SmartEnzymes™

GENOVIS

SialEXO®

Immobilized

STORE AT
+4-8°C

FOR RESEARCH USE ONLY

Instructions for Use

SialEXO® Immobilized Microspin 2×0.5mg (G1-SM6-010)
Process 2×0.5 mg glycoprotein

SialEXO® Immobilized Microspin 5×0.5mg (G1-SM6-025)
Process 5×0.5 mg glycoprotein

SialEXO® Immobilized Microspin 10×0.5mg (G1-SM6-050)
Process 10×0.5 mg glycoprotein

DOWNLOAD INSTRUCTIONS FOR USE
www.genovis.com/ifu-G1-SM6

Immobilized Enzymes for Hydrolysis of Sialic Acids in Spin Columns

The SialEXO products are sialidases for efficient desialylation of N- and O-glycosylated proteins. The SialEXO Immobilized spin columns contain two sialidases covalently coupled to agarose beads, for desialylation of glycoproteins without contaminating the final preparation with enzyme. The enzymes have unique activities, and they work simultaneously. One enzyme has a broad activity for α2-3, α2-6 and α2-8-linked sialic acids, and the other one quickly hydrolyzes sialic acids with α2-3-linkages.

The enzymes in SialEXO Immobilized are derived from *Akkermansia muciniphila* and expressed in *E. coli*. Both enzymes contain a His-tag and the molecular weights of the enzymes are 43 kDa and 66 kDa.

CONTENT AND STORAGE

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

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

SialEXO Immobilized is for R&D use only.

QUALITY CONTROL

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

SialEXO 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

SialEXO® Lyophilized
Lyophilized enzyme mix for hydrolysis of sialic acids

SialEXO® 2-3 Lyophilized
Lyophilized enzyme for hydrolysis of α2-3-linked sialic acids

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

GalactEXO™
Hydrolysis of β1-3,4 galactose

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. SialEXO displays high activity in buffers at pH 6.5-9.0.
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 Sialic Acids 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 SialEXO 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. 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 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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