

PNGase F Recombinant

Cat.# PT-EZ-PNGaseF-15

Size. 15,000 units **Price:** \$50

Cat.# PT-EZ-PNGaseF-75

Size. 75,000 units **Price:** \$200

Description: PNGase F can be used to cleave N-glycans attached to proteins and antibodies. This PNGase F is tag free cloned from *Elizabethkingia miricola* (formerly known as *Flavobacterium meningosepticum*).

Quality control assay: Denatured RNaseB and human monoclonal antibody were used as substrates to test PNGase F activity. (Figure 1)

Stability: PNGase F retains >60% activity after left at room temperature for over 72 hours. Long term storage at – 20 C or below

Purity: >95% by SDS-PAGE gel

Unit definition: One unit is defined as the amount of enzyme required to removed >95% of the glycans from 10 ug of denatured RNase B in 1 hour at 37 C.

Concentration: 50,000 units/mL

Formulation: 20 mM Tris pH8, 50% glycerol.

Source: E. Coli

Reference:

1. Maley, F. et al. (1989). Anal. Biochem. 180, 195-204.

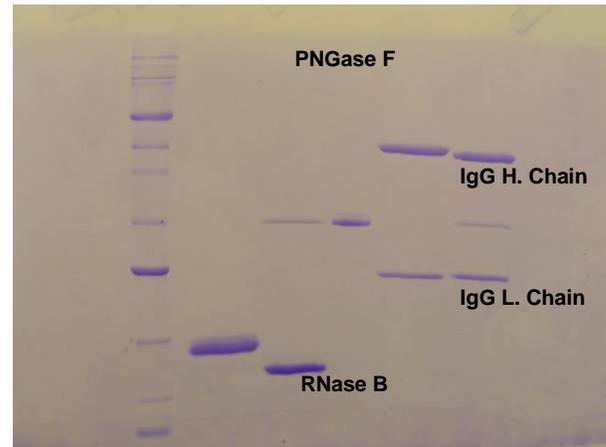


Figure 1. Deglycosylation results of denatured RNaseB and human monoclonal antibody

Lane 1. Marker

Lane 2. RNase B

Lane 3. RNase B PNGase F treated

Lane 4. PNGase F

Lane 5. Human mab

Lane 6. Human mab PNGase F treated

Protocol for denaturing conditions

1. Denature 20 ug of glycoprotein in 0.5% SDS, 50mM DTT and 50mM Tris buffer pH8 by heating at 95 C for 5 minutes.
2. Then add NP-40 or Triton X-100 to sample (1% final concentration).
3. Add 10 uL PNGase per 20 ug of glycoprotein
4. Incubate at 37 C for 1 hour.

Protocol for non-denaturing conditions

1. Add 50 uL PNGase per 20 ug of glycoprotein in 50mM Tris buffer pH8
2. Incubate at 37 C for 18 hours.

Note:

1. Recommended substrate concentration for reaction is 0.1–1 mg/mL
2. NP-40 or Triton X-100 prevents SDS inhibition of PNGase F activity.
3. PNGase F does not cleave N-glycans containing core α 1-3 fucose.