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GENOME SEQUENCES

Complete Genome Sequence of *Escherichia coli* Strain FEX669, a ColV Plasmid-Containing Isolate from Retail Chicken Meat

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ABSTRACT *Escherichia coli* strain FEX669 was isolated from retail ground chicken and shown to contain the extraintestinal pathogenic *E. coli* (ExPEC) virulence genes *sfaD, focC*, and *iutA*. Because this presumptive ExPEC strain was isolated from a retail food item and it was a weak biofilm former, it was characterized using whole-genome sequencing using the PacBio RS II platform. Genomic analysis showed that the FEX669 chromosome is 4,973,943 bp long, with a GC content of 50.47%, and is accompanied by a ColV plasmid that is 237,102 bp long, with a GC content of 50.49%.

Foodborne extraintestinal pathogenic *Escherichia coli* (ExPEC) represents an underappreciated public health threat. Millions of ExPEC infections occur annually in the United States, producing diseases ranging from urinary tract infections (UTIs) to acute sepsis (1). It is well established that ExPEC strains colonize livestock and contaminate food products. ExPEC strains isolated from food and human patient samples were screened for biofilm production via a microtiter plate assay (2). Here, the *E. coli* strain FEX669, an isolate with scant (8 times less than the positive control) biofilm production, was selected for whole-genome sequencing (Fig. 1). This strain was originally isolated from ground chicken in Minneapolis, Minnesota, in May 2002 through the survey of 1,648 diverse food items from 10 retail markets. In addition, its ExPEC-associated features were identified through PCR-based assays and O serotyping (3).

Genomic DNA (gDNA) was isolated from overnight cultures grown in LB broth at 37°C, with shaking at 180 rpm, by using the Genomic-tip 500/G kit (Qiagen, Valencia, CA). The gDNA concentration was determined using the Qubit 2.0 instrument (Invitrogen, Carlsbad, CA), was sheared with a g-TUBE (Covaris, Inc., Woburn, MA), and underwent size selection for 20-kb fragments using the BluePippin kit (Sage Science, Inc., Beverly, MA). The sequencing library was prepared using the SMRTbell library preparation kit (Pacific Biosciences, Menlo Park, CA) and was sequenced on an RS II machine with P6C4 chemistry at the University of Delaware Sequencing Center (Newark, DE). The sequencing generated 150,292 reads, with 1,519,409,874 bases. For all software analyses, default parameters were used except where otherwise noted. SMRT Portal v2.3.0 of SMRT Link v7.0 was used for quality control and filtering, which resulted in 75,495 reads, 1,316,564,288 bases, a read N_{50} value of 24,304 bases, and a mean read length of 17,439 bases. The post-filtered reads were used for genome *de novo* assembly using Hierarchical Genome Assembly version 3 (HGAP3) (Pacific Biosciences). The genome was polished using Quiver software (4). Benchmarking universal single-copy orthologues (BUSCO) and blast analysis were used to assess the quality and completeness of the assembled genome. The final assembly included one circular, 4,973,943-bp chromosome and one circular, 237,102-bp plasmid.

We annotated the genome using NCBI Prokaryotic Genome Annotation Pipeline

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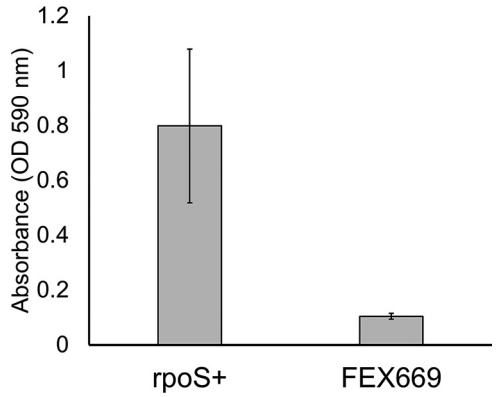

FIG 1 Biofilm production by *Escherichia coli* strain FEX669. Strain 43894OR (OR), a constitutive *rpoS*-producing (rpoS+) strain, was used as a positive control for biofilm (10). Biofilms were stained with crystal violet as previously described (2). Bars represent the mean crystal violet absorbance measured at an optical density at 590 nm (OD_{590}) ± standard deviation (SD) of three independent samples.

(PGAP) (5) and identified 5,013 genes in the genome and 117 RNA sequences, including 85 tRNA genes. PHASTER (6) identified four phages on the chromosome and one on the plasmid. The SerotypeFinder-2.0 (7) and MLST-2.0 databases from the Center for Genomic Epidemiology (https://cge.cbs.dtu.dk/services/) assigned FEX669 to serotype O150:H6, sequence type 6570 (ST6570), and phylogroup B2. ResFinder-3.2 (8) found tetracycline and ampicillin resistance genes on the plasmid and no resistance genes on the chromosome.

A total of 27 virulence genes were identified on the FEX669 chromosome and plasmid using VirulenceFinder-2.0 software (with 90% threshold and 60% minimum length parameters) (9) (accessed 2 November 2020). They included genes for iron acquisition (*chuA*, *fyuA*, *iroN*, *sitA*, *iucC*, *irp2*, and *iutA*), attachment (*hra, focC*, *sfaD*, and *yfcV*), and toxin production (*hlyF, vat*, and *usp*). According to BLAST analysis, the plasmid was highly similar (83% coverage; 99.90% identity) to a ColV plasmid from *E. coli* strain PU-1, a blood isolate from a piglet with acute sepsis (GenBank accession number CP042245.1).

The food product origin of strain FEX669 makes its possession of virulence genes that could facilitate extraintestinal and even systemic infection a cause for concern. Genomic sequencing and characterization of potentially pathogenic *E. coli* are important tools for identifying hazards in the food supply and preventing human illness outbreaks.

Data availability. This whole-genome project has been deposited under BioProject number PRJNA642167, Sequence Read Archive (SRA) number SRP293948, and BioSample number SAMN15822731 with the DDBJ/ENA/GenBank accession numbers CP065152.1 for the FEX669 chromosome and CP065153 for the plasmid.

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