PNGase F

Cat.No.: HYI151

Product Description

PNGase F is the most effective enzymatic method for removing almost all N-linked oligosaccharides from glycoproteins. PNGase F is an amidase, which cleaves between the innermost GlcNAc and asparagine residues of high mannose, hybrid, and complex oligosaccharides from N-linked glycoproteins.

Components

Components	Size
PNGase F	50 µl
10×Glycoprotein Denaturing Buffer	1000 µl
10×GlycoBuffer 2	1000 µl
10% NP-40	1000 µl

Specification

Appearance	Colorless Liquid
Protein purity	≥95% (from SDS-PAGE)
Activity	≥500,000 U/mL
Exoglycosidase	No activity could be detected
Endoglycosidase F1	No activity could be detected
Endoglycosidase F2	No activity could be detected
Endoglycosidase F3	No activity could be detected
Endoglycosidase H	No activity could be detected
Protease	No activity could be detected

Properties

EC number	3.5.1.52 (Recombinant from microorganism)
Molecular weight	35 kDa (SDS-PAGE)
Isoelectric point	8.14

Optimum pH	7.0-8.0
Optimum temperature	65 °C
Substrate specificity	Cleaving glycosidic bonds between GlcNAc and asparagine residues
Recognition sites	N-linked glycans unless containing α1-3 fucose
Activators	DTT
Inhibitor	SDS
Storage temperature	-25～-15 ℃
Heat Inactivation	A 20 μL reaction mixture containing 1 μL of PNGase F is inactivated by incubation at 75 °C for 10 minutes.

Applications

This enzyme is useful for removal of carbohydrate residues from proteins.

Unit definition

One unit(U) is defined as the amount of enzyme required to remove >95% of the carbohydrate from 10 μg of denatured RNase B in 1 hour at 37 °C in a total reaction volume of 10 μL.

Reaction conditions

1. Dissolve 1-20 μg of glycoprotein withdeionized water, add 1 μl 10×Glycoprotein Denaturing Buffer and H_2O (if necessary) to make a 10 μl total reaction volume.
2. Incubate at 100 °C for 10 min, cool it on ice.
3. Add 2 μl 10×GlycoBuffer 2, 2 μl 10% NP-40 and mix.
4. Add 1-2 μl PNGase F and H2O (if necessary) to make a 20 μl total reaction volume and mix.
5. Incubate reaction at 37 °C for 60 min.
6. For SDS-PAGE analysis or HPLC analysis.