

High-Quality Draft Genome Sequence of Low-pH-Active *Veillonella parvula* Strain SHI-1, Isolated from Human Saliva within an *In Vitro* Oral Biofilm Model

Anna Edlund,^{b,c} Quanhui Liu,^a Michael Watling,^d  Thao T. To,^a Roger E. Bumgarner,^d Xuesong He,^c Wenyuan Shi,^c Jeffrey S. McLean^a

Department of Periodontics, University of Washington, Seattle, Washington, USA^a; Microbial and Environmental Genomics, J. Craig Venter Institute, La Jolla, California, USA^b; School of Dentistry, University of California, Los Angeles, Los Angeles, California, USA^c; Department of Microbiology, University of Washington, Seattle, Washington, USA^d

We announce here a draft genome sequence of *Veillonella parvula* strain SHI-1, obtained from healthy human saliva, discovered to be active at low pH using metatranscriptomics within an *in vitro* oral biofilm model. The genome is composed of 7 contigs, for a total of 2,200,064 bp.

Received 15 December 2015 Accepted 6 January 2016 Published 18 February 2016

Citation Edlund A, Liu Q, Watling M, To TT, Bumgarner RE, He X, Shi W, McLean JS. 2016. High-quality draft genome sequence of low-pH-active *Veillonella parvula* strain SHI-1, isolated from human saliva within an *in vitro* oral biofilm model. *Genome Announc* 4(1):e01684-15. doi:10.1128/genomeA.01684-15.

Copyright © 2016 Edlund 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/).

Address correspondence to Anna Edlund, annaedlund@ucla.edu.

Oral bacteria belonging to the genus *Veillonella* are common inhabitants of biofilms on tooth surfaces, the tongue, and the buccal mucosa (1–3). *Veillonella* interacts with a broad group of bacterial taxa and is proposed to promote the growth of both health- and disease-associated bacteria (4–7). In-depth knowledge of these interactions at the gene and molecular levels is lacking. However, in one of our earlier studies of an *in vitro* biofilm community of >100 bacterial species, a previously uncultivated *Veillonella parvula* community member increased in abundance and gene transcription activity at low pH (pH 4.2 to 5.2) in response to sugar amendment (8). *V. parvula* was also previously identified and isolated from patients with chronic periodontitis (2) and severe early childhood caries (9). Here, we report the draft genome sequence of *V. parvula* strain SHI-1, isolated from a diverse *in vitro* biofilm derived from the saliva from a healthy patient (8). The *V. parvula* SHI-1 isolate and genome sequence obtained here will aid in future analysis and annotation efforts of metagenomic and transcriptomic data from the human oral microbiome.

Samples for *Veillonella* isolation were collected 9 h after glucose amendment of the *in vitro* biofilms, grown as described previously (8, 10). Selective agar plates for *Veillonella* were prepared, as described in Egland et al. (11), from Todd-Hewitt broth (THB) supplemented with 0.6% lactic acid. Plates were prepared anaerobically, and 20 μ l of resuspended biofilm sample was spread on each plate and incubated anaerobically at 37°C until colonies appeared. Individual colonies were grown in liquid THB and lactic acid under the same incubation conditions described above (without shaking) prior to DNA extraction. DNA was isolated, as described by McLean et al. (12), using the DNeasy blood and tissue kit (Qiagen, Inc., MD) and eluted in a final volume of 200 μ l of water. Taxonomic identity and culture purity were determined by partial 16S rRNA gene sequencing by Eurofin Genomics (Huntsville, AL), using forward primer 341F (CCTACGGGAGGCAGCAG) (13).

A paired-end Illumina library was prepared from a DNA extract of a 16S-validated growth culture and sequenced on a MiSeq

sequencer (Illumina, Inc.) (2 \times 300 bp). All quality-trimmed reads were *de novo* assembled using SPAdes version 3.61 (14, 15). The contigs were inspected for k-mer frequency consistency, and 7 scaffolds were used for further analyses.

The draft genome is 2.2 Mb, with an overall G+C content of 38.7%. Gene annotation using the Prokaryotic Genome Automatic Annotation Pipeline (PGAAP), provided by the National Center for Biotechnology Information (NCBI), identified a total of 2,055 genes, consisting of 2,005 coding sequences, 45 tRNAs, one small subunit (SSU) 16S rRNA, one large subunit (LSU) 23S rRNA, and 3 SSU 5S rRNAs. The average nucleotide identity (ANI) (16) between SHI-1 and its closest phylogenetic neighbor, ATCC 17748, is 96.51%.

Nucleotide sequence accession numbers. This whole-genome shotgun project has been deposited at DDBJ/EMBL/GenBank under the accession no. [LOJO00000000](https://accession.gtrdb.org/acc/LOJO00000000). The version described in this paper is version LOJO01000000.

ACKNOWLEDGMENTS

This work was supported by grants from the National Institutes of Health, 1R01GM095373 and 1R01DE020102, to J.S.M., and a K99 Pathway to Independence Award (grant K99DE024543) to A.E.

FUNDING INFORMATION

HHS | National Institutes of Health (NIH) provided funding to Anna Edlund under K99 Pathway to Independence Award K99DE024543. HHS | National Institutes of Health (NIH) provided funding to Jeffrey S. McLean under grant number 1R01GM095373. HHS | National Institutes of Health (NIH) provided funding to Jeffrey S. McLean under grant number 1R01DE020102.

REFERENCES

1. Aas JA, Paster BJ, Stokes LN, Olsen I, Dewhirst FE. 2005. Defining the normal bacterial flora of the oral cavity. *J Clin Microbiol* 43:5721–5732. <http://dx.doi.org/10.1128/JCM.43.11.5721-5732.2005>.
2. Mashima I, Nakazawa F. 2015. Draft genome sequence of *Veillonella*

- toetsuensis* ATCC BAA-2400T isolated from human tongue biofilm. Genome Announc 3(4):e00808-15. <http://dx.doi.org/10.1128/genomeA.00808-15>.
3. Arif N, Do T, Byun R, Sheehy E, Clark D, Gilbert SC, Beighton D. 2008. *Veillonella rogosae* sp. nov., an anaerobic, Gram-negative coccus isolated from dental plaque. Int J Syst Evol Microbiol 58:581–584. <http://dx.doi.org/10.1099/ijs.0.65093-0>.
 4. Periasamy S, Kolenbrander PE. 2009. *Aggregatibacter actinomycetem-comitans* builds mutualistic biofilm communities with *Fusobacterium nucleatum* and *Veillonella* species in saliva. Infect Immun 77:3542–3551. <http://dx.doi.org/10.1128/IAI.00345-09>.
 5. Periasamy S, Kolenbrander PE. 2010. Central role of the early colonizer *Veillonella* sp. in establishing multispecies biofilm communities with initial, middle, and late colonizers of enamel. J Bacteriol 192:2965–2972. <http://dx.doi.org/10.1128/JB.01631-09>.
 6. Kolenbrander PE, Palmer RJ, Jr, Rickard AH, Jakubovics NS, Chalmers NI, Diaz PI. 2006. Bacterial interactions and successions during plaque development. Periodontol 2000 42:47–79. <http://dx.doi.org/10.1111/j.1600-0757.2006.00187.x>.
 7. Palmer RJ, Jr, Diaz PI, Kolenbrander PE. 2006. Rapid succession within the *Veillonella* population of a developing human oral biofilm *in situ*. J Bacteriol 188:4117–4124. <http://dx.doi.org/10.1128/JB.01958-05>.
 8. Edlund A, Yang Y, Yooseph S, Hall AP, Nguyen DD, Dorrestein PC, Nelson KE, He X, Lux R, Shi W, McLean JS. 2015. Meta-omics uncover temporal regulation of pathways across oral microbiome genera during *in vitro* sugar metabolism. ISME J 9:2605–2619 <http://dx.doi.org/10.1038/ismej.2015.72>.
 9. Tanner AC, Mathney JM, Kent RL, Jr, Chalmers NI, Hughes CV, Loo CY, Pradhan N, Kanasi E, Hwang J, Dahlan MA, Papadopoulou E, Dewhirst FE. 2011. Cultivable anaerobic microbiota of severe early childhood caries. J Clin Microbiol 49:1464–1474. <http://dx.doi.org/10.1128/JCM.02427-10>.
 10. Edlund A, Yang Y, Hall AP, Guo L, Lux R, He X, Nelson KE, Nealon KH, Yooseph S, Shi W, McLean JS. 2013. An *in vitro* biofilm model system maintaining a highly reproducible species and metabolic diversity approaching that of the human oral microbiome. Microbiome 1:25. <http://dx.doi.org/10.1186/2049-2618-1-25>.
 11. Eglund PG, Palmer RJ, Jr, Kolenbrander PE. 2004. Interspecies communication in *Streptococcus gordonii*-*Veillonella atypica* biofilms: signaling in flow conditions requires juxtaposition. Proc Natl Acad Sci USA 101:16917–16922. <http://dx.doi.org/10.1073/pnas.0407457101>.
 12. McLean JS, Fansler SJ, Majors PD, McAteer K, Allen LZ, Shirliff ME, Lux R, Shi W. 2012. Identifying low pH active and lactate-utilizing taxa within oral microbiome communities from healthy children using stable isotope probing techniques. PLoS One 7:e32219. <http://dx.doi.org/10.1371/journal.pone.0032219>.
 13. Teske A, Sigalevich P, Cohen Y, Muyzer G. 1996. Molecular identification of bacteria from a coculture by denaturing gradient gel electrophoresis of 16S ribosomal DNA fragments as a tool for isolation in pure cultures. Appl Environ Microbiol 62:4210–4215.
 14. 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. <http://dx.doi.org/10.1089/cmb.2012.0021>.
 15. Nurk S, Bankevich A, Antipov D, Gurevich AA, Korobeynikov A, Lapidus A, Prjibelski AD, Pyshkin A, Sirotkin A, Sirotkin Y, Stepanauskas R, Clingenpeel SR, Woyke T, McLean JS, Lasken R, Tesler G, Alekseyev MA, Pevzner PA. 2013. Assembling single-cell genomes and mini-metagenomes from chimeric MDA products. J Comput Biol 20:714–737. <http://dx.doi.org/10.1089/cmb.2013.0084>.
 16. Varghese NJ, Mukherjee S, Ivanova N, Konstantinidis KT, Mavrommatis K, Kyrpides NC, Pati A. 2015. Microbial species delineation using whole genome sequences. Nucleic Acids Res 43:6761–6771. <http://dx.doi.org/10.1093/nar/gkv657>.