Order Information

Product Name	Contents	Code No.
Ab-Capcher ExTra	2 mL	P-003-2
	10 mL	P-003-10
	100 mL	P-003-100
Ab-Rapid SPiN EX (Spin column)	0.1 mL gel/column x 10 (2 mL empty tube x20 included)	P-014-10
Ab-Rapid PuRe EX (For syringe)	2 columns	P-015-2
	10 columns	P-015-10

Related Products

Product Name	Contents	Code No.
Ab-Capcher	2 mL	P-002-2
	10 mL	P-002-10
	100 mL	P-002-100
Ab-Rapid PuRe (For syringe)	2 columns	P-012-2
	10 columns	P-012-10
Ab-Rapid SPiN (Spin column)	0.1 mL gel/column x 10 (2 mL empty tube x 20 included)	P-013-10
	For 50 columns (5 mL bulk gel x1, empty spin column x 50)	P-013-50
Buffer Kit	1 kit (Bind. 200mL, Elut. 30mL, Neutr. 1mL)	P-011

There are cases that prices will be changed without notice.
For research use only.

COSMO BIO CO., LTD.
Inspiration for Life Science
TOYO EKIMAE BLDG. 2-20, TOYO 2CHOME
KOTO-KU, TOKYO 135-0016, JAPAN
TEL : +81-3-5632-9617
FAX : +81-3-5632-9618

ProteNova Co.Ltd. Takamatsu Lab.
2217-44 Hayashi-cho, Takamatsu
Kagawa 761-0301 Japan
TEL +81-87-897-2073 FAX +81-87-816-2073
URL http://protenova.com

ProteNova
INNOVATION BY PROTEIN ENGINEERING

Ab-Capcher ExTra™

Users Manual

Ab-Capcher ExTra Specifications

- Gel matrix : 6% highly crosslinked agarose
- Particle size : 35μm
- Ligand : Alkali-resistant Protein A-derivative
  : (Protein A-R28) (*E.coli*)
- Bind. capacity : >90 mg human IgG /mL gel
- Form : 20% Ethanol (50% gel slurry)
- Storage : 4-8℃

Recommended Reagents

- Buffers
  Binding Buffer : PBS
  Elution Buffer : 0.1 M Glycine-HCl, pH 2.8
  Neutralization Buffer : 1 M Tris

＊Buffer Kit (PN-011) is also available from ProteNova.
Buffer kit contains Binding Buffer, Elution Buffer and Neutralization Buffer.

To increase binding capacity for mouse or rat IgG, 1.5 M Glycine-HCl / 3 M NaCl (pH 9) can be used as Binding Buffer. In some case such as direct purification from the serum, the number of contaminant bands may increase on SDS-PAGE.

Sample preparation (example)

◆ Ascites : 3 x dilution with Binding Buffer.
◆Serum : Ppt. with 50%-saturated $(NH_4)_2SO_4$ or 3-5 x dilution with Binding Buffer
◆Cultured medium : Adjust pH to neutral.

Recommended pre-treatments of sample before applying to the gel.
- Centrifugation ; 10,000 × *g*, 10 min
- Filtration ; 0.45μm filter
  （Please use low-protein-adsorption types）

＊If there are insolubles in the sample, make sure to do pre-treatments.

Preparation for 50% ammonium sulfate precipitation
1. Prepare saturated ammonium sulfate.
   Add equal volume of saturated ammonium sulfate gradually to serum and mix.
2. Stand on ice for more than 1hr.
3. After centrifugation at 4℃, remove the supernatant.
   Wash precipitate with 50%-saturated ammonium sulfate.
4. Resolve the precipitate with small volume of Binding Buffer. The precipitate contains antibody.
5. Exchange to Binding buffer with dialysis or desalting column.

Protocol for IgG Purification

Preparation

Put Ab-Capcher ExTra gel slurry into a column.

Add Neutralization Buffer to an empty microcentrifuge tube.
(1/30 volume of eluate: *e.g.,* 30-35μL to 1 mL of eluate)

Step 1. Equilibration

Equilibrate the column with 5 bed volumes of Binding Buffer.

Step 2. Sample Applying

Apply the sample solution to the column.
(If the bind of the sample seems to be weak, reduce the flow rate.)
Collect the flow through.

Step 3. Column Washing

Wash the column with 5 bed volumes of Binding Buffer.

Step 4. Elution of IgG

Add 4 bed volumes of Elution Buffer to the column.

Discard first 0.5～0.8 bed volume of eluate and collect the following eluate as IgG fraction into a microcentrifuge tube, in which Neutralization Buffer (1/30 volume of eluate) is pre-added, and mix.

Note: ・In some species of antibody, binding to the gel may be weak.
・In some molecular species of Rat IgG2a, binding to the gel may be weak (EX : about 1mg/mL gel)
・In mouse IgM, there are 2 type of molecular species. “High-binding” type can be purified with this protocol, but “low-binding” type is difficult to be purified.

＊Storage and reuse of gel
・Ab-Capcher ExTra is alkali-washable.
・When the gel is reused, wash the column with 3-5 bed volumes of 0.1N NaOH after elution of IgG. If you want to use the gel immediately after washing, equilibrate with 10 bed volumes of Binding Buffer. Then, apply the sample.
・For storage of column, add 5 bed volumes of 20% Ethanol, transfer to a tightly sealed container and store at 4-8℃.