

Certificate of Analysis for NR-2648

Genomic DNA from Escherichia coli, Strain EDL933

Catalog No. NR-2648

Product Description: Genomic DNA was isolated from a preparation of *Escherichia coli* (*E. coli*), strain EDL933, serotype O157:H7. The enterohemorrhagic *E. coli* (EHEC) strain EDL933 was isolated from raw hamburger meat implicated in a multi-state outbreak of hemorrhagic colitis in the United States in 1982.

Lot¹: 61740159 Manufacturing Date: 13MAY2013

TEST	SPECIFICATIONS	RESULTS
Genotypic Analysis Sequencing of 16S ribosomal RNA gene (~ 1490 base pairs)	Consistent with E. coli	Consistent with E. coli ^{2,3}
PCR Assay of Extracted DNA 16S ribosomal RNA gene PCR amplification of plasmid markers ^{4,5}	~ 1500 base pair amplicon	~ 1500 base pair amplicon
hylA (pO157) elt (pJY11) esth (pCS1) estp (pCS1) EAF (pEAF)	Positive Negative Negative Negative Negative	Positive Negative Negative Negative Negative
bfpA (pEAF) invE (pINV) CVD432 (pAA) aggR (pAA) PCR amplification of chromosomal markers ^{4,5}	Negative Negative Negative Negative	Negative Negative Negative Negative
eaeA stx1 stx2 astA	Positive Report results Report results Report results	Positive Positive Positive Negative
Agarose Gel Electrophoresis	High molecular weight chromosomal DNA	High molecular weight chromosomal DNA (Figure 1)
Concentration by PicoGreen® Measurement	0.7 to 1.5 μg in 25 to 100 μL per vial	0.9 μg in 25 μL per vial (0.04 μg/μL)
OD ₂₆₀ /OD ₂₈₀ Ratio	1.7 to 2.0	2.0
Bacterial Inactivation 10% of total yield plated on Tryptic Soy agar with 5% defibrinated sheep blood ^{6,7}	No viable bacteria detected	No viable bacteria detected

¹The bacterial preparation used for extraction of genomic DNA was produced by inoculation of BEI Resources NR-11 lot 3560110. Genomic DNA was extracted using proprietary technology.

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²Also consistent with *Shigella* species

^{3≥ 99%} identical to GenBank: AE005174.2 (E. coli, strain EDL933, serotype O157:H7)

⁴PCR was completed on DNA extracted from NR-11 lot 3560110 for NR-2648 lot 5107305.

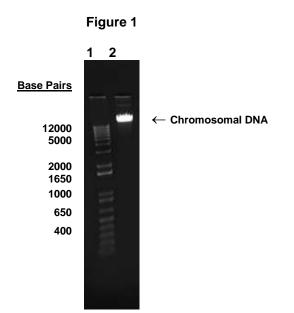
⁵Kimata, K., et al. "Rapid Categorization of Pathogenic *Escherichia coli* by Multiplex PCR." <u>Microbiol. Immunol.</u> 49 (2005): 485-492. PubMed: 15965295.

⁶⁷ days at 37°C and aerobic atmosphere

⁷An extraction procedure was used that has been shown to consistently inactivate 100% of Gram-positive and Gram-negative bacteria.



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Lane 1: Invitrogen™ 1Kb DNA Ladder™

Lane 2: 200 ng of NR-2648

Date: 22 JAN 2014

Signature: (

Title: Technical Manager, BEI Authentication or designee

ATCC®, on behalf of BEI Resources, hereby represents and warrants that the material provided under this certificate has been subjected to the tests and procedures specified and that the results described, along with any other data provided in this certificate, are true and accurate to the best of ATCC®'s knowledge.

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