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GENOME SEQUENCES

Whole-Genome Sequencing of *Salmonella enterica* Serovar Infantis and Kentucky Isolates Obtained from Layer Poultry Farms in Ecuador

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ABSTRACT Five strains of *Salmonella enterica* subsp. *enterica* serovar Infantis and two strains of *S. enterica* subsp. *enterica* serovar Kentucky isolated in 2017 from Ecuadorian layer poultry farms were sequenced using Illumina MiSeq technology. These isolates were collected on layer farms in central Ecuador, one of the most important areas of egg production in the country. The genome sequences of these isolates show valuable information for surveillance purposes.

Nontyphoidal *Salmonella* (NTS) represents one of the leading causes of foodborne gastroenteritis worldwide (1, 2). The global burden of NTS infections is 93.8 million cases, and it is estimated that NTS causes around 155,000 deaths annually (3). Unfortunately, there is a lack of information about surveillance studies of NTS in Latin American countries, and reports on NTS outbreaks are scarce in this region (4). In South American countries, there has been an increase in reports of multidrug-resistant *Salmonella enterica* subsp. *enterica* serovar Infantis in clinical, food, and livestock-related samples. That is the case in Peru (5, 6), Brazil (7), and Ecuador, where reports of NTS infections are related to extended-spectrum β-lactamase (ESBL)-producing *S.* Infantis (8). In Ecuador, this serovar represents the most commonly reported serovar in poultry-related samples (9, 10).

The importance of *S.* Infantis in the epidemiology of nontyphoidal salmonellosis has been described lately in Europe and the United States (2, 11). In the United States, infections caused by CTX-M-65 *S.* Infantis have been linked to travelers to South America (Peru and Ecuador) (12), suggesting that this serotype could play an essential role in the epidemiology of human salmonellosis. No reports of *Salmonella enterica* subsp. *enterica* serovar Kentucky have been published in Ecuador so far (according to the Scopus and Web of Science databases). In this report, we present the genome sequences of five recent strains of *S.* Infantis and two strains of *S.* Kentucky isolated from layer poultry farms in central Ecuador.

From August 2017 to December 2017, an aleatory sampling program in layer farms was carried out as part of routine diagnostics. Briefly, 102 samples from 21 farms in central Ecuador were analyzed by the ISO-6579 method. The samples were preenriched using buffered peptone water (Merck Millipore, Darmstadt, Germany) and incubated at 37°C for 18 to 24 h. Subsequently, the preenrichments were selectively enriched using Rappaport-Vassiliadis broth (Becton, Dickinson, Le Pont de Claix, France) at 41.5°C for 24 h. Next, the selective enrichment was streaked onto xylose lysine deoxycholate (XLD) agar (Merck Millipore) and incubated at 37°C for 24 h. The presumptive colonies were purified in MacConkey agar (Merck Millipore) and incubated at 37°C for 24 h. Biochem-

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TABLE 1 Whole-genome sequencing-based characterization of 7 nontyphoidal *Salmonella enterica* strains isolated from layer poultry farms in Ecuador

Laboratory identifier	Sample type	Predicted *S. enterica* subsp. *enterica* serovar	No. of reads	No. of contigs	Total length (bp)	N_{50} (bp)	GC content (%)	No. of genes	CheckM quality (% completeness [% contamination])	Genome coverage (×)	MLST	Plasmid replicon	Pathogenicity islands	GenBank accession no.	SRA accession no.
U822s	Feed	Infantis	923,027	279	4,970,844	34,389	52.37	5,087	100 (0.08)	71	32		SPI-13, SPI-14, SPI-5	AACYLG000000000	SRR8266241
U824s	Feed	Infantis	663,871	322	4,926,207	29,447	52.35	5,051	100 (0.08)	52	32		SPI-13, SPI-14, SPI-5, SPI-1, C63PI	AADKME000000000	SRR8282892
U825s	Feed	Infantis	1,173,443	220	4,990,278	47,232	52.29	5,100	100 (0.08)	103	32		SPI-13, SPI-14, SPI-5, SPI-1, C63PI	AADPVC000000000	SRR8282855
U827s	Environmental swabs	Kentucky	1,308,655	285	4,744,299	40,935	52.18	4,793	100 (0.39)	99	152	IncX1	SPI-8, C63PI	AADPVF000000000	SRR8282852
U828s	Environmental swabs	Kentucky	926,830	192	4,751,148	52,705	52.15	4,788	100 (0.39)	83	152	IncX1	SPI-8, C63PI	AADFDT000000000	SRR8282866
U842s	Environmental swabs	Infantis	1,163,477	179	5,001,560	52,646	52.22	5,089	100 (0.08)	83	32		SPI-13, SPI-14, C63PI, SPI-5	AACYLE000000000	SRR8266239
U845s	Cloacal swabs	Infantis	760,589	298	4,948,982	30,021	52.34	5,064	100 (0.08)	62	32		SPI-13, SPI-14, C63PI, SPI-3, SPI-5	AADKMC000000000	SRR8282870

ical tests were performed to confirm the genus *Salmonella* (catalase, oxidase, triple sugar iron [TSI], Simmons citrate, Christensen urea agar base, indole reaction). The positive samples were cryopreserved using overnight growth in LB broth (Sigma-Aldrich, St. Louis, USA) supplemented with 30% glycerol (Merck Millipore).

Thirty-one isolates were identified as belonging to the genus *Salmonella*. Seven isolates were selected according to their sampling origin (strains U822s, U824s, and U842s, Latacunga; U825s, Quero; U845s, Ambato; U827s, Pelileo; and U828s, Cevallos). Isolates were recovered from poultry feeders, environmental swabs, and cloacal swabs (Table 1). *Salmonella* cultures were grown in LB broth and incubated at 37°C for 24 h. The biomass was used for genomic DNA extraction using the Wizard genomic DNA purification kit (Promega, Madison, WI, USA) according to the manufacturer's instructions and DNA quantified using the Qubit 3.0 fluorometer (Invitrogen, CA, USA). Genomic libraries were prepared using the Nextera XT library prep kit (Illumina, CA, USA), and whole-genome sequencing (WGS) was performed using 300-bp paired-end read lengths on the MiSeq platform (Illumina). Raw reads were *de novo* assembled via SKESA v2.2 using the NCBI Prokaryotic Genome Annotation Pipeline (13). The draft genome assembly quality was evaluated using CheckM v1.0.18 (14) and QUAST v4.4 (15).

Draft genomes were analyzed using the tools from the Center for Genomic Epidemiology of the Technical University of Denmark. Using WGS data, the serotypes were determined using SeqSero v1.2 (16). Multilocus sequence types (MLSTs) were determined using MLST v2.0 (17). The presence of plasmids was identified using PlasmidFinder v2.1 (18). The prediction of bacterial pathogenicity was evaluated using PathogenFinder v1.1 (19). Finally, the detection of *Salmonella* pathogenicity islands was carried out using SPIFinder v1.0 (20). All analyses were performed using default parameters.

The number of contigs ranged from 179 to 322, and their N_{50} values ranged from 29,447 to 52,705 bp. The draft genomes ranged from 4,744,299 to 5,001,560 bp, with 52.27% average GC content (Table 1). The assemblages showed 100% completeness in all isolates, and the percentage of contamination ranged from 0.08% to 0.39%. The number of annotated genes ranged from 4,788 to 5,100. The SeqSero analysis revealed that the isolates belonged to the *S. enterica* subsp. *enterica* serovars Infantis (5) and Kentucky (2). MLST analysis showed that the *S.* Infantis strains belonged to sequence type 32 (ST32) and the *S.* Kentucky strains to ST152. The plasmid replicon typing detected the plasmid incompatibility group IncX1 in the *S.* Kentucky strains only (Table 1).

These draft genomes provide essential information about the circulating clones of *Salmonella* from layer poultry farms in Ecuador and can serve as references for genome comparison studies.

Data availability. The draft genome sequences reported in this article were deposited in GenBank under the accession numbers AACYLG000000000, AADKME000000000, AADPVC000000000, AADPVF000000000, AADFDT000000000, AACYLE000000000, and AADKMC000000000.

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