



## Genome sequence of the acid-tolerant *Desulfovibrio* sp. DV isolated from the sediments of a Pb-Zn mine tailings dam in the Chita region, Russia



Anastasiia Kovaliova<sup>a</sup>, Vitaly V. Kadnikov<sup>b</sup>, Dmitrii V. Antsiferov<sup>a</sup>, Alexey V. Beletsky<sup>b</sup>, Ehrzena V. Danilova<sup>c</sup>, Marat R. Avakyan<sup>a</sup>, Andrey V. Mardanov<sup>b</sup>, Olga V. Karnachuk<sup>a,\*</sup>

<sup>a</sup> Laboratory of Biochemistry and Molecular Biology, Tomsk State University, 634050 Tomsk, Russia

<sup>b</sup> Institute of Bioengineering, Research Center of Biotechnology of the Russian Academy of Sciences, 119071 Moscow, Russia

<sup>c</sup> Institute of General and Experimental Biology, Siberian Branch Russian Academy of Sciences, 670047 Ulan-Ude, Buryatia, Russia

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### ABSTRACT

Here we report the draft genome sequence of the acid-tolerant *Desulfovibrio* sp. DV isolated from the sediments of a Pb-Zn mine tailings dam in the Chita region, Russia. The draft genome has a size of 4.9 Mb and encodes multiple K<sup>+</sup>-transporters and proton-consuming decarboxylases. The phylogenetic analysis based on concatenated ribosomal proteins revealed that strain DV clusters together with the acid-tolerant *Desulfovibrio* sp. TomC and *Desulfovibrio magneticus*. The draft genome sequence and annotation have been deposited at GenBank under the accession number MLBG00000000.

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Specifications	
Organism/cell line/tissue	<i>Desulfovibrio</i> sp. DV
Sex	N/A
Sequencer or array type	GS FLX (Roche)
Data format	Analyzed
Experimental factors	Bacterial strain
Experimental features	Assembled and annotated draft genome of <i>Desulfovibrio</i> sp. strain DV
Consent	N/A
Sample source location	Sediments of a water seepage from a tailing dam at a Zn-Pb mine in Novii Akatui, Chita Region, Russia (51° 06'12"; 117° 77'84")

### 1. Direct link to deposited data

<https://www.ncbi.nlm.nih.gov/nucleotide/MLBG000000000>.

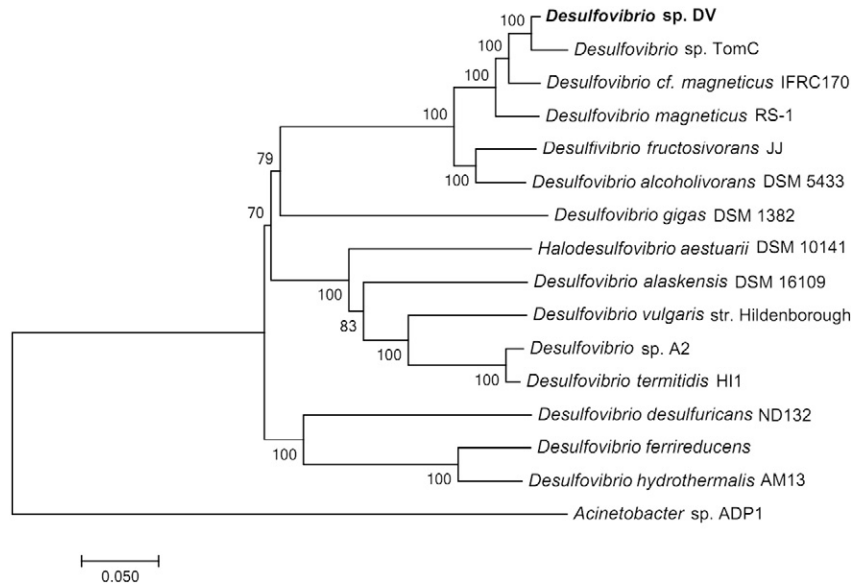
### 2. Introduction

The sulfate-reducing bacteria (SRB) are important components of microbial communities in mine drainage waters and sediments. These types of environments are often characterized by low pH

values and high concentration of dissolved metals originating from the oxidation of residual sulfide minerals in mine waste. SRB can be exploited to mitigate acid mine drainage (AMD) by metal precipitation as insoluble sulfides and proton consumption due to the biogenic H<sub>2</sub>S production [1,2]. However, only few acidophilic/acid-tolerant SRB have been isolated and characterized. The only two validly described, moderately acidophilic SRB isolated from AMD belong to the genus *Desulfosporosinus* [3,4]. At least six different phyla contain prokaryotes capable of dissimilatory sulfate reduction. The majority of known species belong to Firmicutes, including the genera *Desulfosporosinus* and *Desulfotomaculum*, and to *Deltaproteobacteria*. The *deltaproteobacterial* *Desulfovibrio* spp. are prospective for bioremediation purposes due to their relatively fast growth (compared to other SRB), tolerance to oxygen [5] and, of all SRB, the best understood metabolic features and stress response mechanisms [6]. However, the metal-tolerant *Desulfovibrio* isolates characterized so far do not tolerate low pH values [7–9]. Recently the first acid-tolerant member of *Desulfovibrio*, *Desulfovibrio* sp. TomC, was isolated and its genome was made available [10]. Here we report the draft genome sequence of a novel acid-tolerant strain DV, which was isolated from the sediments of a Pb-Zn mine waste at Novii Akatui, Chita region, Russia. The 16S rRNA sequencing and phylogenetic analysis showed that strain DV belongs to the genus *Desulfovibrio* and its closest relative is *Desulfovibrio* sp. TomC (Karnachuk et al., unpublished). The genome sequence will allow to verify the phylogenetic relationships of the two strains and other *Desulfovibrio* isolates and to explore the mechanisms, which enable these bacteria to withstand low pH values.

\* Corresponding author.

E-mail address: [olga.karnachuk@green.tsu.ru](mailto:olga.karnachuk@green.tsu.ru) (O.V. Karnachuk).



**Fig. 1.** The evolutionary history was inferred using the Neighbor-Joining method [14]. The optimal tree with the sum of branch length = 1.87228742 is shown. The percentages of replicate trees in which the associated taxa clustered together in the bootstrap test (1000 replicates) are shown next to the branches [15]. The tree is drawn to scale, with branch lengths in the same units as those of the evolutionary distances used to infer the phylogenetic tree. The evolutionary distances were computed using the Poisson correction method [16] and are in the units of the number of amino acid substitutions per site. The analysis involved 16 amino acid sequences. All positions containing gaps and missing data were eliminated. There were a total of 4398 positions in the final dataset. Evolutionary analyses were conducted in MEGA7 [17].

### 3. Experimental design, materials and methods

#### 3.1. Sequencing and assembly of the *Desulfovibrio* sp. DV genome

Genomic DNA was isolated from *Desulfovibrio* sp. DV biomass using the SDS-CTAB method [11]. The shotgun genomic DNA library was sequenced with a Roche Genome Sequencer FLX using the Titanium XL+ protocol. The reads were de novo assembled into contigs using the Newbler Assembler version 2.9 (454 Life Sciences, Branford, CT). The draft genome of *Desulfovibrio* sp. DV consists of 199 contigs longer than 500 bp, with a total length of 4,848,582 bp. The total length of all 219 obtained contigs is 4,854,132 bp. The N50 contig size of the genome is 34,234 bp. Gene search and annotation were performed using the RAST server [12] following manual curation.

#### 3.2. Features of the *Desulfovibrio* sp. DV genome

The draft genome of *Desulfovibrio* sp. DV of 4.9 Mb is smaller by comparison to 5.07 Mb of *Desulfovibrio* sp. TomC [10] and 5.25 Mb of *Desulfovibrio magneticus* RS-1 [13], but approximately the same size as 4.8 Mb of *Desulfovibrio* cf. *magneticus* IFRC170 (NZ\_JAGC00000000). The GC content of the genome is 62.95%. The genome includes 4350 protein-coding genes, 48 tRNA genes, and 3 rRNA genes. The phylogenetic analysis of 36 ribosomal proteins showed that *Desulfovibrio* sp. TomC was the closest relative of strain DV and they both clustered with *Desulfovibrio magneticus* (Fig. 1).

Known mechanisms for the acid-tolerance have been found in the *Desulfovibrio* sp. DV genome. The K<sup>+</sup>-transporting KdpABC ATPase (genes DVDV\_0453-0449) participates in the generation of internal positive membrane potential, which prevents proton influx to the cytoplasm. Interestingly, the phylogeny of KdpABC proteins from *Desulfovibrio* is not congruent to that one inferred from ribosomal proteins. The closest relatives of K<sup>+</sup>-ATPase from strain DV were found in three strains of *Desulfovibrio magneticus*, whereas the ATPase from *Desulfovibrio* sp. TomC is more distantly related. The K<sup>+</sup>-ATPase occurs only in a few *Desulfovibrio* sp., all belonging to the “*D. magneticus*” cluster. Other K<sup>+</sup>-transporters found in the genome included: TrkA (DVDV\_0267 and DVDV\_3671); KefB (DVDV\_0709); KefA (DVDV\_0704, DVDV\_3097,

DVDV\_3865, DVDV\_4281); Kup (DVDV\_2466); Kch (DVDV\_3664); TrkH (DVDV\_3670). Some of them have orthologs not only in “*D. magneticus*” cluster, but also in other available *Desulfovibrio* genomes.

The proton-consuming decarboxylases can participate in the tolerance to low pH in the manner described for enterobacteria [18]. Lysine decarboxylase (DVDV\_0270) occurs only in the “*D. magneticus*” cluster and is likely acquired from Firmicutes via lateral gene transfer. Highly conservative arginine decarboxylase from strain DV (DVDV\_1220) has 100% amino acid sequence similarity with that from *Desulfovibrio* sp. TomC and 99% with *D. magneticus*.

In conclusion, *Desulfovibrio* sp. DV as well as *Desulfovibrio* sp. TomC and *D. magneticus* encode for multiple K<sup>+</sup>-transporters, which differ them from the rest of desulfovibrios and can enable them to tolerate low pH conditions.

#### Conflict of interest

The authors declare no conflicts of interest in this study.

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#### References

- [1] A.H. Kaksonen, M. Dopson, O.V. Karnachuk, O.H. Tuovinen, J.A. Puhakka, Biological iron oxidation and sulfate reduction in the treatment of acid mine drainage at low temperatures. in: R. Margesin, F. Schinner, J.-C. Marx, C. Gerday (Eds.), *Psychrophiles: From Biodiversity to Biotechnology*, Springer Verlag, Berlin 2008, pp. 429–454.
- [2] O.P. Ikkert, A.L. Gerasimchuk, P.A. Bukhtiyarova, O.H. Tuovinen, O.V. Karnachuk, Characterization of precipitates formed by H<sub>2</sub>S-producing, Cu-resistant Firmicute isolates of *Tissierella* from human gut and *Desulfosporosinus* from mine waste. *Antonie Van Leeuwenhoek* 103 (2013) 1221–1234.
- [3] D. Alazard, M. Joseph, F. Battaglia-Brunet, J.-L. Cayol, B. Ollivier, *Desulfosporosinus acidiphilus* sp. nov.: a moderately acidophilic sulfate-reducing bacterium isolated from acid mining drainage sediments. *Extremophiles* 14 (2010) 305–312.

- [4] I. Sánchez-Andrea, A.J.M. Stams, S. Hedrich, I. Nancuqueo, D.B. Johnson, *Desulfosporosinus acididurans* sp. nov.: an acidophilic sulfate-reducing bacterium isolated from acidic sediments. *Extremophiles* 19 (2015) 39–47.
- [5] F. Ramel, G. Brasseur, L. Pieulle, O. Valette, A. Hirschler-Réa, M.L. Fardeau, A. Dolla, Growth of the obligate anaerobe *Desulfovibrio vulgaris* Hildenborough under continuous low oxygen concentration sparging: impact of the membrane-bound oxygen reductases. *PLoS One* 10 (2015), e0123455.
- [6] J. Zhou, Q. He, C.L. Hemme, A. Mukhopadhyay, K. Hillesland, A. Zhou, Z. He, J.D. Van Nostrand, T.C. Hazen, D.A. Stahl, J.D. Wall, A.P. Arkin, How sulphate-reducing microorganisms cope with stress: lessons from system biology. *Nat. Rev. Microbiol.* 9 (2011) 452–466.
- [7] O.V. Karnachuk, S.Y. Kurochkina, D. Nicomrat, Y.A. Frank, D.A. Ivashenko, E.A. Phyllipenko, O.H. Tuovinen, Copper resistance in *Desulfovibrio* strain R2. *Antonie Van Leeuwenhoek* 83 (2003) 99–106.
- [8] O.V. Karnachuk, K. Sasaki, A.L. Gerasimchuk, O. Sukhanova, D.A. Ivashenko, A.H. Kaksonen, J.A. Puhakka, O.H. Tuovinen, Precipitation of Cu-sulfides by copper-tolerant *Desulfovibrio* isolates. *J. Geomicrobiol.* 25 (2008) 219–227.
- [9] H.K. Abicht, S. Mancini, O.V. Karnachuk, M. Solioz, Genome sequence of *Desulfosporosinus* sp. OT, an acidophilic sulfate-reducing bacterium from copper mining waste in Norilsk, Northern Siberia. *J. Bacteriol.* 193 (2011) 6104–6105.
- [10] O.V. Karnachuk, A.V. Mardanov, M.R. Avakyan, V.V. Kadnikov, M. Vlasova, A.V. Beletsky, A.L. Gerasimchuk, N.V. Ravin, Draft genome sequence of the first acid-tolerant sulfate-reducing deltaproteobacterium *Desulfovibrio* sp. TomC having potential for minewater treatment. *FEMS Microbiol. Lett.* 362 (2015) 1–3.
- [11] E.M. Möller, G. Bahnweg, H. Sandermann, H.H. Geiger, A simple and efficient protocol for isolation of high molecular weight DNA from filamentous fungi, fruit bodies, and infected plant tissues. *Nucleic Acids Res.* 20 (1992) 6115–6116.
- [12] T. Brettin, J.J. Davis, T. Disz, R.A. Edwards, S. Gerdes, G.J. Olsen, R. Olson, R. Overbeek, B. Parrello, G.D. Pusch, M. Shukla, J.A. Thomason III, R. Stevens, V. Vonstein, A.R. Wattam, F. Xia, *RASTtk*: a modular and extensible implementation of the RAST algorithm for building custom annotation pipelines and annotating batches of genomes. *Sci. Rep.* 5 (2015) 8365, <http://dx.doi.org/10.1038/srep08365>.
- [13] H. Nakazawa, A. Arakaki, S. Narita-Yamada, I. Yashiro, K. Jinno, N. Aoki, A. Tsuruyama, Y. Okamura, S. Tanikawa, N. Fujita, H. Takeyama, T. Matsunaga, Whole genome sequence of *Desulfovibrio magneticus* strain RS-1 revealed common gene clusters in magnetotactic bacteria. *Genome Res.* 19 (2009) 1801–1808.
- [14] N. Saitou, M. Nei, The neighbor-joining method: a new method for reconstructing phylogenetic trees. *Mol. Biol. Evol.* 4 (1987) 406–425.
- [15] J. Felsenstein, Confidence limits on phylogenies: an approach using the bootstrap. *Evolution* 39 (1985) 783–791.
- [16] E. Zuckerkandl, L. Pauling, Evolutionary divergence and convergence in proteins. in: V. Bryson, H.J. Vogel (Eds.), *Evolving Genes and Proteins*, Academic Press, New York 1965, pp. 97–166.
- [17] S. Kumar, G. Stecher, K. Tamura, MEGA7: molecular evolutionary genetics analysis version 7.0 for bigger datasets. *Mol. Biol. Evol.* 33 (2016) 1870–1874.
- [18] E.M. Fontenot, K.E. Ezelle, L.N. Gabreski, E.R. Giglio, J.M. McAfee, A.C. Mills, M.N. Qureshi, K.M. Salmon, C.G. Toyota, YfdW and YfdU are required for oxalate-induced acid tolerance in *Escherichia coli* K-12. *J. Bacteriol.* 195 (2013) 1446–1455.