



Draft Genome Sequence of *Clostridium* sp. Strain FP1, with Similarity to *Clostridium tagluense*, Isolated from Spoiled Lamb

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ABSTRACT *Clostridium* sp. strain FP1 was isolated from vacuum-packaged refrigerated spoiled lamb, and this article describes its 5.4-Mb draft genome sequence. The FP1 genome was sequenced to facilitate source tracking and attribution studies, adding to our understanding of the role of *Clostridium* species in premature spoilage of red meats.

Clostridium sp. strain FP1 is a Gram-positive, spore-forming, and slow-growing psychrotrophic anaerobe that was originally isolated from vacuum-packaged refrigerated spoiled lamb at AgResearch Ltd. (Palmerston North, New Zealand). Blown pack spoilage (BPS) is a major issue for the meat industry, and the etiological agents of BPS are numerous members of the *Clostridium* species, including *Clostridium estertheticum*. FP1 was positive by the *C. estertheticum*-like real-time PCR used by industry (1) and was selected for genome sequencing to examine its role in BPS (2). Recent amplified ribosomal DNA restriction analysis (ARDRA) carried out on 90 New Zealand psychrotolerant *Clostridium* isolates derived from three meat production animal types and their environments (3) placed strain FP1 into the *Clostridium tagluense*-like cluster, with >95% similarity to the *C. tagluense* type strain A121 (4). Here, we present the draft annotated genome sequence of *Clostridium* sp. strain FP1, with similarity to *C. tagluense*, isolated from spoiled lamb.

Strain FP1 was isolated from the meat drip of vacuum-packaged lamb in 2017 that had no pack distension, with meat discoloration (some green spots) and sweaty feet odor. Meat drip was cultured anaerobically at 10°C in 10-fold suspensions in pre-reduced peptone-yeast extract-glucose-starch broth (1). Genomic DNA was extracted using a modified phenol-chloroform procedure, as described previously (5). Whole-genome sequencing of FP1 was performed with an Illumina TruSeq Nano library on the Illumina MiSeq platform (2 × 250-bp paired-end reagent kit v2) at the Massey Genome Service (Massey University, Palmerston North, New Zealand). In total, 3,511,896 paired-end raw reads were generated, corrected, trimmed, and *de novo* assembled using the A5-miseq pipeline v20169825 with standard parameters (6). The result of the FP1 assembly was 210 scaffolds with an N_{50} value of 126,678 bp, with the largest scaffold being 470,803 bp long. Based on the assembly information, the FP1 draft genome consists of 5,384,978 bp, with a G+C content of 31.0% and 145× coverage. The genomes were annotated with GAMOLA2 (7), DIAMOND v0.9.21.122 (8), InterProScan v5.36-75.0 (9), and OmicsBox v1.1.164 (10) software packages, using default parameters. A total of 5,116 putative protein-coding genes were predicted, along with 97 tRNAs, 45 rRNAs, and 231 noncoding RNA elements.

The dbCAN2 (11) software was used to determine the carbohydrate-active enzyme (CAZY) (12) profile of FP1 using default settings. The FP1 genome encodes a total of 17 glycoside hydrolases, 36 glycosyl transferases, 7 carbohydrate esterases, and 9 carbohydrate-binding protein module families. When compared to the recently se-

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quenced *C. estertheticum* subsp. *laramiense* strain DSM 14864^T (ATCC 51254^T) (13) and *C. estertheticum* strain DSM 8809^T (ATCC 51377^T) (14), FP1 encodes one-half as many carbohydrate-active enzymes. We propose that related *Clostridium* species may differ in their abilities to metabolize more complex insoluble polysaccharides, as both FP1 and *C. tagluense* A121^T lack genes encoding polysaccharide lyases and the genes required for uronic acid metabolism.

Data availability. The genome sequence data for *Clostridium* sp. strain FP1 were deposited under GenBank accession number [JAAMNJ000000000](https://ncbi.nlm.nih.gov/GenBank/entry/JAAMNJ000000000), BioProject accession number [PRJNA574489](https://ncbi.nlm.nih.gov/BioProject/entry/PRJNA574489), and Sequence Read Archive (SRA) accession number [SRR11113223](https://ncbi.nlm.nih.gov/SRA/entry/SRR11113223).

REFERENCES

- Brightwell G, Clemens R. 2012. Development and validation of a real-time PCR assay specific for *Clostridium estertheticum* and *C. estertheticum*-like psychrotolerant bacteria. *Meat Sci* 92:697–703. <https://doi.org/10.1016/j.meatsci.2012.06.025>.
- Mills J, Donnison A, Brightwell G. 2014. Factors affecting microbial spoilage and shelf-life of chilled vacuum-packed lamb transported to distant markets: a review. *Meat Sci* 98:71–80. <https://doi.org/10.1016/j.meatsci.2014.05.002>.
- Brightwell G, Horváth KM. 2018. Molecular discrimination of New Zealand sourced meat spoilage associated psychrotolerant *Clostridium* species by ARDRA and its comparison with 16S RNA gene sequencing. *Meat Sci* 138:23–27. <https://doi.org/10.1016/j.meatsci.2017.12.007>.
- Murakami T, Mori H, Shcherbakova VA, Yoshimura Y, Segawa T. 2019. Draft genome sequence of *Clostridium tagluense* strain A121^T, isolated from a permafrost core in the Canadian High Arctic. *Microbiol Resour Announc* 8:e01630-18. <https://doi.org/10.1128/MRA.01630-18>.
- Bouillaut L, McBride SM, Sorg JA. 2011. Genetic manipulation of *Clostridium difficile*. *Curr Protoc Microbiol* Chapter 9:Unit 9A.2.
- Coil D, Jospin G, Darling AE. 2015. A5-miseq: an updated pipeline to assemble microbial genomes from Illumina MiSeq data. *Bioinformatics* 31:587–589. <https://doi.org/10.1093/bioinformatics/btu661>.
- Altermann E, Lu J, McCulloch A. 2017. GAMOLA2, a comprehensive software package for the annotation and curation of draft and complete microbial genomes. *Front Microbiol* 8:346. <https://doi.org/10.3389/fmicb.2017.00346>.
- Buchfink B, Xie C, Huson DH. 2015. Fast and sensitive protein alignment using DIAMOND. *Nat Methods* 12:59–60. <https://doi.org/10.1038/nmeth.3176>.
- Jones P, Binns D, Chang H-Y, Fraser M, Li W, McAnulla C, McWilliam H, Maslen J, Mitchell A, Nuka G, Pesseat S, Quinn AF, Sangrador-Vegas A, Scheremetjew M, Yong S-Y, Lopez R, Hunter S. 2014. InterProScan 5: genome-scale protein function classification. *Bioinformatics* 30:1236–1240. <https://doi.org/10.1093/bioinformatics/btu031>.
- Conesa A, Götz S, García-Gómez JM, Terol J, Talón M, Robles M. 2005. Blast2GO: a universal tool for annotation, visualization and analysis in functional genomics research. *Bioinformatics* 21:3674–3676. <https://doi.org/10.1093/bioinformatics/bti610>.
- Zhang H, Yohe T, Huang L, Entwistle S, Wu P, Yang Z, Busk PK, Xu Y, Yin Y. 2018. dbCAN2: a meta server for automated carbohydrate-active enzyme annotation. *Nucleic Acids Res* 46:W95–W101. <https://doi.org/10.1093/nar/gky418>.
- Cantarel BL, Coutinho PM, Rancurel C, Bernard T, Lombard V, Henrissat B. 2009. The Carbohydrate-Active EnZymes database (CAZy): an expert resource for glycogenomics. *Nucleic Acids Res* 37:D233–D238. <https://doi.org/10.1093/nar/gkn663>.
- Pavevich N, Pavevich FP, Maclean PH, Jauregui R, Altermann E, Mills J, Brightwell G. 2019. Draft genome sequence of *Clostridium estertheticum* subsp. *laramiense* DSM 14864^T, isolated from spoiled uncooked beef. *Microbiol Resour Announc* 8:e01275-19. <https://doi.org/10.1128/MRA.01275-19>.
- Yu Z, Gunn L, Brennan E, Reid R, Wall PG, Gaora PÓ, Hurley D, Bolton D, Fanning S. 2016. Complete genome sequence of *Clostridium estertheticum* DSM 8809, a microbe identified in spoiled vacuum packed beef. *Front Microbiol* 7:1764. <https://doi.org/10.3389/fmicb.2016.01764>.