Selectin Biosciences Inc.

β(1-3, 4, 6) Galactosidase

Source: Bovine Testes

Product Code: GE 13

Specifications

Activity: 10.2 U/mg, 2.5 U/mL

Storage: Store at -20°C.

Formulation: The enzyme is provided as a sterile-filtered solution in 20 mM Tris-HCl, 50 mM NaCl, pH 7.5, 0.5 mg/ml BSA.

Stability: Stable at least 12 months when stored frozen at -20°C

Product Description

Specificity: Cleaves all β(1-3) and β(1-4) linked non-reducing, terminal galactose. β(1-6) linked galactose is released at a slower rate. The enzyme is a glycoprotein.

Purity: Each lot of β(1-3, 4, 6) Galactosidase is tested for contaminating protease as follows: 10 µg of denatured BSA is incubated for 24 hours at 37°C with 2 µL of enzyme. SDS-PAGE analysis of the treated BSA shows no evidence of degradation.

Each lot is also tested for contaminating activities by incubating the enzymes with the appropriate substrates for 24 hours; the detection limit is 5 µU/mL (IUB). A passing lot will have no detectable activity.

Assay

One unit of β(1-3, 4, 6) Galactosidase is defined as the amount of enzyme required to produce 1 µmole of p-nitrophenol (pNP) in 1 minute at 37°C, pH 4.0 from p-nitrophenyl-β-D-galactopyranoside.

Contaminant	Substrate
β-N-acetylglucosaminidase	p-nitrophenyl-β-D-N-acetylglucosaminide
α-Galactosidase	p-nitrophenyl-α-D - galactopyranoside
Protease	Denatured BSA

Reagents

- 5X Reaction buffer - 500 mM sodium citrate/phosphate pH 4.0

The supplied buffer concentrate provides the optimal pH for enzyme activity with the standard substrate. If glycosidase treatment is performed at suboptimal pH because of glycoprotein solubility or activity requirements, expect some diminution in enzyme activity.

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Suggestions for Use

Prodecure for De-galactosylation

1. Add up to 100 µg of asialoglycoprotein or 1 nmol of oligosaccharide to tube.
2. Add water to a total of 14 µL
3. Add 4 µL 5X Reaction Buffer 4.
4. Add 2 µL β(1-3, 4, 6) Galactosidase.
5. Incubate at 37°C for 1 hour.

For glycoproteins, cleavage may be monitored by SDS-PAGE if the size differential between native and de-galactosylated protein is sufficient for detection.

References

1. Guile GR, Rudd PM, Wing DR, Prime SB, Dwek RA. A rapid high resolution high-performance liquid chromatographic method for separating glycan mixtures and analyzing oligosaccharide profiles. **Anal Biochem. 1996; 240(2):**210-26.
2. Jacob GS, Scudder P. Glycosidases in structural analysis. **Methods Enzymol. 1994;230:**280-99.
3. Distler JJ, Jourdian GW. The purification and properties of beta-galactosidase from bovine testes. **J Biol Chem. 1973; 248(19):**6772-80.

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