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SUPPORTING INFECTIOUS DISEASE RESEARCH

N1 Neuraminidase (NA) Protein with N-Terminal Histidine Tag from Influenza Virus, A/Brisbane/59/2007 (H1N1), Recombinant from Baculovirus

Catalog No. NR-43785

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

For research use only. Not for use in humans.

Contributor and Manufacturer:

BEI Resources

Product Description:

A recombinant form of the N1 neuraminidase (NA) protein from influenza A virus, A/Brisbane/59/2007 (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 vasodilatorstimulated phosphoprotein (VASP) and a thrombin cleavage site, as described for the 1918 pandemic virus.^{1,2} The predicted protein sequence of NR-43785 is shown in Figure 1. The full-length NA precursor protein is 470 residues (GenPept: <u>ADE28752</u>).

Material Provided:

Each vial contains approximately 100 micrograms of purified recombinant NA protein in 16 mM Na₂HPO₄, 400 mM NaCl (pH 7.5) with 20% glycerol. The protein content in μ g and the concentration, expressed as micrograms per milliliter, are shown on the Certificate of Analysis.

Packaging/Storage:

Purified recombinant NA protein was packaged aseptically in screw-capped plastic cryovials. Lot 62190122 and lot 63417493 are provided on refrigerated bricks and should be stored at 2°C to 8°C immediately upon arrival. Lot 70043130 is shipped on dry ice and should be stored at -20°C immediately upon arrival.

Functional Activity:

NR-43785 was demonstrated to be functionally active based on its ability to cleave the fluorogenic substrate 2'-(4-methylumbelliferyl)- α -D-N-acetylneuraminic acid.³

Citation:

Acknowledgment for publications should read "The following reagent was obtained through BEI Resources, NIAID, NIH: N1 Neuraminidase (NA) Protein with N-Terminal Histidine Tag from Influenza Virus, A/Brisbane/59/2007 (H1N1), Recombinant from Baculovirus, NR-43785."

Biosafety Level: 1

Appropriate safety procedures should always be used with this material. Laboratory safety is discussed in the following publication: U.S. Department of Health and Human Services, Public Health Service, Centers for Disease Control and Prevention, and National Institutes of Health. <u>Biosafety in Microbiological and Biomedical Laboratories</u>. 6th ed. Washington, DC: U.S. Government Printing Office, 2020; see www.cdc.gov/biosafety/publications/bmbl5/index.htm.

Disclaimers:

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References:

- Kühnel, K., et al. "The VASP Tetramerization Domain is a Right-Handed Coiled Coil Based on a 15-Residue Repeat." <u>Proc. Natl. Acad. Sci. USA</u> 101 (2004): 17027-17032. PubMed: 15569942.
- Xu, X., et al. "Structural Characterization of the 1918 Influenza Virus H1N1 Neuraminidase." <u>J. Virol.</u> 82 (2008): 10493-10501. PubMed: 18715929.
- Wetherall, N. T., et al. "Evaluation of Neuraminidase Enzyme Assays Using Different Substrates to Measure Susceptibility of Influenza Virus Clinical Isolates to

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Neuraminidase Inhibitors: Report of the Neuraminidase Inhibitor Susceptibility Network." J. Clin. Microbiol. 41 (2003): 742-750. PubMed: 12574276.

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Figure 1: Predicted Protein Sequence

1	АДРНННННН	HSSSDYSDLQ	RVKQELLEEV	KKELQKVKEE	IIEAFVQELR
51	KRGSLVPRGS	PSRSEF VTLA	GNSSLCSISG	WAIYTKDNSI	RIGSKGDVFV
101	IREPFISCSH	LECRTFFLTQ	GALLNDKHSN	GTVKDRSPYR	ALMSCPLGEA
151	PSPYNSKFES	VAWSASACHD	GMGWLTIGIS	GPDNGAVAVL	KYNGIITGTI
201	KSWKKQILRT	QESECVCMNG	SCFTIMTDGP	SNKAASYKIF	KIEKGKVTKS
251	IELNAPNFHY	EECSCYPDTG	IVMCVCRDNW	HGSNRPWVSF	NQNLDYQIGY
301	ICSGVFGDNP	RPEDGEGSCN	PVTVDGANGV	KGFSYKYDNG	VWIGRTKSNR
351	LRKGFEMIWD	PNGWTNTDSD	FSVKQDVVAI	TDWSGYSGSF	VQHPELTGLD
401	CIRPCFWVEL	VRGLPRENTT	IWTSGSSISF	CGVNSDTANW	SWPDGAELPF
451	TIDK				

Plasmid-derived amino acids – <u>Residues 1 to 3 and 61 to 66</u> Octa-histidine tag – Residues 4 to 11 Tetramerization domain – <u>Residues 12 to 54</u> Thrombin cleavage sequence – Residues 55 to 60 **NA protein – Residues 67 to 454** (represents amino acid residues 83 to 470)