

Spike Glycoprotein (Stabilized) from SARS-Related Coronavirus 2, Theta Variant with C-Terminal Histidine and Avi Tags, Recombinant from HEK293 Cells

Catalog No. NR-55633

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Contributor:

BEI Resources

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), Theta variant (P.3 lineage) was produced in human embryonic kidney HEK293 cells and purified by immobilized metal affinity chromatography.^{1,2,3,4} NR-55633 lacks the signal sequence and contains 1191 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 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-55633 includes del141-143 (LGV), delA243-L244, Y265C, E484K, N501Y, D614G, P681H, E1092K, H1101Y and V1176F mutations in the S glycoprotein as compared to the SARS-CoV-2 reference sequence (GenPept: [QHD43416](#)).^{1,5,6} The predicted protein sequence is shown in Figure 1.¹ NR-55633 has a theoretical molecular weight of 139,300 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.⁷ P.3 is one of several lineages and sublineages designated Theta by the World Health Organization (WHO) and was first detected in the Philippines and Japan in February 2021.^{8,9} The P.3 lineage is characterized by a novel deletion (del141-143) and key mutations in the S glycoprotein, including E484K, N501Y and D614G, which have been linked to increased transmissibility and immune escape.^{6,10}

Material Provided:

Each vial contains approximately 100 µL of NR-55633 in 10 mM HEPES, pH 7, 150 mM NaCl and 2 mM

ethylenediamine-tetraacetic acid (EDTA). The concentration, expressed as mg per mL, is shown on the Certificate of Analysis.

Packaging/Storage:

NR-55633 was packaged aseptically in cryovials. The product is provided on dry ice and should be stored at -20°C or colder immediately upon arrival. Storage at warmer temperatures is not recommended due to a low bioburden. 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, Theta Variant with C-Terminal Histidine and Avi Tags, Recombinant from HEK293 Cells, NR-55633."

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.

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Figure 1: Predicted Protein Sequence

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1  SQCVNLTRT QLPPAYTNSF TRGVYYPDKV FRSSVLHSTQ DLFLPFFSNV
51  TWFHAIHVSG TNGTKRFDNP VLPFNDGVYF ASTEKSNIIR GWIFGTTLDS
101  KTQSLLIVNN ATNVVIKVCE FQFCNDPFYY HKNNKSWMES EFRVYSSANN
151  CTFEYVSQPF LMDLEGKQGN FKNLREFVFK NIDGYFKIYS KHTPINLVRD
201  LPQGFSALEP LVDLPIGINI TRFQTLHRS YLTPGDSSSG WTAGAAACYV
251  GYLQPRTFLL KYNENGTITD AVDCALDPLS ETKCTLKSFT VEKGIYQTSN
301  FRVQPTESIV RFPNITNLCP FGEVFNATRF ASVYAWNRKR ISNCVADYSV
351  LYNASAFSTF KCYGVSPTKL NDLCFTNVYA DSFVIRGDEV RQIAPGQTGK
401  IADYNYKLPD DFTGCVIAWN SNNLDSKVG G NYNYLYRLFR KSNLKPFFERD
451  ISTEIYQAGS TPCNGVKGFN CYFPLQSYGF QPTYGVGYQP YRVVLSFEL
501  LHAPATVCGP KKSTNLVKNK CVNFNFNGLT GTGVLTESNK KFLPFQQFGR
551  DIADTTDAVR DPQLEILDI TPCSEGGVSV ITPGTNTSNQ VAVLYQGVNC
601  TEVPVAIHAD QLTPTWRVYS TGSNVFQTRA GCLIGAEHVN NSYECDIPIG
651  AGICASYQTQ TNSHGSASSV ASQSIIAYTM SLGAENSVAY SNNIAIPTN
701  FTISVTTEIL PVSMTKTSVD CTMYICGDST ECSNLLLQYG SFCTQLNRAL
751  TGIAVEQDKN TQEVFAQVKQ IYKTPPIKDF GGFNFSQILP DPSKPSKRSF
801  IEDLLENKVT LADAGFIKQY GDCLGDIAAR DLICAQKFNG LTVLPLLLTD
851  EMIAQYTSAL LAGTITSGWT FGAGAALQIP FAMQMAYRFN GIGVTQNVLY
901  ENQKLIANQF NSAIGKIQDS LSSTASALGK LQDVVNQNAQ ALNTLVKQLS
951  SNFGAISSVL NDILSRLDPP EAEVQIDRLI TGRLQSLQTY VTQQLIRAAE
1001  IRASANLAAT KMSECVLGQS KRVDFCGKGY HLMSFPQSAP HGVVFLHVTY
1051  VPAQEKNFTT APAICHDGKA HFPRKGVFVS NGTYWFVTQR NFYEPQIITT
1101  DNTFVSGNCD VVIGIVNNTV YDPLQPELDS FKEELDKYFK NHTSPDVDLG
1151  DISGINASFV NIQKEIDRLN EVAKNLNESL IDLQELGKYE QSGGYIPEAP
1201  RDGQAYVRKD GEWVLLSTFL GRSLEVLVFG PGSHHHHHHH HGLNDIFEAQ
1251  KIEWHE

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Spike ectodomain – **Residues 1 to 1191** (represents WT amino acid residues 13 to 1208)

RRAR to GSAS substitution of S1/S2 cleavage site – Residues 665 to 668

KV to PP stabilizing mutations – Residues 969 and 970

Y265C, E484K, N501Y, D614G, P681H, E1092K, H1101Y and V1176F mutations –

Residues 248, 467, 484, 597, 664, 1075, 1084, and 1159

T4 foldon trimerization domain – Residues 1194 to 1220

HRV3C protease cleavage site – Residues 1224 to 1231

Octa-histidine tag and AviTag™ – Residues 1234 to 1256