# **FabDELLO**™

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**Smart**Enzymes<sup>™</sup>



# **FabDELLO**™

#### INSTRUCTIONS FOR PRODUCTS

FabDELLO 8×100 units (B1-BD1-008) Digestion of up to 8×100 µg human IgG1 (8 well strip)

FabDELLO 96×100 units (B1-BD1-096)
Digestion of up to 96×100 ug human lgG1 (96 well plate)

## **Quick Guide**

- The Quick Guide (p. 3) is intended for experienced users. First time users are recommended to follow the detailed protocol (p. 6).
- Cover the enzyme vial with adhesive plastic or move the reaction to a new vial during the incubation.

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# WORKFLOW

#### **Quick Guide**

- 1 Add IgG
  - Add 100 µg lgG to the enzyme vial.



- 2 Add Buffer and CaCl<sub>2</sub>
  - Add TBS buffer to a total volume of 95 µl.
  - Add 5 µl 20× CaCl<sub>2</sub> solution (200 mM).



- 3 Digestion
  - Incubate for 2h at 37°C.



# PRODUCT DESCRIPTION

FabDELLO is an enzyme that digests human IgG1, yielding intact Fab and Fc fragments. Under native conditions, FabDELLO digests human IgG1 at one single site in the upper hinge (..KSCDK / THTCPPCP..). A second digestion site on the Fc may appear if the N-glycans are removed. FabDELLO requires the presence of calcium ions to be active. Optimal activity is obtained at 37°C and pH 7-8.5.

FabDELLO is cloned from *Bdellovibrio bacteriovorus* and expressed in *E. coli*. The enzyme contains a His-tag and the molecular weight is 32 kDa.

#### **Unit Definition**

One unit FabDELLO digests  $\geq$  90% of 1  $\mu$ g human lgG1, when incubated in Tris buffered saline (TBS), pH 7.6, containing 10 mM CaCl<sub>2</sub>, at 37°C for 2 h.

# FabDELLO"

## **Content and Storage**

FabDELLO is supplied lyophilized in 50 mM Tris-HCl, 150 mM NaCl, pH 7.6, with no preservatives added. The enzyme is supplied together with 1 vial 20× CaCl<sub>2</sub> solution (200 mM). FabDELLO is shipped cold and the FabDELLO enzyme should be stored at -20°C upon arrival. The CaCl<sub>2</sub> solution should be stored at +4-8°C upon arrival. FabDELLO has autoproteolytic activity. Therefore, the enzyme should be kept cold and used within the day of reconstitution

#### **Additional Materials Required**

 Digestion buffer: TBS (50 mM Tris-HCl, 150 mM NaCl), pH 7.6.

# DETAILED PROTOCOL

#### Sample Preparation

Prepare the human IgG1 in the digestion buffer. The final IgG concentration in the digestion reaction should be 0.5-5 mg/ml.

# 1 Add IgG

- · Pierce the aluminium foil with a pipet tip.
- Add 100 µg IgG to the enzyme vial<sup>1</sup>.

## 2 Add Buffer

Add TBS buffer to a total volume of 95 µl<sup>2,3</sup>.

# 3 Add CaCl<sub>2</sub>

Add 5 µl 20× CaCl<sub>2</sub> solution (200 mM)<sup>4</sup>.

# 4 Digestion

- Mix the solution by aspirating and dispensing the liquid a few times.
- Cover the vial with adhesive plastic or move the content to another vial.
- Incubate for 2h at 37°C<sup>5</sup>.

# FabDELLO"

# **Quality Control**

FabDELLO is tested to meet the specifications and lot-to-lot consistency.

#### **Related Products**

#### FabALACTICA®

Above hinge digestion of human IgG1

#### Notes

- The enzyme may also be dissolved in ultrapure water and added to a digestion in another vial if digestion of smaller amounts of IgG is desired.
- Optimal activity is achieved in TBS buffer pH 7-8.5. Buffers containing phosphate should be avoided since the phosphate ions will form the insoluble calcium phosphate salt together with the calcium ions required for enzymatic activity.
- 3. Another buffer volume may be added. The total volume in the enzyme vial should be 20-200 ul.
- The final CaCl<sub>2</sub> concentration should be 10 mM. If the reaction volume is adjusted, the volume of the CaCl<sub>2</sub> solution must also be adjusted.
- 5. The digestion time may need to be optimized for individual antibodies.

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