

# Draft Genome Sequence of *Lactobacillus rossiae* DSM 15814<sup>T</sup>

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**The draft genome sequence of *Lactobacillus rossiae* DSM 15814<sup>T</sup> (CS1, ATCC BAA-88) was determined by a whole-genome shotgun approach. Reads were assembled to a 2.9-Mb draft version. RAST genome annotation evidenced 2,723 predicted coding sequences. Many carbohydrate, amino acid, and amino acid derivative subsystem features were found.**

The genus *Lactobacillus* belongs to the phylum *Firmicutes*, class *Bacillus*, order *Lactobacillales*, and family *Lactobacillaceae*. *Lactobacilli* are Gram-positive, catalase-negative, non-spore-forming, rod-shaped bacteria that produce lactic acid as the major end product of fermentation. *Lactobacillus* is the largest genus within the group of lactic acid bacteria (5).

*Lactobacillus rossiae* was usually found within the autochthonous microbiota of sourdoughs (4, 11, 13, 14), spelt flour (3), pineapple (9), and the gastrointestinal tracts of humans (8) and animals (6). The genotypic and phenotypic diversity of *L. rossiae* strains isolated from sourdough was described previously (7, 13). Some strains were selected for antifungal activity (17) and used in sourdough biotechnology for glutamate production (15) and wheat germ fermentation (12). The genome sequence of *L. rossiae* DSM 15814<sup>T</sup> (CS1, ATCC BAA-822) will be useful to explore its biotechnology properties.

A total of 30,544,098 whole-genome shotgun, 100-bp paired-end reads were generated using Illumina sequencing technology. Library preparation was carried out with minor modifications to the TruSeq DNA sample preparation protocol (Illumina, Inc., San Diego, CA). Briefly, 1 µg of bacterial DNA was sheared to an average length of 500 to 600 bp using the Diagenode Biorupter XL sonicator system (Sparta, NJ), and standard blunt ending with “A” base (paired-end DNA sample preparation kit; Illumina, Inc.) was performed. Illumina index adapters were ligated to the ends of the fragments. After ligation reaction and separation of nonligated adapters, samples were amplified by PCR to selectively enrich those fragments in the library having adapter molecules at both ends. The sample was quantified and the quality was tested using a NanoDrop ND-1000 UV-Vis spectrophotometer (Thermo Scientific, Wilmington, DE) and an Agilent 2100 bioanalyzer (Agilent Technologies, Santa Clara, CA). The library was pooled with the other eight bacterial genomes in equimolar ratios to yield a total concentration of 10 nM. Aliquots of pooled libraries (2 pmol) were processed with cBot (Illumina, Inc.) by following the manufacturer’s recommendations. The HiSeq 2000 system was programmed for a paired-end sequencing run of 101 cycles. Raw images were processed using Illumina Pipeline software version RTA 2.8.0/OLB 1.8.0/CASAVA 1.7.0.

After filtering low-quality reads, 30,017,879 high-quality reads were assembled into contigs using CLC Genomics Workbench version 5.01 (CLC Bio, Denmark).

The annotation was done by merging the results obtained from the RAST server (1) and checked by BLAST analysis when needed. In addition, the scaffolds were searched against the KEGG (10),

UniProt (2), and COG (16) databases to annotate the gene descriptions.

The draft genome includes 278 contigs covering 2,946,462 bp ( $N_{50}$  of 150,537 bp, average contig size of 11,466 bp, maximum contig size of 528,241 bp, with an average coverage of 1,000×). A total number of 2,723 predicted coding sequences were annotated.

There are 287 subsystems that are represented in the genome, and this information was used to reconstruct the metabolic network. The closest genome is that of *Lactobacillus brevis* (genome identification number 387344.13 [SEED Viewer version 2.0]). Many carbohydrate, amino acid, and amino acid derivative subsystem features were found, including genes involved in central carbohydrate, monosaccharide, and fermentation metabolisms. Many protein and DNA metabolism subsystem features were also identified.

**Nucleotide sequence accession number.** The whole-genome shotgun project has been deposited at DDBJ/EMBL/GenBank under the accession number [AKZK00000000](http://dx.doi.org/10.1093/nucleic/acc000).

## ACKNOWLEDGMENT

This work was supported by the project “IDEA Giovani Ricercatori” at the University of Bari Aldo Moro.

## REFERENCES

1. Aziz RK, et al. 2008. The RAST server: rapid annotations using subsystems technology. *BMC Genomics* 9:75.
2. Bairoch A, et al. 2005. The Universal Protein Resource (UniProt). *Nucleic Acids Res.* 33:D154–D159.
3. Coda R, et al. 2010. Spelt and emmer flours: characterization of the lactic acid bacteria microbiota and selection of mixed starters for bread making. *J. Appl. Microbiol.* 108:925–935.
4. Corsetti A, et al. 2005. *Lactobacillus rossii* sp. nov. isolated from wheat sourdough. *Int. J. Syst. Evol. Microbiol.* 55:35–40.
5. De Angelis M, Gobbetti M. 2012. *Lactobacillus* spp.: general characteristics, p 78–90. In Fuquay JW, Fox PF, McSweeney PLH (ed), *Encyclopedia of dairy sciences*, 2nd ed, vol 3. Academic Press, San Diego, CA.
6. De Angelis M, et al. 2006. Selection of potential probiotic lactobacilli from pig feces to be used as additives in pelleted feeding. *Res. Microbiol.* 157:792–801.
7. Di Cagno R, et al. 2007. Genotypic and phenotypic diversity of *Lactobacillus rossiae* strains isolated from sourdough. *J. Appl. Microbiol.* 10:821–835.

Received 11 July 2012 Accepted 17 July 2012

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doi:10.1128/JB.01248-12

8. Di Cagno R, et al. 2009. Different fecal microbiotas and volatile organic compounds in treated and untreated children with celiac disease. *Appl. Environ. Microbiol.* 75:3963–3971.
9. Di Cagno R, et al. 2010. Taxonomic structure of the yeasts and lactic acid bacteria microbiota of pineapple (*Ananas comosus* L. Merr.) and use of autochthonous starters for minimally processing. *Food Microbiol.* 27:381–389.
10. Kanehisa M, Goto S, Kawashima S, Okuno Y, Hattori M. 2004. The KEGG resource for deciphering the genome. *Nucleic Acids Res.* 32:D277–D280.
11. Minervini F, et al. 2010. Robustness of *Lactobacillus plantarum* starters during daily propagation of wheat flour sourdough type I. *Food Microbiol.* 27:897–908.
12. Rizzello CG, Nionelli L, Coda R, De Angelis M, Gobbetti M. 2010. Effect of sourdough fermentation on stabilisation, and chemical and nutritional characteristics of wheat germ. *Food Chem.* 119:1079–1089.
13. Scheirlinck I, et al. 2009. Polyphasic taxonomic characterization of *Lactobacillus rossiae* isolates from Belgian and Italian sourdoughs reveals intraspecific heterogeneity. *Syst. Appl. Microbiol.* 32:151–156.
14. Siragusa S, et al. 2009. Taxonomic structure and monitoring of the dominant population of lactic acid bacteria during wheat flour sourdough type I propagation using *Lactobacillus sanfranciscensis* starters. *Appl. Environ. Microbiol.* 75:1099–1109.
15. Stromeck A, Hu Y, Chen L, Gänzle MG. 2011. Proteolysis and bioconversion of cereal proteins to glutamate and  $\gamma$ -aminobutyrate (GABA) in rye malt sourdoughs. *J. Agric. Food Chem.* 59:1392–1399.
16. Tatusov RL, et al. 2003. The COG database: an updated version includes eukaryotes. *BMC Bioinformatics* 4:41.
17. Valerio F, et al. 2009. Antifungal activity of strains of lactic acid bacteria isolated from a semolina ecosystem against *Penicillium roqueforti*, *Aspergillus niger* and *Endomyces fibuliger* contaminating bakery products. *Syst. Appl. Microbiol.* 32:438–448.