

Lawrence Berkeley National Laboratory

LBL Publications

Title

Draft genome sequence of Nitrobacter vulgaris DSM 10236T.

Permalink

<https://escholarship.org/uc/item/5rz8p66x>

Journal

Microbiology Resource Announcements, 13(8)

Authors

Soghomonian, Mark
Soghomonian, Angela
Escobar, Matthew
et al.

Publication Date

2024-08-13

DOI

10.1128/mra.00305-24

Peer reviewed

Draft genome sequence of *Nitrobacter vulgaris* DSM 10236^T

Mark Soghomonian,¹ Angela Soghomonian,¹ Matthew Escobar,² Vera Thiel,³ Markus Göker,³ Natalia Ivanova,⁴ Rekha Seshadri,⁴ Kalyani Maitra¹

AUTHOR AFFILIATIONS See affiliation list on p. 2.

ABSTRACT Here, we report the draft genome sequence of *Nitrobacter vulgaris* DSM 10236^T, a nitrite-oxidizing bacterium isolated from a sewage system in Hamburg, Germany. The genome is 4.3 Mb in size with 4,585 predicted genes, including the full complement of genes necessary for growth on nitrite (*nark*, *nxrA*, *nxB*, *nxC*, and *nxD*).

KEYWORDS *Nitrobacter vulgaris*, chemolithotrophy, draft genome

Nitrite-oxidizing bacteria of the genus *Nitrobacter* play essential roles in nitrogen cycling in both terrestrial and aquatic environments. They are facultative lithoautotrophs that can grow in the presence or absence of oxygen (1–3). *Nitrobacter vulgaris* is a Gram-negative mesophile that has been isolated from many environments, including freshwater and soil (4). To date, genome sequencing has been performed on only one strain of *N. vulgaris* (Ab₁) (5). The type strain *N. vulgaris* DSM 10236^T (also known as *N. vulgaris* strain Z^T) was isolated from a Bauersberg waterworks sand filter in Hamburg, Germany (4). The genome sequence of *N. vulgaris* DSM 10236^T will support further study of its role in the nitrogen cycle.

N. vulgaris DSM 10236^T was grown in mixotrophic *Nitrobacter* medium DSMZ M.756a [<https://mediadive.dsmz.de/medium/756a>] at 28°C for 10 days. Genomic DNA was extracted using the MasterPure Gram-positive DNA Purification Kit (Lucigen) and sent to the Department of Energy, Joint Genome Institute for sequencing.

An Illumina short-insert DNA library was prepared with a PerkinElmer Sciclone robotic liquid handling system using a Roche KAPA Biosystems library preparation kit. DNA (200 ng) was sheared to 300 bp using a Covaris LE220, size-selected by double-SPRI, and then end-repaired, A-tailed, and ligated with Illumina-compatible sequencing adaptors containing a unique molecular index barcode. The library was quantified using KAPA Biosystems' next-generation sequencing library qPCR kit and run on a Roche LightCycler 480 real-time PCR instrument. The library was then multiplexed with other libraries, and the pool was sequenced on an Illumina NovaSeq 6000 using NovaSeq XP v1 reagent kits (Illumina), S4 flow cell, following a 2 × 150 indexed run recipe. In total, 17,899,282 sequence reads were generated. Raw sequences were quality filtered using BBTools v.38.86 per JGI standard operating practice (SOP) protocol 1061 (6), producing 1,499,468,893 bp of sequence. The filtered and normalized reads were assembled using SPAdes (version v3.13.0) with the assembly parameters `--phred--offset 33 --cov--cutoff auto -t 16 -m 64 --careful -k 25,55,95` (7). Contigs with lengths <1 kb were discarded (BBTools `reformat.sh: minlength`). The final draft assembly was then annotated using the IMG Annotation Pipeline v.5 (8) (Table 1).

Genome analyses were performed using IMG/M (9). The genome sequence of *N. vulgaris* DSM 10236^T has a pairwise average nucleotide identity of 98.8% and 86.5% with the sequences of *N. vulgaris* Ab₁ and *N. hamburgensis* X14, respectively (10). The genome contains all genes required for chemolithotrophic growth on nitrite (*nark*, *nxrA*, *nxB*, *nxC*, and *nxD*), and its nitrite-oxidizing enzyme (NXR) operon is organized identically to the NXR operon in *N. hamburgensis* X14 and *N. vulgaris* Ab₁ (1). Interestingly, *N. vulgaris* DSM 10236^T

Editor Irene L. G. Newton, Indiana University, Bloomington, Indiana, USA

Address correspondence to Rekha Seshadri, rseshadri@ibl.gov, or Kalyani Maitra, kmaitra@mail.fresnostate.edu.

The authors declare no conflict of interest.

Received 25 March 2024

Accepted 13 June 2024

Published 11 July 2024

Copyright © 2024 Soghomonian 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/).

TABLE 1 Genome features of *Nitrobacter vulgaris* DSM 10236^T

Total scaffold sequence length (bp)	4,293,395
Number of contigs	98
Contig N ₅₀ (bp)	110,847
Average fold coverage (x)	349
GC content (%)	59.5
Total genes	4,585
Protein-coding genes	4,491
rRNA genes	3
tRNA genes	61
JGI IMG/M taxon ID	2829791209
NCBI WGS accession number	JAVDPZ000000000.1
NCBI BioProject accession number	PRJNA583244
NCBI SRA accession number	SRR10872729
NCBI BioSample number	SAMN13172834

appears to be the only *Nitrobacter* genome (of seven sequenced to date) with a predicted nitrous oxide reductase gene (*nosZ*, JGI gene ID 2829793416). It is located in an operon containing a *nosR* nitrous oxide reductase transcriptional regulator and a nitrous oxidase accessory protein. These genes are typically associated with denitrifying bacteria, and therefore further research is needed to explore possible connections between *N. vulgaris* DSM 10236^T and denitrification (11).

ACKNOWLEDGMENTS

The work (proposal DOI: <https://doi.org/10.46936/10.25585/60001087>) was conducted by the US Department of Energy Joint Genome Institute (<https://ror.org/04xm1d337>), a DOE Office of Science User Facility that is supported by the Office of Science of the US Department of Energy operated under contract no. DE-AC02-05CH11231. This announcement was largely prepared by undergraduate students, and we gratefully acknowledge JGI for initiating and supporting it as an educational project (the “Adopt-a-genome” Project). We are also grateful to Meike Döppner, DSMZ, for DNA quality control.

AUTHOR AFFILIATIONS

¹Department of Chemistry and Biochemistry, California State University, Fresno, California, USA

²Department of Biological Sciences, California State University, San Marcos, California, USA

³Leibniz Institute DSMZ, German Collection of Microorganisms and Cell Cultures, Braunschweig, Germany

⁴DOE Joint Genome Institute, Lawrence Berkeley National Laboratory, Berkeley, California, USA

AUTHOR ORCIDs

Matthew Escobar  <http://orcid.org/0000-0002-6559-1222>

Vera Thiel  <http://orcid.org/0000-0003-2833-8911>

Markus Göker  <http://orcid.org/0000-0002-5144-6200>

Natalia Ivanova  <http://orcid.org/0000-0002-5802-9485>

Rekha Seshadri  <http://orcid.org/0000-0003-3219-2900>

Kalyani Maitra  <http://orcid.org/0000-0001-5491-843X>

REFERENCES

1. Starckenburg SR, Spieck E, Bottomley PJ. 2011. Metabolism and genomics of nitrite-oxidizing bacteria: emphasis on studies of pure cultures and of *Nitrobacter* species, p 267–293. In Ward BB, Arp DJ, Klotz MG (ed), Nitrification. ASM Press, Washington, DC.

2. Bock E, Sundermeyer-Klinger H, Stackebrandt E. 1983. New facultative lithoautotrophic nitrite-oxidizing bacteria. *Arch Microbiol* 136:281–284. <https://doi.org/10.1007/BF00425217>
3. Bock E, Wilderer PA, Freitag A. 1988. Growth of *Nitrobacter* in the absence of dissolved oxygen. *Water Res* 22:245–250. [https://doi.org/10.1016/0043-1354\(88\)90085-1](https://doi.org/10.1016/0043-1354(88)90085-1)
4. Bock E, Koops H-P, Möller UC, Rudert M. 1990. A new facultatively nitrite oxidizing bacterium, *Nitrobacter vulgaris* sp. nov. *Arch Microbiol* 153:105–110. <https://doi.org/10.1007/BF00247805>
5. Mellbye BL, Davis EW, Spieck E, Chang JH, Bottomley PJ, Sayavedra-Soto LA. 2017. Draft genome sequence of *Nitrobacter vulgaris* strain Ab₁, a nitrite-oxidizing bacterium. *Genome Announc* 5:e00290-17. <https://doi.org/10.1128/genomeA.00290-17>
6. Bushnell B, Rood J, Singer E. 2017. BBMerge - accurate paired shotgun read merging via overlap. *PLoS One* 12:e0185056. <https://doi.org/10.1371/journal.pone.0185056>
7. Bankevich A, Nurk S, Antipov D, Gurevich AA, Dvorkin M, Kulikov AS, Lesin VM, Nikolenko SI, Pham S, Prjibelski AD, Pyshkin AV, Sirotkin AV, Vyahhi N, Tesler G, Alekseyev MA, Pevzner PA. 2012. SPAdes: a new genome assembly algorithm and its applications to single-cell sequencing. *J Comput Biol* 19:455–477. <https://doi.org/10.1089/cmb.2012.0021>
8. Huntemann M, Ivanova NN, Mavromatis K, Tripp HJ, Paez-Espino D, Palaniappan K, Szeto E, Pillay M, Chen I-M, Pati A, Nielsen T, Markowitz VM, Kyrpides NC. 2015. The standard operating procedure of the DOE-JGI microbial genome annotation pipeline (MGAP v.4). *Stand Genomic Sci* 10:86. <https://doi.org/10.1186/s40793-015-0077-y>
9. Chen I-MA, Markowitz VM, Chu K, Palaniappan K, Szeto E, Pillay M, Ratner A, Huang J, Andersen E, Huntemann M, Varghese N, Hadjithomas M, Tennessen K, Nielsen T, Ivanova NN, Kyrpides NC. 2017. IMG/M: integrated genome and metagenome comparative data analysis system. *Nucleic Acids Res* 45:D507–D516. <https://doi.org/10.1093/nar/gkw929>
10. Konstantinidis KT, Tiedje JM. 2005. Genomic insights that advance the species definition for prokaryotes. *Proc Natl Acad Sci U S A* 102:2567–2572. <https://doi.org/10.1073/pnas.0409727102>
11. Orellana LH, Rodriguez-R LM, Higgins S, Chee-Sanford JC, Sanford RA, Ritalahti KM, Löffler FE, Konstantinidis KT. 2014. Detecting nitrous oxide reductase (*nosZ*) genes in soil metagenomes: method development and implications for the nitrogen cycle. *mBio* 5:e01193-14. <https://doi.org/10.1128/mBio.01193-14>