

SUPPORTING INFECTIOUS DISEASE RESEARCH

## **Product Information Sheet for NR-55416**

Spike Glycoprotein S1 Domain from SARS-Related Coronavirus 2, HV69-70del Variant with C-Terminal Histidine Tag, Recombinant from HEK293 Cells

Catalog No. NR-55416 ACROBiosystems Catalog No. S1N-C52Hd

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), HV69-70del variant was produced by transient transfection in human embryonic kidney HEK293 cells and purified by affinity chromatography. 1 NR-55416 lacks the signal sequence, contains 668 residues of the SARS-CoV-2 S glycoprotein (amino acid residues V16 to R685) and features a C-terminal poly-histidine tag. NR-55416 is a variant of SARS-CoV-2 which contains the HV69-70del 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-55416 has a theoretical molecular weight of approximately 76,700 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.<sup>4</sup> New SARS-CoV-2 mutations in the S glycoprotein are currently under study, and a United Kingdom variant (also known as 20B/501Y.V1, VOC202012/01 or B.1.1.7 lineage) includes the HV69-70del mutation.<sup>1,5</sup>

## **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-55416 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 -20°C 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-55416 was measured by its binding ability in a functional ELISA (Figure 3), in which immobilized NR-55416 at 2  $\mu$ g per mL (100  $\mu$ L per well) can

bind human ACE2 protein (Fc tag) (ACROBiosystems AC2-H5257); the linear range is 0.1 to 2 ng per mL.¹ Immobilized NR-55416 at 2 μg per mL (100 μL per well) can bind anti-SARS-CoV-2 RBD neutralizing antibody, human IgG1 (ACROBiosystems SAD-S35) with a linear range of 0.1 to 2 ng per mL (Figure 4) and biotinylated human ACE2, his, Avitag™ (ACROBiosystems AC2-H82E6) with a linear range of 0.4 to 13 ng per mL (Figure 5).

## Reconstitution:

NR-55416 should be reconstituted with 167  $\mu$ L sterile deionized water to a stock solution of 600  $\mu$ g 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-55416 is aliquoted to less than 10  $\mu$ g per vial. Note: Avoid vigorous shaking or vortexing.

## Storage of Reconstituted Protein:

Reconstituted NR-55416 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, HV69-70del Variant with C-Terminal Histidine Tag, Recombinant from HEK293 Cells, NR-55416."

## 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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#### References:

- 1. Chen, J., Personal Communication.
- Wu, F., et al. "A New Coronavirus Associated with Human Respiratory Disease in China." <u>Nature</u> 579 (2020): 265-269. PubMed: 32015508.
- Walls, A. C., et al. "Structure, Function, and Antigenicity of the SARS-CoV-2 Spike Glycoprotein." <u>Cell</u> 181 (2020): 281-292. PubMed: 32155444.
- Hulswit, R. J. G., C. A. M. de Haan and B. -J. Bosch. "Coronavirus Spike Protein and Tropism Changes." <u>Adv. Virus Res.</u> 96 (2016): 29-57. PubMed: 27712627.
- Shen, L., et al. "Rapidly Emerging SARS-CoV-2 B.1.1.7 Sub-Lineage in the United States of America with Spike Protein D178H and Membrane Protein V70L Mutations." <u>Emerg. Microbes Infect.</u> (2021): in press. PubMed: 34125658.

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Figure 1: Predicted Protein Sequence

| 1   | VNLTTRTQLP | PAYTNSFTRG         | VYYPDKVFRS                           | SVLHSTQDLF         | LPFFSNVTWF |
|-----|------------|--------------------|--------------------------------------|--------------------|------------|
| 51  | HAISGTNGTK | ${\tt RFDNPVLPFN}$ | DGVYFASTEK                           | SNIIRGWIFG         | TTLDSKTQSL |
| 101 | LIVNNATNVV | IKVCEFQFCN         | DPFLGVYYHK                           | NNKSWMESEF         | RVYSSANNCT |
| 151 | FEYVSQPFLM | DLEGKQGNFK         | NLREFVFKNI                           | DGYFKIYSKH         | TPINLVRDLP |
| 201 | QGFSALEPLV | DLPIGINITR         | FQTLLALHRS                           | YLTPGDSSSG         | WTAGAAAYYV |
| 251 | GYLQPRTFLL | KYNENGTITD         | AVDCALDPLS                           | ETKCTLKSFT         | VEKGIYQTSN |
| 301 | FRVQPTESIV | RFPNITNLCP         | FGEVFNATRF                           | ASVYAWNRKR         | ISNCVADYSV |
| 351 | LYNSASFSTF | KCYGVSPTKL         | NDLCFTNVYA                           | DSFVIRGDEV         | RQIAPGQTGK |
| 401 | IADYNYKLPD | DFTGCVIAWN         | SNNLDSKVGG                           | ${\tt NYNYLYRLFR}$ | KSNLKPFERD |
| 451 | ISTEIYQAGS | TPCNGVEGFN         | CYFPLQSYGF                           | QPTNGVGYQP         | YRVVVLSFEL |
| 501 | LHAPATVCGP | KKSTNLVKNK         | CVNFNFNGLT                           | GTGVLTESNK         | KFLPFQQFGR |
| 551 | DIADTTDAVR | DPQTLEILDI         | TPCSFGGVSV                           | ITPGTNTSNQ         | VAVLYQDVNC |
| 601 | TEVPVAIHAD | QLTPTWRVYS         | TGSNVFQTRA                           | GCLIGAEHVN         | NSYECDIPIG |
| 651 | AGICASYQTQ | TNSPRRAR GG        | ${\tt GSGGGS}{\underline{\tt HHHH}}$ | <u>НННННН</u>      |            |

S1 domain – **Residues 1 to 668** (represents amino acid residues 16 to 685) Poly-histidine tag – <u>Residues 677 to 686</u>

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# **Product Information Sheet for NR-55416**

Figure 2: Representative SDS-PAGE

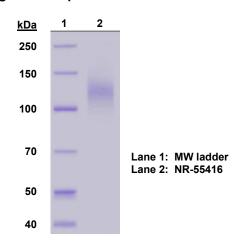
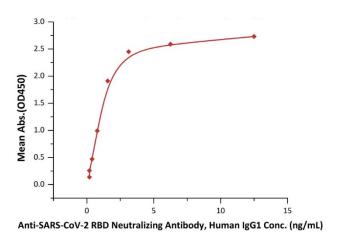


Figure 4: Representative ELISA



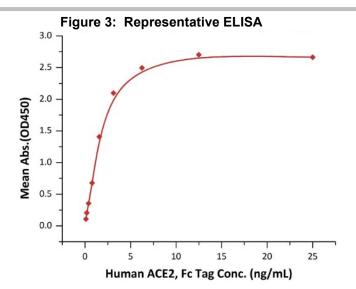
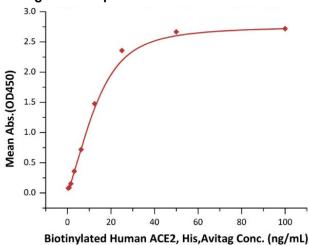


Figure 5: Representative ELISA



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