

7-28-2016

Draft Genome Sequence of the Emerging Bivalve Pathogen *Vibrio tubiashii* subsp. *europaeus*

Edward J. Spinard

Javier Dubert

David R. Nelson
University of Rhode Island, dnelson@uri.edu

Marta Gomez-Chiarri
University of Rhode Island, gomezchi@uri.edu

Juan L. Barja

Follow this and additional works at: https://digitalcommons.uri.edu/cmb_facpubs

Creative Commons License



This work is licensed under a [Creative Commons Attribution 4.0 License](https://creativecommons.org/licenses/by/4.0/).

Citation/Publisher Attribution

Spinard, E. J., Dubert, J., Nelson, D. R., Gomez-Chiarri, M., and Barja, J. L. (2016). Draft genome sequence of the emerging bivalve pathogen *Vibrio tubiashii* subsp. *europaeus*. *Genome Announcements*, 4 (4): e00625-16. doi: 10.1128/genomeA.00625-16
Available at: <http://dx.doi.org/10.1128/genomeA.00625-16>

This Article is brought to you for free and open access by the Cell and Molecular Biology at DigitalCommons@URI. It has been accepted for inclusion in Cell and Molecular Biology Faculty Publications by an authorized administrator of DigitalCommons@URI. For more information, please contact digitalcommons@etal.uri.edu.

Draft Genome Sequence of the Emerging Bivalve Pathogen *Vibrio tubiashii* subsp. *europaeus*

Edward J. Spinard,^a Javier Dubert,^b  David R. Nelson,^a Marta Gomez-Chiarri,^c Juan L. Barja^b

Department of Cell and Molecular Biology, University of Rhode Island, Kingston, Rhode Island, USA^a; Departamento de Microbiología y Parasitología, CIBUS, Facultad de Biología, Universidade de Santiago de Compostela, Santiago de Compostela, Spain^b; Department of Fisheries, Animal and Veterinary Sciences, University of Rhode Island, Kingston, Rhode Island, USA^c

***Vibrio tubiashii* subsp. *europaeus* is a bivalve pathogen isolated during episodes of mortality affecting larval cultures in different shellfish hatcheries. Here, we announce the draft genome sequence of the type strain PP-638 and describe potential virulence factors, which may provide insight into the mechanism of pathogenicity.**

Received 10 May 2016 Accepted 10 June 2016 Published 28 July 2016

Citation Spinard EJ, Dubert J, Nelson DR, Gomez-Chiarri M, Barja JL. 2016. Draft genome sequence of the emerging bivalve pathogen *Vibrio tubiashii* subsp. *europaeus*. *Genome Announc* 4(4):e00625-16. doi:10.1128/genomeA.00625-16.

Copyright © 2016 Spinard 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 David R. Nelson, dnelson@uri.edu.

Vibrio tubiashii subsp. *europaeus* is an emerging bivalve pathogen identified recently as the etiological agent responsible for larval and spat mortalities in clam, oyster, and abalone cultures detected in Spanish and French hatcheries (1, 2). This pathogen is a causative agent of vibriosis, inducing mass mortalities and important economic losses, representing the main bottleneck for the production process in shellfish aquaculture (1, 2).

V. tubiashii subsp. *europaeus* PP-638^T (= CECT 8136^T = DSM 27349^T) was originally isolated from a culture tank of flat oyster (*Ostrea edulis*) during an episode of larval mortality in a shellfish hatchery (Galicia, Northwest Spain) (1). DNA was isolated from *V. tubiashii* subsp. *europaeus* PP-638^T grown overnight in YP30 using the Wizard genomic DNA purification kit (Promega), according to the manufacturer's instructions, except DNA was resuspended in 2 mM Tris-HCl buffer (Bio Basic). Genomic DNA was sequenced using an Illumina MiSeq at the Genomics and Sequencing Center at the University of Rhode Island, Kingston, RI. Reads were trimmed using CLC Genomics Workbench (version 8.5.1) for quality, ambiguous nucleotides, and adapters. A total of 2,943,708 paired-end and 3,234,516 mate-paired reads providing 199× coverage were assembled using SPAdes (version 3.1.1), using default parameters (3). Contigs were filtered based on 34× coverage and 4,000-bp length, resulting in 10 contigs with an N_{50} of 1,788,614 and an average G+C content of 45.37%. The assembly was mapped to *Vibrio tubiashii* ATCC 19109 using the CLC Microbial Genome Finishing module and resulted in six contigs mapping to chromosome 1, one complete contig representing chromosome 2, one complete contig representing the p251-like megaplasmid, and one contig mapping to the p57-like plasmid (4). One 4,885-bp contig did not map to the reference genome. The draft genome was submitted to Rapid Annotations using Subsystems Technology (RAST) for annotation, resulting in 5,157 open reading frames (5–7).

Encoded on chromosome 2 of the *V. tubiashii* subsp. *europaeus* PP-638^T genome is a putative metalloprotease with 75% similarity to VtpA found in *Vibrio coralliilyticus* RE22 (8). Another protease with 71% similarity to Epp in *Vibrio anguillarum* M93Sm is en-

coded on chromosome 2 (9). Three putative hemolysins and phospholipases are encoded in the genome. One hemolysin located on chromosome 2 has 67% similarity to Plp in *V. anguillarum* M93Sm (10, 11). In *V. anguillarum* M93Sm, *plp* is divergently transcribed from the pore-forming hemolysin/cytolysin *vah1* (11). In *V. tubiashii* subsp. *europaeus* PP-638^T, the Plp homolog is also divergently transcribed away from a pore-forming cytolysin, although it has 42% similarity to aerolysin in *Aeromonas eucrenophila* (NCBI Reference Sequence WP_042642875.1), not *vah1*. The genome encodes two secretion systems (type III secretion system [T3SS] and T6SS) that are used to deliver effector molecules directly into the host. The T3SS-secreted virulence factor has a domain similar to the GTPase-activating domain found on YopE from *Yersinia pestis* (12–16). While the T6SS structural components are encoded on the p251-like megaplasmid, the protein responsible for forming the puncturing tip of the T6SS secretion system, VgrG, appears to be encoded by two genes. One VgrG-encoding gene is on chromosome 1, and the second is on chromosome 2.

Nucleotide sequence accession numbers. This whole-genome shotgun project has been deposited in DDBJ/EMBL/GenBank under the accession no. [LUAX00000000](https://www.ncbi.nlm.nih.gov/nuclink/LUAX00000000). The version described in this paper is the first version LUAX01000000.

ACKNOWLEDGMENTS

This material is based upon work conducted at a Rhode Island NSF EPS-CoR research facility, the Genomics and Sequencing Center, supported in part by the National Science Foundation EPSCoR Cooperative Agreement no. EPS-1004057 and by grant AGL2014–59655 from the Ministry of Economy and Competitiveness of Spain.

FUNDING INFORMATION

This work, including the efforts of Juan L. Barja, was funded by Ministry of Economy and Competitiveness of Spain (AGL2014-59655).

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

REFERENCES

- Prado S, Dubert J, Barja JL. 2015. Characterization of pathogenic vibrios isolated from bivalve hatcheries in Galicia, NW Atlantic coast of Spain. Description of *Vibrio tubiashii* subsp. *europaensis* subsp. nov. Syst Appl Microbiol 38:26–29. <http://dx.doi.org/10.1016/j.syapm.2014.11.005>.
- Travers MA, Mersni Achour R, Haffner P, Tourbiez D, Cassone AL, Morga B, Doghri I, Garcia C, Renault T, Fruitier-Arnaudin I, Saulnier D. 2014. First description of French *V. tubiashii* strains pathogenic to mollusk: I. Characterization of isolates and detection during mortality events. J Invertebr Pathol 123:38–48. <http://dx.doi.org/10.1016/j.jip.2014.04.009>.
- Nurk S, Bankevich A, Antipov D, Gurevich AA, Korobeynikov A, Lapidus A, Pribelski AD, Pyshkin A, Sirotkin A, Sirotkin Y, Stepanauskas R, Clingenpeel SR, Woyke T, McLean JS, Lasken R, Tesler G, Alekseyev MA, Pevzner PA. 2013. Assembling single-cell genomes and mini-metagenomes from chimeric MDA products. J Comput Biol 20: 714–737. <http://dx.doi.org/10.1089/cmb.2013.0084>.
- Richards GP, Needleman DS, Watson MA, Bono JL. 2014. Complete genome sequence of the larval shellfish pathogen *Vibrio tubiashii* type strain ATCC 19109. Genome Announc 2(6):e01252-14. <http://dx.doi.org/10.1128/genomeA.01252-14>.
- Aziz RK, Bartels D, Best AA, DeJongh M, Disz T, Edwards RA, Formsma K, Gerdes S, Glass EM, Kubal M, Meyer F, Olsen GJ, Olson R, Osterman AL, Overbeek RA, McNeil LK, Paarmann D, Paczian T, Parrello B, Pusch GD, Reich C, Stevens R, Vassieva O, Vonstein V, Wilke A, Zagnitko O. 2008. The RAST server: Rapid Annotations using Subsystems Technology. BMC Genomics 9:75. <http://dx.doi.org/10.1186/1471-2164-9-75>.
- Brettin T, Davis JJ, Disz T, Edwards RA, Gerdes S, Olsen GJ, Olson R, Overbeek R, Parrello B, Pusch GD, Shukla M, Thomason JA, III, Stevens R, Vonstein V, Wattam AR, Xia F. 2015. RASTtk: a modular and extensible implementation of the RAST algorithm for building custom annotation pipelines and annotating batches of genomes. Sci Rep 5:8365. <http://dx.doi.org/10.1038/srep08365>.
- Overbeek R, Olson R, Pusch GD, Olsen GJ, Davis JJ, Disz T, Edwards RA, Gerdes S, Parrello B, Shukla M, Vonstein V, Wattam AR, Xia F, Stevens R. 2014. The SEED and the rapid annotation of microbial genomes using subsystems technology (RAST). Nucleic Acids Res 42: D206–D214. <http://dx.doi.org/10.1093/nar/gkt1226>.
- Hasegawa H, Lind EJ, Boin MA, Häse CC. 2008. The extracellular metalloprotease of *Vibrio tubiashii* is a major virulence factor for pacific oyster (*Crassostrea gigas*) larvae. Appl Environ Microbiol 74:4101–4110. <http://dx.doi.org/10.1128/AEM.00061-08>.
- Varina M, Denkin SM, Staroscik AM, Nelson DR. 2008. Identification and characterization of Epp, the secreted processing protease for the *Vibrio anguillarum* EmpA metalloprotease. J Bacteriol 190:6589–6597. <http://dx.doi.org/10.1128/JB.00535-08>.
- Li L, Mou X, Nelson DR. 2013. Characterization of Plp, a phosphatidylcholine-specific phospholipase and hemolysin of *Vibrio anguillarum*. BMC Microbiol 13:271. <http://dx.doi.org/10.1186/1471-2180-13-271>.
- Rock JL, Nelson DR. 2006. Identification and characterization of a hemolysin gene cluster in *Vibrio anguillarum*. Infect Immun 74:2777–2786. <http://dx.doi.org/10.1128/IAI.74.5.2777-2786.2006>.
- Marchler-Bauer A, Anderson JB, Chitsaz F, Derbyshire MK, DeWeese-Scott C, Fong JH, Geer LY, Geer RC, Gonzales NR, Gwadz M, He S, Hurwitz DI, Jackson JD, Ke Z, Lanczycki CJ, Liebert CA, Liu C, Lu F, Lu S, Marchler GH, Mullokandov M, Song JS, Tasneem A, Thanki N, Yamashita RA, Zhang D, Zhang N, Bryant SH. 2009. CDD: specific functional annotation with the Conserved Domain Database. Nucleic Acids Res 37:D205–D210. <http://dx.doi.org/10.1093/nar/gkn845>.
- Marchler-Bauer A, Bryant SH. 2004. CD-search: protein domain annotations on the fly. Nucleic Acids Res 32:W327–W331. <http://dx.doi.org/10.1093/nar/gkh454>.
- Marchler-Bauer A, Derbyshire MK, Gonzales NR, Lu S, Chitsaz F, Geer LY, Geer RC, He J, Gwadz M, Hurwitz DI, Lanczycki CJ, Lu F, Marchler GH, Song JS, Thanki N, Wang Z, Yamashita RA, Zhang D, Zheng C, Bryant SH. 2015. CDD: NCBI's conserved domain database. Nucleic Acids Res 43:D222–D226. <http://dx.doi.org/10.1093/nar/gku1221>.
- Marchler-Bauer A, Lu S, Anderson JB, Chitsaz F, Derbyshire MK, DeWeese-Scott C, Fong JH, Geer LY, Geer RC, Gonzales NR, Gwadz M, Hurwitz DI, Jackson JD, Ke Z, Lanczycki CJ, Lu F, Marchler GH, Mullokandov M, Omelchenko MV, Robertson CL, Song JS, Thanki N, Yamashita RA, Zhang D, Zhang N, Zheng C, Bryant SH. 2011. CDD: a Conserved Domain Database for the functional annotation of proteins. Nucleic Acids Res 39:D225–D229. <http://dx.doi.org/10.1093/nar/gkq1189>.
- Evdokimov AG, Tropea JE, Routzahn KM, Waugh DS. 2002. Crystal structure of the *Yersinia pestis* GTPase activator YopE. Protein Sci 11: 401–408. <http://dx.doi.org/10.1110/ps.34102>.