

Alpha(2-3,6) Neuraminidase

α (2-3,6) Neuraminidase (N-acetylneuraminate glycohydrolase EC 3.2.1.18) cleaves all non-reducing terminal non-branched α (2-3) and α (2-6) sialic acid residues from complex carbohydrates and glycoproteins. There is no detectable activity on α (2-8) or α (2-9) linkages or on branched α (2-3) or α (2-6) linkages (Figure 1). The relative cleavage rates for different linkages are: α (2-3) > α (2-6).

α (2-3,6) Neuraminidase will not cleave branched sialic acids (linked to an internal residue). Use α (2-3,6,8,9) Neuraminidase for α (2-8) or branched sialic acids. To cleave only nonreducing terminal α (2-3) unbranched sialic acid residues, use α (2-3) Neuraminidase.

α (2-3,6) Neuraminidase is isolated from a clone of *Clostridium perfringens*. The enzyme has been extensively characterized using oligosaccharide standards

α (2-3,6) Neuraminidase is useful for:

- Structural analysis of oligosaccharides
- Determining sialic acid linkages
- Glycoprotein desialylation
- Removing heterogeneity from glycoproteins

Specifications

Activity

≥ 250 U/mg, ≥15 U/mL

Storage

Store at 4°C. Do not freeze.

Formulation

The enzyme is provided as a sterile-filtered solution in 20 mM Tris HCl, 25 mM NaCl pH 7.5.

Stability

Stable at least 12 months when stored properly. Several days exposure to ambient temperatures will not reduce activity.

Product Description

Molecular Weight

~41,000 Daltons

Specificity

Only non-reducing terminal α (2-3) and α (2-6) unbranched sialic acids (see Figure 1).

Purity

Each lot of α (2-3,6) Neuraminidase is tested for contaminating protease as follows: 10 µg of denatured BSA is incubated for 24 hours at 37°C with 2 µL of enzyme. SDS-PAGE analysis of the treated BSA shows no evidence of degradation.

The production host strain has been extensively tested and does not produce any detectable glycosidases.

pH Range:

50 mM sodium phosphate (pH 6.0) provides the optimal buffer for enzyme activity with 3'-siayllactose, a standard substrate. If glycosidase treatment is performed at suboptimal pH because of glycoprotein solubility or activity requirements, expect some diminution in enzyme activity.

Assay

One unit of α (2-3,6) Neuraminidase is defined as the amount of enzyme required to produce 1 µmole of methylumbelliferone in 1 minute at 37°C, pH 5.0 from MU-NANA [2'-(4-methylumbelliferyl)- α -D-N-acetylneuraminic acid].

Reagents

- 5X Reaction buffer 6.0 - 250 mM sodium phosphate, pH 6.0

Suggestions for Use

Procedure for De-sialylation

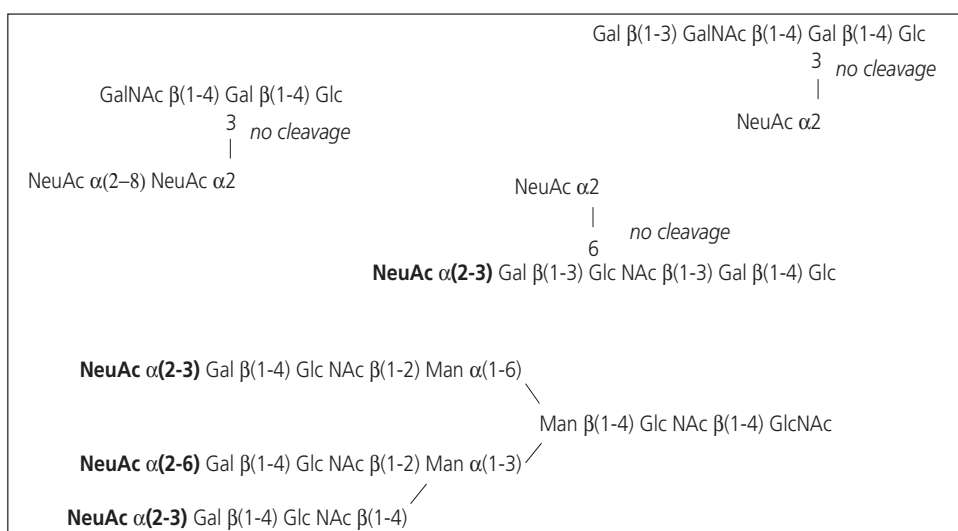
1. Add up to 100 µg of glycoprotein or 1 nmol of oligosaccharide to a tube.
2. Add water to a total of 14 µL.
3. Add 4 µL 5X Reaction Buffer 6.0.
4. Add 2 µL of α (2-3,6) Neuraminidase.
5. Incubate at 37°C for 1 hour.

De-sialylation may be monitored by SDS-PAGE if the size differential between native and desialylated protein is sufficient for detection.

References

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2. Dwek, R. A., C. J. Edge, D. J. Harvey, M. R. Wormald and R. B. Parekh. Analysis of glycoprotein-associated oligosaccharides. *Ann Rev Biochem* 62:65-100 (1993).
3. Kobata, A. Use of endo- and exoglycosidases for structural studies of glycoconjugates. *Anal Biochem* 100:1-14 (1979).
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5. Roggentin, P, B. Rothe, F Lottspeich and R. Schauer. Cloning and sequencing of a *Clostridium perfringens* sialidase gene. *FEBS Lett* 238:31-34 (Sep 1988).
6. Roggentin P., R. G. Kleineidam and R. Schauer. Diversity in the properties of two sialidase isoenzymes produced by *Clostridium perfringens* spp. *Biol chem Hoppe Seyler* 376:569-575 (1995).

Figure 1 - Linkage specificities showing cleavable residues (in bold) for $\alpha(2-3,6)$ Neuraminidase



Gal = Galactose; Glc = Glucose; Man = Mannose; GalNAc = N-acetylgalactosamine; GlcNAc = N-acetylglucosamine; NeuAc = N-acetylneuraminic Acid (Sialic Acid)

Order Information

Catalog No.	Product Description	Package Size	Temp. °C
120056-1	$\alpha(2-3,6)$ Neuraminidase (<i>Clostridium perfringens</i> recombinant)	60 μ L	+4

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