biei resources

SUPPORTING INFECTIOUS DISEASE RESEARCH

Vector pCMV/R Containing the SARS-Related Coronavirus 2, Spike Glycoprotein Gene, Lineage B.1.1.529, Omicron Variant

Catalog No. NR-56470

This reagent is the tangible property of the U.S. Government.

For research use only. Not for use in humans.

Contributor:

John R. Mascola, M.D., Director, Humoral Immunology Section, Vaccine Research Center, National Institute of Allergy and Infectious Diseases, National Institutes of Health, Bethesda, Maryland, USA

Manufacturer:

BEI Resources

Product Description:

<u>NR-56470 expresses the full-length, Omicron variant spike (S)</u> <u>glycoprotein, and is intended for producing pseudotyped</u> <u>particles/pseudovirions.^{1,2} NR-56470 is not intended for</u> recombinant protein expression.

The vector for the S glycoprotein gene from severe acute respiratory syndrome-related coronavirus 2 (SARS-CoV-2), Wuhan-Hu-1 (GenBank: MN908947) was designed by codon optimizing the full-length S sequence (residues 1 to 1273) for mammalian expression and introducing point mutations found in the B.1.1.529 lineage, resulting in a spike glycoprotein gene representative of the Omicron variant. The spike gene was subcloned into the pCMV/R mammalian expression vector (also referred to as VRC8400).^{1,3,4} The protein encoded by NR-56470 contains the following lineage-defining point mutations: A67V, T95I, G339D, S371L, S373P, K417N, N440K, G446S, S477N, T478K, E484A, Q493R, G496S, Q498R, N501Y, T547K, D614G, H655Y, N679K, P681H, N764K, D796Y, N856K, Q954H and N969K.¹ The kanamycin resistance gene, aph, provides transformant selection through kanamycin resistance in Escherichia coli (E. coli).1 The resulting size of the plasmid is approximately 8240 base pairs. The complete plasmid sequence and map are provided on the BEI Resources webpage. The plasmid was produced in E. coli and extracted.

The S glycoprotein mediates viral binding to the host angiotensin converting enzyme 2 (ACE2). This protein forms a trimer, and when bound to a host receptor, allows fusion of the viral and cellular membranes. The S protein is a target for neutralizing antibodies.⁵ The Omicron variant of SARS-CoV-2 includes multiple S glycoprotein mutations that were first identified in Botswana, Hong Kong, and South Africa in November 2021.⁶

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. <u>Note</u>: The contents of the vial should be used to replicate the plasmid in *E. coli* prior to mammalian expression.

Packaging/Storage:

NR-56470 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 pCMV/R Containing the SARS-Related Coronavirus 2, Spike Glycoprotein Gene, Lineage B.1.1.529, Omicron Variant, NR-56470."

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. <u>Biosafety in Microbiological and Biomedical Laboratories</u>. 6th ed. Washington, DC: U.S. Government Printing Office, 2020; see www.cdc.gov/biosafety/publications/bmbl5/index.htm.

Disclaimers:

You are authorized to use this product for research use only. It is not intended for human use.

Use of this product is subject to the terms and conditions of the BEI Resources Material Transfer Agreement (MTA). The MTA is available on our Web site at <u>www.beiresources.org</u>.

While BEI Resources uses reasonable efforts to include accurate and up-to-date information on this product sheet, neither ATCC[®] nor the U.S. Government makes any warranties or representations as to its accuracy. Citations from scientific literature and patents are provided for informational purposes only. Neither ATCC[®] nor the U.S. Government warrants that such information has been confirmed to be accurate.

This product is sent with the condition that you are responsible for its safe storage, handling, use and disposal. ATCC[®] and the U.S. Government are not liable for any damages or injuries arising from receipt and/or use of this product. While reasonable effort is made to ensure authenticity and reliability of materials on deposit, the U.S. Government, ATCC[®], their suppliers and contributors to BEI Resources are not liable for damages arising from the misidentification or misrepresentation of products.

Use Restrictions:

This material is distributed for internal research, non-commercial purposes only. This material, its product or its derivatives may not be distributed to third parties. Except as performed under a U.S. Government contract, individuals contemplating commercial use of the material, its products or

E-mail: <u>contact@beiresources.org</u> Tel: 800-359-7370 Fax: 703-365-2898 **b**|**e**|**i** resources

SUPPORTING INFECTIOUS DISEASE RESEARCH

its derivatives must contact the contributor to determine if a license is required. U.S. Government contractors may need a license before first commercial sale.

References:

- 1. Mascola, J. R., Personal Communication.
- Millet, J. K., et al. "Production of Pseudotyped Particles to Study Highly Pathogenic Coronaviruses in a Biosafety Level 2 Setting." <u>J. Vis. Exp.</u> 145 (2019): doi: 10.3791/59010. PubMed: 30882796.
- Wu, F., et al. "A New Coronavirus Associated with Human Respiratory Disease in China." <u>Nature</u> 579 (2020): 265-269. PubMed: 32015508.
- Barouch, D. H., et al. "A Human T-Cell Leukemia Virus Type 1 Regulatory Element Enhances the Immunogenicity of Human Immunodeficiency Virus Type 1 DNA Vaccines in Mice and Nonhuman Primates." <u>J.</u> <u>Virol.</u> 79 (2005): 8828-8834. PubMed: 15994776.
- Hulswit, R. J. G., C. A. M. de Haan and B.-J. Bosch. "Coronavirus Spike Protein and Tropism Changes." <u>Adv.</u> <u>Virus Res.</u> 96 (2016): 29-57. PubMed: 27712627.
- 6. <u>WHO</u>

ATCC[®] is a trademark of the American Type Culture Collection.

