

1-14-2016

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Recommended Citation

Oshone, R. S. Hurst IV, F. Abebe-Akele, S. Simpson, K. Morris, W. K. Thomas, and L. S. Tisa. 2016. Permanent draft genome sequences for two variants of Frankia sp. strain CpI1, the first Frankia strain isolated from the root nodule of *Comptonia peregrina*. *Genome Announc.* 4(1):e01588-15 (doi: 10.1128/genomeA.01588-15).

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Permanent Draft Genome Sequences for Two Variants of *Frankia* sp. Strain CpI1, the First *Frankia* Strain Isolated from Root Nodules of *Comptonia peregrina*

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***Frankia* strains CpI1-S and CpI1-P are members of *Frankia* lineage Ia that are able to reinfect plants of the *Betulaceae* and *Myricaceae* families. Here, we report two 7.6-Mbp draft genome sequences with 6,396 and 6,373 candidate protein-coding genes for CpI1-S and CpI1-P, respectively.**

Received 17 November 2015 Accepted 18 November 2015 Published 14 January 2016

Citation Oshone R, Hurst SG, IV, Abebe-Akele F, Simpson S, Morris K, Thomas WK, Tisa LS. 2016. Permanent draft genome sequences for two variants of *Frankia* sp. strain CpI1, the first *Frankia* strain isolated from root nodules of *Comptonia peregrina*. *Genome Announc* 4(1):e01588-15. doi:10.1128/genomeA.01588-15.

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Actinorhizal symbiosis is the association between an actinobacterium from the genus *Frankia* and a wide variety of dicotyledonous plants representing 8 different families of angiosperms, resulting in the formation of root nodule structure (1–3). The symbiosis allows these ecologically important pioneer plants to colonize harsh environments that are found worldwide (4). Molecular phylogenetic approaches have identified four major *Frankia* lineages (5–8). Cluster I contains two subclusters: one subcluster (cluster Ia) represents *Frankia* strains with the ability to infect a wider range of host plants, including members of the *Betulaceae* and *Myricaceae* families, and the other subcluster (cluster Ib) contains strains limited to *Casuarina* and *Allocasuarina* host plants. Members of cluster II infect host plants of the subfamily *Dryadoideae* (*Rosaceae*), *Coriariaceae*, and *Datisceae*, and of the genus *Ceanothus* (*Rhamnaceae*). The members of cluster III are the most promiscuous and infect *Elaeagnaceae*, *Rhamnaceae*, *Myricaceae*, *Gymnostoma* (*Casuarinaceae*), and occasionally *Alnus* species. The fourth *Frankia* lineage consists of the atypical strains, which are unable to reinfect actinorhizal host plants or form ineffective root nodule structures that are unable to fix nitrogen. The genomes for representatives from each cluster have been sequenced (9–24).

In 1978, the first *Frankia* isolate, CpI1, was obtained from root nodules of *Comptonia peregrina* and fulfilled Koch's postulates (25). Following this initial success, many other strains from different host plants have been isolated, including the recent isolation of a member of cluster II (20). Two variants of *Frankia* sp. strain CpI1 were identified that had different carbon source requirements for growth. The *Frankia* sp. strain CpI1 succinate variant (CpI1-S) will use succinate and other dicarboxylic acids for growth, while the *Frankia* sp. strain CpI1 propionate variant (CpI1-P) will not (26). These two variants also show differences in their heavy metal tolerances and antibiotic resistance patterns (27, 28). *Frankia* sp. strains CpI1-S and CpI1-P were chosen for sequencing to provide more information on the differences between these two variants.

The draft genome sequences of *Frankia* sp. strains CpI1-S and

CpI1-P were generated at the Hubbard Genome Center (University of New Hampshire, Durham, NH) using Illumina technology techniques (29). A standard Illumina shotgun library was constructed and sequenced using the Illumina HiSeq 2000 platform, which generated 10,867,000 reads (260-bp insert size) totaling 1,628.6 Mbp for CpI1-S and 20,978,588 reads (260-bp insert size) totaling 3,146.7 Mbp for CpI1-P. The Illumina sequence data were assembled using CLC Genomics Workbench (version 8.0.1) and AllPaths-LG (version r41043) (30). The final draft assembly for *Frankia* sp. CpI1-S consisted of 153 contigs containing a total sequence of 7,614,630 bp, with an N_{50} contig size of 99.4 kb and $206.3\times$ coverage of the genome. For *Frankia* sp. CpI1-P, the final draft assembly consisted of 181 contigs containing 7,593,325 bp, with an N_{50} contig size of 98.1 kb and $287\times$ coverage of the genome.

The assembled *Frankia* sp. CpI1-S and CpI1-P genomes were annotated via the Integrated Microbial Genomes (IMG) platform developed by the Joint Genome Institute, Walnut Creek, CA, USA (31, 32), and resulted in 6,396 and 6,373 candidate protein-coding genes for strains CpI1-S and CpI1-P, respectively.

Nucleotide sequence accession numbers. This whole-genome shotgun project has been deposited at DDBJ/EMBL/GenBank under the accession numbers JYFN00000000 (CpI1-S) and LJXX00000000 (CpI1-P). The versions described in this paper are versions JYFN01000000 and LJXX01000000, respectively.

ACKNOWLEDGMENTS

Partial funding was provided by the New Hampshire Agricultural Experiment Station. This is Scientific Contribution no. 2641. This work was supported by the USDA National Institute of Food and Agriculture Hatch 022821 (to L.S.T.), Agriculture and Food Research Initiative grant 2015-67014-22849 from the USDA National Institute of Food and Agriculture (to L.S.T.), and the College of Life Science and Agriculture at the University of New Hampshire-Durham. Sequencing was performed on an Illumina HiSeq 2500 purchased with an NSF MRI grant DBI-1229361 to W.K.T.

FUNDING INFORMATION

The National Science Foundation (NSF) provided funding to W. Kelley Thomas under grant number DBI-122936. USDA | The National Institute of Food and Agriculture (NIFA) provided funding to Louis S. Tisa under grant numbers Hatch 02281 and 2015-67014-22849.

REFERENCES

- Schwencke J, Carú 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. <http://dx.doi.org/10.1080/153249801753127615>.
- Chaia EE, Wall LG, Huss-Danell K. 2010. Life in soil by the actinorhizal root nodule endophyte *Frankia*. A review. *Symbiosis* 51:201–226. <http://dx.doi.org/10.1007/s13199-010-0086-y>.
- Normand P, Benson DR, Berry AM, Tisa LS. 2014. Family *Frankiaceae*, p 339–356. In Rosenberg E, DeLong EF, Lory S, Stackebrandt E, Thompson F (ed), *The prokaryotes—Actinobacteria*, 4th ed. Springer-Verlag, Berlin, Germany.
- Benson DR, Dawson JO. 2007. Recent advances in the biogeography and genecology of symbiotic *Frankia* and its host plants. *Physiol Plant* 130: 318–330. <http://dx.doi.org/10.1111/j.1399-3054.2007.00934.x>.
- 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. <http://dx.doi.org/10.1099/00207713-46-1-1>.
- Cournoyer B, Lavire C. 1999. Analysis of *Frankia* evolution radiation using *glnII* sequences. *FEMS Microbiol Lett* 117:29–34.
- 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. <http://dx.doi.org/10.1007/s10482-011-9613-y>.
- 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.
- Normand P, Lapiere 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. <http://dx.doi.org/10.1101/gr.5798407>.
- Normand P, Queiroz C, Tisa LS, Benson DR, Rouy Z, Cruveiller S, Médigue C. 2007. Exploring the genomes of *Frankia*. *Physiol Plant* 130: 331–343. <http://dx.doi.org/10.1111/j.1399-3054.2007.00918.x>.
- 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 I, Pitluck S, Pennacchio L, Nolan M, Ivanova N, Pati A, Land ML, Pawlowski K, Berry AM. 2011. Genome sequence of “*Candidatus* *Frankia datiscæ*” Dg1, the uncultured Microsymbiont from nitrogen-fixing root nodules of the dicot *Datisca glomerata*. *J Bacteriol* 193:7017–7018. <http://dx.doi.org/10.1128/JB.06208-11>.
- 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 LG, Woyke T, Tisa LS. 2013. Draft genome sequence of *Frankia* sp. strain CN3, an atypical, non-infective (Nod-) ineffective (Fix-) isolate from *Coriaria nepalensis*. *Genome Announc* 1(2):00085-13. <http://dx.doi.org/10.1128/genomeA.00085-13>.
- Sen A, 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, Sur S, Szeto E, Tavares F, Teshima H, Thakur S, Wall LG, Woyke T, 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. <http://dx.doi.org/10.1128/genomeA.00103-13>.
- Nouioui I, Beauchemin N, Cantor MN, Chen A, Detter JC, Furnholm T, Ghodhbane-Gtari F, Goodwin L, Gtari M, Han C, Han J, Huntemann M, Hua SX, Ivanova N, Kyrpides N, Markowitz V, Mavrommatis K, Mikhailova N, Nordberg HP, Ovchinnikova G, Pagani I, Pati A, Sen A, Sur S, Szeto E, Thakur S, Wall L, Wei C-L, Woyke T, Tisa LS. 2013. Draft genome sequence of *Frankia* sp. strain BMG5.12, a nitrogen-fixing actinobacterium isolated from Tunisian soils. *Genome Announc* 1: <http://dx.doi.org/10.1128/genomeA.00468-13>.
- Wall LG, Beauchemin N, Cantor MN, Chaia E, Chen A, Detter JC, Furnholm T, Ghodhbane-Gtari F, Goodwin L, Gtari M, Han C, Han J, Huntemann M, Hua SX, Ivanova N, Kyrpides N, Markowitz V, Mavrommatis K, Mikhailova N, Nordberg HP, Nouioui I, Ovchinnikova G, Pagani I, Pati A, Sen A, Sur S, Szeto E, Thakur S, Wei C-L, Woyke T, Tisa LS. 2013. Draft genome sequence of *Frankia* sp. strain BCU110501, a nitrogen-fixing actinobacterium isolated from nodules of *Discaria trinevis*. *Genome Announc* 1(4): <http://dx.doi.org/10.1128/genomeA.00503-13>.
- Mansour SR, Oshone R, Hurst SG, IV, Morris K, Thomas WK, Tisa LS. 2014. Draft genome sequence of *Frankia* sp. strain Ccl6, a salt-tolerant nitrogen-fixing actinobacterium isolated from the root nodule of *Casuarina cunningghamiana*. *Genome Announc* 2(1):e01205-13. <http://dx.doi.org/10.1128/genomeA.01205-13>.
- Hurst SG, IV, Oshone R, Ghodhbane-Gtari F, Morris K, Abebe-Akele F, Thomas WK, Ktari A, Salem K, Mansour S, Gtari M, Tisa LS. 2014. Draft genome sequence of *Frankia* sp. strain Thr, a nitrogen-fixing actinobacterium isolated from the root nodules of *Casuarina cunningghamiana* grown in Egypt. *Genome Announc* 2(3): <http://dx.doi.org/10.1128/genomeA.00493-14>.
- Ghodhbane-Gtari F, Hurst SG, IV, Oshone R, Morris K, Abebe-Akele F, Thomas WK, Ktari A, Salem K, Gtari M, Tisa LS. 2014. Draft genome sequence of *Frankia* sp. strain BMG5.23, a salt-tolerant nitrogen-fixing actinobacterium isolated from the root nodules of *Casuarina glauca* grown in Tunisia. *Genome Announc* 2(3):e00520-14. <http://dx.doi.org/10.1128/genomeA.00520-14>.
- Tisa LS, Beauchemin N, Gtari M, Sen A, Wall LG. 2013. What stories can the *Frankia* genomes start to tell us? *J Biosci* 38:719–726. <http://dx.doi.org/10.1007/s12038-013-9364-1>.
- Gtari M, Ghodhbane-Gtari F, Nouioui I, Ktari A, Hezbri K, Mimiouni W, Bissli I, Ayari A, Yamanaka T, Normand P, Tisa LS, Boudabous A. 2015. Cultivating the uncultured: growing the recalcitrant cluster-2 *Frankia* strains. *Sci Rep* 5:13112. <http://dx.doi.org/10.1038/srep13112>.
- Tisa LS, Beauchemin N, Cantor MN, Furnholm T, Ghodhbane-Gtari F, Goodwin LL, Copeland A, Gtari M, Huntemann M, Ivanova N, Kyrpides N, Markowitz V, Mavrommatis K, Mikhailova N, Nouioui I, Oshone R, Ovchinnikova G, Pagani I, Palaniappan K, Pati A, Sen A, Shapiro N, Szeto E, Wall LG, Wishart J, Woyke T. 2015. Draft genome sequence of *Frankia* sp. strain DC12, an atypical, non-infective, ineffective isolate from *Datisca cannabina*. *Genome Announc* 3(4):e00889-15. <http://dx.doi.org/10.1128/genomeA.00889-15>.
- Swanson E, Oshone R, Simpson S, Morris K, Abebe-Akele F, Thomas WK, Tisa LS. 2015. Draft genome sequence of *Frankia* sp. strain ACN1^{ag}, a nitrogen-fixing actinobacterium isolated from the root nodule of *Alnus glutinosa*. *Genome Announc* 3:e01483-15. <http://dx.doi.org/10.1128/genomeA.01483-15>.
- Swanson E, Oshone R, Simpson S, Morris K, Abebe-Akele F, Thomas WK, Tisa LS. 2015. Permanent draft genome sequence of *Frankia* sp. strain avci1, a nitrogen-fixing actinobacterium isolated from the root nodule of *Alnus viridis* ssp. *crispa* grown in Canada. *Genome Announc* 3: <http://dx.doi.org/10.1128/genomeA.01511-15>.
- Pujic P, Bolotin A, Fournier P, Sorokin A, Lapidus A, Richau KH, Briolay J, Mebarki F, Normand P, Sellstedt A. 2015. Genome sequence of the atypical symbiotic *Frankia* R43 strain, a nitrogen-fixing and hydrogen-evolving actinobacterium. *Genome Announc* 3:e01387-15. <http://dx.doi.org/10.1128/genomeA.01387-15>.
- Callaham D, Deltredici P, Torrey JG. 1978. Isolation and cultivation *in vitro* of the actinomycete causing root nodulation in *Comptonia*. *Science* 199:899–902. <http://dx.doi.org/10.1126/science.199.4331.899>.
- Tisa L, McBride M, Ensign JC. 1983. Studies of growth and morphology of *Frankia* strains EAN1^{pec}, Eu1^c, Cp1¹ and ACN1^{ag}. *Can J Bot* 61: 2768–2773. <http://dx.doi.org/10.1139/b83-306>.
- Tisa LS, Chval MS, Krumholz GD, Richards J. 1999. Antibiotic resistance patterns of *Frankia* strains. *Can J Bot* 77:1257–1260.

28. Richards JW, Krumholz GD, Chval MS, Tisa LS. 2002. Heavy metal resistance patterns of *Frankia* strains. *Appl Environ Microbiol* **68**: 923–927.
29. Bennett S. 2004. Solexa Ltd. *Pharmacogenomics* **5**:433–438. <http://dx.doi.org/10.1517/14622416.5.4.433>.
30. Gnerre S, MacCallum I, Przybylski D, Ribeiro FJ, Burton JN, Walker BJ, Sharpe T, Hall G, Shea TP, Sykes S, Berlin AM, Aird D, Costello M, Daza R, Williams L, Nicol R, Gnirke A, Nusbaum C, Lander ES, Jaffe DB. 2011. High-quality draft assemblies of mammalian genomes from massively parallel sequence data. *Proc Natl Acad Sci USA* **108**: 1513–1518. <http://dx.doi.org/10.1073/pnas.1017351108>.
31. Markowitz VM, Korzeniewski F, Palaniappan K, Szeto E, Werner G, Padki A, Zhao XL, Dubchak I, Hugenholtz P, Anderson I, Lykidis A, Mavromatis K, Ivanova N, Kyrpides NC. 2006. The integrated microbial genomes (IMG) system. *Nucleic Acids Res* **34**:D344–D348. <http://dx.doi.org/10.1093/nar/gkj024>.
32. Markowitz VM, Mavromatis K, Ivanova NN, Chen IA, Chu K, Kyrpides NC. 2009. IMG er: a system for microbial genome annotation expert review and curation. *Bioinformatics* **25**:2271–2278. <http://dx.doi.org/10.1093/bioinformatics/btp393>.