b|e|i RESOURCES
SUPPORTING INFECTIOUS DISEASE RESEARCH

Certificate of Analysis for NR-3050

Genomic DNA from *Escherichia coli*, Strain CDC (ATCC® 12807™)

Catalog No. NR-3050

Product Description: Genomic DNA was extracted from a preparation of *Escherichia coli* (*E. coli*), strain CDC (ATCC® 12807™), serotype O126:K71(B16):H.

Lot[1]: 64204561 **Manufacturing Date: 24MAY2016**

TEST	SPECIFICATIONS	RESULTS
Genotypic Analysis		
Sequencing of 16S rRNA gene (~ 880 base pairs)	≥ 99% sequence identity to *E. coli* type strain (GenBank: X80725)	99.5% sequence identity to *E. coli* type strain (GenBank: X80725)[2]
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	0.95 µg in 54 µL per vial (17.6 µg/mL)
Functional Activity by PCR Amplification		
16S ribosomal RNA gene	~ 1500 base pairs amplicon	~ 1500 base pairs amplicon
Genotypic Analysis of Virulence Markers[3,4]		
PCR amplification of plasmid markers		
EAF (pEAF)	Report results	Positive
bfpA (pEAF)	Report results	Positive
elt (pJY11)	Negative	Negative
esth (pCS1)	Negative	Negative
estp (pCS1)	Negative	Negative
invE (pINV)	Negative	Negative
CVD432 (pAA)	Negative	Negative
aggR (pAA)	Negative	Negative
PCR amplification of chromosomal markers		
eaeA	Report results	Negative
stx1	Negative	Negative
sx2	Negative	Negative
astA	Report results	Negative
OD_{260}/OD_{280} Ratio	1.7 to 2.1	1.8
Bacterial Inactivation		
10% of total yield plated on agar[5,6]	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 NRS-99 (Lot 3663830) into Tryptic Soy broth. After incubation for 24 hours at 37°C, genomic DNA was extracted using proprietary technology.
[2]Also consistent with *Shigella* species.
[3]Kimata, K., et al. "Rapid Categorization of Pathogenic *Escherichia coli* by Multiplex PCR." Microbiol. Immunol. 49 (2005): 485-492. PubMed: 15965295.
[4]Virulence marker results were obtained from the source organism used to produce the seed lot used for nucleic acid extraction (NR-99, lot 3663825).
[5]An extraction procedure was used that has been shown to consistently inactivate 100% of Gram-negative and Gram-positive bacteria.
[6]Plates were incubated for 14 days under propagation conditions.

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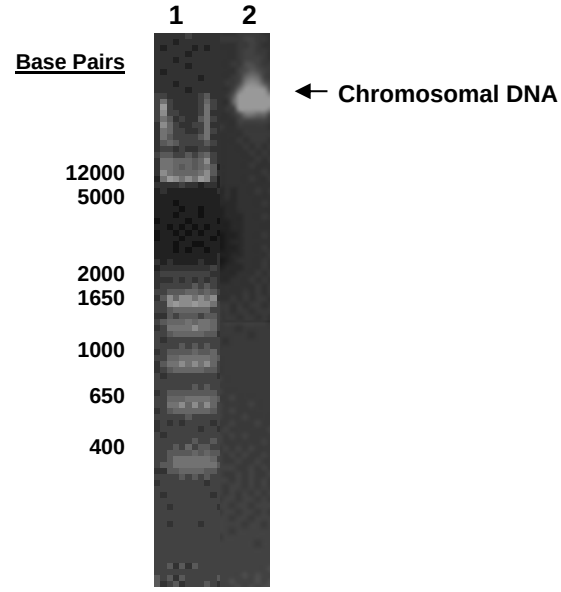
Figure 1: Agarose Gel Electrophoresis



Lane 1: Invitrogen™ 1Kb Plus DNA Ladder™
Lane 2: 200 ng of NR-3050

Heather Couch

Digitally signed by Heather Couch
DN: cn=Heather Couch, o=ATCC, ou=ATCC Federal Solutions, email=hcouch@atcc.org, c=US
Date: 2018.02.28 09:00:58 -05'00'

28 FEB 2018

Program Manager or designee, ATCC Federal Solutions

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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