

Neuraminidase Cp

Sialidase, NANase, N-acetylneuraminate glycohydrolase

Source

recombinant from Clostridium perfringens

Catalog Number

E-S005	60 µl
E-S005-20	20 µl
E-S005-200	200 µl

EC

3.2.1.18

Applications

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

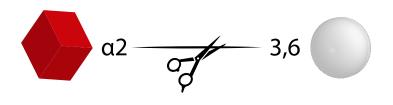
Recommended Reagents

included with 20μL and 60 μL pack sizes: 1 vial: Reaction buffer – 400 μl 250mM Sodium phosphate, pH 6.0

Activity ≥ 15 U/ml Specific Activity ≥ 250 U/mg

Molecular Weight ~41,000 daltons **pH optimum** 6.0, active over the range 4.5-7.

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



Specific Activity

One unit of QA-Bio Neuraminidase is defined as the amount of enzyme required to produce 1 μ mole of methylumbelliferone in 1 minute at 37°C, pH 5.0 from MU-NANA (2'-(4-methyl-umbelliferyl)-*alpha*-D-N acetylneuraminic acid].

Specificity

 $\alpha(2-3,6)$ Neuraminidase Cp cleaves all non- reducing terminal non-branched &alpha(2-3)- and $\alpha(2-6)$ 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).

 $\alpha(2-3,6)$ Neuraminidase Cp will not cleave branched sialic acids (linked to an internal residue). Use $\alpha(2-3,6,8,9)$ Neuraminidase (E-S001) for $\alpha(2-8)$ or branched sialic acids. To cleave only non-reducing terminal $\alpha(2-3)$ unbranched sialic acid residues, use $\alpha(2-3)$ Neuraminidase (E-S007).

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

Relative activity α -(2-3) > α -(2-6)

Formulation

The enzyme is provided as a sterile-filtered solution in 20 m*M* Tris-HCl, 25 m*M* NaCl (pH 7.5).

Neuraminidase Cp Specifications - Protocol

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Stability

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

Storage

Store enzyme at 4°C. Do not freeze.

Purity

QA-Bio Neuraminidase Cp is tested for contaminating protease as follows: $10 \mu g$ of denatured BSA is incubated at 37°C for 24 hours with $2 \mu l$ 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.

Directions for use

- 1. Add up to 100 µg of glycoprotein or 1 nmol of oligosaccharide to tube.
- 2. Add de-ionized water to a total of 14 μ l.
- 3. Add 4 µl 5x Reaction Buffer 6.0.
- 4. Add 2 µl Neuraminidase Cp.
- 5. Incubate at 37°C for 1 hour.

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

References:

Corfield, A. P., H. Higa, J. C. Paulson and R. Schauer. The specificity of viral and bacterial sialidases for alpha(2-3) and alpha(2-6)-linked sialic acids in glycoproteins. Biochim Biophys Acta 744:121-126 (1983).

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).

Kobata, A. Use of endo- and exoglycosidases for structural studies of glycoconjugates. Anal Biochem 100:1-14 (1979).

Prime, S. J. Dearnley, A. M. Venton, R. B. Parekh and C. J. Edge. Oligosaccharide sequencing based on exo- and endoglycosidase digestion and liquid chromatographic analysis of the products. J Chromatogr A 720:263-274 (1996).

Roggentin, P, B. Rothe, F. Lottspeich and R. Schauer. Cloning and sequencing of a Clostridium perfringens neuraminidase gene. FEBS Lett 238: 31-34 (Sept 1988).

Roggentin P., R. G. Kleineidam and R. Schauer. Diversity in the properties of two neuraminidase isoenzymes produced by Clostridium perfringens spp . Biol Chem Hoppe-Seyler 376: 569-575 (1995).

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This product is intended for *in vitro* research only.

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