Whole-Genome Sequences of Vibrio Species from Warm-Water Shrimps Imported into Canada: Detection of Genetic Elements Associated with Antimicrobial Resistance and Potential Mobilizing Capacities

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ABSTRACT We present draft genome sequences of Vibrio species (Vibrio alginolyticus, Vibrio cholerae, and two Vibrio parahaemolyticus strains) that were isolated from warm-water shrimps imported into Canada. All four isolates harbor genetic elements associated with antimicrobial resistance (AMR), including mobile genetic elements that can promote horizontal transfer of AMR genes.

Vibrio species are Gram-negative bacteria associated with plankton and estuary-harvested seafood. The genus Vibrio includes more than 100 known species, a dozen of which are known to be capable of infecting humans (1). Infections are associated with exposure to seawater or consumption of seafood containing infectious loads of pathogenic biotypes of Vibrio species (1, 2). Globally, Vibrio parahaemolyticus is implicated in most seafood-linked foodborne illnesses (3–5). Some Vibrio cholerae strains express cholera toxins, particularly the serovars O1 and O139, which have been associated with multiple pandemics to date and cause life-threatening severe diarrhea (6). Vibrio alginolyticus is an emerging pathogen that is known to be linked to skin infections and mild diarrhea (2).

As part of our surveillance analysis of warm-water shrimps imported into Canada, we isolated and characterized clinically significant Vibrio species and identified a subset of strains that exhibited multidrug resistance (MDR), defined as resistance to three or more antibiotics. Here, we report the whole-genome sequences (WGS) of four MDR Vibrio strains. These strains were isolated and characterized at the species level as described previously (7) and were stored frozen at −80°C, with antimicrobial resistance (AMR) profiles determined by the Kirby-Bauer disk diffusion method (8, 9). The antibiotic susceptibility test results showed resistance to up to nine different antibiotics (Table 1). For DNA isolation, stock cultures were struck onto tryptic soy agar with 2% NaCl (TSA-2N) (Difco BD, Franklin Lakes, NJ, USA), and single colonies were grown overnight at 35°C on TSA-2N. Genomic DNA was extracted using the Maxwell 16-cell DNA purification kit (Promega, Madison, WI, USA), and indexed libraries were prepared using the Nextera XT kit and sequenced on a MiSeq instrument (2 × 300-bp paired-end reads, v3 chemistry) according to the manufacturer’s protocol (Illumina, San Diego, CA, USA). Sequence analysis tools were used with default settings for read adapter trimming, quality filtering, and error correction (BBMap v38.26 [BBDuk and Tadpole]) (https://sourceforge.net/projects/bbmap), de novo assembly (SKESA v2.3 [SVN 551987:557549M] and Pilon v1.22) (10), gene prediction (Prodigal [commit fe80417]) (11), and summary metrics (QUAST v5.0.0 [de6973bb]) (12). AMR gene and plasmid characterizations were predicted by in-house scripts using the Resistance Gene Identifier (RGI) v5.0.0/Comprehensive Antibiotic Resistance Database (CARD)

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**TABLE 1** Genome profiles and AMR results for the four *Vibrio* strains

| Strain no. | Organism       | GenBank accession no. | SRA accession no. | Genome size (Mbp) | No. of reads | Genome coverage (x) | No. of contigs | GC content (%) | No. of genes | AMR phenotype* | Plasmid type | Predicted mobility | Plasmid genotype |
|------------|----------------|-----------------------|-------------------|------------------|--------------|---------------------|---------------|----------------|--------------|----------------|-------------|----------------|------------------|
| ISF-22-6   | *V. alginolyticus* | JAILX0100000000       | SRR15526828       | 5.2              | 468,108      | 90                  | 73            | 343,022        | 44.4         | AMP, SXT, TE, SF, OT (E, KF) | Unknown     | Nonmobilizable   | CARB-19, tet(3S), aph(3')-Ib, aph (6)-Id, floR, sul2, varG, catB9, sul1, aac(6')-IId, catB3, sul1, mphiA, PER3 |
| ISF-208-6  | *V. cholerae*    | JAILXN0100000000      | SRR15526827       | 4.4              | 726,913      | 160                 | 82            | 206,565        | 47.4         | AMP, CTX, K, S, KF, CTF, PIP, OX, E (ENO) | IncA/C2     | Conjugative       | varG, catB9, sul1, aac(6')-IId, catB3, sul1, mphiA, PER3 |
| ISF-232-3  | *V. parahaemolyticus* | JAILXM0100000000     | SRR15526826       | 5.1              | 190,219      | 30                  | 161           | 80,238         | 45.3         | AMP, CTX, K, KF, CTF (E) | IncQ1       | Mobilizable       | aac(6')-Ilo, ANT (Z')-io, VEB-5 |
| ISF-238-3  | *V. parahaemolyticus* | JAILXL0100000000     | SRR15526825       | 5.4              | 592,288      | 465                 | 85            | 195,494        | 45.3         | AMP, S, SF, OT (CTF, E, KF) | Unknown     | Nonmobilizable   | aac(6')-Ib, aph(6)-Id, floR, sul2 |

*AMP, ampicillin; CTF, ceftiofur; CTX, cefotaxime; E, erythromycin; ENO, enrofloxacin; K, kanamycin; KF, cephalothin; OT, oxytetracycline; OX, oxolinic acid; PIP, piperacillin; S, streptomycin; SF, sulfafurazole; SXT, sulfamethoxazole-trimethoprim; TE, tetracycline. Abbreviations shown in parentheses refer to intermediate resistance.*
v3.0.3 (13) and MOB-suite v1.4.9 tools and database (14). Isolate details are summarized in Table 1.

A V. parahaemolyticus strain isolated in-house from Canadian molluscan shellfish was shown to harbor an integrative and conjugative element (ICE) belonging to the SXT/R391 family, located on chromosome I (15). This type of mobilizing capacity is known to be associated with adaptation and evolution (16), including acquired AMR traits. The widespread detection of several vector systems in Gram-negative bacteria has been reported and reviewed (17). WGS of the submitted Vibrio species with diverse extrachromosomal elements are available for the assessment of various in silico tools to predict phenotypic AMR detected in the Vibrio isolates using standard laboratory procedures (8, 9) and for the advancement and improvement of WGS-based prediction of AMR phenotypes.

Data availability. The WGS data have been deposited in EMBL/GenBank as BioProject PRJNA645603, under the accession numbers listed in Table 1.

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