

Certificate of Analysis for NR-56450

Spike Glycoprotein (Stabilized) from SARS-Related Coronavirus 2, (Lineage AY.4.2; Delta Variant) with C-Terminal Histidine and Avi Tags, Recombinant from HEK293 Cells

Catalog No. NR-56450

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

Product Description:

A recombinant form of the spike (S) glycoprotein from severe acute respiratory syndrome-related coronavirus 2 (SARS-CoV-2), lineage AY.4.2 (Delta variant) was produced in human embryonic kidney HEK293 cells and purified by immobilized metal affinity and gel filtration chromatography. NR-56450 lacks the signal sequence and contains 1194 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. NR-56450 includes T19R, T95I, G142D, Y145H, E156G, delF157-R158, A222V, L452R, T478K, D614G, P681R and D950N mutations in the S glycoprotein as compared to the SARS-CoV-2 reference sequence (GenPept: QHD43416). Quality control testing was completed just prior to vialing.

Lot: 70049505 Manufacturing Date: 09DEC2021

TEST	SPECIFICATIONS	RESULTS
Appearance	Report results	Clear and colorless
Purity Analytical Fast Protein Liquid Chromatography (FPLC)	Report results	Peak observed at expected retention time (Figure 1)
Protein Concentration (A ₂₈₀)	Report results	0.25 mg/mL
Final Product		
Amount per vial	Report results	100 μg
Volume per vial	Report results	400 μL
Functional Activity by Direct ELISA		
SARS-CoV-2 spike (S309) antibody ¹	Report results	Reactive (Figure 2)
SARS-CoV-2 spike S1 antibody ²	Report results	Reactive (Figure 3)

¹Pinto, D., et al. "Cross-Neutralization of SARS-CoV-2 by a Human Monoclonal SARS-CoV Antibody." <u>Nature</u> 583 (2020): 290-295. PubMed: 32422645

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Fax: 703-365-2898

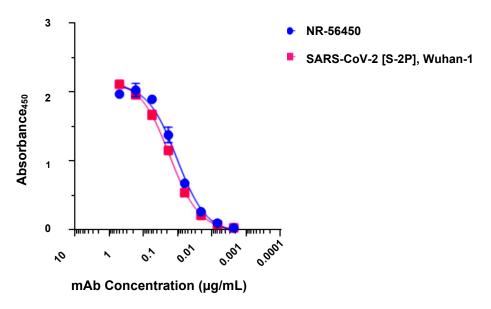
²Using SARS-CoV-2 (2019-nCoV) Spike S1 Antibody, Rabbit mAb (Sino Biological catalog number 40150-R007)

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9.40 7 6 5 Abs₂₈₀ (mAU) 3 2 1 0 5 15 10 20 25 30 mL **Elution Volume**

Figure 1: Analytical FPLC

Figure 2: Direct ELISA with S309 mAb



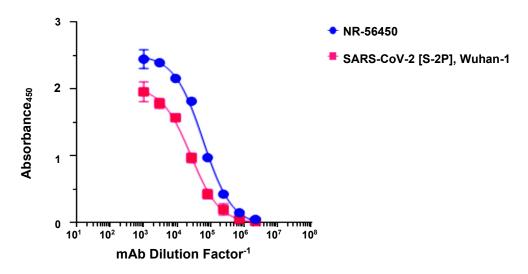
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Figure 3: Direct ELISA with Spike S1 mAb (Sino Biological 40150-R007)



/Sonia Bjorum Brower/ Sonia Bjorum Brower

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Technical Manager or designee, ATCC Federal Solutions

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