

Complete Genome Sequence of Rahnella aquatilis CIP 78.65

Robert J. Martinez,^a David Bruce,^b Chris Detter,^b Lynne A. Goodwin,^b James Han,^c Cliff S. Han,^b Brittany Held,^b Miriam L. Land,^d Natalia Mikhailova,^c Matt Nolan,^c Len Pennacchio,^c Sam Pitluck,^c Roxanne Tapia,^b Tanja Woyke,^c and Patricia A. Sobecky^a

Department of Biological Sciences, University of Alabama, Tuscaloosa, Alabama, USA^a; Los Alamos National Laboratory, Bioscience Division, Los Alamos, New Mexico, USA^b; US DOE Joint Genome Institute, Walnut Creek, California, USA^c; and Oak Ridge National Laboratory, Bioscience Division, Oak Ridge, Tennessee, USA^d

Rahnella aquatilis CIP 78.65 is a gammaproteobacterium isolated from a drinking water source in Lille, France. Here we report the complete genome sequence of *Rahnella aquatilis* CIP 78.65, the type strain of *R. aquatilis*.

Rahnella aquatilis CIP 78.65 (CUETM 77-115, CDC 1327-79, DSM 4594, ATCC 33071) is a gammaproteobacterium isolated from a drinking water source in Lille, France (7, 15). Strains belonging to the genus *Rahnella* have been isolated from soil, freshwater, food, and clinical samples. As a result, members of this genus have been studied in plant biology as they relate to plant growth and nutrient acquisition (13, 16, 18, 26, 27), food sciences (4, 17, 23, 24), medical microbiology (1, 10, 12), and more recently, in studies that demonstrate the potential use of *Rahnella* isolates for bioremediation (2, 8, 19, 21, 25). Therefore, whole-genome sequencing was conducted on *Rahnella aquatilis* CIP 78.65 to support current research that utilizes this strain as well as to support comparative genomics studies.

The complete genome of Rahnella aquatilis CIP 78.65 was generated at the DOE Joint Genome Institute (JGI) using a combination of Illumina (3) and 454 technologies (20). For this genome, an Illumina GAII shotgun library (79,438,616 reads totaling 6,037.3 Mb), a 454 Titanium standard library (335,154 reads), and a paired-end 454 library (370,318 reads totaling 202.7 Mb) were generated and sequenced. All general aspects of library construction and sequencing can be found on the JGI website (http://www .jgi.doe.gov/). The initial draft assembly contained 50 contigs in 2 scaffolds. The 454 Titanium standard data and the 454 paired-end data were assembled together with Newbler, version 2.3. Illumina sequencing data were assembled with Velvet, version 1.0.13 (28). We integrated the 454 Newbler consensus shreds, the Illumina Velvet consensus shreds, and the read pairs in the 454 paired-end library using parallel Phrap, version SPS-4.24 (High Performance Software, LLC). The software Consed (5, 6, 9) was used in the following finishing process. Illumina data were used to correct potential base errors and increase consensus quality using the software Polisher, developed at JGI (Alla Lapidus, unpublished). Possible misassemblies were corrected using gapResolution (Cliff Han, unpublished) or Dupfinisher (11) or by sequencing subcloned PCR products. Gaps between contigs were closed by editing in Consed, by PCR, and by Bubble PCR (J.-F. Cheng, unpublished) primer walks. Protein-encoding genes were determined by Prodigal (14) and GenePRIMP (22). The total size of the genome is 5,447,000 bp, and the final assembly is based on 176 Mb of 454 draft data (32.6× coverage) and 5,847 Mb of Illumina draft data $(1,083 \times \text{coverage}).$

The *Rahnella aquatilis* CIP 78.65 genome consists of a 4,861,101-bp (G+C content, 52.12%) circular chromosome and three plasmids, pRahaq201, pRahaq202, and pRahaq203, of 463,906 bp (G+C content, 52.29%), 115,487 bp (G+C content, 50.69%), and 8,604 bp (paired-end data indicate a circular plas-

mid with a gap spanning a known length of 1,950 bp), respectively. The genome contains 4,970 protein-encoding genes, 104 pseudogenes, 76 tRNA genes, and 7 rRNA gene operons.

Nucleotide sequence accession numbers. The complete genome sequence of *Rahnella aquatilis* CIP 78.65 is available in DDBJ/EMBL/GenBank under the accession number CP003244, in the IMG database under 2506520038, and in the Genomes OnLine Database under Gc02077. The GenBank accession numbers for plasmids pRahaq201, pRahaq202, and pRahaq203 are CP003245, CP003246, and CP003247, respectively.

ACKNOWLEDGMENTS

The work conducted by the U.S. Department of Energy Joint Genome Institute is supported by the Office of Science of the U.S. Department of Energy under contract no. DE-AC02-05CH11231 and U.S. Department of Energy grant no. DE-FG02-04ER63906.

REFERENCES

- 1. Alballaa SR, et al. 1992. Urinary tract infection due to *Rahnella aquatilis* in a renal transplant patient. J. Clin. Microbiol. **30**:2948–2950.
- 2. Beazley MJ, et al. 2009. Nonreductive biomineralization of uranium(VI) phosphate via microbial phosphatase activity in anaerobic conditions. Geomicrobiol. J. 26:431–441.
- 3. Bennett, S. 2004. Solexa Ltd. Pharmacogenomics 5:433-438.
- Ercolini D, et al. 2006. Changes in the spoilage-related microbiota of beef during refrigerated storage under different packaging conditions. Appl. Environ. Microbiol. 72:4663–4671.
- Ewing B, Green P. 1998. Base-calling of automated sequencer traces using phred. II. Error probabilities. Genome Res. 8:186–194.
- Ewing B, et al. 1998. Base-calling of automated sequencer traces using phred. I. Accuracy assessment. Genome Res. 8:175–185.
- Gavini F, et al. 1976. Taxonomic study of enterobacteria belonging or related to genus *Enterobacter*. Ann. Microbiol. (Paris) 127B:317–335.
- 8. Geissler A, et al. 2009. Biogeochemical changes induced in uranium mining waste pile samples by uranyl nitrate treatments under anaerobic conditions. Geobiology 7:282–294.
- 9. Gordon D, et al. 1998. Consed: a graphical tool for sequence finishing. Genome Res. 8:195–202.
- Goubau P, et al. 1988. Septicemia caused by *Rahnella aquatilis* in an immunocompromised patient. Eur. J. Clin. Microbiol. Infect. Dis. 7:697– 699.
- 11. Han CS, Chain P. 2006. Finishing repetitive regions automatically with Dupfinisher, p 142–147. *In* Arabnia HR, Valafar H (ed), Proceedings of

Received 9 March 2012 Accepted 22 March 2012 Address correspondence to Robert J. Martinez, rmartinez@ua.edu. Copyright © 2012, American Society for Microbiology. All Rights Reserved.

doi:10.1128/JB.00380-12

the 2006 International Conference on Bioinformatics and Computational Biology. CSREA Press, Las Vegas, NV.

- Harrell LJ, et al. 1989. Rahnella aquatilis, an unusual Gram-negative rod isolated from the bronchial washing of a patient with acquired immunodeficiency syndrome. J. Clin. Microbiol. 27:1671–1672.
- 13. Heulin T, et al. 1994. *Bacillus polymyxa* and *Rahnella aquatilis*, the dominant N2-fixing bacteria associated with wheat rhizosphere in French soils. Eur. J. Soil Biol. **30**:35–42.
- Hyatt D, et al. 2010. Prodigal: prokaryotic gene recognition and translation initiation site identification. BMC Bioinformatics 11:119.
- 15. Izard D, et al. 1979. *Rahnella aquatilis*, new member of the *Enterobacteriaceae*. Ann. Microbiol. (Paris) **130A**:163–177.
- Izumi H, et al. 2006. Endobacteria in some ectomycorrhiza of Scots pine (*Pinus sylvestris*). FEMS Microbiol. Ecol. 56:34–43.
- Kang SA, et al. 2004. Secretory production of *Rahnella aquatilis* ATCC 33071 levansucrase expressed in *Escherichia coli*. J. Microbiol. Biotechnol. 14:1232–1238.
- Kim KY, et al. 1997. *Rahnella aquatilis*, a bacterium isolated from soybean rhizosphere, can solubilize hydroxyapatite. FEMS Microbiol. Lett. 153: 273–277.
- Kourtev PS, et al. 2006. Responses of the anaerobic bacterial community to addition of organic C in chromium(VI)- and iron(III)-amended microcosms. Appl. Environ. Microbiol. 72:628–637.

- Margulies M, et al. 2005. Genome sequencing in microfabricated highdensity picolitre reactors. Nature 437:376–380.
- Martinez RJ, et al. 2007. Aerobic uranium(VI) bioprecipitation by metalresistant bacteria isolated from radionuclide- and metal-contaminated subsurface soils. Environ. Microbiol. 9:3122–3133.
- 22. Pati A, et al. 2010. GenePRIMP: a gene prediction improvement pipeline for prokaryotic genomes. Nat. Methods 7:455–457.
- 23. Raphael E, et al. 2011. Extended spectrum beta-lactamase gene sequences in Gram-negative saprophytes on retail organic and nonorganic spinach. Appl. Environ. Microbiol. 77:1601–1607.
- 24. Ruimy R, et al. 2010. Organic and conventional fruits and vegetables contain equivalent counts of Gram-negative bacteria expressing resistance to antibacterial agents. Environ. Microbiol. 12:608–615.
- Valverde A, et al. 2011. Diversity and community structure of culturable arsenic-resistant bacteria across a soil arsenic gradient at an abandoned tungsten-tin mining area. Chemosphere 85:129–134.
- Vyas P, et al. 2010. Cold-adapted and rhizosphere-competent strain of *Rahnella* sp. with broad-spectrum plant growth-promotion potential. J. Microbiol. Biotechnol. 20:1724–1734.
- Warmink JA, et al. 2009. Universal and species-specific bacterial 'fungiphiles' in the mycospheres of different basidiomycetous fungi. Environ. Microbiol. 11:300–312.
- 28. Zerbino DR, Birney E. 2008. Velvet: algorithms for de novo short read assembly using de Bruijn graphs. Genome Res. 18:821–829.