

N1 Neuraminidase (NA) Protein with N-Terminal Histidine Tag from Influenza Virus, A/New Caledonia/20/1999 (H1N1), Recombinant from Baculovirus

Catalog No. NR-43779

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

Product Description:

A recombinant form of the N1 neuraminidase (NA) protein from influenza A virus, A/New Caledonia/20/1999 (H1N1) containing an N-terminal histidine tag was produced in Sf9 insect cells using a baculovirus expression vector system and purified by nickel affinity chromatography. The predicted ectodomain coding region of the NA gene was fused to a synthetic gene segment encoding an N-terminal octa-histidine tag followed by a 43 amino acid tetramerization domain from vasodilator-stimulated phosphoprotein (VASP) and a thrombin cleavage site, as described for the 1918 pandemic virus. NR-43779 lot 70041698 is provided in Dulbecco's-Phosphate buffered saline (D-PBS; pH 7.4) supplemented with 50% glycerol.

Lot: 70041698

Manufacturing Date: 28JUN2021

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 ~ 55 kDa represents > 95% of total staining intensity (Figure 1)
Identification by Western Blot Analysis Polyclonal anti-N1 NA ¹ Ferret hyperimmune sera ² Monoclonal anti-histidine tag ⁴	Reactive Reactive Reactive	Reactive (Figure 2A) Reactive (Figure 2B) ³ Reactive (Figure 2C)
Concentration by Bradford Assay Bovine Serum Albumin (standard)	Report results	220 µg per mL
Final Product Amount per vial Volume per vial	Report results Report results	110 µg 500 µL
Functional Activity Neuraminidase activity in fluorescent enzymatic assay ⁵	Report results	9.8 × 10 ⁸ relative fluorescent units per hour per mg protein
Endotoxin Content (Limulus Amebocyte Lysate Assay)	Report results	< 22.73 EU per mg
Filtration	0.2 µm sterile-filtered	0.2 µm sterile-filtered

¹Using a 1:1000 dilution of goat polyclonal anti-N1 NA (BEI Resources NR-3136) as primary antibody and a 1:1000 dilution of HRP-conjugated donkey anti-goat IgG (R&D Systems HAF109) as secondary antibody

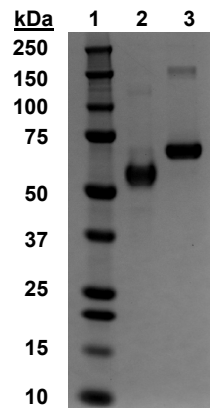
²Using a 1:1000 dilution of ferret hyperimmune sera to Influenza A/New Caledonia/20/1999 (H1N1) (BEI Resources NR-19263) as primary antibody and a 1:1000 dilution of HRP-conjugated goat anti-ferret IgG (Abcam ab112770) as secondary antibody

³NR-19263 is a pooled polyclonal antibody from serum, and might produce a non-specific band

⁴Using 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

⁵Using serial dilutions of NR-43779 and 2'-(4-methylumbelliferyl)-α-D-N-acetylneuraminic acid (4-MUNANA), 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." *J. Clin. Microbiol.* 41 (2003): 742-750. PubMed: 12574276.

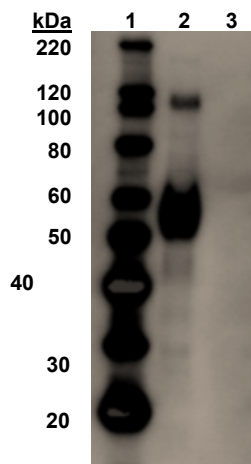
Figure 1: SDS-PAGE Analysis



Lane 1: Precision Plus Protein™ Standard
 Lane 2: NR-43779 (1.0 µg)
 Lane 3: BSA (1.0 µg)

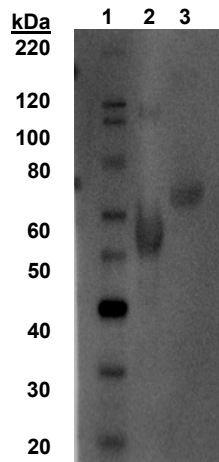
Figure 2: Western Blot Analysis

A: Polyclonal Anti-N1 NA



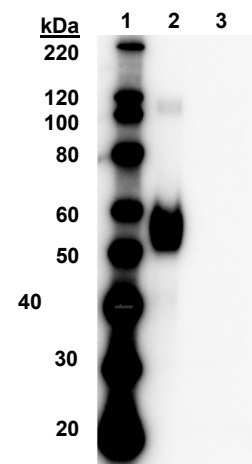
Lane 1: MagicMark™ XP Protein Standard
 Lane 2: NR-43779 (1.0 µg)
 Lane 3: BSA (1.0 µg)

B: Ferret Hyperimmune Sera



Lane 1: MagicMark™ XP Protein Standard
 Lane 2: NR-43779 (1.0 µg)
 Lane 3: BSA (1.0 µg)

C: Monoclonal Anti-Histidine Tag



Lane 1: MagicMark™ XP Protein Standard
 Lane 2: NR-43779 (1.0 µg)
 Lane 3: BSA (1.0 µg)

/Sonia Bjorum Brower/
Sonia Bjorum Brower

Technical Manager or designee, ATCC Federal Solutions

21 OCT 2022

ATCC®, on behalf of BEI Resources, hereby represents and warrants that the material provided under this certificate has been subjected by the contributor to the tests and procedures specified and that the results described, along with any other data provided in this certificate, are true and accurate to the best of ATCC®'s knowledge.

ATCC® is a trademark of the American Type Culture Collection.

You are authorized to use this product for research use only. It is not intended for human use.

