

Vector pcDNA3.1 Containing β -Lactamase Fused to Ebolavirus, Zaire VP40 (Bla-VP40)

Catalog No. NR-19813

For research use only. Not for human use.

Contributor:

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Manufacturer:

BEI Resources

Product Description:

The viral matrix protein VP40 gene from ebolavirus (EBOV), Zaire was synthesized by multiple rounds of overlapping PCR based on the EBOV, Zaire genome sequence (GenBank accession L11365). The β -lactamase gene was PCR amplified from the pcDNA3.1 vector (Invitrogen™) and fused to the N-terminal of VP40 by a short linker sequence (GSGGGSGGT) to create a modified β -lactamase-VP40 fusion protein (Bla-VP40) which was subcloned into pcDNA3.1. The modified β -lactamase lacks the N-terminal 24 amino acid secretion signal and His24 was substituted by Asp to create an optimal Kozak consensus sequence.¹ The plasmid was produced in *Escherichia coli* 5-alpha F'F cells (New England Biolabs®) and extracted using a QIAGEN® plasmid DNA extraction kit.

VP40 drives the budding of filovirus particles. 293T cells cotransfected with NR-19813 and the EBOV glycoprotein (NR-19814) or the Marburg virus (MARV) glycoprotein (NR-19815) produce EBOV or MARV virus-like particles (VLPs), respectively. Fusion of these VLPs with target cells can be detected by monitoring β -lactamase activity using a fluorogenic substrate, permitting study of the cell entry steps of these highly pathogenic viruses without the need for BSL-4 containment.²

NR-19813 has been qualified for use in bacterial transformations.

Material Provided:

Each vial contains 20 to 50 ng of plasmid DNA in TE buffer (10 mM Tris-HCl, 1 mM EDTA, pH 7.0). The concentration is shown on the Certificate of Analysis. The vial should be centrifuged prior to opening.

Packaging/Storage:

NR-19813 was packaged aseptically in screw-capped plastic cryovials. The product is provided frozen on dry ice and should be stored at -20°C or colder immediately upon arrival. Freeze-thaw cycles should be minimized.

Citation:

Acknowledgment for publications should read "The following reagent was obtained through BEI Resources, NIAID, NIH:

Vector pcDNA3.1 Containing β -Lactamase Fused to Ebolavirus, Zaire VP40 (Bla-VP40), NR-19813."

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. Biosafety in Microbiological and Biomedical Laboratories. 5th ed. Washington, DC: U.S. Government Printing Office, 2009; see www.cdc.gov/biosafety/publications/bmb15/index.htm.

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References:

1. Manicassamy, B., and L. Rong. "Expression of Ebolavirus Glycoprotein on the Target Cells Enhances Viral Entry." *Virology* 6 (2009): 75. PubMed: 19505320.
2. Tschernie, D.M., et al. "An Enzymatic Virus-like Particle Assay for Sensitive Detection of Virus Entry." *J. Virol. Methods* 163 (2010): 336-343. PubMed: 19879300.

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