

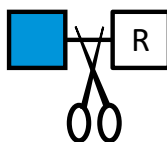
**$\beta$ -N-Acetylglucosaminidase contents**

Catalog #	Description	Size	M. W.	Purity	pH range	Storage
GE1201	$\beta$ -N-Acetylglucosaminidase	500 units, lyophilized	101,594	> 95%	3.5-7.5	-20°C, up to 12 months
BA0801	10X Reaction Buffer 4	1 mL			7.5	4 to 25°C
BA1101	10X Reaction Buffer 7	1 mL			5.0	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 recombinant  $\beta$ -N-Acetylglucosaminidase (glycosyl hydrolase family GH20; E.C. 3.2.1.52), cloned from *Streptococcus pneumoniae* and expressed in *Escherichia coli* with an N-terminal 8xHis tag. The 8xHis tag may be removed by digestion with FasTEV™ (Cat #GE0501), a TEV protease with enhanced stability and catalytic activity.

This enzyme catalyzes the hydrolysis of the non-reducing terminal N-Acetylglucosamine (GlcNAc) from oligosaccharides and glycoprotein substrates.



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

**$\beta$ -N-Acetylglucosaminidase** 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 7 (Cat #BA1101) is the optimal buffer for most digestions.

**Unit definition:** One unit is defined as the amount of enzyme required to catalyze the release of 1 nmole of *p*-nitrophenol (*p*NP) from *p*-nitrophenyl-N-acetyl- $\beta$ -D-glucosaminide (*p*NP-GlcNAc) in 1 min at 37°C in 100  $\mu$ 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 *p*NP-GlcNAc in 1X Reaction Buffer 7 (50 mM sodium citrate, pH 5.0) at 37°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 molecular grade water to make a 5,000 units/mL (Cat #GE1201) solution in 1X Reaction Buffer 4. Once reconstituted, store at 4°C for up to 7 days or -20°C for up to 3 months. Aliquoting is recommended to avoid repeated freeze-thaw cycles.

**Suggested protocol for removal of GlcNAc from glycoproteins:**

- Mix the following components in a microfuge tube:

Glycoprotein (e.g. G0N-BSA; Cat #NG0101)	10 $\mu$ g
10X Reaction Buffer 7 (Cat #BA1101)	2 $\mu$ L
$\beta$ -N-Acetylglucosaminidase (Cat #GE1201)	1 $\mu$ L (5 units)
Molecular grade water	to 20 $\mu$ L final volume
- Incubate at 37°C for 1h.
- Analyze 2  $\mu$ L reaction by Western/lectin blot (or ELISA) to determine the completion of reaction. Suggested 1° probe: lectin GSL II, biotinylated.

**Reference:** Clarke VA, et al. J Biol Chem. 1995 Apr 14;270(15):8805-14. PMID: 7721787

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