

Complete genome sequence of *Limosilactobacillus fermentum* JNU532 as a probiotic candidate for the functional food and feed supplements

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Abstract

Lactic acid bacteria (LAB) have been reported to possess various beneficial properties and are commonly used as probiotics. LAB play a crucial role in milk fermentation, industrial lactic acid fermentation, and health and medicine. *Limosilactobacillus fermentum* isolated from fermented dairy and food products is considered as 'Generally Recognized as Safe' by FDA. *Limosilactobacillus fermentum* plays an important role in modulation of the intestinal microbiota, enhancing the host immune system and improving feed digestibility. We isolated a probiotic candidate that was identified and named *Limosilactobacillus fermentum* JNU532. In a previous report, cell-free culture of *L. fermentum* JNU532 exhibited anti-melanogenic and antioxidant activities. In this study, we present the complete genome assembly of the bacterial strain JNU532. The final genome consists of one circular chromosome (2,077,416 base pairs) with a GC ratio of 51.5%.

Keywords

Limosilactobacillus fermentum, Probiotics, Food, Feed, Supplements.

Recently, the genus *Lactobacillus* was divided into several genera [1], with the species *Lactobacillus fermentum* being assigned to the genus *Limosilactobacillus*. *Limosilactobacillus fermentum* is one of hetero-fermentatives and is used in the fermentation of milk, plants, and silage. *Limosilactobacillus fermentum* strains not only enhance the nutritional value and flavor of food but also its functional properties. This species has strong pH tolerance and good bile tolerance, and it can also reduce cholesterol content in the human body [2]. In addition, *L. fermentum* can inhibit harmful intestinal microbiota, lessen the activity of food allergens, reduce mutagenic and carcinogenic activities, display immunomodulatory activity, and lower cholesterol [3, 4]. *Limosilactobacillus fermentum* JNU532 was isolated from local fermented kimchi in this study. In our previous study, an *L. fermentum* JNU532–derived fermentation broth demonstrated antioxidant properties and the ability to reduce melanin production by inhibiting the activity of tyrosinase in B16F10 melanoma cells. Therefore, *L. fermentum* JNU532 may be considered a natural depigmentation agent. [5].

L. fermentum JNU532 was cultivated in Man-Rogosa-Sharpe (MRS) medium (Becton, Dickinson, France) for 24 h at 37 °C. Genomic DNA was extracted with a HiGene™ Genomic DNA Prep kit (BIOFACT, Daejeon, South Korea), according to the manufacturer's instructions.

The short-read sequencing library was prepared with an Illumina® DNA Prep kit (Illumina, San Diego, USA). Sequencing was performed on the Illumina MiSeq platform (Illumina) using the Illumina MiSeq reagent kit V3 (300 bp, paired end). The long-read sequencing library was prepared using an Oxford Nanopore Ligation Sequencing Kit (Oxford Nanopore, Oxford, UK). Long-read sequencing was carried out on a MinION sequencing device (Oxford Nanopore) equipped with a MinION flow cell (R9.4.1) (Oxford Nanopore). Illumina sequencing produced 1,244,607 paired end reads (433,289,083 bp), while 122,763 reads with an average length of 1,839 bases were obtained from Oxford Nanopore sequencing.

De novo genome sequence assembly was assembled performed using a Flye assembler (v. 2.9) [6] with default options. Adapter sequences from short reads were removed using TrimGalore (v. 0.6.7) [7] with the "paired" parameter. Errors in the draft genome assembly were corrected with Pilon (v. 1.24) [8] with default parameters. Gene prediction of the chromosomal sequence was performed with Prokka (v. 1.14.5) [9]. The completeness of the genome assembly was assessed using BUSCO (v. 5.2.2) [10] with the OrthoDB v10 bacterial (bacteria_odb10) database.

The complete genome sequence of *L. fermentum* JNU532 consisted of one circular chromosome with a GC ratio of 51.5%. A total of 2,113 genes, including 15 rRNA and 57 tRNA, were predicted in the genome (Fig 1). The genomic information of *L. fermentum* JNU532 could provide insight to future research on the characteristics of this strain for functional food and feed supplements.

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DATA AVAILABILITY

The complete genome sequence has been deposited in the NCBI GenBank under the accession number GCA_024800585.1. The BioProject accession number is PRJNA872884 and the BioSample accession number is SAMN30472492.

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126 Table 1. Genome features of *Limosilactobacillus fermentum* JNU532

<i>L. fermentum</i> JNU532	
Total genome length (bp)	2,077,416
GC content (%)	51.5
Depth	96.0
Genome completeness (%)	99.2
Protein-coding genes	2,041
tRNA	57
rRNA	15

127 bp, base pair; G, guanine; C, cytosine; tRNA, transfer RNA; rRNA, ribosomal RNA.

128

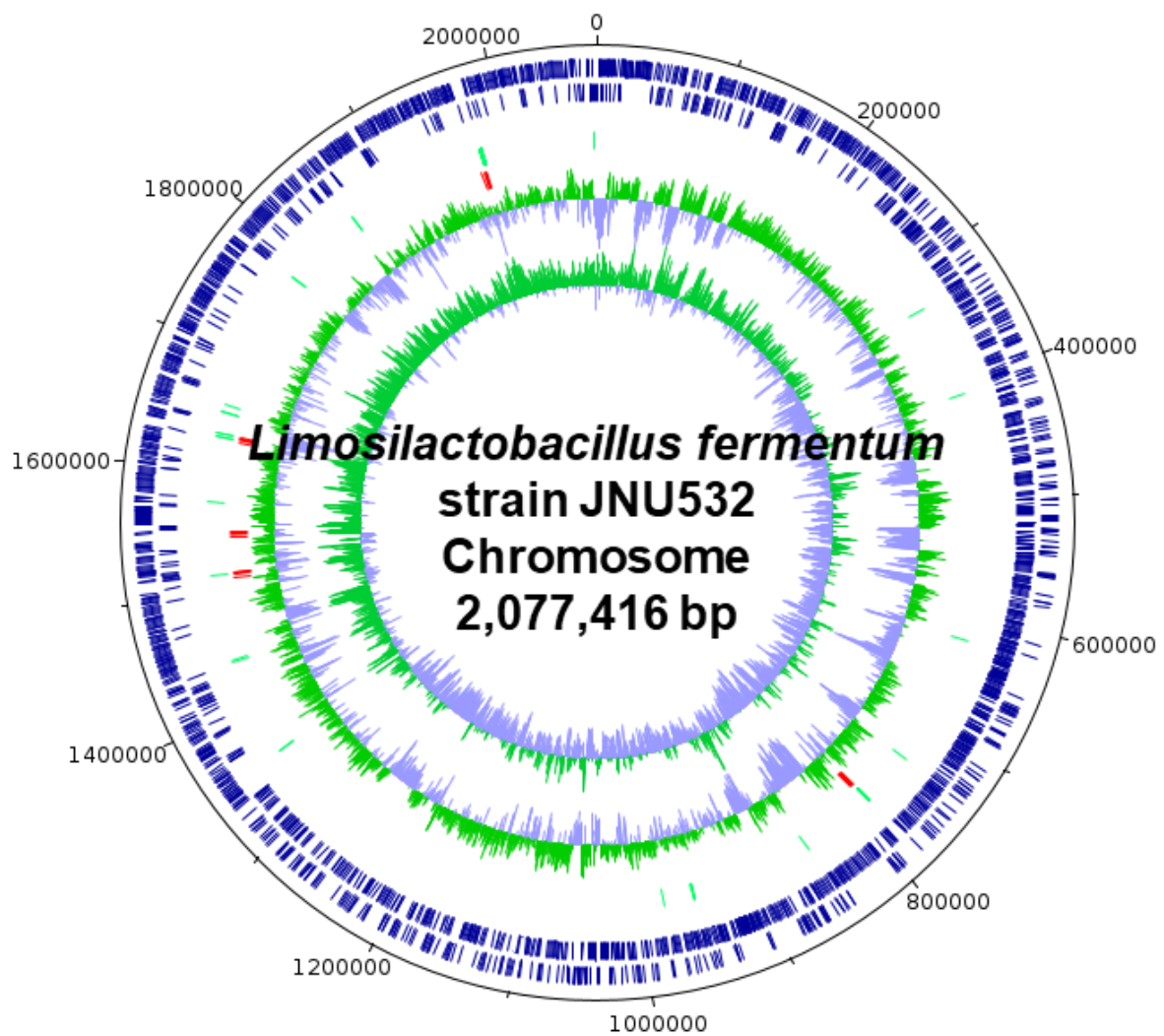


Fig 1. Circular chromosome map of *Limosilactobacillus fermentum* JNU532. From the periphery to the center, marked features are as follows: protein--coding sequences on the forward strand, protein--coding sequences on the reverse strand, tRNA, rRNA, GC ratio, and GC skew. bp, base pair; G, guanine; C, cytosine; tRNA, transfer RNA; rRNA, ribosomal RNA.