



AlligBAITOR™

Lyophilized

STORE AT

-20°C



FOR RESEARCH USE ONLY

Instructions for Use

AlligBAITOR™ Lyophilized 2000 units (A4-AB1-020)

Binds 2 mg Ig



Lyophilized Protein for All Ig Capture and Antigen Release

AlligBAITOR is an immunoglobulin-binding protein that binds all human immunoglobulin isotypes and subclasses. Upon binding, AlligBAITOR Lyophilized enables native antigen release from immune complexes.

AlligBAITOR is active in a wide range of physiological relevant buffer conditions, including PBS and TBS, in a salt concentration up to 500 mM and in the pH range 5.5–8.5.

UNIT DEFINITION

One unit AlligBAITOR Lyophilized captures 1 µg Ig and releases any bound antigen when incubated in PBS buffer at 37°C for 30 minutes.

CONTENT AND STORAGE

AlligBAITOR Lyophilized is supplied lyophilized in 50 mM Tris-HCl, 150 mM NaCl, pH 7.6, with no preservatives added.

AlligBAITOR Lyophilized is shipped cold and should be stored at -20°C upon arrival. After reconstitution, AlligBAITOR Lyophilized is stable for at least 1 month at +4-8°C.

AlligBAITOR Lyophilized is for R&D use only.

QUALITY CONTROL

AlligBAITOR Lyophilized is tested to meet the specifications and lot-to-lot consistency.

AlligBAITOR Lyophilized 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 (IdeS)

IgMBRAZOR™

Digestion of IgM

IgASAP™

Digestion of IgA

Preparations

Additional Materials Required

- Antibody Fc-binding affinity resin on a microspin column¹.
- Binding buffer: PBS² (10–150 mM sodium phosphate, 150 mM NaCl, pH 7.4) or TBS (10–50 mM, 150 mM NaCl).
- Microcentrifuge tubes: 1.5–2 ml.

1. Only required for the ‘Release of Antigens from Antibody-Antigen Complexes’ workflow, which was developed and verified using a CaptureSelect™ Fc column. Equivalent antibody Fc-binding affinity columns may be used.
2. Optimal binding may also be obtained in Bis-Tris (50 mM), and Tris (50 mM) buffers at pH 6.5–8.5. NaCl up to 500 mM can be added without affecting the performance of the binding capacity. Optimization of buffer conditions may be required depending on the sample.

Pan-Immunoglobulin Binding

Sample Preparation

Determine the antibody concentration by titration or quantitative assay. Prepare the antibody solution in binding buffer. The final concentration of antibody should be 0.2–5 mg/mL.

A1. Prepare AlligBAITOR

A1.1 Reconstitute AlligBAITOR in 100 µl ddH₂O to a concentration of 20 units/µl.

A2. Add AlligBAITOR

A2.1 Add 1 unit AlligBAITOR / 1 µg antibody.

A3. Immunoglobulin Binding

A3.1 Incubate for 30 min at 37°C.

Note: Optimization of protein concentration and incubation time may be needed depending on the substrate.

Release of Antigens from Antibody-Antigen Complexes

B1. Capture Antibody Antigen Complex using Antibody Fc Binding Column

- B1.1 Determine antibody concentration. Equilibrate Fc binding column, bind antibody-antigen complexes and wash resin according to manufacturers instructions.

B2. Prepare AlligBAITOR

- B2.1 Reconstitute AlligBAITOR in 100 µl ddH₂O to a concentration of 20 units/µl.

B3. Releasing Antigens

- B3.1 Add 1 unit AlligBAITOR / 1 µg antibody to the prepared column.
- B3.2 Seal the column with the lid.
- B3.3 Fully suspend the media, mix it by inversion and make sure there is a flow in the column.
- B3.4 Incubate the column with end-over-end mixing for 1h at 37°C.
- B3.5 Remove the bottom cap and place the column in a new microcentrifuge tube. Loosen the lid.
- B3.6 Centrifuge at 1000×g for 1 min to collect the flow-through containing the released antigens.
- B3.7 Insert the bottom cap.

B4. Elution of Ig- and AlligBAITOR Fraction (Optional)

- B4.1 Add 100 µl elution buffer to the column.
- B4.2 Seal the column with the lid and mix by tapping the tube and incubate for 2 min at RT.
- B4.3 Remove the bottom cap and place the column in a new microcentrifuge tube. Loosen the lid.
- B4.4 Centrifuge at 1000×g for 1 min to collect the eluted bound material.

Note: The eluted material contains the Igs and AlligBAITOR as a strong affinity complex, meaning the antibody is not in a functional state.

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