



Endo F2 (Endoglycosidase F2)

Endo-beta-N-acetylglucosaminidase F2

Source

recombinant gene from *Elizabethkingia miricola* in *E. Coli*

Catalog Number E-EF02

E-EF02	60 µl
E-EF02-20	20 µl
E-EF02-200	200 µl

EC 3.2.1.96

Recommended Reagents

included with E-EF02:

1 vial: 5x Reaction Buffer - 400 µl
250 mM sodium acetate, pH4.5

Activity ≥ 5 U/ml

Specific Activity ≥ 20 U/mg

Molecular Weight 32 kD

Specific Activity

Defined as the amount of enzyme required to catalyze the release of N-linked oligosaccharides from 1 micro-mole of denatured porcine fibrinogen in 1 minute at 37°C, pH 5.5. Cleavage is monitored by SDS-PAGE (cleaved fibrinogen migrates faster).

Formulation

The enzyme is provided as a sterile-filtered solution in 10 mM sodium acetate, 25mM NaCl, pH 4.5

Storage

Store enzyme at 4°C. Do not freeze.

Specificity

QA-Bio™ Endo F2 cleaves Asparagine-linked biantennary and high mannose glycans (at a 40X reduced rate). 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.

Endoglycosidase F2 is less sensitive to protein conformation than PNGase F and is therefore more suitable for deglycosylation of native proteins. However for optimal results, denaturation of the glycoprotein is recommended.

Quality & Purity

QA-Bio Endo F2 is tested for contaminating protease as follows: 10 µg of denatured BSA is incubated at 37°C 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.

Directions for use

1. Add up to 200 µg of glycoprotein to an Eppendorf tube. Adjust to 38 µl final volume with de-ionized water.
2. Add 10 µl 5x Reaction Buffer 4.5
3. Add 2.0 µl of Endo F2 to the reaction. Incubate 1 hour at 37°C.

Monitor cleavage by SDS-PAGE.

References:

Maley P., R. B. Trimble, A. L. Tarentino and T. H. Plummer Jr. Characterization of glycoproteins and their associated oligosaccharides through the use of endoglycosidases. **Anal Biochem** **180**:195-204 (1989).

Plummer, T. H. Jr, A. W. Phelan and A. L. Tarentino. Porcine fibrinogen glycopeptides: substrates for detecting endo-N-acetylglucosaminidases F2 and F3. **Anal Biochem** **235**:98-101 (1996).

Reddy A., B. G. Grimwood, T. H. Plummer Jr and A. L. Tarentino. High-level expression of the Endo-N-Acetylglucosaminidase F2 gene in E.coli: one step purification to homogeneity. **Glycobiology** **8**:633-636 (1998).

Tarentino, A. L., C. M. Gomez and T. H. Plummer Jr. Deglycosylation of Asparagine-Linked Glycans by Peptide:N-Glycosidase F. **Biochemistry** **24**:4665-4671 (1985).

Tarentino A. L., G. Quinones, W. P. Schrader, L. M. Changchien and T. H. Plummer Jr. Multiple endoglycosidase (Endo) F activities expressed by *Flavobacterium meningosepticum*. Endo F1: molecular cloning, primary sequence, and structural relationship to Endo H. **J Biol Chem** **267**:3868-3872 (1992).

Tarentino A. L., G. Quinones, L. M. Changchien, and T. H. Plummer Jr. Multiple endoglycosidase F activities expressed by *Flavobacterium meningosepticum* endoglycosidases F2 and F3: Molecular cloning, primary sequence, and enzyme expression. **J Biol Chem** **268**(13):9702-9708 (1993).

Tarentino A. L. and T. H. Plummer Jr. Substrate specificity of *Flavobacterium meningosepticum*: Endo F2 and endo F3: purity is the name of the game. **Glycobiology** **4**:771-773 (1994).

Tarentino, A. L. and T. H. Plummer Jr. Enzymatic deglycosylation of asparagine-linked glycans: purification, properties and specificity of oligosaccharidecleaving enzymes from *Flavobacterium meningosepticum*. **Methods in Enzymology** **230**:44-57 (1994).

Tarentino A. L., G. Quinones and T. H. Plummer Jr. Overexpression and purification of non-glycosylated recombinant endo-N-acetylglucosaminidase F3. **Glycobiology** **5**:599-601 (1995).

Trimble, R. B. and A. L. Tarentino. Identification of Distinct Endoglycosidase (Endo) Activities in *Flavobacterium meningosepticum*: Endo F1, Endo F2 and Endo F3. **J. Biol Chem** **266**:1646-1651 (1991).

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This product is intended for *in vitro* research only.

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