

Complete Genome Sequence of Vibrio campbellii DS40M4

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ABSTRACT We present the complete genome sequence of Vibrio campbellii DS40M4, assembled from Illumina and Oxford Nanopore data. This effort improves upon a previous draft assembly to resolve this organism's two-chromosome and one-plasmid genetic structure and to provide valuable context for evaluating the gene arrangement and evolution of this species.

*V*ibrio campbellii is a core member of the Vibrio Harveyi clade, the members of which have been studied as models for quorum sensing and the role of this regulatory process in biological events such as bioluminescence and virulence [\(1](#page-1-0)[–](#page-1-1)[5\)](#page-1-2). V. campbellii DS40M4 was isolated from the open ocean in a tropical region of the Northeast Atlantic Ocean off the northwestern African coast between Cape Verde and the Canary Islands, and its taxonomic assignment as V. campbellii, as well as specific isolation and cultivation methods, were previously reported [\(6,](#page-1-3) [7\)](#page-1-4).

Genomic DNA was extracted using the Gentra Puregene Yeast/Bact. kit (Qiagen) and sequenced using an Illumina MiSeq platform (Nextera XT kit and version 2 300-cycle kit $[2 \times 150$ -bp paired-end reads]) and a MinION Mk1B device (1D ligation kit and SpotON flow cell R9.4; Oxford Nanopore Technologies). A total of 515,350 Illumina paired-end reads and 88,896 MinION reads were used for hybrid de novo assembly with Unicycler (version 0.4.4 beta) in conservative mode [\(8,](#page-1-5) [9\)](#page-1-6). The final assembly was annotated using the NCBI Prokaryotic Genome Annotation Pipeline (version 4.6) and the RAST server for comparison [\(10\)](#page-1-7). Genome coverages were 59 \times and 95 \times , and the mean read lengths were 300 bp and 12,300 bp for the Illumina and MinION reads, respectively.

The resulting hybrid assembly consists of three circular replicons, with a total size of 5.21 Mb and 45.1% GC content (chromosome [Chr] I, 3.33 Mb; Chr II, 1.88 Mb; plasmid, 77.3 kb). The closest complete genome sequence determined by average nucleotide identity [\(11\)](#page-1-8) is V. campbellii ATCC 25920^T (98.02% identity). This assembly was aligned with all available closed V. campbellii genomes in GenBank to determine overall completeness and compare gene organization using QUAST [\(12\)](#page-1-9) and progressive-Mauve [\(13\)](#page-1-10). Of the 56,338 open reading frames (ORFs) identified, the NCBI annotation predicted 4,694 protein-coding sequences. In addition to confirming the genetic content previously reported [\(6\)](#page-1-3), including the presence of a number of potential virulence factors using VFanalyzer [\(14\)](#page-1-11), this assembly resolved the presence and locations of 12 rRNA operons and 135 tRNAs, as opposed to the 4 rRNA operons and 83 tRNAs from the previous assembly. The numerous rRNA operons are characteristic of this clade and represent an interesting factor possibly affecting growth rates and habitat adaptation. PHASTER predicts one intact prophage sequence (39.8 kb) and two other possible chromosomal regions for phage elements [\(15\)](#page-1-12). The putative plasmid, which was not identified in the previous assembly, harbors a type II toxin-antitoxin locus that may be important in natural plasmid maintenance and is similar to loci found

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on the reported plasmids of V. campbellii strains BAA-1116 and ATCC 25920^T. Since phage and plasmid-mediated lateral gene transfer may contribute to shaping the genetic content in bacteria, assembling these elements may lend insight into acquired fitness advantages despite maintenance costs, as in the case of plasmids. In comparison to a previous draft genome sequence assembly that generated 121 contigs [\(6\)](#page-1-3), this effort has provided a complete assembly allowing for improved evaluations of gene synteny, regulation, and evolution and further insight into the genetic underpinnings of niche response and adaptation.

Data availability. The complete genome sequence was deposited in DDBJ/EMBL/ GenBank under the accession numbers [CP030788](https://www.ncbi.nlm.nih.gov/nuccore/CP030788) to [CP030790.](https://www.ncbi.nlm.nih.gov/nuccore/CP030790) The versions described in this paper are versions CP030788.1 to CP030790.1. The raw reads were deposited in the SRA under BioProject number [PRJNA479421.](https://www.ncbi.nlm.nih.gov/bioproject/PRJNA479421)

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