

N-Glycanase (EDTA-Free) from Elizabethkingia meningoseptica, Recombinant

Cat. No. NATE-0483

Lot. No. (See product label)

Introduction

Synonyms N-Glycanase

Product Information

Species Elizabethkingia meningoseptica

Source E. coli

Form A sterile-filtered solution in 20 mM Tris-HCl, 50 mM NaCl (pH 7.5).

Molecular Weight ~35 kDa daltons

Purity The absence of exoglycosidase contaminants was confirmed by extended incubations with the corresponding pNP or MU-glycosides. No protease activity was detectable after incubation of the enzyme with 0.2 mg resorufin-labeled casein for ~18 hours at 37°C according to the method described by Twining.

Activity > 10 U/mg

Optimum pH Optimum: pH 8.6 Range: pH 7.5-9.5

Specificity N-Glycanase (EDTA-Free) releases intact N-linked oligosaccharides from glycoproteins and glycopeptides. Prior denaturation of the glycoprotein substrate by treatment with heat/SDS greatly enhances the rate and reliability of N-glycan removal, although at high concentrations the enzyme can remove intact glycans from undenatured glycoproteins. The site of enzyme cleavage is highly specific, with hydrolysis occurring between asparagine and proximal N-acetyl-glucosamine of most oligomannose, hybrid-and complex-type N-glycans. The enzyme releases 1-amino oligosaccharide, which is hydrolyzed non-enzymatically to form ammonia and free oligosaccharides having an intact chitobiose reducing terminus. The peptide backbone is an important structural determinant since glycan cleavage will not occur from an asparagine having unsubstituted α -amino and carboxyl groups. While di-N-acetylchitobiose is the minimum glycan structural determinant cleavage does not occur if there is core α (1-3)-linked fucose as commonly encountered in plant glycoproteins. Phosphate, sulfate and sialic acid groups attached to the oligosaccharide do not affect cleavage. As a consequence of hydrolysis the asparagine on the peptide is converted to aspartic acid, but otherwise the polypeptide remains intact. True endoglycosidases, such as endo F and endo H, have more restricted specificities and do not release intact oligosaccharides since they cleave within the chitobiose core and leave a single N-acetylglucosamine attached to the polypeptide.

Storage and Shipping Information

Storage Shipped on ice pack for next day delivery. Store enzyme at 2-8°C or -20°C, but avoid repeated freeze-thawing.

Stability Extended incubations may be performed at 25°C rather than 37°C to promote stability of the N-Glycanase.