

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

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Contributor and Manufacturer:

BEI Resources

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 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 470 residues (GenPept: AFO65030).

Material Provided:

Each vial contains approximately 100 µg of purified recombinant NA protein in D-PBS (pH 7.4) with 1 mM EDTA and an additional 500 mM NaCl. 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.

Functional Activity:

NR-43779 was demonstrated to be functionally active based on its ability 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/New Caledonia/20/1999 (H1N1), Recombinant from Baculovirus, NR-43779."

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/bmbli5/index.htm.

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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.
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.

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Table 1 – Predicted Protein Sequence

| | | | | | |
|-----|--------------------------|-------------|-------------|------------|-------------|
| 1 | ADPHHHHHH | HSSSDYSDLQ | RVKQELLEEV | KKELQKVKEE | IIEAFVQELR |
| 51 | KRGS ^u LVPRGS | PSRSEFVTLA | GNSSLCSISG | WAIYTKDNSI | RIGSKGDV FV |
| 101 | IREFFISCSH | LECRFFFLTQ | GALLNDKHSN | GTVKDRSPYR | ALMSCPLGEA |
| 151 | PSPYNSKFES | VAWSASACHD | GMGWL TIGIS | GPDNGAVAVL | KYNGIITETI |
| 201 | KSWKKRILRT | QESECVCVNG | SCFTIMTDGP | SNGAASYKIF | KIEKGKVTKS |
| 251 | IELNAPNFHY | EECSCYPDTG | TVMCVC RDNW | HGSNRPWVSF | NQNLDYQIGY |
| 301 | ICSGVFGDNP | RPKDGE GSCN | PVTVDGADGV | KGFSYKYGNG | VWIGRTKSNR |
| 351 | LRKGFEMIWD | PNGWTD TSD | FSVKQDVVAI | TDWSGYSGSF | VQHELTGLD |
| 401 | CIRPCFWVEL | VRGLPRENTT | IWTSGSSISF | CGVNSDTANW | SWPDGAELPF |
| 451 | TIDK | | | | |

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 454*

*This represents amino acid residues 83 to 470 of the A/New Caledonia/20/1999 (H1N1) NA protein.