A genome

Journals.ASM.org



Complete Genome Sequence of the Cyanogenic Phosphate-Solubilizing *Pseudomonas* sp. Strain CCOS 191, a Close Relative of *Pseudomonas* mosselii

Theo H. M. Smits,^a Joël F. Pothier,^a Michela Ruinelli,^a Jochen Blom,^b David Frasson,^c Chantal Koechli,^{d*} Carlotta Fabbri,^d Helmut Brandl,^d Brion Duffy,^a Martin Sievers^c

Environmental Genomics and Systems Biology Research Group, Institute of Natural Resource Sciences, Zürich University of Applied Sciences, Wädenswil, Switzerland^a; Bioinformatics and Systems Biology, Justus-Liebig-Universität, Giessen, Germany^b; Microbiology and Molecular Biology Research Group, Institute of Biotechnology, Zürich University of Applied Sciences, Wädenswil, Switzerland^c; Institute of Evolutionary Biology and Environmental Studies, University of Zürich, Zürich, Switzerland^d

* Present address: Chantal Koechli, Department of Microbiology, Cornell University, Ithaca, New York, USA.

We sequenced the complete genome of the isolate *Pseudomonas* sp. CCOS 191. This strain is able to dissolve phosphate minerals and form cyanide. The genome sequence is used to establish the phylogenetic relationship of this species.

Received 8 May 2015 Accepted 13 May 2015 Published 11 June 2015

Citation Smits THM, Pothier JF, Ruinelli M, Blom J, Frasson D, Koechli C, Fabbri C, Brandl H, Duffy B, Sievers M. 2015. Complete genome sequence of the cyanogenic phosphate-solubilizing *Pseudomonas* sp. strain CCOS 191, a close relative of *Pseudomonas mosselii*. Genome Announc 3(3):e00616-15. doi:10.1128/genomeA.00616-15. Copyright © 2015 Smits et al. This is an open-access article distributed under the terms of the Creative Commons Attribution 3.0 Unported license. Address correspondence to Theo H. M. Smits, theo.smits@zhaw.ch.

e recently isolated *Pseudomonas* sp. strain CCOS 191 from a water sample in Zürich, Switzerland, which is able to form large amounts of cyanide and dissolve phosphate minerals. Cyanide-forming microbes, many of which are members of the Pseudomonas family (1), are highly present in the environment, at times making up 50% of the soil community (2). Whereas most pseudomonads form cyanide, hypothesized to be a defense mechanism and/or a result of secondary metabolism (3), only in the presence of glycine as direct precursor, Pseudomonas sp. CCOS 191 is able to generate cyanide from glucose. Phosphorus is an essential macronutrient for the growth and reproduction of plants and microorganisms and is assimilated mostly from soluble phosphate. In cases in which phosphate-containing rocks (e.g., apatite) are the source of phosphorus in terrestrial environments, minerals have to be converted into a biologically available form (4). Pseudomonas sp. CCOS 191 is efficient in the solubilization of phosphate minerals.

Total genomic DNA was isolated according to the method of Pitcher et al. (5). The genome was sequenced using PacBio singlemolecule real-time (SMRT) reads (FGCZ, Zurich, Switzerland), as well as >80,000,000 50-bp Illumina paired-end reads (GATC, Konstanz, Germany). Five SMRT cells yielded 371,914 reads, with an average length of 5,416 bp (for a total of 2,014,434,330 bp). These reads were assembled into one contig of 6,030,031 bp using the HGAP approach (6), which, after manual inspection, was found to be circular. To check the assembly and correct for eventual incorrectly assigned bases, a subset of 8,000,000 Illumina reads were mapped against the HGAP unique contig using the Lasergene genomics package (DNAStar, Madison, WI). The final assembly gave a 6,012,947-bp circular chromosome. The sequence was annotated automatically using GenDB (7) with manual optimization, which yielded a total of 5,302 genes.

Based on the phylogeny of the 16S rRNA, *gyrB*, *rpoB*, and *rpoD* housekeeping genes (8), we generated a phylogeny with known

members of the *Pseudomonas putida* group, revealing that this strain might represent a novel species. Comparative genomics using EDGAR (9) indicated that the mean average amino acid identity (95.92%) is close to that of *Pseudomonas mosselii* DSM 17497^T, at the border of the species delineation (10). Based on multilocus sequence analysis (MLSA) data, the closest relative might be *Pseudomonas soli* (11), but no genome sequence of this species is available.

The production of cyanide from glucose may be a phenotypic feature that could be identified using the genome sequence of *Pseudomonas* sp. strain CCOS 191 and exploited for biotechnological applications in the future.

Nucleotide sequence accession number. The nucleotide sequence of the genome of *Pseudomonas* sp. CCOS 191 was deposited at EMBL-EBI under accession no. LN847264.

ACKNOWLEDGMENTS

The research was carried out with the help of initial funding of the Technology Transfer Office of the ZHAW School of Life Sciences and Facility Management. C.F. was supported by a university grant (Forschungskredit der Universität Zürich).

REFERENCES

- Knowles CJ, Bunch AW. 1986. Microbial cyanide metabolism. Adv Microb Physiol 27:73–111. http://dx.doi.org/10.1016/S0065-2911(08)60304 -5.
- Faramarzi MA, Stagars M, Pensini E, Krebs W, Brandl H. 2004. Metal solubilization from metal-containing solid materials by cyanogenic *Chromobacterium violaceum*. J Biotechnol 113:321–326. http://dx.doi.org/ 10.1016/j.jbiotec.2004.03.031.
- Blumer C, Haas D. 2000. Mechanism, regulation, and ecological role of bacterial cyanide biosynthesis. Arch Microbiol 173:170–177. http:// dx.doi.org/10.1007/s002039900127.
- Browne P, Rice O, Miller SH, Burke J, Dowling DN, Morrissey JP, O'Gara F. 2009. Superior inorganic phosphate solubilization is linked to phylogeny within the *Pseudomonas fluorescens* complex. Appl Soil Ecol 43:131–138. http://dx.doi.org/10.1016/j.apsoil.2009.06.010.

- Pitcher DG, Saunders NA, Owen RJ. 1989. Rapid extraction of bacterial genomic DNA with guanidium thiocyanate. Lett Appl Microbiol 8:151–156. http://dx.doi.org/10.1111/j.1472-765X.1989.tb00262.x.
- Chin C-S, Alexander DH, Marks P, Klammer AA, Drake J, Heiner C, Clum A, Copeland A, Huddleston J, Eichler EE, Turner SW, Korlach J. 2013. Nonhybrid, finished microbial genome assemblies from long-read SMRT sequencing data. Nat Methods 10:563–569. http://dx.doi.org/ 10.1038/nmeth.2474.
- Meyer F, Goesmann A, McHardy AC, Bartels D, Bekel T, Clausen J, Kalinowski J, Linke B, Rupp O, Giegerich R, Pühler A. 2003. GenDB—an open source genome annotation system for prokaryote genomes. Nucleic Acids Res 31:2187–2195. http://dx.doi.org/10.1093/nar/ gkg312.
- Mulet M, Gomila M, Scotta C, Sánchez D, Lalucat J, García-Valdés E. 2012. Concordance between whole-cell matrix-assisted laser-desorption/

ionization time-of-flight mass spectrometry and multilocus sequence analysis approaches in species discrimination within the genus *Pseudomonas*. Syst Appl Microbiol **35**:455–464. http://dx.doi.org/10.1016/ j.sy-apm.2012.08.007.

- Blom J, Albaum SP, Doppmeier D, Pühler A, Vorhölter F-J, Zakrzewski M, Goesmann A. 2009. EDGAR: a software framework for the comparative analysis of prokaryotic genomes. BMC Bioinformatics 10:154. http:// dx.doi.org/10.1186/1471-2105-10-154.
- Konstantinidis KT, Tiedje JM. 2005. Towards a genome-based taxonomy for prokaryotes. J Bacteriol 187:6258–6264. http://dx.doi.org/10.1128/ JB.187.18.6258-6264.2005.
- Pascual J, García-López M, Carmona C, Sousa Tda S, de Pedro N, Cautain B, Martín J, Vicente F, Reyes F, Bills GF, Genilloud O. 2014. *Pseudomonas soli* sp. nov., a novel producer of xantholysin congeners. Syst Appl Microbiol 37:412–416. http://dx.doi.org/10.1016/j.syapm.2014.07.003.