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Endo H Denatured Protocol for Deglycosylating Glycoproteins

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ABSTRACT

Endo H cleaves Asparagine-linked hybrid or high mannose oligosaccharides, but not complex oligosaccharides. It cleaves between the two N-acetylglucosamine residues in the diacetylchitobiose core of the oligosaccharide, generating a truncated sugar molecule with one N-acetylglucosamine residue remaining on the asparagine. In contrast, PNGase F removes the oligosaccharide intact. Detergent and heat denaturation may increase the rate of cleavage for some glycoproteins.

MATERIALS

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Endo H QA-Bio Inc Catalog #E-EH02

OPEN ACCESS

DOI:

dx.doi.org/10.17504/protocols.io.sedeba6

External link: <http://www.qa-bio.com/docs/E-EH02.QA-Bio.specsheet.pdf>

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Protocol status: Working
We use this protocol and it's working

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14501

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- 1** Add up to 200 µg of glycoprotein to an Eppendorf tube. Adjust to 37.5 µl final volume with de-ionized water.
- 2** Add 10 µl 5x Reaction Buffer 5.5 and 2.5 µl of Denaturation Solution. Heat at 100°C for 5 minutes. (NOTE: It is not necessary to add Triton X-100. SDS will not inactivate Endo H.)
- 3** Add 2.0 µl of Endo H to the reaction. Incubate 3 hours at 37°C.
- 4** Monitor cleavage by SDS-PAGE.