Journal of Bacteriology

Complete Genome Sequence of Bifidobacterium breve CECT 7263, a Strain Isolated from Human Milk

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J. Bacteriol. **2012, 194(14):3762. DOI: 10.1128/JB.00691-12.**

JB Journals.ASM.org

GENOME ANNOUNCEMENT

Complete Genome Sequence of *Bifidobacterium breve* CECT 7263, a Strain Isolated from Human Milk

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***Bifidobacterium breve* is an actinobacterium frequently isolated from colonic microbiota of breastfeeding babies. Here, we report the complete and annotated genome sequence of a *B. breve* strain isolated from human milk, *B. breve* CECT 7263. The genome sequence will provide new insights into the biology of this potential probiotic organism and will allow the characterization of genes related to beneficial properties.**

Bifidobacteria are common members of the human gut microbiota and are believed to play a beneficial role in maintaining the health of the host. They were first isolated a century ago (14) from infant feces and were soon associated with a healthy infant gut because of their predominance in breastfed infants in comparison to formula-fed ones (4, 5). The health-promoting effects of breast milk have been linked, at least partly, to different components but, also, to the presence of live bacteria in this biological fluid, including lactic acid bacteria (8, 10) and bifidobacteria (7, 9, 11).

Here, we report the first annotated genome of a *Bifidobacterium breve* strain isolated from human milk, which will allow improved knowledge of its potential probiotic traits. The entire genome of *B. breve* CECT 7263 was sequenced by 454 Pyrosequencing on a GS-FLX sequencer to a coverage of 24-fold (454 Life Sciences, Banford, CT). The initial draft assembly provided 46 contigs using the Newbler program version 2.3 (Roche Applied Science). The incomplete draft genome includes 2,314,396 bases with a GC content of 58.9%, a total of 1,868 protein-encoding sequences, and 57 RNA encoding sequences. Coding regions were predicted using the BG7 prediction system (Era7 Technologies, Granada, Spain) that goes from protein similarity detection to open reading frame (ORF) prediction. The BG7 system avoids the loss of genes with frameshifts or alterations in the start or stop codons and is tolerant to fragmentation of genes in different contigs (frequent in next-generation-sequencing [NGS] genome projects). The semiautomatic annotation of the sequences resulted in 34 final contigs, 1,725 protein-coding genes, 53 tRNA-encoding genes, and 3 rRNA operons.

In recent years, several studies have shown the ability of bifidobacteria to produce conjugated linoleic acid (CLA) and conjugated linolenic acid (CLNA) isomers, a property that seems particularly associated with *B. breve* strains (1, 3, 12). CLA and CLNA isomers are interesting because of their anticarcinogenic properties, immune modulation, and antiobesity activities (2, 15). In this context, the human milk *B. breve* CECT 7263 strain showed a 75% bioconversion of linoleic acid into *cis*-9,*trans*-11-CLA and approximately 96% bioconversion of α-linolenic acid into *cis*-9,*trans*-11,*cis*-15-CLNA (unpublished results). Comparative analysis of orthologous neighborhood genomic regions revealed that CECT 7263 shares with other *B. breve* strains the distinctive clustering of genes encoding a FAD-dependent fatty acid hydratase and a NAD(P)(H) oxidoreductase, which might be part of a multicomponent enzyme machinery for conjugated fatty acid synthesis (6, 13).

Nucleotide sequence accession number. The genome sequence of *B. breve* CECT 7263 has been deposited in DDBJ/EMBL/GenBank under accession number AFVV00000000.

ACKNOWLEDGMENTS

This study was partly supported by the FUN-C-FOOD (Consolider-Ingenio 2010) and AGL2010-15420 projects from the Ministerio de Economía y Competitividad (Spain) and by a research contract funded by Lactalis Puleva.

REFERENCES

1. **Barrett E, Ross RP, Fitzgerald GF, Stanton C.** 2007. Rapid screening method for analyzing the conjugated linoleic acid production capabilities of bacterial cultures. Appl. Environ. Microbiol. **73**:2333–2337.
2. **Bhattacharya A, Banu J, Rahman M, Causey J, Fernandes G.** 2006. Biological effects of conjugated linoleic acids in health and disease. J. Nutr. Biochem. **17**:789–810.
3. **Gorissen L, De Vuyst L, Raes K, De Smet S, Leroy F.** 2012. Conjugated linoleic and linolenic acid production kinetics by bifidobacteria differ among strains. Int. J. Food Microbiol. **155**:234–240.
4. **Harmsen HJM, et al.** 2000. Analysis of intestinal flora development in breast-fed infants by using molecular identification and detection methods. J. Pediatr. Gastroenterol. Nutr. **30**:61–67.
5. **Hopkins MJ, Macfarlane GT, Furrie E, Fite A, Macfarlane S.** 2005. Characterisation of intestinal bacteria in infant stools using real-time PCR and northern hybridisation analyses. FEMS Microbiol. Ecol. **54**:77–85.
6. **Kishino S, et al.** 2011. Novel multi-component enzyme machinery in lactic acid bacteria catalyzing C=C double bond migration useful for conjugated fatty acid synthesis. Biochem. Biophys. Res. Commun. **416**:188–193.
7. **Makino H, et al.** 2011. Transmission of intestinal *Bifidobacterium longum* subsp. *longum* strains from mother to infant, determined by multilocus sequencing typing and amplified fragment length polymorphism. Appl. Environ. Microbiol. **77**:6788–6793.
8. **Martín R, et al.** 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.
9. **Martín R, et al.** 2009. Isolation of bifidobacteria from breast milk and

Received 24 April 2012 **Accepted** 30 April 2012

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doi:10.1128/JB.00691-12

assessment of the bifidobacterial population by PCR-denaturing gradient gel electrophoresis and quantitative real-time PCR. Appl. Environ. Microbiol. **75**:965–969.
10. **Martín R, et al.** 2003. Human milk is a source of lactic acid bacteria for the infant gut. J. Pediatr. **143**:754–758.
11. **Martín V, et al.** 2012. Sharing of bacterial strains between breast milk and infant faeces. J. Hum. Lact. **28**:36–44.
12. **Park HG, et al.** 2009. Characterization of conjugated linoleic acid production by *Bifidobacterium breve* LMC 520. J. Agric. Food Chem. **57**:7571–7575.
13. **Rosberg-Cody E, et al.** 2011. Myosin-cross-reactive antigen (MCRA) protein from *Bifidobacterium breve* is a FAD-dependent fatty acid hydratase which has a function in stress protection. BMC Biochem. **12**:9–20. doi:10.1186/1471-2091-12-9.
14. **Tissier H.** 1906. Traitement des infections intestinales par la méthode de la flore bactérienne de l'intestin. Crit. Rev. Soc. Biol. **60**:359–361.
15. **Tricon S, Burdge GC, Williams CM, Calder PC, Yaqoob P.** 2005. The effects of conjugated linoleic acid on human health-related outcomes. Proceed. Nutr. Soc. **64**:171–182.