



## Draft Genome Sequence of Multidrug-Resistant Strain Citrobacter portucalensis MBTC-1222, Isolated from Uziza (Piper guineense) Leaves in Nigeria

Etinosa O. Igbinosa, a,b Jana Rathie, a Diana Habermann, Erik Brinks, a Gyu-Sung Cho, a Charles M.A.P. Franza

<sup>a</sup>Department of Microbiology and Biotechnology, Max Rubner-Institut, Kiel, Germany

**ABSTRACT** In this work, we report the draft whole-genome sequence of the multiply antibiotic-resistant *Citrobacter portucalensis* strain MBTC-1222 isolated from the uziza leafy vegetable in Nigeria. Sequence analysis showed the assembled genome size to be 4,881,935 bp, containing 4,603 protein-coding genes, 131 pseudogenes, 7 rRNAs, 74 tRNAs, and 9 noncoding RNAs (ncRNAs).

"itrobacter portucalensis is a Gram-negative facultative anaerobic bacterium belonging to the family Enterobacteriaceae, and the type strain, Citrobacter portucalensis A60<sup>T</sup>, was isolated from water in Portugal (1). Members of the genus Citrobacter occur in the intestines of humans and animals, and due to consequent fecal shedding, they can also be found in varied environments, such as water, soil, and sewage. Some Citrobacter species strains have antibiotic resistance genes, i.e., the beta-lactamase gene ampC and guinolone resistance gene qnrB, on their chromosomal DNA (2, 3). The closely related species Citrobacter freundii is an emerging opportunistic pathogen and is known to cause infections involving the urinary, gastrointestinal, or respiratory tract (4). In addition, multidrug-resistant C. freundii strains have been isolated from various vegetables, such as lettuce, salad, and ready-to-eat-salad (5, 6). In this work, we studied the antibiotic resistance of C. portucalensis MBTC-1222, isolated from uziza leaves (also known as ashanti pepper [Piper guineense]), in Nigeria using whole-genome sequencing. So far, to our knowledge, a genome sequence of a highly multidrug-resistant C. portucalensis strain isolated from vegetables has not been available. It is therefore imperative to identify and characterize antimicrobial resistance genes of C. portucalensis strains associated with vegetables.

The total genomic DNA of C. portucalensis MBTC-1222 was isolated using the peqGOLD bacterial DNA kit (Peglab, Erlangen, Germany). The sequencing library was prepared with an Illumina Nextera XT library prep kit (Illumina, San Diego, CA, USA) and run on the MiSeq platform (Illumina) with  $2 \times 250$  paired-end reads. In total, 943,130 paired-end and 39,707 single-end sequence reads were obtained with approximately 54-fold coverage. The reads were de novo assembled using SPAdes version 3.11.1 (7). The draft genome assembly consisted of 49 contigs, and the  $N_{50}$  value was 330,308 bp. The genome size of C. portucalensis is 4,881,935 bp, with a 52.03 mol% G+C content. The genome sequence was annotated using the Rapid Annotations using Subsystems Technology (RAST) and NCBI servers (8). It contained 4,603 protein-coding sequences, 7 rRNAs, 74 tRNAs, and 9 noncoding RNAs (ncRNAs). The comparison of antibiotic resistance genes between C. portucalensis A60<sup>T</sup> and C. portucalensis MBTC-1222 was confirmed using the ResFinder server (version 2.1) (9). Both C. portucalensis strains carry genes bla<sub>CMY</sub> and qnrB5, which encode resistance to beta-lactam antibiotics and fluoroquinolone antibiotics, respectively. The isolate C. portucalensis MBTC-1222 contains further antibiotic resistance genes, including those for aminoglycosides

**Received** 30 January 2018 **Accepted** 6 February 2018 **Published** 1 March 2018

Citation Igbinosa EO, Rathje J, Habermann D, Brinks E, Cho G-S, Franz CMAP. 2018. Draft genome sequence of multidrug-resistant strain Citrobacter portucalensis MBTC-1222, isolated from uziza (Piper guineense) leaves in Nigeria. Genome Announc 6:e00123-18. https://doi.org/10.1128/genomeA.00123-18.

Copyright © 2018 Igbinosa et al. This is an open-access article distributed under the terms of the Creative Commons Attribution 4.0 International license.

Address correspondence to Gyu-Sung Cho, gyusung.cho@mri.bund.de.

<sup>&</sup>lt;sup>b</sup>Department of Microbiology, Faculty of Life Sciences, University of Benin, Benin City, Nigeria

lgbinosa et al. genameAmouncements™

(strA and strB), beta-lactams (bla<sub>TEM</sub>), chloramphenicol (catA2), sulfonamide (sul2), tetracycline (tetA), and trimethoprim (dfrA14). The nucleotide sequence similarities of all antibiotic resistance genes were higher than 95% compared to the GenBank database.

**Accession number(s).** The whole-genome shotgun project has been deposited at DDB/ENA/GenBank under the accession no. PJEP01000000.

## **ACKNOWLEDGMENTS**

E. O. Igbinosa gratefully acknowledges the Alexander von Humboldt Stiftung's Georg Forster Stipendium for Experienced Researchers for financing his research stay at the Max Rubner-Institut (MRI).

Adrian Prager, Jessica Obermeyer, and Lena Niclausen are acknowledged for their technical assistance.

## **REFERENCES**

- Ribeiro TG, Gonçalves BR, da Silva MS, Novais Â, Machado E, Carriço JA, Peixe L. 2017. Citrobacter portucalensis sp. nov., isolated from an aquatic sample. Int J Syst Evol Microbiol 67:3513–3517. https://doi.org/10.1099/ ijsem.0.002154.
- Ribeiro TG, Novais A, Branquinho R, Machado E, Peixe L. 2015. Phylogeny and comparative genomics unveil independent diversification trajectories of *qnrB* and genetic platforms within particular *Citrobacter* species. Antimicrob Agents Chemother 59:5951–5958. https://doi.org/10.1128/AAC 00027-15
- Porres-Osante N, Sáenz Y, Somalo S, Torres C. 2015. Characterization of beta-lactamases in faecal *Enterobacteriaceae* recovered from healthy humans in Spain: Focusing on *AmpC* polymorphisms. Microb Ecol 70: 132–140. https://doi.org/10.1007/s00248-014-0544-9.
- 4. Lavigne JP, Defez C, Bouziges N, Mahamat A, Sotto A. 2007. Clinical and molecular epidemiology of multidrug-resistant *Citrobacter* spp. infections in a French university hospital. Eur J Clin Microbiol Infect Dis 26:439 441. https://doi.org/10.1007/s10096-007-0315-3.
- van Hoek AH, Veenman C, van Overbeek WM, Lynch G, de Roda Husman AM, Blaak H. 2015. Prevalence and characterization of ESBL- and AmpC-

- producing *Enterobacteriaceae* on retail vegetables. Int J Food Microbiol 204:1–8. https://doi.org/10.1016/j.ijfoodmicro.2015.03.014.
- Campos J, Mourão J, Pestana N, Peixe L, Novais C, Antunes P. 2013. Microbiological quality of ready-to-eat salads: an underestimated vehicle of bacteria and clinically relevant antibiotic resistance genes. Int J Food Microbiol 166:464–470. https://doi.org/10.1016/j.ijfoodmicro.2013.08.005.
- 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. https://doi.org/10.1089/cmb.2012.0021.
- Aziz RK, Bartels D, Best AA, DeJongh M, Disz T, Edwards RA, Formsma K, Gerdes S, Glass EM, Kubal M, Meyer F, Olsen GJ, Olson R, Osterman AL, Overbeek RA, McNeil LK, Paarmann D, Paczian T, Parrello B, Pusch GD, Reich C, Stevens R, Vassieva O, Vonstein V, Wilke A, Zagnitko O. 2008. The RAST server: rapid annotations using subsystems technology. BMC Genomics 9:75. https://doi.org/10.1186/1471-2164-9-75.
- Zankari E. 2014. Comparison of the web tools ARG-ANNOT and ResFinder for detection of resistance genes in bacteria. Antimicrob Agents Chemother 58:4986. https://doi.org/10.1128/AAC.02620-14.