

Diarrheagenic *Escherichia coli* Organism Panel

Catalog No. NR-9545

For research use only. Not for human use.

Contributor:

ATCC®

Product Description:

NR-9545 consists of five organisms representing different diarrheagenic *Escherichia coli* (*E. coli*) pathotypes (Table 1). The indicated pathotypes have been confirmed by PCR amplification of pathogenic sequences from extracted nucleic acid.

A brief description of each organism may be found in Appendix I. Each organism is available individually by requesting the indicated BEI Resources NR number.

A companion panel containing extracted nucleic acid from these five organisms is available as BEI Resources NR-9546 (Diarrheagenic *Escherichia coli* Nucleic Acid Panel).

Material Provided:

Each vial contains approximately 0.5 mL of bacterial culture in 0.5X Tryptic Soy Broth supplemented with 10% glycerol.

Packaging/Storage:

NR-9545 was packaged aseptically, in screw-capped plastic cryovials. The product is provided frozen and should be stored at -60°C or colder immediately upon arrival. For long-term storage, the vapor phase of a liquid nitrogen freezer is recommended. Freeze-thaw cycles should be avoided.

Growth Conditions:

Media:

Tryptic Soy Broth or equivalent
Tryptic Soy Agar or equivalent

Incubation:

Temperature: 37°C

Atmosphere: Aerobic

Propagation:

1. Keep vial frozen until ready for use; then thaw.
2. Transfer the entire thawed aliquot into a single tube of Tryptic Soy Broth.
3. Use several drops of the suspension to inoculate a Tryptic Soy Agar slant and/or plate.
4. Incubate the slant and/or plate at 37°C for 24 hours.

Citation:

Acknowledgment for publications should read "The following reagent was obtained through the NIH Biodefense and Emerging Infections Research Resources Repository, NIAID,

NIH: Diarrheagenic *Escherichia coli* Organism Panel, NR-9545."

Biosafety Level: 2

Appropriate safety procedures should always be used with this material. Laboratory safety is discussed in the following publication: U.S. Department of Health and Human Services, Public Health Service, Centers for Disease Control and Prevention, and National Institutes of Health. Biosafety in Microbiological and Biomedical Laboratories. 5th ed. Washington, DC: U.S. Government Printing Office, 2007; see www.cdc.gov/od/ohs/biosfty/bmb15/bmb15toc.htm.

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Table 1

BEI Resources	Strain	Serotype	Pathotype	Marker(s) Confirmed by PCR Amplification of Extracted DNA
NR-4	H10407	O78:H11	ETEC	<i>esth</i> and <i>estp</i> on pCS1; <i>elt</i> on pJY11 <i>astA</i> on chromosome
NR-11	EDL 933	O157:H7	EHEC	<i>hlyA</i> on pO157 <i>stx1</i> , <i>stx2</i> and <i>eaeA</i> on chromosome
NR-99	CDC	O126:K71(B16):H	EPEC	EAF and <i>bfpA</i> on pEAF <i>eaeA</i> on chromosome
NR-100	1885-77 (EDL 1282)	O29:NM	EIEC	<i>invE</i> on pINV
NR-102	NCDC U14-41	O3:K2a,2b(L):H2	EAEC	CVE432 and <i>aggR</i> on pAA

APPENDIX I

ETEC (BEI Resources NR-4)

The enterotoxigenic *E. coli* (ETEC) strain H10407 was isolated from a patient with diarrhea in Bangladesh¹ and was deposited at ATCC® in the early 1980s by Samuel B. Formal, Department of Bacterial Diseases, Walter Reed Army Institute of Research, Washington, DC. It produces at least two types of virulence factors: 1) colonization factor antigen I (CFA/I), which is responsible for adhesion of bacterial cells to intestinal epithelial cells, and 2) heat-labile (LT) and heat-stable (ST) enterotoxins which cause diarrhea.¹ *E. coli* H10407 carries three plasmid species: 1) pCS1 (CFA/I+ST⁺; 95,000 bp), 2) pJY11 (LT+ST⁺; 65,000 bp), and 3) pTRA1 (65,000 bp), a self-transmissible plasmid which mobilizes pCS1 and pJY11.¹ The gene for another heat-stable enterotoxin (EAST1) has been found in *E. coli* H10407, sequenced (GenBank: AB042004), and reported to have enterotoxin activity.^{2,3}

References:

1. Yamamoto, T. and T. Yokota. "Plasmids of Enterotoxigenic *Escherichia coli* H10407: Evidence for Two Heat-Stable Enterotoxin Genes and a Conjugal Transfer System." J. Bacteriol. 153 (1983): 1352–1360. PubMed: 6298182.
2. Yamamoto, T. and P. Echeverria. "Detection of the Enteraggregative *Escherichia coli* Heat-stable Enterotoxin 1 Gene Sequences in Enterotoxigenic *E. coli* Strains Pathogenic for Humans." Infect. Immun. 64 (1996): 1441–1445. PubMed: 8606115. GenBank: AB042004.
3. McVeigh, A., et al. "IS1414, an *Escherichia coli* Insertion Sequence with a Heat-Stable Enterotoxin Gene Embedded in a Transposase-Like Gene." Infect. Immun. 68 (2000): 5710–5715. PubMed: 10992475.

EHEC (BEI Resources NR-11)

The enterohemorrhagic *E. coli* (EHEC) strain EDL933 was isolated from raw hamburger meat¹ implicated in a multi-state outbreak of hemorrhagic colitis in the United States in 1982. It was deposited at ATCC in 2000 by Guy Plunkett III, Ph.D., Laboratory of Genetics, University of Wisconsin-Madison, Madison, Wisconsin. *E. coli* strain EDL933 and many other EHEC strains encode potent toxins, similar to those of *Shigella dysenteriae*, which can cause severe intestinal, kidney and central nervous system disease. *E. coli* EDL933 carries two plasmid species: 1) pO157 (92,077 bp) and 2) pEDL933 (3,306 bp). Virulence-associated genes are located on both the chromosome and pO157. The complete sequence of the chromosome (5,528,445 bp; GenBank: NC_002655)² and pO157 (GenBank: AF074613)³ from *E. coli* strain EDL933 have been determined.

References:

1. Riley, L. W., et al. "Hemorrhagic Colitis Associated with a Rare *Escherichia coli* Serotype." N. Engl. J. Med. 308 (1983): 681–685. PubMed: 6338386.
2. Perna, N. T., et al. "Genome Sequence of Enterohaemorrhagic *Escherichia coli* O157:H7." Nature 409 (2001): 529–533. PubMed: 11206551. GenBank: NC_002655.
3. Burland, V., et al. "The Complete DNA Sequence and Analysis of the Large Virulence Plasmid of *Escherichia coli* O157:H7." Nucleic Acids Res. 26 (1998): 4196–4204. PubMed: 9722640. GenBank: AF074613.

EPEC (BEI Resources NR-99)

The enteropathogenic *E. coli* (EPEC) strain CDC was deposited at ATCC® by the National Communicable Disease Center, Atlanta, Georgia. EPEC strains cause diarrheal outbreaks and chronic diarrhea, especially in infants. EPEC pathogenesis requires the expression of genes present both on the chromosome and on an adherence factor plasmid, pEAF.^{1,2} The complete sequence of the pEAF plasmid (also referred to as pB171) from EPEC strain B171 has been determined (68,817 bp; GenBank: AB024946).²

References:

1. Momenteau, H. "Study on Enteropathogenic *E. coli* of the O126:K71 (B16) Serotype." Ann. Inst. Pasteur (Paris) 113 (1967): 128–131. PubMed: 4864790.
2. Tobe, T., et al. "Complete DNA Sequence and Structural Analysis of the Enteropathogenic *Escherichia coli* Adherence Factor Plasmid." Infect. Immun. 67 (1999): 5455-5462. PubMed: 10496929. GenBank: AB024946.

EIEC (BEI Resources NR-100)

The enteroinvasive *E. coli* (EIEC) strain 1885-77 was isolated from human stool¹ in 1977 and was deposited at ATCC® in 1988 by Dr. Nancy A. Strockbine, Enteric Bacteriology Section, Centers for Disease Control and Prevention, Atlanta, Georgia. A high-molecular-weight plasmid and a positive Serény test have both been associated with EIEC strains.² EIEC strains invade and multiply within intestinal epithelial cells, resulting in a dysentery-like enteritis in humans, similar to that caused by *Shigella* species. EIEC pathogenesis requires the expression of genes present both on the chromosome and on a large invasion plasmid, pINV (220,000 bp).^{3,4} The plasmid shares a significant degree of DNA homology with the virulence plasmid described in *Shigella* species, and is structurally and functionally equivalent.^{3,4}

References:

1. Toledo, M. R., et al. "Invasive Strain of *Escherichia coli* Belonging to O Group 29." *J. Clin. Microbiol.* 9 (1979): 288–289. PubMed: 372230.
2. Harris, J. R., I. K. Wachsmuth, B. R. Davis, and M. L. Cohen. "High-Molecular-Weight Plasmid Correlates with *Escherichia coli* Enteroinvasiveness." *Infect. Immun.* 37 (1982): 1295–1298. PubMed: 6752026.
3. Hsia, R.-C., P. L. C. Small, and P. M. Bavoil. "Characterization of Virulence Genes of Enteroinvasive *Escherichia coli* by TnphoA Mutagenesis: Identification of *invX*, a Gene Required for Entry into HEp-2 Cells." *J. Bacteriol.* 175 (1993): 4817–4823. PubMed: 8393007.
4. Lan, R., et al. "Molecular Evolutionary Relationships of Enteroinvasive *Escherichia coli* and *Shigella* spp." *Infect. Immun.* 72 (2004): 5080–5088. PubMed: 15322001.

EAEC (BEI Resources NR-102)

The enteroaggregative *E. coli* (EAEC) strain NCDC U14-41 was isolated from human urine¹ in 1943 by Dr. F. Kauffmann and was deposited at ATCC® in 1967 by Dr. William H. Ewing, Bacteriology Section, National Communicable Disease Center, Atlanta, Georgia. PCR probes have been developed to identify the presence of aggregative adherence pattern associated plasmid (pAA) and the virulence marker *aggR*, which are commonly associated with EAEC.

References:

1. *Acta Pathol. Microbiol. Scand.* 20 (1943): 21–44.
2. Brenner, D. J., et al. "Atypical Biogroups of *Escherichia coli* Found in Clinical Specimens and Description of *Escherichia hermannii* sp. nov." *J. Clin. Microbiol.* 15 (1982): 703–713. PubMed: 7040466.
3. Schmidt, H., et al. "Development of PCR for Screening of Enteroaggregative *Escherichia coli*." *J. Clin. Microbiol.* 33 (1995): 701–705. PubMed: 7751380.
4. Tsai, C. C., S. Y. Chen, and H. Y. Tsen. "Screening the Enteroaggregative *Escherichia coli* Activity and Detection of the *aggA*, *aafA*, and *astA* Genes with Novel PCR Primers for the *Escherichia coli* Isolates from Diarrhea Cases in Taiwan." *Diagn. Microbiol. Infect. Dis.* 46 (2003): 159–165. PubMed: 12867090.
5. Ogata, K., et al. "Prevalence of *Escherichia coli* Possessing the *eaeA* Gene of Enteropathogenic *E. coli* (EPEC) or the *aggR* Gene of Enteroaggregative *E. coli* (EAggEC) in Traveler's Diarrhea Diagnosed in Those Returning to Tama, Tokyo from Other Asian Countries." *Jpn. J. Infect. Dis.* 55 (2002): 14–18. PubMed: 11971156.
6. Moon, J. Y., J. H. Park, and Y. B. Kim. "Molecular Epidemiological Characteristics of Virulence Factors on Enteroaggregative *E. coli*." *FEMS Microbiol. Lett.* 253 (2005): 215–220. PubMed: 16257141.