

FabRICATOR®

Instructions for product no
A0-FR1-020 2000 units
A0-FR1-050 5000 units

Product Description

FabRICATOR® is an enzyme used for preparation of F(ab')₂. FabRICATOR® is a digestive enzyme that cleaves IgG only at one specific site below the hinge region resulting in F(ab')₂ and Fc-fragments.

FabRICATOR® cleaves all subclasses of human, monkey, rabbit and sheep IgG but only subclass IgG3 of mouse IgG.

Since FabRICATOR® only cleaves at one specific site below the hinge region, there is no risk of getting other fragments than Fc and F(ab')₂ if the incubation time is increased for several hours.

Content and storage

FabRICATOR® is supplied as lyophilized powder from 50 mM sodium phosphate, 150 mM NaCl, pH 6.6, with no preservatives added.

Contains sufficient material to produce F(ab')₂ fragments from up to 2 mg IgG (A0-FR1-020) or up to 5mg IgG (A0-FR1-050).

FabRICATOR® is shipped on ice. It should be stored at -20 °C upon arrival.

After reconstitution FabRICATOR® is stable for 1 month at 2-8 °C.

FabRICATOR® is for R&D use only.

Unit Definition

One unit cleaves ≥ 95% of 1µg human IgG when incubated in 50mM sodium phosphate pH 6.6, 150mM NaCl at 37 °C for 30min.

Quality Control

FabRICATOR® is tested to ensure lot-to-lot consistency.

FabRICATOR® is tested for absence of microbial contamination with blood agar plates, Sabaraud dextrose agar plates and fluid thioglycolate medium.

Additional Materials Required

- ✓ 50mM sodium phosphate, 150mM NaCl, pH 6.6.

Method

Reconstitute **A0-FR1-020** in **30 µl** double distilled H₂O and **A0-FR1-050** in **75µl** double distilled H₂O. This gives a concentration of 67 units/µL. To prevent microbial contamination, sodium azide can be added to the solution to a final concentration of 0.02 - 0.05% (w/v).

1mg human IgG is digested by adding 1000 units (15µL) FabRICATOR® and incubating for 30 minutes in 50mM sodium phosphate pH 6.6, 150mM NaCl at 37 °C.

FabRICATOR® contains a His-tag and can therefore easily be removed after digestion. The Fc fragments can be removed with protein A or protein G.

Best activity is obtained at pH 6.6 and 37 °C. It is possible to use a buffer with a higher pH and increasing the reaction time. Digestion can also be done at room temperature with prolonged incubation time.

Product References

Mary H. Ryana, Diane Petrone, Jennifer F. Nemetha, Evan Barnathan, Lars Björck, Robert E. Jordan: *Proteolysis of purified IgGs by human and bacterial enzymes in vitro and the detection of specific proteolytic fragments of endogenous IgG in rheumatoid synovial fluid*, Molecular Immunology, October 2007.

Application note

FabRICATOR - perfect F(ab')₂ fragments in minutes