Draft Genome Sequencing of *Stenotrophomonas indicatrix* BOVIS40 and *Stenotrophomonas maltophilia* JVB5, Two Strains with Identifiable Genes Involved in Plant Growth Promotion

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ABSTRACT Here, plant growth-promoting *Stenotrophomonas* strains isolated from the sunflower root endosphere were studied. Bacterial DNA was sequenced on Illumina’s NextSeq platform. The gene prediction reveals diverse functional genes involved in plant growth promotion from each bacterial genome. The exploration of bacterial resources as bioinoculants is promising for agricultural biotechnology.

*Stenotrophomonas* species are free-living, Gram-negative nonsporeformers that are commonly found in the soil and plant environments (1). *Stenotrophomonas* species can be involved in plant growth promotion (PGP) (2), although some are yet to be explored. Therefore, genomic elucidation can help predict the diverse genes responsible for bacterial functions in agricultural biotechnology.

Bacteria isolated from sunflower roots were sourced from farmlands in Lichtenburg, South Africa (26°4′31.266″S, 25°58′44.442″E), in February 2020. The healthy sunflower plants were carefully uprooted, placed inside sterile ziplock bags, transported to the laboratory, and stored at 4°C. The root samples were cut into small pieces with a sterile scalpel and washed in sterile distilled water. To ensure complete removal of the epiphytic bacteria, surface sterilization was achieved by soaking the samples in 70% ethanol for 3 min, followed by 3% sodium hypochlorite for 3 min, 70% ethanol for 30 s, and rinsing with sterile distilled water. The level of sterility of the samples was assessed by pour plating onto Luria-Bertani (LB) medium using the last water used to rinse the plant samples. One gram of plant material was weighed, suspended in 1 M phosphate-buffered solution, and manually macerated in a mortar and pestle until a smooth suspension was obtained. Sample suspensions were serially diluted up to 10⁻⁵ dilutions, and 0.1 ml of an aliquot from dilutions 10⁻⁵ and 10⁻⁶ was pipetted into petri dishes and pour plated with sterilized LB agar. The inoculated petri plates were incubated at 28°C for 24 h. Distinct bacterial colonies that formed on the plates were counted and selected based on their morphological appearance. A pure culture of the bacterial isolate was obtained by repeated streaking onto sterile LB agar and incubated at 28°C for 24 h. The pure bacterial strains were kept on an agar slant and stored at 4°C for further use.

The pure strains were used for DNA extraction, using a commercial Quick-DNA miniprep kit specific for fungi or bacteria (Zymo Research, Irvine, CA, USA; catalog number D6005). Whole-genome sequencing was performed on Illumina’s NextSeq platform at Inqaba Biotechnical Industries (Pty.) Ltd. (Pretoria, South Africa). The sample preparation (DNA library) was performed using a NextSeq midoutput kit, and a data set (2 × 150-bp paired-end reads) was generated for each sample. Genomic sequences were analyzed on the KBase platform (https://kbase.us/) (3). The quality of the sequence reads was evaluated using FastQC version 0.11.5 (4), while sequence adaptors and low-quality bases were removed using Trimmomatic version 0.36 (5). Furthermore, the sequence reads were assembled using SPAdes version 3.13.0 (6). Gene annotation and prediction were performed using the RASTtk (Rapid Annotations using Subsystems Technology) toolkit version 1.073 and the publicly accessible catalog (RASTtk website) for each strain.

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available NCBI (https://www.ncbi.nlm.nih.gov/) Prokaryotic Genome Annotation Pipeline (PGAP) (7). All analyses were performed using default parameters unless otherwise specified.

Secondary metabolites were determined using antiSMASH version 6.0.0 (8). The sequence analysis of strain BOVIS40 yielded a sequence read count of 7,301,524 bp, a genome size of 4,427,090 bp, a G+C content of 66.4%, 4,446 coding sequence genes, 62 tRNAs, and 2 rRNAs. In addition, a sequence read count of 8,764,890 bp, a genome size of 4,771,305 bp, a G+C content of 66%, 57 tRNAs, and 4,160 coding genes were obtained from the genome assembly of strain JVB5. The predicted PGP genes and nonribosomal peptide/polyketide (NRPS/PKS) monomers are presented in Table 1 and Fig. 1, respectively. The detection of PGP traits and secondary metabolites (NRPS/PKS) in strains BOVIS40 and JVB5 will help in understanding the mechanisms employed by bacterial endophytes within the endosphere for plant growth through phytohormone production and the secretion of biocontrol compounds. In addition, the expression of PGP genes in the genomes may modulate their multiple functions for enhancing plant growth for improved crop production. Furthermore, the antibiosis potential of endophytic bacteria against phytopathogens has been linked to their ability to produce NRPS/PKS antimicrobial compounds (9). Hence, the potential function of NRPS/PKS reveals novel information about endophytic bacteria as excellent candidates in ensuring sustainable plant health.

**Data availability.** The draft genome sequences for strains BOVIS40 and JVB5 are available in GenBank under the BioProject accession numbers PRJNA706595 and PRJNA706608.
respectively. The Sequence Read Archive (SRA) accession number for strain BOVIS40 is SRR13883846, while that for strain JVB5 is SRR13908543. Genome accession numbers JAGENA00000000.0 and JAGEKL00000000.0 were assigned to strains BOVIS40 and JVB5, respectively.

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All authors contributed substantially and intellectually to this work. O.O.B. designed the research, revised the work critically for important intellectual content, performed quality assurance, and provided funding, project administration, and resources. B.S.A. was involved in data curation, formal analysis, investigation, visualization of data, and writing of the original draft of the manuscript. A.S.A. was involved in data curation, visualization of data, reviewing and thoroughly editing the initial draft, validation, and formal analysis.

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