

EZAlbumin™ Depletion Kit (Spin Column)

10/14

(Store at 4°C)

Cat. No.
K6573-25 EZAlbumin™ Depletion Kit (Spin Column), contains 25 columns and sufficient Binding and Elution Buffer for 25 columns

I. Introduction:

BioVision's Albumin Depletion Kit provides easy and user-friendly spin columns for 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 sample 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.

II. Applications:

- Removal of albumin from plasma, serum, antibody or other samples
- Purification of Cibacron Blue 3G-A binding proteins
- Purification of albumin

III. Kit Contents:

Components	K6573-25	Cap Code	Part Number
EZAlbumin Binding Buffer	33 ml	Clear	K6573-25-1
EZAlbumin Binding Spin Column	25 columns	-	K6573-25-2
EZAlbumin Elution Buffer	25 ml	Brown	K6573-25-3

IV. User Supplied Reagents and Equipment:

- Microcentrifuge and microcentrifuge tubes

V. Storage Conditions and Reagent Preparation:

Store kit at 4°C, protected from light. Read the entire protocol before performing the assay.

- **EZAlbumin Binding Buffer and EZAlbumin Elution Buffer:** Bring to room temperature before use.
- **EZAlbumin Binding Spin Column:** Supplied as 50% beads slurry. Bring to room temperature before use.

VI. Albumin Depletion Protocol:

1. EZAlbumin Binding Spin Column Preparation and Equilibration: 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 min. to remove the storage buffer. Discard the storage buffer. Wash the column 3x by adding 0.2 ml of EZAlbumin Binding Buffer. Centrifuge for 1,500 x g for 1 min. and discard the flow through.

2. Sample Application:

- a. Apply sample to spin column. Close the cap tightly and invert the column a few times. Incubate the column for 30 min. on a rotary shaker at room temperature.

Notes:

- Dilute (if necessary) plasma or serum sample 10 times by adding 10 µl of sample into 90 µl of EZAlbumin Binding Buffer (maximum sample protein concentration 5-7 mg/ml, maximum sample loading volume 100 µl).
- EZAlbumin Binding Spin Column can also be used to purify Cibacron Blue 3G-A binding proteins. For large-scale albumin depletion/purification, Hi-Bind™ Cibacron Blue-Agarose Beads (Cat # 7923) are also available from BioVision.

- b. Place the column in a new microcentrifuge tube and centrifuge at 1,500 x g for 1 min. Retain flow-through. This is albumin-depleted sample.

Notes:

- a) Make sure that there is no residual solution at the bottom of column before putting the column in new microcentrifuge tube.

b) Optional:

- To recover more albumin-depleted sample, wash the column 3x with 0.2 ml of EZAlbumin Binding Buffer using new microcentrifuge tube each time. Combine wash fractions with flow-through and concentrate.
- Elute the bound albumin from the column with 0.2 ml of EZAlbumin Elution buffer. Repeat the elution 4x using new microcentrifuge tube. Combine all the elution fractions, concentrate if necessary and dialyze against 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.

3. 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.

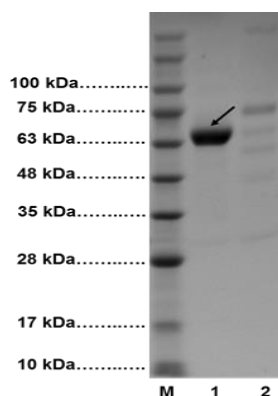


Figure: Depletion of albumin from plasma using EZAlbumin™ Depletion Kit (Spin Column). Samples were analyzed by 15% SDS-PAGE. M: Protein Marker, 1: Eluted albumin fraction (20 µl of concentrated eluted albumin, primarily containing albumin), 2: Albumin-depleted plasma sample (20 µl of combined and concentrated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

VII. RELATED PRODUCTS:

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| Hi-Bind Ni QR Agarose Beads (6562) | Hi-Bind™ Cibacron Blue-Agarose Beads (7923) |
| Ready-to-Use Ni QR Agarose Beads Buffer Kit (K6563) | Ni-IDA Spin Columns (6567) |
| Ready-to-Use Ni-IDA Spin Purification Kit (K6567) | Albumin, Human Plasma (7546) |
| Human Serum Albumin (4016) | BSA (10% in H ₂ O) (2119) |
| AGE-BSA (2221) | Glucose AGE-BSA-II (2223) |
| BSA Control for AGE-BSA (2221-BSA) | Biotinylated Bovine Serum Albumin (12 biotin/BSA) (7099) |
| Biotinylated Bovine Serum Albumin (3 biotin/BSA) (7097) | Biotinylated Bovine Serum Albumin (5 biotin/BSA) (7098) |
| Bovine Serum Albumin-Cohn Factor V, Endotoxin Low, pH-5.2 (7910) | Bovine Serum Albumin-Cohn Factor V, pH-5.2 (7904) |
| Bovine Serum Albumin-Cohn Factor V, Endotoxin Low, pH-7 (7909) | Bovine Serum Albumin-Cohn Factor V, pH-7 (7905) |
| Bovine Serum Albumin-Cohn Factor V, Fatty Acid Free, pH-5.2 (7908) | Bovine Serum Albumin-Factor V, 30% solution (7914) |
| Bovine Serum Albumin-Cohn Factor V, Fatty Acid Free, pH-7 (7907) | Thrombin, Active, Bovine Plasma (7592) |
| Bovine Serum Albumin-Cohn Factor V, Fatty Acid Low, pH-5.2 (7913) | Thrombin, Active, Human Plasma (7593) |
| Bovine Serum Albumin-Cohn Factor V, Fatty Acid Low, pH-5.2 (7912) | Thrombin, Active, Porcine blood (7590) |
| Bovine Serum Albumin-Heat Shock, Diagnostic Grade, pH 7 (7918) | Thrombin, Active, Bovine Plasma (Technical grade) (7591) |
| Bovine Serum Albumin-Heat Shock, Fatty Acid Free, pH 7 (7921) | |
| Bovine Serum Albumin-Heat Shock, Low Endotoxin, pH 7 (7922) | |
| Bovine Serum Albumin-Heat Shock, Protease DNASE Free, pH 7 (7920) | |
| Bovine Serum Albumin-Heat Shock, Protease Free, pH 7 (7919) | |
| Bovine Serum Albumin-Heat Shock, Reagent Grade, pH 7 (7917) | |
| Bovine Serum Albumin-Heat Shock, Standard Grade, pH 7 (7915) | |
| Bovine Serum Albumin-Heat Shock, Standard Grade, pH 5.2 (7916) | |
| Bovine Serum Albumin-Cohn Factor V, Immunoassay Grade, Protease Free pH-5.2 (7911) | |
| Bovine Serum Albumin-Cohn Factor V, Immunoassay Grade, Protease Free pH-7 (7906) | |

FOR RESEARCH USE ONLY! Not to be used on humans.