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Draft Genome Sequence of the Marine Pathogen *Vibrio coralliilyticus* RE22

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***Vibrio coralliilyticus* RE22 is a causative agent of vibriosis in larval bivalves. We report here the draft genome sequence of *V. coralliilyticus* RE22 and describe additional virulence factors that may provide insight into its mechanism of pathogenicity.**

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Vibrio coralliilyticus RE22 (formerly *Vibrio tubiashii* RE22) is a marine pathogen and a causative agent of vibriosis in larval bivalves (1). The disease is characterized by high mortality rates leading to a severe loss of production in shellfish hatcheries (2–4). Currently, only two proteases (VtpA and VtpB) and one hemolysin (VthA) have been characterized in RE22 (5–7). To better understand the mechanisms of pathogenicity, it is necessary to discover additional potential virulence factors. Here, we announce the draft genome sequence of *V. coralliilyticus* RE22 and selectively describe some potential virulence factors.

V. coralliilyticus RE22Sm (a spontaneous mutant resistant to streptomycin) was grown overnight in yeast-peptone broth supplemented with 3% NaCl (YP30) at 27°C in a shaking water bath. Genomic DNA was isolated using the Wizard genomic DNA purification kit (Promega), according to the manufacturer's instructions, except DNA was resuspended into 100 μl of a 2 mM Tris-HCl (pH 8) solution. DNA was sequenced at the Rhode Island Genomics Sequencing Center, Kingston, RI, using an Illumina MiSeq Sequencer. Reads were trimmed using the CLC Genomics Workbench (version 8.0.1) for quality, ambiguous base pairs, adapters, duplicates, and size, resulting in 7,602,646 paired-end and mate-paired reads averaging 235.84 bp in size. The reads were assembled using the *de novo* assembly algorithm of CLC Genomics Workbench and SPAdes genomic assembler (version 3.1.1) (8). Contigs with an average coverage of >110 reads were joined using the CLC Microbial Genome Finishing module using *V. coralliilyticus* OCN014 as a reference genome. In total, the draft genome is composed of five contigs. Three contigs totaling 3.46 Mbp and having an average G+C content of 46% mapped to chromosome 1 of *V. coralliilyticus* OCN014. The complete chromosome 2 is represented by one 1.90-Mbp contig with a G+C content of 45%. A megaplasmid is represented by one 0.32-Mbp contig with a G+C content of 50%. The draft genome was annotated using Rapid Annotations using Subsystems Technology (RAST) and resulted in 5,234 open reading frames (9–11).

The genome of *V. coralliilyticus* RE22 encodes two extracellular metalloproteases besides those encoded by the previously described *vtpA* and *vtpB* genes. One protease shows similarity to the

Epp protease in *Vibrio anguillarum* (12), while the other contains a domain conserved in the M4 family of metalloproteases (13–17). In addition to *vthA*, three putative hemolysin/cytolysin genes were discovered. A putative MARTX toxin operon encoding three type 1 secretion system (T1SS) transport proteins, a MARTX toxin, and a hypothetical protein is on the megaplasmid. Unlike typical MARTX toxin gene clusters, the transporter genes are not transcribed divergently from the MARTX toxin (18). Instead, they seem to be in the MARTX operon, upstream of the MARTX toxin gene. Unlike most MARTX toxin gene clusters, no *rtxC* (acyltransferase) is present in the operon. Additional putative hemolysins include a phospholipase/hemolysin located on chromosome 2 that shows similarity to *plp* in *V. anguillarum* (19) and a hemolysin annotated as *hlyA* located on chromosome 1 that shows similarity to *vah1* in *V. anguillarum* (20).

Nucleotide sequence accession numbers. This whole-genome shotgun project has been deposited in DDBJ/ENA/GenBank under the accession no. [LGLS00000000](https://www.ncbi.nlm.nih.gov/nuclink/LGLS00000000). The version described in this paper is the first version, [LGLS01000000](https://www.ncbi.nlm.nih.gov/nuclink/LGLS01000000).

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REFERENCES

1. Wilson B, Muirhead A, Bazanella M, Huete-Stauffner C, Vezzulli L, Bourne DG. 2013. An improved detection and quantification method for the coral pathogen *Vibrio coralliilyticus*. *PLoS One* 8:e81800. <http://dx.doi.org/10.1371/journal.pone.0081800>.
2. Estes R, Friedman C, Elston R, Herwig R. 2004. Pathogenicity testing of shellfish hatchery bacterial isolates on Pacific oyster *Crassostrea gigas* larvae. *Dis Aquat Org* 58:223–230. <http://dx.doi.org/10.3354/dao058223>.
3. Elston R, Hasegawa H, Humphrey K, Polyak I, Häse C. 2008. Re-

- emergence of *Vibrio tubiashii* in bivalve shellfish aquaculture: severity, environmental drivers, geographic extent and management. *Dis Aquat Org* 82:119–134. <http://dx.doi.org/10.3354/dao01982>.
4. Sindermann CJ, Lightner DV. 1988. Disease diagnosis and control in North American marine aquaculture, 2nd ed. Elsevier, Amsterdam, The Netherlands.
 5. Hasegawa H, Hase CC. 2009. TetR-type transcriptional regulator VtpR functions as a global regulator in *Vibrio tubiashii*. *Appl Environ Microbiol* 75:7602–7609. <http://dx.doi.org/10.1128/AEM.01016-09>.
 6. Hasegawa H, Lind EJ, Boin MA, Hase 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>.
 7. Hasegawa H, Hase CC. 2009. The extracellular metalloprotease of *Vibrio tubiashii* directly inhibits its extracellular haemolysin. *Microbiology* 155: 2296–2305. <http://dx.doi.org/10.1099/mic.0.028605-0>.
 8. Nurk S, Bankevich A, Antipov D, Gurevich AA, Korobeynikov A, Lapidus A, Prjibelski 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>.
 9. 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>.
 10. 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>.
 11. 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>.
 12. 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>.
 13. Adekoya OA, Sylte I. 2009. The thermolysin family (M4) of enzymes: therapeutic and biotechnological potential. *Chem Biol Drug Des* 73:7–16. <http://dx.doi.org/10.1111/j.1747-0285.2008.00757.x>.
 14. 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>.
 15. 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>.
 16. 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>.
 17. 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>.
 18. Satchell KJF. 2007. MARTX, multifunctional autoprocessing repeats-in-toxin toxins. *Infect Immun* 75:5079–5084. <http://dx.doi.org/10.1128/IAI.00525-07>.
 19. 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>.
 20. 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>.