Journal of Bacteriology

Complete Genome Sequence of Clinical Isolate Pantoea ananatis LMG 5342

Pieter De Maayer, Wai Yin Chan, Fabio Rezzonico, Andreas Bühlmann, Stephanus N. Venter, Jochen Blom, Alexander Goesmann, Jürg E. Frey, Theo H. M. Smits, Brion Duffy and Teresa A. Coutinho *J. Bacteriol.* 2012, 194(6):1615. DOI: 10.1128/JB.06715-11.

	Updated information and services can be found at: http://jb.asm.org/content/194/6/1615
REFERENCES	<i>These include:</i> This article cites 14 articles, 8 of which can be accessed free at: http://jb.asm.org/content/194/6/1615#ref-list-1
CONTENT ALERTS	Receive: RSS Feeds, eTOCs, free email alerts (when new articles cite this article), more»

Information about commercial reprint orders: http://journals.asm.org/site/misc/reprints.xhtml To subscribe to to another ASM Journal go to: http://journals.asm.org/site/subscriptions/





Complete Genome Sequence of Clinical Isolate Pantoea ananatis LMG 5342

Pieter De Maayer,^{a,b} Wai Yin Chan,^{a,b} Fabio Rezzonico,^c Andreas Bühlmann,^c Stephanus N. Venter,^{a,b} Jochen Blom,^d Alexander Goesmann,^d Jürg E. Frey,^c Theo H. M. Smits,^c Brion Duffy,^c and Teresa A. Coutinho^{a,b}

Forestry and Agricultural Biotechnology Institute (FABI)^a and Department of Microbiology and Plant Pathology,^b University of Pretoria, Pretoria, South Africa; Agroscope Changins-Wädenswil (ACW), Plant Protection Division, Wädenswil, Switzerland^c; and Computational Genomics, CeBiTec, Bielefeld University, Bielefeld, Germany^d

The enterobacterium *Pantoea ananatis* is an ecologically versatile species. It has been found in the environment, as plant epiphyte and endophyte, as an emerging phytopathogen, and as a presumptive, opportunistic human pathogen. Here, we report the complete genome sequence of *P. ananatis* LMG 5342, isolated from a human wound.

embers of the genus Pantoea are Gram-negative, motile rods belonging to the family *Enterobacteriaceae* and display ecological versatility, as they are commonly recovered from water, soil, insects, and plants (11, 14). Pantoea ananatis is associated primarily with plants, as epiphyte or endophyte, and as an emerging phytopathogen infecting a wide range of important crop and forest plants (e.g., maize, onion, rice, pineapple, Eucalyptus) (4). P. ananatis has also been reported as an occasional clinical isolate and a presumptive opportunistic human pathogen, associated with septicemia following penetrating trauma with plant material, nosocomial infections due to exposure to contaminated hospital materials, and secondary complications of preexisting illnesses (5, 11, 12). Enterobacter agglomerans LMG 5342 (ATCC 29920; CDC6070-69) was isolated from a human wound in Georgia (3, 7) and has been taxonomically reassigned to P. ananatis (9, 11). Here, we describe the complete genome sequence of P. ananatis LMG 5342, the first clinical isolate of this species and genus to be sequenced.

Whole-genome pyrosequencing using a single run on the Roche 454 GS-Junior sequencer (454 Life Sciences) yielded 165,189 high-quality reads with an average read length of 430 bp (total of 72,654,851 bases), providing 15-times genome coverage. *De novo* assembly using Newbler assembler version 2.5 (454 Life Sciences) yielded 68 contigs which were further assembled using the Lasergene package, version 8.1 (DNAstar, Madison, WI), and by scaffolding against the reference genome of the *Eucalyptus* phytopathogen *P. ananatis* LMG 20103 (6). PCR amplification and Sanger sequencing were performed to close all gaps. Protein coding sequences (CDS) were predicted (8) using Glimmer (13) and Critica (1). The GenDB annotation pipeline (10) was used for automated function assignment, followed by manual optimization.

The genome of *P. ananatis* LMG 5342 consists of a 4,604-kb chromosome with a G+C content of 53.4% and a 303-kb circular plasmid with 51.5% G+C. Primers based on the LMG 5342 plasmid were utilized for gap closure of the homologous plasmid in the *Eucalyptus* pathogen *P. ananatis* LMG 20103, which was previously misassembled into the chromosome (6). A total of 4,675 CDS, seven ribosomal DNA operons, and 77 tRNAs are encoded on the LMG 5342 genome. Comparative genomic analysis using EDGAR (2) indicated that the genome of the clinical strain is highly conserved both in synteny and gene content with that of LMG 20103 (6), with 3,843 (82.2%) orthologous CDS and 833 (17.8%) singletons. Some of these singletons may potentially be

linked to human pathogenesis. Detailed genome comparisons and functional analyses will be performed to characterize these factors. The genome sequence of *P. ananatis* LMG 5342 thus represents an extensive resource to broaden our understanding of *P. ananatis* plant and human associations.

Nucleotide sequence accession numbers. The *P. ananatis* LMG 5342 chromosome and plasmid sequences are deposited in EMBL under accession numbers HE617160 (chromosome) and HE617161 (pPANA10 plasmid).

ACKNOWLEDGMENTS

We thank M. Oggenfuss and B. Frey (ACW) for technical support.

This study was funded by the National Research Foundation (NRF), the Tree Protection Cooperative Programme (TPCP), the NRF/Department of Science and Technology Centre of Excellence in Tree Health Biotechnology (CTHB), the THRIP support program of the Department of Trade and Industry, South Africa, and the Swiss Federal Office of Agriculture (FOAG Project Fire Blight Pathogen).

This work was conducted in part within the European research network COST Action 873.

REFERENCES

- 1. Badger JH, Olsen GJ. 1999. CRITICA: coding region identification tool invoking comparative analysis. Mol. Biol. Evol. 16:512–524.
- 2. Blom J, et al. 2009. EDGAR: a software framework for the comparative analysis of prokaryotic genomes. BMC Bioinform. 10:154.
- Brenner DJ, Fanning GR, Leete Knutson JK, Steigerwalt AG, Krichevsky MI. 1984. Attempts to classify herbicola group-*Enterobacter* agglomerans strains by deoxyribonucleic acid hybridization and phenotypic tests. Int. J. Syst. Bacteriol. 34:45–55.
- Coutinho TA, Venter SN. 2009. Pantoea ananatis: an unconventional plant pathogen. Mol. Plant Pathol. 10:325–335.
- 5. De Baere T, et al. 2004. Bacteremic infection with *Pantoea ananatis*. J. Clin. Microbiol. 42:4393–4395.
- De Maayer P, et al. 2010. Genome sequence of *Pantoea ananatis* LMG20103, the causative agent of *Eucalyptus* blight and dieback. J. Bacteriol. 192:2936–2937.
- 7. Lind E, Ursing J. 1986. Clinical strains of Enterobacter agglomerans (syn-

Received 13 December 2011 Accepted 4 January 2012

Address correspondence to Pieter De Maayer, Pieter.DeMaayer@fabi.up.ac.za. P.D.M. and W.Y.C. contributed equally to this publication.

Copyright © 2012, American Society for Microbiology. All Rights Reserved. doi:10.1128/JB.06715-11

onyms: *Erwinia herbicola, Erwinia milletiae*) identified by DNA-DNA hybridization. Acta Pathol. Microbiol. Immunol. Scand. B **94**:205–213.

- McHardy AC, Goesmann A, Pühler A, Meyer F. 2004. Development of joint application strategies for two microbial gene finders. Bioinformatics 20:1622–1631.
- Mergaert J, Verdonck L, Kersters K. 1993. Transfer of Erwinia ananas (synonym, Erwinia uredovora) and Erwinia stewartii to the genus Pantoea emend. as Pantoea ananas (Serrano 1928) comb. nov. and Pantoea stewartii (Smith 1898) comb. nov., respectively, and description of Pantoea stewartii subsp. indologenes subsp. nov. Int. J. Syst. Bacteriol. 43:162–173.
- Meyer F, et al. 2003. GenDB—an open source genome annotation system for prokaryote genomes. Nucleic Acids Res. 31:2187–2195.
- Rezzonico F, Smits THM, Montesinos E, Frey JE, Duffy B. 2009. Genotypic comparison of *Pantoea agglomerans* plant and clinical strains. BMC Microbiol. 9:204.
- Rezzonico F, Stockwell VO, Tonolla M, Duffy B, Smits THM. 2011. Pantoea clinical isolates cannot be accurately assigned to species based on metabolic profiling. Transpl. Infect. Dis. [Epub ahead of print.] doi: 10.1111/j.1399-3062.2011.00684.x.
- Salzberg SL, Delcher AL, Kasif S, White O. 1998. Microbial gene identification using interpolated Markov models. Nucleic Acids Res. 26:544– 548.
- Smits THM, et al. 2011. Metabolic versatility and antibacterial metabolite biosynthesis are distinguishing genomic features of the fire blight antagonist *Pantoea vagans* C9-1. PLoS One 6:e22247.