

# Draft Genome Sequence of *Frankia* sp. Strain BMG5.12, a Nitrogen-Fixing Actinobacterium Isolated from Tunisian Soils

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**Members of the actinomycete genus *Frankia* form a nitrogen-fixing symbiosis with 8 different families of actinorhizal plants. We report a draft genome sequence for *Frankia* sp. strain BMG5.12, a nitrogen-fixing actinobacterium isolated from Tunisian soils with the ability to infect *Elaeagnus angustifolia* and *Myrica gale*.**

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Among the *Actinobacteria*, the genus *Frankia* is well known for its facultative lifestyle as a plant symbiont of dicotyledonous plants and as a free-living soil dweller (1–3). The symbiosis allows actinorhizal plants to colonize harsh environmental terrains under diverse ecological conditions. Based on several criteria, including the 16S rRNA gene (4), *glnII* (5, 6), *gyrB* (6), and a 16S-23S rRNA intertranscribed spacer region (7), phylogenetic analysis has identified four distinct clusters among the *Frankia* strains. Genomes for representatives from each of these clusters have been sequenced (8–12) and have provided vital baseline information for genomic approaches toward understanding these novel bacteria.

Members of cluster III *Frankia* strains are considered broad-host-range symbionts and are known to associate with five plant families (*Betulaceae*, *Myricaceae*, *Elaeagnaceae*, *Rhamnaceae*, and *Casuarinaceae*). *Frankia* sp. strain BMG5.12 was chosen for sequencing as another cluster III representative with broad-host-range properties. Strain BMG 5.12 was isolated from Tunisian soils that were devoid of *Elaeagnus* plants and it was found to infect these host plants (13). Strain BMG5.12 was sequenced to find information about the potential ecological roles of the *Frankia* strains and their interactions with actinorhizal plants. As a representative from a harsh dry climate, the *Frankia* sp. BMG5.12 genome may provide insight on its ability to adapt to these arid hot soils that have high salinity.

The draft genome sequence of *Frankia* sp. BMG5.12 was generated at the Department of Energy (DOE) Joint Genome Institute (JGI) using Illumina technology (14). An Illumina standard shotgun library was constructed and sequenced using the Illumina HiSeq 2000 platform, which generated 9,945,424 reads totaling 1,491.8 Mbp. All techniques for DNA isolation, library construction, and sequencing were performed according to JGI standards and protocols (<http://www.jgi.doe.gov>). The Illumina sequence

data were assembled using Velvet (version 1.1.04) (14) and Allpaths-LG (version r41043) (15). The final draft assembly contained 139 contigs in 139 scaffolds. The total size of the genome is 7.6 Mbp, and the final assembly is based on 932.3 Mbp of Illumina data, which provide an average 122.9× coverage of the genome.

The draft genome sequence of *Frankia* sp. BMG5.12 was resolved to 139 scaffolds consisting of 7,589,313 bp, with a G+C content of 71.67%, 6,253 candidate protein-coding genes, 51 tRNA genes, and 2 rRNA regions.

**Nucleotide sequence accession numbers.** The *Frankia* sp. BMG5.12 genome sequence has been deposited at DDBJ/EMBL/GenBank under the accession no. [ARFH000000000](https://www.ncbi.nlm.nih.gov/nuccore/ARFH000000000). The version described in this paper is the first version, accession no. [ARFH010000000](https://www.ncbi.nlm.nih.gov/nuccore/ARFH010000000).

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## REFERENCES

1. Benson DR, Silvester WB. 1993. Biology of *Frankia* strains, actinomycete symbionts of actinorhizal plants. *Microbiol. Rev.* 57:293–319.
2. Schwencke J, Caru M. 2001. Advances in actinorhizal symbiosis: host plant-*Frankia* interactions, biology, and applications in arid land reclamation. A review. *Arid Land Res. Manag.* 15:285–327.
3. Chaia EE, Wall LG, Huss-Danell K. 2010. Life in soil by actinorhizal root nodule endophyte *Frankia*. A review. *Symbiosis* 51:201–226.

4. Normand P, Orso S, Cournoyer B, Jeannin P, Chapelon C, Dawson J, Evtushenko L, Misra AK. 1996. Molecular phylogeny of the genus *Frankia* and related genera and emendation of the family *Frankiaceae*. *Int. J. Syst. Bacteriol.* **46**:1–9.
5. Cournoyer B, Lavire C. 1999. Analysis of *Frankia* evolution radiation using *glnII* sequences. *FEMS Microbiol. Lett.* **117**:29–34.
6. Nouioui I, Ghodhbane-Gtari F, Beauchemin NJ, Tisa LS, Gtari M. 2011. Phylogeny of members of the *Frankia* genus based on *gyrB*, *nifH* and *glnII* sequences. *Antonie Van Leeuwenhoek* **100**:579–587.
7. Ghodhbane-Gtari F, Nouioui I, Chair M, Boudabous A, Gtari M. 2010. 16S-23S rRNA intergenic spacer region variability in the genus *Frankia*. *Microb. Ecol.* **60**:487–495.
8. Normand P, Lapierre P, Tisa LS, Gogarten JP, Alloisio N, Bagnarol E, Bassi CA, Berry AM, Bickhart DM, Choisine N, Couloux A, Cournoyer B, Cruveiller S, Daubin V, Demange N, Francino MP, Goltsman E, Huang Y, Kopp OR, Labarre L, Lapidus A, Lavire C, Marechal J, Martinez M, Mastrorunzio JE, Mullin BC, Niemann J, Pujic P, Rawnsley T, Rouy Z, Schenowitz C, Sellstedt A, Tavares F, Tomkins JP, Vallet D, Valverde C, Wall LG, Wang Y, Medigue C, Benson DR. 2007. Genome characteristics of facultatively symbiotic *Frankia* sp. strains reflect host range and host plant biogeography. *Genome Res.* **17**:7–15.
9. Normand P, Queiroux C, Tisa LS, Benson DR, Rouy Z, Cruveiller S, Medigue C. 2007. Exploring the genomes of *Frankia*. *Physiol. Plant.* **130**:331–343.
10. Persson T, Benson DR, Normand P, Vanden Heuvel B, Pujic P, Chertkov O, Teshima H, Bruce DC, Detter C, Tapia R, Han S, Han J, Woyke T, Pitluck S, Pennacchio L, Nolan M, Ivanova N, Pati A, Land ML, Pawlowski K, Berry AM. 2011. Genome sequence of “*Candidatus Frankia datisciae*” Dg1, the uncultured microsymbiont from nitrogen-fixing root nodules of the dicot *Datisca glomerata*. *J. Bacteriol.* **193**:7017–7018.
11. Ghodhbane-Gtari F, Beauchemin N, Bruce D, Chain P, Chen A, Walston Davenport K, Deshpande S, Detter C, Furnholm T, Goodwin L, Gtari M, Han C, Han J, Huntemann M, Ivanova N, Kyrpides N, Land ML, Markowitz V, Mavrommatis K, Nolan M, Nouioui I, Pagani I, Pati A, Pitluck S, Santos CL, Sen A, Sur S, Szeto E, Tavares F, Teshima H, Thakur S, Wall L, Woyke T, Tisa LS. 2013. Draft genome sequence of *Frankia* sp. strain CN3, an atypical, noninfective (Nod<sup>-</sup>) ineffective (Fix<sup>-</sup>) isolate from *Coriaria nepalensis*. *Genome Announc.* **1**(2):e00085-13. doi:[10.1128/genomeA.00085-13](https://doi.org/10.1128/genomeA.00085-13).
12. Sen A, Beauchemin N, Bruce D, Chain P, Chen A, Walston Davenport K, Deshpande S, Detter C, Furnholm T, Ghodhbane-Gtari F, Goodwin L, Gtari M, Han C, Han J, Huntemann M, Ivanova N, Kyrpides N, Land ML, Markowitz V, Mavrommatis K, Nolan M, Nouioui I, Pagani I, Pati A, Pitluck S, Santos CL, Sur S, Szeto E, Tavares F, Teshima H, Thakur S, Wall L, Woyke T, Wishart J, Tisa LS. 2013. Draft genome sequence of *Frankia* sp. strain QA3, a nitrogen-fixing actinobacterium isolated from the root nodule of *Alnus nitida*. *Genome Announc.* **1**(2):e00103-13. doi:[10.1128/genomeA.00103-13](https://doi.org/10.1128/genomeA.00103-13).
13. Gtari M, Brusetti L, Skander G, Mora D, Boudabous A, Daffonchio D. 2004. Isolation of *Elaeagnus*-compatible *Frankia* from soils collected in Tunisia. *FEMS Microbiol. Lett.* **234**:349–355.
14. Bennett S. 2004. Solexa Ltd. *Pharmacogenomics* **5**:433–438.
15. Zerbino DR, Birney E. 2008. Velvet: algorithms for *de novo* short read assembly using de Bruijn graphs. *Genome Res.* **18**:821–829.