b|e|i RESOURCES

SUPPORTING INFECTIOUS DISEASE RESEARCH

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* (*E. coli*; ETEC), strain H10407, serotype O78:H11.

Lot[1]: 64204551 **Manufacturing Date: 15JUN2016**

TEST	SPECIFICATIONS	RESULTS
Genotypic Analysis		
Sequencing of 16S ribosomal RNA gene (~ 1480 base pairs)	≥ 99% sequence identity to *E. coli*, strain H10407 (GenBank: FN649414)	99.8% sequence identity to *E. coli*, strain H10407 (GenBank: FN649414)
PCR amplification of plasmid markers[2,3]		
elt (pJY11)	Positive	Positive
esth (pCS1)	Positive	Positive
estp (pCS1)	Positive	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[2,3]		
eaeA	Negative	Negative
stx1	Negative	Negative
stx2	Negative	Negative
astA	Positive	Positive
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	0.93 µg in 46.5 µL per vial (20 µg/mL)
Functional Activity by PCR Amplification 16S ribosomal RNA gene	~ 1500 base pairs amplicon	~ 1500 base pairs amplicon
OD_{260}/OD_{280} Ratio	1.7 to 2.1	1.8
Bacterial Inactivation 10% of total yield plated on Tryptic Soy agar with 5% defibrinated sheep blood[4,5]	No viable bacteria detected	No viable bacteria detected

[1]The bacterial preparation used for extraction of genomic DNA was produced by inoculation of BEI Resources NR-4, Lot 3561329, into Tryptic Soy broth. After incubation for 24 hours at 37°C, genomic DNA was extracted using proprietary technology.

[2]PCR amplification was not performed on NR-2647, Lot 64204551. PCR amplification data was obtained from NR-2647, Lot 5107284 (also produced from extraction of BEI Resources NR-4, Lot 3561329) and NR-4, Lot 3561329.

[3]Kimata, K., et al. "Rapid Categorization of Pathogenic *Escherichia coli* by Multiplex PCR." Microbiol. Immunol. 49 (2005): 485-492. PubMed: 15965295.

[4]14 days at 37°C in an aerobic atmosphere

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

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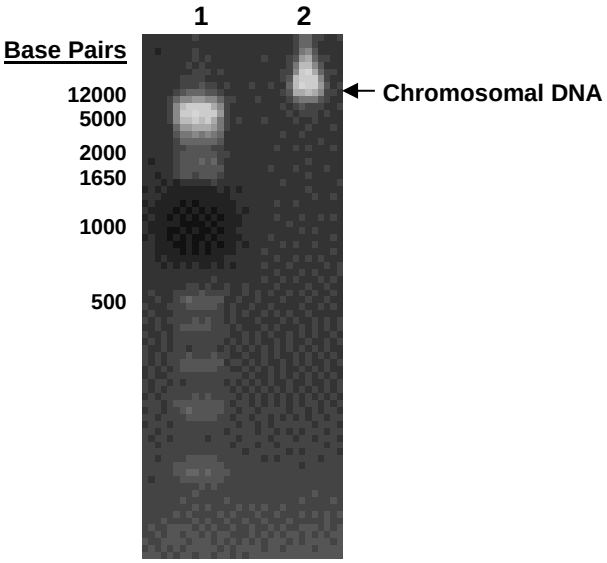
Figure 1: Agarose Gel Electrophoresis



Lane 1: Invitrogen™ 1Kb Plus DNA Ladder
Lane 2: 200 ng of NR-2647

Date: 03 OCT 2016

Signature:

BEI Resources Authentication

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.

Support Provided by NIAID

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