



Complete Genome Sequence and Methylome of the Type Strain of *Shewanella algae*

Christian Tellgren-Roth,^a Kaisa Thorell,^b  Michael Y. Galperin,^c  Tino Krell,^d  Ute Römling,^e Åsa Sjöling,^e
 Alberto J. Martín-Rodríguez^e

^aDepartment of Immunology, Genetics, and Pathology, Uppsala Genome Center, Uppsala University, Uppsala, Sweden

^bInstitute of Biomedicine, Department of Microbiology and Immunology, University of Gothenburg, Gothenburg, Sweden

^cNational Center for Biotechnology Information, National Library of Medicine, National Institutes of Health, Bethesda, Maryland, USA

^dDepartment of Environmental Protection, Estación Experimental del Zaidín, Spanish National Research Council, Granada, Spain

^eDepartment of Microbiology, Tumor and Cell Biology, Karolinska Institutet, Stockholm, Sweden

ABSTRACT We report the complete genome sequence and base modification analysis of the *Shewanella algae* type strain CECT 5071 (= OK-1 = ATCC 51192 = DSM 9167 = IAM 14159). The genome is composed of a single chromosome of 4,924,764 bp, with a GC content of 53.10%.

The gammaproteobacterium *Shewanella algae* was first described as a tetrodotoxin-producing epiphyte of the red alga *Jania* sp. (1), and the type strain was designated OK-1 following the original nomenclature by Kotaki et al. (2). *S. algae* is the most relevant human pathogen within the genus *Shewanella*, causing bacteremia, otitis, skin and soft tissue infections, and a variety of other diseases, with the emergence of multi-drug-resistant isolates being a concern (3, 4). *S. algae* is also known for its ability to produce secondary metabolites like iron-scavenging siderophores (5). Its versatile physiology is considered a hallmark of the genus *Shewanella*. In the past 3 years, genomes of several clinical and environmental isolates of *S. algae* have been sequenced (6–9), but no complete genome sequence of a reference strain has been available. We have investigated *S. algae* from different angles (10–14). Here, we report the complete genome sequence of the *S. algae* type strain OK-1 (1), which was obtained from the Spanish Type Culture Collection (strain CECT 5071^T).

The strain was grown in LB medium at 37°C to exponential phase, and the DNA was isolated with the Genomic-tip 500/G kit (Qiagen). Multiplexed sequencing libraries with a target insert size of 8 kb were prepared using the SMRTbell Express template preparation kit v2.0 and barcoded overhang adapter kit 8A (Pacific Biosciences [PacBio], Menlo Park, CA, USA). After removal of adapter dimers, the libraries were sequenced on a Sequel single-molecule real-time (SMRT) cell following the manufacturer's recommendations (PacBio). Barcode splitting was done using SMRT Link v8.0 (PacBio) and resulted in 622,115 reads totaling 3,045,548,325 bases, with an N_{50} value of 7,709 bases. A total of 693,216,159 bases from unique reads were used in the microbial assembly pipeline included in SMRT Link v8.0 (PacBio) with default parameters.

The genome of the *S. algae* type strain CECT 5071 is composed of a single chromosome of 4,924,764 bp, with a GC content of 53.10% (Fig. 1). No extrachromosomal elements were detected. Genome annotation by the NCBI Prokaryotic Genome Annotation Pipeline (PGAP) v5.0 (15) predicted 4,400 genes, of which 4,225 are protein-coding genes. A total of 136 RNAs were predicted, including 107 tRNAs, 25 rRNAs (5S, 9 copies; 16S, 8 copies; 23S, 8 copies), and 4 noncoding RNAs. SMRT sequencing offers the possibility of assessing genome-wide DNA methylation patterns. Detected methylation motifs are summarized in Table 1, and the methylome is available in the REBASE database (16).

Citation Tellgren-Roth C, Thorell K, Galperin MY, Krell T, Römling U, Sjöling Å, Martín-Rodríguez AJ. 2021. Complete genome sequence and methylome of the type strain of *Shewanella algae*. Microbiol Resour Announc 10:e00559-21. <https://doi.org/10.1128/MRA.00559-21>.

Editor David A. Baltus, University of Arizona

Copyright © 2021 Tellgren-Roth 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 Alberto J. Martín-Rodríguez, jonatan.martin.rodriguez@ki.se.

Received 7 June 2021

Accepted 6 July 2021

Published 5 August 2021

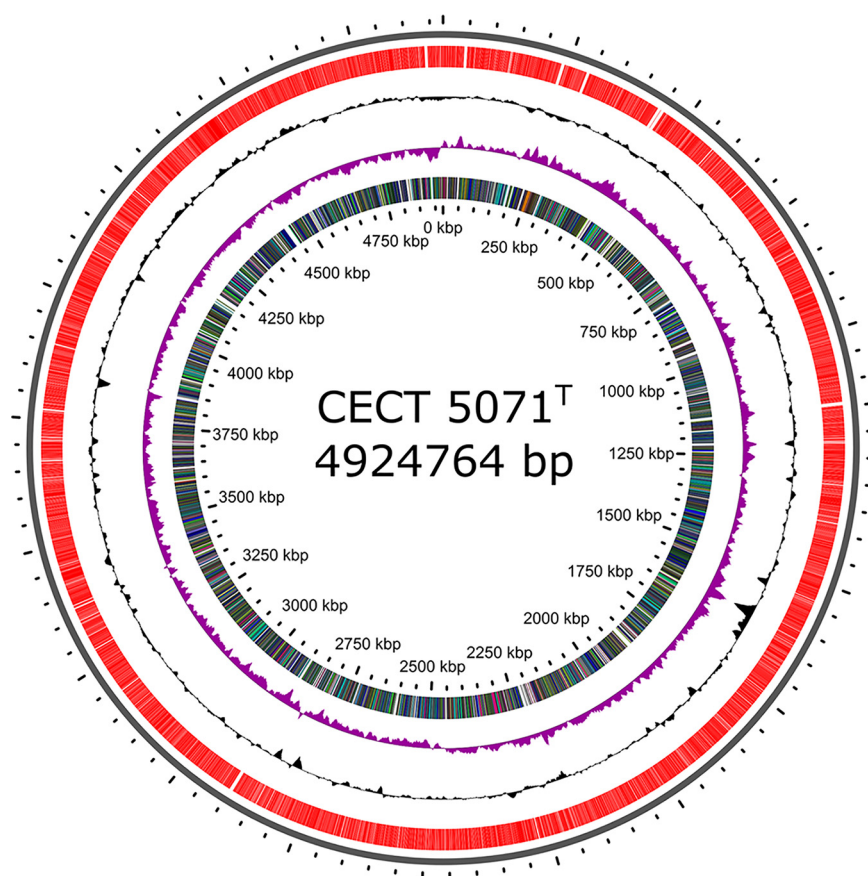


FIG 1 Circular map of the *Shewanella algae* CECT 5071^T chromosome. Circles from outside to inside represent the coding sequences (red), GC content (black), GC skew (purple), and color-coded Clusters of Orthologous Groups (COG) assignments of protein-coding genes. The map was generated with CGView (20).

Of note, the *rpoS* gene is truncated in the type strain of *S. algae* (10), consistent with evidence from the draft genomes of equivalent strains, namely, ATCC 51192 (GenBank accession no. [GCA_012396675.1](https://www.ncbi.nlm.nih.gov/nuccore/GCA_012396675.1)), NBRC 103173 (GenBank accession no. [GCA_001598875.1](https://www.ncbi.nlm.nih.gov/nuccore/GCA_001598875.1)), and JCM 21037 (GenBank accession no. [GCA_000615045.1](https://www.ncbi.nlm.nih.gov/nuccore/GCA_000615045.1)), suggesting that this truncation was already present in the original OK-1 isolate. In *Escherichia coli*, the RpoS protein is the stress sigma factor of the RNA polymerase required for stationary-phase transcription (17). However, natural *E. coli* *rpoS* mutants exist (18, 19). The ecological and physiological significance of the *rpoS* truncation in the *S. algae* type strain remains to be determined.

Data availability. The complete genome sequence of *S. algae* CECT 5071^T was deposited in DDBJ/ENA/GenBank under the accession no. [CP068230](https://www.ncbi.nlm.nih.gov/nuccore/CP068230). Sequencing raw data are available at the SRA under the accession no. [SRR14739658](https://www.ncbi.nlm.nih.gov/sra/SRR14739658). The methylome of *S. algae* CECT 5071^T is available at the REBASE database under the organism accession no. [46337](https://www.ncbi.nlm.nih.gov/rebase/methyl/46337).

TABLE 1 Summary of methylation motifs detected in the complete genome sequence of *S. algae* CECT 5071^T and associated methyltransferases

Motif	Center position	Modification type	No. detected	No. in genome	Type	Methyltransferase locus tag
GATC	2	m6A	41,451	41,456	II	— ^a
TGANNNNNNTTCC	3	m6A	827	827	I _γ	E1N14_007370
TGGCCA	4	m4C	4,218	8,820	II _α	E1N14_009720

^a—, GATC could not be matched unambiguously by REBASE because there is more than one candidate.

ACKNOWLEDGMENTS

This work was supported by grants from Stiftelsen Lars Hiertas Minne (grant FO2019-0293), Stiftelsen Långmanska Kulturfonden (grant BA20-0736), the Karolinska Institutet Research Foundation (grant 2020-01556), Stiftelsen Anna och Gunnar Vidfelts Fond för Biologisk Forskning (grant 2019-051-Vidfelts fond/SOJOH), and Hans Dahlbergs Stiftelsen för Miljö och Hälsa to A.J.M.-R. M.Y.G. was supported by the Intramural Research Program of the National Library of Medicine, National Institutes of Health. We acknowledge the National Genomics Infrastructure (NGI)/Uppsala Genome Center and UPPMAX for providing assistance in massive parallel sequencing and computational infrastructure. Work performed at NGI/Uppsala Genome Center has been funded by the Council for Research Infrastructure/Swedish Research Council and Science for Life Laboratory, Sweden.

REFERENCES

1. Simidu U, Kita-Tsukamoto K, Yasumoto T, Yotsu M. 1990. Taxonomy of four marine bacterial strains that produce tetrodotoxin. *Int J Syst Bacteriol* 40:331–336. <https://doi.org/10.1099/00207713-40-4-331>.
2. Kotaki Y, Oshima Y, Yasumoto T. 1985. Bacterial transformation of paralytic shellfish toxins in coral reef crabs and a marine snail. *Bull Jpn Soc Sci Fish* 51:1009–1013. <https://doi.org/10.2331/suisan.51.1009>.
3. Martín-Rodríguez AJ, Martín-Pujol O, Artiles-Campelo F, Bolaños-Rivero M, Römling U. 2017. *Shewanella* spp. infections in Gran Canaria, Spain: retrospective analysis of 31 cases and a literature review. *JMM Case Rep* 4: e005131. <https://doi.org/10.1099/jmmcr.0.005131>.
4. Janda JM, Abbott SL. 2014. The genus *Shewanella*: from the briny depths below to human pathogen. *Crit Rev Microbiol* 40:293–312. <https://doi.org/10.3109/1040841X.2012.726209>.
5. Rütshlin S, Gunesch S, Böttcher T. 2017. One enzyme, three metabolites: *Shewanella algae* controls siderophore production via the cellular substrate pool. *Cell Chem Biol* 24:598–604.e10. <https://doi.org/10.1016/j.chembiol.2017.03.017>.
6. Chen YJ, Tung KC, Hong YK, Chen SY, Huang YT, Liu PY. 2019. Genome sequence of colistin-resistant bacteremic *Shewanella algae* carrying the beta-lactamase gene *bla_{OXA-55}*. *Can J Infect Dis Med Microbiol* 2019:3840563. <https://doi.org/10.1155/2019/3840563>.
7. Tamez AM, McLaughlin RW, Li J, Wan XL, Zheng JS. 2021. Searching for putative virulence factors in the genomes of *Shewanella indica* and *Shewanella algae*. *Arch Microbiol* 203:683–692. <https://doi.org/10.1007/s00203-020-02060-1>.
8. de Andrade Alves VB, Carvalho E, Madureira PA, Marino ED, Vaz ACN, Vidal AMC, de Azevedo Ruiz VL. 2020. First isolation and whole-genome sequencing of a *Shewanella algae* strain from a swine farm in Brazil. *BMC Microbiol* 20:360. <https://doi.org/10.1186/s12866-020-02040-x>.
9. Endimiani A, Bernasconi OJ, Büdel T, Campos-Madueno EI, Kuenzli E, Hatz C, Carattoli A. 2020. Whole-genome characterization of a *Shewanella algae* strain coharboring *bla_{CTX-M-15}* and *armA* genes on a novel IncC plasmid. *Antimicrob Agents Chemother* 64:e00267-20. <https://doi.org/10.1128/AAC.00267-20>.
10. Martín-Rodríguez AJ, Suárez-Mesa A, Artiles-Campelo F, Römling U, Hernández M. 2019. Multilocus sequence typing of *Shewanella algae* isolates identifies disease-causing *Shewanella chilikensis* strain 614. *FEMS Microbiol Ecol* 95:fiy210. <https://doi.org/10.1093/femsec/fiy210>.
11. Thorell K, Meier-Kolthoff JP, Sjöling Å, Martín-Rodríguez AJ. 2019. Whole-genome sequencing redefines *Shewanella* taxonomy. *Front Microbiol* 10:1861. <https://doi.org/10.3389/fmicb.2019.01861>.
12. Martín-Rodríguez AJ, Villion K, Yilmaz-Turan S, Vilaplana F, Sjöling Å, Römling U. 2021. Regulation of colony morphology and biofilm formation in *Shewanella algae*. *Microb Biotechnol* 14:1183–1200. <https://doi.org/10.1111/1751-7915.13788>.
13. Martín-Rodríguez AJ, Reyes-Darias JA, Martín-Mora D, González JM, Krell T, Römling U. 2021. Reduction of alternative electron acceptors drives biofilm formation in *Shewanella algae*. *NPJ Biofilms Microbiomes* 7:9. <https://doi.org/10.1038/s41522-020-00177-1>.
14. Martín-Rodríguez AJ, González-Orive A, Hernández-Creus A, Morales A, Dorta-Guerra R, Norte M, Martín VS, Fernández JJ. 2014. On the influence of the culture conditions in bacterial antifouling bioassays and biofilm properties: *Shewanella algae*, a case study. *BMC Microbiol* 14:102. <https://doi.org/10.1186/1471-2180-14-102>.
15. 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>.
16. Roberts RJ, Vincze T, Posfai J, Macelis D. 2015. REBASE: a database for DNA restriction and modification: enzymes, genes and genomes. *Nucleic Acids Res* 43:D298–D299. <https://doi.org/10.1093/nar/gku1046>.
17. Battesti A, Majdalani N, Gottesman S. 2011. The RpoS-mediated general stress response in *Escherichia coli*. *Annu Rev Microbiol* 65:189–213. <https://doi.org/10.1146/annurev-micro-090110-102946>.
18. Carter MQ, Louie JW, Huynh S, Parker CT. 2014. Natural *rpoS* mutations contribute to population heterogeneity in *Escherichia coli* O157:H7 strains linked to the 2006 US spinach-associated outbreak. *Food Microbiol* 44:108–118. <https://doi.org/10.1016/j.fm.2014.05.021>.
19. Uhlich GA, Chen CY, Cottrell BJ, Hofmann CS, Dudley EG, Strobaugh TP, Nguyen LH. 2013. Phage insertion in *mIra* and variations in *rpoS* limit curli expression and biofilm formation in *Escherichia coli* serotype O157:H7. *Microbiology (Reading)* 159:1586–1596. <https://doi.org/10.1099/mic.0.066118-0>.
20. Stothard P, Wishart DS. 2005. Circular genome visualization and exploration using CGView. *Bioinformatics* 21:537–539. <https://doi.org/10.1093/bioinformatics/bti054>.