GENOVIS

GalactEXO™

Immobilized

STORE AT
+4-8°C

FOR RESEARCH USE ONLY

Instructions for Use

GalactEXO™ Immobilized
Microspin 5 × 0.5 mg (G1-GM6-025)
Process 5 × 0.5 mg glycoprotein

GalactEXO™ Immobilized
Microspin 10 × 0.5 mg (G1-GM6-050)
Process 10 × 0.5 mg glycoprotein

DOWNLOAD INSTRUCTIONS FOR USE
www.genovis.com/ifu-G1-GM6

Immobilized Enzyme for Hydrolysis of β1-3,4-linked Galactose in Spin Columns

GalactEXO is a β-Galactosidase mix that hydrolyses β1-3,4-linked terminal galactose in N- and O-glycosylated proteins. The GalactEXO Immobilized spin columns contain two galactosidases covalently coupled to agarose beads, for efficient hydrolysis of galactoses without contaminating the final preparation with enzyme. GalactEXO hydrolyzes galactose on glycoproteins under native conditions and display activity in pH 5.5-7.5.

The enzymes in GalactEXO are derived from *Akkermansia muciniphila* and expressed in *E. coli*. Both enzymes contain a His-tag and the molecular weights of the enzymes are 87 kDa and 109 kDa.

CONTENT AND STORAGE

The GalactEXO Immobilized columns contain sufficient material to process 0.5 mg glycoprotein per column. The resin is supplied in 20% ethanol with no preservatives added.

GalactEXO Immobilized is shipped cold and should be stored at +4-8°C upon arrival. **Do not freeze the product!**

GalactEXO Immobilized is for R&D use only.

QUALITY CONTROL

GalactEXO Immobilized is tested to meet the specifications and lot-to-lot consistency.

GalactEXO Immobilized is tested for absence of microbial contamination with blood agar plates, Sabouraud dextrose agar plates and fluid thioglycollate medium.

YOU MIGHT ALSO BE INTERESTED IN

GalactEXO™ Lyophilized
Lyophilized enzyme mix for hydrolysis of β1-3,4-linked galactose

SialEXO™
Hydrolysis of sicalic acids

GalNAcEXO™
Hydrolysis of α-linked GalNAcs

FucosEXO™
Hydrolysis of α1-2,3,4 fucose

Preparations

Important Information

- Use lids and bottom caps during the incubation.
- Before centrifugation, remove the bottom cap and loosen the lid (do not remove the lid).

Additional Materials Required

- Reaction buffer: 20 mM Tris pH 6.8.[1,2]
- Microcentrifuge tubes (1.5-2 ml)

1. GalactEXO Immobilized displays high activity in buffers with pH 5.5-7.5, and over a wide range of ionic strengths (0-500 mM NaCl). Optimization might be required if a buffer other than the recommended reaction buffer is used.
2. If the glycoprotein sticks to the resin, a buffer with higher salt concentration can be used in the reaction and/or in the wash steps.

Hydrolysis of β1-3,4-linked Galactose in Spin Columns

Sample Preparation
Prepare the glycoprotein in 100-300 µl reaction buffer per column. Use 0.5 mg of glycoprotein per column.

1. Equilibration

1.1 Break off the bottom cap of the column (save the cap) and place the column in a microcentrifuge tube. Loosen the lid.

1.2 Centrifuge at 200 × g for 1 min to remove the storage solution. Discard the flow-through.

1.3 Equilibrate the column by adding 300 µl reaction buffer and centrifuge at 200 × g for 1 min. Discard the flow-through.

1.4 Perform step 1.3 two additional times.

1.5 Insert the bottom cap.

2. Enzymatic Reaction

2.1 Add the glycoprotein in a volume of 100-300 µl reaction buffer. Use 0.5 mg glycoprotein per column.

2.2 Seal the column with the lid.

2.3 Fully suspend the media, mix by inversion and make sure there is a flow in the column.

2.4 Incubate the column with end-over-end mixing at room temperature for 30-60 min[3].

3. Collection of Processed Material

3.1 Remove the bottom cap and place the column in a new microcentrifuge tube. Loosen the lid.

3.2 Centrifuge at 1000 × g for 1 min to recover the processed material.

4. For Maximum Recovery of the Sample

4.1 Insert the bottom cap.

4.2 Add 100 µl reaction buffer.[2]

4.3 Seal the column with the lid and invert it a couple of times.

4.4 Remove the bottom cap and place the column in a new microcentrifuge tube. Loosen the lid.

4.5 Centrifuge at 1000 × g for 1 min to collect the processed material.

4.6 Repeat steps 4.1-4.5.

4.7 Pool the collected fractions, including the sample from step 3.2.

3. Longer incubation times may be required depending on the glycoprotein.

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