

Product Information Sheet for NR-52966

Vector pLVX-EF1 α -IRES-Puro Containing the SARS-Related Coronavirus 2, USA-WA1/2020 Open Reading Frame 3b Gene

Catalog No. NR-52966

For research use only. Not for use in humans.

Contributor:

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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 3b (orf3b) 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-EF1 \$\alpha\$ -IRES-Puro](#) lentiviral expression plasmid.^{1,2,3,4} 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 (EF1 α) 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 9080 base pairs. NR-52966 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.

ORF3b is involved in inducing inflammatory response and is a potent interferon antagonist, suppressing the induction of type I interferon more efficiently than its SARS-CoV ortholog.^{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-52966 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-EF1 α -IRES-Puro Containing the SARS-Related Coronavirus 2, USA-WA1/2020 Open Reading Frame 3b Gene, NR-52966."

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.

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. Kim, D.-K., et al. "A Comprehensive, Flexible Collection of SARS-CoV-2 Coding Regions." G3 (Bethesda) 10 (2020): 3399-3402. PubMed: 32763951.
5. Konno, Y., et al. "SARS-CoV-2 ORF3b Is a Potent Interferon Antagonist Whose Activity Is Increased by a Naturally Occurring Elongation Variant." Cell Rep. 32 (2020): 108185. PubMed: 32941788.
6. Michel, C. J., et al. "Characterization of Accessory Genes in Coronavirus Genomes." Virology 17 (2020): 131. PubMed: 32854725.
7. Hachim, A., et al. "ORF8 and ORF3b Antibodies Are Accurate Serological Markers of Early and Late SARS-CoV-2 Infection." Nat. Immunol. 21 (2020): 1293-1301. PubMed: 32807944.
8. 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.

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