Korean Journal of Microbiology (2018) Vol. 54, No. 2, pp. 171-173
DOI https://doi.org/10.7845/kjm.2018.8033

pISSN 0440-2413
eISSN 2383-9902

Draft genome sequences of *Enterococcus faecium* JB00008 (KACC 92186P) isolated from Korean fermented soybean paste (Cheonggukjang)

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한국 전통유래식품(청국장)에서 분리한 *Enterococcus faecium* JB00008 (KACC 92186P) 유산균주의 유전체 분석

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(Received June 1, 2018; Accepted June 14, 2018)

Enterococcus faecium was commonly used as a probiotics and feed additives to human and animals because of their beneficial effects. We sequenced the genome of *E. faecium* JB00008 (KACC 92186P) isolated from a Korean fermented soybean paste (Cheonggukjang) that showed antibacterial activity against *Escherichia coli*. A 2,847,295-bp draft genome was obtained, and it has in 37.84% G + C content in 34 contigs (length, ≥ 500 bp).

Keywords: *Enterococcus faecium*, antibacterial activity, genome sequencing, hybrid assembly, probiotics

Enterococcus spp. are Gram-positive cocci and facultative anaerobes (Fisher and Phillips, 2009). *Enterococcus faecium* normally colonizes the human/animal gut (Hammerum, 2012). Due to their beneficial effects, such as heat tolerance and antibacterial activity, probiotics including *Enterococcus* spp. are commonly used in the food/feed industry (Lee, 2002; Marciňáková *et al*., 2004; Goh *et al*., 2005). We isolated 87 *E. faecium* strains from a traditional Korean fermented soybean paste (Cheonggukjang). All *E. faecium* strains were identified using PCR-based method (Park *et al*., 2017). The antibacterial activity of these strains were tested against pathogenic bacteria (K88 antigen-positive *Escherichia coli*). Among all strains, *E. faecium* JB00008 (KACC 92186P) showed the highest antibacterial activity. For genome sequencing, single colony of *E. faecium* JB00008 cultured on an Enterococcosel agar plate was inoculated into the BHI (Brain-Heart Infusion) broth. The cells were incubated at 37°C for 24 h and harvested by centrifuging at 13,000 × *g* for 1 min. The purely cultured cells were washed twice with 1 × PBS (Phosphate-buffered saline) solution. The genomic DNA of the strain was extracted by using the G-spin Total DNA Extraction kit (iNtRON Biotechnology, Cat #17121), according to the manufacturer's instructions. We constructed libraries and sequenced using HiSeq X system (Illumina) and MinION (Oxford Nanopore), respectively. An Illumina sequencing library contained ~350-bp inserts by using the Nextera XT DNA Library Preparation kit (Illumina Inc., Cat #FC-131-1096), according to the manufacturer's instructions.

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For a MinION sequencing library, genomic DNA was processed by using the Rapid Low Input by PCR Barcoding Kit (SQK-RLB001, Oxford Nanopore). The libraries were independently sequenced on the HiSeq X system (Illumina) for 150-bp paired-end reads and on the MinION flow cell with R9 chemistry (Oxford Nanopore), respectively. After sequencing, the adapter sequences were trimmed out by using in-house Perl scripts and Cutadapt 1.14 (Martin, 2011). The Illumina and MinION reads, in which 95% of bases have quality scores of ≥ 31, were selected. Both reads were hybrid-assembled *de novo* by using SPAdes version 3.11.1 with the options –careful and –nanopore.

Finally, the 2,847,295-bp draft genome was assembled into 34 contigs (Table 1, contig length, ≥ 500 bp; G + C content, 37.84%). A total of 2,680 protein-coding sequences (CDS) and 46 RNAs were predicted by web-based annotation server, Rapid Annotation Using Subsystems Technology (RAST) (Overbeek *et al*., 2013). Enterocin-related genes (*entIM*, *entL50A*, *entL50B*, *entIT*, *entP*) associated with antimicrobial activities thereby producing enterocin are present in the draft genome. Enterocin is a particular bacteriocin produced by the *Enterococcus* species and has an antimicrobial activity against a broad range of Gram-positive and Gram-negative bacteria (Parente and Hill, 1992). Due to safety issues on *Enterococcus*, we examined the presence of virulence factors (VF) and antibiotic resistance (AR) genes in the draft genome. Among 22,189 antibiotic resistance genes, reported in the Comprehensive Antibiotic Resistance Database (Liu and Pop, 2008), only one AR gene (*bacA*) responsible for resistance against bacitracin, was detected in the draft genome. The genome sequencing data will be useful for a deeper understanding of *E. faecium* strain isolated from the Korean fermented soybean pastes. The *E. faecium* JB00008 (KACC 92186P) strain was deposited in the Korean Agricultural Culture Collection, Rural Development Administration, Wanju, Korea (accession No. KACC 92186P) for a domestic patent.

Table 1. Genome features of the *E. faecium* JB00008 (KACC 92186P)

Features	Chromosome
Genome size (bp)	2,847,295
No. of contigs (≥ 500 bp)	34
GC content (%)	37.84
No. of RNA genes	46
No. of protein-coding genes	2,680

Nucleotide sequence accession numbers

This whole-genome shotgun project has been deposited at GenBank under the accession number NPOO00000000. The version of the draft genome described in this paper is the second version replaced accession number NPOO00000000.2 (https://www.ncbi.nlm.nih.gov/Traces/wgs/?val=NPOO02#contigs).

적 요

본 연구에서 이용된 *Enterococcus faecium* JB00008 (KACC 92186P) 균주는 전통발효식품인 청국장에서 분리되었다. 이 균주는 *Escherichia coil*에 대해 높은 항균활성능력을 보였다. 우리는 이 균주의 유전체의 염기 서열을 분석하였다. 유전체 초안은 500 bp 이상의 contig 34개로 조립되었으며, 크기가 2,847,295 bp이고, G + C 함량(%)는 37.84%이다. 유전체 초안에서 항균활성물질(bacteriocin) 중 하나인 enterocin과 연관된 유전자가 5개 확인되었다.

Acknowledgements

This study was supported by a grant from the (1) Strategic Initiative for Microbiomes in Agriculture and Food, Ministry of Agriculture, Food and Rural Affairs, Republic of Korea as part of the multi-ministerial Genome Technology to Business Translation Program (Grant ID: 914005–04) and (2) the Basic Science Research Program through the National Research Foundation of Korea (NRF) funded by the Ministry of Education (NRF-2016R1C1B2016246) and Brain Korea 21 Plus Project and (3) the 2017 Research Grant from Kangwon National University (No. 520170234). Jongbin Park and Gwi-Deuk Jin were supported by Brain Korea 21 plus program.

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