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Complete genome sequence of *Lactococcus taiwanensis* strain K_LL004, encoding hydrolytic enzymes of plant polysaccharides isolated from grasshopper (*Oxya chinensis sinuosa*)

Hyunok Doo[1#], Hyeri Kim[1#], Jin Ho Cho[2#], Minho Song[3#], Eun Sol Kim[1], Jae Hyoung Cho[1], Sheena Kim[1], Gi Beom Keum[1], Jinok Kwak[1], Sriniwas Pandey[1], Hyeun Bum Kim[1]* and Ju-Hoon Lee[4]*

[1]*Department of Animal Resources Science, Dankook University, Cheonan 31116, Korea*
[2]*Department of Animal Science, Chungbuk National University, Cheongju 28644, Korea*
[3]*Division of Animal and Dairy Science, Chungnam National University, Daejeon 34134, Korea*
[4]*Department of Food Animal Biotechnology, Department of Agricultural Biotechnology, Center for Food and Bioconvergence, Seoul National University, Seoul 08826, Korea*

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#These authors contributed equally to this work.

***Corresponding author**
Hyeun Bum Kim
Department of Animal Resources Science, Dankook University, Cheonan 31116, Korea.
Tel: +82-41-550-3653
E-mail: hbkim@dankook.ac.kr

Ju-Hoon Lee
Department of Food Animal Biotechnology, Department of Agricultural Biotechnology, Center for Food and Bioconvergence, Seoul National University, Seoul 08826, Korea.
Tel: +82-2-880-4854
E-mail: juhlee@snu.ac.kr

Abstract

The *Lactococcus taiwanensis* strain K_LL004 was isolated from the gut of a grasshopper (*Oxya chinensis sinuosa*) collected from local farm in Korea. *L. taiwanensis* strain K_LL004 is the functional probiotic candidate with an ability to hydrolyse plant polysaccharides. The complete genome of the *L. taiwanensis* strain K_LL004 contains one circular chromosome (1,995,099 bp) with a guanine + cytosine (GC) content of 38.8%. Moreover, 1,929 Protein-coding sequence, 19 rRNA genes, and 62 tRNA genes were identified based on results of annotation. *L. taiwanensis* strain K_LL004 has a gene, which encodes hydrolytic enzymes such as beta-glucosidase and beta-xylosidase, that hydrolyzes plant polysaccharides.

Keywords: *Lactococcus taiwanensis*, Grasshopper, Beta-glucosidase, Beta-xylosidase, Whole genome sequencing

Lactococcus is a genus of lactic acid bacteria (LAB) that are present on grass and other plant material and in the gastrointestinal tracts [1]. Twenty-two species of the genus *Lactococcus* are established till date. In particular, *Lactococcus lactis* is the most common strain which is used as a starter in food fermentation [2]. *Lactococcus taiwanensis*, a type of Lactic acid Bacteria, has not been studied in detail, and therefore the genomic information of *Lactococcus taiwanensis* is limited.

In the present study, the *L. taiwanensis* strain K_LL004 was isolated from the gut of a grasshopper (*Oxya chinensis sinuosa*), an insect preferring to feed on plants, collected from local farm in Yangyang, Gangwon-do, Korea. The *L. taiwanensis* strain K_LL004 was grown in de Man-Rogosa-Sharpe

ORCID
Hyunok Doo
https://orcid.org/0000-0003-4329-4128
Hyeri Kim
https://orcid.org/0000-0002-6560-2390
Jin Ho Cho
https://orcid.org/0000-0001-7151-0778
Minho Song
https://orcid.org/0000-0002-4515-5212
Eun Sol Kim
https://orcid.org/0000-0001-8801-421X
Jae Hyoung Cho
https://orcid.org/0000-0002-1128-3451
Sheena Kim
https://orcid.org/0000-0002-5410-1347
Gi Beom Keum
https://orcid.org/0000-0001-6006-9577
Jinok Kwak
https://orcid.org/0000-0003-1217-3569
Sriniwas Pandey
https://orcid.org/0000-0002-6947-3469
Hyeun Bum Kim
https://orcid.org/0000-0003-1366-6090
Ju-Hoon Lee
https://orcid.org/0000-0003-0405-7621

Competing interests
No potential conflict of interest relevant to this article was reported.

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Availability of data and material
Upon reasonable request, the datasets of this study can be available from the corresponding author.

Authors' contributions
Conceptualization: Doo H, Kim HB, Lee JH.
Formal analysis: Kim ES, Cho Jae Hyoung, Kim S, Keum GB.
Methodology: Cho Jin Ho, Song M.
Validation: Kim S, Kwak J, Pandey S.
Writing - original draft: Doo H, Kim H, Cho Jin Ho, Song M.
Writing - review & editing: Doo H, Kim H, Cho Jin Ho, Song M, Kim ES, Cho Jae Hyoung, Kim S, Keum GB, Kwak J, Pandey S, Kim HB, Lee JH.

Ethics approval and consent to participate
This article does not require IRB/IACUC approval because there are no human and animal participants.

broth at 37℃ for 24 h. Genomic DNA was extracted using the MagAttract HMW DNA Kit (QIAGEN, Hilden, Germany), according to the manufacturer's instructions. The complete genome of the *L. taiwanensis* strain K_LL004 was sequenced using the PacBio RS II (Pacific Biosciences, Menlo Park, CA, USA) platform at Insilicogen (Yongin, Korea). Library preparation was performed using SMRTbell™ Template Prep Kit 1.0 following the manufacturer's instructions (Pacific Biosciences). PacBio sequencing produced 161,058 of long reads and 1,143,521,995 base pairs after subreads filtering. *De novo* assemble was performed using the hierarchical genome assembly process (HGAP v2.3.0) workflow and polished using Quiver. The quality of genome assembly was assessed by using Quality Assessment Tool for Genome Assemblies (QUAST) v5.0.2 [3]. The quantitative assessment of the genome completeness was conducted by using Benchmarking Universal Single-Copy Orthologs (BUSCO) v3.0.2 [4]. Protein coding genes, rRNA and tRNA genes of *L. taiwanensis* strain K_LL004 were functionally annotated and predicted through Rapid Annotation using Subsystem Technology (RAST) v2.0 [5]. The functional categorization of all predicted Protein coding genes was performed using Clusters of Orthologous Groups (COG)-based EggNOG-mapper v2 [6]. Potential virulence factors and antibiotic resistance in *L. taiwanensis* strain K_LL004 were predicted using the BLASTn method according to the Virulence Factor Database (VFDB) and the Comprehensive Antibiotic Resistance Database (CARD) [7,8].

The complete genome of the *L. taiwanensis* strain K_LL004 contains one circular chromosome (1,995,099 bp) with a guanine + cytosine (GC) content of 38.8%, 1,929 predicted protein-coding sequence, 19 rRNA genes, and 62 tRNA genes. The genome feature and map of *L. taiwanensis* strain K_LL004 are illustrated in Table 1, Figs. 1A and 1B.

It was confirmed that the *L. taiwanensis* strain K_LL004 has genes which encodes enzymes like beta-glucosidase (EC 3.2.1.21 BG) and beta-xylosidase (EC 3.2.1.37 xyl3), which plays an important role in beta-glycoside metabolism and xylose utilization, respectively. These enzymes are known to hydrolyze the plant cell wall polysaccharides [9]. In addition, the genome of *L. taiwanensis* strain K_LL004 didn't show presence of any virulence factors and antibiotic resistant genes, indicating that *L. taiwanensis* strain K_LL004 can be speculated as a potential probiotic candidate with an ability to hydrolyse plant polysaccharides.

NUCLEOTIDE SEQUENCE ACCESSION NUMBERS

The complete genome sequences of *Lactococcus taiwanensis* K_LL004 were deposited in GeneBank under the accession numbers CP070872.1. The BioSample accession number is SAMN17981207, and BioProject accession number is PRJNA224116.

Table 1. Genome features of *Lactococcus taiwanensis* strain K_LL004

Property	Term
Average genome coverage	449x
Chromosome length (bp)	1,995,099 bp
No. of contig	1 (chromosome)
Guanine + cytosine (G + C) content (%)	38.8
Protein-coding genes	1,929
rRNA genes	19
tRNA genes	62
Genbank Accession No.	CP070872

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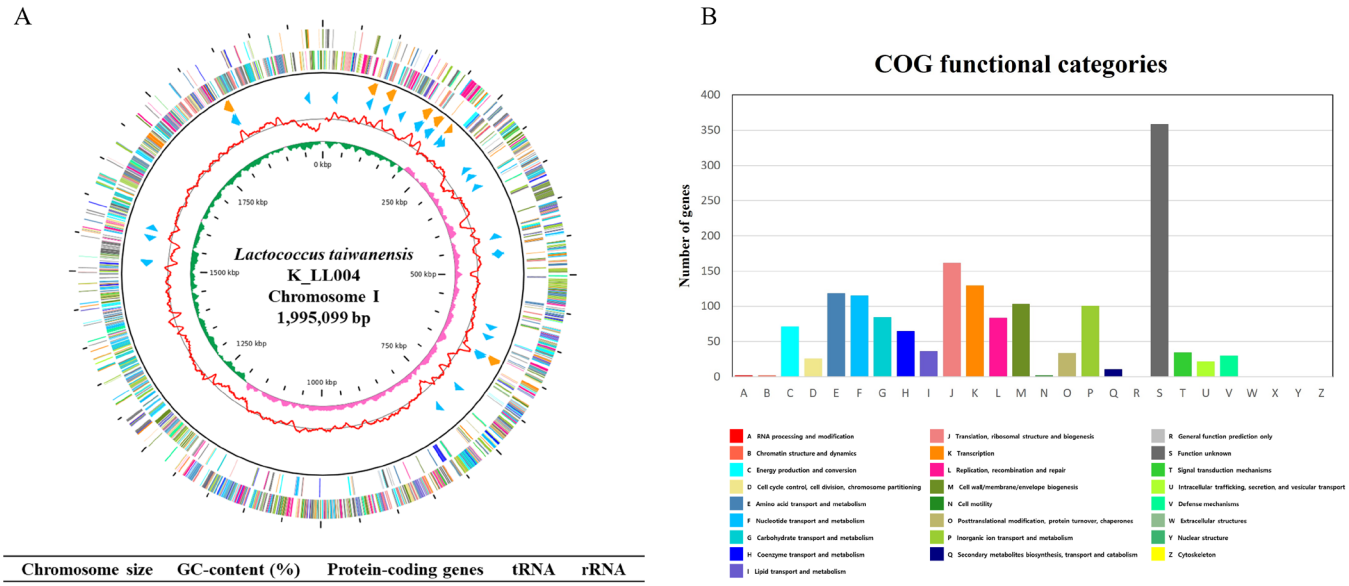

Chromosome size	GC-content (%)	Protein-coding genes	tRNA	rRNA
1,995,099 bp	38.8	1,929	62	19

Fig. 1. Genome map of *Lactococcus taiwanensis* K_LL004 and the functional categorization of predicted protein coding genes. The outer circle denotes the locations of all annotated open reading frames (ORFs), and the inner circle with the red denotes guanine + cytosine (GC) content. Pink, and green peaks denote GC skew. The orange, and sky-blue arrows denote the rRNAs, and tRNA operons, respectively. The annotated ORFs are colored differently based on the Clusters of Orthologous Groups (COG) assignments (A). COG functional categories of predicted protein coding genes (B).

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