

# Genome Sequence of *Methylobacterium* sp. Strain GXF4, a Xylem-Associated Bacterium Isolated from *Vitis vinifera* L. Grapevine

Han Ming Gan,<sup>a</sup> Teong Han Chew,<sup>b</sup> André O. Hudson,<sup>c</sup> and Michael A. Savka<sup>c</sup>

ScienceVision SB, Selangor, Malaysia<sup>a</sup>; Department of Biological Sciences, Faculty of Biosciences and Bioengineering, Universiti Teknologi Malaysia, Johor, Malaysia<sup>b</sup>; and The Thomas H. Gosnell School of Life Sciences, Rochester Institute of Technology, Rochester, New York, USA<sup>c</sup>

***Methylobacterium* sp. strain GXF4 is an isolate from grapevine. Here we present the sequence, assembly, and annotation of its genome, which may shed light on its role as a grapevine xylem inhabitant. To our knowledge, this is the first genome announcement of a plant xylem-associated strain of the genus *Methylobacterium*.**

The soil and insect feeding on plants are important reservoirs of microbes that may be selected and propagated within host plants. Microorganisms that predominantly inhabit xylem systems are called xylem limited and generally rely on insect vectors or wounding for dissemination (1). Important examples of pathogenic xylem-limited bacteria are strains of *Xylella fastidiosa* that are vectored by sap-feeding insects of the Cicadellidae family (1, 9). *X. fastidiosa* causes economically important diseases in a wide variety of plants, including grape, citrus, plum, peach, coffee, and others (1, 9).

This sequenced strain was isolated from xylem fluids of Riesling grapevines grafted onto rootstock Courderc 3309 from a Cornell University vineyard located at the New York State Agricultural Experiment Station, Geneva, NY. It has been previously identified by sequencing of the full-length 16S rRNA gene, is related to bacteria of the genus *Methylobacterium*, and was designated GXF4 (4). Few studies have characterized bacteria from xylem fluids of grapevines. Strain GXF4 was shown to produce bacterial cell-to-cell communication signals of the acyl-homoserine lactone (AHL) family that may influence communication and colonization outcomes within the xylem at the bacterial community level (4). Such interactions may have implications for both biotic and abiotic disease management of vineyards (1, 9). In this study, the genome sequence of *Methylobacterium* sp. strain GXF4, an endophyte from grapevine xylem, was determined to provide novel insight into the molecular principles of xylem endophytes. This work has the potential to enhance the development of novel approaches to improve disease resistance to xylem-limited plant pathogens (1, 9).

The genome sequencing of *Methylobacterium* sp. strain GXF4 was performed with the Illumina Genome Analyzer IIx with 100-bp paired-end reads. The paired-end reads were trimmed and assembled using CLC Genomics Workbench 4.8 (CLC Bio, Aarhus, Denmark). The prediction of open reading frames (ORFs), tRNAs, and rRNAs was performed by using Prodigal 2.50, tRNAscan-SE 1.3, and RNAmmer 1.2 (5, 6, 8), respectively. Subsequent genome annotation was done using Blast2GO 2.5.0 (3). The *de novo* assembly yielded 123 contigs with an accumulated length of 6,116,340 bp (97× coverage) and an average GC content of 69.64%. The contig N50 was 113 kb, and the largest assembled contig was approximately 373 kb. The draft genome contains 5,933 ORFs, 46 tRNAs, and 3 rRNAs.

Similar to *Methylobacterium extorquens* AM1, strain GXF4 contains the complete gene set for methanol oxidation, which is

located at contigs 54 (*mxoQE*), 66 (*mxoMD* and *pqqABCDE*), 78 (*mxoKLDEHB*), and 124 (*mxoFJGIRSAC*) (2). Consistent with its AHL-producing phenotype, two putative AHL synthase genes were identified in the genome of strain GXF4. Interestingly, strain GXF4 contains a gene that codes for β-galactosidase that, to the best of our knowledge, has not been reported in the genomes of the genus *Methylobacterium*. β-Galactosidase catalyzes the hydrolysis of galactan, a common polysaccharide component of the plant cell wall (7). On the basis of the isolation source of strain GXF4, it is reasonable to suggest that this gene endows strain GXF4 with the ability to modify the xylem cell wall structure.

**Nucleotide sequence accession numbers.** This Whole Genome Shotgun project has been deposited at DDBJ/EMBL/GenBank under accession no. [AKFK00000000](https://doi.org/10.1093/nuclemta/ktr000). The version described in this paper is the first version, AKFK01000000.

## ACKNOWLEDGMENTS

The genome sequencing described here was supported by funds from the School of Life Sciences, College of Science, Rochester Institute of Technology, and by a 2012 FEAD grant from the College of Science, Rochester Institute of Technology.

M.A.S. thanks Thomas Burr, Departments of Plant Pathology and Plant-Microbe Biology, Cornell University, New York State Agricultural Experiment Station, Geneva, NY, for his generosity in sharing bacterial strains and for his sponsorship during M.A.S.'s sabbatical. We also thank Yea-Ling Tay and Lian Shien Lee for assistance with this project.

## REFERENCES

1. Bisztray GD, et al. 2011. Grapevine pathogens spreading with propagating plant stock: detection and methods for elimination, p 1–88. In Szabo PV, Shojania J (ed), *Grapevines: varieties, cultivation, and management*. Nova Science Publishers, Hauppauge, NY.
2. Chistoserdova L, Chen SW, Lapidus A, Lidstrom ME. 2003. Methylo-trophy in *Methylobacterium extorquens* AM1 from a genomic point of view. *J. Bacteriol.* **185**:2980–2987.
3. Conesa A, et al. 2005. Blast2GO: a universal tool for annotation, visualization and analysis in functional genomics research. *Bioinformatics* **21**:3674–3676.
4. Hudson AO, Ahmad NH, Van Buren R, Savka MA. 2010. Sugarcane and grapevine endophytic bacteria: isolation, detection of quorum sensing signals and identification by 16S v3 rDNA sequence analysis, p 801–806. *In*

Received 3 July 2012 Accepted 6 July 2012

Address correspondence to Michael A. Savka, [massbi@rit.edu](mailto:massbi@rit.edu).

Copyright © 2012, American Society for Microbiology. All Rights Reserved.

doi:10.1128/JB.01201-12

- Vilas AM (ed), Current research, technology and education topics in applied microbiology and microbial biotechnology, vol 2. Formatex Research Center, Badajoz, Spain,
5. Hyatt D, et al. 2010. Prodigal: prokaryotic gene recognition and translation initiation site identification. *BMC Bioinformatics* **11**:119.
  6. Lagesen K, et al. 2007. RNAmmer: consistent and rapid annotation of ribosomal RNA genes. *Nucleic Acids Res.* **35**:3100–3108.
  7. Lee KJD, Marcus SE, Knox JP. 2011. Cell wall biology: perspectives from cell wall imaging. *Mol. Plant* **4**:212–219.
  8. Lowe TM, Eddy SR. 1997. tRNA-scan-SE: a program for improved detection of transfer RNA genes in genomic sequence. *Nucleic Acids Res.* **25**: 955–964.
  9. Purcell AH, Hopkins DL. 1996. Fastidious xylem-limited bacterial plant pathogens. *Annu. Rev. Phytopathol.* **34**:131–151.