

FragIT™ Z

FOR RESEARCH
USE ONLY
www.genovis.com

STORE AT
+4-8°C



SmartEnzymes™



GENOVIS

INSTRUCTIONS FOR PRODUCTS

FragIT™ Z Microspin 2 columns (A0-FZ6-010)

Digestion of up to 2 × 0.5 mg IgG

FragIT™ Z Microspin 5 columns (A0-FZ6-025)

Digestion of up to 5 × 0.5 mg IgG

FragIT™ Z Microspin 10 columns (A0-FZ6-050)

Digestion of up to 10 × 0.5 mg IgG

Quick Guide

- The Quick Guide (p. 3) is intended for experienced users. First time users are recommended to follow the detailed protocol (p. 5).
- Use lids and bottom caps during the incubation.
- Before centrifugation, remove the bottom cap and slightly open the lid.

Sample Preparation

- Prepare the antibody in 100-300 µl digestion buffer. Max 0.5 mg IgG per column.

Digestion – FragIT™ Z Microspin

1 Equilibration

Equilibrate the column with 3 x 300 μ l digestion buffer. Centrifuge at 200 x g for 1 min.



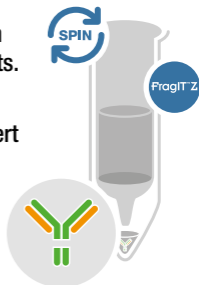
2 Digestion

- Add the antibody to the FragIT Z column and cap the column.
- Incubate at room temperature with end-over-end mixing for 60 min.



3 Collection

- Centrifuge at 1000 x g for 1 min to collect the antibody fragments.
- For maximum recovery, add 100 μ l digestion buffer, invert and centrifuge at 1000 x g for 1 min.
- Repeat once.



PRODUCT DESCRIPTION

FragIT Z is a resin with FabRICATOR® Z enzyme covalently coupled to agarose beads for subunit generation of mouse IgG2a and IgG3. Pure F(ab')₂ and Fc fragments are generated without the enzyme in the final preparation.

The IgG is incubated with the FragIT Z resin and fragments are then easily collected by a centrifugation step.

FragIT Z digests IgG at a specific site below the hinge region, and there is no risk of overdigestion if the incubation time is prolonged. The recommended buffer for FragIT Z is 10 mM sodium phosphate pH 6.5 with 50 mM NaCl¹. The protocol may need optimization for individual antibodies.

Content and Storage

FragIT Z Microspin columns contain sufficient material to digest 0.5 mg mouse IgG2a or IgG3. The resin is supplied in 20% EtOH with no preservatives added.

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

FragIT Z Microspin is for R&D use only.

- 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

- Digestion buffer¹: 10 mM sodium phosphate, 50 mM NaCl, pH 6.5.
- Collection tubes: Microcentrifuge tubes (1.5-2 ml).

Sample Preparation

- Prepare the antibody in 100-300 μ l digestion buffer² per column. Max amount of IgG is 0.5 mg per column.

Digestion of Mouse IgG2a and IgG3 on FragIT™ Column

1 Equilibration

- Break off the bottom cap of the column (save the cap) and place the column in a collection tube. Loosen the lid.
- Centrifuge at $200 \times g$ for 1 min to remove the storage solution.
- Equilibrate the column by adding 300 μ l digestion buffer and centrifuge at $200 \times g$ for 1 min.
- Repeat the equilibration step two times.
- Seal the spin column with the bottom cap.

2 Digestion

- Add the antibody to be digested in a volume of 100-300 μ l digestion buffer^{1,2}. Max 0.5 mg IgG per column.
- Seal the column with the top lid.
- Fully suspend the media, mix it by inversion and make sure there is a flow in the column.
- Incubate the column with end-over-end mixing at room temperature for 60 min³.

3 Collection of Fragments

- Remove the bottom cap and place the column in a collection tube. Loosen the top lid.
- Centrifuge the column at $1000 \times g$ for 1 min to recover the fragments.

For Maximum Recovery of the Sample:

- Seal the spin column with the bottom cap.
- Add 100 μl digestion buffer.
- Seal the column and invert the column a couple of times.
- Remove the bottom cap and place the column in a collection tube. Loosen the top lid.
- Centrifuge the column at $1000 \times g$ for 1 min to recover the fragments.
- Repeat once.
- Pool the collected fractions.

Notes

1. A digestion buffer with 50-150mM NaCl at pH 6.5-7.5 can be used, but the digestion time needs to be increased (2-24 h) with increasing NaCl concentration and pH.
2. The volume should be at least 100 μl / column, but it can be increased to up to 300 μl / column. Max 0.5 mg IgG per column.
3. Increasing the temperature to up to 37°C will increase the digestion efficiency. The incubation time can be increased without overdigestion of the IgG.

Quality Control

FragIT Z is tested to to meet specifications and lot-to-lot consistency.

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

Related Products

FragIT™ Z kit

Digestion of mouse IgG2a and IgG3, and purification of F(ab')₂ and Fc fragments

FragIT™

Immobilized FabRICATOR®, digestion of IgG

FragIT™ kit

Digestion of IgG and and purification of F(ab')₂ and Fc fragments

Immobilized FabALACTICA®

Generation of Fab fragments from human IgG1

FabALACTICA® Fab kit

Generation and purification of intact Fab fragments from human IgG1

FragIT™ Z

Legal and Disclaimers

All rights reserved. Genovis products may be covered by one or more patents, trademarks and copyrights owned or controlled by Genovis AB.

For more information about commercial rights, please contact the Genovis team at info@genovis.com.

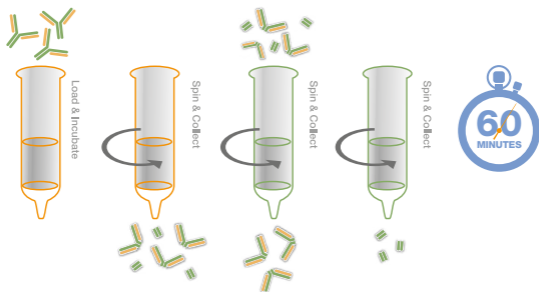
Genovis products are intended for research use only. They are not intended to be used for therapeutic or diagnostic purposes in humans or animals.

FragIT™ kit

Digestion of IgG and Purification of F(ab')₂ and Fc Fragments

FragIT kit consists of an IgG digestion column, FragIT, and an affinity purification column, CaptureSelect™*. FragIT is a resin with FabRICATOR® enzyme covalently coupled to agarose beads for digestion of IgG to generate F(ab')₂ and Fc fragments. After digestion, the fragments can easily be purified using the CaptureSelect™* column supplied in the kit.

- Digestion of IgG on a column.
- Purification of F(ab')₂ and Fc fragments.



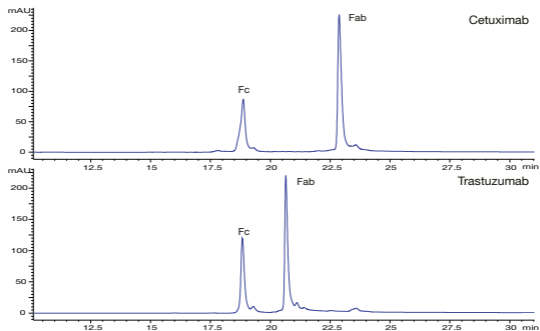
**Made with Thermo Scientific™ CaptureSelect™ resin from Thermo Fisher Scientific Inc. and its subsidiaries. Thermo Scientific and CaptureSelect are trademarks of Thermo Fisher Scientific Inc. and its subsidiaries.*

FabALACTICA®

Generation of hIgG1 Fab Fragments – Digestion above the Hinge

FabALACTICA is a cysteine protease that digests human IgG1 at a specific site in the upper hinge region, generating intact Fab and Fc fragments.

- Specific – one precise digestion site in the upper hinge of human IgG1.
- No need for reducing agents or co-factors.
- Easy to use, with no risk of overdigestion.





USA & Canada

Genovis Inc.
245 First Street, Suite 1800
Cambridge, MA 02142
USA

Customer service: 1-617-444-8421
Order phone (toll free): 1-855-782-0084
Order fax: 1-858-524-3006
Email: orders.us@genovis.com

EMEA & Asia

Genovis AB
Box 790
SE-220 07 Lund
Sweden

Customer service: +46 46 10 12 30
Order phone: +46 46 10 12 30
Order fax: +46 46 12 80 20
Email: order@genovis.com