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## ChroPlate - ProteinA/G V.1

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## Abstract

### **Purification Guide for the Isolation of Antibodies with ChroPlate Filtration Plates by DALEX Biotech.**

Easy and quick high throughput antibody purification from various sources and species.

Each well of the ChroPlate has a binding capacity of > 1 mg (tested with human polyclonal Ig, binding varies between species and clones).

The proprietary resin does not shrink or swell in aqueous buffers.

High pressure stability.

pH stability short term 2 - 8, long term 3 - 8.

Excellent thermal stability up to 15 minutes at 80 °C in aqueous buffers at neutral pH.

Can be dried for long term storage (80 °C for > 2 h).

## Guidelines

For optimal binding and purity, the pH of the sample should be 7.5-8.5 and should contain 150-300 mM NaCl. An easy way to achieve this is by adding 1/11 volume 0.5 M Tris, 2 M NaCl (pH 8.0) to your sample. For screening of binding condition for e.g. a monoclonal antibody this parameters might be varied.

Purification works best with an antibody concentration of 1-2 mg/ml in your sample.

It is advisable that all fractions are collected (Sample, flow through, wash, and eluate) in separate plates for analysis, e.g. SDS-PAGE.

## Materials

Materials provided in the kit:

ChroPlate

Dummy plate

Wash buffer

Elution buffer

Neutralization buffer

Materials not provided in the kit:

Tween-20

0.5 M Tris, 2 M NaCl (pH 8.0)



## Safety warnings

! The buffers in the kit include sodium azide (CAS No. 26628-22-8) as a preservative.  
For safety information on this chemical(s) check <http://www.dguv.de/ifa/gestis-database>

## Before start

Make sure your sample is free of particulate matter. You can remove particles by centrifugation or filtration (0.45  $\mu\text{m}$ ).



- 1 Do you want to purify by centrifugation or by vacuum filtration? Please choose below.

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## STEP CASE

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### Centrifugation 6 steps

#### Equilibration

- 2 Add 500  $\mu$ l wash buffer to each well, place the ChroPlate on top of a deep-well plate, and centrifuge 5 minutes at 1000 g in a swing-out rotor. For counterbalance of the centrifuge a dummy filter plate is included in the kit.

#### Load and Wash

- 3 Place the ChroPlate on a clean deep-well plate. Add up to 1.5 ml sample to every well. Centrifuge 5 minutes at 1000 g in a swing-out rotor.

##### Note

It is advisable to keep the flow-through and wash fractions for later analysis, e.g. SDS-PAGE.

The centrifugation time depends on the sample's volume and viscosity. Volumes larger than 1 ml and viscous samples e.g. serum usually requires more than 5 minutes centrifugation time.

For optimal binding and purity, the pH of the sample should be 7.5 - 8.5 and should contain 150 - 300 mM NaCl. An easy way to achieve this is by adding 1/11 volume 0.5 M Tris, 2 M NaCl (pH 8.0) to your sample.

- 4 Empty the deep-well plate or place the ChroPlate on a clean one. Add 500  $\mu$ l wash buffer to each well and centrifuge 5 minutes at 1000 g in a swing-out rotor.

##### Note

In case of unspecific hydrophobic and/or ionic interactions include up to 1 % Tween-20 and/or up to 0.5 M NaCl in the wash buffer.



- 5 Repeat the previous step.

#### Note

For increased purity, repeat the washing step a third time.

## Elution

- 6 Place the ChroPlate on a clean deep-well plate. Add 100  $\mu$ l elution buffer to each well and centrifuge 5 minutes at 1000 g in a swing-out rotor.  
Repeat two more times.
- 7 Add one drop of neutralization solution to every well of the deep-well plate and mix gently.