Journal of Bacteriology

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J. Bacteriol. **2011, 193(17):4555. DOI: 10.1128/JB.05459-11. Published Ahead of Print 24 June 2011.**

JOURNAL OF BACTERIOLOGY, Sept. 2011, p. 4555–4556 Vol. 193, No. 17
0021-9193/11/$12.00 doi:10.1128/JB.05459-11

Genome Sequence of *Salinisphaera shabanensis*, a Gammaproteobacterium from the Harsh, Variable Environment of the Brine-Seawater Interface of the Shaban Deep in the Red Sea▽

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Received 4 June 2011/Accepted 13 June 2011

We present the genome of *Salinisphaera shabanensis*, isolated from a brine-seawater interface and representing a new order within the *Gammaproteobacteria*. Its adaptations to physicochemical and nutrient availability fluctuations include six genes encoding heavy metal-translocating P-type ATPases and multiple genes involved in iron uptake, siderophore production, and poly-β-hydroxybutyrate synthesis.

Salinisphaera shabanensis was isolated from the brine-seawater interface of Shaban Deep in the Red Sea (1) and represents a new order within the *Gammaproteobacteria* (4). The versatility of *S. shabanensis* is a likely advantage for and adaptation to the environmental complexity and drastic physicochemical gradients observed in this biotope.

S. shabanensis was grown under optimal conditions (1). Genomic DNA was extracted from the biomass obtained by using a blood & cell culture DNA minikit (Qiagen), following the manufacturer's instructions. The genome was sequenced using a combination of Roche 454 GS (FLX Titanium) and Illumina sequencing platforms (single and paired end). A total of 89,611,845 bp (mean read length, 280 bp) was obtained from Roche 454, providing approximately 21-fold genome coverage. Single and paired-end Illumina data provided 190,119,660 bp (mean read length, 30 bp) and 205,030,490 bp (mean read length, 35 bp), corresponding to 406-fold coverage. Roche 454 data were assembled using Newbler Assembler, version 2.5 (Roche), while Illumina data were assembled with SOAPdenovo (http://soap.genomics.org.cn/soapdenovo.html). Assemblies were merged using AMOS Minimus2 (http://sourceforge.net/apps/mediawiki/amos/index.php?title=Minimus2).

The assembly contained 69 scaffolds, with an N50 contig size (where N50 is the contig length such that at least 50% of the bases of the assembly are contained within contigs of this size or greater) of approximately 131 kb. Genes were identified using Prodigal software (5) followed by mpiBLAST (http://www.mpiblast.org/)- and EBI-Interproscan (http://www.ebi.ac.uk/InterProScan/)-based annotation. This provided annotation for 98% of all 4,110 predicted genes. Additional analysis was done using the RAST server (3). The draft genome has a G+C content of 61%.

Data from this study provide insights into how *S. shabanensis* copes with life at the brine-seawater interface and deals with the multiple stresses resulting from drastic changes in environmental conditions. As observed for other deep-sea brines of the Red Sea, Shaban Deep displays a notable increase in salinity but also in heavy metal concentration (2).

Osmoadaptation by accumulation of ectoine and betaine has been previously reported in *S. shabanensis* (1). Accordingly, our study detected the *ectABC* gene cluster, responsible for the synthesis of ectoine, *ectD*, responsible for hydroxyectoine synthesis, and genes for an almost complete pathway for betaine synthesis from choline and for multiple betaine and choline transporters. A marked increase in genes associated with K^+ metabolism and homeostasis is also probably a reflection of the need to cope with osmotic stress.

Several genes implicated in heavy metal resistance and detoxification were detected, with a high number of resistance-nodulation-cell division (RND) permeases and transporters involved in ion efflux. The detection of six genes encoding Cu/heavy metal-translocating P-type ATPases is particularly noteworthy. An average of two to three genes encoding P1-type ATPases had been previously reported, although higher numbers had been detected in microbes living in environments contaminated with heavy metals (e.g., *Cupriavidus metalliduran*s) (6). Additional detoxification genes included multiple mercuric and arsenate reductases and genes for transporters and resistance proteins involved in resistance to zinc, cobalt, cadmium, magnesium, mercury, and arsenate.

Multiple genes involved in iron uptake, siderophore production, and poly-β-hydroxybutyrate synthesis and degradation were detected, which can be seen as additional adaptations to the drastic spatial fluctuation in environmental conditions and nutrient availability.

Nucleotide sequence accession number. Nucleotide sequences are available in GenBank, project identification number 66981, with accession number AFNV00000000.

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▽ Published ahead of print on 24 June 2011.

This work was partially supported by the External Lab Access program of the King Abdullah University of Science and Technology Global Collaborative Research (GCR).

We would also like to acknowledge the support of R. Huber, G. Gmeinwieser, H. Huber, M. Thomm, and the rest of the former and current team at the Lehrstuhl für Mikrobiologie und Archaeenzentrum, University of Regensburg.

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