

Certificate of Analysis for NR-55277

Spike Glycoprotein Receptor Binding Domain (RBD) from SARS-Related Coronavirus 2, Alpha Variant with C-Terminal Histidine Tag, Recombinant from HEK293 Cells

Catalog No. NR-55277

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

Product Description:

A recombinant form of the spike (S) glycoprotein receptor binding domain (RBD) from severe acute respiratory syndrome-related coronavirus 2 (SARS-CoV-2), Alpha variant [also referred to as United Kingdom (UK) variant; B.1.1.7 lineage] was produced by transfection of purified plasmid (derived from BEI Resources NR-54006) in human embryonic kidney HEK293F (Expi293F™; Gibco™ A14527) cells, purified by affinity chromatography (Ni-NTA agarose) and vialed in phosphate buffered saline (PBS), pH 7.4. NR-55277 lacks the signal sequence and contains 223 residues of the SARS-CoV-2 S glycoprotein RBD and features a C-terminal hexa-histidine tag. NR-55277 is an Alpha variant of SARS-CoV-2, which includes a N501Y mutation in the S glycoprotein RBD as compared to the SARS-CoV-2 reference sequence (GenPept: QHD43416).

Lot: 70042540 Manufacturing Date: 21APR2021

TEST	SPECIFICATIONS	RESULTS
Appearance	Clear and colorless	Clear and colorless
SDS-PAGE Analysis (Coomassie Blue)	Protein band of interest represents > 90% of total staining intensity	Protein band of approximately 35 kDa represents 100 % of total staining intensity (Figure 1)¹
Concentration by Bradford Assay		
Bovine Serum Albumin (standard)	Report results	0.326 mg per mL
Vial Contents		
Amount per vial	Report results	0.033 mg
Volume per vial	Report results	0.10 mL
Functional Activity by Western Blot Analysis		
Monoclonal anti-histidine tag	Reactive	Reactive (Figure 2) ²
Filtration	0.2 µm sterile-filtered	0.2 µm sterile-filtered

¹The recombinant protein migrated to a slightly larger size than was expected, likely caused by glycosylation common in recombinant spike proteins derived from coronaviruses. For more information, please see Chakraborti, S., et al. "The SARS Coronavirus S Glycoprotein Receptor Binding Domain: Fine Mapping and Functional Characterization." <u>Virol. J.</u> 2 (2005): 73. PubMed: 16122388.

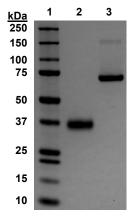
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²Using a 1:5000 dilution of 6xHis Monoclonal Antibody (Takara 631212) as primary antibody and a 1:1000 dilution of HRP-conjugated goat anti-mouse IgG (R&D Systems HAF007) as secondary antibody.



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Figure 1: SDS-PAGE Analysis

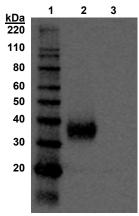


Lane 1: Precision Plus Protein™ Standard (6 µL)

Lane 2: NR-55277 (1 µg)

Lane 3: Bovine serum albumin (1 µg)

Figure 2: Anti-Histidine Western Blot Analysis



Lane 1: MagicMark™ XP Protein Standard (5 µL)

Lane 2: NR-55277 (0.5 μg)

Lane 3: Bovine serum albumin (0.5 µg)

/Heather Couch/

Heather Couch 15 JUL 2021

Program Manager or designee, ATCC Federal Solutions

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NR-55277_70042540_15JUL2021