

## Vector pαH Containing the Human Coronavirus, NL63 Recombinant Spike Ectodomain Gene

### Catalog No. NR-54976

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

**For research use only. Not for use in humans.**

#### Contributor:

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

#### Manufacturer:

BEI Resources

#### Product Description:

NR-54976 is an expression vector containing the human coronavirus, NL63 recombinant spike ectodomain gene insert (codon optimized) encoding S1 ectodomain residues 1-1291 (GenPept: [Q6Q1S2.1](#)) linked to C-terminal T4 fibrin trimerization domain (foldon), an HRV3C cleavage site, octa His-tag and 2X Strep-tag<sup>®</sup> II. Recombinant S ectodomain trimer is stabilized in the prefusion conformation by two proline substitutions (S1052P and I1053P).<sup>1,2</sup> NR-54976 contains the beta-lactamase gene, *bla*, to provide transformant selection through ampicillin resistance in *Escherichia coli* (*E. coli*). NR-54976 is also referred to as VRC7498.<sup>1</sup> The plasmid is approximately 8080 base pairs and 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.<sup>3,4</sup>

#### 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-54976 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 pαH Containing the Human Coronavirus, NL63 Recombinant Spike Ectodomain Gene, NR-54976."

#### 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](#). 6th ed. Washington, DC: U.S. Government Printing Office, 2020.

#### 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](http://www.beiresources.org).

While BEI Resources uses reasonable efforts to include accurate and up-to-date information on this product sheet, neither ATCC<sup>®</sup> 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<sup>®</sup> 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<sup>®</sup> 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<sup>®</sup>, 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 license is required. U.S. Government contractors may need a license before first commercial sale.

NR-54976 is claimed in U.S. Provisional Patent Application number 16/344774 and Global Patent Index publication number EP3532095 and the continuations, continuations-in-part, re-issues and foreign counterparts thereof. NR-54976 cannot be transferred to for-profit entities. For-profit entities wishing to obtain this material must inquire to NIAID's Technology Transfer and Intellectual Property Office with reference to NIH Ref. No. E-234-2016 by e-mailing [jstein@mail.nih.gov](mailto:jstein@mail.nih.gov) and [matthew.reiber@nih.gov](mailto:matthew.reiber@nih.gov). The

Scripps Research Institute and Dartmouth College have rights to this material.

**References:**

1. Graham, B., Personal Communication.
2. Graham, B., et al. "Prefusion Coronavirus Spike Proteins and their Use." [U.S. Provisional Patent Application 16/344774](#), 2020.
3. Wrapp, D., et al. "Cryo-EM Structure of the 2019-nCoV Spike in the Prefusion Conformation." *Science* 367 (2020): 1260-1263. PubMed: 32075877.
4. 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.

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

