



Complete Genome Assembly of a Quality Control Reference Isolate, Moraxella catarrhalis Strain ATCC 25240

H. E. Daligault,^a K. W. Davenport,^a T. D. Minogue,^b K. A. Bishop-Lilly,^{c,d} D. C. Bruce,^a P. S. Chain,^a S. R. Coyne,^b K. G. Frey,^{c,d} J. Jaissle,^b G. I. Koroleva,^e J. T. Ladner,^e C.-C. Lo,^a L. Meincke,^a C. Munk,^a G. F. Palacios,^e C. L. Redden,^{c,d} S. L. Johnson^a

Los Alamos National Laboratory, Los Alamos, New Mexico, USA^a; Diagnostic Systems Division, USAMRIID, Fort Detrick, Maryland, USA^b; Naval Medical Research Center, NMRC-Frederick, Fort Detrick, Maryland, USA^c; Henry M. Jackson Foundation, Bethesda, Maryland, USA^d; Center for Genome Sciences, USAMRIID, Fort Detrick, Maryland, USA^e

Generally an opportunistic pathogen in the United States, *Moraxella catarrhalis* has acquired resistance to multiple antibacterial/antimicrobial agents. Here, we present the complete 1.9-Mb genome of *M. catarrhalis* strain ATCC 25240, as deposited in NCBI under the accession number CP008804.

Received 18 August 2014 Accepted 20 August 2014 Published 18 September 2014

Citation Daligault HE, Davenport KW, Minogue TD, Bishop-Lilly KA, Bruce DC, Chain PS, Coyne SR, Frey KG, Jaissle J, Koroleva GI, Ladner JT, Lo C-C, Meincke L, Munk C, Palacios GF, Redden CL, Johnson SL. 2014. Complete genome assembly of a quality control reference isolate, *Moraxella catarrhalis* strain ATCC 25240. Genome Announc. 2(5):e00938-14. doi:10.1128/genomeA.00938-14.

Copyright © 2014 Daligault et al. This is an open-access article distributed under the terms of the Creative Commons Attribution 3.0 Unported license.

Address correspondence to S. L. Johnson, shannonj@lanl.gov.

Moraxella catarrhalis, previously known as Branhamella catarrhalis, is the causative agent of many upper respiratory tract infections in the United States. *M. catarrhalis* often exacerbates chronic obstructive pulmonary disease (COPD) and may cause nearly one-fifth of bacterial ear infections in the United States (1–3). Infections are generally limited to children or elderly people; however, the pathogen exhibits high-level β -lactamase resistance, making it a concern for immunocompromised adults (4, 5). To increase the number of reference genomes for diagnostic development and phylogenetic reconstructions (as of this writing only one complete genome is available in public databases), we sequenced and assembled the genome of *M. catarrhalis* strain 25240 into a single closed chromosome (6).

High quality genomic DNA was extracted from a purified isolate using QIAgen Genome Tip-500 at USAMRIID's Diagnostic Systems Division (DSD). Specifically, a 100-mL bacterial culture was grown to stationary phase and nucleic acid extracted as per the manufacturer's recommendations. Sequence data were generated using a combination of Illumina and 454 technologies (7, 8). For this genome assembly, we constructed and sequenced an Illumina library of 100-bp reads to high coverage (300-fold genomecoverage) as well as a separate long-insert paired-end library (average insert size 7,431 \pm 1,858 bp, run on the Roche 454 Titanium platform to 17-fold genome coverage). The two libraries were assembled together in Newbler (Roche) and the consensus sequences computationally shredded into 2-kbp overlapping fake reads (shreds). The raw reads were also assembled in Velvet and those consensus sequences computationally shredded into 1.5kbp overlapping shreds (9). Draft data from all platforms were then assembled together with ALLPATHS and the consensus sequences computationally shredded into 10-kbp overlapping shreds (10). We then integrated the Newbler consensus shreds, Velvet consensus shreds, ALLPATHS consensus shreds, and a subset of the long-insert read-pairs using parallel Phrap (High Performance Software, LLC). Possible misassemblies were corrected and some gap closure was accomplished with manual editing in Consed (11–13).

Automatic annotation for the *M. catarrhalis* ATCC 25240 genome utilized an Ergatis-based workflow at LANL with minor manual curation. Annotation located 1,742 coding genes, 50 tRNAs, and 12 rRNAs. The final 1,941,566-bp assembly has 41.5% G+C content. Preliminary review of the annotated genome suggests that over 30 drug resistance genes are present in the genome.

Nucleotide sequence accession number. The final sequence has been deposited to GenBank under the accession number CP008804.

ACKNOWLEDGMENTS

Funding for this effort was provided by the Defense Threat Reduction Agency's Joint Science and Technology Office (DTRA J9-CB/JSTO). This manuscript is approved by LANL for unlimited release (LA-UR-14-25174).

The views expressed in this article are those of the authors and do not necessarily reflect the official policy or position of the Department of the Navy, Department of Defense, or the U.S. Government.

REFERENCES

- de Vries SPW, van Hijum SAFT, Schueler W, Riesbeck K, Hays JP, Hermans PWM, Bootsma HJ. 2010. Genome analysis of *Moraxella catarrhalis* strain RH4, a human respiratory tract pathogen. J. Bacteriol. 192:3574–3583. http://dx.doi.org/10.1128/JB.00121-10.
- Vergison A. 2008. Microbiology of otitis media: a moving target. Vaccine 26(Suppl 7):G5–G10. http://dx.doi.org/10.1016/j.vaccine.2008.11.006.
- Wang W, Reitzer L, Rasko DA, Pearson MM, Blick RJ, Laurence C, Hansen EJ. 2007. Metabolic analysis of *Moraxella catarrhalis* and the effect of selected *in vitro* growth conditions on global gene expression. Infect. Immun. 75:4959–4971. http://dx.doi.org/10.1128/IAI.00073-07.
- Thornsberry C, Sahm DF, Kelly LJ, Critchley IA, Jones ME, Evangelista AT, Karlowsky JA. 2002. Regional trends in antimicrobial resistance among clinical isolates of *Streptococcus pneumoniae*, *Haemophilus influenzae*, and *Moraxella catarrhalis* in the United States: results from the TRUST surveillance program, 1999–2000. Clin. Infect. Dis. 34(Suppl 1): S4–S16. http://dx.doi.org/10.1086/324525.

- Verduin CM, Hol C, Fleer A, van Dijk H, van Belkum A. 2002. Moraxella catarrhalis: from emerging to established pathogen. Clin. Microbiol. Rev. 15: 125–144. http://dx.doi.org/10.1128/CMR.15.1.125-144.2002.
- de Vries SPW, Bootsma HJ, Hays JP, Hermans PWM. 2009. Molecular aspects of *Moraxella catarrhalis* pathogenesis. Microbiol. Mol. Biol. Rev. 73:389–406. http://dx.doi.org/10.1128/MMBR.00007-09.
- 7. Margulies M, Egholm M, Altman WE, Attiya S, Bader JS, Bemben LA, Berka J, Braverman MS, Chen Y-J, Chen Z, Dewell SB, Du L, Fierro JM, Gomes XV, Godwin BC, He W, Helgesen S, Ho CH, Irzyk GP, Jando SC, Alenquer MLI, Jarvie TP, Jirage KB, Kim J-B, Knight JR, Lanza JR, Leamon JH, Lefkowitz SM, Lei M, Li J, Lohman KL, Lu H, Makhijani VB, McDade KE, McKenna MP, Myers EW, Nickerson E, Nobile JR, Plant R, Puc BP, Ronan MT, Roth GT, Sarkis GJ, Simons JF, Simpson JW, Srinivasan M, Tartaro KR, Tomasz A, Vogt KA, Volkmer GA, Wang SH, Wang Y, Weiner MP, Yu P, Begley RF, Rothberg JM. 2005. Genome sequencing in microfabricated high-density picolitre reactors. Nature 437:376–380. http://dx.doi.org/10.1038/nature03959.
- Bennett S. 2004. Solexa Ltd. Pharmacogenomics 5:433–438. http:// dx.doi.org/10.1517/14622416.5.4.433.
- Zerbino DR, Birney E. 2008. Velvet: algorithms for *de novo* short read assembly using de Bruijn graphs. Genome Res. 18:821–829. http:// dx.doi.org/10.1101/gr.074492.107.
- Butler J, MacCallum I, Kleber M, Shlyakhter IA, Belmonte MK, Lander ES, Nusbaum C, Jaffe DB. 2008. ALLPATHS: *de novo* assembly of wholegenome shotgun microreads. Genome Res. 18:810–820. http:// dx.doi.org/10.1101/gr.7337908.
- Ewing B, Hillier L, Wendl MC, Green P. 1998. Base-calling of automated sequencer traces using Phred. I: accuracy assessment. Genome Res. 8:175–185. http://dx.doi.org/10.1101/gr.8.3.175.
- Ewing B, Green P. 1998. Base-calling of automated sequencer traces using Phred. II: error probabilities. Genome Res. 8:186–194.
- Gordon D, Abajian C, Green P. 1998. Consed: a graphical tool for sequence finishing. Genome Res. 8:195–202. http://dx.doi.org/10.1101/ gr.8.3.195.