Product Information Sheet for NR-19235

N1 Neuraminidase (NA) Protein with N-Terminal Histidine Tag from Influenza Virus, A/Puerto Rico/8/1934 (H1N1), Recombinant from Baculovirus

Catalog No. NR-19235

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

For research use only. Not for human use.

Contributor and Manufacturer:

BEI Resources

Product Description:

A recombinant form of the N1 neuraminidase (NA) protein from influenza A virus, A/Puerto Rico/8/1934 (H1N1) containing an N-terminal histidine tag was produced in High Five™ insect cells using a baculovirus expression vector system and was purified by nickel affinity chromatography under denaturing conditions. The purified protein was refolded by dialysis and filtered. The predicted ectodomain coding region of the NA gene was fused to a synthetic gene segment encoding an N-terminal eight-histidine tag followed by a 43 amino acid tetramerization domain from vasodilator-stimulated phosphoprotein (VASP)[1] and a thrombin cleavage site, as described for the 1918 pandemic virus.[2] The predicted protein sequence is shown in Table 1. The full-length NA precursor protein is 454 residues (GenPept: ABD77678).

Material Provided:

Each vial contains approximately 50 to 150 µg of purified recombinant NA protein in 50 mM Tris-HCl (pH 8.5), 240 mM NaCl, 10 mM KCl, 1 mM EDTA, 0.5 M Arginine, 0.5% Triton X-100, and 1 mM DTT. The protein content in µg and the concentration, expressed as µg/mL, are shown on the Certificate of Analysis.

Packaging/Storage:

Purified recombinant NA protein was packaged aseptically in screw-capped plastic cryovials. This product is provided on refrigerated bricks and should be stored at 2°C to 8°C immediately upon arrival. Do not freeze.

Functional Activity:

NR-19235 has not been tested for enzymatic activity. Previous work at BEI Resources indicated that other influenza virus neuraminidases purified under denaturing conditions and refolded by dialysis are not able to cleave the fluorogenic substrate 2'-(4-methylumbelliferyl)-α-D-N-acetylneuraminic acid (4-MUNANA).[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/Puerto Rico/8/1934 (H1N1), Recombinant from Baculovirus, NR-19235."

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. Biosafety in Microbiological and Biomedical Laboratories. 5th ed. Washington, DC: U.S. Government Printing Office, 2009; see www.cdc.gov/biosafety/publications/bmbl5/index.htm.

Disclaimers:

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References:

1. Kühnel, K., et al. "The VASP Tetramerization Domain is a Right-Handed Coiled Coil Based on a 15-Residue Repeat." Proc. Natl. Acad. Sci. USA 101 (2004): 17027-17032. PubMed: 15569942.

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2. Xu, X., et al. "Structural Characterization of the 1918 Influenza Virus H1N1 Neuraminidase." J. Virol. 82 (2008): 10493-10501. PubMed: 18715929.
3. 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.

Support Provided by NIAID

Table 1 – Predicted Protein Sequence

1	ADPHHHHHHH	HSSSDYSDLQ	RVKQELLEEV	KKELQKVKEE	IIEAFVQELR
51	KRGSLVPRGS	PSRSEFVILT	GNSSLCPIRG	WAIYSKDNSI	RIGSKGDVFV
101	IREPFISCSH	LECRTFFLTQ	GALLNDKHSS	GTVKDRSPYR	ALMSCPVGEA
151	PSPYNSRFES	VAWSASACHD	GMGWLTIGIS	GPDNGAVAVL	KYNGIITETI
201	KSWRKKILRT	QESECACVNG	SCFTIMTDGP	SDGLASYKIF	KIEKGKVTKS
251	IELNAPNSHY	EECSCYPDTG	KVMCVCRDNW	HGSNRPWVSF	DQNLDYQIGY
301	ICSGVFGDNP	RPEDGTGSCG	PVYVDGANGV	KGFSYRYGNG	VWIGRTKSHS
351	SRHGFEMIWD	PNGWTETDSK	FSVRQDVVAM	TDWSGYSGSF	VQHPELTGLD
401	CMRPCFWVEL	IRGRPKEKTI	WTSASSISFC	GVNSDTVDWS	WPDGAELPFS
451	IDK				

Plasmid-derived amino acids – Residues 1 to 3 and 61 to 66
His Tag – Residues 4 to 11
Tetramerization domain – Residues 12 to 54
Thrombin cleavage sequence – Residues 55 to 60
NA protein – **Residues 67 to 453***

*This represents amino acid residues 68 to 454 of the A/Puerto Rico/8/1934 (H1N1) NA protein.

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