GENOVIS

FucosEXO™

Immobilized

STORE AT
+4-8°C

FOR RESEARCH USE ONLY

Instructions for Use

FucosEXO™ Immobilized
Microspin 5 × 0.5 mg (G1-FM6-025)
Process 5 × 0.5 mg glycoprotein

FucosEXO™ Immobilized
Microspin 10 × 0.5 mg (G1-FM6-050)
Process 10 × 0.5 mg glycoprotein

DOWNLOAD INSTRUCTIONS FOR USE
www.genovis.com/ifu-G1-FM6

Immobilized Enzyme for Hydrolysis of α1-2,3,4 Fucose in Spin Columns

FucosEXO is a mix of α-Fucosidases for efficient hydrolysis of α1-2, α1-3 and α1-4-linked fucose residues on native N- and O-glycosylated proteins or free oligosaccharides. The FucosEXO Immobilized spin columns contain two fucosidases covalently coupled to agarose beads, for defucosylation of glycoproteins or free oligosaccharides without contaminating the final preparation with enzyme. FucosEXO hydrolyzes fucose on glycoproteins under native conditions and display a high activity in pH 6.0-8.0.

The enzymes in FucosEXO are derived from *Akkermansia muciniphila* and *Streptococcus oralis*, and expressed in *E. coli*. Both enzymes contain a His-tag and the molecular weights of the enzymes are 87 kDa and 64 kDa.

CONTENT AND STORAGE

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

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

FucosEXO Immobilized is for R&D use only.

QUALITY CONTROL

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

FucosEXO 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

FucosEXO™ Lyophilized
Lyophilized enzyme mix for hydrolysis of α1-2,3,4 fucose

SialEXO®
Hydrolysis of sialic acids

GalactEXO™
Hydrolysis of β1-3,4 galactose

GalNAcEXO™
Hydrolysis of α-linked GalNAcs

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. FucosEXO Immobilized displays high activity in buffers with pH 6.0-8.0, 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 NaCl can be used in the reaction and/or in the wash steps.

Hydrolysis of α1-2,3,4 Fucose in Spin Columns

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

1. Equilibration

1.1 Break off the bottom cap of the FucosEXO Immobilized 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. Max 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 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 collect 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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