



Draft Genome Sequence of *Massilia* sp. Strain ONC3, a Novel Bacterial Species of the *Oxalobacteraceae* Family Isolated from Garden Soil

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ABSTRACT From garden soil, we isolated and sequenced *Massilia* sp. strain ONC3, a new member of the *Oxalobacteraceae* within the *Massilia* genus. Sequence analysis showed an assembled genome size of 5,622,601 bp, with a predicted total of 5,104 protein-coding sequences, 3,194 functionally assigned genes, 2 rRNA operons, and 56 tRNAs.

Members of the genus *Massilia* have been isolated from soil, air, and water samples, as well as from the plant rhizosphere and endosphere, and they are typically Gram-negative, rod-shaped aerobes (1, 2). Certain species of *Massilia* can promote plant growth through their ability to solubilize recalcitrant phosphate sources in soils (3) or through their positive impact on the colonization of plants by beneficial root symbionts such as arbuscular mycorrhizal fungi. *Massilia* sp. strain RK4, for example, increases root colonization by arbuscular mycorrhizal fungi and their nutritional benefits to maize plants under salt stress (4). Other *Massilia* species are highly resistant to heavy metals and have been isolated from mines (5).

We isolated a putative new species of *Massilia* from unplanted garden soil (pH 5.2) near Maxton, NC (34.6494, -79.4327). The isolate was an interesting target for whole-genome sequencing, since *Massilia* strains often show plant growth-promoting capabilities. The soil samples were dried for 3 days prior to bacterial isolation, mixed with 1× phosphate-buffered saline, streaked out several times until single colonies were isolated on R2A medium, and cultured at 30°C. Cultures were grown in R2A broth at 30°C for 2 days before genomic DNA (gDNA) was extracted using the Agencourt GenFind v2 kit (Beckman Coulter Life Sciences, Indianapolis, IN) and the protocol for bacterial gDNA extraction. The genomic library was prepared using the Illumina Nextera platform (San Diego, CA), size selected to an average fragment length of 475 bp, and sequenced using Illumina NextSeq paired-end v2 chemistry on v2.5 flow cells at 150 bp per read. The target coverage for the reads was 20×. We used the default settings of BayesHammer and SPAdes 3.13.0 (6) for quality trimming and *de novo* assembly of the 10,528,120 total reads. This resulted in 155 contigs with an N_{50} value of 56,531 bp (contig size range, 942 to 207,795 bp) and a total assembled size of 5,622,601 bp, with a GC content of 63.82%. Assembly quality with 40 reference proteins through BUSCO (7, 8) revealed a measured completeness (40 single-copy BUSCOs) of 100%. Genome assembly and annotation were carried out using the PATRIC 3.5.28 pipeline (9) and confirmed by Galaxy (10) and RAST 2.0 (11), which identified 5,104 protein-coding sequences, 3,194 proteins with functional assignments, 2 rRNA operons, and 56 tRNA genes.

Based on NCBI BLAST (12) searches of the 16S rRNA gene sequences, *Massilia* sp. strain ONC3 was found to have the highest similarities (98% sequence identity) with *Massilia solisilvae* J18 and *Massilia terrae* J11. The Genome-to-Genome Distance Calculator (GGDC) (13) calculates intergenomic distances using three formulas (the sum of all identities found in aligned high-scoring segment pairs divided by the total genome

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length and expressed as a percentage) and was used to compare the ONC3 genome to the 5 most related genomes (*Massilia albidiflava* DSM 17472, *Massilia armeniaca* ZMN-3, *Massilia putida* 6NM-7^T, *Massilia* sp. strain NR 4-1, and *Massilia* sp. strain WG5 [14]). The results for the ONC3 genome relative to the other selected genomes showed a 0% match (or less than 70% of the scientific community threshold) (15, 16). This suggests that *Massilia* sp. ONC3 is a novel species within the *Massilia* genus.

Data availability. This genome has been deposited at DDBJ/EMBL/GenBank under the BioProject number [PRJNA529408](https://www.ncbi.nlm.nih.gov/bioproject/PRJNA529408), BioSample number [SAMN11265771](https://www.ncbi.nlm.nih.gov/biosample/SAMN11265771), accession number [NZ_SPUM000000000](https://www.ncbi.nlm.nih.gov/nuccore/NZ_SPUM000000000), and SRA accession number [SRR9320523](https://www.ncbi.nlm.nih.gov/sra/SRR9320523).

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