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ab241023
Albumin Depletion Kit

View Albumin Depletion Kit datasheet:
www.abcam.com/ab241023

For the removal of albumin from plasma, serum and other biological samples and for the purification of albumin and Cibacron Blue 3G-A-binding proteins.

This product is for research use only and is not intended for diagnostic use.

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1. Overview

Albumin Depletion Kit (ab241023) provides easy and user-friendly spin columns for the depletion/removal of abundant albumin protein from samples thus enhancing the sample analysis. The kit is based on the interaction of Cibacron Blue 3G-A (immobilized on beads) with albumin. Each spin column can bind ~2-3 mg of albumin and processes samples in less than one hour. After albumin depletion, samples can be used for subsequent ELISA, immunoassays and other downstream analyses such as gel electrophoresis and functional assays. These columns can also be used for purification and enrichment of a variety of Cibacron Blue dye-binding proteins.

2. Protocol Summary

Remove storage buffer from Cibacron Blue Column by centrifugation at 1,500 x *g* for 1 minute at room temperature.

Equilibrate Cibacron Blue Column in Albumin Binding Buffer (3 x 0.2 mL) by centrifugation.

Apply sample to Cibacron Blue Column and incubate on a rotary shaker at room temperature for 30 minutes.

Centrifuge Cibacron Blue Column and collect albumin-depleted sample in the flow-through. Optionally, wash column (by centrifugation) with Albumin Binding Buffer (3 x 0.2 mL) to increase the yield of albumin-depleted sample.

Elute albumin (and other bound proteins) by centrifugation using Cibacron Elution Buffer (5 x 0.2 mL).

3. Materials Supplied and Storage

Store kit at 4°C in the dark immediately on receipt and check below for storage for individual components.

Item	Quantity	Storage temperature (before prep)
Albumin Binding Buffer	33 mL	4°C
Cibacron Blue Column	25 vials	4°C
Cibacron Elution Buffer	25 mL	4°C

4. Materials Required, Not Supplied

These materials are not included in the kit, but will be required to successfully perform this assay:

- Microcentrifuge
- Microcentrifuge tubes

5. General guidelines, precautions, and troubleshooting

Please observe safe laboratory practice and consult the safety datasheet.

For general guidelines, precautions, limitations on the use of our assay kits and general assay troubleshooting tips, particularly for first time users, please consult our guide:
www.abcam.com/assaykitguidelines

For typical data produced using the assay, please see the assay kit datasheet on our website.

6. Reagent Preparation

6.1 Albumin Binding Buffer

Ready to use as supplied. Bring to room temperature before use.

6.2 Cibacron Blue Column

Supplied as a 50% bead slurry. Bring to room temperature before use.

6.3 Cibacron Elution Buffer

Ready to use as supplied. Bring to room temperature before use.

7. Assay Procedure

– Equilibrate prepared reagents to room temperature just prior to use.

7.1 Cibacron Blue Column preparation and equilibration:

1. Snap off the bottom closure and slightly loosen the cap. Place the column in a 1.5 or 2.0 mL microcentrifuge tube and centrifuge at 1,500 x *g* for 1 minute to remove the storage buffer. Discard the storage buffer.
2. Wash the column 3x by adding 0.2 mL of Albumin Binding Buffer. Centrifuge for 1,500 x *g* for 1 minute each time and discard the flow through.

7.2 Sample application:

1. Apply sample to Cibacron Blue Column. Close the cap tightly and invert the column a few times.
2. Incubate the column for 30 minutes on a rotary shaker at room temperature.

ΔNote: Dilute (if necessary) plasma or serum sample 10 times by adding 10 µL of sample into 90 µL of Albumin Binding Buffer (maximum sample protein concentration 5-7 mg/mL, maximum sample loading volume 100 µL).
ΔNote: Cibacron Blue Column can also be used to purify Cibacron Blue 3G-A binding proteins.

3. Place the column in a new microcentrifuge tube and centrifuge at 1,500 x *g* for 1 minute. Retain flow-through. <u>This is the albumin-depleted sample.</u>

ΔNote: Make sure that there is no residual solution at the bottom of column before putting the column in new microcentrifuge tube (Step 7.2.3, above).

Optional:

- To recover more albumin-depleted sample, wash the column 3x with 0.2 mL of Albumin Binding Buffer using new

microcentrifuge tube each time. Combine the wash fractions with the flow-through and concentrate.

- Elute the bound albumin from the column with 0.2 mL of Cibacron Elution Buffer. Repeat the elution 4x using a new microcentrifuge tube. Combine all the elution fractions, concentrate if necessary and dialyze against an appropriate storage buffer.
- If purifying Cibacron Blue 3G-A binding proteins, further optimization of the number of washes or elutions may be necessary depending upon the type of protein being purified.

8. Data Analysis

Measure the OD280 of albumin-depleted plasma or serum and eluted albumin samples. Use these samples for downstream 2D gel electrophoresis or any other downstream application.

9. Typical Data

Data provided **for demonstration purposes only**.

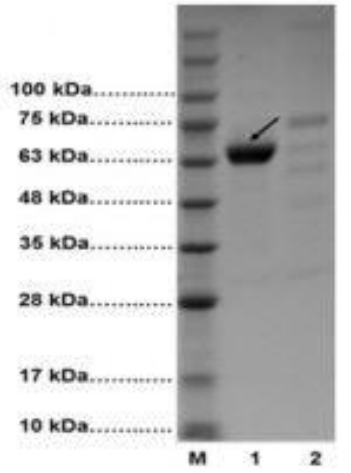

Figure 1. Depletion of albumin from plasma using Albumin Depletion Kit (ab241023). Samples were analyzed by 15% SDS-PAGE. M: Protein Marker, 1: Eluted albumin fraction (20 µL of concentrated eluted albumin, primarily containing albumin), 2: Albumin-depleted plasma sample (20 µL of combined and concentrated flow-through and wash fractions).

10. Notes

Technical Support

For all technical or commercial enquiries please go to:
https://www.abcam.com/en-us/contact-us
https://www.abcam.cn/contact-us (China)
https://www.abcam.co.jp/contact-us (Japan)