Active Recombinant Elizabethkingia miricola PNGase F

Cat. No. PNGase F-014E **Lot. No.** (See product label)

SPECIFICATION

Product Overview	Recombinant PNGase F gene from Elizabethkingia miricola expressed in E. coli
Species	Elizabethkingia miricola
Source	E. coli
Description	PNGase F is suitable for the release of all types (high-mannose, hybrid and complex) N-linked glycans from glycoproteins and glycopeptides. PNGase F will not remove oligosaccharides containing α(1-3) linked core fucose commonly found on plant glycoproteins. PNGase F cleaves asparagine-linked (N-linked) oligosaccharides from glycoproteins. PNGase F deaminates asparagine to aspartic acid, leaving the oligosaccharides intact. Denaturation increases the rate of cleavage up to 100×. Most native proteins can still be completely N-deglycosylated but incubation time must be increased. PNGase F will remain active under incubation conditions for at least 72 hours. PNGase F will not remove oligosaccharides containing α(1,3)-linked core fucose commonly found on plant glycoproteins; for this purpose, use peptide N-glycosidase A.
Form	Sterile-filtered solution
EC	3.5.1.52
Specificity	PNGase F cleaves asparagine-linked (N-linked) oligosaccharides from glycoproteins. PNGase F deaminates asparagine to aspartic acid, leaving the oligosaccharides intact. Denaturation increases the rate of cleavage up to 100×. Most native proteins

Tel: 1-631-559-9269 1-516-512-3133
Email: info@creative-biomart.com Fax: 1-631-938-8127
45-1 Ramsey Road, Shirley, NY 11967, USA

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Contents	60 µL aliquot of recombinant PNGase F (0.3 U) in 20 mM Tris-HCl, pH 7.5 5× PNGase F Reaction Buffer 7.5 for PNGase F-250 mM sodium phosphate, pH 7.5 PNGase F Denaturation Solution-2% SDS, 1 M Beta-mercaptoethanol PNGase F Triton X-100-15% solution
Bio-activity	Activity: 5 U/mL Specific Activity: > 25 U/mg Defined as the amount of enzyme required to catalyze the release of N-linked oligosaccharides from 1 micromole of denatured RNase B in 1 minute at 37 centigrade, pH 7.5. Cleavage is monitored by SDS-PAGE (cleaved RNase B migrates faster).
Molecular Mass	36 kDa
Suggested usage	1. Add up to 200 µg of glycoprotein to an Eppendorf tube. Adjust to 35 µL final volume with de-ionized water. 2. Add 10 µL 5× PNGase F Reaction Buffer 7.5 and 2.5 µL of PNGase F Denaturation Solution. Heat at 100 centigrade for 5 minutes. 3. Cool. Add 2.5 µL of PNGaseF Triton X-100 and mix. 4. Add 2.0 µL of PNGaseF to the reaction. Incubate 3 hours at 37 centigrade.
Unit Definition	One unit of PNGase F activity is defined as the amount of enzyme required to catalyze the release of N-linked oligosaccharides from 1 micromole of RNase B in 1 minute at 37 centigrade, pH 7.5. Cleavage is monitored by SDS-PAGE (cleaved RNase B migrates faster).
Purity	PNGase F is tested for contaminating protease as follows: 10 µg of denatured BSA is

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	incubated at 37 centigrade for 24 hours with 2 µL of enzyme. SDS-PAGE analysis of the treated BSA shows no evidence of degradation. The production host strain has been extensively tested and does not produce any detectable glycosidases.
Stability	Several days exposure to ambient temperatures will not reduce activity. Stable at least 12 months when stored properly.
Storage	Store enzyme at 4 centigrade.
Storage Buffer	The enzyme is provided as a sterile-filtered solution in 20 mM Tris-HCl, pH 7.5

GENE INFORMATION

Synonyms	PNGase F; Peptide N Glycosidase F; N-Glycosidase; N-Glycanase

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