

Spike Glycoprotein (Stabilized) from SARS-Related Coronavirus 2, Wuhan-Hu-1 HexaPro with C-Terminal Histidine and Twin-Strep® Tags, Recombinant from CHO Cells

Catalog No. NR-53769

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For research use only. Not for use in humans.

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: [QHD43416](#)) was produced by transfection of purified plasmid in Chinese hamster ovary (CHO) cells, purified by immobilized nickel affinity chromatography and dialyzed into buffer.^{1,2,3} NR-53769 lacks the signal sequence and contains 1194 residues (ectodomain) of the SARS-CoV-2 S glycoprotein; the recombinant protein was stabilized by substitution at the furin S1/S2 cleavage site (RRAR→GSAS; residues 682 to 685) and KV→PP mutations (residues 986 and 987) as well as the additional proline substitutions that create the HexaPro variant (F817P, A892P, A899P and A942P), and includes a T4 foldon trimerization domain, HRV3C protease cleavage site, and C-terminal octa-histidine and Twin-Strep® (TST) tags.^{1,2,3,4} The predicted protein sequence is shown in Figure 1. NR-53769 has a theoretical molecular weight of 140,700 daltons. The crystal structure for trimeric S glycoprotein from SARS-CoV-2 has been solved at 3.46 Å resolution (PDB: [6VSB](#)).⁵

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 80 µL of NR-53769 in 20 mM Tris, pH 8.0 and 500 mM NaCl. The concentration, expressed as mg per mL, is shown on the Certificate of Analysis.

Packaging/Storage:

NR-53769 was packaged aseptically in cryovials. The product is provided on dry ice and should be stored at -60°C or colder immediately upon arrival. Freeze-thaw cycles should be avoided.

Citation:

Acknowledgment for publications should read "The following reagent was obtained through BEI Resources, NIAID, NIH: Spike Glycoprotein (Stabilized) from SARS-Related Coronavirus 2, Wuhan-Hu-1 HexaPro with C-Terminal Histidine and Twin-Strep® Tags, Recombinant from CHO Cells, NR-53769."

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; see www.cdc.gov/biosafety/publications/bmbl5/index.htm.

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NR-53769 was produced from a plasmid (available as BEI Resources NR-53587) which is claimed in U.S. Provisional Patent Application numbers 62/412,703, 62/972,886 and 63/032,502 and the continuations, continuations-in-part,

re-issues and foreign counterparts thereof. The University of Texas-Austin, the National Institutes of Health and Dartmouth College all have rights to this material.

References:

1. McLellan, J., Personal Communication.
2. Wu, F., et al. "A New Coronavirus Associated with Human Respiratory Disease in China." *Nature* 579 (2020): 265-269. PubMed: 32015508.
3. Hsieh, C. -L., et al. "Structure-Based Design of Prefusion-Stabilized SARS-CoV-2 Spikes." *Science* 369 (2020): 1501-1505. PubMed: 32703906.

4. Walls, A. C., et al. "Structure, Function, and Antigenicity of the SARS-CoV-2 Spike Glycoprotein." *Cell* 181 (2020): 281-292. PubMed: 32155444.
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 FLPFFSNVTW
51  FHAIHVSQTN GTKRFDNPVL PFNDGVYFAS TEKSNIIRGW IFGTTLDSTK
101 QSLIVNNAT NVVIKVECFQ FCNDPFLGVY YHKNNKSWME SEFRVYSSAN
151 NCTFEYVSQP FLMDLEGKQG NFKNLREFVF KNIDGYFKIY SKHTPINLVR
201 DLPQGFSALE PLVDLPIGIN ITRFQTLLAL HRSYLTGDS SSGWTAGAAA
251 YYVGYLQPR FLLKYNENGT ITDAVDCALD PLSETKCTLK SFTVEKGIYQ
301 TSNFRVQPT SIVRFPNITN LCPFGEVFNA TRFASVYAWN RKRISNCVAD
351 YSVLYNSASF STFCKYGVSP TKLNDLCFTN VYADSFVIRG DEVRQIAPGQ
401 TGKIADYNYK LPDDFTGCVI AWNSNNLDSK VGGNYNYLYR LFRKSNLKP
451 ERDISTEIQ AGSTPCNGVE GFNCYFPLQS YGFQPTNGVG YQPYRVVLS
501 FELLHAPATV CGPKKSTNLV KNKCVNFNEN GLTGTGVLTE SNKKFLPFQ
551 FGRDIADTTD AVRDPQTL EITPCSFSG VSVITPGTNT SNQVAVLYQD
601 VNCTEVPVAI HADQLTPTWR VYSTGSNVFQ TRAGCLIGAE HVNNSYECDI
651 PIGAGICASY QTQNSPGSA SSVASQSI IATMSLGAENS VAYSNNIAI
701 PTNFTISVTT EILPVSMTKT SVDCTMYICG DSTECSNLLL QYGSFCTQLN
751 RALTGIAVEQ DKNTQEVFAQ VKQIYKTPPI KDFGGFNFSQ ILPDPSKPSK
801 RSPIEDLLFN KVTADAGFI KQYGDCLGDI AARDLICAQK FNGTLVLPPL
851 LTDEMIAQYT SALLAGTITS GWTFGAGPAL QIPFPMQ MAY RFNGIGVTQN
901 VLYENQKLI NQFNSAIGKI QDSLSTPSA LGKLQDVVNQ NAQALNTLVK
951 QLSSNFGAIS SVLNDILSRL DPPEAEVQID RLITGRLQSL QTYVTQQLIR
1001 AAEIRASANL AATKMSECVL GQSKRVDFCG KGYHLMSFPQ SAPHGVVFLH
1051 VTYVPAQEK FTTAPAICHD GKAHFPREGV FVSNGTHWV TQRNFYEPQI
1101 ITTDNTFVSG NCDVVIGIVN NTVYDPLQPE LDSFKEELDK YFKNHTSPDV
1151 DLGDISGINA SVVNIQKEID RLNEVAKNLN ESLIDLQELG KYEQSGSYIP
1201 EAPRDGQAYV RKDGEWVLLS TFLGRSLEVL FQGPQH HHHH HHHSAWSHPQ
1251 FEKGGGSGG GSGGSAWSHP QFEK

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Spike ectodomain – **Residues 1 to 1194** (represents WT amino acid residues 15 to 1208)

RRAR to GSAS substitution of S1/S2 cleavage site – Residues 668 to 671

Hexapro mutations: Residues 803, 878, 885, 928, 972 and 973

T4 foldon trimerization domain – Residues 1197 to 1223

HRV3C protease cleavage site – Residues 1227 to 1234

Octa-histidine tag – Residues 1236 to 1243

Twin-Strep® tag – Residues 1246 to 1274