





Draft Genome Sequence of *Curtobacterium* sp. Strain UCD-KPL2560 (Phylum *Actinobacteria*)

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Here, we present the draft genome sequence of the actinobacterium *Curtobacterium* sp. strain UCD-KPL2560, which was isolated from the running surface of an indoor track field house in Medford, MA, USA (42.409716°N, -71.115169°W). The genome assembly contains 3,480,487 bp in 156 contigs.

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embers of the genus *Curtobacterium*, which was accepted as a genus in 1972 and refined in 1986 (1, 2), have been previously isolated from soil (3, 4), plants (5), cheese-processing equipment (6), and residential carpet (7). *Curtobacterium* spp. are characterized as Gram-positive aerobic bacilli that frequently demonstrate yellow-orange pigment when grown *in vitro* (2). *Curtobacterium* spp. are not common pathogens of humans; however, *Curtobacterium* spp. are recognized plant pathogens (8), and rare plant-to-human infections, as well as occasional human host isolation, are reported (9, 10).

We isolated Curtobacterium sp. strain UCD-KPL2560 from the flooring of an indoor track facility in Medford, MA, on 15 October 2015 as part of a project to produce reference genomes for microorganisms residing in the built environment. A nylon-flocked swab (Copan) dipped in sterile buffer (0.1 M NaCl and 0.1% Tween) was rubbed over the track surface starting block area (5 cm²) for 1 min, inoculated onto brain heart infusion agar containing fosfomycin (20 µg/ml), and incubated aerobically at 37°C for five days. A small round orange colony was selected from the original isolation plate and subcultured for purity. For 16S rRNA identification, an initial DNA extraction was performed with the DNeasy purification kit (Qiagen). A putative genus name was assigned to the isolate following PCR amplification (primers 27F and 1492R) and subsequent Sanger sequencing of the 16S rRNA gene. Genomic DNA for whole-genome sequencing was extracted using the MasterPure complete DNA/RNA purification kit (Epi-

Illumina paired-end libraries were generated using a Nextera DNA sample prep kit (Illumina). We selected 600- to 900-bp fragments using a Pippin Prep (Sage Science). Resultant libraries were sequenced on an Illumina MiSeq, with a read length of 300 bp. This produced a total of 3,920,338 paired-end reads. Quality trimming and error correction of the reads resulted in 3,506,299 high-quality reads using the A5-MiSeq assembly pipeline (version 05/22/2015) (11, 12). The resulting assembly contained 156 scaffolds (minimum, 598 bp; maximum, 215,854 bp; N_{50} , 36,322 bp). The

final assembly contained 3,480,487 bp, with a G+C content of 72% and a coverage estimate of 114% (EC value). Genome completeness was assessed using PhyloSift and CheckM (13, 14); all 37 PhyloSift marker genes were present, with 36 in single copy, and the ribosomal subunit L14 present in four copies. CheckM gave a 99% completeness estimation.

Annotation was performed using the RAST server (default settings on 14 April 2016) (15). *Curtobacterium* sp. strain UCD-KPL2560 contains 3,201 predicted coding sequences (CDSs), four predicted rRNAS, and 53 predicted tRNAs. It has 12 type IV pilus genes, 34 motility genes, and four auxin biosynthesis genes. At least two partial phages are predicted, and no clustered regularly interspaced short palindromic repeat (CRISPR) systems were identified (15–17).

We attempted to assign a putative species designation to *Curtobacterium* sp. UCD-KPL2560 using PhyloPhlAn and by generating a 16S-rRNA-gene-based phylogeny of *Curtobacterium* species curated by the Ribosomal Database Project (18, 19). However, neither yielded an assignment. PhyloPhlAn generated "*Clavibacter michiganensis*" taxonomic assignments for 16 of 17 *Curtobacterium* spp., with available NCBI sequencing data (one reclassified as *Rothia*). Additionally, *Curtobacterium* sp. UCD-KPL2560 did not fall within a well-supported clade of a single species within the genus *Curtobacterium* using 16S rRNA alone.

Accession number(s). This whole-genome shotgun project has been deposited at DDBJ/ENA/GenBank under the accession no. MCIG00000000. The version described in this paper is version MCIG01000000.

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REFERENCES

- Yamada K, Komagata K. 1972. Taxonomic studies on coryneform bacteria V. Classification of coryneform bacteria. J Gen Appl Microbiol 18: 417–431. http://dx.doi.org/10.2323/jgam.18.417.
- Komagta K, Suzuki K. 1986. Genus Curtobacterium, p 1313–1317. In Bergey's manual of systematic bacteriology. Williams & Wilkins, Philadelphia, PA.
- Kim MK, Kim YJ, Kim HB, Kim SY, Yi TH, Yang DC. 2008. Curtobacterium ginsengisoli sp. nov., isolated from soil of a ginseng field. Int J Syst Evol Microbiol 58:2393–2397. http://dx.doi.org/10.1099/ijs.0.65591-0.
- Mariita RM, Bhatnagar S, Hanselmann K, Hossain MJ, Korlach J, Boitano M, Roberts RJ, Liles MR, Moss AG, Leadbetter JR, Newman DK, Dawson SC. 2015. Complete genome sequence of *Curtobacterium* sp. strain MR_MD2014, isolated from topsoil in Woods Hole, Massachusetts. Genome Announc 3(6):e01504-15. http://dx.doi.org/10.1128/ genomeA.01504-15.
- 5. Bulgari D, Minio A, Casati P, Quaglino F, Delledonne M, Bianco PA. 2014. *Curtobacterium* sp. genome sequencing underlines plant growth promotion-related traits. Genome Announc 2(4):e00592-14. http://dx.doi.org/10.1128/genomeA.00592-14.
- Didienne R, Defargues C, Callon C, Meylheuc T, Hulin S, Montel MC. 2012. Characteristics of microbial biofilm on wooden vats ("gerles") in PDO Salers cheese. Int J Food Microbiol 156:91–101. http://dx.doi.org/ 10.1016/j.ijfoodmicro.2012.03.007.
- 7. Flanagan JC, Lang JM, Darling AE, Eisen JA, Coil DA. 2013. Draft genome sequence of *Curtobacterium flaccumfaciens* strain UCD-AKU (phylum *Actinobacteria*). Genome Announc 1(3):e00244-13. http://dx.doi.org/10.1128/genomeA.00244-13.
- Agarkova IV, Lambrecht PA, Vidaver AK, Harveson RM. 2012. Genetic diversity among Curtobacterium flaccumfaciens pv. flaccumfaciens popu-

- lations in the American high plains. Can J Microbiol 58:788–801. http://dx.doi.org/10.1139/w2012-052.
- Francis MJ, Doherty RR, Patel M, Hamblin JF, Ojaimi S, Korman TM. 2011. Curtobacterium flaccumfaciens septic arthritis following puncture with a Coxspur hawthorn thorn. J Clin Microbiol 49:2759–2760. http:// dx.doi.org/10.1128/JCM.00340-11.
- Funke G, Aravena-Roman M, Frodl R. 2005. First description of *Curto-bacterium* spp. isolated from human clinical specimens. J Clin Microbiol 43:1032–1036. http://dx.doi.org/10.1128/JCM.43.3.1032-1036.2005.
- 11. Tritt A, Eisen JA, Facciotti MT, Darling AE. 2012. An integrated pipeline for *de novo* assembly of microbial genomes. PLoS One 7:e42304. http://dx.doi.org/10.1371/journal.pone.0042304.
- 12. Coil D, Jospin G, Darling AE. 2015. A5-miseq: an updated pipeline to assemble microbial genomes from Illumina MiSeq data. Bioinformatics 31:587–589. http://dx.doi.org/10.1093/bioinformatics/btu661.
- 13. Darling AE, Jospin G, Lowe E, Matsen FA, Bik HM, Eisen JA. 2014. PhyloSift: phylogenetic analysis of genomes and metagenomes. PeerJ 2:e243. http://dx.doi.org/10.7717/peerj.243.
- Parks DH, Imelfort M, Skennerton CT, Hugenholtz P, Tyson GW. 2015. CheckM: assessing the quality of microbial genomes recovered from isolates, single cells, and metagenomes Genome Res 25:1043–1055. http:// dx.doi.org/10.1101/gr.186072.114.
- 15. Overbeek R, Olson R, Pusch GD, Olsen GJ, Davis JJ, Disz T, Edwards RA, Gerdes S, Parrello B, Shukla M, Vonstein V, Wattam AR, Xia F, Stevens R. 2014. The SEED and the rapid annotation of microbial genomes using subsystems technology (RAST). Nucleic Acids Res 42: D206–D214. http://dx.doi.org/10.1093/nar/gkt1226.
- Arndt D, Grant JR, Marcu A, Sajed T, Pon A, Liang Y, Wishart DS. 2016. PHASTER: a better, faster version of the PHAST phage search tool. Nucleic Acids Res 44:W16–W21. http://dx.doi.org/10.1093/nar/gkw387.
- 17. **Grissa I, Vergnaud G, Pourcel C.** 2007. CRISPRFinder: a Web tool to identify clustered regularly interspaced short palindromic repeats. Nucleic Acids Res 35:W52–W57. http://dx.doi.org/10.1093/nar/gkm360.
- Segata N, Börnigen D, Morgan XC, Huttenhower C. 2013. PhyloPhlAn is a new method for improved phylogenetic and taxonomic placement of microbes. Nat Commun 4:2304. http://dx.doi.org/10.1038/ncomms3304.
- Cole JR, Wang Q, Cardenas E, Fish J, Chai B, Farris RJ, Kulam-Syed-Mohideen AS, McGarrell DM, Marsh T, Garrity GM, Tiedje JM. 2009. The Ribosomal Database Project: improved alignments and new tools for rRNA analysis. Nucleic Acids Res 37:D141–D145. http://dx.doi.org/10.1093/nar/gkn879.