JOURNAL OF BACTERIOLOGY, Sept. 2010, p. 4800 Vol. 192, No. 18
0021-9193/10/$12.00 doi:10.1128/JB.00702-10

Complete Genome Sequence of *Lactobacillus fermentum* CECT 5716, a Probiotic Strain Isolated from Human Milk[▽]

Esther Jiménez, Susana Langa, Virginia Martín, Rebeca Arroyo, Rocío Martín,
Leónides Fernández, and Juan M. Rodríguez*

Departamento de Nutrición, Bromatología y Tecnología de los Alimentos, Universidad Complutense de Madrid, 28040 Madrid, Spain

Received 18 June 2010/Accepted 7 July 2010

Lactobacillus fermentum **is a heterofermentative lactic acid bacterium and is frequently isolated from mucosal surfaces of healthy humans.** ***Lactobacillus fermentum*** **CECT 5716 is a well-characterized probiotic strain isolated from human milk and, at present, is used in commercial infant formulas. Here, we report the complete and annotated genome sequence of this strain.**

Breast milk is the best food for neonates because it provides a unique combination of nutrients and bioactive compounds, ensuring correct growth and development of the infant. In addition, it also contains probiotic bacteria (4, 5). In a previous study, we isolated *Lactobacillus fermentum* CECT 5716 from such biological fluid (3). Subsequent studies revealed that this strain was a good probiotic candidate since it reached high survival rates when exposed to gastrointestinal tract-like conditions, showed a strong adherence to intestinal cells, stimulated the expression of mucin-encoding genes, produced antimicrobial compounds, and displayed *in vivo* and *in vitro* immunomodulatory and antibacterial properties against pathogenic bacteria (1, 5, 7). *L. fermentum* CECT 5716 showed a beneficial effect in a murine model of intestinal inflammation, reducing the inflammatory response and the intestinal damage (2). In addition, consumption of this strain enhances the response to influenza vaccination in healthy volunteers and reduces the incidence of influenza-like illness (8).

In order to interrogate the genome sequence of *Lactobacillus fermentum* CECT 5716 with regard to its probiotic properties, the complete genome sequence was determined by a whole-genome shotgun strategy using 454 pyrosequencing technology (454 Life Sciences, Banford, CT). The initial draft assembly provided by 454 Life Sciences was based on 193,362 pyrosequencing reads with an average read length of 250 nucleotides which assembled into 1,343 contigs. Sequence reads were assembled automatically with the Life Sciences GS FLX (Newbler) program. The genome sequence of *Lactobacillus fermentum* IFO 3956 (6) was used to order these contigs into large scaffolds. The assembling process was relatively complex due to the 83 transposase-encoding regions that were found in the CECT 5716 genome.

The complete genome of *Lactobacillus fermentum* CECT 5716 consists of a circular chromosome of 2,100,449 bp, with a GC content of 51.49%, and has no plasmid. Its chromosome contains 1,109 predicted protein-encoding genes, 54 tRNA-encoding genes, and 20 rRNA-encoding genes. The comparison of the CECT 5716 and IFO 3956 genomes revealed that they were highly similar, with the exception of 16 protein-encoding genes that are present in CECT 5716 but not in IFO 3956. Among them, there are putative enzymes involved in the metabolism of purines (allantoinase, GMP oxidoreductase, GMP synthase), amino acids (serine-pyruvate transaminase, 3 glutamate synthases), lipids (acyltransferase), and carbohydrates (mannose-6-phosphate isomerase).

Nucleotide sequence accession number. The sequence data for the *L. fermentum* CECT 5716 genome are available in GenBank/EMBL under accession no. CP002033.

This study was partly supported by the FUN-C-FOOD (Consolider-Ingenio 2010) and AGL2007-62042 projects from the Ministerio de Educación y Ciencia (Spain) and by a research contract funded by Puleva Biotech.

REFERENCES

1. **Diaz-Ropero, M. P., R. Martin, S. Sierra, F. Lara-Villoslada, J. M. Rodriguez, J. Xaus, and M. Olivares.** 2007. Two *Lactobacillus* strains, isolated from breast milk, differently modulate the immune response. J. Appl. Microbiol. **102:**337–343.
2. **Mañé, J., V. Lorén, E. Pedrosa, I. Ojanguren, J. Xaus, E. Cabré, E. Domènech, and M. A. Gassull.** 2009. *Lactobacillus fermentum* CECT5716 prevents and reverts intestinal damage on TNBS-induced colitis in mice. Inflamm. Bowel Dis. **15:**1155–1163.
3. **Martín, R., S. Langa, C. Reviriego, E. Jiménez, M. L. Marín, J. Xaus, L. Fernández, and J. M. Rodríguez.** 2003. Human milk is a source of lactic acid bacteria for the infant gut. J. Pediatr. **143:**754–758.
4. **Martín, R., M. Olivares, M. L. Marín, L. Fernández, J. Xaus, and J. M. Rodríguez.** 2005. Probiotic potential of 3 lactobacilli strains isolated from breast milk. J. Hum. Lact. **21:**8–17.
5. **Martín, R., E. Jiménez, M. Olivares, M. L. Marín, L. Fernández, J. Xaus, and J. M. Rodríguez.** 2006. *Lactobacillus salivarius* CECT 5713, a potential probiotic strain isolated from infant feces and breast milk of a mother-child pair. Int. J. Food Microbiol. **112:**35–43.
6. **Morita, H., H. Toh, S. Fukuda, H. Horikawa, K. Oshima, T. Suzuki, M. Murakami, S. Hisamatsu, Y. Kato, T. Takizawa, H. Fukuoka, T. Yoshimura, K. Itoh, D. J. O'Sullivan, L. L. McKay, H. Ohno, J. Kikuchi, T. Masaoka, and M. Hattori.** 2008. Comparative genome analysis of *Lactobacillus reuteri* and *Lactobacillus fermentum* reveal a genomic island for reuterin and cobalamin production. DNA Res. **15:**151–161.
7. **Olivares, M., M. P. Díaz-Ropero, R. Martín, J. M. Rodríguez, and J. Xaus.** 2006. Antimicrobial potential of four *Lactobacillus* strains isolated from breast milk. J. Appl. Microbiol. **101:**72–79.
8. **Olivares, M., M. P. Díaz-Ropero, S. Sierra, F. Lara-Villoslada, J. Fonollá, M. Navas, J. M. Rodríguez, and J. Xaus.** 2007. Oral intake of *Lactobacillus fermentum* CECT5716 enhances the effects of influenza vaccination. Nutrition **23:**254–260.

---

* Corresponding author. Mailing address: Departamento de Nutrición, Bromatología y Tecnología de los Alimentos, Universidad Complutense de Madrid, Ciudad Universitaria, Avda. Puerta de Hierro, s/n. 28040 Madrid, Spain. Phone: 34-91-3943837. Fax: 34-91-3943743. E-mail: jmrodrig@vet.ucm.es.

[▽] Published ahead of print on 16 July.