

Draft Genome Sequence of *Rhodococcus rhodochrous* Strain ATCC 21198

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Rhodococcus rhodochrous is a Gram-positive red-pigmented bacterium commonly found in the soil. The draft genome sequence for *R. rhodochrous* strain ATCC 21198 is presented here to provide genetic data for a better understanding of its lipid-accumulating capabilities.

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Rhodococcus rhodochrous is a red pigment-forming Grampositive bacterium belonging to the Actinobacteria phylum and the Nocardiaceae family. This metabolically diverse bacterium has been most notably used for the industrial production of acrylamide from acrylonitrile and in the bioremediation of hydrocarbons, polychlorinated biphenyls, and other aromatic compounds (1–4). Other members of the Actinobacteria, such as Streptomyces, Nocardia, Rhodococcus, and Gordonia, have been shown to synthesize and accumulate triacylglycerols (TAGs) (5). The potential for *R. rhodochrous* to utilize a variety of carbon sources for the accumulation of TAGs is of interest due to the increasing global need for alternative biofuels. Despite the widespread use of *R. rhodochrous* in degradation research, the understanding of its ability to lipid accumulation is limited.

A draft genome sequence was generated for strain ATCC 21198 to develop a better understanding of the lipid-accumulating capabilities of *R. rhodochrous*. The isolate *R. rhodochrous* ATCC 21198 was purchased from American Type Culture Collection (ATCC). Genomic DNA was isolated from frozen cell pellets using the Mo-Bio PowerSoil DNA isolation kit (Mo-Bio Laboratories, Inc., Carlsbad, CA), modified by the addition of lysozyme.

Sequence data were generated using an Illumina MiSeq instrument (6) according to the manufacturer's instructions, using a paired-end approach with an approximate insert library size of 400 bp and read lengths of 250 bp. The CLC Genomics Workbench (version 6.5) was used to trim and filter reads for quality sequence data and subsequent assembly. The draft genome sequence for *R. rhodochrous* ATCC 21198 is represented by 161 DNA contigs, with an estimated genome size of ~6.4 Mb and a G+C DNA content of 70.2%. The average sequence depth coverage across the genome was ~214 times the genome size, which was annotated as described previously (7) for 6,039 predicted proteincoding gene models.

In response to an environment with little nitrogen and excess carbon, oleaginous microbes will store carbon as lipids (5, 8).

Rhodococcus species have been shown to produce large amounts of lipids in the form of TAGs due to the high activity of the enzyme acyl-coenzyme A (CoA):diacylglycerol acyltransferase (DGAT) (9). In this draft sequence of R. rhodochrous, there are several predicted wax ester synthase/diacylglycerol acyltransferase genes showing sequence identity to other Rhodococcus species. Other putative fatty acid and TAG synthesis genes were also discovered in the R. rhodochrous genome, such as acetyl-CoA carboxylase, acyl carrier proteins, (1-acyl-G3P) acyltransferase, glycerol kinase, and glycerol-3-phosphate dehydrogenase (10). Several cytochrome P450-like enzymes were predicted in the genome of R. rhodochrous, as well as the associated genes flavodoxin and feredoxin. Other predicted genes related to degradation mechanisms include oxygenases, dioxygenases, and a putative aromatic degradation protein. The R. rhodochrous draft genome offers insight into the potential metabolic capabilities of this organism and will facilitate further studies.

Nucleotide sequence accession numbers. This draft genome sequence has been deposited in GenBank under the accession no. AZHI00000000. The version described in this paper is version AZHI01000000.

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For strain and DNA requests, contact W.T.F.

REFERENCES

 Yamada H, Kobayashi M. 1996. Nitrile hydratase and its application to industrial production of Acrylamide. Biosci. Biotechnol. Biochem. 60: 1391–1400. http://dx.doi.org/10.1271/bbb.60.1391.

- Kobayashi M, Shimizu S. 1998. Metalloenzyme nitrile hydratase: structure, regulation, and application to biotechnology. Nat. Biotechnol. 16: 733–736. http://dx.doi.org/10.1038/nbt0898-733.
- Martínková L, Uhnáková B, Pátek M, Nesvera J, Kren V. 2009. Biodegradation potential of the genus *Rhodococcus*. Environ. Int. 35:162–177. http://dx.doi.org/10.1016/j.envint.2008.07.018.
- 4. Larkin MJ, Kulakov LA, Allen CC. 2005. Biodegradation and *Rhodococ-cus*—masters of catabolic versatility. Curr. Opin. Biotechnol. 16:282–290. http://dx.doi.org/10.1016/j.copbio.2005.04.007.
- Alvarez HM, Steinbüchel A. 2002. Triacylglycerols in prokaryotic microorganisms. Appl. Microbiol. Biotechnol. 60:367–376. http://dx.doi.org/1 0.1007/s00253-002-1135-0.
- 6. Quail MA, Smith M, Coupland P, Otto TD, Harris SR, Connor TR, Bertoni A, Swerdlow HP, Gu Y. 2012. A tale of three next generation sequencing platforms: comparison of Ion Torrent, Pacific Biosciences, and Illumina MiSeq sequencers. BMC Genomics 13:341. http://dx.doi.org /10.1186/1471-2164-13-341.
- Brown SD, Klingeman DM, Lu TY, Johnson CM, Utturkar SM, Land ML, Schadt CW, Doktycz MJ, Pelletier DA. 2012. Draft genome sequence of *Rhizobium* sp. strain PDO1-076, a bacterium isolated from *Populus deltoides*. J. Bacteriol. 194:2383–2384. http://dx.doi.org/10.1128/ JB.00198-12.
- Ratledge C, Wynn JP. 2002. The biochemistry and molecular biology of lipid accumulation in oleaginous microorganisms, Adv. Appl. Microbiol. 51:1–51. http://dx.doi.org/10.1016/S0065-2164(02)51000-5.
- Alvarez AF, Alvarez HM, Kalscheuer R, Wältermann M, Steinbüchel A. 2008. Cloning and characterization of a gene involved in triacylglycerol biosynthesis and identification of additional homologous genes in the oleaginous bacterium *Rhodococcus opacus* PD630. Microbiology 154: 2327–2335. http://dx.doi.org/10.1099/mic.0.2008/016568-0.
- Kosa M, Ragauskas AJ. 2011. Lipids from heterotrophic microbes: advances in metabolism research. Trends Biotechnol. 29:53–61. http://dx .doi.org/10.1016/j.tibtech.2010.11.002.