Spike Glycoprotein S1 Domain from SARS-Related Coronavirus 2, P681H Variant with C-Terminal Histidine Tag, Recombinant from HEK293 Cells

Catalog No. NR-55420
ACROBiosystems Catalog No. S1N-C52Hb

For research use only. Not for use in humans.

Contributor and Manufacturer:
ACROBiosystems, Newark, Delaware, USA

Product Description:
A recombinant form of the spike (S) glycoprotein S1 domain from severe acute respiratory syndrome-related coronavirus 2 (SARS-CoV-2), P681H variant was produced by transient transfection in human embryonic kidney HEK293 cells and purified by affinity chromatography.[1] NR-55420 lacks the signal sequence, contains 670 residues of the SARS-CoV-2 S glycoprotein (amino acid residues V16 to R685) and features a C-terminal poly-histidine tag. NR-55420 is a variant of SARS-CoV-2 which contains the P681H mutation in the S glycoprotein as compared to the SARS-CoV-2 reference sequence (GenPept: QHD43416).[1,2] The predicted protein sequence is shown in Figure 1.[1] NR-55420 has a theoretical molecular weight of 76,900 daltons. The crystal structure for the wild-type S glycoprotein from SARS-CoV-2 has been solved at 2.8 Å resolution (PDB: 6VXX).[3] Representative SDS-PAGE results are shown in Figure 2.[1]

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.[4] The P681H mutation was identified in the SARS-CoV-2 variant (known as 20B/501Y.V1, VOC 202012/01 or B.1.1.7 lineage) which emerged in the United Kingdom. P681 is part of the S1/S2 proteolytic cleavage site for furin proteases, and the mutation P681H results in increased cleavage efficiency and may alter antibody recognition sites.[5,6]

Material Provided:
Each vial contains approximately 100 µg of purified recombinant protein lyophilized in phosphate-buffered saline, pH 7.4 and 10% trehalose.

Packaging/Storage:
NR-55420 was packaged aseptically in glass vials. The product is provided lyophilized and should be placed in a closed, dry environment with desiccants and stored at -20ºC or colder immediately upon arrival. A frost-free freezer should be avoided, since changes in moisture and temperature may affect protein stability.

Functional Activity:
The biological activity of NR-55420 was measured by its binding ability in a functional ELISA (Figure 3), in which immobilized NR-55420 at 2 µg per mL (100 µL per well) can bind human ACE2 protein (Fc tag) (ACROBiosystems AC2-H5257); the linear range is 0.1 to 3 ng per mL.[1]

Reconstitution:
NR-55420 should be reconstituted with 167 µL sterile deionized water to a stock solution of 600 µg per mL. Add water at room temperature with occasional gentle mixing. Carrier protein [e.g. 0.1% (w/v) bovine serum albumin] must be included in the reconstitution buffer if the final protein concentration is lower than recommended or NR-55420 is aliquoted to less than 10 µg per vial. Note: Avoid vigorous shaking or vortexing.

Storage of Reconstituted Protein:
Reconstituted NR-55420 should be stored at -70°C or colder immediately and used within 3 months. Avoid repeated freeze-thaw cycles.

Citation:
Acknowledgment for publications should read "The following reagent was obtained through BEI Resources, NIAID, NIH: Spike Glycoprotein S1 Domain from SARS-Related Coronavirus 2, P681H Variant with C-Terminal Histidine Tag, Recombinant from HEK293 Cells, NR-55420."

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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Product Information Sheet for NR-55420

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References:

1. Chen, J., Personal Communication.
2. Wu, F., et al. "A New Coronavirus Associated with Human Respiratory Disease in China." Nature 579 (2020): 265-269. PubMed: 32015508.
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. 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. Lubinski, B., et al. "Functional Evaluation of Proteolytic Activation for the SARS-CoV-2 Variant B.1.1.7: Role of the P681H Mutation." bioRxiv (2021). PubMed: 33851153.
6. Voss, C., et al. "Epitope-Specific Antibody Responses Differentiate COVID-19 Outcome and Variants of Concern." JCI Insight (2021): *in press*. PubMed: 34081630.

Support Provided by NIAID

Figure 1: Predicted Protein Sequence

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1   VNLTTRTQLP PAYTNSFTRG VYYPDKVFRS SVLHSTQDLF LPFFSNVTWF
51  HAIHVSGTNG TKRFDNPVLP FNDGVYFAST EKSNIIRGWI FGTTLDSKTQ
101 SLLIVNNATN VVIKVCEFQF CNDPFLGVYY HKNNKSWMES EFRVYSSANN
151 CTFEYVSQPF LMDLEGKQGN FKNLREFVFK NIDGYFKIYS KHTPINLVRD
201 LPQGFSALEP LVDLPIGINI TRFQTLLALH RSYLTPGDSS SGWTAGAAAY
251 YVGYLQPRTF LLKYNENGTI TDAVDCALDP LSETKCTLKS FTVEKGIYQT
301 SNFRVQPTES IVRFPNITNL CPFGEVFNAT RFASVYAWNR KRISNCVADY
351 SVLYNSASFS TFKCYGVSPT KLNDLCFTNV YADSFVIRGD EVRQIAPGQT
401 GKIADYNYKL PDDFTGCVIA WNSNNLDSKV GGNYNYLYRL FRKSNLKPFE
451 RDISTEIYQA GSTPCNGVEG FNCYFPLQSY GFQPTNGVGY QPYRVVVLSF
501 ELLHAPATVC GPKKSTNLVK NKCVNFNFNG LTGTGVLTES NKKFLPFQQF
551 GRDIADTTDA VRDPQTLEIL DITPCSFGGV SVITPGTNTS NQVAVLYQDV
601 NCTEVPVAIH ADQLTPTWRV YSTGSNVFQT RAGCLIGAEH VNNSYECDIP
651 IGAGICASYQ TQTNSHRRAR GGGSGGGSHH HHHHHHHH
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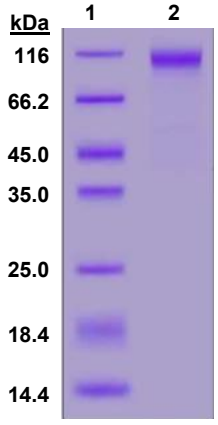
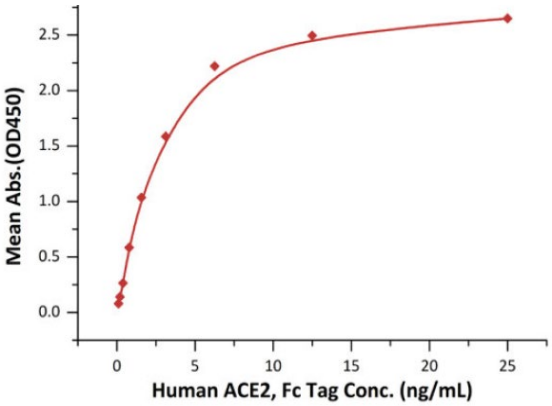
S1 domain – **Residues 1 to 670** (represents amino acid residues 16 to 685)
P681H mutation – **Residue 666**
Poly-histidine tag – Residues 679 to 688

Figure 2: Representative SDS-PAGE



Lane 1: MW ladder
Lane 2: NR-55420

Figure 3: Representative ELISA



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