JAST (Journal of Animal Science and Technology) TITLE PAGE

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ARTICLE INFORMATION	**Fill in information in each box below**
Article Type	Research article
Article Title (within 20 words without abbreviations)	Complete genome of blaCTX-M-65-carrying *Salmonella* Infantis strain Z01323CSL0015 isolated from broiler chicken
Running Title (within 10 words)	Complete genome of bla$_{CTX-M-65}$-carrying *Salmonella* Infantis Z01323CSL0015
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Competing interests	No potential conflict of interest relevant to this article was reported.
Funding sources State funding sources (grants, funding sources, equipment, and supplies). Include name and number of grant if available.	This study was supported by Korea Disease Control and Prevention Agency (KCDA) (Project no. 2022-ER2103-00)
Acknowledgements	This study was supported by Korea Disease Control and Prevention Agency (KCDA) (Project no. 2022-ER2103-00).
Availability of data and material	Upon reasonable request, the datasets of this study can be available from the corresponding author.
Authors' contributions Please specify the authors' role using this form.	Conceptualization: Oh JY, Chae J-C. Data curation: Jeong JH, Kwak SM. Formal analysis: Jeong JH. Methodology: Jeong JH, Kwak SM. Software: Jeong JH. Validation: Oh JY. Investigation: Oh JY, Jeong JH, Kwak SM, Chae J-C. Writing - original draft: Oh JY. Writing - review & editing: Oh JY, Jeong JH, Kwak SM, Chae J-C.
Ethics approval and consent to participate	This article does not require IRB/IACUC approval because there are no human and animal participants.

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(Research Article)

Complete genome of *bla*$_{\text{CTX-M-65}}$-carrying *Salmonella* Infantis strain Z01323CSL0015 isolated from broiler chicken

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Abstract

Salmonella Infantis strain Z01323CSL0015 was isolated from broiler chickens in Korea. The complete genome of the strain was obtained from the hybrid assembly of sequences generated using Illumina and Oxford Nanopore sequencing platforms. It contains one circular chromosome (4,726,490 bp with a guanine + cytosine (GC) content of 52.3%) and one circular plasmid, pZ01323CSL0015 (310,238 bp with 50.4% GC content). A total of 5,087 protein coding sequences, 22 rRNA genes, and 83 tRNA genes were identified based on the annotation results. The large plasmid contained resistance genes against various antibiotics, such as cephalosporins, aminoglycosides, trimethoprim, sulfonamide, and tetracycline.

Keywords: *Salmonella* Infantis, broiler chicken, large plasmid, antibiotic resistance genes

Salmonella spp. are globally recognized as acute pathogens that cause gastroenteritis in humans [1-4]. Some *Salmonella* serovars have been detected in the food production stage and may colonize poultry and swine farms [2, 3]. *Salmonella* Infantis has emerged as the fourth most prevalent serovar associated with human illness after *S.* Enteritidis, *S.* Typhimurium, and *S.* Typhimurium monophasic variants [4]. *Salmonella* Infantis strain Z01323CSL0015 was isolated from a cloacal swab specimen of a 21-day-old broiler chicken from Korea. This strain was cultivated according to the Food Code (Ministry of Food and Drug Safety, Korea). First, the swab sample was suspended in Rappaport-Vassiliadis Salmonella (RVS) enrichment broth (Difco, USA) and cultivated under aerobic conditions at 42°C for 48 h. The culture was inoculated on Rambach agar (CHROMagar, France) and incubated at 37°C for 24 h. Resulting well-isolated violet colonies were consecutively cultivated on MacConkey agar (Difco, USA). Genomic DNA of the Z01323CSL0015 strain was extracted using the DNeasy Blood and Tissue kit (QIAGEN, Germany) according to the manufacturer's instructions. The genome sequencing of *S.* Infantis strain Z01323CSL0015 was conducted in our laboratory and Macrogen Inc. (Seoul, Korea) for long- and short-read sequencing. A MinION sequencer (Oxford Nanopore Technologies, UK) equipped with a Flow Cell R10 Version (Oxford Nanopore Technologies, UK) was used to generate long-read sequences, whereas short-read sequences were obtained using HiSeq X (Illumina, USA). The Illumina and Nanopore sequencing platforms produced 12,500,648 and 1,248,961 reads corresponding to the sequencing depth of 287.6X and 330.0X, respectively. Short-read raw sequences were processed for trimming adapters/primers and low-quality sequences using Trimmomatic (v. 0.39) [5]. The long-read raw products were treated to trim low-quality sequences with Filtlong (v. 0.2.0) and adapter sequences with Porechop (v. 0.2.4) [5]. Hybrid *de novo* assembly was performed using Unicycler (v. 0.4.9b), followed by polishing with Pilon (v. 1.21) [5]. All the predicted protein coding genes were assigned to the Clusters of Orthologous Groups (COGs) database [6] using COGclassifier version 1.0.5 (https://github.com/moshi4/COGclassifier). Potential virulence factors and antimicrobial resistance genes in the Z01323CSL0015 strain were predicted using the analysis algorithms in the bacterial and viral bioinformatics resource center (BV-BRC) [7]. The complete genome of the Z01323CSL0015 strain consisted of one circular chromosome (4,726,490 bp, 52.3% guanine-cytosine [GC] content) and one plasmid designated as pZ1322SSL0063-1 (310,238 bp, 50.4% GC content) (Table 1 and Fig. 1). The best match among the similar strains based on 16S rRNA gene sequences was *Salmonella enterica* subsp. *enterica* Serovar Infantis strain CVM N17S1509 (GenBank Nos. CP052817 and CP052818) which also contained a mega-plasmid, pN17S1509 [8]. Average nucleotide identity (ANI) value was 99.97% between the genomes. The complete genome of strain Z01323CSL0015 comprised 5,087 protein coding sequences (CDSs) and 105 non-coding genes (22 rRNA and 83 tRNA genes). A total of 3,841 proteins were classified into functional categories based on the COGs database. The most abundant COG categories were carbohydrate transport and metabolism (category G, 396 genes, 10.3%), followed by amino acid transport and metabolism (category E, 356 genes, 9.3%), transcription (category K, 314 genes, 8.2%), energy production and conversion (category C, 290 genes, 7.6%), and cell wall/membrane/envelop biogenesis (category M, 289 genes, 7.5%) (Fig. 2). The plasmid of Z01323CSL0015 strain contained various antimicrobial resistance genes such as *bla*$_{\text{CTX-M-65}}$ (cefotaxime), *aph(4)-la* (atypical aminoglycoside), *aac(3)-V* (gentamicin and netilmicin), *aadA* (streptomycin and spectinomycin), *tetC* (tetracycline), *dfrA14* (trimethoprim), *sul1* (sulfonamide), and *qacEdelta1* (quaternary ammonium compounds). These genes were also found in pN17S1509 mentioned above except for *bla*$_{\text{CTX-M-65}}$. *Salmonella* Infantis carrying multidrug-resistant pESI (plasmid for emerging *S*. Infantis)-like mega plasmids have been reported in many countries [2]. In a view of horizontal gene transfer, the plasmid of Z01323CSL0015 could be disseminated among the serotypes conferring antibiotic resistance. Therefore, our genomic information might be useful for the surveillance of plasmid-mediated antibiotic resistome.

Acknowledgments

This study was supported by Korea Disease Control and Prevention Agency (2022-ER2103-00).

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Nucleotide sequence accession numbers

The completed genome sequences of *Salmonella enterica* serovar Infantis strain Z01323CSL0015 were deposited at the NCBI GenBank under the accession numbers, CP133181 (chromosome) and CP133182 (plasmid).

Competing interests

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

Funding sources

This study was supported by Korea Disease Control and Prevention Agency (KCDA) (Project no. 2022-ER2103-00).

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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: Oh JY, Chae J-C.
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Software: Jeong JH.
Validation: Oh JY.
Investigation: Oh JY, Jeong JH, Kwak SM, Chae J-C.
Writing - original draft: Oh JY.
Writing - review & editing: Oh JY, Jeong JH, Kwak SM, Chae J-C.

Ethics approval and consent to participate

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Figure legends

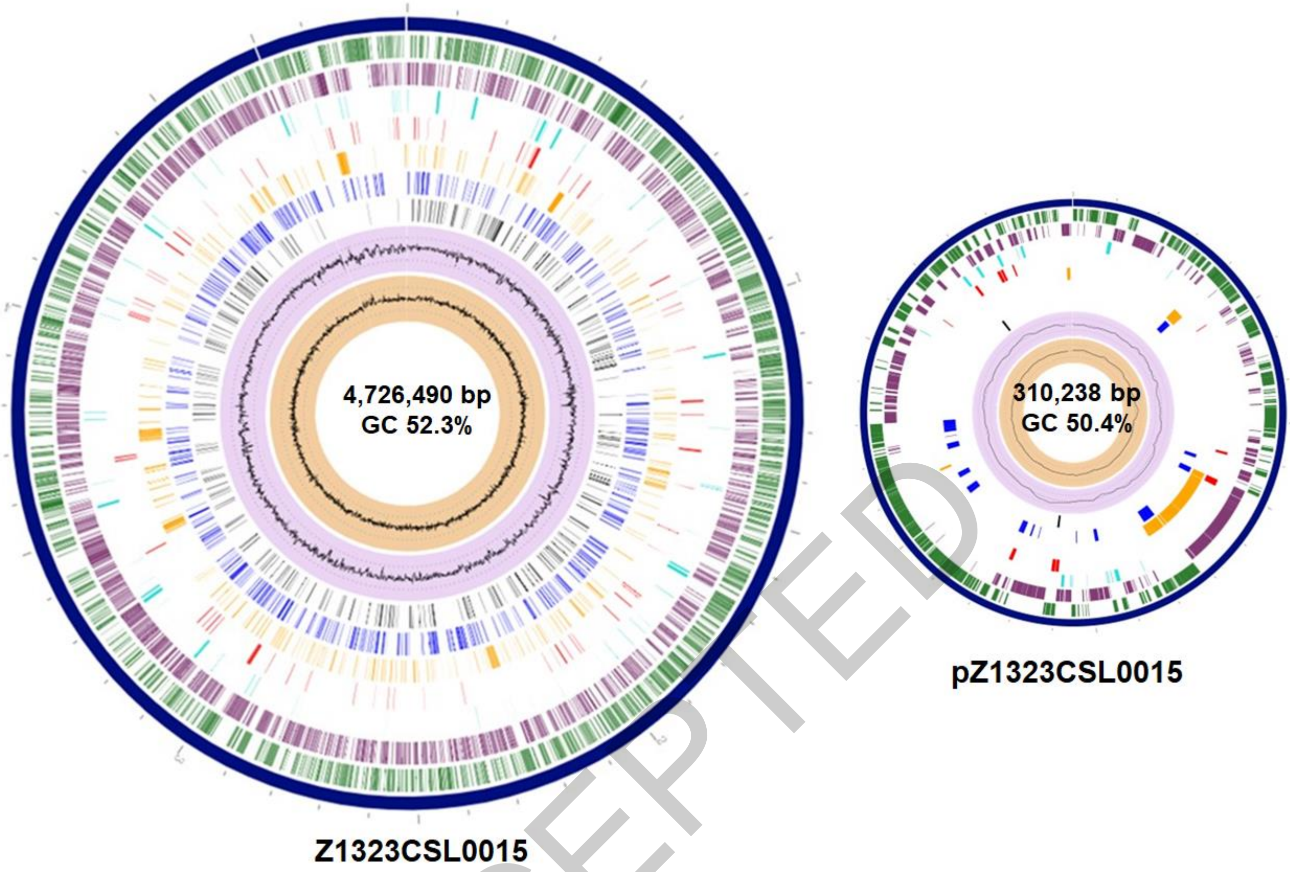

Fig. 1. The circular graphical genomic map of *Salmonella* Infantis strain Z01323CSL0015. Circles from the outside to the center denote green color, the coding sequences (CDS) on the forward strand; violet, CDS on the reverse strand; red, antimicrobial resistance (AMR) genes; reddish brown, virulence factor (VF) genes; blue, transporters; black, drug targets; black on a light purple background, GC content; black on apricot background, GC skew.

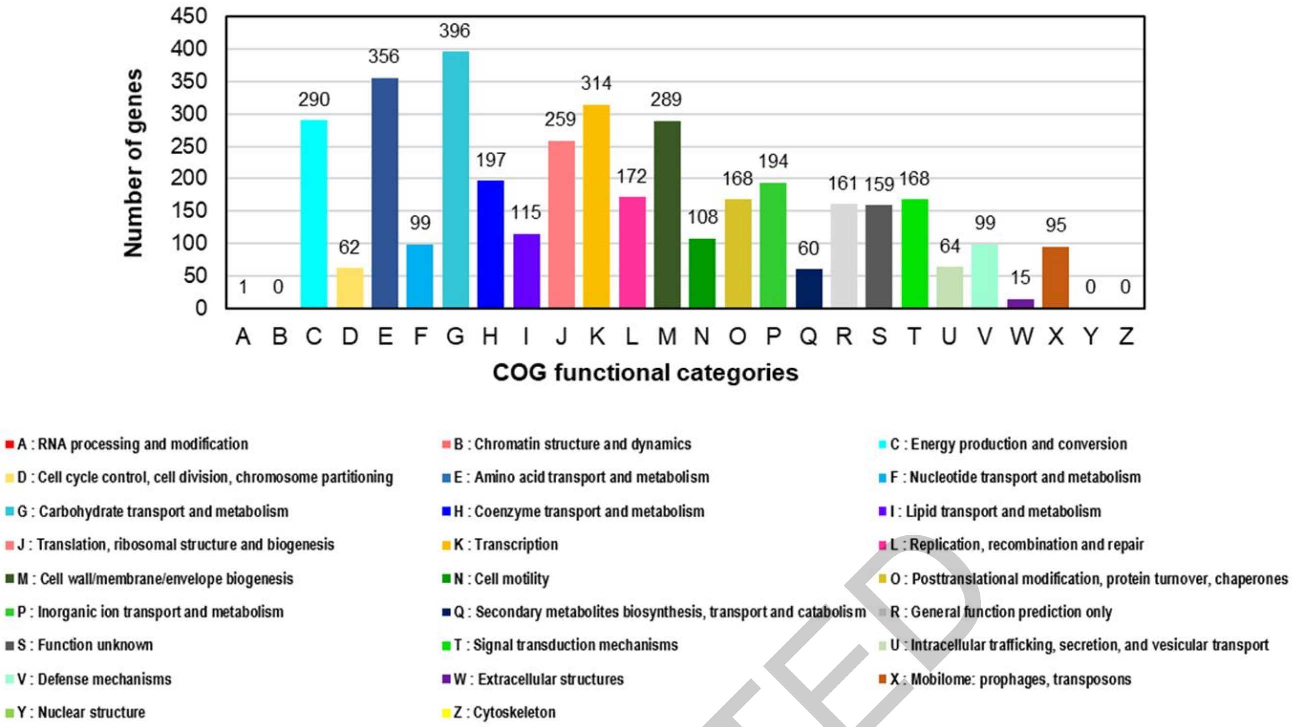

Fig. 2. COG (Cluster of Orthologous Group) functional categories of predicted protein coding sequences in the genome of *Salmonella* Infantis strain Z01323CSL0015.

Table 1. Genome features of *Salmonella enterica* serovar Infantis strain Z01323CSL0015

Properties	Chromosome	Plasmid
	Z1323CSL0015	pZ1323CSL0015
Accession no.	CP133181	CP133182
Genome size (bp)	4,726,490	310,238
GC content (%)	52.3	50.4
No. of CDSs	4,671	416
rRNA genes	22	ND
tRNA genes	82	1

ND, not detected.