Endo F Multi-Kit
includes: Endo F1, Endo F2, Endo F3

Source recombinant *Elizabethkingia meningosepticum* in *E. Coli* (was *Chryseobacterium meningosepticum)*

Catalog Number KE-EFX3

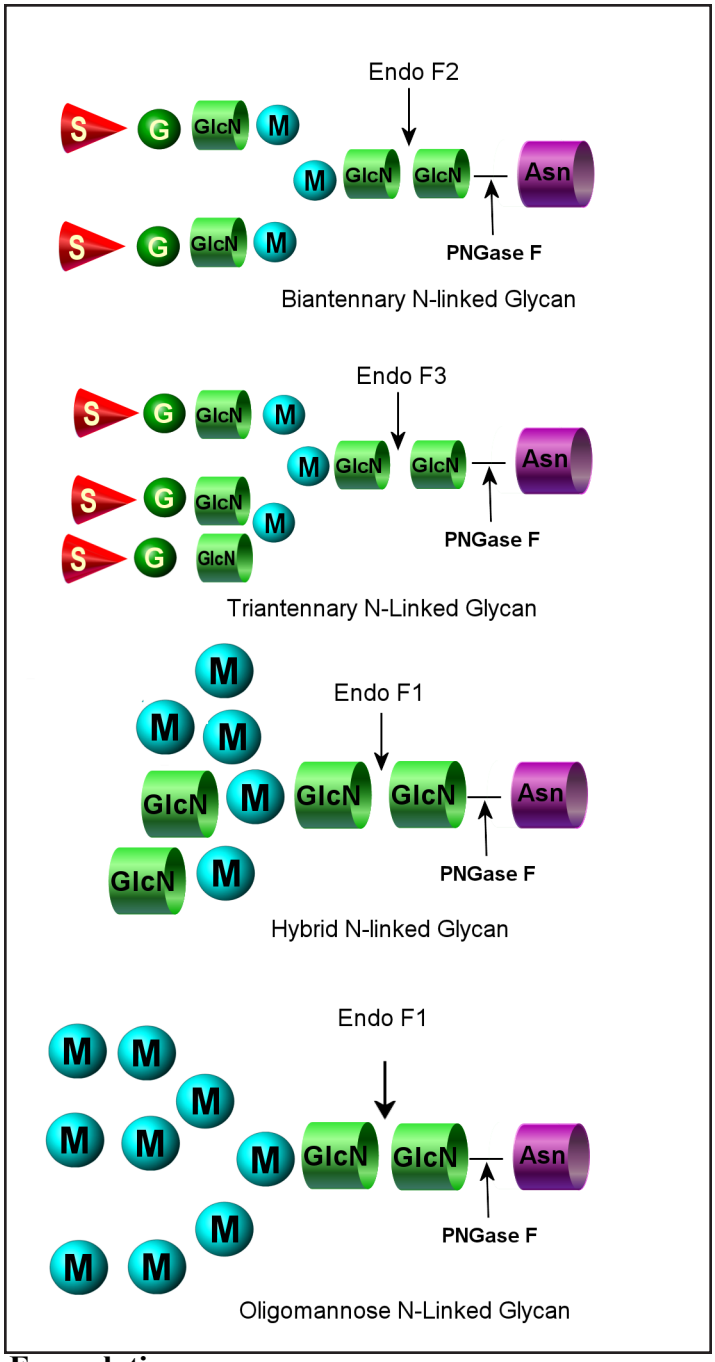
EC 2.7.7.43

Contents
1 vial: Endo F1- 20 µl (0.3 U)
20 mM Tris-HCl pH 7.5
1 vial: Endo F2- 20 µl (0.1 U)
10 mM sodium acetate, 25 mM NaCl, pH 4.5
1 vial: Endo F3- 20 µl (0.1 U)
20 mM Tris-HCl pH 7.5
1 vial: 5x Reaction Buffer - 400 µl
250 mM sodium acetate, pH4.5
1 vial: 5x Reaction Buffer - 400 µl
250 mM sodium phosphate, pH5.5

Specific activity
Endo F1 ≥16 U/mg
Endo F2 ≥20 U/mg
Endo F3 ≥25 U/mg

Activity
Endo F1 ≥17 U/ml
Endo F2 ≥ 5 U/ml
Endo F3 ≥ 5 U/ml

Specific Activity
Defined as the amount of enzyme required to catalyze the release of N-linked oligosaccharides from 1 micro-mole of denatured Ribonuclease B (Endo F1) or porcine fibrinogen peptides (Endo F2/F3) in 1 minute at 37°C, pH 5.5 (PH 4.5 for Endo F3). Cleavage is monitored by SDS-PAGE.



Formulation
The enzymes are provided as a sterile-filtered solution.

Storage
Store enzyme at 4°C. Do not freeze.

Stability
Several days exposure to ambient temperatures will not reduce activity. Stable at least 12 months when stored properly.

KE-EFX3 Endo F Multi-Kit

Specifications - Protocol

QA-Bio
www.QA-Bio.com
info@QA-Bio.com

phone/fax 866-384-2272
phone 760-565-3057

Specificity

QA-Bio™ Endo F1 cleaves Asparagine-linked (N-linked) high mannose or hybrid oligosaccharides. Endo F2 cleaves N-linked biantennary oligosaccharides and high mannose (at a 40X reduced rate).

Endo F3 cleaves free or N-linked fucosylated biantennary or triantennary oligosaccharides,as well as triamannosylchitobiose core structures.

These enzymes cleave 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.

The recombinant version is not glycosylated, which may result in properties differing from the native protein.

Quality & Purity

QA-Bio Endo F1, Endo F2, and Endo F3 are 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 absence of exoglycosidase contaminants is confirmed by extended incubations with the corresponding pNP-glycosides.

Directions for use

1. Add up to 200 µg of glycoprotein to an Eppendorf tube. Adjust to 34 µl final volume with de-ionized water.

2. Add 10 µl Endo F2 &F3 5x Reaction Buffer, 250 mM sodium acetate pH 4.5. Use Endo F1 buffer, 250 mM sodium phosphate pH 5.5 if you are using the Endo F1 enzyme alone.

4. Add 2.0 µl of each enzyme to the reaction. Incubate 3 hours 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).

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).

Warranties and liabilities

QA-Bio warrants that the above product conforms to the specifications described herein. Should the product fail for reasons other than through misuse QA-Bio will, at its option, replace free of charge or refund the purchase price. This warranty is exclusive and QA-Bio makes no other warrants, expressed or implied, including any implied conditions or warranties of merchantability or fitness for any particular purpose. QA-Bio shall not be liable for any incidental, consequential or contingent damages.

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

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QA-Bio
www.QA-Bio.com
info@QA-Bio.com
phone/fax 866-384-2272
phone 760-565-3057