**α2-3 Neuraminidase**

**Source:** Cloned from *Salmonella typhimurium* LT2 and overexpressed in *E. coli* (1).

Supplied in: 50 mM NaCl, 20 mM Tris-HCl (pH 7.5 @ 25°C), and 5 mM Na₂EDTA.

**Reagents Supplied with Enzyme:**
- 10X G4 Reaction Buffer
- 100X BSA

**Reaction Conditions:**
- 1X G4 Reaction Buffer: 50 mM Sodium Citrate (pH 6.0 @ 25°C), 100 mM NaCl. Supplement with 100 µg/ml BSA. Incubate at 37°C.

Note: To hydrolyze α2-3 linkages selectively, an initial 10-fold dilution of this enzyme, using 1X G4 Reaction Buffer supplemented with 100 µg/ml BSA, is recommended.

Optimal incubation times and enzyme concentrations must be determined empirically for a particular substrate.

**Quality Controls**

**Glycosidase Assays:** 500 units of α2-3 Neuraminidase were incubated with 0.1 mM of fluorescently-labeled oligosaccharides and glycopeptides, in a 10 µl reaction for 20 hours at 37°C. The reaction products were analyzed by TLC for digestion of substrate.

No other glycosidase activities were detected (ND) with the following substrates:

- β-N-Acetyl-glucosaminidase: GlcNAcβ1-4GlcNAcβ1-4GlcNAc-AMC ND
- α-Fucosidase: Fucx1-2Galβ1-4Glc-AMCGalβ1-4 (Fucx1-3)GlcNAcβ1-3Galβ1-4Glc-AMC ND
- β-Galactosidase: Galβ1-3GlcNAcβ1-4Galβ1-4Glc-AMC ND
- α-Galactosidase: Galα1-3Galβ1-4Galβ1-3Gal-AMC ND

**Specific Activity:** ~11,300,000 units/mg.

**Molecular Weight:** 41,000 daltons.

**Quality Assurance:** No contaminating exoglucosidase or proteolytic activity could be detected.

---

**α2-3 Neuraminidase**

**Source:** Cloned from *Salmonella typhimurium* LT2 and overexpressed in *E. coli* (1).

Supplied in: 50 mM NaCl, 20 mM Tris-HCl (pH 7.5 @ 25°C), and 5 mM Na₂EDTA.

**Reagents Supplied with Enzyme:**
- 10X G4 Reaction Buffer
- 100X BSA

**Reaction Conditions:**
- 1X G4 Reaction Buffer: 50 mM Sodium Citrate (pH 6.0 @ 25°C), 100 mM NaCl. Supplement with 100 µg/ml BSA. Incubate at 37°C.

Note: To hydrolyze α2-3 linkages selectively, an initial 10-fold dilution of this enzyme, using 1X G4 Reaction Buffer supplemented with 100 µg/ml BSA, is recommended.

Optimal incubation times and enzyme concentrations must be determined empirically for a particular substrate.

**Quality Controls**

**Glycosidase Assays:** 500 units of α2-3 Neuraminidase were incubated with 0.1 mM of fluorescently-labeled oligosaccharides and glycopeptides, in a 10 µl reaction for 20 hours at 37°C. The reaction products were analyzed by TLC for digestion of substrate.

No other glycosidase activities were detected (ND) with the following substrates:

- β-N-Acetyl-glucosaminidase: GlcNAcβ1-4GlcNAcβ1-4GlcNAc-AMC ND
- α-Fucosidase: Fucx1-2Galβ1-4Glc-AMCGalβ1-4 (Fucx1-3)GlcNAcβ1-3Galβ1-4Glc-AMC ND
- β-Galactosidase: Galβ1-3GlcNAcβ1-4Galβ1-4Glc-AMC ND
- α-Galactosidase: Galα1-3Galβ1-4Galα1-3Gal-AMC ND

**Specific Activity:** ~11,300,000 units/mg.

**Molecular Weight:** 41,000 daltons.

**Quality Assurance:** No contaminating exoglucosidase or proteolytic activity could be detected.
**α-Mannosidase:**
Manα1-3Manβ1-4GlcNAC-AMC  
Manα1-6Manα1-6(Manα1-3)Man-AMC  

**β-Glucosidase:**
Glcβ1-4Glcβ1-4Glc-AMC  

**β-Xylosidase:**
Xylβ1-4Xylβ1-4Xylβ1-4Xyl-AMC  

**β-Mannosidase:**
Manβ1-4Manβ1-4Man-AMC  

**Endo F₁, F₂, H:**
Dansylated invertase high mannose.  

**Endo F₁, F₂:**
Dansylated fibrinogen biantennary.  

**PNGase F:**
Fluoresceinated fetuin triantennary.  

**Protease Assay:** After incubation of 500 units of α2-3 Neuraminidase with 0.2 nmol of a standard mixture of proteins in a 20 µl reaction, for 20 hours at 37°C, no proteolytic activity could be detected by SDS-PAGE.

**Note:** Store at 4°C or in small aliquots at –20°C. Avoid repeated freeze/thaw cycles.

**References:**