

Product Information Sheet for NR-54978

Modified pαH Vector Containing the Human Coronavirus, HKU1 Spike Glycoprotein

Catalog No. NR-54978

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:

The vector for the spike (S) glycoprotein gene from human coronavirus, HKU1 was designed by codon optimizing the fulllength S sequence (residues 1 to 1277; GenPept: Q0ZME7) for mammalian expression and subcloning into the pαH mammalian expression vector, which was modified by subcloning a HRV3C protease cleavage site, T4 foldon trimerization domain, and the octa-histidine and Strep-tag® II tags downstream of the open reading frame.1,2 recombinant protein is stabilized by substitution at the furin S1/S2 cleavage site (RRKRR→GGSGS; residues 752 to 756) and NL→PP mutations (residues 1067 and 1068). NR-54978 contains the beta-lactamase gene, bla, to provide transformant selection through ampicillin resistance in Escherichia coli (E. coli). NR-54978 is also referred to as VRC7576.1 The resulting size of the plasmid is approximately 7970 base pairs. The complete plasmid sequence and map are provided on the BEI Resources webpage. The plasmid was produced in E. coli and extracted.

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-54978 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 p α H Vector Containing the Human Coronavirus, HKU1 Spike Glycoprotein, NR-54978."

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

Disclaimers:

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NR-54978 is claimed in U.S. Provisional Patent Application number 16/344774 and Global Patent Index publication number EP 3532095 and the continuations, continuations-inpart, re-issues and foreign counterparts thereof. NR-54978 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.
- 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.

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