

Spike Glycoprotein (Stabilized) from SARS-Related Coronavirus 2, with C-Terminal Histidine and Twin-Strep® Tags, Recombinant from HEK293F Cells

Catalog No. NR-56132

This reagent is the tangible property of the U.S. Government.

For research use only. Not for use in humans.

Contributor:

S. Mark Tompkins, Ph.D., Professor, Department of Infectious Diseases, Center for Vaccines and Immunology (CVI), College of Veterinary Medicine, University of Georgia (UGA), Athens, Georgia, USA, supported under government contract HHSN272201400004C

Manufacturer:

UGA Bioexpression and Fermentation Facility

Product Description:

Note: The Strep tag designation on the label is incorrect; the correct Strep tag is a Twin-Strep® tag.

A recombinant form of the spike (S) glycoprotein from severe acute respiratory syndrome-related coronavirus 2 (SARS-CoV-2; GenPept: [QIZ14569.1](#)) was produced by transient transfection into human embryonic kidney HEK293F cells and purified by immobilized metal affinity chromatography.^{1,2,3} NR-56132 lacks the signal sequence and contains 1196 residues (ectodomain) of the SARS-CoV-2 spike glycoprotein; the recombinant protein was stabilized by substitution at the furin S1/S2 cleavage site (RRAR→GSAS; residues 670 to 673) and KV→PP mutations (residues 974 and 975), and includes a HRV3C protease cleavage site, T4 foldon trimerization domain, and C-terminal octa-histidine and Twin-Strep® tags. The predicted protein sequence is shown in Figure 1.¹ NR-56132 has a theoretical molecular weight of 140.9 kilodaltons.

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.⁴

Material Provided:

Each vial contains approximately 100 µL of NR-56132 in phosphate buffered saline (PBS). The concentration, expressed as milligrams per milliliter, is shown on the Certificate of Analysis.

Packaging/Storage:

NR-56132 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.

Citation:

Acknowledgment for publications should read "The following reagent was obtained through BEI Resources, NIAID, NIH: Spike Glycoprotein (Stabilized) from SARS-Related Coronavirus 2, with C-Terminal Histidine and Twin-Strep® Tags, Recombinant from HEK293F Cells, NR-56132."

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.

Disclaimers:

You are authorized to use this product for research use only. It is not intended for human use.

Use of this product is subject to the terms and conditions of the BEI Resources Material Transfer Agreement (MTA). The MTA is available on our Web site at www.beiresources.org.

While BEI Resources uses reasonable efforts to include accurate and up-to-date information on this product sheet, neither ATCC® nor the U.S. Government makes any warranties or representations as to its accuracy. Citations from scientific literature and patents are provided for informational purposes only. Neither ATCC® nor the U.S. Government warrants that such information has been confirmed to be accurate.

This product is sent with the condition that you are responsible for its safe storage, handling, use and disposal. ATCC® and the U.S. Government are not liable for any damages or injuries arising from receipt and/or use of this product. While reasonable effort is made to ensure authenticity and reliability of materials on deposit, the U.S. Government, ATCC®, their suppliers and contributors to BEI Resources are not liable for damages arising from the misidentification or misrepresentation of products.

Use Restrictions:

This material is distributed for internal research, non-commercial purposes only. This material, its product or its derivatives may not be distributed to third parties. Except as performed under a U.S. Government contract, individuals contemplating commercial use of the material, its products or its derivatives must contact the contributor to determine if a license is required. U.S. Government contractors may need a license before first commercial sale.

References:

1. Tompkins, S. M., 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.* 26 (2020): 1033-1036. PubMed: 32398876.
4. 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.

ATCC® is a trademark of the American Type Culture Collection.



Figure 1: Predicted Protein Sequence

```

1  SQCVNLTTRT QLPPAYTNSF TRGVYYPDKV FRSSVLHSTQ DLFLPFFSNV
51  TWFHAIHVSG TNGTKRFDNP VLPFNDGVYF ASTEKSNIIR GWIFGTTLDS
101 KTQSLIVNN ATNVVIKVE FQFCNDPFLG VYYHKNNKSW MESEFRVYSS
151 ANNCTFEYVS QPFLMDLEGK QGNFKNLREF VFKNIDGYFK IYSKHTPINL
201 VRDLPQGFSA LEPLVDLPIG INITRFQTL ALHRSYLTPG DSSSGWTAGA
251 AAYYVGYLQP RTFLLKYEN GTITDAVDCA LDPLSETKCT LKSFTVEKGI
301 YQTSNFRVQP TESIVRFPNI TNLCPFGEVF NATRFASVYA WNRKRISNCV
351 ADYSVLYNSA SFSTFKCYGV SPTKLNLCF TNVYADSFVI RGDEVRQIAP
401 GQTGKIADYN YKLPDDFTGC VIAWNSNLD SKVGGNYNYL YRLFRKSNLK
451 PFERDISTEI YQAGSTPCNG VEGFCYFPL QSYGFQPTNG VGYQPYRVVV
501 LSFELLHAPA TVCGPKKSTN LVKNKCVNFN FNGLTGTGVL TESNKKFLPF
551 QQFGRDIADT TDAVRDPQTL EILDITPCSF GGVSVITPGT NTSNQVAVLY
601 QDVNCTEVPV AIHADQLTPT WRVYSTGSNV FQTRAGCLIG AEHVNNSYEC
651 DIPIGAGICA SYQTQTNSPG SASSVASQSI IAYTMSLGAE NSVAYSNSI
701 AIPTNFTISV TTEILPVSMT KTSVDCTMYI CGDSTECNL LLQYGSFCTQ
751 LNRALTGIAV EQDKNTQEVF AQVKQIYKTP PIKDFGGFNF SQILPDPSKP
801 SKRSFIEDLL FNKVTLADAG FIKQYGDCLG DIAARDLICA QKFNGLTVLP
851 PLLTDEMIAQ YTSALLAGTI TSGWTFGAGA ALQIPFAMQM AYRFNGIGVT
901 QNVLYENQKL IANQFNSAIG KIQDSLSTA SALGKLQDVV NQNAQALNTL
951 VKQLSSNFGA ISSVLNDILS RLDPEAEVQ IDRLITGRLQ SLQTYVTQQL
1001 IRAAEIRASA NLAATKMSEC VLGQSKRVDF CGKGYHLMSF PQSAPHGVVF
1051 LHVTYVPAQE KNFTTAPAIC HDGKAHFPRE GVFVSNGTHW FVTQRNFYEP
1101 QIITDNTFV SGNCDVIGI VNNTVYDPLQ PELDSFKEEL DKYFKNHTSP
1151 DVDLGDISGI NASVVNIQKE IDRLNEVAKN LNESLIDLQE LGKYEQGSY
1201 IPEAPRDGQA YVRKDGEWVL LSTFLGRSLE VLFQGPHHH HHHHSAWSH
1251 PQFEKGGGSG GGGSGGSAWS HPQFEK
    
```

Spike ectodomain – Residues 1 to 1196 (representing WT residues 13 to 1208)

RRAR to GSAS substitution of S1/S2 cleavage site – Residues 670 to 673

KV to PP stabilizing mutations – Residues 974 and 975

T4 foldon trimerization domain – Residues 1199 to 1225

HRV3C Protease cleavage site – Residues 1229 to 1236

Octa-histidine tag – Residues 1237 to 1244

Twin-Strep® tag – Residues 1248 to 1276