



# Draft Genome Sequence of Strain R\_RK\_3, an Iron-Depositing Isolate of the Genus *Rhodomicrobium*, Isolated from a Dewatering Well of an Opencast Mine

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**ABSTRACT** *Rhodomicrobium* sp. strain R\_RK\_3 is an iron-depositing bacterium from which we report the draft genome. This strain was isolated from ochrous depositions of a mining well pump in Germany. The Illumina NextSeq technique was used to sequence the genome of the strain.

A 16S rRNA gene sequence comparison of strain R\_RK\_3 revealed a 94% similarity to *Rhodomicrobium vannielii* ATCC 17100 (GenBank accession no. CP002292). The search was conducted using BLASTn (1) and EZBioCloud (2). However, determination of the most abundant taxon for the genome bin based on weighted scaffold length revealed 27.0% similarity to *Bradyrhizobium icense*.

The family *Hyphomicrobiaceae* contains the three phototrophic genera *Rhodoplanes* (3), *Blastochloris* (4), and *Rhodomicrobium* (5). Within the genus *Rhodomicrobium*, *R. vannielii* (5) and *R. udaipurensis* (6) are currently the only recognized species. The draft genomes have been published for both of these species (7, 8). For *Rhodomicrobium vannielii*, the oxidation of ferrous iron has been described as a side reaction (9). The current work presents the third draft genome of a *Rhodomicrobium* strain, isolated from a novel habitat, i.e., the well pump of a dewatering well that also showed iron-depositing activity.

Strain R\_RK\_3 was isolated from ochrous deposits from a dewatering well pump at the Hambach opencast mine, in the Rhenish lignite-mining area. Sequential dilutions of sampled ochrous material were spread on modified Leptothrix medium (10) (deionized water was replaced by sterile well water), and reddish-brown colonies were selected after incubation for 17 to 21 days at room temperature. The iron- and manganese-depositing activity of the isolate was verified by dissolving the colonies with oxalic acid (10%) as previously described by Schmidt et al. (11). Strain R\_RK\_3 is a small Gram-negative bacterium with a coccus shape of 2- to 4- $\mu\text{m}$  length and a width of 1.0  $\mu\text{m}$ .

Extraction of genomic DNA was done as previously described (12). The paired-end library was prepared by following the Illumina Nextera XT DNA library prep kit protocol. Genome sequencing was done on an Illumina NextSeq 500 sequencer using the NextSeq mid output kit v2 (300 cycles) by generating 74,681,576 raw reads. Demultiplexing was done with bcl2fastq v2.18.0.12, and quality filtering of raw reads was performed using Trimmomatic v0.36 (13), resulting in 48,202,264 filtered reads. Reads were then checked for ambiguous base calls and low complexity, employing the DUST algorithm (14), and filtered accordingly with an R script in Microsoft R Open v3.3.2 (15), followed by preassembly with SPAdes v3.10.0 (16) using default k-mer lengths up to 99 bp. Scaffolds  $\geq 500$  bp of this preassembly were subject to extension and second-round scaffolding with SSPACE standard v3.0 (17).

Received 11 July 2017 Accepted 12 July 2017 Published 24 August 2017

**Citation** Braun B, Künzel S, Schröder J, Szewzyk U. 2017. Draft genome sequence of strain R\_RK\_3, an iron-depositing isolate of the genus *Rhodomicrobium*, isolated from a dewatering well of an opencast mine. *Genome Announc* 5:e00864-17. <https://doi.org/10.1128/genomeA.00864-17>.

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Scaffolds  $\geq 2,500$  bp were assigned to genome bins by MetaBAT v0.32.4 (18), and functional annotation of draft genomes was performed with Prokka v1.12b (19). Tentative contig layout authentication (CLA) was conducted with Contig-Layout-Authenticator v1.0 (20) using the reference genome of *Rhodomicrobium vannielii* ATCC 17100.

The draft genome included 39 contigs with an  $N_{50}$  assembly quality of 226,044 and  $L_{50}$  of 10. The shortest scaffold was 2,822 bp, and the longest scaffold was 401,427 bp. The total size of the scaffolds was 5,786,497 bp with a G+C content of 65%. Annotation resulted in 39 contigs including 5,135 coding regions for 5,205 genes, 587 signal peptides, no clustered regularly interspaced short palindromic repeat (CRISPR) unit, 4 rRNAs (16S, 23S), 48 tRNAs, 1 transfer-messenger RNA (tmRNA), and 17 miscellaneous RNAs.

**Accession number(s).** This whole-genome shotgun project has been deposited at DDBJ/ENA/GenBank under the accession no. [MWLK00000000](https://doi.org/10.1093/nar/25.17.3389). The version described in this paper is version MWLK01000000.

## ACKNOWLEDGMENTS

Bioinformatic consulting was provided by omics2view.consulting GbR, Kiel, Germany.

This work was supported by a grant of the “Bundesministerium für Bildung und Forschung (BMBF),” 02WT1184.

## REFERENCES

- Altschul SF, Madden TL, Schäffer AA, Zhang J, Zhang Z, Miller W, Lipman DJ. 1997. Gapped BLAST and PSI-BLAST: a new generation of protein database search programs. *Nucleic Acids Res* 25:3389–3402. <https://doi.org/10.1093/nar/25.17.3389>.
- Yoon SH, Ha SM, Kwon S, Lim J, Kim Y, Seo H, Chun J. 2017. Introducing EzBioCloud: a taxonomically united database of 16S rRNA gene sequences and whole-genome assemblies. *Int J Syst Evol Microbiol* 67: 1613–1617. <https://doi.org/10.1099/ijsem.0.001755>.
- Hiraishi A, Ueda Y. 1994. Transfer of the bacteriochlorophyll b-containing phototrophic bacteria *Rhodospseudomonas viridis* and *Rhodospseudomonas sulfoviridis* to the genus *Blastochloris* gen. nov. *Int J Syst Bacteriol* 44:665–673. <https://doi.org/10.1099/00207713-44-4-665>.
- Hiraishi A. 1997. Transfer of the bacteriochlorophyll & containing phototrophic bacteria *Rhodospseudomonas viridis* and *Rhodospseudomonas sulfoviridis* to the genus *Blastochloris* gen. nov. *Int J Syst Bacteriol* 47:217–219. <https://doi.org/10.1099/00207713-47-1-217>.
- Duchow E, Douglas HC. 1949. *Rhodomicrobium vannielii*, a new photoheterotrophic bacterium. *J Bacteriol* 58:409–416.
- Ramana VV, Raj PS, Tushar L, Sasikala Ch, Ramana ChV. 2013. *Rhodomicrobium udaipurense* sp. nov., a psychrotolerant, phototrophic alphaproteobacterium isolated from a freshwater stream. *Int J Syst Evol Microbiol* 63:2684–2689. <https://doi.org/10.1099/ijvs.0.046409-0>.
- Brown PJB, Kysela DT, Buechlein A, Hemmerich C, Brun YV. 2011. Genome sequences of eight morphologically diverse alphaproteobacteria. *J Bacteriol* 193:4567–4568. <https://doi.org/10.1128/JB.05453-11>.
- Tushar L, Sasikala Ch, Ramana ChV. 2014. Draft genome sequence of *Rhodomicrobium udaipurense* JA643T with special reference to hopanoid biosynthesis. *DNA Res* 21:639–647. <https://doi.org/10.1093/dnares/dsu026>.
- Heising S, Schink B. 1998. Phototrophic oxidation of ferrous iron by a *Rhodomicrobium vannielii* strain. *Microbiology* 144:2263–2269. <https://doi.org/10.1099/00221287-144-8-2263>.
- Atlas R. 2010. *Handbook of microbiological media*. 4th ed. CRC Press, Washington, DC.
- Schmidt B, Sánchez LA, Fretschner T, Kreps G, Ferrero MA, Siñeriz F, Szewzyk U. 2014. Isolation of *Sphaerotilus-Leptothrix* strains from iron bacteria communities in Tierra del Fuego wetlands. *FEMS Microbiol Ecol* 90:454–466. <https://doi.org/10.1111/1574-6941.12406>.
- Braun B, Szewzyk U. 2016. Complete genome sequence of “*Candidatus Viadribacter manganicus*” isolated from a German floodplain area. *Genome Announc* 4(5):3402. <https://doi.org/10.1128/genomeA.00897-16>.
- Bolger AM, Lohse M, Usadel B. 2014. Trimmomatic: a flexible trimmer for Illumina sequence data. *Bioinformatics* 30:2114–2120. <https://doi.org/10.1093/bioinformatics/btu170>.
- Morgulis A, Gertz EM, Schäffer AA, Agarwala R. 2006. A fast and symmetric DUST implementation to mask low-complexity DNA sequences. *J Comput Biol* 13:1028–1040. <https://doi.org/10.1089/cmb.2006.13.1028>.
- R Core Team. 2014. R: a language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. <http://www.r-project.org/>.
- Bankevich A, Nurk S, Antipov D, Gurevich AA, Dvorkin M, Kulikov AS, Lesin VM, Nikolenko SI, Pham SON, Prjibelski AD, Pyshkin AV, Sirotkin AV, Vyahhi N, Tesler G, Alekseyev MAXA, Pevzner PA. 2012. SPAdes: a new genome assembly algorithm and its applications to single-cell sequencing. *J Comput Biol* 19:455–477. <https://doi.org/10.1089/cmb.2012.0021>.
- Boetzer M, Henkel CV, Jansen HJ, Butler D, Pirovano W. 2011. Scaffolding pre-assembled contigs using SSPACE. *Bioinformatics* 27:578–579. <https://doi.org/10.1093/bioinformatics/btq683>.
- Kang DD, Froula J, Egan R. 2014. A robust statistical framework for reconstructing genomes from metagenomic data. *bioRxiv* <https://doi.org/10.1101/011460>.
- Seemann T. 2014. Prokka: rapid prokaryotic genome annotation. *Bioinformatics* 30:2068–2069. <https://doi.org/10.1093/bioinformatics/btu153>.
- Shaik S, Kumar N, Lankapalli AK, Tiwari SK, Baddam R, Ahmed N. 2016. Contig-Layout-Authenticator (CLA): a combinatorial approach to ordering and scaffolding of bacterial contigs for comparative genomics and molecular epidemiology. *PLoS One* 11:e0155459. <https://doi.org/10.1371/journal.pone.0155459>.