3050 Spruce Street, St. Louis, MO 63103 USA Tel: (800) 521-8956 (314) 771-5765 Fax: (800) 325-5052 (314) 771-5757 email: techservice@sial.com sigma-aldrich.com

# **Product Information**

α(2→3,6,8,9)-Neuraminidase from *Arthrobacter ureafaciens*, recombinant, expressed in *E. coli* 

Catalog Number **N8271** Storage Temperature 2–8 °C

CAS RN 9001-67-6 EC 3.2.1.18

Synonyms: Sialidase, *N*-Acetylneuraminidase, *N*-Acetylneuraminate glycohydrolase

# **Product Description**

Two major classes of oligosaccharides (glycans) may be attached to glycoproteins. *N*-linked glycans are attached to the amide side chain of some asparagine (Asn) residues, which form part of the consensus sequence, AsnXaaSer/Thr, while *O*-linked glycans may be added to the hydroxyl side-chain of Ser or Thr residues. The terminal residues on these glycan chains are commonly *N*-acylneuraminic acids (sialic acids). Neuraminidase can be used directly on intact glycoproteins or purified glycans as a gentle means of removing sialic acid (see Figure 1).

Recombinant neuraminidase from *Arthrobacter ureafaciens*, expressed in a glycosidase-free *E. coli* host, is a highly purified enzyme preparation, which releases  $\alpha(2\rightarrow 3)$ ,  $\alpha(2\rightarrow 6)$ ,  $\alpha(2\rightarrow 8)$ , and  $\alpha(2\rightarrow 9)$  linked sialic acids from complex glycans. The relative rates of cleavage are reported to be as follows:<sup>1,2</sup>

$$\alpha(2\rightarrow6) > \alpha(2\rightarrow3) > \alpha(2\rightarrow8)$$
 and  $\alpha(2\rightarrow9)$ 

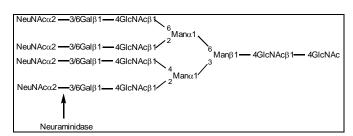
These rates make little difference in practice, as sufficient enzyme is used to ensure that all linkages are released. This broad spectrum of activity makes the enzyme ideal for complete non-specific removal of sialic acid groups prior to analysis. Glycolipids can also be digested with this enzyme in the presence of a detergent, such as sodium taurodeoxycholate.<sup>3</sup>

Molecular mass: ~88 and ~95 kDa (two active species)

pH Range: 4.5–8.0 (depending upon the substrate)
Optimal pH: 5.0–5.5 (with *N*-acetylneuraminyllactose)
Optimal pH: 4.3–4.5 (with colominic acid,
poly-*N*-acetylneuraminic acid)

Figure 1.

Neuraminidase reactions



The enzyme is supplied in 20 mM Trizma®-HCl, pH 7.5.

Specific activity: ≥135 units/mg protein

Unit Definition: One unit will hydrolyze 1  $\mu$ mole of 4-methylumbelliferyl  $\alpha$ -D-N-acetylneuraminide per minute at pH 5.0 at 37 °C.

Other activities:

Protease (not detected)

β-Galactosidase (not detected)

 $\alpha$ -mannosidase (not detected)

β-hexosaminidase (not detected)

 $\alpha$ -fucosidase (not detected)

#### Components

 $\alpha(2\rightarrow3,6,8,9)$ -Neuraminidase (Catalog Number N8271)

5× Reaction Buffer (Catalog Number R0266) – 250 mM sodium phosphate, pH 6.0

## **Precautions and Disclaimer**

This product is for R&D use only, not for drug, household, or other uses. Please consult the Safety Data Sheet for information regarding hazards and safe handling practices.

## Storage/Stability

It is recommended to store the product at 2–8 °C. Both the enzyme and buffer are expected to retain activity for at least 1 year if stored unopened at 2–8 °C.

Do Not Freeze.

Once diluted, the enzyme solution should be stored at 2–8 °C and used within 7 days. The diluted buffer should be stored at 2–8 °C and used within 7 days.

#### **Procedure**

- Dispense one nanomole (or less) of substrate in a total volume of 14 μL into a microcentifuge tube.
- 2. Add to this 4 µL of 5× Reaction Buffer.
- 3. Then add 2  $\mu$ L of neuraminidase solution.
- 4. Cap the tube and incubate at 37 °C for 1 hour.
- 5. Stop the reaction by heating for 5 minutes at 100 °C.
- 6. Allow the sample to cool and centrifuge briefly to ensure that all liquid is at the base of the tube.

## References

- 1. Uchida, Y. et al., J. Biochem., **86(5)**, 1573-1585 (1979).
- 2. Supplier data
- 3. Saito, N. et al., J. Biol. Chem., **254(16)**, 7845-7854 (1979).

Trizma is a registered trademark of Sigma-Aldrich Co. LLC.

ADM, CS, AE, GCY, MAM 10/18-1