

# Complete Genome Sequence of *Bacillus methylotrophicus* Strain B25, a Potential Plant Growth-Promoting Rhizobacterium

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**The complete genome of *Bacillus methylotrophicus* strain B25, isolated in Switzerland, was sequenced. Its size is 3.85 Mb, and several genes that may contribute to plant growth-promoting activities were identified *in silico*.**

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*Bacillus methylotrophicus* strain B25 (formerly *Bacillus amyloliquefaciens* subsp. *plantarum*) (1) is a Gram-positive bacterium isolated in Switzerland from the inner wood tissues of a decaying *Platanus x acerifolia* tree. Bacterial strains belonging to this clade are plant-associated microorganisms known to have beneficial effects on their hosts, such as growth promotion and health enhancement (2–4). The complete genome sequence of *B. methylotrophicus* strain B25 is reported here and will provide genomic information on the specific properties of this strain.

DNA was extracted with the GenElute bacterial genomic DNA kit (Sigma-Aldrich), according to the manufacturer's instructions. Two libraries were constructed for sequencing at Genoscope. First, genomic DNA was fragmented, and inserts between 300 and 600 nucleotides (nt) were selected to construct a paired-end indexed library. Second, genomic DNA was fragmented, and inserts of around 8 kb were selected to construct a mate-paired indexed library. These libraries were loaded on an Illumina MiSeq sequencing device flowcell and sequenced on paired-ends, at 300 nt in length.

The reads from the paired-end library were merged with a homemade program and were assembled with mate-paired data using Newbler (overlap layout consensus [OLC]) (Roche). To reduce the number of undetermined bases, GapCloser was used (<http://soap.genomics.org.cn/soapdenovo.html>).

The remaining gaps were then covered using PCRs and specific primers for gap edges. The PCR fragments were sequenced by primer walking. The whole-assembly data were integrated into the MicroScope platform (<http://www.genoscope.cns.fr/agc/microscope>) for automatic annotation (5).

The complete genome of *B. methylotrophicus* strain B25 contains a 3,854,619-bp circular chromosome and an 8,138-bp plasmid. The genome contains 3,679 coding sequences (CDSs), with an average length of 934 bp, 21 rRNAs, and 68 tRNAs, and has a G+C content of 46.7%. The annotation predicted gene clusters coding for nonribosomal peptide synthetases (NRPS) and polyketide synthases (PKS). Among all predicted protein-coding genes, the strain B25 genome includes interesting genes involved in bacterium-plant interactions and plant growth promotion ac-

tivities, i.e., the bacillibactin siderophore, an operon required for biofilm formation (*epsA-0*), and a phytate-degrading enzyme (phytase) (6). Gene resistance analysis was conducted with the CARD system (<http://arpcard.mcmaster.ca/>) (7). The B25 genome contains only natural resistance genes conserved at the species level. Gene content comparisons and synteny conservations indicate that the closest neighbors of B25 are *B. methylotrophicus* strains FZB42 (6), IT-45 (6), LFB112 (8), and Y2 (9). Further studies and genome comparisons of *B. methylotrophicus* strain B25 will enable the elucidation of the mechanisms supporting its plant-benefiting properties.

**Nucleotide sequence accession numbers.** This whole-genome shotgun project has been deposited at DDBJ/EMBL/GenBank under the accession numbers [LN999829](https://doi.org/10.1101/050226) and [LN999830](https://doi.org/10.1101/050230).

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## REFERENCES

- Dunlap CA, Kim SJ, Kwon SW, Rooney AP. 2015. Phylogenomic analysis shows that *Bacillus amyloliquefaciens* subsp. *plantarum* is a later heterotypic synonym of *Bacillus methylotrophicus*. *Int J Syst Evol Microbiol* 65: 2104–2109. <http://dx.doi.org/10.1099/ijs.0.000226>.
- Borriss R, Chen XH, Rueckert C, Blom J, Becker A, Baumgarth B, Fan B, Pukall R, Schumann P, Spröer C, Junge H, Vater J, Pühler A, Klenk HP. 2011. Relationship of *Bacillus amyloliquefaciens* clades associated with strains DSM7 and FZB42: a proposal for *Bacillus amyloliquefaciens* subsp. *amyloliquefaciens* sp. nov. and *Bacillus amyloliquefaciens* subsp. *plantarum* subsp. nov. based on complete genome sequence comparisons. *Int J Syst Evol Microbiol* 61:1786–1801. <http://dx.doi.org/10.1099/ijs.0.023267-0>.
- Madhaiyan M, Poonguzhali S, Kwon SW, Sa TM. 2010. *Bacillus methylotrophicus* sp. nov., a methanol-utilizing, plant-growth-promoting bacterium isolated from rice rhizosphere soil. *Int J Syst Evol Microbiol* 60: 2490–2495. <http://dx.doi.org/10.1099/ijs.0.015487-0>.
- Niazi A, Manzoor S, Asari S, Bejai S, Meijer J, Bongcam-Rudloff E. 2014. Genome analysis of *Bacillus amyloliquefaciens* subsp. *plantarum*

- UCMB5113: a rhizobacterium that improves plant growth and stress management. *PLoS One* 9:e104651. <http://dx.doi.org/10.1371/journal.pone.0104651>.
5. Vallenet D, Belda E, Calteau A, Cruveiller S, Engelen S, Lajus A, Le Fèvre F, Longin C, Mornico D, Roche D, Rouy Z, Salvignol G, Scarpelli C, Thil Smith AA, Weiman M, Médigue C. 2013. MicroScope: an integrated microbial resource for the curation and comparative analysis of genomic and metabolic data. *Nucleic Acids Res* 41:D636–D647. <http://dx.doi.org/10.1093/nar/gks1194>.
  6. Chen XH, Koumoutsi A, Scholz R, Eisenreich A, Schneider K, Heinemeyer I, Morgenstern B, Voss B, Hess WR, Reva O, Junge H, Voigt B, Jungblut PR, Vater J, Süßmuth R, Liesegang H, Strittmatter A, Gottschalk G, Borriss R. 2007. Comparative analysis of the complete genome sequence of the plant growth-promoting bacterium *Bacillus amyloliquefaciens* FZB42. *Nat Biotechnol* 25:1007–1014. <http://dx.doi.org/10.1038/nbt1325>.
  7. McArthur AG, Waglechner N, Nizam F, Yan A, Azad MA, Baylay AJ, Bhullar K, Canova MJ, De Pascale G, Ejim L, Kalan L, King AM, Koteva K, Morar M, Mulvey MR, O'Brien JS, Pawlowski AC, Piddock LJ, Spanogiannopoulos P, Sutherland AD, Tang I, Taylor PL, Thaker M, Wang W, Yan M, Yu T, Wright GD. 2013. The comprehensive antibiotic resistance database. *Antimicrob Agents Chemother* 57:3348–3357. <http://dx.doi.org/10.1128/AAC.00419-13>.
  8. Cai J, Liu F, Liao X, Zhang R. 2014. Complete genome sequence of *Bacillus amyloliquefaciens* LFB112 isolated from Chinese herbs, a strain of a broad inhibitory spectrum against domestic animal pathogens. *J Biotechnol* 175: 63–64. <http://dx.doi.org/10.1016/j.jbiotec.2014.01.013>.
  9. Hao K, He P, Blom J, Rueckert C, Mao Z, Wu Y, He Y, Borriss R. 2012. The genome of plant growth-promoting *Bacillus amyloliquefaciens* subsp. *plantarum* strain YAU B9601-Y2 contains a gene cluster for mersacidin synthesis. *J Bacteriol* 194:3264–3265. <http://dx.doi.org/10.1128/JB.00545-12>.