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Draft genome of a *Pseudovibrio* sp. isolated from the skeleton of *Pachyseris speciosa* from a Singaporean reef

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ABSTRACT A *Pseudovibrio* sp. was isolated from the skeleton of the heat resilient coral *Pachyseris speciosa*. Genome analysis revealed the presence of the complete denitrification pathway and potential dimethylsulfoniopropionate metabolism which enhance coral resilience and production of tropodithietic acid, an antibiotic implicated in host defense.

KEYWORDS corals, marine microbiology, *Pseudovibrio*

Pseudovibrio spp. belong to the family *Stappiaceae* (1) (class *Alphaproteobacteria*) (2) and are thought to be beneficial symbionts to marine invertebrates, such as sponges and corals (3). They can survive at a wide range of temperatures and fluctuating nutrients (4). They have the potential to act as probiotics in different marine systems due to the metabolites they produce, like tropodithietic acid (TDA), which inhibits *Vibrio* pathogens (5).

Pachyseris speciosa samples were collected from the seawall at the northern side of Kusu Island, Singapore, and maintained in an outdoor aquaria tank with a constant seawater flow. Samples were fragmented into ~9 cm^2 (~2 g) fragments and ground with a mortar and pestle while mixing ~5 mL of sterile artificial seawater (macerate). Samples were treated with proteinase K, vortexed with glass beads, diluted with sterile artificial seawater, then spread plated on marine agar (Difco 2216) and incubated at 33°C until colonies were observed. Individual bacterial colonies were subcultured to ensure purity, and a single colony was used to inoculate marine broth for subsequent processing and sequencing.

Genomic DNA extraction used the DNeasy Powersoil Pro Kit (Qiagen, Hilden, Germany). DNA library preparation used the Accel-NGS 2S Plus Low Input DNA Library Prep Kit (Swift Biosciences, Ann Arbor, USA) before sequencing using an Illumina HiSeq X Ten platform, version 2.5 (Illumina, San Diego, CA, USA), generating 150-bp paired-end reads. DNA library preparation and sequencing were performed at Singapore Centre for Environmental Life Sciences Engineering, Nanyang Technological University.

Paired-end reads were trimmed using Trimmomatic, version 0.39 (6). Default parameters were used for all software unless specified. Genome assembly was performed using the Shovill pipeline, version 1.1.0 (https://github.com/tseemann/shovill), and the St. Petersburg genome Assembler (SPAdes), version 3.15.5 (7). Genome annotation was performed using PGAP, version 6.7 (8). The assembly (Table 1) was checked for plasmids with PlasmidFinder, version 2.0.1 (9), but no plasmids were found.

Identification of the isolate was performed on the Microbial Genomes Atlas (10) web server using the NCBI-Prok function, with the closest homology to *Pseudovibrio* sp. FO-BEG1 based on average amino acid identity. Phylogenetic relatedness between the isolate and all publicly available *Pseudovibrio* genomes was determined using both the Orthologous Average Nucleotide Identity Tool, version 0.93.1 (11) and the

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TABLE 1 Genomic features and accession numbers of *Pseudovibrio* sp.[a]

Isolate name	Genome size (bp)	Average coverage	No. of CDS	No. of rRNAs	No. of tRNAs	G + C content (%)	No. of contigs	N_{50} (bp)	L_{50}	Assembly accession no.	SRA accession no.	No. of reads	Average read length (bp)
SCP19[b]	5,167,238	31.4	4,671	8	70	52.5	33	892,587	2	JBDZYJ000000000	SRR28745652	12,626,488	151

[a]CDS, coding DNA sequences; SCP, Singapore coral probiotic.
[b]SCP, Singapore coral probiotic.

Genome-To-Genome Distance Calculator, version 3.0 (12), to calculate the average nucleotide identity (ANI) and DNA-DNA hybridization (DDH) scores, respectively. Results showed a >95% ANI score with *Pseudovibrio denitrificans* DSM 17465 but did not share ≥70% DDH (species threshold) (13) with any *Pseudovibrio* genomes.

Pseudovibrio sp. SCP19 (for Singapore coral probiotics) produced brown colonies, and its genome encoded genes for the core enzymes of TDA production (*tdaABCDEF*) (5). The genome also encoded a dimethylsulfoniopropionate (DMSP) lyase gene (*dddD*) (14), indicating a potential to cleave DMSP into the cloud nucleating agent dimethylsulfide (15) and 3-hydroxypropionate, which can be converted into acrylate (16), the anti-herbivory compound (17). Analysis of the genome using GhostKOALA, version 2.0 (18), revealed the complete pathway for denitrification, thereby having the potential to reduce nitrates and nitrites into dinitrogen (19). These characteristics support the hypothesis that *Pseudovibrio* sp. SCP19 is a strong probiotic candidate.

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AUTHOR CONTRIBUTIONS

Aaron An Rong Loh, Conceptualization, Data curation, Formal analysis, Investigation, Project administration, Supervision, Validation, Writing – original draft, Writing – review and editing | Dalong Hu, Methodology, Software | Jabez Mason Yong Jun Law, Investigation | Elton Lim Wen Xiong, Methodology | Lindsey Kane Deignan, Conceptualization, Funding acquisition, Resources | Stephen Summers, Data curation, Funding acquisition, Methodology, Resources | Joao Paulo Andre Pereyra, Conceptualization, Data curation, Formal analysis, Methodology, Project administration, Software, Supervision, Validation, Writing – review and editing | Rebecca Josephine Case, Conceptualization, Funding acquisition, Methodology, Project administration, Resources, Supervision, Writing – review and editing

DATA AVAILABILITY

The SRA data were deposited to the National Center for Biotechnology Information, and the complete genome sequences were deposited to GenBank under the accession numbers listed in Table 1.

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