

Certificate of Analysis for NR-52724

Spike Glycoprotein (Stabilized) from SARS-Related Coronavirus 2, Wuhan-Hu-1 with C-Terminal Histidine and Twin-Strep® Tags, Recombinant from HEK293 Cells

Catalog No. NR-52724

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

Product Description:

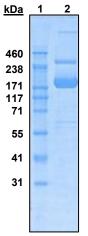
A recombinant form of the spike (S) glycoprotein from severe acute respiratory syndrome-related coronavirus 2 (SARS-CoV-2), Wuhan-Hu-1 (GenPept: QHD43416) was produced by transient transfection of the plasmid VRC7471 into human embryonic kidney HEK293 (Expi293F™) cells, purified by immobilized metal affinity and StrepTactin™ Sepharose® chromatography, and dialyzed into phosphate buffered saline (PBS; pH 7.0). NR-52724 lacks the signal sequence and contains 1194 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), and includes a T4 foldon trimerization domain, HRV3C protease cleavage site, and C-terminal octa-histidine and Twin-Strep® (TST) tags.

Lot: 70037503 Manufacturing Date: 20JUN2020

TEST	SPECIFICATIONS	RESULTS
Appearance	Clear and colorless	Clear and colorless
SDS-PAGE Analysis	Protein band of interest represents > 90% of total staining intensity	Dominant band of ~ 190 kDa represents ≥ 95% of total staining intensity (Figure 1)¹
Protein Concentration (A ₂₈₀)	Report results	0.28 mg per mL
Final Product Amount per vial Volume per vial	Report results Report results	28 μg 100 μL
Filtration	0.22 µm sterile-filtered	0.22 µm sterile-filtered
Endotoxin	Report results	3.87 U per mL

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.

Figure 1: SDS-PAGE Analysis



Lane 1: HiMark™ Pre-stained Protein Standard (ThermoFisher LC5699; 10 µL)

Lane 2: NR-52724 (2.5 µg)

BEI Resources www.beiresources.org E-mail: contact@beiresources.org Tel: 800-359-7370

Fax: 703-365-2898



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/Heather Couch/ Heather Couch

17 JUL 2020

Program Manager or designee, ATCC Federal Solutions

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Tel: 800-359-7370

Fax: 703-365-2898