

Draft Genome Sequence of Low-Passage Clinical Isolate *Porphyromonas gingivalis* MP4-504

Thao T. To,^a Quanhui Liu,^a Michael Watling,^b Roger E. Bumgarner,^b Richard P. Darveau,^a Jeffrey S. McLean^{a*}

Department of Periodontics, University of Washington School of Dentistry, Seattle, Washington, USA^a; Department of Microbiology, University of Washington, Seattle, Washington, USA^b

* Present address: Jeffrey S. McLean, 1959 NE Pacific St., Seattle, Washington, USA.

We present the draft genome of *Porphyromonas gingivalis* MP4-504, a low-passage clinical isolate obtained from a periodontitis patient. The genome is composed of 92 contigs for a length of 2,373,453 bp and a G+C of 48.3%. The *traA-Q* conjugative transfer locus is genetically distinct from W83 but highly similar to ATCC 33277.

Received 22 February 2016 Accepted 24 February 2016 Published 7 April 2016

Citation To TT, Liu Q, Watling M, Bumgarner RE, Darveau RP, McLean JS. 2016. Draft genome sequence of low-passage clinical isolate *Porphyromonas gingivalis* MP4-504. *Genome Announc* 4(2):e00256-16. doi:10.1128/genomeA.00256-16.

Copyright © 2016 To 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 Jeffrey S. McLean, jsmclean@uw.edu.

The Gram-negative oral anaerobe *Porphyromonas gingivalis* is highly associated with periodontal disease and inflammation (1, 2). Considered a “keystone pathogen,” *P. gingivalis* contributes to development and stabilization of a dysbiotic microbial community, leading to chronic inflammatory responses indicative of periodontitis (3–5). However, the various genetic mechanisms of this species are not fully understood and increased availability of genomic sequences is needed. Here, we report the draft genome sequence of low-passage clinical isolate MP4-504, which properties include stable adherence to oral streptococci (6), enhanced invasion of gingival epithelial cells (GECs) (7), strong inhibition of IL-8 production by GECs (8), and the ability to transfer DNA by conjugation at high efficiencies (9).

MP4-504 was originally sampled from the University of Washington Graduate Periodontics Clinic from the periodontal pocket (8-mm probing depth) of a chronic periodontitis patient and immediately transported to anaerobic conditions. The sample was then serially diluted on blood agar plates for bacterial isolation and anaerobically incubated at 35°C for 7 days before preliminary biochemical identification (10) and storage in a –80°C freezer collection.

For this study, isolate MP4-504 was grown as previously described and subjected to two additional passages beyond the primary freezer stock (7). Genomic DNA was extracted using the Qiagen DNeasy blood and tissue kit. Paired-end 300 bp reads were sequenced using the Illumina MiSeq platform. All quality-trimmed reads were *de novo* assembled using SPAdes v3.61 using default parameters (11, 12).

The final assembly consists of 92 contigs with a length of 2,373,453 bp (N_{50} , 57,689 bp) and overall G+C content of 48.3%. Gene annotation using the Prokaryotic Genome Automatic Annotation Pipeline (PGAAP) provided by the National Center for Biotechnology Information (NCBI) identified a total of 2,070 genes, consisting of 1,891 coding sequences, 47 tRNAs, 3 rRNAs, and 3 clustered regularly interspaced short palindromic repeats (CRISPRs). MP4-504 shares 98.84% average nucleotide identity

(ANI) (13) with closest phylogenetic neighbor, W83 (14), and 98.67% ANI with type strain ATCC 33277 (15). MP4-504 also shares 98.62% ANI with JCVI SC001, a *P. gingivalis* isolate recovered from a hospital sink biofilm (16).

MP4-504 is also capable of transferring shuttle vectors of *Bacteroides-Escherichia coli* origin to *E. coli* by conjugation, similar to ATCC 33277 and unlike type strain W83 (9, 17). Consistent with these findings, comparative analysis of the MP4-504 draft genome using the Rapid Annotation and Subsystem Technology (RAST) (18) server against the genomes of other *P. gingivalis* strains revealed that MP4-504 bears a similar genomic region to cTnPg1 reported in ATCC 33277 (17), with similarity of 93.3% at the protein level. When compared to W83, this MP4-504 *traA-Q* region bears only 42.8% similarity. Likewise, this region contains a gene encoding conjugative transposon protein, TraP, necessary for the conjugation of plasmids in ATCC 33277 but missing in W83. Further comparative analysis with other available *P. gingivalis* genomes will expand our understanding of genetic mechanisms underlying this bacterium’s pathogenesis.

Nucleotide sequence accession numbers. This whole-genome shotgun project has been deposited at DDBJ/EMBL/GenBank under the accession no. [LOEL00000000](https://www.ncbi.nlm.nih.gov/nuclink/LOEL00000000). The version described in this paper is version [LOEL01000000](https://www.ncbi.nlm.nih.gov/nuclink/LOEL01000000).

ACKNOWLEDGMENTS

This work was supported by National Institutes of Health grants R01DE023810 and R01DE020102 to J.S.M. and T90DE21984 to T.T.T.

REFERENCES

- Socransky SS, Haffajee AD, Cugini MA, Smith C, Kent RL. 1998. Microbial complexes in subgingival plaque. *J Clin Periodontol* 25: 134–144. <http://dx.doi.org/10.1111/j.1600-051X.1998.tb02419.x>.
- Socransky SS, Haffajee AD. 2005. Periodontal microbial ecology. *Periodontol* 38:135–187. <http://dx.doi.org/10.1111/j.1600-0757.2005.00107.x>.
- Hajishengallis G, Darveau RP, Curtis MA. 2012. The keystone-pathogen hypothesis. *Nat Rev Microbiol* 10:717–725. <http://dx.doi.org/10.1038/nrmicro2873>.

4. Darveau RP. 2010. Periodontitis: a polymicrobial disruption of host homeostasis. *Nat Rev Microbiol* 8:481–490. <http://dx.doi.org/10.1038/nrmicro2337>.
5. Darveau RP, Hajishengallis G, Curtis MA. 2012. *Porphyromonas gingivalis* as a potential community activist for disease. *J Dent Res* 91:816–820.
6. Lamont RJ, Hersey SG, Rosan B. 1992. Characterization of the adherence of *Porphyromonas gingivalis* to oral streptococci. *Oral Microbiol Immunol* 7:193–197. <http://dx.doi.org/10.1111/j.1399-302X.1992.tb00024.x>.
7. Lamont RJ, Chan A, Belton CM, Izutsu KT, Vasel D, Weinberg A. 1995. *Porphyromonas gingivalis* invasion of gingival epithelial cells. *Infect Immun* 63:3878–3885.
8. Darveau RP, Belton CM, Reife RA, Lamont RJ. 1998. Local chemokine paralysis, a novel pathogenic mechanism for *Porphyromonas gingivalis*. *Infect Immun* 66:1660–1665.
9. Tribble GD, Lamont GJ, Progulsk-Fox A, Lamont RJ. 2007. Conjugal transfer of chromosomal DNA contributes to genetic variation in the oral pathogen *Porphyromonas gingivalis*. *J Bacteriol* 189:6382–6388. <http://dx.doi.org/10.1128/JB.00460-07>.
10. Moncla BJ, Motley ST, Braham P, Ewing L, Adams TH, Vermeulen NMJ. 1991. Use of synthetic oligonucleotide DNA probes for identification and direct detection of *Bacteroides forsythus* in plaque samples. *J Clin Microbiol* 29:2158–2162.
11. 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.
12. 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>.
13. Varghese NJ, Mukherjee S, Ivanova N, Konstantinidis KT, Mavromatis 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>.
14. Nelson KE, Fleischmann RD, DeBoy RT, Paulsen IT, Fouts DE, Eisen JA, Daugherty SC, Dodson RJ, Durkin AS, Gwinn M, Haft DH, Kolonay JF, Nelson WC, Mason T, Tallon L, Gray J, Granger D, Tettelin H, Dong H, Galvin JL, Duncan MJ, Dewhirst FE, Fraser CM. 2003. Complete genome sequence of the oral pathogenic bacterium *Porphyromonas gingivalis* strain W83. *J Bacteriol* 185:5591–5601. <http://dx.doi.org/10.1128/JB.185.18.5591-5601.2003>.
15. Naito M, Hirakawa H, Yamashita A, Ohara N, Shoji M, Yukitake H, Nakeyama K, Toh H, Yoshimura F, Kuhara S, Hattori M, Hayashi T, Nakayama K. 2008. Determination of the genome sequence of *Porphyromonas gingivalis* strain ATCC 33277 and genomic comparison with strain W83 revealed extensive genome rearrangements in *P. gingivalis*. *DNA Res* 15:215–225.
16. McLean JS, Lombardo MJ, Ziegler MG, Novotny M, Yee-Greenbaum J, Badger JH, Tesler G, Nurk S, Lesin V, Bami D, Hall AP, Edlund A, Allen LZ, Durkin S, Reed S, Torriani F, Neelson KH, Pevzner PA, Friedman R, Venter JC, Lasken RS. 2013. Genome of the pathogen *Porphyromonas gingivalis* recovered from a biofilm in a hospital sink using a high-throughput single-cell genomics platform. *Genome Res* 23:867–877. <http://dx.doi.org/10.1101/gr.150433.112>.
17. Tribble GD, Kerr JE, Wang B. 2013. Genetic diversity in the oral pathogen *Porphyromonas gingivalis*: molecular mechanisms and biological consequences. *Future Microbiol* 8:607–620. <http://dx.doi.org/10.2217/fmb.13.30>.
18. Overbeek R, Olson R, Pusch GD, Olsen GJ, Davis JJ, Disz T, Edwards RA, Gerdes S, Parrello B, Shukla M, Vonstein V, Wattam AR, Xia F, Stevens R. 2014. The SEED and the Rapid Annotation of microbial genomes using Subsystems Technology (RAST). *Nucleic Acids Res* 42:D206–D214. <http://dx.doi.org/10.1093/nar/gkt1226>.