Product Information Sheet for NR-52971

Vector pLVX-EF1α-IRES-Puro Containing the SARS-Related Coronavirus 2, USA-WA1/2020 Open Reading Frame 7b Gene

Catalog No. NR-52971

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 open reading frame 7b (orf7b) 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 N-terminal 2X Strep tag and cloned into the pLVX-EF1α-IRES-Puro lentiviral expression plasmid.[1,2,3] 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. The resulting size of the plasmid is approximately 9330 base pairs. NR-52971 can be used for transient expression and lentivirus generation.[1] The complete plasmid sequence and map are provided on the BEI Resources webpage. The plasmid was produced in *E. coli* and extracted.

ORF7b is a protein of unknown function and is not essential for viral replication *in vitro*. It is believed to be a transmembrane protein and has been shown to localize to the Golgi complex as well as intracellular virus particles and purified virus particles. The production of ORF7b is the result of ribosomal leaky scanning of gene 7 mRNA.[4,5]

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-52971 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 Open Reading Frame 7b Gene, NR-52971."

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. Busby, M., et al. "Optimisation of a Multivalent Strep Tag for Protein Detection." Biophys. Chem. 152 (2010): 170-177. PubMed: 20970240.
3. Gordon, D. E., et al. "A SARS-CoV-2 Protein Interaction Map Reveals Targets for Drug Repurposing." Nature 583 (2020): 459-468. PubMed: 32353859.
4. Yoshimoto, F. K. "The Proteins of Severe Acute Respiratory Syndrome Coronavirus-2 (SARS CoV-2 or n-COV19), the Cause of COVID-19." Protein J. 39 (2020): 198-216. PubMed: 32447571.
5. Schaecher, S. R., J. M. Mackenzie and A. Pekosz. "The ORF7b Protein of Severe Acute Respiratory Syndrome Coronavirus (SARS-CoV) is Expressed in Virus-Infected Cells and Incorporated into SARS-CoV Particles." J. Virol. 81 (2007): 718-731. PubMed: 17079322.

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