SmartEnzymes™

GENOVIS

PNGase F

Lyophilized

STORE AT

-20°C

FOR RESEARCH USE ONLY

Instructions for Use

PNGase F Lyophilized 1000 units (G1-PF1-010)
Process 1 mg glycoprotein

PNGase F Lyophilized 5 × 1000 units (G1-PF1-050)
Process 5 × 1 mg glycoprotein

DOWNLOAD INSTRUCTIONS FOR USE

www.genovis.com/ifu-G1-PF1

www.genovis.com

Lyophilized Enzyme for Hydrolysis of N-glycans on Glycoproteins

PNGase F (Peptide N-glycosidase F) is a glycoamidase hydrolyzing the amide bond between the polypeptide asparagine and the innermost GlcNAc of all mammalian asparagine-linked complex, hybrid, or high-mannose oligosaccharides. The activity of PNGase F on some glycoproteins can be slow or inhibited due to steric hindrance - longer incubation times or denaturation of the glycoprotein may in these cases be required.

PNGase F is derived from *Elizabethkingia meningoseptica* and expressed in *E. coli*. The enzyme contains a His-tag and has a molecular weight of 36 kDa.

UNIT DEFINITION

1 unit PNGase F Lyophilized removes N-glycans from ≥95% of 1 µg glycoprotein (etanercept) when incubated in PBS (10 mM sodium phosphate, 150 mM NaCl) pH 7.4 at 37°C for 1 hour.

CONTENT AND STORAGE

PNGase F Lyophilized is supplied lyophilized in 50 mM HEPES buffer pH 7.5, with no preservatives added.

PNGase F Lyophilized is shipped cold and should be stored at -20°C upon arrival. After reconstitution, PNGase F Lyophilized is stable for at least 1 month at +4-8°C.

PNGase F Lyophilized is for R&D use only.

QUALITY CONTROL

PNGase F Lyophilized is tested to meet the specifications and lot-to-lot consistency.

PNGase F Lyophilized is tested for absence of microbial contamination with blood agar plates, Sabouraud dextrose agar plates and fluid thioglycollate medium.

YOU MIGHT ALSO BE INTERESTED IN

PNGase F Immobilized
Immobilized enzyme for hydrolysis of N-glycans in spin columns

OglyZOR®
Hydrolysis of core 1 O-glycans

GlycINATOR®
Hydrolysis of all Fc N-glycans

OmniGLYZOR™
Hydrolysis of N- and mucin-type O-glycans

Preparations

Additional Materials Required

- Reaction buffer: PBS pH 7.4.[1]

1. Other common buffers in pH 6.0-9.0 over a wide range of ionic strengths (0-1000 mM NaCl) may also be used. Optimization may be required if buffers other than the recommended are used.

Hydrolysis of N-glycans on Glycoproteins

Sample Preparation
Prepare the glycoprotein in the reaction buffer. The final glycoprotein concentration in the reaction should be 0.5-5 mg/ml.

1. Prepare PNGase F

1.1 Reconstitute PNGase F in 50 µl ddH_2O to a concentration of 20 units/µl.

2. Add PNGase F

2.1 Add 1 unit PNGase F / 1 µg glycoprotein.[2]

3. Enzymatic Reaction

3.1 Incubate for 1 h to overnight at 37°C.[3]

Note: Optimization of the enzyme concentration, incubation time and incubation temperature may be needed depending on the substrate. Some glycoproteins require denaturing and/or reducing reaction conditions to reach full deglycosylation.

2. A higher enzyme concentration may increase digestion efficiency of individual glycoproteins. This requires optimization.
3. Incubation temperature can be increased to up to 50°C for a faster deglycosylation. A surfactant may be required to prevent precipitation of the substrate protein.

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