

Spike Glycoprotein (Stabilized) from SARS-Related Coronavirus 2, Wuhan-Hu-1 with C-Terminal Histidine Tag, Recombinant from HEK293F Cells

Catalog No. NR-52397

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Contributor:

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Manufacturer:

BEI Resources

Product Description:

A recombinant form of the spike (S) glycoprotein from severe acute respiratory syndrome-related coronavirus 2 (SARS-CoV-2), Wuhan-Hu-1 (GenPept: [QJE37812](#)) was produced by transfection of purified plasmid in human embryonic kidney HEK293F cells and purified by immobilized metal affinity chromatography.^{1,2,3} NR-52397 lacks the signal sequence and contains 1196 residues (ectodomain) of the SARS-CoV-2 spike glycoprotein; the recombinant protein was modified to remove the polybasic S1/S2 cleavage site (RRAR to A; residues 682 to 685), stabilized with a pair of mutations (K986P and V987P, wild type numbering; GenPept: [YP_009724390](#)) and includes a thrombin cleavage site, T4 foldon trimerization domain and C-terminal hexa-histidine tag.^{1,4} The predicted protein sequence is shown in Figure 1. NR-52397 has a theoretical molecular weight of 137,600 daltons. The crystal structure for trimeric S glycoprotein from SARS-CoV-2 has been solved at 3.46 Å resolution (PDB: [6VSB](#)).⁵

Note: For a detailed protocol and list of related items, see <https://labs.icahn.mssm.edu/krammerlab/covid-19/>

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.⁶

Material Provided:

Each vial contains approximately 100 µL of NR-52397 in phosphate buffered saline (PBS). The concentration, expressed as mg per mL, is shown on the Certificate of Analysis.

Note: The long-term stability of this preparation is not known at this time. It is recommended that users confirm the

activity of the product if not used within three months of receipt.

Packaging/Storage:

NR-52397 was packaged aseptically in cryovials. The product is provided on dry ice and should be stored at -20°C or colder immediately upon arrival. Freeze-thaw cycles should be avoided.

Functional Activity:

NR-52397 reacts with monoclonal anti-histidine tag in western blot analysis. NR-52397 is intended for western blot, ELISA and animal vaccination.^{1,3}

Citation:

Acknowledgment for publications should read “The following reagent was produced under HHSN272201400008C and obtained through BEI Resources, NIAID, NIH: Spike Glycoprotein (Stabilized) from SARS-Related Coronavirus 2, Wuhan-Hu-1 with C-Terminal Histidine Tag, Recombinant from HEK293F Cells, NR-52397.”

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/bmb15/index.htm.

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References:

1. Krammer, F., F. Amanat and S. Strohmeier, Personal Communication.

2. Walls, A. C., et al. "Structure, Function, and Antigenicity of the SARS-CoV-2 Spike Glycoprotein." *Cell* 181 (2020): 281-292. PubMed: 32155444.
 3. Amanat, F., et al. "A Serological Assay to Detect SARS-CoV-2 Seroconversion in Humans." *Nat. Med.* (2020): *in press*. PubMed: 32398876.
 4. Wu, F., et al. "A New Coronavirus Associated with Human Respiratory Disease in China." *Nature* 579 (2020): 265-269. PubMed: 32015508.
 5. Wrapp, D., et al. "Cryo-EM Structure of the 2019-nCoV Spike in the Prefusion Conformation." *Science* 367 (2020): 1260-1263. PubMed: 32075877.
 6. 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.

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Figure 1 – Predicted Protein Sequence

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1  CVNLTTRTQL PPAYTNSFTR GVYYPDKVFR SSVLHSTQDL FLPPFSNVTW
51  FHAIHVSQTN GTKRFDPVPL PFNDGVYFAS TEKSNIIRGW IFGTTLDSKT
101 QSLLIIVNAT NVVIKVCVFQ FCNDPFLGVY YHKNNKSWME SEFRVYSSAN
151 NCTFEYVSQP FLMDLEGKQG NFKNLREFVF KNIDGYFKIY SKHTPINLVR
201 DLPQGFSALE PLVDLPIGIN ITRFQTLAL HRSYLTPGDS SSGWTAGAAA
251 YYVGYLQPRF FLLKYNENGT ITDAVDCALD PLSETKCTLK SFTVEKGIYQ
301 TSNFRVQPTF SIVRFPNITN LCPFGEVFNA TRFASVYAWN RKRISNCVAD
351 YSVLYNSASF STFVKYGVSP TKLNDLCFTN VYADSFVIRG DEVRQIAPGQ
401 TGKIADYNYK LPDDFTGCVI AWNSNLDLSD VGGNYNYLYR LFRKSNLKPFL
451 ERDISTEIQY AGSTPCNGVE GFNCYFPLQS YGFQPTNGVG YQPYRVVVL
501 FELLHAPATV CGPKKSTNLV KNCVNFNFN GLTGTGVLTE SNKKFLPFQQ
551 FGRDIADTTD AVRDPQTLFI LDITPCSFSG VSVITPGTNT SNQVAVLYQD
601 VNCTEVPVAI HADQLTPTWR VYSTGSNVFQ TRAGCLIGAE HVNNSYECDI
651 PIGAGICASY QTQTNPASV ASQSIIAYTM SLGAENSVAY SNNNSIAIPTN
701 FTISVTTEIL PVSMTKTSVD CTMYICGDST ECSNLLLQYG SFCTQLNRAL
751 TGIAVEQDKN TQEVFAQVKQ IYKTPPIKDF GGFNFSQILP DPSKPSKRSF
801 IEDLLFNKVT LADAGFIKQY GDCLGDIAAR DLICAQKFNG LTVLPPLLLTD
851 EMIAQYTSAL LAGTITSGWT FGAGAALQIP FAMQMAYRFN GIGVTQNVLY
901 ENQKLIANQF NSAIGKIQDS LSSTASALGK LQDVVNQNAQ ALNLTLVKQLS
951 SNFGAISSVL NDILSRDPP EAEVQIDRLI TGRLQSLQTY VTQQLIRAAE
1001 IRASANLAAT KMSECVLGQS KRVDFCGKGY HLMSFPQSAP HGVVFLHVTY
1051 VPAQEKNFIT APAICHGKA HFPREGVFSV NGTHWFVTQR NFEYEQIITF
1101 DNTFVSGNCD VVIGIVNNTV YDPLQPELDS FKEELDKYFK NHTSPDVLG
1151 DISGINASVV NIQKEIDRLN EVAKNLNESL IDLQELGKYE QYIKWPSGRL
1201 VPRGSPGSGY IPEAPRDGQA YVRKDGWVLL LSTFLGHHHH HH
    
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Spike ectodomain – **Residues 1 to 1196** (represents WT amino acid residues 15 to 1213)

RRAR to A substitution of S1/S2 cleavage site – Residue 671

KV to PP stabilizing mutations – Residues 969 and 970

Thrombin cleavage site – Residues 1200 to 1205

T4 foldon trimerization domain – Residues 1206 to 1236

Hexa-histidine tag – Residues 1237 to 1242