Spike Glycoprotein (Stabilized) from Human Coronavirus, HKU1 with C-Terminal Histidine and Avi Tags, Recombinant from HEK293F Cells

Catalog No. NR-53713

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

For research use only. Not for human use.

Contributor and Manufacturer:

Roland K. Strong, Ph.D., Professor, Division of Basic Sciences, Fred Hutchinson Cancer Research Center, Seattle, Washington, USA

Product Description:

A recombinant form of the spike (S) glycoprotein from human coronavirus (HCoV), HKU1 (GenPept: ABC70719) was produced in human embryonic kidney HEK293F cells and purified by immobilized metal affinity and size exclusion chromatography.[1,2] NR-53713 lacks the signal sequence and contains 1264 residues (ectodomain) of the HCoV spike glycoprotein; the recombinant protein was stabilized by substitution at the furin S1/S2 cleavage site (RRKRR to GGSGS; residues 752 to 756) and with a pair of mutations (N1067P and L1068P, wild type numbering), and includes a thrombin cleavage site, T4 foldon trimerization domain and C-terminal hexa-histidine tag fused to an AviTag™ BirA biotinylation acceptor sequence.[1,2] The predicted protein sequence is shown in Figure 1.[1] NR-53713 has a theoretical molecular weight of 147,070 daltons. The crystal structure for trimeric S glycoprotein from HCoV, HKU1 has been solved at 4.04 Å resolution (PDB: 5I08).[3]

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 50 µL of NR-53713 in phosphate buffered saline (PBS; pH ~ 7). The concentration, expressed as mg per mL, is shown on the Certificate of Analysis.

Packaging/Storage:

NR-53713 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, HKU1 with C-Terminal Histidine and Avi Tags, Recombinant from HEK293F Cells, NR-53713."

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.

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.

At this time, this product is only available to non-profit entities for research purposes only. Please contact Fred Hutchinson licensing for commercial use purposes and for commercial company access at mta@fredhutch.org.

References:

1. Strong, R. K., Personal Communication.

BEI Resources
www.beiresources.org

E-mail: contact@beiresources.org
Tel: 800-359-7370
Fax: 703-365-2898

2. Woo, P. C., et al. "Comparative Analysis of 22 Coronavirus HKU1 Genomes Reveals a Novel Genotype and Evidence of Natural Recombination in Coronavirus HKU1." J. Virol. 80 (2006): 7136-7145. PubMed: 16809319.
3. Kirchdoerfer, R. N., et al. "Pre-Fusion Structure of a Human Coronavirus Spike Protein. Version 2." Nature 531 (2016): 118-121. PubMed: 26935699.
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.
5. Woo, P. C., et al. "Characterization and Complete Genome Sequence of a Novel Coronavirus, Coronavirus HKU1, from Patients with Pneumonia." J. Virol. 79 (2005): 884-895. PubMed: 15613317.
6. Llanes, A., et. al. "*Betacoronavirus* Genomes: How Genomic Information has been Used to Deal with Past Outbreaks and the COVID-19 Pandemic." Int. J. Mol. Sci. 21 (2020): 4546. PubMed: 32604724.

Support Provided by NIAID

Figure 1 – Predicted Protein Sequence

```
1    VIGDFNCTNS FINDYNKTIP RISEDVVDVS LGLGTYYVLN RVYLNTTLLF
51   TGYFPKSGAN FRDLALKGSI YLSTLWYKPP FLSDFNNGIF SKVKNTKLYV
101  NNTLYSEFST IVIGSVFVNT SYTIVVQPHN GILEITACQY TMCEYPHTVC
151  KSKGSIRNES WHIDSSEPLC LFKKNFTYNV SADWLYFHFY QERGVFYAYY
201  ADVGMPTTFL FSLYLGTILS HYYVMPLTCN AISSNTDNET LEYWVTPLSR
251  RQYLLNFDEH GVITNAVDCS SSFLSEIQCK TQSFAPNTGV YDLSGFTVKP
301  VATVYRRIPN LPDCDIDNWL NNVSVPSPLN WERRIFSNCN FNLSTLLRLV
351  HVDSFSCNNL DKSKIFGSCF NSITVDKFAI PNRRRDDLQL GSSGFLQSSN
401  YKIDISSSSC QLYYSLPLVN VTINNFNPSS WNRRYGFGSF NLSSYDVVYS
451  DHCFSVNSDF CPCADPSVVN SCAKSKPPSA ICPAGTKYRH CDLDTTLYVK
501  NWCRCSCLPD PISTYSPNTC PQKKVVVGIG EHCPGLGINE EKCGTQLNHS
551  SCFCSPDAFL GWSFDSCISN NRCNIFSNFI FNGINSGTTC SNDLLYSNTE
601  ISTGVCVNYD LYGITGQGIF KEVSAAYYNN WQNLLYDSNG NIIGFKDFLT
651  NKTYTILPCY SGRVSAAFYQ NSSSPALLYR NLKCSYVLNN ISFISQPFYF
701  DSYLGCVLNA VNLTSYSVSS CDLRMGSGFC IDYALPSSGG SGSGISSPYR
751  FVTFEPFNVS FVNDSVETVG GLFEIQIPTN FTIAGHEEFI QTSSPKVTID
801  CSAFVCSNYA ACHDLLSEYG TFCDNINSIL NEVNDLLDIT QLQVANALMQ
851  GVTLSSNLNT NLHSDVDNID FKSLLGCLGS QCGSSSRSLL EDLLFNKVKL
901  SDVGFVEAYN NCTGGSEIRD LLCVQSFNGI KVLPPILSET QISGYTTAAT
951  VAAMFPPWSA AAGVPFSLNV QYRINGLGVT MDVLNKNQKL IANAFNKALL
1001 SIQNGFTATN SALAKIQSVV NANAQALNSL LQQLFNKFGA ISSSLQEILS
1051 RLDPPEAQVQ IDRLINGRLT ALNAYVSQQL SDITLIKAGA SRAIEKVNEC
1101 VKSQSPRINF CGNGNHILSL VQNAPYGLLF IHFSYKPTSF KTVLVSPGLC
1151 LSGDRGIAPK QGYFIKQNDS WMFTGSSYYY PEPISDKNVV FMNSCSVNFT
1201 KAPFIYLNNS IPNLSDFEAE LSLWFKNHTS IAPNLTFNSH INATFLDLYY
1251 EMNVIQESIK SLNSGRLVPR GSPGSGYIPE APRDGQAYVR KDGEWVLLST
1301 FLGHHHHHHG LNDIFEAQKI EWHE
```

Spike ectodomain – Residues 1 to 1264 (representing residues 14 to 1277)
RRKRR to GGSGS substitution of S1/S2 cleavage site – Residues 739 to 743
NL to PP stabilizing mutations – Residues 1054 and 1055
Thrombin cleavage site – Residues 1267 to 1272
T4 foldon trimerization domain – Residues 1276 to 1302
Hexa-histidine tag and AviTag™ – Residues 1304 to 1324

BEI Resources
www.beiresources.org

E-mail: contact@beiresources.org
Tel: 800-359-7370
Fax: 703-365-2898