SmartEnzymes™

GENOVIS

GlySERIAS®

Immobilized

STORE AT
+4-8°C

FOR RESEARCH USE ONLY

Instructions for Use

GlySERIAS® Immobilized
Microspin 5×0.2mg (A0-GS6-010)
Process 5×0.2mg fusion protein

GlySERIAS® Immobilized
Microspin 10×0.2mg (A0-GS6-020)
Process 10×0.2mg fusion protein

DOWNLOAD INSTRUCTIONS FOR USE
www.genovis.com/ifu-A0-GS6

Immobilized Enzyme for Hydrolysis of Flexible Linkers in Spin Columns

GlySERIAS is a unique enzyme that digests flexible glycine-rich fusion protein linkers such as Gly_4Ser and Gly_xSer_y (GS), and polyglycine (G) linkers. The GlySERIAS Immobilized spin columns contain the GlySERIAS enzyme covalently coupled to agarose beads for digestion of flexible linkers in fusion proteins. The repetitive design of the linker will lead to several simultaneous digestion sites and separation of the previously linked components.

GlySERIAS is derived from phage K and expressed in *E. coli*. The enzyme contains a His-tag and has a molecular weight of 18 kDa.

CONTENT AND STORAGE

The GlySERIAS Immobilized columns contain sufficient material to digest 0.2 mg fusion protein. The resin is supplied in 20% ethanol with no preservatives added.

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

GlySERIAS Immobilized is for R&D use only.

QUALITY CONTROL

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

GlySERIAS 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

FabRICATOR®
Below hinge digestion of IgG

FabALACTICA™
Above hinge digestion of human IgG1

FabDELLO®
Above hinge digestion of human IgG1, including hinge-mutated IgG

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: TBS (50 mM Tris-HCl, 150 mM NaCl) pH 7.6 or PBS[1] (10 mM sodium phosphate, 150 mM NaCl) pH 7.4.
- Microcentrifuge tubes (1.5-2 ml).

1. PBS including 2.7 mM KCl can also be used.

Hydrolysis of Flexible Linkers in Spin Columns

Sample Preparation
Prepare the fusion protein in 100-300 µl reaction buffer[2]. Recommended amount of fusion protein is 0.2 mg per column.

1. Equilibration

1.1 Break off the bottom cap of the GlySERIAS 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 fusion protein to the column (0.2 mg fusion protein in 100-300 µl reaction buffer).

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[3] for 1 h to overnight[4].

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.

4.3 Seal the column with the lid and make sure the media is fully resuspended.

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 Pool the collected fraction with the sample from step 3.2.

2. Optimization may be required if a reaction buffer other than the recommended is used.
3. Increasing the incubation temperature to 37°C may increase the digestion efficacy for some proteins but may induce artifacts during longer incubation times.
4. A shorter incubation time will allow for a more complete coverage of the linker sequence whereas a longer incubation time will reduce the complexity and result in more homogeneous subunits. The incubation time required for complete linker digestion differs between fusion proteins and linkers. The linker may not be completely removed from the linked proteins.

USA & Canada
Genovis Inc.
10919 Technology Place Suite C, San Diego, CA 92127, USA
Phone: 1-855-782-0084 (toll free)
Fax: 1-858-524-3006

EMEA & Asia
Genovis AB
Box 4, SE-244 21 Kävlinge, Sweden
Phone: +46 46 10 12 30
Fax: +46 46 12 80 20

support@genovis.com
www.genovis.com

GENOVIS