

SUPPORTING INFECTIOUS DISEASE RESEARCH

Product Information Sheet for NR-54977

Vector pαH Containing the Human Coronavirus, 229E Recombinant Spike Ectodomain Gene

Catalog No. NR-54977

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

For research use only. Not for use in humans.

Contributor:

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Manufacturer:

BEI Resources

Product Description:

NR-54977 is an expression vector containing the human coronavirus, 229E recombinant spike ectodomain gene insert (codon optimized) encoding S1 ectodomain residues 1-1108 (GenPept: AOG74783.1) linked to C-terminal T4 fibritin trimerization domain (foldon), an HRV3C cleavage site, octa His-tag and 2X Strep-tag® II. Recombinant S ectodomain trimer is stabilized in the prefusion conformation by two proline substitutions (I869P and I870P)^{1,2} NR-54977 contains the beta-lactamase gene, *bla*, to provide transformant selection through ampicillin resistance in *Escherichia coli* (*E. coli*). NR-54977 is also referred to as VRC7449.¹ The plasmid is approximately 7530 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.^{3,4}

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-54977 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, 229E Recombinant Spike Ectodomain Gene, NR-54977."

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.

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NR-54977 is claimed in U.S. Provisional Patent Application number 16/344774 and Global Patent Index publication number EP 3532095 and the continuations, continuations-in-part, re-issues and foreign counterparts thereof. NR-54977 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 and matthew.reiber@nih.gov. The Scripps Research Institute and Dartmouth College have rights to this material.

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References:

- 1. Graham, B., Personal Communication.
- Graham, B., et al. "Prefusion Coronavirus Spike Proteins and their Use." <u>U.S. Provisional Patent Application</u> 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.
- Hulswit, R. J. G., C. A. M. de Haan and B.-J. Bosch. "Coronavirus Spike Protein and Tropism Changes." <u>Adv. Virus Res.</u> 96 (2016): 29-57. PubMed: 27712627.

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