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SUPPORTING INFECTIOUS DISEASE RESEARCH

Vector pαH Containing the SARS Coronavirus, Recombinant Spike Ectodomain Gene

Catalog No. NR-54975

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For research use only. Not for use in humans.

Contributor:

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Manufacturer:

BEI Resources

Product Description:

NR-54975 is an expression vector containing the SARS recombinant spike ectodomain gene coronavirus. insert (codon optimized) encoding S1 ectodomain residues 1-1190 (GenPept: AAP41037.1) linked to C-terminal T4 fibritin trimerization domain (foldon), an HRV3C cleavage site, octa His-tag and Strep-tag[®] II. Recombinant S ectodomain trimer is stabilized in the prefusion conformation by two proline substitutions (K968P and V969P).^{1,2} NR-54975 contains the beta-lactamase gene, bla, to provide transformant selection through ampicillin resistance in Escherichia coli (E. coli). NR-54975 is also referred to as VRC7575.1 The plasmid is approximately 7710 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. <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-54975 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 $p\alpha H$ Containing the SARS Coronavirus, Recombinant S Ectodomain Gene, NR-54975."

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 (BMBL)</u>. 6th ed. Washington, DC: U.S. Government Printing Office, 2020.

Disclaimers:

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NR-54975 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-54975 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.

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References:

- 1. Graham, B., Personal Communication.
- 2. 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." <u>Science</u> 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.</u> <u>Virus Res.</u> 96 (2016): 29-57. PubMed: 27712627.

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