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α(2-3, 6, 8, 9) Neuraminidase

α (2-3, 6, 8, 9) neuraminidase (EC 3.2.1.18) cleaves all cleaves all non-reducing terminal sialic acid residues from complex carbohydrates and glycoproteins. The relative cleavage rates for different linkages are: α(2-6) > α(2-3) > α(2-8), α(2-9).

In addition, the enzyme will cleave branched sialic acids (linked to an internal residue). This property makes it unique among sialidases (Figure 1). High concentrations of enzymes and prolonged incubation times may be required for cleaving branched residues. To cleave only non-reducing terminal α(2-3) unbranched sialic acid residues, use α (2-3) neuraminidase(GE20). α (2-3, 6, 8, 9) neuraminidase is isolated from a clone of *Arthrobacter ureafaciens*. The enzyme has been extensively characterized using oligosaccharide standards.

α (2-3, 6, 8, 9) neuraminidase is useful for:

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

Product Code: GE 23

Specifications

Activity: ≥135 U/mg, 5 U/mL

Storage: Store at 4°C. Do not freeze.

Formulation: The enzyme is provided as a sterile solution in 20 mM Tris pH 7.5, 25 mM sodium chloride.

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

Product Description

Molecular weight: 70,000 Daltons

Purity: Each lot of α(2-3, 6, 8, 9) 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 BSA shows no evidence of degradation.

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

Specificity: All non-reducing terminal branched and unbranched sialic acids (see Figure 1).

pH Range:	Optimum:	pH 6
	Range:	pH 4.5 - 7

The recommended buffer concentrate provides the optimal pH for enzyme activity with the 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, 8, 9) neuraminidase activity is defined as the amount of enzyme required to produce 1 μmole of methylumbelliferone in 1 minute at 37°C, pH 5

from 2'-(4-methylumbelliferyl)-α-D-N-acetylneuraminic acid.

Reagents

- 5X Reaction buffer 6 – 250 mM sodium phosphate pH 6.0

Suggestions For Use

Procedure for Desialylation

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

1. Add up to 100 µg of glycoprotein or 1 nmol of oligosaccharide to tube.
2. Add water to 13 µL and 4 µL 5X Reaction Buffer.
3. Add 2 µL α (2-3, 6, 8, 9) neuraminidase.
4. Incubate at 37°C for 1 hour.

NOTE: longer incubation times are necessary if branched sialic acids are present.

References

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This product is intended for in vitro research only.

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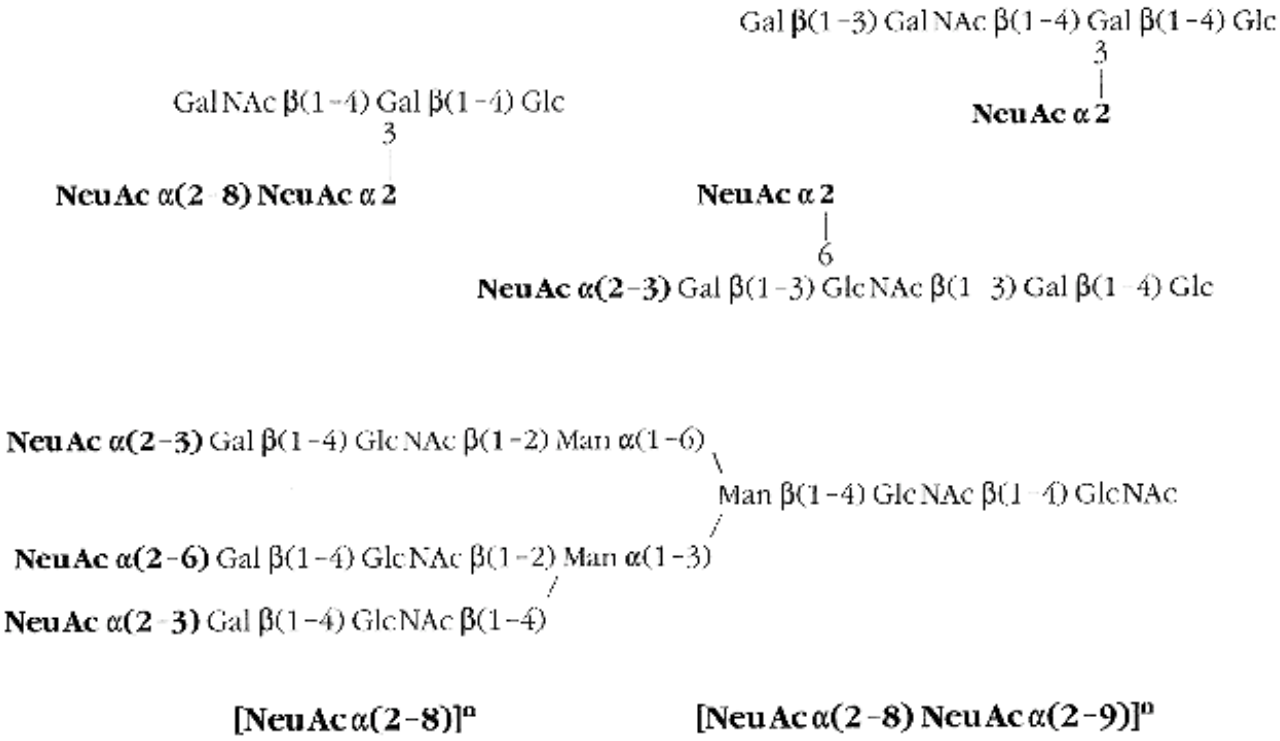
Figure 1

Linkage specificities showing cleavable residues (in bold) for α(2-3, 6, 8, 9) Neuraminidase



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