Modified pαH Vector Containing the SARS-Related Coronavirus 2, Wuhan-Hu-1 Spike Glycoprotein

Catalog No. NR-52564

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

For research use only. Not for human use.

Contributor:

Barney Graham, Deputy Director and Chief, Vaccine Research Center, National Institutes of Health, Bethesda, Maryland, USA

Manufacturer:

BEI Resources

Product Description:

Note: The label on the vial is incorrect; the expressed protein is untagged.[1,2]

NR-52564 expresses the full-length, unmodified S glycoprotein, and is intended for producing pseudotyped particles/pseudovirions.[1] NR-52564 is not intended for recombinant protein expression.

The vector for the spike (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 subcloning into the pαH mammalian expression vector, which was modified by subcloning a T4 fibritin trimerization motif, HRV3C protease cleavage site, and the tags Twin-Strep-tag® and octa-histidine downstream of the open reading frame.[1,2] However, NR-52564 expresses the full-length S protein without any additional tags or modifications. NR-52564 contains the beta-lactamase gene, *bla*, to provide transformant selection through ampicillin resistance in *Escherichia coli (E. coli)*. NR-52564 is also referred to as VRC7475.[1] The resulting size of the plasmid is approximately 8570 base pairs. The complete plasmid sequence and map (Figure 1) 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.[3]

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.

Packaging/Storage:

NR-52564 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: Modified pαH Vector Containing the SARS-Related Coronavirus 2, Wuhan-Hu-1 Spike Glycoprotein, NR-52564."

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:

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 www.beiresources.org.

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 its derivatives must contact the contributor to determine if a

BEI Resources
www.beiresources.org

E-mail: contact@beiresources.org
Tel: 800-359-7370
Fax: 703-365-2898

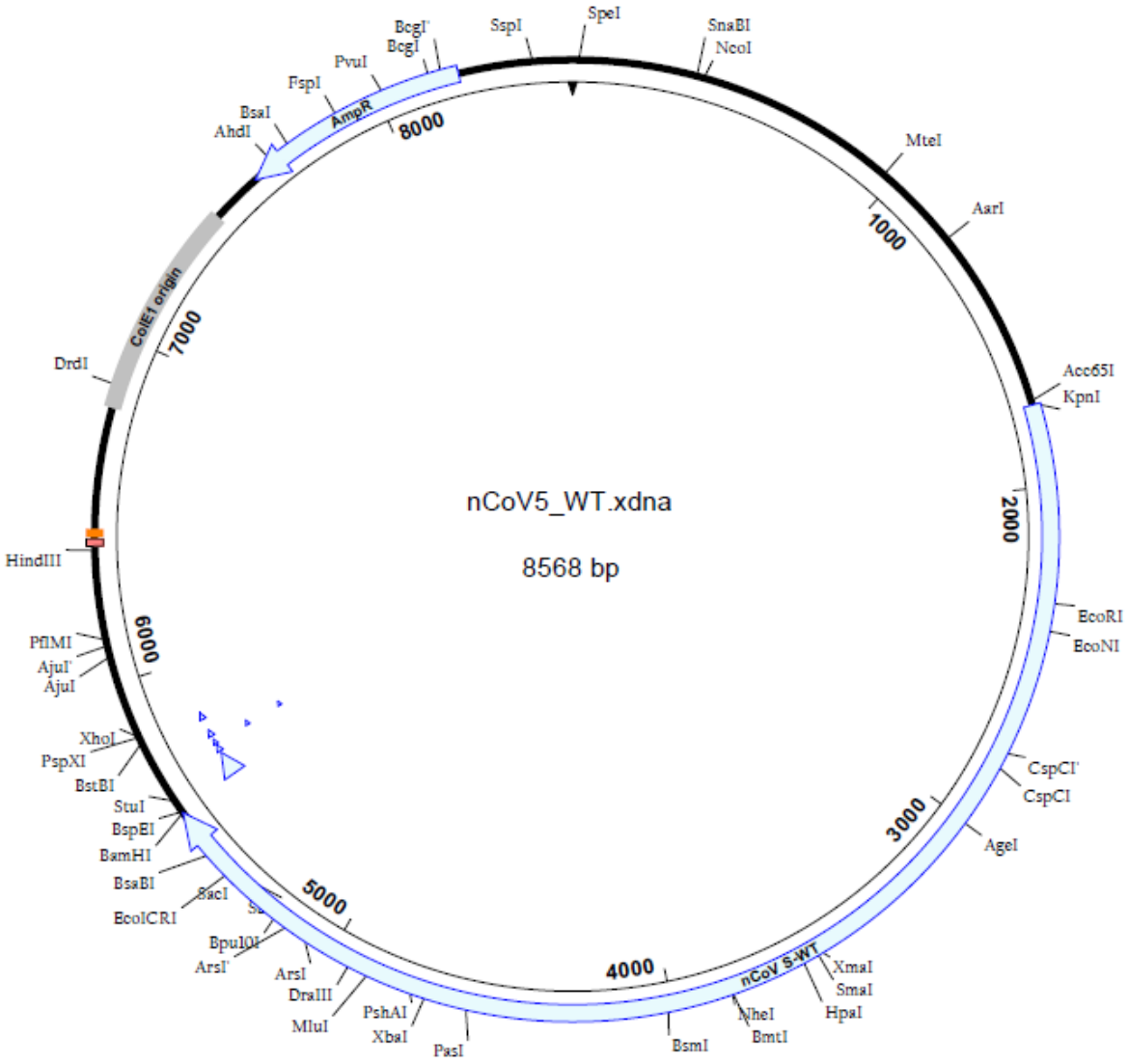
license is required. U.S. Government contractors may need a license before first commercial sale.

References:

1. Graham, B., Personal Communication.
2. Wrapp, D., et al. "Cryo-EM Structure of the 2019-nCoV Spike in the Prefusion Conformation." Science 367 (2020): 1260-1263. PubMed: 32075877.
3. Hulswit, R. J. G., C. A. M. de Haan and B.-J. Bosch. "Coronavirus Spike Protein and Tropism Changes." Adv. Virus Res. 96 (2016): 29-57. PubMed: 27712627.

Support Provided by NIAID

Figure 1: Plasmid Map of NR-52564



BEI Resources
www.beiresources.org

E-mail: contact@beiresources.org
Tel: 800-359-7370
Fax: 703-365-2898