Rev. 2025-06-17

α2.3 Sialidase contents

Catalog #	Description	Size	M. W.	Purity	pH Range	Storage
GE0302	α2,3 Sialidase	5,000 units, lyophilized	77,512	> 95%	4.0-8.0	-20°C, up to 12 months
BA0801	10X Reaction Buffer 4	1 mL			7.5	4 to 25°C
BA1201	10X Reaction Buffer 8	1 mL			6.0	4 to 25°C

This product is for research use only and not for resale or for any use in the manufacture of a therapeutic or for any diagnostic purpose.

Product description: This product is a recombinant neuraminidase (exo- α -sialidase; glycosyl hydrolase family GH33, EC #3.2.1.18), cloned from *Streptococcus pneumoniae* and expressed in *Escherichia coli* with an *N*-terminal 8xHis tag. The 8xHis tag may be removed by digestion with FasTEVTM (Cat #GE0501), a TEV protease with enhanced stability and catalytic activity.

This enzyme preferentially releases terminal $\alpha 2,3$ -linked *N*-acetylneuraminic acid (Neu5Ac) from oligosaccharides, complex carbohydrates, and glycoproteins.



This product does not contain any detectable activities of proteases or other glycosidases.

 α 2,3 Sialidase is supplied with two 10X Reaction Buffers to ensure optimal digestion and ease of use. Reaction Buffer 4 (Cat #BA0801) is used for reactions that require higher than neutral pH buffering, and Reaction Buffer 8 (Cat #BA1201) is the optimal buffer for most digestions.

Unit definition: One unit is defined as the amount of α 2,3 Sialidase required to catalyze the release of 1 nanomole of p-nitrophenol (pNP) from 2-0-(p-nitrophenyl)- α -D-N-acetylneuraminic acid (pNP-Neu5Ac) in 1 min at 37°C in 100 μ L 1X Reaction Buffer 4 (50 mM Tris-HCl, 100 mM NaCl, pH 7.5).

Activity assay: One unit of enzyme is added to $100~\mu L$ of $500~\mu M$ pNP-Neu5Ac in 1X Reaction Buffer 8 (50mM Bis-Tris, 100mM NaCl, pH 6.0) at $37^{\circ}C$ for 30 min, followed by addition of $100~\mu L$ of a stop solution (0.2~M sodium borate, pH 9.8). Measure absorption at 405~nm on a plate reader.

Product reconstitution: Dissolve the lyophilized product in $100 \, \mu L$ of molecular grade water to make a 50,000 units/ml (Cat #GE0302) solution in 1X Reaction Buffer 4. Once reconstituted, store at 4°C for up to 5 days or -20°C for up to 6 months. Aliquoting is recommended to avoid repeated freeze-thaw cycles.

Suggested protocol for protein desialylation:

1. Mix the following components in a microfuge tube:

Glycoprotein (e.g., fetuin; user supplied) 10X Reaction Buffer 8 (Cat #BA1201) α2,3 Sialidase (Cat #GE0302) Molecular grade water

1 nanomole (2-100 μg) 10 μL

 $1.0~\mu L$ (50 units) to $100~\mu L$ final volume

- 2. Incubate at 37°C for 1 h.
- 3. Analyze by Western blot or other method to determine the extent of desialylation on the substrate. Suggested 1° probes: biotinylated SiaFindTM α 2,3-Specific Lectenz® (Cat #SK2301B), SiaFindTM Pan-Specific Lectenz® (Cat #SK0501B), or SiaFindTM Pan-Specific Lectenz® 2.0 (Cat #SK0502B).

Reference: Gut H, et al. FEBS Lett. 2008 Oct 15;582(23-24):3348-52. PMID: 18775704.