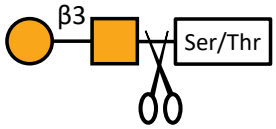
_O_-Glycosidase contents

Catalog #	Description	Size	M. W.	Purity	pH range	Storage
GE1301	*O*-Glycosidase	1,000 units (0.1 mL)	147,000	> 90%	4.5-7.5	4°C, up to 6 months
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 a recombinant *O*-Glycosidase (endo-α-*N*-acetylgalactosaminidase, *O*-glycanase; glycosyl hydrolase family GH101; E.C. 3.2.1.97), cloned from *Enterococcus faecalis* 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 unsubstituted *O*-glycans from Ser/Thr residues of glycoprotein substrates. It predominantly cleaves the Core 1 structure (T antigen).



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

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 500 µM *p*-nitrophenyl 2-acetamido-2-deoxy-3-O-(β-D-galactopyranosyl)-α-D-galactopyranoside (Galβ1,3GalNAc-α-pNP) in 1 min at 37°C in 100 µL 1X Reaction Buffer 7 (50 mM sodium citrate, pH 5.0). The reaction is stopped by the addition of 100 µL 0.2 M borate buffer, pH 9.8, followed by measuring absorption at 405 nm.

Form and Storage: The product (Cat #GE1301) is supplied as a solution in 0.1 mL storage buffer (20 mM Tris-HCl, 100 mM NaCl, pH 7.5) with 10X Reaction Buffer 7 to ensure optimal digestion and ease of use. It is stable at 4°C for up to 6 months.

Suggested protocol for removal of unsubstituted _O_-glycan from glycoproteins:

1. Mix the following components in a microfuge tube:

Glycoprotein (e.g. asialofetuin)	10 µg
10X Reaction Buffer 7 (Cat #BA1101)	2 µL
O-Glycosidase (Cat #GE1301)	1 µL (10 units)
Molecular grade water	to 20 µL final volume

2. Incubate at 37°C for 1h.
3. Analyze 2 µL reaction by ELISA (or Western blot) to determine the completion of reaction. Suggested 1° probe: lectin PNA, biotinylated.

Reference: Goda HM, et al. Biochem Biophys Res Commun. 2008 Oct 31;375(4):441-6. PMID: 18725192