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α (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. 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 non-reducing 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) is useful for:

- Structural analysis of oligosaccharides
- Determining sialic acid linkage
- Glycoprotein deglycosylation
- Removing heterogeneity from glycoproteins

Product Code: GE 24

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 (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 with 2 µL of enzyme. SDS-PAGE analysis of the treated BSAshows 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 from MU-NANA [2'-(4-methylumbelliferyl)-α-D-N-acetylneuraminic acid].

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Reagents

- 5X reaction buffer 6.0 – 250 mM sodium phosphate, pH 6.0

Suggestions for Use

Procedure for De-sialylation

Add up to 100 μg of glycoprotein or 1 nmol of oligosaccharide to tube

Add water to a total of 14 μL

Add 5 μL 5X Reaction Buffer 5.0

Add 2 μL of α(2-3,6) Neuraminidase

Incubate at 37°C for 1 hour.

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

References

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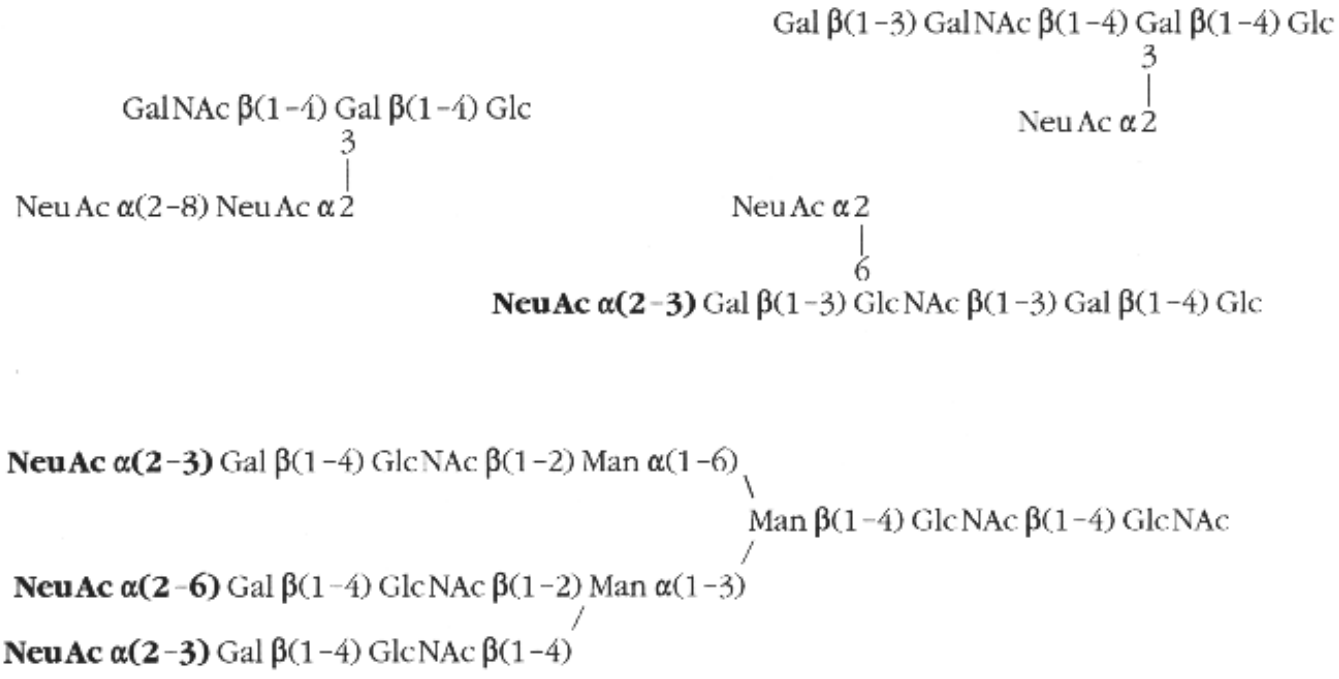
Figure 1 – Linkage specificities showing cleavable residues (in bold) for α(2-3, 6) Neuraminidase



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