



# Draft Genome Sequences of Seven Strains of Shiga Toxin-Producing *Escherichia coli* O111 with Variation in Their Sensitivity to Novobiocin

Luca Rotundo,<sup>a</sup> Federica Boccia,<sup>b</sup> Pina M. Fratamico,<sup>a</sup> Aixia Xu,<sup>a</sup> Christopher H. Sommers,<sup>a</sup> Yanhong Liu,<sup>a</sup> James L. Bono,<sup>c</sup> Tiziana Pepe<sup>b</sup>

<sup>a</sup>USDA, Agricultural Research Service, Eastern Regional Research Center, Wyndmoor, Pennsylvania, USA

<sup>b</sup>Department of Veterinary Medicine and Animal Production, University of Naples Federico II, Naples, Italy

<sup>c</sup>USDA, Agricultural Research Service, U.S. Meat Animal Research Center, Clay Center, Nebraska, USA

**ABSTRACT** Inclusion of novobiocin as a selective agent for enrichment media and selective agars inhibits the growth of some Shiga toxin-producing *Escherichia coli* (STEC) strains, particularly non-O157 STEC, which can yield false-negative detection results. Here, we report the draft genomic sequences of seven STEC O111 isolates with different sensitivities to novobiocin.

Shiga toxin-producing *Escherichia coli* (STEC) strains are responsible for serious outbreaks and sporadic cases of foodborne illness worldwide. Diseases associated with STEC infection include gastroenteritis, hemorrhagic colitis, and hemolytic uremic syndrome, a life-threatening complication that occurs primarily in children, the elderly, and immunocompromised individuals (1). Serotype O157:H7 and serogroups O26, O103, O111, and O145 are the most epidemiologically relevant forms of STEC implicated in causing severe human illness (2) due to their low infectious dose and high virulence. The genes encoding the Shiga toxins (*stx*<sub>1</sub> and *stx*<sub>2</sub>), as well as the *eae* gene encoding the intimin protein, are important STEC virulence factors.

To ensure consumer safety, methods have been reported for the detection of STEC in food (3–7). They rely on use of an enrichment step, using a selective broth to allow growth and improved recovery of the target STEC. The ISO13136:2012 method includes the use of novobiocin as a supplement in the enrichment broth due to its antimicrobial effect against Gram-positive bacteria (8). However, the growth of some serogroups, including O111, could be inhibited due to their sensitivity to novobiocin (9–11), which would result in false-negative detection results. Therefore, the genomes of seven STEC O111 strains isolated from humans and cattle (Table 1), previously shown to have variation in resistance to novobiocin (11), were sequenced. This information is important for understanding the characteristics of this pathogen at the molecular level, as well as for development of enrichment media and selective agars that will allow improved detection of STEC O111.

STEC O111 strains (Table 1) were grown in tryptic soy broth (TSB; Becton, Dickinson and Company, Sparks, MD, USA) at 37°C for 18 h, and genomic DNA was extracted using a DNeasy blood and tissue kit (Qiagen, Valencia, CA, USA) and a QIAcube instrument (Qiagen). The Nextera DNA Flex library prep kit (Illumina, San Diego, CA, USA) was used for library preparation, and the library concentration was assessed using a Qubit 3.0 fluorometer (Life Technologies, Carlsbad, CA, USA). The libraries were denatured and loaded onto a flow cell (for cluster generation), and sequencing was performed using the Illumina MiniSeq platform with a 2 × 150-bp paired-end read protocol with approximately 100× coverage. BaseSpace Software (version 2.0.2018) found on the Illumina webpage was used to access the application for analysis of the raw data. This included FastQC (version 1.0.0) to improve the

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Address correspondence to Yanhong Liu, [yanhong.liu@ars.usda.gov](mailto:yanhong.liu@ars.usda.gov).

**TABLE 1** Characteristics and accession numbers of seven *E. coli* serogroup O111 strains

Characteristic	Data for strain:						
	00-4748	98-8338	TB226A	95-0586	3007-85	CL-37	7-14 10A
GenBank accession no.	QMHY00000000	QMHZ00000000	QMIA00000000	QMIB00000000	QMIC00000000	QMHX00000000	QMHW00000000
SRA no. <sup>a</sup>	SRP155839	SRP155841	SRP155842	SRP155848	SRP155849	SRP155838	SRP155833
Genome coverage (×) <sup>b</sup>	76	100	88	94	82	81	100
Genome size (bp) <sup>b</sup>	4,966,944	5,288,007	5,429,255	5,242,733	5,179,065	5,281,527	5,212,079
GC content (%) <sup>c</sup>	50.50	50.40	50.40	50.40	50.40	50.40	50.40
<i>In silico</i> serotype <sup>d</sup>	O111:H8	O111:H8	O111:H8	O111:H8	O111:H8	O111:H8	O111:H8
Previously reported serotype	O111:NM	O111:NM	O111:H-	O111:H-	O111:NM	O111:H8	O111:H8
Shiga toxin variant(s) <sup>e</sup>	stx <sub>2a</sub> ; stx <sub>2b</sub>	stx <sub>1a</sub> ; stx <sub>1b</sub>	stx <sub>1a</sub> ; stx <sub>1b</sub> ; stx <sub>2a</sub> ; stx <sub>2b</sub>	stx <sub>1a</sub> ; stx <sub>1b</sub>	stx <sub>1a</sub> ; stx <sub>1b</sub> ; stx <sub>2a</sub> ; stx <sub>2b</sub>	stx <sub>1a</sub> ; stx <sub>1b</sub>	stx <sub>1a</sub> ; stx <sub>1b</sub>
<i>eae</i> gene <sup>e</sup>	Positive	Positive	Positive	Positive	Positive	Positive	Positive
No. of contigs <sup>b</sup>	254	295	333	316	300	305	279
Total no. of genes <sup>b</sup>	5,410	5,882	6,099	5,788	5,730	5,853	5,797
No. of pseudogenes <sup>b</sup>	316	343	353	318	339	356	331
<i>N</i> <sub>50</sub> <sup>f</sup>	105,049	99,577	99,577	99,880	104,026	104,100	104,239
No. of CDSs <sup>b</sup>	5,295	5,769	5,979	5,668	5,612	5,736	5,673
No. of rRNAs <sup>b</sup>	10	12	11	11	11	10	10
No. of tRNAs <sup>b</sup>	93	89	97	97	95	95	102
No. of ncRNAs <sup>b</sup>	12	12	12	12	12	12	12
MLST <sup>g</sup>	ST16; ST480	ST16; ST480	ST16; ST480	ST16; ST480	ST16; ST480	ST16; ST480	ST16; ST480
Resistance gene(s) <sup>h</sup>	None found	<i>aac(3)-IId</i> ; <i>aadA5</i> ; <i>sul1</i> ; <i>dfrA17</i> ; <i>bla</i> <sub>TEM-1B1</sub> ; <i>catA1</i>	<i>aph(3)-Ia</i> ; <i>aadA1</i> ; <i>sul1</i> ; <i>tet(A)</i>	None found	None found	<i>aph(3)-Ia</i> ; <i>strA</i> ; <i>strB</i> ; <i>aph(6)-Ic</i>	None found
Origin	Human	Human	Human	Human	Human	Human	Cattle
Source or reference	17	17	18	Laboratory Centre for Disease Control, Canada	CDC, STEC, Nebraska	STEC Center, Canada	Nebraska

<sup>a</sup>Sequence Read Archive (SRA) accession number.

<sup>b</sup>Genome annotation was performed via the NCBI Prokaryotic Genome Annotation Pipeline (12). CDSs, coding sequences; ncRNAs, noncoding RNAs.

<sup>c</sup>The GC content (%) was obtained using the Genome Assembler and Annotation Report.

<sup>d</sup>The *in silico* serotype was obtained using *E. coli* Serotyping version 1.0.0 (13).

<sup>e</sup>Virulence genes and Shiga toxin subtype were determined using VirulenceFinder (14).

<sup>f</sup>The *N*<sub>50</sub> value is the size of the shortest contig in the set of longest contigs that together cover at least 50% of the total genome size.

<sup>g</sup>Multilocus sequence typing (MLST) information was obtained using the Bacterial Analysis Pipeline (15).

<sup>h</sup>Antimicrobial resistance genes were identified using the Bacterial Analysis Pipeline (16).

quality of the raw data, the SPAdes genome assembler (version 3.9.0), and the Bacterial Analysis Pipeline (version 1.0.4). Genome annotation was performed via the NCBI Prokaryotic Genome Annotation Pipeline (12). Strains used, genome size, GC content, coverage of the sequenced strains, serotype identification, virulence genes, antimicrobial resistance genes, multilocus sequencing typing (sequence types), and other characteristics are listed in Table 1.

**Data availability.** This whole-genome shotgun project has been deposited at DDBJ/ENA/GenBank under the accession numbers listed in Table 1. The versions described in this paper are the first versions.

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