### Certificate of Analysis for NR-52308

# Spike Glycoprotein (Stabilized) from SARS-Related Coronavirus 2, Wuhan-Hu-1 with C-Terminal Histidine Tag, Recombinant from Baculovirus

#### Catalog No. NR-52308

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

#### **Product Description:**

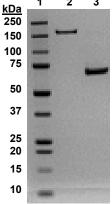
A recombinant form of the spike (S) glycoprotein from severe acute respiratory syndrome-related coronavirus 2 (SARS-CoV-2), Wuhan-Hu-1 (GenPept: QJE37812) was produced in SF9 insect cells using a baculovirus expression system and purified by nickel affinity chromatography. NR-52308 lacks the signal sequence and contains 1196 residues (ectodomain) of the SARS-CoV-2 spike glycoprotein; the recombinant protein was modified to remove the polybasic S1/S2 cleavage site (RRAR to A; residues 682 to 685), stabilized with a pair of mutations (K986P and V987P, wild type numbering; GenPept: QHD43416) and includes a thrombin cleavage site, T4 foldon trimerization domain and C-terminal hexa-histidine tag.

Lot: 70037653 Manufacturing Date: 20SEP2020

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 ~ 195 kDa represents > 90% of total staining intensity (Figure 1) <sup>1</sup>
Concentration by Bicinchoninic Acid Assay		
Bovine Serum Albumin (standard)	Report results	0.12 mg per mL
Final Product		
Amount per vial	Report results	23 μg
Volume per vial	Report results	190 µL
Functional Activity by Western Blot Analysis		
Monoclonal anti-histidine tag	Reactive	Reactive (Figure 2) <sup>2</sup>
Sterility	0.22 µm sterile-filtered	0.22 µm sterile-filtered

<sup>&</sup>lt;sup>1</sup>The 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

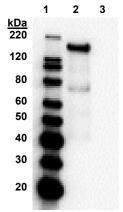


Lane 1: Precision Plus Protein™ Standard (5 µL)

Lane 2: NR-52308 (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-52308 (0.2 μg)

Lane 3: Bovine serum albumin (0.2 µg)

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<sup>&</sup>lt;sup>2</sup>Using a 1:1000 dilution of mouse monoclonal anti-histidine tag (R&D Systems MAB050) as primary antibody and a 1:1000 dilution of HRP-conjugated goat anti-mouse IgG (R&D Systems HAF007) as secondary antibody.



## **Certificate of Analysis for NR-52308**

/Heather Couch/ Heather Couch

28 OCT 2020

Program Manager or designee, ATCC Federal Solutions

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