**Description:** β-N-Acetyl-hexosaminidase is a recombinant protein fusion of β-N-Acetyl-hexosaminidase (1) and maltose binding protein. It has identical activity to β-N-Acetyl-hexosaminidase. β-N-Acetyl-hexosaminidase catalyzes the hydrolysis of terminal β-N-acetyl-galactosamine and glucosamine residues from oligosaccharides.

**Reagents Supplied with Enzyme:**
10X GlycoBuffer 1

**Reaction Conditions:**
1X GlycoBuffer 1:
50 mM Sodium Acetate (pH 5.5 @ 25°C) and 5 mM CaCl₂. Incubate at 37°C.

**Optimal incubation times and enzyme concentrations must be determined empirically for a particular substrate.**

**Glycosidase Assays:**
- **β-N-Acetyl-hexosaminidase:**
  - Unit Definition: One unit is defined as the amount of enzyme required to cleave > 95% of the terminal β-N-acetyl-galactosamine from 1 nmol of GalNAcβ1-4Galβ1-4Glc-7-amino-4-methyl-coumarin (AMC), in 1 hour at 37°C in a total reaction volume of 10 µl.
  - Unit Definition Assay: Two fold dilutions of β-N-Acetyl-hexosaminidase, are incubated with 1 nmol AMC-labeled substrate in 1X GlycoBuffer 1 in a 10 µl reaction. The reaction mix is incubated for 1 hour at 37°C. Separation of reaction products are visualized via thin layer chromatography (3).
  - Specific Activity: ~ 10,000 units/mg
  - Molecular Weight: 100,000 daltons
  - Quality Assurance: No contaminating exoglycosidase or proteolytic activity could be detected.

- **α-Fucosidase:**
  - Unit Definition: One unit is defined as the amount of enzyme required to cleave > 95% of the terminal β-N-acetyl-galactosamine from 1 nmol of GalNAcβ1-3Galβ1-4Glc-AMC, in 1 hour at 37°C in a total reaction volume of 10 µl.
  - Unit Definition Assay: Two fold dilutions of α-Fucosidase, are incubated with 1 nmol AMC-labeled substrate in 1X GlycoBuffer 1 in a 10 µl reaction. The reaction mix is incubated for 1 hour at 37°C. Separation of reaction products are visualized via thin layer chromatography (3).
  - Specific Activity: ~ 10,000 units/mg
  - Molecular Weight: 100,000 daltons
  - Quality Assurance: No contaminating exoglycosidase or proteolytic activity could be detected.

**Quality Controls**

No other glycosidase activities were detected (ND) with the following substrates:
- **α-Fucosidase:**
  - Fucox1-2Galβ1-4Glc-AMC Ga1β1-4
  - (Fucox1-3)GalNAcβ1-3Galβ1-4Glc-AMC

- **β-Galactosidase:**
  - Galβ1-3GalNAcβ1-4Galβ1-4Glc-AMC

- **α-Galactosidase:**
  - Galα1-3Galβ1-4Galα1-3Gal-AMC

**Quality Assurances**

No other glycosidase activities were detected (ND) with the following substrates:
- **α-Fucosidase:**
  - Fucox1-2Galβ1-4Glc-AMC Ga1β1-4
  - (Fucox1-3)GalNAcβ1-3Galβ1-4Glc-AMC

- **β-Galactosidase:**
  - Galβ1-3GalNAcβ1-4Galβ1-4Glc-AMC

- **α-Galactosidase:**
  - Galα1-3Galβ1-4Galα1-3Gal-AMC

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*(See other side)*
### α-Neuraminidase:
Neu5Acα2-3Galβ1-3GlcNAcβ1-3Galβ1-4Glc-AMC ND

### α-Mannosidase:
Manα1-3Manβ1-4GlcNAc-AMC ND
Manα1-6Manα1-6(Manα1-3)Man-AMC ND

### β-Glucosidase:
Glcβ1-4Glcβ1-4Glc-AMC ND

### β-Xylosidase:
Xylβ1-4Xylβ1-4Xylβ1-4Xyl-AMC ND

### β-Mannosidase:
Manβ1-4Manβ1-4Man-AMC ND

### Endo F₁, F₂, H:
Dansylated invertase high mannose. ND

### Endo F₃, F₄:
Dansylated fibrinogen biantennary. ND

### PNGase F:
Fluoresceinated fetuin triantennary. ND

### Protease Assay:
After incubation of 50 units of β-N-Acetyl-hexosaminidase, with 0.2 nmol of a standard mixture of proteins in a 20 µl reaction, for 20 hours at 37°C, no proteolytic activity could be detected by SDS-PAGE.

### *Note:
Non-branched oligosaccharides only.

### References: