AMERICAN SOCIETY FOR MICROBIOLOGY | Microbiology Resource Announcements

GENOME SEQUENCES

Genome Sequence of the Radiation-Resistant Bacterium *Deinococcus radiophilus* ATCC 27603^T

Claudia R. Maynard,[a] Kyle S. MacLea[a,b,c]

[a]Biotechnology Program, University of New Hampshire, Manchester, New Hampshire, USA
[b]Biology Program, University of New Hampshire, Manchester, New Hampshire, USA
[c]Department of Life Sciences, University of New Hampshire, Manchester, New Hampshire, USA

ABSTRACT The pigmented bacterium *Deinococcus radiophilus*, which is highly resistant to radiation exposure, was first isolated from irradiated lizardfish. We report a genome assembly of *D. radiophilus* UWO 1055^T (=ATCC 27603^T), with a predicted genome size of 2.7 Mbp (62.66% G+C content). A number of CRISPR-associated proteins and two CRISPR arrays were identified.

Compared to highly sequenced bacterial phyla, such as the *Proteobacteria*, to date, only about 53 genome assemblies from the phylum *Deinococcus-Thermus* have been made publicly available. Within the phylum, organisms of the radiation-resistant genus *Deinococcus* were separated from the actinobacterial genus *Micrococcus* (which has superficial similarities to the deinococci on the basis of pigmentation and Gram-positive staining) when the new genus was created (1). These unusual Gram-positive organisms (that also possess an outer membrane) include the most studied radiation-resistant organism, *Deinococcus radiodurans* (1), for which original (2) and improved (3) genome assemblies are available. Few other deinococci have available genome sequences, and there is some controversy on the relatedness of these organisms that was recognized early on by Brooks and Murray (1) and for which they suggested a possible need to split the genus at a later date. Additional genomes in this genus will help settle this and other issues.

First found in an irradiated lizardfish (the species known as Bombay duck or bummalo [*Harpadon nehereus*]), *Deinococcus radiophilus* is an obligate aerobic deinococcus, stains Gram positive, and is pigmented and non-spore forming. *D. radiophilus* (4–7) differs from *D. radiodurans* and other members of the genus by a lack of nitrate reduction and no production of acid from glucose in standard media (1). *D. radiophilus* is also of note because of its remarkable DNA repair abilities and its production of useful restriction endonucleases (8, 9). There is also some interest in the use of deinococci, including *D. radiophilus,* in the bioremediation of accidents involving radioactive materials and in novel biotechnology applications (10, 11).

Freeze-dried *D. radiophilus* ATCC 27603^T was rehydrated and streaked for single colonies on Trypticase soy agar at 30°C for 48 h at 1 atm. A Trypticase soy broth culture was grown from a single colony and used to isolate genomic DNA (gDNA) by means of the QIAamp DNA minikit (Qiagen, Valencia, CA, USA). The Kapa HyperPlus kit (KR1145, v.3.16; Wilmington, MA, USA) was used to fragment gDNA and tag it with sequence adapters for sequencing on an Illumina HiSeq 2500 instrument (Hubbard Center for Genome Studies, Durham, NH, USA). A total of 4,543,128 raw 250-bp reads were trimmed using Trimmomatic (settings, paired-end mode with a window size of 4, quality requirement of 15, and minimum read length of 36) before being assembled with the default parameters for bacterial assembly using SPAdes v.3.11.1 (12, 13). Once small contigs (<500 bp) and contaminants (flagged by the NCBI using the PGAP,

Citation Maynard CR, MacLea KS. 2019. Genome sequence of the radiation-resistant bacterium *Deinococcus radiophilus* ATCC 27603^T. Microbiol Resour Announc 8:e00627-19. https://doi.org/10.1128/MRA.00627-19.

Editor Catherine Putonti, Loyola University Chicago

Address correspondence to Kyle S. MacLea, Kyle.MacLea@UNH.edu.

K.S.M. dedicates this work to Robert Gordon MacLea (1917–2000), U.S. Army veteran and carpenter.

Received 28 May 2019
Accepted 2 July 2019
Published 25 July 2019

described below) were removed, QUAST (14) identified 100 contigs, with the largest one being 325,917 bp, with an N_{50} value of 77,319 bp and an estimated genome coverage of 754×. Annotation was conducted using the NCBI Prokaryotic Genome Annotation Pipeline (PGAP) (15). PGAP revealed a total sequence length of 2,753,082 bp, a total of 2,789 genes, 2,731 protein-coding sequences, 78 pseudogenes, 50 tRNAs, 5 rRNAs, 3 noncoding RNAs (ncRNAs), 2 CRISPR arrays, and a G+C content of 62.66%, in line with the published value (determined by the melting temperature [T_m]) of 62% (1).

Data availability. The *Deinococcus radiophilus* ATCC 27603^T whole-genome shotgun sequence (WGS) project has been deposited at DDBJ/ENA/GenBank under the accession number RXPE00000000. The version described in this paper is version RXPE01000000. The raw Illumina data from BioProject number PRJNA509618 were submitted to the NCBI Sequence Read Archive (SRA) under experiment accession number SRX5889021.

ACKNOWLEDGMENTS

Sequencing and bioinformatics analysis were undertaken at the Hubbard Center for Genome Studies at UNH, supported by NH-INBRE, with the kind assistance of Kelley Thomas and Stephen Simpson.

This work was a project of the Microbiology Education through Genome Annotation-New Hampshire (MEGA-NH) program.

This work was supported by the Department of Life Sciences at the University of New Hampshire, by a Manchester Undergraduate Project Support grant to C.R.M., and by New Hampshire-INBRE through Institutional Development Award (IDeA) P20GM103506 from the National Institute of General Medical Sciences of the NIH.

The funders had no role in the study design, data collection and interpretation, or decision to submit the work for publication.

REFERENCES

1. Brooks BW, Murray R. 1981. Nomenclature for Micrococcus radiodurans and other radiation-resistant cocci: Deinococcaceae fam. nov. and Deinococcus gen. nov., including five species. Int J Syst Bacteriol 31:353–360. https://doi.org/10.1099/00207713-31-3-353.
2. White O, Eisen JA, Heidelberg JF, Hickey EK, Peterson JD, Dodson RJ, Haft DH, Gwinn ML, Nelson WC, Richardson DL, Moffat KS, Qin H, Jiang L, Pamphile W, Crosby M, Shen M, Vamathevan JJ, Lam P, McDonald L, Utterback T, Zalewski C, Makarova KS, Aravind L, Daly MJ, Minton KW, Fleischmann RD, Ketchum KA, Nelson KE, Salzberg S, Smith HO, Venter JC, Fraser CM. 1999. Genome sequence of the radioresistant bacterium Deinococcus radiodurans R1. Science 286:1571–1577. https://doi.org/10.1126/science.286.5444.1571.
3. Hua X, Hua Y. 2016. Improved complete genome sequence of the extremely radioresistant bacterium Deinococcus radiodurans R1 obtained using PacBio single-molecule sequencing. Genome Announc 4:e00886-16. https://doi.org/10.1128/genomeA.00886-16.
4. Lewis NF. 1971. Studies on a radio-resistant coccus isolated from Bombay duck (Harpodon nehereus). J Gen Microbiol 66:29–35. https://doi.org/10.1099/00221287-66-1-29.
5. Lewis NF, Kumta US. 1972. Evidence for extreme UV resistance of Micrococcus sp. NCTC 10785. Biochem Biophys Res Commun 47:1100–1105. https://doi.org/10.1016/0006-291x(72)90947-3.
6. Zimmerman JM, Battista JR. 2005. A ring-like nucleoid is not necessary for radioresistance in the Deinococcaceae. BMC Microbiol 5:17. https://doi.org/10.1186/1471-2180-5-17.
7. Lavy A, Neeman Y, Fuhrman B. 2005. The antioxidative effect of the bacteria Dienococcus radiophilus against LDL lipid peroxidation. Eur J Nutr 44:281–284. https://doi.org/10.1007/s00394-004-0522-y.
8. Purvis IJ, Moseley B. 1983. Isotation and characterisation of DraI, a type II restriction endonuclease recognisting a aequence containing only A:T basepairs, and inhibition of its activity by UV irradiation of substrate DNA. Nucleic Acids Res 11:5467–5474. https://doi.org/10.1093/nar/11.16.5467.
9. Grosskopf R, Wolf W, Kessler C. 1985. Two new restriction endonucleases DraII and DraIII from *Deinococcus radiophilus*. Nucleic Acids Res 13:1517–1528. https://doi.org/10.1093/nar/13.5.1517.
10. Daly MJ. 2000. Engineering radiation-resistant bacteria for environmental biotechnology. Curr Opin Biotechnol 11:280–285. https://doi.org/10.1016/S0958-1669(00)00096-3.
11. Gerber E, Bernard R, Castang S, Chabot N, Coze F, Dreux-Zigha A, Hauser E, Hivin P, Joseph P, Lazarelli C, Letellier G, Olive J, Leonetti J-P. 2015. *Deinococcus* as new chassis for industrial biotechnology: biology, physiology and tools. J Appl Microbiol 119:1–10. https://doi.org/10.1111/jam.12808.
12. 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.
13. Bankevich A, Nurk S, Antipov D, Gurevich AA, Dvorkin M, Kulikov AS, Lesin VM, Nikolenko SI, Pham S, Prjibelski AD, Pyshkin AV, Sirotkin AV, Vyahhi N, Tesler G, Alekseyev MA, 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.
14. Gurevich A, Saveliev V, Vyahhi N, Tesler G. 2013. QUAST: quality assessment tool for genome assemblies. Bioinformatics 29:1072–1075. https://doi.org/10.1093/bioinformatics/btt086.
15. Tatusova T, DiCuccio M, Badretdin A, Chetvernin V, Nawrocki EP, Zaslavsky L, Lomsadze A, Pruitt KD, Borodovsky M, Ostell J. 2016. NCBI Prokaryotic Genome Annotation Pipeline. Nucleic Acids Res 44:6614–6624. https://doi.org/10.1093/nar/gkw569.