

Certificate of Analysis for NR-44366

N9 Neuraminidase (NA) Protein with N-Terminal Histidine Tag from Influenza A Virus, A/Anhui/1/2013 (H7N9), Recombinant from Baculovirus

Catalog No. NR-44366

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

Product Description:

A recombinant form of the N9 neuraminidase (NA) protein from influenza A virus, A/Anhui/1/2013 (H7N9) containing an N-terminal histidine tag was produced in Sf9 insect cells using a baculovirus expression vector system and purified by nickel affinity chromatography.

Lot: 70046220 Manufacturing Date: 21OCT2021

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TEST	SPECIFICATIONS	RESULTS
Appearance	Clear and colorless	Clear and colorless
SDS-PAGE Analysis	Protein band of interest represents > 90% of total staining intensity	Protein band of interest accounts for ~91.5% of total staining intensity (Figure 1)
Identification by Western Blot Analysis Monoclonal anti-histidine Polyclonal anti-influenza A virus N9 Neuraminidase (NA), A/tern/Australia/G70C/1975 (H11N9)	Reactive Reactive	Reactive (Figure 2A) ¹ Reactive (Figure 2B) ²
Concentration by Bradford Assay Bovine Serum Albumin (BSA; standard)	Report results	147 μg/mL
Final Product Quantity per vial Volume per vial	Report results Report results	69 μg 470 μL
Functional Activity Neuraminidase activity in fluorescent enzymatic assay	Report results	6.3 × 10 ¹⁰ relative fluorescence units per hour per mg protein ³
Endotoxin Content Limulus Amoebocyte Lysate Assay	Report results	34.01 EU/mg
Filtration	0.2 µm sterile-filtered	0.2 µm sterile-filtered

¹Using a 1:1000 dilution of mouse monoclonal anti-histidine tag (R&D Systems, Catalog No. MAB050) as primary antibody and a 1:1000 dilution of HRP-conjugated goat anti-mouse IgG (R&D Systems, Catalog No. HAF007) as secondary antibody

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²Using a 1:10,000 dilution of goat polyclonal anti-A/tern/Australia/G70C/1975 (BEI Resources Catalog No. NR-667) as primary antibody and a 1:1000 dilution of HRP-conjugated anti-goat IgG (R&D Systems, Catalog No. HAF109) as secondary antibody

³Using serial dilutions of NR-44366 and 0.15 mM 2'-(4-methylumbelliferyl)-α-d-N-acetylneuraminic acid (4-MUNANA) (Sigma, Catalog No. M8639), as described in Wetherall, N. T., et al. "Evaluation of Neuraminidase Enzyme Assays Using Different Substrates to Measure Susceptibility of Influenza Virus Clinical Isolates to Neuraminidase Inhibitors: Report of the Neuraminidase Inhibitor Susceptibility Network." <u>J. Clin. Microbiol.</u> 41 (2003): 742-750. PubMed: 12574276.



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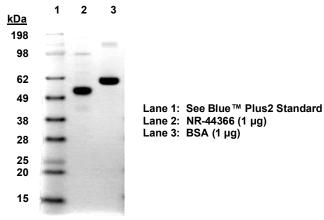
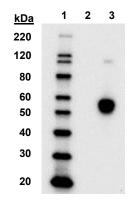


Figure 2: Western Blot Analysis

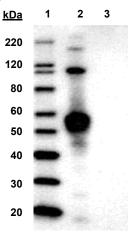
A: Monoclonal Anti-Histidine Tag



Lane 1: MagicMark™ XP Protein Standard

Lane 2: BSA (0.5 μg) Lane 3: NR-44366 (0.5 μg)

B: Polyclonal Anti-Influenza A Virus



Lane 1: MagicMark™ XP Protein Standard

Lane 2: NR-44366 (0.5 μg) Lane 3: BSA (0.5 μg)

/Sonia Bjorum Brower/ Sonia Bjorum Brower

24 APR 2024

Technical Manager or designee, ATCC Federal Solutions

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