**α1-2,3 Mannosidase**

**P0729S**

640 units 32,000 U/ml Lot: 0161304

RECOMBINANT Store at 4°C Exp: 4/14

Description: α1-2,3 Mannosidase is a highly specific exoglycosidase that catalyzes the hydrolysis of α-1-2 and α-1-3 linked α-D-mannopyranosyl residues from oligosaccharides (1).

Specificity:

\[
\begin{align*}
\text{Man} & \rightarrow \text{Man} \\
\alpha & \quad \alpha
\end{align*}
\]

**Detailed Specificity:**

Specificity can vary depending on incubation time and concentration of substrate (Figure 1).

A. 0.1 nm/µl substrate, 1 hour incubation

- Manα1–6 \(\text{Man}_1\)
- Manα1–3 \(\text{Man}_2\)
- Manα1–6 \(\text{Man}_3\)

B. 0.1 nm/µl substrate, 1 hour incubation

- Manα1–6 \(\text{Man}_1\)
- Manα1–6 \(\text{Man}_2\)
- Manα1–3 \(\text{Man}_3\)

C. 0.1 nm/µl substrate, 18 hour incubation

- Manα1–6 \(\text{Man}_1\)
- Manα1–3 \(\text{Man}_2\)
- Manα1–6 \(\text{Man}_3\)

**D. 0.1 nm/µl substrate, 18 hour incubation**

- Manα1–2 \(\text{Man}_1\) Manα1–6 \(\text{Man}_2\)
- Manα1–3 \(\text{Man}_3\)

**E. 0.045 nm/µl substrate, 18 hour incubation**

- Manα1–2 \(\text{Man}_1\) Manα1–3 \(\text{Man}_2\) Manα1–6 \(\text{Man}_3\)

**Figure 1:** Detailed specificity of α1,2-3 Mannosidase. All reactions contained 32 units of α1,2-3 Mannosidase, 1X G6 Reaction Buffer and 1X BSA in a total reaction volume of 10 μl. Reactions were incubated at 37°C. The substrate depicted in (E) will not cut to completion.

**Note:** p-nitrophenyl-α-D-mannopyranoside is NOT a substrate for this enzyme.

**Source:** Cloned from *Xanthomonas manihotis* and expressed in *E. coli* (2).

Supplied in: 50 mM NaCl, 20 mM Tris-HCl (pH 7.5 @ 25°C) and 1 mM Na₂EDTA.

**Reagents Supplied with Enzyme:**

- 1X G6 Reaction Buffer
- 10X BSA

**Reaction Conditions:**

- 1X G6 Reaction Buffer
- 50 mM Sodium Acetate (pH 5.5 @ 25°C), 5 mM CaCl₂, Supplement with 100 μg/ml BSA. Incubate at 37°C.

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

Glycosidase Assays:
32 units of α1-2,3 Mannosidase were incubated with 0.1 mM of fluorescently-labeled oligosaccharides and glycopeptides, in a 10 µl reaction for 20 hours at 37°C. The reaction products were analyzed by TLC for digestion of substrate.

No other glycosidase activities were detected (ND) with the following substrates:
- β-N-Acetylglucosaminidase:
  GlcNAcb1-4GlcNAcb1-4GlcNAc-AMC ND
- α-N-Acetylgalactosaminidase:
  GalNacα1-3(Fucα1-2)Galβ1-4Glc-AMC ND
- α-Fucosidase:
  Fucα1-2Galβ1-4Glc-AMC
  Galβ1-4(Fucα1-3)GlcNAcβ1-3Galβ1-4Glc-AMC ND
- β-Galactosidase:
  Galβ1-3GlcNAcβ1-4Galβ1-4Glc-AMC
  Galβ1-4GlcNAcβ1-2Manα1-6Manβ1-4GlcNAc-AMC ND
- α-Galactosidase:
  Galα1-3Galβ1-4Glc-AMC ND
- α-Mannosidase:
  Manα1-6Manα1-6(Manα1-3)Man-AMC ND
- α-Neuraminidase:
  Neu5Acα2-3Galβ1-3GlcNAcβ1-3Galβ1-4Glc-AMC ND
- β-Glucosidase:
  Glcβ1-4Glcβ1-4Glc-AMC ND
- α-Glucosidase:
  Glcα1-6Glcα1-4Glc-AMC ND
- β-Xylosidase:
  Xylβ1-4Xylβ1-4Xylβ1-4Xyl-AMC ND
- β-Mannosidase:
  Manβ1-4Manβ1-4Man-AMC ND
- Endo F1, F2, H:
  Dansylated fibrinogen biantennary. ND
- Endo F1, F2:
  Dansylated invertase high mannose. ND
- PNGase F:
  Fluoresceinated fetuin triantennary. ND
- Protease F:
  After incubation of 220 units of α1-2,3 Mannosidase with 0.2 nmol of a standard mixture of proteins for 20 hours at 37°C, no proteolytic activity could be detected by SDS-PAGE. 

References:

U.S. Patent No. 7,094,563