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### Title

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### Author

Zhou, Aifen

### Publication Date

2009-02-08

# The molecular mechanism of adaptation to salt stress revealed by the long-term evolution of *Desulfovibrio vulgaris* Hildenborough

Aifen Zhou<sup>1,6</sup>, Zhili He<sup>1,6</sup>, Marcin P. Joachimiak<sup>2,6</sup>, Paramvir S Dehal<sup>2,6</sup>, Adam P. Arkin<sup>2,6</sup>, Kristina Hillesland<sup>4,6</sup>, David Stahl<sup>4,6</sup>, Judy Wall<sup>5,6</sup>, Terry C. Hazen<sup>3,6</sup>, and Jizhong Zhou<sup>1,6</sup>

<sup>1</sup>Institute for Environmental Genomics, Department of Botany and Microbiology, University of Oklahoma, Norman, OK

<sup>2</sup>Physical Biosciences Division, Lawrence Berkeley National Laboratory, Berkeley, CA

<sup>3</sup>Earth Science Division, Lawrence Berkeley National Laboratory, Berkeley, CA

<sup>4</sup>Civil & Environmental Engineering, University of Washington, Seattle, WA

<sup>5</sup>Departments of Biochemistry and Molecular Microbiology & Immunology, University of Missouri-Columbia, Columbia, MO

<sup>6</sup>Virtual Institute for Microbial Stress and Survival, <http://vimss.lbl.gov>

## Acknowledgements

This work was part of the Virtual Institute for Microbial Stress and Survival (<http://VIMSS.lbl.gov>) supported by the U. S. Department of Energy, Office of Science, Office of Biological and Environmental Research, Genomics:GTL program through contract DE-AC02-05CH11231 between Lawrence Berkeley National Laboratory and the U. S. Department of Energy.

One of the greatest challenges in biology is to understand the interaction between genotype and environment to determine the fitness of an organism. With the recent advances in genome sequencing and high-throughput genomic technologies, now it is possible to link sub-cellular molecular/metabolic processes with the population-level processes, functions and evolution. In order to investigate the long-term evolutionary responses, diversifications and adaptation to salt stress, sulfate reducing bacteria *Desulfovibrio vulgaris* Hildenborough, a model environmental organism, is used to address such fundamental questions by mimicking the stress condition in the lab culture. Control lines and stressed lines (6 lines each, from single colony based pure culture) grown in medium LS4D and LS4D + 100 mM NaCl were transferred every 48 hrs with one to one hundred dilutions. Phenotype of all the cell lines in terms of salt tolerance was tested with LS4D supplemented with 250 mM NaCl. Results demonstrated that the adaptation to salt stress is a dynamical process. The enhanced salt tolerance of stressed lines was observed at 300 generations and became more obvious with the increase of generations. The de-adaptation experiment on 500, 1000 and 1200 generation cell lines not only provided strong evidence that the phenotype was due to the genetic change instead of physiological adaptation, but also indicated that there is also a dynamic trend for genetic adaptation and the genetic mutation might become stable at 1000 generation. In order to further understand the molecular mechanisms of adaptation to salt stress in long-term evolution process, gene expression profiles of the 500 and 1000 generation samples were examined by *D. vulgaris* whole genome oligo microarray. “Energy production and conversion” and “signal transduction mechanisms” are among the gene categories with most genes up-regulated. Statistical analysis also showed that gene expression profiling between evolved lines with salt stress vs control evolved lines and the evolved lines with salt stress vs ancestor are more closer at 1000 generation. Whole genome sequencing on selected colonies is underway to identify the beneficial genetic mutation.

## The molecular mechanism of adaptation to salt stress revealed by the long-term evolution of *Desulfovibrio vulgaris* Hildenborough

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