



## Closed Genome Sequence of *Escherichia coli* K-12 Group Strain C600

Anna Allué-Guardia,<sup>a,b</sup> Emmanuel C. Nyong,<sup>a</sup> Sara S. K. Koenig,<sup>a,b</sup> Sean M. Vargas,<sup>a</sup> James L. Bono,<sup>c</sup> Mark Eppinger<sup>a,b</sup>

<sup>a</sup>Department of Biology, University of Texas at San Antonio, San Antonio, Texas, USA <sup>b</sup>South Texas Center for Emerging Infectious Diseases (STCEID), San Antonio, Texas, USA <sup>c</sup>U.S. Department of Agriculture, Agricultural Research Service (USDA-ARS), Clay Center, Nebraska, USA

**ABSTRACT** *Escherichia coli* strain C600 is a prototypical K-12 derived laboratory strain which has been broadly used for molecular microbiology and bacterial physiology studies since its isolation in 1954. Here, we present the closed genome sequence of *E. coli* strain C600, retrieved from the American Type Culture Collection (ATCC 23724).

**E** scherichia coli strain C600 (Migula) Castellani and Chalmers (ATCC 23724) is a bacteriophage  $\lambda$ -sensitive strain first described in 1954 (1) and derived from progenitor strain K-12, which was originally isolated in 1922 (2). Since its isolation in 1954, strain C600 has been used as a model *E. coli* strain in laboratories worldwide for molecular microbiology, genetic, and physiology studies (3, 4). Its genome can incorporate foreign DNA by transformation and conjugation (5, 6). Strain C600 is also susceptible to phage transduction and serves as a prime bacterial host in bacteriophage infectivity assays, producing stable lysogens (7, 8). Recovered Shiga-toxigenic lysogens in the C600 genome background that were transduced by Shiga toxin (Stx)-converting bacteriophages have shown an increased ability to produce Shiga toxin under spontaneous noninduced conditions (8) and when cocultured with Stx-producing *E. coli* (9, 10).

*E. coli* strain C600 was cultured overnight in Luria-Bertani broth (37°C, 180 rpm), and total genomic DNA was extracted using the QIAamp DNA minikit (Qiagen), following the manufacturer's instructions. To generate a high-quality closed genome sequence, we pursued a hybrid approach using long-read PacBio RS II and short-read Illumina MiSeq sequencing. Briefly, for PacBio sequencing, genomic DNA was sheared into 20-kb fragments using a g-Tube device (Covaris), and a library was prepared based on the 20-kb PacBio sample preparation protocol and sequenced using P6/C4 chemistry on a single-molecule real-time (SMRT) cell with a 240-min collection time. The 113,777 long reads ( $N_{50} = 7,800$ ) were assembled *de novo* using the PacBio Hierarchical Genome Assembly Process (HGAP v3.0) (11) with default parameters in SMRT Analysis v2.3.0 and consensus polishing with Quiver (11). For Illumina sequencing, a paired-end 2 × 250-bp library was prepared using the Nextera XT DNA library preparation kit (Illumina) and sequenced using the 500-cycle MiSeq reagent kit v2 (Illumina), yielding a total of 697,116 paired-end reads. Illumina reads were used for sequence error correction of the PacBio assembly, using Pilon v1.18 with default settings (12).

The assembly yielded a circular chromosome of 4,599,824 bp, with a coverage of  $107 \times$  and a GC content of 50.8%. The chromosomal origin of replication, *oriC*, was designated the start point (+1) of the closed molecule and determined using the Web-based tool Ori-Finder (13). Genome annotation with the NCBI Prokaryotic Genome Annotation Pipeline (PGAP) (14) predicted a total of 4,636 protein-coding sequences, 22 rRNAs, 86 tRNAs, and 15 noncoding RNAs (ncRNAs).

Citation Allué-Guardia A, Nyong EC, Koenig SSK, Vargas SM, Bono JL, Eppinger M. 2019. Closed genome sequence of *Escherichia coli* K-12 group strain C600. Microbiol Resour Announc 8:e01052-18. https://doi.org/10.1128/ MRA.01052-18.

Editor Julia A. Maresca, University of Delaware Copyright © 2019 Allué-Guardia et al. This is an open-access article distributed under the terms of the Creative Commons Attribution 4.0 International license.

Address correspondence to Mark Eppinger, mark.eppinger@utsa.edu.

Received 22 August 2018 Accepted 6 December 2018 Published 10 January 2019 Strain C600 belongs to sequence type 10 (ST10), as inferred from MLST (v2.0) (15) using the Achtman scheme (16). Prophage profiling with PHASTER identified six prophage-associated regions, including three intact prophages and three incomplete phage remnants (17). *In silico* antimicrobial susceptibility testing using ResFinder (v3.0) with default settings detected no antibiotic resistance genes (18).

**Data availability.** The annotated chromosome has been deposited in NCBI GenBank under the accession number CP031214. Illumina and PacBio reads are available under the accession numbers SRX4909245 and SRX4908799, respectively, in the Sequence Read Archive (SRA).

## ACKNOWLEDGMENTS

This work was supported by the South Texas Center of Emerging Infectious Diseases (STCEID), the Department of Biology at the University of Texas at San Antonio, the National Institute of Allergy and Infectious Diseases of the National Institutes of Health (SC2AI120941), and the U.S. Department of Homeland Security (2014-ST-062-000058). The contributions of J.L.B. were funded by the U.S. Department of Agriculture, Agricultural Research Service (CRIS project 3040-42000-015-00D).

We thank Sandy Fryda-Bradley and Kristin Kuhn at the USDA for providing technical assistance.

The mention of a trade name, proprietary product, or specific equipment does not constitute a guarantee or warranty by the USDA and does not imply approval to the exclusion of other products that might be suitable.

## REFERENCES

- 1. Appleyard RK. 1954. Segregation of new lysogenic types during growth of a doubly lysogenic strain derived from *Escherichia coli* K12. Genetics 39:440–452.
- Bachmann BJ. 1996. Derivations and genotypes of some mutant derivatives of *Escherichia coli* K-12, p 2460–2488. *In* Neidhardt FC (ed), Escherichia coli and Salmonella, 2nd ed. ASM Press, Washington, DC.
- 3. Lederberg J, Tatum EL. 1946. Gene recombination in *Escherichia coli*. Nature 158:558.
- 4. Lennox ES. 1955. Transduction of linked genetic characters of the host by bacteriophage P1. Virology 1:190–206.
- Brambach BC, Michels A, Franzke J, Kettler R. 2013. Current density and conductivity dependent electroporation of *Escherichia coli* C600. Prog Biophys Mol Biol 111:46–54. https://doi.org/10.1016/j.pbiomolbio.2012 .12.006.
- Starodub ME, Trevors JT. 1990. Mobilization of *Escherichia coli* R1 silverresistance plasmid pJT1 by Tn5-Mob into *Escherichia coli* C600. Biol Met 3:24–27.
- Serra-Moreno R, Acosta S, Hernalsteens JP, Jofre J, Muniesa M. 2006. Use of the lambda Red recombinase system to produce recombinant prophages carrying antibiotic resistance genes. BMC Mol Biol 7:31. https://doi.org/10.1186/1471-2199-7-31.
- Iversen H, LA-L TM, Aspholm M, Arnesen LP, Lindback T. 2015. Commensal *E. coli* Stx2 lysogens produce high levels of phages after spontaneous prophage induction. Front Cell Infect Microbiol 5:5. https://doi .org/10.3389/fcimb.2015.00005.
- Goswami K, Chen C, Xiaoli L, Eaton KA, Dudley EG. 2015. Coculture of *Escherichia coli* 0157:H7 with a nonpathogenic *E. coli* strain increases toxin production and virulence in a germfree mouse model. Infect Immun 83:4185–4193. https://doi.org/10.1128/IAI.00663-15.
- Xiaoli L, Figler HM, Goswami Banerjee K, Hayes CS, Dudley EG. 2018. Non-pathogenic *Escherichia coli* enhance Stx2a production of *E. coli* O157:H7 through both *bamA*-dependent and independent mechanisms. Front Microbiol 9:1325. https://doi.org/10.3389/fmicb.2018.01325.

- Chin CS, Alexander DH, Marks P, Klammer AA, Drake J, Heiner C, Clum A, Copeland A, Huddleston J, Eichler EE, Turner SW, Korlach J. 2013. Nonhybrid, finished microbial genome assemblies from long-read SMRT sequencing data. Nat Methods 10:563–569. https://doi.org/10.1038/ nmeth.2474.
- Walker BJ, Abeel T, Shea T, Priest M, Abouelliel A, Sakthikumar S, Cuomo CA, Zeng Q, Wortman J, Young SK, Earl AM. 2014. Pilon: an integrated tool for comprehensive microbial variant detection and genome assembly improvement. PLoS One 9:e112963. https://doi.org/10.1371/journal .pone.0112963.
- Gao F, Zhang CT. 2008. Ori-Finder: a Web-based system for finding *oriCs* in unannotated bacterial genomes. BMC Bioinformatics 9:79. https://doi .org/10.1186/1471-2105-9-79.
- Tatusova T, DiCuccio M, Badretdin A, Chetvernin V, Nawrocki EP, Zaslavsky L, Lomsadze A, Pruitt KD, Borodovsky M, Ostell J. 2016. NCBI Prokaryotic Genome Annotation Pipeline. Nucleic Acids Res 44: 6614–6624. https://doi.org/10.1093/nar/gkw569.
- Larsen MV, Cosentino S, Rasmussen S, Friis C, Hasman H, Marvig RL, Jelsbak L, Sicheritz-Ponten T, Ussery DW, Aarestrup FM, Lund O. 2012. Multilocus sequence typing of total-genome-sequenced bacteria. J Clin Microbiol 50:1355–1361. https://doi.org/10.1128/JCM.06094-11.
- Wirth T, Falush D, Lan R, Colles F, Mensa P, Wieler LH, Karch H, Reeves PR, Maiden MC, Ochman H, Achtman M. 2006. Sex and virulence in *Escherichia coli*: an evolutionary perspective. Mol Microbiol 60:1136–1151. https://doi.org/10.1111/j.1365-2958.2006.05172.x.
- Arndt D, Grant JR, Marcu A, Sajed T, Pon A, Liang Y, Wishart DS. 2016. PHASTER: a better, faster version of the PHAST phage search tool. Nucleic Acids Res 44:W16–W21. https://doi.org/10.1093/nar/gkw387.
- Zankari E, Hasman H, Cosentino S, Vestergaard M, Rasmussen S, Lund O, Aarestrup FM, Larsen MV. 2012. Identification of acquired antimicrobial resistance genes. J Antimicrob Chemother 67:2640–2644. https://doi .org/10.1093/jac/dks261.