

Certificate of Analysis for NR-2647

Genomic DNA from Escherichia coli, Strain H10407

Catalog No. NR-2647

Product Description: Genomic DNA was isolated from a preparation of enterotoxigenic *Escherichia coli* (ETEC), strain H10407, serotype O78:H11. The bacterial preparation was produced by propagation of BEI Resources NR-4.

Lot¹: 5107284 Manufacturing Date: 22FEB2006

TEST	SPECIFICATIONS	RESULTS
Genotypic Analysis		
Sequencing of 16S rRNA gene (~ 500 bp)	Identical to NR-4	Identical to NR-4
	Consistent with Escherichia coli	Consistent with Escherichia coli ²
PCR amplification of plasmid markers ³		
elt (pJY11)	Report results	Positive
esth (pCS1)	Report results	Positive
estp (pCS1)	Report results	Positive
EAF (pEAF)	Negative	Negative
bfpA (pEAF)	Negative	Negative
invE (pINV)	Negative	Negative
CVD432 (pAA)	Negative	Negative
aggR (pAA)	Negative	Negative
PCR amplification of chromosomal markers ³		
eaeA	Negative	Negative
stx1	Negative	Negative
stx2	Negative	Negative
astA	Report results	Positive
Agarose Gel Electrophoresis	High molecular weight chromosomal	High molecular weight chromosoma
	DNA	DNA (Figure 1)
Concentration by PicoGreen® Measurement	4 to 6 μg in 25 to 100 μL per vial	4.8 μg in 62 μL per vial (77 μg/mL)
Functional Activity by PCR Amplification		
16S ribosomal RNA gene	~ 1500 bp amplicon	~ 1500 bp amplicon
Virulence markers ³	The second secon	
elt (pJY11)	~ 263 bp amplicon	~ 263 bp amplicon
esth (pCS1)	~ 179 bp amplicon	~ 179 bp amplicon
estp (pCS1)	~ 123 bp amplicon	~ 123 bp amplicon
astA (chromosome)	~ 106 bp amplicon	~ 106 bp amplicon
OD ₂₆₀ /OD ₂₈₀ Ratio	1.7 to 1.9	1.8
Bacterial Inactivation		
10% of total yield plated on Nutrient Agar and		
incubated for 7 days at 37°C	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-4 (Lot 3561329) into LB Broth (ATCC® 60-2100). After incubation for 16 hours at 37°C, genomic DNA was extracted using proprietary technology.

²Also consistent with *Shigella* species.

Fax: 703-365-2898

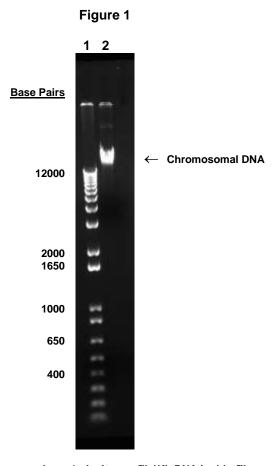
E-mail: contact@beiresources.org

800-359-7370

³Kimata, K., et al. "Rapid Categorization of Pathogenic *Escherichia coli* by Multiplex PCR." <u>Microbiol. Immunol.</u> 49 (2005): 485–492. PubMed: 15965295.

⁴An extraction procedure was used that has been shown to consistently inactivate 100% of Gram-negative bacteria.

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Lane 1: Invitrogen™ 1Kb DNA Ladder™

Lane 2: 200 ng of NR-2647

Date: 10 MAR 2008 Signature: Signature on File

> Title: Technical Manager, BEI Authentication or designee

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