

# Genome Sequence of *Pseudomonas stutzeri* Strain JM300 (DSM 10701), a Soil Isolate and Model Organism for Natural Transformation

## Antonio Busquets,<sup>a</sup> Arantxa Peña,<sup>a</sup> Margarita Gomila,<sup>b</sup> Rafael Bosch,<sup>a</sup> Balbina Nogales,<sup>a</sup> Elena García-Valdés,<sup>a,c</sup> Jorge Lalucat,<sup>a,c</sup> and Antoni Bennasar<sup>a,d</sup>

Microbiologia, Departament de Biologia, Universitat de les Illes Balears, Campus UIB, Palma de Mallorca, Spain<sup>a</sup>; Hospital Son Llàtzer, Palma de Mallorca, Spain<sup>b</sup>; Institut Mediterrani d'Estudis Avançats (IMEDEA, CSIC-UIB), Esporles, Spain<sup>c</sup>; and Instituto Universitario de Investigaciones en Ciencias de la Salud (IUNICS-UIB), Universitat de les Illes Balears, Campus UIB, Palma de Mallorca, Spain<sup>d</sup>

## *Pseudomonas stutzeri* strain JM300 (DSM 10701) is a denitrifying soil isolate and a model organism for natural transformation in bacteria. Here we report the first complete genome sequence of JM300, the reference strain of genomovar 8 for the species.

**P**seudomonas stutzeri is a species of extremely broad phenotypic and genotypic diversity. Eighteen genomic and phylogenetic groupings (genomovars) are recognized within the species (4, 9). *P. stutzeri* strain JM300 (DSM 10701) was derived as a smoothcolony variant of a strain obtained from the collection of C. C. Delwiche of the University of California. That strain was isolated in 1980 from an anaerobic enrichment from soil with succinate and nitrous oxide (1).

Natural transformation is perhaps the most versatile mechanism of horizontal gene transfer (7), and *P. stutzeri* can be considered a naturally transformable bacterium, as one-third of its members have this feature (2, 6, 12). Many relevant advances in the biochemical characterization of natural transformation have been achieved with *P. stutzeri* JM300, a strain considered a model system for this process.

So far, whole-genome sequences of three *P. stutzeri* strains of genomovar 1 and one strain of genomovar 2 are publically available (3, 10, 13, 14). Analysis of strain JM300, the reference strain of genomovar 8, will help us to gain insight into the evolution of the species by analyzing a member of a different genomovar and by studying the mechanisms of niche adaptation by natural transformation of *P. stutzeri* strains.

The genome sequence of *P. stutzeri* strain JM300 was reconstructed *de novo* with Celera Assembler 7.0 (8) from a total of 532,488 reads obtained from 8-kb mate pairs with a 454 Life Sciences genome sequencer FLX Titanium platform. Additional sequencing data were obtained from runs with an Illumina HiSeq 2000 platform and a 500-bp Illumina paired-end library. The finished sequence has a genome size of 4.2 Mb and a G+C mol% of 63.2%.

The genome was annotated using the NCBI Prokaryotic Genomes Automatic Annotation Pipeline (PGAAP). A total of 3,815 coding sequences were identified. The frequency of individual reads is consistent with the chromosomal size, and the presence of 4 copies for 5S, 16S, and 23S rRNA genes was as described for the species strains (5). Genes for complete tricarboxylic acid, glycolysis, and pentose phosphate pathways are present. Furthermore, genes coding for discriminating metabolic and physiological properties of the species were detected: the complete set of genes for the denitrification pathway, for starch metabolism, and for flagellar synthesis. Genes coding for products necessary for natural transformation ability were found (type IV pili, *pilAI, pilC*,

*pilT*, and *pilU*, *comA*, and *exbB*). No nitrogen fixation genes (*nif*) or extrachromosomal elements were found. Predicted phage-related sequences, transposons, and insertion elements were detected.

Comparative genome analysis confirmed that strain JM300 exhibited overall similarity to the previously sequenced 4 strains of genomovars 1 and 2 (genome size, G+C content). Genomovars were discriminated by average nucleotide identities based on BLAST (ANIb) values (11): higher than 96% between strains of the same genomovar, between 79 to 85% for different genomovars, and lower than 77% with other species of *Pseudomonas*.

**Nucleotide sequence accession number.** This whole-genome shotgun project has been deposited at DDBJ/EMBL/GenBank under the accession number CP003725. The version described in this paper is the first version.

#### ACKNOWLEDGMENTS

This work was supported in part by grants CGL 2009-12180 and CSD 2009-00006 (Consolider Program) from the CICYT (Spain) and by FEDER funding. M.G. is the recipient of a contract from the "Juan de la Cierva" Program of the Spanish Ministry for Economy and Competitivity.

### REFERENCES

- Carlson CA, Pierson LS, Rosen JJ, Ingraham JL. 1983. Pseudomonas stutzeri and related species undergo natural transformation. J. Bacteriol. 153:93–99.
- 2. Chen I, Dubnau D. 2004. DNA uptake during bacterial transformation. Nat. Rev. Microbiol. 2:241–249.
- 3. Chen M, et al. 2011. Complete genome sequence of the type strain *Pseudomonas stutzeri* CGMCC 1.1803. J. Bacteriol. **193**:6095.
- 4. Cladera AM, Bennasar A, Barceló M, Lalucat J, García-Valdés E. 2004. Comparative genetic diversity of *Pseudomonas stutzeri* genomovars, clonal structure, and phylogeny of the species. J. Bacteriol. **186**: 5239–5248.
- Lalucat J, Bennasar A, Bosch R, García-Valdés E, Palleroni NJ. 2006. Biology of *Pseudomonas stutzeri*. Microbiol. Mol. Biol. Rev. 70:510–547.
- 6. Lorenz MG, Sikorski J. 2000. The potential for intraspecific horizontal

Received 16 July 2012 Accepted 27 July 2012

Address correspondence to Antoni Bennasar, toni.bennasar@uib.es.

Copyright © 2012, American Society for Microbiology. All Rights Reserved. doi:10.1128/JB.01257-12

gene exchange by natural genetic transformation: sexual isolation among genomovars of *Pseudomonas stutzeri*. Microbiology **146**:3081–3090.

- 7. Lorenz MG, Wackernagel W. 1994. Bacterial gene transfer by natural genetic transformation in the environment. Microbiol. Rev. 58:563–602.
- 8. Miller JR, et al. 2008. Aggressive assembly of pyrosequencing reads with mates. Bioinformatics 24:2818–2824.
- Mulet M, et al. 2008. Phylogenetic analysis and siderotyping as useful tools in the taxonomy of *Pseudomonas stutzeri*: description of a novel genomovar. Int. J. Syst. Evol. Microbiol. 58:2309–2315.
- Peña A, et al. 2012. Draft genome of *Pseudomonas stutzeri* strain ZoBell (CCUG 16156), a marine isolate and model organism for denitrification studies. J. Bacteriol. 194:1277–1278.
- Richter M, Rosselló-Móra R. 2009. Shifting the genomic gold standard for the prokaryotic species definition. Proc. Natl. Acad. Sci. U. S. A. 106: 19126–19131.
- Sikorski J, Teschner N, Wackernagel W. 2002. Highly different levels of natural transformation are associated with genomic subgroups within a local population of *Pseudomonas stutzeri* from soil. Appl. Environ. Microbiol. 68:865–873.
- Yan Y, et al. 2008. Nitrogen fixation island and rhizosphere competence traits in the genome of root-associated *Pseudomonas stutzeri* A1501. Proc. Natl. Acad. Sci. U. S. A. 105:7564–7569.
- Yu H, et al. 2011. Complete genome sequence of the nitrogen-fixing and rhizosphere-associated bacterium *Pseudomonas stutzeri* strain DSM 4166. J. Bacteriol. 193:3422–3423.