



Draft Genome Sequence of the Moderately Heat-Tolerant Lactococcus lactis subsp. lactis bv. diacetylactis Strain GL2 from Algerian **Dromedary Milk**

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Lactococcus lactis subsp. lactis bv. diacetylactis GL2 is a moderately thermotolerant lactic acid bacterium isolated from dromedary raw milk. Here, we present the draft genome sequence of this potential new dairy starter strain, which combines thermotolerance and the capacity to metabolize lactose, casein, and citrate.

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romedary milk constitutes an important protein source in pastoral societies of several countries; it is traditionally fermented by a natural flora that contains a panel of lactic acid bacteria, microorganisms which are widely used in food fermentations. A number of Lactococcus genome sequences have been determined (e.g., 1–3). In this work, we announce the draft genome sequence of the atypical Lactococcus lactis subsp. lactis by. diacetylactis strain GL2, isolated from dromedary raw milk in south Algeria and formerly designated HD9B (4). In contrast to most other lactococci, this strain showed noticeable growth up to a temperature of 45°C. This thermotolerance probably reflects an adaptation to the high temperatures of the Algerian desert and is an interesting technological trait for cheese-making processes. In addition, the strain grows rapidly in cow's milk and is able to ferment citrate, lactose, and many other carbohydrates, properties that have also been reported for L. lactis subsp. lactis and some bv. diacetylactis strains isolated from plant material (5).

L. lactis GL2 was cultured at 30°C in M17 medium (6) with 1% glucose. Total genomic DNA was isolated with the DNeasy blood and tissue kit (Qiagen). TruSeq DNA library preparations led to inserts of 250 to 650 bp. We collected 780-Mb paired-end (PE) raw reads from an Illumina MiSeq, and the quality was checked by FastQC (http://www.bioinformatics.babraham.ac.uk/projects /fastqc). FLASH (7) was applied for merging overlapping pairedend reads, and both adapters (p5 and p9) were clipped from these reads (merged and unconnected PE) using cutadapt (8). All lowquality tails (<25) were removed. *k*-mers were counted with Jellyfish (9), and the results were used to correct reads by the QUAKE tool (10). The assembly was carried out using Velvet (11) and IDBA-UD (12). Overlapping small contigs resulted in 48 contigs (>5 kb), totaling 2,245,404 bp, with an average G+C content of 35.1%.

Phylogenetic analysis using the maximum-likelihood method indicated L. lactis subsp. lactis KLDS4.0325 (3) isolated from ku-

mis as the closest related sequenced strain, and the next closest was IL1403 (1). Genome functional annotation was performed with RAST 4 (13) and resulted in 2,203 protein-coding sequences (CDSs), among them a number of strain-specific CDSs, 43 tRNAs, and 4 rRNAs. The presence of chromosomal genes for diacetyl/ acetoin production was confirmed. In addition, 39 noncoding RNAs were annotated by Infernal/Rfam (14).

A detailed analysis of the genome will give further insights into the genetic basis of the biotechnologically important traits of this strain.

Nucleotide sequence accession numbers. The whole-genome shotgun project has been deposited in GenBank/EMBL/DDBJ under the accession no. **INCC02000000** (chromosome), consisting of sequences JNCC02000001 to JNCC02000048.

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REFERENCES

- 1. Bolotin A, Wincker P, Mauger S, Jaillon O, Malarme K, Weissenbach J, Ehrlich SD, Sorokin A. 2001. The complete genome sequence of the lactic acid bacterium Lactococcus lactis ssp. lactis IL1403. Genome Res 11: 731-753. http://dx.doi.org/10.1101/gr.GR-1697R.
- 2. Falentin H, Naquin D, Loux V, Barloy-Hubler F, Loubière P, Nouaille S, Lavenier D, Le Bourgeois P, Francois P, Schrenzel J, Hernandez D, Even S, Le Loir Y. 2014. Genome sequence of Lactococcus lactis subsp. lactis bv. diacetylactis LD61. Genome Announc 2(1):e01176-13. http:// dx.doi.org/10.1128/genomeA.01176-13.
- 3. Yang X, Wang Y, Huo G. 2013. Complete genome sequence of Lactococcus lactis subsp. lactis KLDS4.0325. Genome Announc 1(6):e00962-13. http://dx.doi.org/10.1128/genomeA.00962-13.
- 4. Drici H, Gilbert C, Kihal M, Atlan D. 2010. Atypical citrate-fermenting

Lactococcus lactis strains isolated from dromedary's milk. J Appl Microbiol **108:**647–657. http://dx.doi.org/10.1111/j.1365-2672.2009.04459.x.

- Nomura M, Kobayashi M, Narita T, Kimoto-Nira H, Okamoto T. 2006. Phenotypic and molecular characterization of *Lactococcus lactis* from milk and plants. J Appl Microbiol 101:396–405. http://dx.doi.org/10.1111/ j.1365-2672.2006.02949.x.
- Terzaghi B, Sandine W. 1975. Improved medium for lactic streptococci and their bacteriophages. Appl Environ Microbiol 29:807–813.
- Magoč T, Salzberg SL. 2011. FLASH: fast length adjustment of short reads to improve genome assemblies. Bioinformatics 27:2957–2963. http:// dx.doi.org/10.1093/bioinformatics/btr507.
- Martin M. 2011. Cutadapt removes adapter sequences from highthroughput sequencing reads. EMBnet.journal 17:10-12. http:// dx.doi.org/10.14806/ej.17.1.200.
- 9. Marçais G, Kingsford C. 2011. A fast, lock-free approach for efficient parallel counting of occurrences of *k*-mers. Bioinformatics 27:764–770. http://dx.doi.org/10.1093/bioinformatics/btr011.

- 10. Kelley DR, Schatz MC, Salzberg SL. 2010. Quake: quality-aware detection and correction of sequencing errors. Genome Biol 11:R116.
- Zerbino DR, Birney E. 2008. Velvet: algorithms for *de novo* short read assembly using de Bruijn graphs. Genome Res 18:821–829. http:// dx.doi.org/10.1101/gr.074492.107.
- Peng Y, Leung HCM, Yiu SM, Chin FYL. 2012. IDBA-UD: a *de novo* assembler for single-cell and metagenomic sequencing data with highly uneven depth. Bioinformatics 28:1420–1428. http://dx.doi.org/10.1093/ bioinformatics/bts174.
- 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.
- Nawrocki EP, Eddy SR. 2013. Infernal 1.1: 100-fold faster RNA homology searches. Bioinformatics 29:2933–2935. http://dx.doi.org/10.1093/ bioinformatics/btt509.