.1111/j.1462-2920.2009.01947.x>.
10. Margesin R, Sproer C, Zhang DC, Busse HJ. 2012. *Polaromonas glacialis* sp. nov. and *Polaromonas cryoconiti* sp. nov., isolated from alpine glacier cryoconite. *Int J Syst Evol Microbiol* 62:2662–2668. <https://doi.org/10.1099/ijs.0.037556-0>.
11. Xing T, Yao T, Liu Y, Wang N, Xu B, Shen L, Gu Z, Gu B, Liu H, Zhou Y. 2016. *Polaromonas eurypsychrophila* sp. nov., isolated from an ice core. *Int J Syst Evol Microbiol* 66:2497–2501. <https://doi.org/10.1099/ijsem.0.001079>.
12. Sun X, Qiu L, Kolton M, Haggblom M, Xu R, Kong T, Gao P, Li B, Jiang C, Sun W. 2020. V^+ reduction by *Polaromonas* spp. in vanadium mine tailings. *Environ Sci Technol* 54:14442–14454. <https://doi.org/10.1021/acs.est.0c05328>.
13. Bondici VF, Lawrence JR, Khan NH, Hill JE, Yergeau E, Wolfaardt GM, Warner J, Korber DR. 2013. Microbial communities in low permeability, high pH uranium mine tailings: characterization and potential effects. *J Appl Microbiol* 114:1671–1686. <https://doi.org/10.1111/jam.12180>.
14. Tritt A, Eisen JA, Facciotti MT, Darling AE. 2012. An integrated pipeline for de novo assembly of microbial genomes. *PLoS ONE* 7:e42304. <https://doi.org/10.1371/journal.pone.0042304>.
15. Coil D, Jospin G, Darling AE. 2015. A5-miseq: an updated pipeline to assemble microbial genomes from Illumina MiSeq data. *Bioinformatics* 31:587–589. <https://doi.org/10.1093/bioinformatics/btu661>.
16. Tatusova T, DiCuccio M, Badretdin A, Chetvernin V, Nawrocki EP, Zaslavsky L, Lomsadze A, Pruitt KD, Borodovsky M, Ostell J. 2016. NCBI Prokaryotic Genome Annotation Pipeline. *Nucleic Acids Res* 44:6614–6624. <https://doi.org/10.1093/nar/gkw569>.
17. Li W, O'Neill KR, Haft DH, DiCuccio M, Chetvernin V, Badretdin A, Coulouris G, Chitsaz F, Derbyshire MK, Durkin AS, Gonzales NR, Gwadz M, Lanczycki CJ, Song JS, Thanki N, Wang J, Yamashita RA, Yang M, Zheng C, Marchler-Bauer A, Thibaud-Nissen F. 2021. RefSeq: expanding the Prokaryotic Genome Annotation Pipeline reach with protein family model curation. *Nucleic Acids Res* 49:D1020–D1028. <https://doi.org/10.1093/nar/gkaa1105>.
18. O'Leary NA, Wright MW, Brister JR, Ciufu S, Haddad D, McVeigh R, Rajput B, Robbertse B, Smith-White B, Ako-Adjei D, Astashyn A, Badretdin A, Bao Y, Blinkova O, Brover V, Chetvernin V, Choi J, Cox E, Ermolaeva O, Farrell CM, Goldfarb T, Gupta T, Haft D, Hatcher E, Hlavina W, Joardar VS, Kodali VK, Li W, Maglott D, Masterson P, McGarvey KM, Murphy MR, O'Neill K, Pujar S, Rangwala SH, Rausch D, Riddick LD, Schoch C, Shkeda A, Storz SS, Sun H, Thibaud-Nissen F, Tolstoy I, Tully RE, Vatsan AR, Wallin C, Webb D, Wu W, Landrum MJ, Kimchi A, Tatusova T, DiCuccio M, Kitts P, et al. 2016. Reference sequence (RefSeq) database at NCBI: current status, taxonomic expansion, and functional annotation. *Nucleic Acids Res* 44:D733–D745. <https://doi.org/10.1093/nar/gkv1189>.
19. Zhang Z, Schwartz S, Wagner L, Miller W. 2000. A greedy algorithm for aligning DNA sequences. *J Comput Biol* 7:203–214. <https://doi.org/10.1089/10665270050081478>.
20. Aziz RK, Bartels D, Best AA, DeJongh M, Disz T, Edwards RA, Formsma K, Gerdes S, Glass EM, Kubal M, Meyer F, Olsen GJ, Olson R, Osterman AL, Overbeek RA, McNeil LK, Paarmann D, Paczian T, Parrello B, Pusch GD, Reich C, Stevens R, Vassieva O, Vonstein V, Wilke A, Zagnitko O. 2008. The RAST server: Rapid Annotations using Subsystems Technology. *BMC Genomics* 9:75. <https://doi.org/10.1186/1471-2164-9-75>.
21. Overbeek R, Olson R, Pusch GD, Olsen GJ, Davis JJ, Disz T, Edwards RA, Gerdes S, Parrello B, Shukla M, Vonstein V, Wattam AR, Xia F, Stevens R. 2014. The SEED and the Rapid Annotation of microbial genomes using Subsystems Technology (RAST). *Nucleic Acids Res* 42:D206–D214. <https://doi.org/10.1093/nar/gkt1226>.
22. Tamura K, Stecher G, Kumar S. 2021. MEGA11: Molecular Evolutionary Genetics Analysis version 11. *Mol Biol Evol* 38:3022–3027. <https://doi.org/10.1093/molbev/msab120>.
23. Edgar RC. 2004. MUSCLE: multiple sequence alignment with high accuracy and high throughput. *Nucleic Acids Res* 32:1792–1797. <https://doi.org/10.1093/nar/gkh340>.