



Genome Sequence of *Providencia rettgeri* NVIT03, Isolated from *Nasonia vitripennis*

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ABSTRACT *Providencia rettgeri* is a common insect-associated Gram-negative bacterium. Here, we present the draft genome sequence of *P. rettgeri* NVIT03, the most common bacterial symbiont of the insect hymenopteran model *Nasonia vitripennis*. This symbiont is also part of the *Sarcophaga bullata* pupal microbiome that *Nasonia* spp. parasitize and that critically influences the development of the wasp.

The motile Gram-negative bacillus *Providencia rettgeri* was previously identified as a member of the gut microbiome of nematodes, insects, birds, and humans (1) while also being associated with opportunistic infections in humans and insects (2, 3). *P. rettgeri* is found throughout development in the dipteran genus *Sarcophaga*, a carrion-breeding necrophagous insect (4). These same insects serve as a host for the parasitic wasps of the genus *Nasonia*, which also maintain *P. rettgeri* in their gut microbial communities throughout development (5). *P. rettgeri* is the most dominant bacterium (67.00%) in the *Nasonia vitripennis* microbiome (5). This genome will help elucidate host-microbe interactions that are important to the affected insects' physiology, development, behavior, reproduction, and evolution (6, 7).

We present here a draft genome sequence of *P. rettgeri* NVIT03, a gut bacterium isolated from the *N. vitripennis* strain AsymCx. After surface sterilizing whole *Nasonia* animals with 10% bleach for 2 min and then homogenizing them in sterile phosphate-buffered saline (PBS), we plated 1× aliquots of the homogenate onto brain heart infusion agar medium and confirmed the taxonomic identity by Sanger sequencing of the 16S rRNA before sending the isolate from the AsymCx strain for whole-genome sequencing.

Bacterial genomic DNA was isolated from *P. rettgeri* NVIT03 cells cultured in 100 ml of nutrient broth medium (catalog no. 1.05443.0500; Sigma-Aldrich) at 30°C and 250 rpm for 18 h in a flask. The culture was centrifuged at 6,000 rpm for 5 min, and the resulting cell pellet was used for genome extraction using the DNeasy blood and tissue kit (Qiagen, Hilden, Germany). DNA was quantified using the double-stranded DNA (dsDNA) high-sensitivity (HS) assay kit on the Qubit 2.0 fluorometer (Life Technologies, Waltham, MA, USA).

Library preparation and genome sequencing were performed at MicrobesNG (Birmingham, United Kingdom). The DNA library was prepared using the Nextera XT library prep kit (Illumina) and sequenced on the Illumina HiSeq platform using a 250-bp paired-end protocol. Sequencing resulted in 191,241 reads which were trimmed using Trimmomatic 0.36 (8) and quality filtered using SAMtools (9), BEDTools (10), and bwa-mem (11). Seventy-three contigs were assembled with SPAdes (version 3.8) (12), with an N_{50} value of 787,417 bp, 4,400,855 nucleotides in total, and about 164-fold coverage. This strain is most closely related to the previously sequenced *P. rettgeri* strain AR_0082 (GenBank accession no. CP029736). The genome of *P. rettgeri* NVIT03 consists presumably of a single chromosome (4,380,957 bp) and exhibits a G+C content of 40.30%. No evidence of plasmids was found based on analysis with PlasmidFinder version 1.3 (13).

Citation Wang G-H, Brucker RM. 2019. Genome sequence of *Providencia rettgeri* NVIT03, isolated from *Nasonia vitripennis*. Microbiol Resour Announc 8:e01157-18. <https://doi.org/10.1128/MRA.01157-18>.

Editor Vincent Bruno, University of Maryland School of Medicine

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Received 12 September 2018

Accepted 11 December 2018

Published 17 January 2019

Annotation was performed using Prokka 1.12 (14). The genome contains 4,097 predicted protein-encoding genes with deduced functions and 1,242 genes (30.31%) coding for hypothetical proteins. Coding genes were identified using RNAmmer (15) and tRNAscan (16). The draft genome encodes 8 rRNAs and 72 tRNAs.

Using the CosmosID package (CosmosID, Inc., Rockville, MD, USA), no putative virulence factors were identified from *P. rettgeri* NVIT03. As the dominant bacterium of the microbiome of *Nasonia* spp., annotation of the *P. rettgeri* genome increases our insight into the mechanisms of this host-bacterium association.

Data availability. This whole-genome shotgun project has been deposited at DDBJ/ENA/GenBank under the accession no. [QUAF00000000](https://doi.org/10.1093/bioinformatics/btr509). The version described in this paper is version QUAF01000000. The BioProject number is [PRJNA484797](https://doi.org/10.1093/bioinformatics/btp324).

ACKNOWLEDGMENTS

This research was funded by the Rowland Fellowship at Harvard University.

We thank Brittany Berdy of the Rowland Institute at Harvard for reviewing and editing the manuscript.

REFERENCES

- Jackson TJ, Wang HY, Nugent MJ, Griffin CT, Burnell AM, Dowds BCA. 1995. Isolation of insect pathogenic bacteria, *Providencia rettgeri*, from *Heterorhabditis* spp. *J Appl Bacteriol* 78:237–244. <https://doi.org/10.1111/j.1365-2672.1995.tb05022.x>.
- Washington MA, Barnhill J, Griffin JM. 2015. A case of wound infection with *Providencia rettgeri* and coincident gout in a patient from Guam. *Hawaii J Med Public Health* 74:375–377.
- O'Hara CM, Brenner FW, Miller JM. 2000. Classification, identification, and clinical significance of *Proteus*, *Providencia*, and *Morganella*. *Clin Microbiol Rev* 13:534–546. <https://doi.org/10.1128/CMR.13.4.534-546.2000>.
- Gupta AK, Rastogi G, Nayduch D, Sawant SS, Bhonde RR, Shouche YS. 2014. Molecular phylogenetic profiling of gut-associated bacteria in larvae and adults of flesh flies. *Med Vet Entomol* 28:345–354. <https://doi.org/10.1111/mve.12054>.
- Brucker RM, Bordenstein SR. 2012. The roles of host evolutionary relationships (genus: *Nasonia*) and development in structuring microbial communities. *Evolution* 66:349–362. <https://doi.org/10.1111/j.1558-5646.2011.01454.x>.
- Moran NA. 2007. Symbiosis as an adaptive process and source of phenotypic complexity. *Proc Natl Acad Sci U S A* 104:8627–8633. <https://doi.org/10.1073/pnas.0611659104>.
- Douglas AE. 2015. Multiorganismal insects: diversity and function of resident microorganisms. *Annu Rev Entomol* 60:17–34. <https://doi.org/10.1146/annurev-ento-010814-020822>.
- 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>.
- Li H. 2011. A statistical framework for SNP calling, mutation discovery, association mapping and population genetical parameter estimation from sequencing data. *Bioinformatics* 27:2987–2993. <https://doi.org/10.1093/bioinformatics/btr509>.
- Quinlan AR. 2014. BEDTools: the Swiss-army tool for genome feature analysis. *Curr Protoc Bioinformatics* 47:11.12.1–11.12.34. <https://doi.org/10.1002/0471250953.bi1112s47>.
- Li H, Durbin R. 2009. Fast and accurate short read alignment with Burrows-Wheeler transform. *Bioinformatics* 25:1754–1760. <https://doi.org/10.1093/bioinformatics/btp324>.
- Bankevich A, Nurk S, Antipov D, Gurevich AA, Dvorkin M, Kulikov AS, Lesin VM, Nikolenko SI, Pham S, Pribelski 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>.
- Carattoli A, Zankari E, Garcia-Fernandez A, Larsen MV, Lund O, Villa L, Aarestrup FM, Hasman H. 2014. In silico detection and typing of plasmids using PlasmidFinder and plasmid multilocus sequence typing. *Antimicrob Agents Chemother* 58:3895–3903. <https://doi.org/10.1128/AAC.02412-14>.
- Seemann T. 2014. Prokka: rapid prokaryotic genome annotation. *Bioinformatics* 30:2068–2069. <https://doi.org/10.1093/bioinformatics/btu153>.
- Lagesen K, Hallin P, Rodland EA, Staerfeldt HH, Rognes T, Ussery DW. 2007. RNAmmer: consistent and rapid annotation of ribosomal RNA genes. *Nucleic Acids Res* 35:3100–3108. <https://doi.org/10.1093/nar/gkm160>.
- Lowe TM, Eddy SR. 1997. tRNAscan-SE: a program for improved detection of transfer RNA genes in genomic sequence. *Nucleic Acids Res* 25:955–964. <https://doi.org/10.1093/nar/25.5.955>.