

# Genome Sequence of *Lentisphaera araneosa* HTCC2155<sup>T</sup>, the Type Species of the Order *Lentisphaerales* in the Phylum *Lentisphaerae*<sup>V</sup>

J. Cameron Thrash,<sup>1†</sup> Jang-Cheon Cho,<sup>2†</sup> Kevin L. Vergin,<sup>1</sup>  
Robert M. Morris,<sup>1#</sup> and Stephen J. Giovannoni<sup>1\*</sup>

Department of Microbiology, Oregon State University, Corvallis, Oregon 97331,<sup>1</sup> and Division of  
Biology and Ocean Sciences, Inha University, Incheon 402-751, Republic of Korea<sup>2</sup>

Received 26 February 2010/Accepted 18 March 2010

**Information on the genome content of deeply branching phyla with very few cultured members is invaluable for expanding understanding of microbial evolution. *Lentisphaera araneosa* HTCC2155<sup>T</sup> was isolated from the Oregon coast using dilution-to-extinction culturing. It is a marine heterotroph found in surface and mesopelagic waters in both the Pacific and Atlantic oceans and has the unusual property of producing a net-like matrix of secreted exopolysaccharide. Here we present the genome sequence of *L. araneosa* HTCC2155<sup>T</sup>, importantly, one of only two sequenced members of the phylum *Lentisphaerae*.**

The phylum *Lentisphaerae* was designated in 2004 with five isolated organisms, of which two were characterized and served as the basis for the designation of two novel orders (2). The phylum is most closely related to *Verrucomicrobia*, *Chlamydiae*, *Planctomycetes*, and the candidate division OP3; these phyla make up a recently designated monophyletic superphylum, PVC (11, 15). Within the phylum *Lentisphaerae*, clone sequences have been obtained from a variety of environments (2 [and references therein], 16). Isolated from surface waters off the Oregon coast by dilution-to-extinction culturing (3, 13), two organisms with identical 16S rRNA gene sequences were named *Lentisphaera araneosa* and make up the order *Lentisphaerales* (2). Here we present the genome sequence of the type strain, *L. araneosa* HTCC2155<sup>T</sup>.

*L. araneosa* HTCC2155<sup>T</sup> was isolated from seawater samples collected at 10 m using low-nutrient heterotrophic medium (2). The genome sequence was determined by shotgun sequencing at the J. Craig Venter Institute as part of the Moore Foundation Microbial Genome Sequencing Project (<http://www.moore.org/microgenome>). This draft unclosed genome, consisting of 81 contigs (ABCK01000001 to ABCK01000081), was analyzed with the GenDB program (7) at the Center for Genome Research and Biocomputing at Oregon State University, similarly to Previous analyses (9, 10) and through the Joint Genome Institute IMG/M website (<http://img.jgi.doe.gov/cgi-bin/pub/main.cgi>) (5). The draft genome comprises 5,173 open reading frames (ORFs), 6,023,180 bases, with a G+C content of 40.95%. Forty-nine percent of these ORFs have predicted functions. The genome is predicted to contain 55 tRNA genes, 5 5S rRNA genes, 3 16S rRNA genes, and 1 23S rRNA gene. There are putative genes for

a complete tricarboxylic acid cycle, glycolysis, the pentose phosphate pathway, and amino acid synthesis.

Notably, the *L. araneosa* genome contains 267 putative sulfatases. Since the primary described role of sulfatases is liberating sulfur from sulfate esters during sulfate deprivation (reference 14 and references therein), such a quantity of these genes in this organism is surprising, given the abundance of sulfate in marine environments. The genome of the marine planctomycete *Pirellula* sp. strain 1 contained 110 putative sulfatases which the authors hypothesized were involved in sulfur scavenging from marine snow (4).

*L. araneosa* was so named because the exopolysaccharide (EPS) secreted during growth forms a web-like matrix between cells. The genome contains several putative genes connected with EPS production—22 predicted glycosyltransferases, several of which are located two genes downstream from putative UDP-*N*-acetylglucosamine 2-epimerases (*epsC* homologs) in *eps* gene cluster-like configurations (12, 14). EPS production has been connected to psychrotolerance in several strains (1, 6, 8) and with increased pressure (6). The highest relative abundances of *L. araneosa* were measured in upper mesopelagic waters off the Oregon coast and at the Bermuda Atlantic Time Series station (2). We hypothesize that predicted EPS production genes confer increased fitness on *L. araneosa* in mid-ocean environments by stimulating the formation of aggregates (“marine snow”) or by interfering with predation.

**Nucleotide sequence accession number.** The draft genome sequence of *L. araneosa* HTCC2155<sup>T</sup> is available in GenBank under accession number ABCK00000000 and also in the Marine Microbial Genomics database at Oregon State University (<http://bioinfo.cgrb.oregonstate.edu/microbes/>).

Sequencing, assembly, annotation, and data analysis were supported by the Gordon and Betty Moore Foundation Marine Microbiology Initiative as part of its Marine Microbial Sequencing Project (<http://www.moore.org/marine-micro.aspx>), by an individual investigator award to S.J.G., and by the 21C Frontier Program of Microbial Genomics and Applications from the MEST, Republic of Korea.

We thank Scott Givan at the Oregon State University Center for Genome Research and Biocomputing for his work in developing the genome annotation software.

\* Corresponding author. Mailing address: Department of Microbiology, Oregon State University, Corvallis, OR 97331. Phone: (541) 737-1835. Fax: (541) 737-0496. E-mail: [steve.giovannoni@oregonstate.edu](mailto:steve.giovannoni@oregonstate.edu).

† These authors contributed equally to this work.

# Present address: School of Oceanography, University of Washington, Seattle, WA 98195.

<sup>V</sup> Published ahead of print on 2 April 2010.

## REFERENCES

- Allen, M. A., F. M. Lauro, T. J. Williams, D. Burg, K. S. Siddiqui, D. D. Francisci, K. W. Y. Chong, O. Pilak, H. H. Chew, M. Z. D. Maere, L. Ting, M. Katrib, C. Ng, K. R. Sowers, M. Y. Galperin, I. J. Anderson, N. Ivanova, E. Dalin, M. Martinez, A. Lapidus, L. Hauser, M. Land, T. Thomas, and R. Cavicchioli. 2009. The genome sequence of the psychrophilic archaeon, *Methanococcoides burtonii*: the role of genome evolution in cold adaptation. *ISME J.* **3**:1012–1035.
- Cho, J., K. L. Vergin, R. M. Morris, and S. Giovannoni. 2004. *Lentisphaera araneosa* gen. nov., sp. nov., a transparent exopolymer producing marine bacterium, and the description of a novel bacterial phylum, *Lentisphaerae*. *Environ. Microbiol.* **6**:611–621.
- Connon, S. A., and S. J. Giovannoni. 2002. High-throughput methods for culturing microorganisms in very-low-nutrient media yield diverse new marine isolates. *Appl. Environ. Microbiol.* **68**:3878–3885.
- Glöckner, F. O., M. Kube, M. Bauer, H. Teeling, T. Lombardot, W. Ludwig, D. Gade, A. Beck, K. Borzym, K. Heitmann, R. Rabus, H. Schlesner, R. Amann, and R. Reinhardt. 2003. Complete genome sequence of the marine planctomycete *Pirellula* sp. strain 1. *Proc. Natl. Acad. Sci. U. S. A.* **100**:8298–8303.
- Markowitz, V. M., N. N. Ivanova, E. Szeto, K. Palaniappan, K. Chu, D. Dalevi, I.-M. A. Chen, Y. Grechkin, I. Dubchak, I. Anderson, A. Lykidis, K. Mavromatis, P. Hugenholtz, and N. C. Kyrpides. 2008. IMG/M: a data management and analysis system for metagenomes. *Nucleic Acids Res.* **36**:D534–D538.
- Marx, J. G., S. D. Carpenter, and J. W. Deming. 2009. Production of cryoprotectant extracellular polysaccharide substances (EPS) by the marine psychrophilic bacterium *Colwellia psychrelythraea* strain 34H under extreme conditions. *Can. J. Microbiol.* **55**:63–72.
- Meyer, F., A. Goesmann, A. McHardy, D. Bartels, T. Bekel, J. Clausen, J. Kalinowski, B. Linke, O. Rupp, R. Giegerich, and A. PuEhler. 2003. GenDB—an open source genome annotation system for prokaryote genomes. *Nucleic Acids Res.* **31**:2187–2195.
- Nichols, C. M., J. P. Bowman, and J. Guezennec. 2005. Effects of incubation temperature on growth and production of exopolysaccharides by an Antarctic Sea ice bacterium grown in batch culture. *Appl. Environ. Microbiol.* **71**:3519–3523.
- Oh, H.-M., S. J. Giovannoni, S. Ferriera, J. Johnson, and J.-C. Cho. 2009. Complete genome sequence of *Erythrobacter litoralis* HTCC2594. *J. Bacteriol.* **191**:2419–2420.
- Oh, H.-M., S. J. Giovannoni, K. Lee, S. Ferriera, J. Johnson, and J.-C. Cho. 2009. Complete genome sequence of *Robiginitalea biformata* HTCC2501. *J. Bacteriol.* **191**:7144–7145.
- Pilhofer, M., K. Rapp, C. Eckl, A. P. Bauer, W. Ludwig, K.-H. Schleifer, and G. Petroni. 2008. Characterization and evolution of cell division and cell wall synthesis genes in the bacterial phyla *Verrucomicrobia*, *Lentisphaerae*, *Chlamydiae*, and *Planctomycetes* and phylogenetic comparison with rRNA genes. *J. Bacteriol.* **190**:3192–3202.
- Stingele, F., J. Neeser, and B. Mollet. 1996. Identification and characterization of the *eps* (exopolysaccharide) gene cluster from *Streptococcus thermophilus* Sfi6. *J. Bacteriol.* **178**:1680–1690.
- Stingl, U., H. J. Tripp, and S. J. Giovannoni. 2007. Improvements of high-throughput culturing yielded novel SAR11 strains and other abundant marine bacteria from the Oregon coast and the Bermuda Atlantic Time Series study site. *ISME J.* **1**:361–371.
- van Kranenburg, R. V., H. R. Vos, I. I. V. Swam, M. Kleerebezem, and W. Vos. 1999. Functional analysis of glycosyltransferase genes from *Lactococcus lactis* and other gram-positive cocci: complementation, expression, and diversity. *J. Bacteriol.* **181**:6347–6353.
- Wagner, M., and M. Horn. 2006. The *Planctomycetes*, *Verrucomicrobia*, *Chlamydiae* and sister phyla comprise a superphylum with biotechnological and medical relevance. *Curr. Opin. Biotechnol.* **17**:241–249.
- Zoetendal, E. G., C. M. Plugge, A. D. L. Akkermans, and W. M. de Vos. 2003. *Vicivallis vadensis* gen. nov., sp. nov., a sugar-fermenting anaerobe from human faeces. *Int. J. Syst. Evol. Microbiol.* **53**(Pt. 1):211–215.