

PRODUCT INFORMATION**PNGase F, recombinant (lyophilized)****Cat. no. 36405****PRODUCT DESCRIPTION**

PNGase F is a mutant recombinant glycosidase from *Flavobacterium meningosepticum* and expressed and purified from *E. coli*. The enzyme catalyzes the cleavage of N-linked oligosaccharides between the innermost GlcNAc and asparagine residues of high mannose, hybrid and complex oligosaccharides from N-linked glycoproteins. The proprietary changes made to PNGase F have been shown to have unique characteristics when compared to other commercially-available sources of PNGase F:

- No need for refrigerated transport, storage at room temperature
- Does not need a denaturing step
- Works on native glycoproteins and serum glycoproteins in only minutes at room temperature
- Digestion leads to a more complete glycan release as compared to other commercially-available enzymes

Especially designed and tested for mass spectrometry imaging of tissue samples

Concentration after reconstitution with 50 µl H₂O dist.: 10⁶ units/ml (2.0 mg/ml)

Molecular Weight: approx. 36 kDa

Storage of the lyophilisate: + 15 °C to + 30 °C

Storage after reconstitution: + 2 °C to - 20 °C (avoid multiple freeze-thaw cycles).**1. Deglycosylation of proteins under denaturing conditions**

Deglycosylation may be visualized by gel-shift on SDS PAGE.

Required Materials:

- 5 % (w/v) SDS
 - 1 M DTT
 - 1x Phosphate Buffered Saline (PBS), pH 7.4
 - 10 % (w/v) NP-40
 - Blue ice
- Add up to 50 µg of the target glycoprotein in 1x PBS to a final volume of 11 µl.
 - Add 1 µl of 5 % SDS and 1 µl of 1M DTT.
 - Denature the sample for 10 min at 95 °C and cool the sample by incubation on Ice.

Note: Other buffers can be used if pH ranges between 6 - 10.

- Add 2 µl of 10 % NP-40.
- Add 1 µl of recombinant PNGase F.
- Incubate at 37 °C for 30 minutes.

2. Deglycosylation of proteins under native conditions

Deglycosylation under native (non-denaturing) conditions may require increasing both the amount of PNGase F used and the incubation time.

Required Materials:

- 1x Phosphate Buffered Saline (PBS), pH 7.4
- Add up to 20 µg of glycoprotein in 1x PBS to a final volume of 18 µl.
 - Add 2 µl of recombinant PNGase F.
 - Incubate at 37 °C for 0.5 - 24 hours.

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