

Spike Glycoprotein (Stabilized) from Human Coronavirus NL63, with C-Terminal Histidine and Twin-Strep® Tags, Recombinant from HEK293 Cells

Catalog No. NR-56134

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 human coronavirus NL63, (HCoV-NL63; GenPept: [AFV53148](#)) was produced by transient transfection into human embryonic kidney HEK293 cells and purified by immobilized metal affinity chromatography.^{1,2,3} NR-56134 lacks the signal sequence and contains 1273 residues (ectodomain) of the HCoV-NL63 spike glycoprotein; the recombinant protein was stabilized by SI→PP mutations (residues 1034 and 1035), 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-56134 has a theoretical molecular weight of 149.0 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-56134 in phosphate buffered saline (PBS). The concentration, expressed as milligrams per milliliter, is shown on the Certificate of Analysis.

Packaging/Storage:

NR-56134 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 Human Coronavirus NL63, with C-Terminal Histidine and Twin-Strep® Tags, Recombinant from HEK293 Cells, NR-56134."

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. van der Hoek, L., et al. "Identification of a New Human Coronavirus." Nat. Med. 10 (2004): 368-373. PubMed: 15034574.

3. Gallagher, T. M. and M. J. Buchmeier. "Coronavirus Spike Proteins in Viral Entry and Pathogenesis." *Virology* 279 (2001): 371-374. PubMed: 11162792.
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  CNSNANLSML QLGVPDSSST IVTGLLPHTW FCANQSTSVY SANGFFYIDV
51  GNHRSAFALH TGYDANQYY IYVTNEIGLN ASVTLKICKF SRNTTFDFLS
101 NASSSFDCIV NLLFTEQLGA PLGITISGET VRLHLYNVTR TFYVPAAYKL
151 TKLSVKCYFN YSCVFSVUNA TVTVNVTTTHN GRVVNYTVCD DCNGYTDNIF
201 SVQQDGRIPN GFPPNNWFLN TNGSTLVDGV SRLYQPLRLT CLWPVPGLKS
251 STGFVYFNAT GSDVNCNGYQ HNSVVDVMRY NLNFSANSLD NLKSGVIVFK
301 TLQYDVLFCY SNSSSGVLDT TIPFGPSSQP YYCFINSTIN TTHVSTFVGI
351 LPPTVREIVV ARTGQFYING FKYFDLGFIE AVNFNVTTAS ATDFWTVAFA
401 TFVDVLVNVS ATNIQNLLYC DSPFEKLQCE HLQFGLQDGF YSANFLDDNV
451 LPETYVALPI YYQHTDINFT ATASFGGSCY VCKPHQVNIS LNGNTSVCVR
501 TSHFSIRYIY NRVKSGSPGD SSWHIYLSKG TCPFSFSKLN NFQKFKTICF
551 STVEVPGSCN FPLEATWHYT SYTIVGALYV TWSEGNISITG VPPVPSGIRE
601 FSNLVLNNCT KYNIYDYVGT GIIRSSNQSL AGGITYVSNS GNLLGFKNVS
651 TGNIFIVTPC NQPDQVAVYQ QSIIGAMTAV NESRYGLQNL LQLPNFYVVS
701 NGGNNCTTAV MTYSNFGICA DGSLIPVRPR NSSDNGISAI ITANLSIPSN
751 WTTSVQVEYL QITSTPIVVD CATYVCNGNP RCKNLLKQYT SACKTIEDAL
801 RLSAHLETND VSSMLTFDSN AFSLANVTSF GDYNLSSVLP QRNIRSSRIA
851 GRSLEDLLF SKVVTSGGLT VDVDYKSC TKGLSIADLACA QYNGIMVLP
901 GVADAERMAM YTGSLIGGMV LGGLTSAAAI PFSLALQARL NYVALQTDVL
951 QENQKILAAS FNKAINNIVA SFSSVNDAIT QTAEAIHTVT IALNKIQDVV
1001 NQQSALNHL TSQLRHNFQA ISNSIQAIYD RLDPPQADQQ VDLRLITGRLA
1051 ALNAFVSQVL NKYTEVRGSR RLAQQKINEC VKSQSNRYGF CGNGTHIFSI
1101 VNSAPDGLLF LHTVLLPTDY KNVKAWSGIC VDGIIYGYVLR QPNLVLYSDN
1151 GVFRVTSRIM FQPRLPVLS D FVQIYNCNVT FVNISRVELH TVIPDYVDVN
1201 KTLQEFQNL PKYVKPNFDL TPFNLTYLNL SSELKQLEAK TASLFQTTVE
1251 LQGLIDQINS TYVDLKLNR FENGSGYIPE APRDGQAYVR KDGEWVLLST
1301 FLGRSLEVL F QGPGHHHHHH HHSASWHPQF EKGGGSGGGG SGGSAWHPQ
1351 FEK

```

Spike ectodomain – Residues 1 to 1273 (representing WT residues 19 to 1291)

SI to PP stabilizing mutations – Residues 1034 and 1035

T4 foldon trimerization domain – Residues 1276 to 1302

HRV3C Protease cleavage site – Residues 1306 to 1313

Octa-histidine tag– Residues 1315 to 1322

Twin-Strep® tag – Residues 1325 to 1353