

Spike Glycoprotein (Stabilized) from SARS-Related Coronavirus 2, (Lineage BA.2.75; Omicron Variant) with C-Terminal Histidine and Avi Tags, Recombinant from HEK293 Cells

Catalog No. NR-58991

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

For research use only. Not for use in humans.

Contributor:

BEI Resources

Manufacturer:

D. Noah Sather, Associate Professor, Center for Global Infectious Disease Research, Seattle Children's Research Institute, Seattle, Washington, USA

Product Description:

A recombinant form of the spike (S) glycoprotein from severe acute respiratory syndrome-related coronavirus 2 (SARS-CoV-2), lineage BA.2.75 (Omicron variant) was produced in human embryonic kidney HEK293 cells and purified by immobilized metal affinity and gel filtration chromatography.^{1,2,3,4} NR-58991 lacks the signal sequence and contains 1193 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; wild type numbering), and includes a T4 foldon trimerization domain, HRV3C protease cleavage site and C-terminal octa-histidine tag fused to an AviTag™ BirA biotinylation acceptor sequence.^{1,2,3} NR-58991 includes T19I, delL24-P26, A27S, G142D, K147E, W152R, F157L, I210V, V213G, G257S, G339H, S371F, S373P, S375F, T376A, D405N, R408S, K417N, N440K, G446S, N460K, S477N, T478K, E484A, Q498R, N501Y, Y505H, D614G, H655Y, N679K, P681H, N764K, D796Y, Q954H, and N969K mutations in the S glycoprotein as compared to the SARS-CoV-2 reference sequence (GenPept: [QHD43416](#)).^{1,5} The predicted protein sequence is shown in Figure 1. NR-58991 has a theoretical molecular weight of 139,727 daltons. The crystal structure for trimeric S glycoprotein from SARS-CoV-2 has been solved at 3.46 Å resolution (PDB: [6VSB](#)).²

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.⁶ BA.2.75 is one of several subvariant of the SARS-CoV2 Omicron variant.^{7,8} BA.2.75 has increased affinity to ACE2, compared to BA.2 and BA.4/5, resulting in its humoral evasion capability and transmission advantage. Antibody evasion might be due to the mutations in N460K, G446S and G339H.⁸

Material Provided:

Each vial contains approximately 400 microliters of NR-58991 in 10 mM HEPES, pH 7, 150 mM NaCl and 2 mM ethylenediamine-tetraacetic acid (EDTA). The concentration, expressed as mg/mL, is shown on the Certificate of Analysis.

Packaging/Storage:

NR-58991 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 Spike Glycoprotein (Stabilized) from SARS-Related Coronavirus 2, (Lineage BA.2.75; Omicron Variant) with C-Terminal Histidine and Avi Tags, Recombinant from HEK293 Cells, NR-58991."

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 (BMBL). Current Edition. Washington, DC: U.S. Government Printing Office.

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. Sather, D. N., Personal Communication.
2. Wrapp, D., et al. "Cryo-EM Structure of the 2019-nCoV Spike in the Prefusion Conformation." Science 367 (2020): 1260-1263. PubMed: 32075877.
3. Walls, A. C., et al. "Structure, Function, and Antigenicity of the SARS-CoV-2 Spike Glycoprotein." Cell 181 (2020): 281-292. PubMed: 32155444.
4. Rambaut, A., et al. "A Dynamic Nomenclature Proposal for SARS-CoV-2 Lineages to Assist Genomic Epidemiology." Nat. Microbiol. 5 (2020): 1403-1407. PubMed: 32669681.
5. Wu, F., et al. "A New Coronavirus Associated with Human Respiratory Disease in China." Nature 579 (2020): 265-269. PubMed: 32015508.
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.
7. [WHO](#)
8. Cao, Y., et al. "Characterization of the Enhanced Infectivity and Antibody Evasion of Omicron BA.2.75." Cell. Host. Microbe. 30(2022): 1527-1539. PubMed: 36270286.

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



Figure 1: Predicted Protein Sequence

1 SQCVNLITRT QSYTNSFTRG VYYDPKVFRS SVLHSTQDLF LPFFSNVTWF
 51 HAIHVSGTNG TKRFDNPVLP FNDGVYFAST EKSNIIRGWI FGTTLDSKTQ
 101 SLLIVNNATN VVIKVCEFQF CNDPFLDVYY HENNKSRMES ELRVYSSANN
 151 CTFEYVSQPF LMDLEGKQGN FKNLREFVFK NIDGYFKIYS KHTPVNLGRD
 201 LPQGFSALEP LVDLPIGINI TRFQTLLALH RSYLTPGDSS SSWTAGAAAY
 251 YVGYLQPRTF LLKYNENGTI TDAVDCALDP LSETKCTLKS FTVEKGIYQT
 301 SNFRVQPTES IVRFPNITNL CPFHEVFNAT RFASVYAWNR KRISNCVADY
 351 SVLYNFAPFE AFKCYGVSPT KLNDLCFTNV YADSFVIRGN EVSQIAPGQT
 401 GNIADYNYKL PDDFTGCVIA WNSNKLDSKV SGNYNYLYRL FRKSKLKPFE
 451 RDISTEIYQA GNKPCNGVAG FNCYFPLQSY GFRPTYGVGH QPYRVVLSF
 501 ELLHAPATVC GPKKSTNLVK NKCVNFNENG LTGTGVLTES NKKFLPFQQF
 551 GRDIADTTDA VRDPQLEIL DITPCSFGGV SVITPGTNTS NOVAVLYQGV
 601 NCTEVPVAIH ADQLTPTWRV YSTGSNVFQT RAGCLIGAEY VNNSYECDIP
 651 IGAGICASYQ TQTKSHGSAS SVASQSIIAY TMSLGAEVSV AYSNNSIAIP
 701 TNFTISVTTE ILPVSMTKTS VDCTMYICGD STECSNLLLQ YGSFCTQLKR
 751 ALTGIAVEQD KNTQEVFAQV KQIYKTPPIK YFGGFNFSQI LPDPSKPSKR
 801 SFIEDLLFNK VTLADAGFIK QYGDCLGDIA ARDLICAQKF NGLTVLPLLL
 851 TDEMIAQYTS ALLAGTITSG WTFGGAGALQ IPFAMQMAYR FNGIGVTONV
 901 LYENQKLIAN QFNSAIKIQ DSLSSTASAL GKLQDVVNHN AQALNTLVKQ
 951 LSSKFGAISS VLNDDILSRD PPEAEVQIDR LITGRLQSLQ TYVTQQLIRA
 1001 AEIRASANLA ATKMSECVLG QSKRVDFCGK GYHLMSFPQS APHGVVFLHV
 1051 TYVPAQEKNF TTAPAICHDG KAHFPREGVF VSNGTHWFTV QRNFYEPQII
 1101 TTDNTFVSGN CDVVIGIVNN TVYDPLQPEL DSFKEELDKY FKNHTSPDVD
 1151 LGDISGINAS VVNIQKEIDR LNEVAKNLNE SLIDLQELGK YEQGSGYIPE
 1201 APRDGQAYVR KDGEWVLLST FLGRSLEVLV QGPGSHHHHH HHHGLNDIFE
 1251 AQKIEWHE

Spike ectodomain – Residues 1 to 1193 [represents amino acid residues 13 to 1208 of the native S protein (GenPept: [QHD43416](#))]

RRAR to GSAS substitution of S1/S2 cleavage site – Residues 667 to 670

KV to PP stabilizing mutations – Residues 971 and 972

T19I, A27S, G142D, K147E, W152R, F157L, I210V, V213G, G257S, G339H, S371F, S373P, S375F, T376A, D405N, R408S, K417N, N440K, G446S, N460K, S477N, T478K, E484A, Q498R, N501Y, Y505H, D614G, H655Y, N679K, P681H, N764K, D796Y, Q954H and N969K mutations–

Residues 7, 16, 127, 132, 137, 142, 195, 198, 242, 324, 356, 358, 360-361, 390, 393, 402, 425, 431, 445, 462-463,

469, 483, 486, 490, 599, 640, 664, 666, 749, 781, 939 and 954

T4 foldon trimerization domain – Residues 1196 to 1222

HRV3C protease cleavage site – Residues 1226 to 1233

Octa-histidine tag and AviTag™ – Residues 1236 to 1258