



Draft Genome Sequence of *Pseudomonas* sp. Strain DrBHI1 (Phylum *Proteobacteria*)

Alexandria K. Wilson,^a Virginia G. Watral,^b Michael L. Kent,^b Thomas J. Sharpton,^{b,c} Christopher A. Gaulke^b

BioResource Research, Oregon State University, Corvallis, Oregon, USA^a; Department of Microbiology, Oregon State University, Corvallis, Oregon, USA^b; Department of Statistics, Oregon State University, Corvallis, Oregon, USA^c

ABSTRACT Here, we report the draft genome sequence of *Pseudomonas* sp. strain DrBHI1. The total assembly length is 5,649,751 bp in 146 contigs. This strain was isolated from zebrafish (*Danio rerio*) feces.

Pseudomonas is a large genus of Gram-negative, aerobic, rod-shaped bacteria known to inhabit a diversity of environmental and host-associated habitats (1, 2). Several species of this genus have been shown to be associated with the intestinal tracts of a variety of fish species (3), and some have been proposed as probiotic biocontrol agents in aquaculture (4).

Pseudomonas sp. strain DrBHI1 was isolated from the feces of zebrafish (*Danio rerio*). The isolation of this microbe was part of an undergraduate research project designed to increase the number of reference genomes from zebrafish-associated microbial communities represented in genome databases. Fecal pellets were collected from 9-liter tanks housing 12-month-old 5D line zebrafish using an aseptic technique. Fecal pellets were diluted, plated on brain heart infusion (BHI) agar, and incubated for 24 h at 27°C, and individual colonies were isolated. Isolates were inoculated in BHI broth and incubated overnight at 27°C. DNA was obtained using the UltraClean Microbial DNA isolation kit and the UltraClean PCR cleanup kit (Mo Bio). Universal 16S primers, 27F and 1492R (5), were used to PCR amplify the 16S rRNA gene, and an approximately 1,400-nucleotide product was gel purified using the UltraClean GelSpin DNA extraction kit (Mo Bio) and subsequently sequenced using an ABI 3730 DNA analyzer (Thermo Fisher Scientific). We then generated a list of full-length rRNA sequences closely related (>90% identity) to our isolate using BLAST (6) and the NCBI's 16S rRNA database. These sequences were used to construct a maximum likelihood phylogeny using FastTree (7), which demonstrated that our isolate likely belonged to the genus *Pseudomonas*.

We next constructed a DNA library using the Nextera XT kit (Illumina), which we sequenced on an Illumina MiSeq platform. A total of 1,922,050 250-bp paired-end reads were generated, quality trimmed, filtered using ea-utils (8), and assembled using Velvet (9). The total assembly size was 5,649,751 bp in 146 contigs with an N_{50} value of 105,138 bp, average contig coverage of 125×, and 64.25% GC content. Genome completeness was quantified with PhyloSift (10), which approximated that this genome was >99% complete based on the presence of 37 universal single-copy genes. The genome was then annotated using the integrated microbial genomes (IMG) system (11) and predicted to contain 5,229 coding and 129 noncoding genes. Eight biosynthetic clusters were also identified (five nonribosomal peptide synthetase clusters, two bacteriocins, and one other cluster). To infer taxonomy, we used PhyloSift to place concatenated marker gene alignments on a reference phylogeny. This analysis suggested that our isolate was closely related to *Pseudomonas entomophila* L48 (https://figshare.com/articles/bhi1_concat_tree_pdf/5362594). However, the average nucleotide identity (ANI) (11) between our isolate and *P. entomophila* was low (89%). Given the

Received 31 August 2017 Accepted 1 September 2017 Published 28 September 2017

Citation Wilson AK, Watral VG, Kent ML, Sharpton TJ, Gaulke CA. 2017. Draft genome sequence of *Pseudomonas* sp. strain DrBHI1 (phylum *Proteobacteria*). *Genome Announc* 5:e01090-17. <https://doi.org/10.1128/genomeA.01090-17>.

Copyright © 2017 Wilson et al. This is an open-access article distributed under the terms of the [Creative Commons Attribution 4.0 International license](https://creativecommons.org/licenses/by/4.0/).

Address correspondence to Christopher A. Gaulke, gaulkec@oregonstate.edu.

discordance between the marker gene sequence placement and ANI, we were unable to classify this isolate beyond its genus and propose that it be classified *Pseudomonas* sp. strain DrBH11.

Accession number(s). This whole-genome shotgun project has been deposited at DDBJ/ENA/GenBank under accession number [NIBE00000000](https://doi.org/10.1099/NIBE01000000). The version described in this paper is the first version, NIBE01000000.

ACKNOWLEDGMENTS

We thank the staff of the Center for Genome Research and Biocomputing at Oregon State University for assistance with Sanger and Illumina sequencing. We also acknowledge Duncan Millard for his assistance with Bash and Python scripting.

Funding for this project was provided by a professional development award from the Oregon State University Postdoctoral Association to C.A.G., an NSF DEB award (1557192) to T.J.S., and an NIH ORIP award (R24OD010998) to M.L.K.

REFERENCES

1. Anzai Y, Kim H, Park JY, Wakabayashi H, Oyaizu H. 2000. Phylogenetic affiliation of the pseudomonads based on 16S rRNA sequence. *Int J Syst Evol Microbiol* 50:1563–1589. <https://doi.org/10.1099/00207713-50-4-1563>.
2. Vodovar N, Vallenet D, Cruveiller S, Rouy Z, Barbe V, Acosta C, Cattolico L, Jubin C, Lajus A, Segurens B, Vacherie B, Wincker P, Weissenbach J, Lemaitre B, Médigue C, Boccard F. 2006. Complete genome sequence of the entomopathogenic and metabolically versatile soil bacterium *Pseudomonas entomophila*. *Nat Biotechnol* 24:673–679. <https://doi.org/10.1038/nbt1212>.
3. Llewellyn MS, Boutin S, Hoseinifar SH, Derome N. 2014. Teleost microbiomes: the state of the art in their characterization, manipulation and importance in aquaculture and fisheries. *Front Microbiol* 5:207. <https://doi.org/10.3389/fmicb.2014.00207>.
4. Verschuere L, Rombaut G, Sorgeloos P, Verstraete W. 2000. Probiotic bacteria as biological control agents in aquaculture. *Microbiol Mol Biol Rev* 64:655–671. <https://doi.org/10.1128/MMBR.64.4.655-671.2000>.
5. Jiang H, Dong H, Zhang G, Yu B, Chapman LR, Fields MW. 2006. Microbial diversity in water and sediment of Lake Chaka, an athalassohaline lake in northwestern China. *Appl Environ Microbiol* 72:3832–3845. <https://doi.org/10.1128/AEM.02869-05>.
6. Altschul SF, Gish W, Miller W, Myers EW, Lipman DJ. 1990. Basic local alignment search tool. *J Mol Biol* 215:403–410. [https://doi.org/10.1016/S0022-2836\(05\)80360-2](https://doi.org/10.1016/S0022-2836(05)80360-2).
7. Price MN, Dehal PS, Arkin AP. 2010. FastTree 2—approximately maximum-likelihood trees for large alignments. *PLoS One* 5:e9490. <https://doi.org/10.1371/journal.pone.0009490>.
8. Aronesty E. 2013. Comparison of sequencing utility programs. *Open Bioinform J* 7:1–8. <https://doi.org/10.2174/1875036201307010001>.
9. Zerbino DR, Birney E. 2008. Velvet: algorithms for de novo short read assembly using de Bruijn graphs. *Genome Res* 18:821–829. <https://doi.org/10.1101/gr.074492.107>.
10. Darling AE, Jospin G, Lowe E, Matsen FA, Bik HM, Eisen JA. 2014. PhyloSift: phylogenetic analysis of genomes and metagenomes. *PeerJ* 2:e243. <https://doi.org/10.7717/peerj.243>.
11. Huntemann M, Ivanova NN, Mavromatis K, Tripp HJ, Paez-Espino D, Palaniappan K, Szeto E, Pillay M, Chen IM, Pati A, Nielsen T, Markowitz VM, Kyrpides NC. 2016. Erratum to: the standard operating procedure of the DOE-JGI Microbial Genome Annotation Pipeline (MGAP v.4). *Stand Genomic Sci* 11:27. <https://doi.org/10.1186/s40793-016-0148-8>.