Spike Glycoprotein (Stabilized) from SARS-Related Coronavirus 2, Wuhan-Hu-1 with C-Terminal Histidine Tag, Recombinant from HEK293 Cells

Catalog No. NR-53257

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Contributor and Manufacturer:

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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: YP_009724390) was produced in human embryonic kidney HEK293 cells, purified by immobilized metal affinity chromatography and dialyzed into buffer.[1,2,3] NR-53257 contains a cleaved N-terminal mu-phosphatase signal sequence and 1198 residues (ectodomain) of the SARS-CoV-2 spike glycoprotein.[4] The recombinant protein is stabilized by substitution at the furin S1/S2 cleavage site (RRAR to GSAS; residues 682 to 685) and stabilizing mutations (K986P and V987P, wild type numbering) and includes a C-terminal tobacco etch virus (TEV)-cleavage site, glycine-serine linker, T4 foldon trimerization domain and octa-histidine tag.[1,2,3] The predicted protein sequence is shown in Figure 1.[1] NR-53257 has a theoretical molecular weight of 138,600 daltons. The crystal structure for trimeric S glycoprotein from SARS-CoV-2 has been solved at 2.8 Å resolution (PDB: 6VXX).

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.[5]

Material Provided:

Each vial contains approximately 325 µL of NR-53257 in 50 mM Tris (pH 8.0), 150 mM NaCl and 0.25% L-histidine. The concentration, expressed as mg per mL, is shown on the Certificate of Analysis. Note: It is recommended that the vial be thawed on ice and centrifuged at 14,000 rpm for 5 minutes at 4°C prior to opening.[1]

Packaging/Storage:

NR-53257 was packaged aseptically in cryovials. The product is provided on dry ice and should be stored at -60°C or colder immediately upon arrival. Do not store at 2-8°C for longer than 12 hours.[1] 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 with C-Terminal Histidine Tag, Recombinant from HEK293 Cells, NR-53257."

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/bmbl5/index.htm.

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References:

1. Carter, L., Personal Communication.

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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. Wu, F., et al. "A New Coronavirus Associated with Human Respiratory Disease in China." Nature 579 (2020): 265-269. PubMed: 32015508.
4. Berrow, N. S., et al. "A Versatile Ligation-Independent Cloning Method Suitable for High-Throughput Expression Screening Applications." Nucleic Acids Res. 35 (2007): e45. PubMed: 17317681.
5. 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.

Support Provided by NIAID

Figure 1 – Predicted Protein Sequence

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1    ETGTQCVNLT TRTQLPPAYT NSFTRGVYYP DKVFRSSVLH STQDLFLPFF
51   SNVTWFHAIH VSGTNGTKRF DNPVLPFNDG VYFASTEKSN IIRGWIFGTT
101  LDSKTQSLLI VNNATNVVIK VCEFQFCNDP FLGVYYHKNN KSWMESEFRV
151  YSSANNCTFE YVSQPFLMDL EGKQGNFKNL REFVFKNIDG YFKIYSKHTP
201  INLVRDLPQG FSALEPLVDL PIGINITRFQ TLLALHRSYL TPGDSSSGWT
251  AGAAAYYVGY LQPRTFLLKY NENGTITDAV DCALDPLSET KCTLKSFTVE
301  KGIYQTSNFR VQPTESIVRF PNITNLCPFG EVFNATRFAS VYAWNRKRIS
351  NCVADYSVLY NSASFSTFKC YGVSPTKLND LCFTNVYADS FVIRGDEVRQ
401  IAPGQTGKIA DYNYKLPDDF TGCVIAWNSN NLDSKVGGNY NYLYRLFRKS
451  NLKPFERDIS TEIYQAGSTP CNGVEGFNCY FPLQSYGFQP TNGVGYQPYR
501  VVVLSFELLH APATVCGPKK STNLVKNKCV NFNFNGLTGT GVLTESNKKF
551  LPFQQFGRDI ADTTDAVRDP QTLEILDITP CSFGGVSVIT PGTNTSNQVA
601  VLYQDVNCTE VPVAIHADQL TPTWRVYSTG SNVFQTRAGC LIGAEHVNNS
651  YECDIPIGAG ICASYQTQTN SPSGAGSVAS QSIIAYTMSL GAENSVAYSN
701  NSIAIPTNFT ISVTTEILPV SMTKTSVDCT MYICGDSTEC SNLLLQYGSF
751  CTQLNRALTG IAVEQDKNTQ EVFAQVKQIY KTPPIKDFGG FNFSQILPDP
801  SKPSKRSFIE DLLFNKVTLA DAGFIKQYGD CLGDIAARDL ICAQKFNGLT
851  VLPPLLTDEM IAQYTSALLA GTITSGWTFG AGAALQIPFA MQMAYRFNGI
901  GVTQNVLYEN QKLIANQFNS AIGKIQDSLS STASALGKLQ DVVNQNAQAL
951  NTLVKQLSSN FGAISSVLND ILSRLDPPEA EVQIDRLITG RLQSLQTYVT
1001 QQLIRAAEIR ASANLAATKM SECVLGQSKR VDFCGKGYHL MSFPQSAPHG
1051 VVFLHVTYVP AQEKNFTTAP AICHDGKAHF PREGVFVSNG THWFVTQRNF
1101 YEPQIITTDN TFVSGNCDVV IGIVNNTVYD PLQPELDSFK EELDKYFKNH
1151 TSPDVDLGDI SGINASVVNI QKEIDRLNEV AKNLNESLID LQELGKYEQY
1201 IKGSGRENLY FQGGGGSGYI PEAPRDGQAY VRKDGEWVLL STFLGHHHHH
1251 HHH
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Cleaved signal sequence – Residues 1 to 3
Spike ectodomain – Residues 5 to 1202 (representing residues 14 to 1211)
TEV cleavage site – Residues 1207 to 1212
Gly-Ser linker – Residues 1213 to 1218 and 1245
T4 foldon trimerization domain – Residues 1219 to 1244
Octa-histidine tag – Residues 1246 to 1253

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