G-Biosciences ♦ 1-800-628-7730 ♦ 1-314-991-6034 ♦ technical@GBiosciences.com

A Geno Technology, Inc. (USA) brand name

AlbuminOUT™

(Cat. # 786-251, 786-252, 786-251T)



INTRODUCTION 3
ITEM(S) INCLUDED 3
STORAGE CONDITION 3
ADDITIONAL ITEMS REQUIRED 3
PROTOCOL 4
ELUTION OF COLUMN BOND ALBUMIN AND OTHER PROTEINS (OPTIONAL) 5
REMOVAL OF ALBUMIN FROM OTHER SPECIES: 5
RELATED PRODUCTS 5

INTRODUCTION

Samples that contain a large abundance of albumin, such as plasma and cerebrospinal fluid, tend to mask identification and discovery of other less abundant proteins in 2D gel electrophoresis. AlbuminOUT™ has been specifically developed for substantial removal of albumin from such samples. The albumin removal method is based on binding of albumin with Cibachron Blue dye. AlbuminOUT™ has been optimized for removal of human albumin from samples. AlbuminOUT™ is rapid spin column method, each column contains (0.2ml) dye bond resins with capacity > 2mg human albumin per column. AlbuminOUT™ will remove over >98% albumin from 5-50µl human plasma. Other nucleotide binding proteins may also be captured and removed by Cibachron Blue dye resins. AlbuminOUT™ may also be used for removal of albumin from other species. AlbuminOUT™ is suitable for processing 25 (Cat # 786-251) and 50 (Cat# 786-252) samples respectively.

ITEM(S) INCLUDED

Description	Cat. # 786-251	Cat. # 786-252	Cat. # 786-251T
Albumin Binding Buffer	25ml	2x25ml	5ml
Albumin Elution Buffer	6ml	2x 6ml	2ml
AlbuminOUT™ Spin Column	25	50	4
Collection Tubes (2ml)	50	100	10

STORAGE CONDITION

The kit is shipped at ambient temperature. Upon arrival, store it at 4°C. When stored and used as recommended, the kit is good for use for 12 months.

ADDITIONAL ITEMS REQUIRED

- Micro centrifuge
- 1.5ml collection tubes.

PROTOCOL

Please note that albumin content in samples may vary. Typically, each column will bind and remove >98% albumin (~2mg albumin) from 5-50µl human serum. Other factors, such as salt concentration, may influence the albumin retention capacity of the column, therefore, the total human serum load on the column must not exceed 50µl (undiluted human serum) - whenever possible, keep the human serum load under 50µl.

1. Transfer 50µl Albumin Binding Buffer into a 1.5ml tube. Add 5-50µl sample containing albumin (serum or cerebrospinal fluid) into the tube. Mix the content and centrifuge 10,000xg for 5 minutes in a cold centrifuge. Collect the clear supernatant and store in an ice-bucket until used.
2. Spin the AlbuminOUT™ Spin Column for a brief 5 seconds. Break off and open the bottom plug of the column. Reposition the column in a 2ml Collection Tube provided with this kit.
3. Add 100µl Albumin Binding Buffer and spin the AlbuminOUT™ Spin Column at 1000xg for 10-15 seconds. Repeat this step two more times. Empty the collection tube and replace the column in the collection tube.
4. Load the diluted sample into the AlbuminOUT™ Spin Column. Incubate the column for 1-2 minutes at room temperature.
5. Collect any flow-through and re-apply to the AlbuminOUT™ Spin Column. Incubate the column for 1-2 minutes at room temperature.
6. Centrifuge the AlbuminOUT™ Spin Column at 1000xg for 5 seconds. Collect the albumin free flow-through for further processing.

NOTE: *The flow through sample contains a salt concentration may not be suitable for 2D gel analysis. Dialyze the albumin free sample using 1-4Kd molecular weight cut off Tube-O-Dialyzer (Cat. # 786-141-143) against a salt free buffer. Alternatively, clean the sample with Perfect-FOCUS (Cat #786-124) before 2D gel analysis.*

Elution Of Column Bond Albumin And Other Proteins (Optional)

1. Wash the column 3 times with Albumin Binding Buffer, 200µl each wash.
2. Add 200µl Albumin Binding Buffer and spin the column at 1000xg for 5 seconds. Repeat this step twice. Empty the collection tube and replace the column in the collection tube.
3. Add 100-200µl Albumin Elution Buffer. Incubate the column for 5 minutes at room temperature. Centrifuge the spin column at 1000xg for 5 seconds.
4. Collect the flow-through. The flow-through predominantly contains albumin. Contamination with other proteins cannot be ruled-out. Other nucleotide binding proteins may also be captured by Cibachron™ Blue dye resins (AlbuminOUT™ Spin Column) and co-eluted with albumin.

For step elution of the proteins captured by AlbuminOUT™ Spin Column, use sodium chloride concentration between 250mM - 1.5M, at pH 7.2.

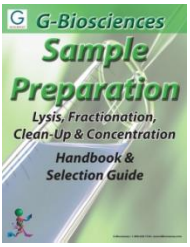
NOTE:*The eluted albumin contains a salt concentration may not be suitable for 2D gel analysis. Dialyze the eluent albumin using 1-4Kd molecular weight cut off Tube-O-Dialyzer (Cat.# 786-141-143) against a salt free buffer. Alternatively, clean the sample with Perfect-FOCUS (Cat#786-124) before 2D gel analysis.*

REMOVAL OF ALBUMIN FROM OTHER SPECIES:

AlbuminOUT™ may be used for removal of albumin from other sources such as pig, dog, sheep, rabbit, rat, and bovine. Binding efficiency of non-human source may vary between 1-2 mg albumin/column; therefore, it is important to perform a test before use. The protocol, as outlined above, is suitable for removal of albumin from any species or sample. In most cases, reducing the undiluted sample load by 30%-50% would be sufficient.

RELATED PRODUCTS

Download our Sample preparation Handbook.



http://info.gbiosciences.com/complete-protein-sample-preparation-handbook

For other related products, visit our website at www.GBiosciences.com or contact us.

This page is intentionally left blank

This page is intentionally left blank



www.GBiosciences.com