

Draft Genome Sequences of Three Newly Identified Species in the Genus Cronobacter, C. helveticus LMG23732^T, C. pulveris LMG24059, and C. zurichensis LMG23730^T

Naqash Masood,^a Karen Moore,^b Audrey Farbos,^b Sumyya Hariri,^a Konrad Paszkiewicz,^b Ben Dickins,^a Alan McNally,^a Stephen Forsythe^a

Pathogen Research Group, School of Science and Technology, Nottingham Trent University, Nottingham, United Kingdom^a; Wellcome Trust Biomedical Informatics Hub, Biosciences, University of Exeter, Exeter, United Kingdom^b

Cronobacter helveticus, Cronobacter pulveris, and Cronobacter zurichensis are newly described species in the Cronobacter genus, which is associated with serious infections of neonates. This is the first report of draft genome sequences for these species.

Received 29 August 2013 Accepted 30 August 2013 Published 26 September 2013

Citation Masood N, Moore K, Farbos A, Hariri S, Paszkiewicz K, Dickins B, McNally A, Forsythe S. 2013. Draft genome sequences of three newly identified species in the genus Cronobacter, C. helveticus LMG23732^T, C. pulveris LMG24059, and C. zurichensis LMG23730^T. Genome Announc. 1(5):e00783-13. doi:10.1128/genomeA.00783-13.

Copyright © 2013 Masood et al. This is an open-access article distributed under the terms of the Creative Commons Attribution 3.0 Unported license.

Address correspondence to Stephen Forsythe, stephen.forsythe@ntu.ac.uk.

"he Cronobacter genus until recently consisted of seven species, C. sakazakii, C. malonaticus, C. turicensis, C. universalis, C. muytjensii, C. dublinensis, and C. condimenti (1). In 2013, three new Cronobacter species were proposed, C. zurichensis, C. pulveris, and C. helveticus (2). Due to the association of Cronobacter with fatal neonatal infections there is an international requirement for powdered infant formula to be microbiologically tested for all members of the Cronobacter genus (Codex Alimentarius Commission, code of hygienic practice for powdered formulae for infants and young children, http://www.codexalimentarius.net/ download/standards/11026/CXP_066e.pdf). Therefore, the genome sequencing of these newly described species is warranted for better understanding of the genus diversity and improved detection methodology. This was undertaken using C. helveticus strain LMG23732^T, C. zurichensis strain LMG23730^T, and C. pulveris strain LMG24059 isolated from powdered infant formula.

Bacterial DNA was extracted from 1-day cultures using a GenElute bacterial genome kit (Sigma-Aldrich) and sequenced using an Illumina HiSeq 2500 sequencing system. *De novo* assembly was performed using Velvet (3). For further annotation we used the SEED-based automated annotation system provided by the RAST server (4).

C. helveticus LMG23732^T, *C. pulveris* LMG24059, and *C. zurichensis* LMG23730^T generated 8,351,512, 5,000,582, and 6,613,368 high-quality paired-end reads of 150 bp in length, respectively. The genome sizes were 4,530,369 bp, 4,900,556 bp, and 4,246,797 bp with G+C contents of 56%, 56.6%, and 57.8%, respectively. These were in 79, 125, and 103 contigs with 4,315, 4,630, and 3,883 coding sequences (CDS), respectively, and 30-fold coverage. The CDS include traits previously described in *Cronobacter* (5). These included genes associated with iron acquisition, stress response, heavy metal resistance (arsenic, copper cobalt, zinc and cadmium), and phages. Genes encoding proteins associated with several virulence-associated traits such as adhesion and antibiotic resistance, including fluoroquinolones, fosfomycin, β-lactamases, and multidrug efflux pumps, were also determined.

Nucleotide sequence accession numbers.The genome sequences of *C. helveticus*, *C. pulveris*, and *C. zurichensis* have been deposited in GenBank under the accession numbers AWFX00000000, AWFY00000000, and AWFZ000000000, respectively.

ACKNOWLEDGMENTS

We thank Nottingham Trent University for their financial support of N.M. and the Amm-AlQura University for their financial support of S.H.

REFERENCES

- 1. Joseph S, Cetinkaya E, Drahovska H, Levican A, Figueras MJ, Forsythe SJ. 2012. *Cronobacter condimenti* sp. nov., isolated from spiced meat, and *Cronobacter universalis* sp. nov., a species designation for *Cronobacter* sp. genomospecies 1, recovered from a leg infection, water, and food ingredients. Int. J. Syst. Evol. Microbiol. 62:1277–1283.
- 2. Brady C, Cleenwerck I, Venter S, Coutinho T, De Vos P. 2013. Taxonomic evaluation of the genus Enterobacter based on multilocus sequence analysis (MLSA): proposal to reclassify E. nimipressuralis and E. amnigenus into Lelliottia gen. nov. as Lelliottia nimipressuralis comb. nov. and Lelliottia amnigena comb. nov., respectively, E. gergoviae and E. pyrinus into Pluralibacter gen. nov. as Pluralibacter gergoviae comb. nov. and Pluralibacter pyrinus comb. nov., respectively, E. cowanii, E. radicincitans, E. oryzae and E. arachidis into Kosakonia gen. nov. as Kosakonia cowanii comb. nov., Kosakonia radicincitans comb. nov., Kosakonia oryzae comb. nov. and Kosakonia arachidis comb. nov., respectively, and E. turicensis, E. helveticus and E. pulveris into Cronobacter as Cronobacter zurichensis nom. nov., Cronobacter helveticus comb. nov. and Cronobacter pulveris comb. nov., respectively, and emended description of the genera Enterobacter and Cronobacter. Syst. Appl. Microbiol. 36:309–319.
- 3. Zerbino DR, Birney E. 2008. Velvet: algorithms for *de novo* short read assembly using de Bruijn graphs. Genome Res. 18:821–829.
- 4. 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. doi:10.1186/1471-2164-9-75.
- Joseph S, Desai P, Ji Y, Cummings CA, Shih R, Degoricija L, Rico A, Brzoska P, Hamby SE, Masood N, Hariri S, Sonbol H, Chuzhanova N, McClelland M, Furtado MR, Forsythe SJ. 2012. Comparative analysis of genome sequences covering the seven *Cronobacter* species. PLoS One 7:e49455.