Certificate of Analysis for NR-19237

N2 Neuraminidase (NA) Protein with N-Terminal Histidine Tag from Influenza Virus, A/Wisconsin/67/2005 (H3N2), Recombinant from Baculovirus

Catalog No. NR-19237

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

Product Description:

A recombinant form of the N2 neuraminidase (NA) protein from influenza A virus, A/Wisconsin/67/2005 (H3N2) containing an N-terminal octa-histidine tag was produced in Sf9 insect cells using a baculovirus expression vector system and was 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. Semi-purified protein in PBS from the previous lot was thawed on ice overnight, purified by nickel affinity chromatography to remove lipopolysaccharides, sterile-filtered and aliquoted into vials to produce this lot.

Lot: 70046570 **Manufacturing Date: 13AUG2021**

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 ~ 60 kDa represents ~ 92.8% of total staining intensity (Figure 1)
Identification by Western Blot Analysis Monoclonal anti-histidine tag[1] Polyclonal anti-N2 NA[2]	 Reactive Reactive	 Reactive (Figure 2) Reactive (Figure 3)
Concentration by Bradford Assay	Report results	567 µg per mL
Final Product Amount per vial Volume per vial	 Report results Report results	 91 µg 160 µL
Functional Activity Neuraminidase activity in fluorescent enzymatic assay[3]	Report results	1.21 × 10^{10} relative fluorescent units per hour per mg protein
Endotoxin Content (Limulus Amebocyte Lysate Assay)	Report results	48.32 EU per mg
Filtration	0.2 µm sterile-filtered	0.2 µm sterile-filtered

[1]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

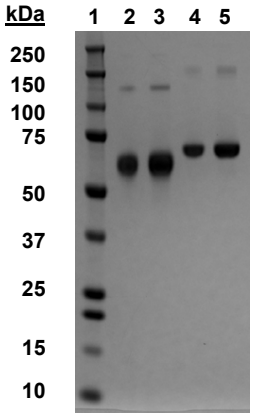
[2]Using a 1:1000 dilution of goat polyclonal anti-N2 NA (BEI Resources NR-3137) as primary antibody and a 1:1000 dilution of HRP-conjugated donkey anti-goat IgG (R&D Systems HAF109) as secondary antibody

[3]Using serial dilutions of NR-19237 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.

BEI Resources
www.beiresources.org
E-mail: contact@beiresources.org
Tel: 800-359-7370
Fax: 703-365-2898

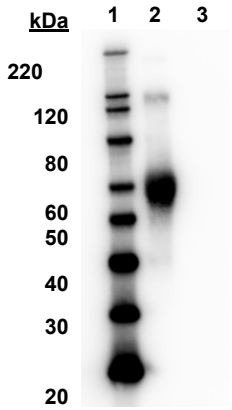
Certificate of Analysis for NR-19237

Figure 1: SDS-PAGE Analysis



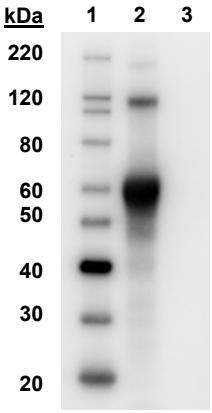
Lane 1: Precision Plus Protein™ Standard
Lane 2: NR-19237 (1.0 µg)
Lane 3: NR-19237 (2.0 µg)
Lane 4: BSA (1.0 µg)
Lane 5: BSA (2.0 µg)

Figure 2: Western Blot with Monoclonal Anti-Histidine Tag



Lane 1: MagicMark™ XP Protein Standard
Lane 2: NR-19237 (1.0 µg)
Lane 3: BSA (1.0 µg)

Figure 3: Western Blot with Polyclonal Anti-N2 NA



Lane 1: MagicMark™ XP Protein Standard
Lane 2: NR-19237 (1.0 µg)
Lane 3: BSA (1.0 µg)

/Sonia Bjorum Brower/
Sonia Bjorum Brower 21 OCT 2022
Technical Manager or designee, ATCC Federal Solutions

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.

Support Provided by NIAID

BEI Resources
www.beiresources.org
E-mail: contact@beiresources.org
Tel: 800-359-7370
Fax: 703-365-2898