Vector pLVX-EF1α-IRES-Puro Containing the SARS-Related Coronavirus 2, USA-WA1/2020 Non-Structural Protein 13 Gene

Catalog No. NR-52961

For research use only. Not for use in humans.

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Manufacturer:
BEI Resources

Product Description:
Note: The vial label indicates this product contains a TST tag. This nomenclature refers to a 2X Strep tag.[1,2] This product does not express the Twin-Strep-tag® that is commonly referred to as a TST tag.

The non-structural protein 13 (nsp13) gene from severe acute respiratory syndrome-related coronavirus 2 (SARS-CoV-2), USA-WA1/2020 (GenBank: MN985325) was codon optimized and modified by the addition of a C-terminal 2X Strep tag and cloned into the pLVX-EF1α-IRES-Puro lentiviral expression plasmid.[1,2] The vector contains an internal ribosomal entry site (IRES) that allows a gene-of-interest and a puromycin resistance gene to be simultaneously co-expressed from a single mRNA transcript. Expression of the transcript is driven by the human elongation factor 1 alpha (EF1α) promoter. The beta-lactamase gene, *bla*, provides transformant selection through ampicillin resistance in *Escherichia coli* (*E. coli)* and the puromycin resistance gene, *pac*, provides transformant selection through puromycin resistance in eukaryotic cells. NR-52961 can be used for transient expression and lentivirus generation.[1] The resulting size of the plasmid is approximately 10,720 base pairs. The complete plasmid sequence and map are provided on the BEI Resources webpage. The plasmid was produced in *E. coli* and extracted.

NSP13 is a multifunctional protein located in the ORF1ab polyprotein. The N-terminus is an NTPase and contains a zinc binding domain. The C-terminus is a helicase that can unwind both RNA and DNA. The helicase activity is stimulated by the RNA polyermase RdRp (NSP12). NSP13 is essential for viral replication and therefore is a potential antiviral drug target.[3,4]

Material Provided:
Each vial contains plasmid DNA in TE buffer (10 mM Tris-HCl, 1 mM EDTA, pH 8.0). The DNA concentration and volume provided are shown on the Certificate of Analysis. The vial should be centrifuged prior to opening. Note: The contents of the vial should be used to replicate the plasmid in *E. coli* prior to mammalian expression studies.

Packaging/Storage:
NR-52961 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 pLVX-EF1α-IRES-Puro Containing the SARS-Related Coronavirus 2, USA-WA1/2020 Non-Structural Protein 13 Gene, NR-52961."

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/bmbl5/index.htm.

Disclaimers:
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References:

1. Krogan, N., Personal Communication.
2. Gordon, D. E., et al. "A SARS-CoV-2 Protein Interaction Map Reveals Targets for Drug Repurposing." Nature 583 (2020): 459-468. PubMed: 32353859.
3. Subissi, L., et al. "SARS-CoV ORF1b-Encoded Nonstructural Proteins 12-16: Replicative Enzymes as Antiviral Targets." Antiviral Res. 101 (2014): 122-130. PubMed: 24269475.
4. Shu, T., et al. "SARS-Coronavirus-2 Nsp13 Possesses NTPase and RNA Helicase Activities That Can Be Inhibited by Bismuth Salts." Virol. Sin. 35 (2020): 321-329. PubMed: 32500504.

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