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GENOME SEQUENCES

Draft Genome Sequence of Terrestrial *Streptomyces* sp. Strain VITNK9, Isolated from Vellore, Tamil Nadu, India, Exhibiting Antagonistic Activity against Fish Pathogens

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ABSTRACT We report the draft genome sequence of *Streptomyces* sp. strain VITNK9, isolated from a soil sample collected in Vellore District (12.9165°N, 79.1325°E), Tamil Nadu, India, with an assembly size of 7,920,076 bp and 72.7% GC content.

Streptomyces spp. have been considered one of the most prolific sources of pharmacologically active compounds for decades, contributing approximately 60% of antibiotics which are in clinical use today. To discover natural products possessing antagonistic activity against fish pathogens, we isolated a new actinomycete strain, *Streptomyces* sp. strain VITNK9, from a terrestrial soil sample from Vellore, Tamil Nadu, India, using actinomycete isolation agar (AIA) plates (1, 2). The strain had a smooth colony surface, an earthy odor, and a spiral spore formation, including aerial mycelia and substrate hyphae penetrating through agar, morphologies typical of *Streptomycetes*. The crude extract of the strain exhibited antimicrobial activity against several fish pathogens, implying that this strain can be an important resource of bioactive metabolites (1). Further, to obtain the genomic potential of VITNK9 to synthesize bioactive compounds, its genome was sequenced.

For DNA isolation, *Streptomyces* sp. strain VITNK9 was incubated on ISP1 broth at 30°C and 250 rpm for 4 days. Genomic DNA was isolated using the High Pure PCR template preparation kit (Roche) following the manufacturer's protocol. DNA libraries were prepared using the TruSeq DNA PCR-free kit. DNA shotgun libraries were sequenced on an Illumina platform (101-bp paired-end reads) at Macrogen (South Korea), yielding a total of 6.05 Gbp of sequence with 59,886,702 reads. Sequences were trimmed using Trimmomatic version 0.36 (minlen 100, sliding window 4:20, ILLUMINACLIP:TruSeq3PE-2.fa:2:30:8) (3), and read quality was assessed using FastQC version 0.11.5 (4) prior to *de novo* assembly using SPAdes version 3.13.0 (parameters: –k 21,33,55,77,91 –careful –only-assembler –cov-cutoff auto) (5). Scaffolds of ≥2 kb were used for further analysis. Completeness and contamination were estimated with CheckM version 1.0.7 (6) based on 460 markers and using lineage workflow. For the phylogenetic analysis, sequences of the closest described type strains were obtained from EzTaxon (https://www.ezbiocloud.net/) (7). Open reading frames (ORFs) for the VITNK9 genome were identified and annotated using Rapid Annotations using Subsystems Technology (RAST) (8) with the RASTtk algorithm (https://rast.nmpdr.org/) (9).

Genome assembly of VITNK9 yielded 18 scaffolds with an N_{50} value of 1,271,901 bp and with the largest contig being 1,996,233 bp. The genome consists of 7,919,889 bp without ambiguous nucleotides (N) with a GC content of 72.7%, 100% estimated

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completeness, 0.06% contamination, and 0% heterogeneity. The VITNK9 genome contains 7,148 predicted and 73 RNA genes (68 tRNA genes). Based on EzTaxon analyses of the extracted 16S rRNA gene (876 bp) from VITNK9, the strain showed 84.11% similarity to *Streptomyces rochei* strain NRRL B-2410^T.

The closest neighbors of strain VITNK9 were *Streptomyces coelicolor* A3(2) (score, 500), *Streptomyces avermitilis* MA-4680 (score, 436), *Streptomyces scabiei* 87.22 (score, 428), *Streptomyces griseus* subsp. *griseus* NBRC 13350 (score, 377), and *Saccharopolyspora erythraea* NRRL 2338 (score, 213) detected by RAST annotation (10). The MICs of the ethyl acetate (EA) crude extract of VITNK9 were found to be moderate against the following fish pathogens: *Aeromonas hydrophila* and *Edwardsiella tarda* (0.03 mg ml^{-1}), *Vibrio anguillarum* and *Vibrio harveyi* (0.06 mg ml^{-1}, and *Aeromonas caviae* (0.125 mg ml^{-1}) (1). Furthermore, an antiSMASH version 5.1.2 analysis (11) of the VITNK9 genome revealed 30 putative biosynthetic gene clusters (BGCs). According to the prediction, 12 gene clusters possess >75% similarity to BGCs, 6 gene clusters possess 50 to 75% similarity, 8 gene clusters showed <50% similarity with known BGCs, and 4 gene clusters were detected as completely unknown. The notable BGCs found using antiSMASH prediction were in the ribosomally synthesized and posttranslationally modified peptide (RiPP) class, (lantipeptide SapB [12]), the terpene class (geosmin [13], isorenieratene [14], hopene [15], albaflavenone [16], siderophore desferrioxamine B/E [17], and ectoine [18]), nonribosmal peptide synthetase (coelibactin and coelichelin [15]), and several polyketide synthase (PKS) classes. Default parameters were used for all the tools used except where otherwise noted.

Overall, *Streptomyces* sp. strain VITNK9 possesses a high potential for diverse and bioactive secondary metabolites and can further be considered an important strain to study for bioactive compounds and their biosynthesis in the future.

Data availability. The genome of the *Streptomyces* sp. strain VITNK9 has been deposited at GenBank under accession number no. JACWAA000000000, assembly accession no. ASM1485402v1, BioProject no. PRJNA661553, and BioSample no. SAMN16063085. The raw reads are available under accession no. SRR12997718 in the Sequence Read Archive (SRA).

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