

Vector pMCSG53 Containing the SARS-Related Coronavirus 2, Wuhan-Hu-1 Non-Structural Protein 8 Gene

Catalog No. NR-52902

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Contributor:

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Manufacturer:

BEI Resources

Product Description:

The non-structural protein 8 (nsp8) gene from severe acute respiratory syndrome-related coronavirus 2 (SARS-CoV-2), Wuhan-Hu-1 (GenBank: [MN908947](#)) was codon optimized and cloned into the [pMCSG53](#) plasmid.^{1,2} pMCSG53 is an *Escherichia coli* (*E. coli*) expression vector that contains an N-terminal hexa-histidine tag, followed by a tobacco etch virus (TEV) protease recognition site prior to the insert coding sequence, resulting in the expression of a cleavable histidine-tagged protein.³ It also contains tRNA genes covering rare codons for arginine (AGG/AGA) and isoleucine (AUA) to improve expression in *E. coli*. The beta-lactamase gene, *bla*, provides transformant selection through ampicillin resistance in *E. coli*. The resulting size of the plasmid is approximately 5390 base pairs. The complete plasmid sequence and map are provided on the BEI Resources webpage. The plasmid was produced in *E. coli* and extracted.

NSP8 is an accessory subunit in the RNA-dependent RNA polymerase (RdRp, also known as NSP12) complex, and together with NSP7 may function as a general processivity factor for RdRp. The RdRp is a target of antiviral medications.^{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 expression studies.

Packaging/Storage:

NR-52902 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 pMCSG53 Containing the SARS-Related Coronavirus 2, Wuhan-Hu-1 Non-Structural Protein 8 Gene, NR-52902."

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.

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References:

1. Joachimiak, A., Personal Communication.
2. Wu, F., et al. "A New Coronavirus Associated with Human Respiratory Disease in China." [Nature](#) 579 (2020): 265-269. PubMed: 32015508.
3. Eschenfeldt, W. H., et al. "New LIC Vectors for Production of Proteins from Genes Containing Rare Codons." [J. Struct. Funct. Genomics](#) 14 (2013): 135-144. PubMed: 24057978.

4. Zhai, Y., et al. "Insights into SARS-CoV Transcription and Replication from the Structure of the Nsp7-Nsp8 Hexadecamer." *Nat. Struct. Mol. Biol.* 12 (2005): 980-986. PubMed: 16228002.
5. Hillen, H. S., et al. "Structure of Replicating SARS-CoV-2 Polymerase." *Nature* (2020): *In press*. PubMed: 32438371.

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