SUPPORTING INFECTIOUS DISEASE RESEARCH

# *Escherichia coli* Virulence Target *invE* Primers

### Catalog No. NR-12200

This reagent is the tangible property of the U.S. Government.

## For research use only. Not for human use.

#### **Contributor and Manufacturer:**

**BEI Resources** 

#### **Product Description:**

Diarrheagenic *Escherichia coli* (*E. coli*) are classified into several pathogenic groups based on their virulence characteristics. NR-12200 contains forward and reverse primers that specifically amplify a region of the virulence target *invE* found on plasmid pINV of enteroinvasive *E. coli* (EIEC).

#### **Material Provided:**

Each vial contains approximately 100  $\mu$ L of a mixture of forward and reverse primers in TE buffer (pH 7.0). The concentration is shown on the Certificate of Analysis.

Note: *E. coli* 12-Target Multiplex PCR 10X Buffer (BEI Resources NR-13440) will be provided with your shipment of NR-12200.

#### Packaging/Storage:

Primers were packaged aseptically in screw-capped plastic cryovials. The product is provided frozen on dry ice and should be stored at -60°C or colder upon arrival. Freeze-thaw cycles should be minimized.

#### Citation:

Acknowledgment for publications should read "The following reagent was obtained through BEI Resources, NIAID, NIH: *Escherichia coli* Virulence Target *invE* Primers, NR-12200."

#### **Biosafety Level: 1**

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. <u>Biosafety in</u> <u>Microbiological and Biomedical Laboratories</u>. 5th ed. Washington, DC: U.S. Government Printing Office, 2009; see www.cdc.gov/biosafety/publications/bmbl5/index.htm.

#### **Disclaimers:**

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#### **References:**

- Kimata, K., et al. "Rapid Categorization of Pathogenic Escherichia coli by Multiplex PCR." <u>Microbiol. Immunol.</u> 49 (2005): 485-492. PubMed: 15965295.
- Harris, J. R., I. K. Wachsmuth, B. R. Davis, and M. L. Cohen. "High-Molecular-Weight Plasmid Correlates with *Escherichia coli* Enteroinvasiveness." <u>Infect. Immun.</u> 37 (1982): 1295-1298. PubMed: 6752026.
- 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. <u>Bacteriol.</u> 175 (1993): 4817-4823. PubMed: 8393007.
- Lan, R., et al. "Molecular Evolutionary Relationships of Enteroinvasive *Escherichia coli* and *Shigella* spp." <u>Infect.</u> <u>Immun.</u> 72 (2004): 5080-5088. PubMed: 15322001.

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# APPENDIX I

# E. coli Virulence Target invE Primers

Reagent	Source	Catalog #		
E. coli Virulence Target invE Primers	BEI Resources	NR -12200		
Positive Control Template, Genomic DNA from <i>E. coli</i> , Strain 1885-77 (EDL1282)	BEI Resources	NR-3051		
10X PCR Buffer	BEI Resources	NR-13440		
GoTaq <sup>®</sup> Polymerase	Promega	M500B		
dNTP Mix	Promega	U151		
Molecular Biology Grade Water	ATCC®	60-2645		

#### **Recommended Reagents/Equipment**

#### **Reaction Mix<sup>1</sup>**

Reagent	Stock Concentration	Volume per Reaction (µL)
Molecular Biology Grade Water		19.2
10X PCR Buffer	10X	3
dNTP Mix	10 mM each	0.6
GoTaq <sup>®</sup> Polymerase		0.2
Primers <sup>2</sup>	10 µM (each primer)	5
Template DNA	1 ng per µL	2
4		Total – 30 µL

<sup>1</sup>Reaction mix should be kept on bench-top cooler until ready for use. <sup>2</sup>Primers are supplied at working stock concentrations.

## **Cycling Protocol**

Cycle	# of Repeats	Step	Conditions
1	1	1	94°C for 5 minutes
2	30	1	94°C for 1 minute
		2	52°C for 1 minute
		3	72°C for 1 minute
3	1	1	72°C for 7 minutes
4	Indefinite	1	Hold at 4°C