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ab193265 Protein A/G/L Sepharose® Column

For the purification of monoclonal and polyclonal antibodies.

View Protein A/G/L Sepharose® Column datasheet:
www.abcam.com/ab193265
[use www.abcam.cn/ab193265 for China, or www.abcam.co.jp/ab193265 for Japan)

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

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1. Overview

Protein A/G/L binds to all IgG subclasses from various mammalian species, including all IgGs that bind to Protein A, Protein G, and Protein L, individually, making it the ideal choice for purification of all kinds of polyclonal or monoclonal IgG antibodies.

The binding capacity is greater than 10 mg/mL of bead; High flow rate; Low falling off of rProtein A/G/L; pH stability 2-10.

Protein A/G/L-Sepharose is prepared by covalently coupling recombinant Protein A/G/L (containing five Ig-binding regions of protein L, five IgG binding domains from Protein A, and three Ig-binding regions of protein G. Cell wall binding regions, albumin binding regions and other nonspecific binding regions have all been eliminated from the fusion protein to ensure the maximum specific IgG binding) to 6% cross-linked sepharose beads. The coupling technique is optimized to give a high binding capacity. The capacity of IgG binding could be greater than 10 mg of human IgG per mL of wet bead.

2. Materials Supplied and Storage

Store at 4°C. Do not freeze. Stable, as supplied, for at least 1 year.

Ready-to-use pre-packed columns.

Item	Quantity	Storage temperature
Protein A/G/L Sepharose® Column	1 mL/5 mL	4°C

3. Materials Required but not supplied

Binding Buffers: 50 mM sodium borate, 0.15 M sodium chloride pH 8.0.

Elution Buffers: 0.1 M citric acid, pH 3.0

4. 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.

5. Assay Procedure

1.1 Equilibrate the column to room temperature.
1.2 Remove the upper (first) then lower cap and allow the preservative to drain by gravity flow.
1.3 Equilibrate the column with 5 – 10 bed volumes of degassed Binding Buffer.
1.4 Add sample in Binding Buffer and recycle through column 15-20 times.
1.5 Wash with 4 – 5 column volumes of Binding Buffer containing 0.5 M NaCl.
1.6 Wash with at least 2 column volumes of Binding Buffer and ensure the effluent reaches the same Absorbance (280 nm) as the Binding Buffer.
1.7 Drain the column to the top frit. Elute with one bed volume of Elution Buffer pH 3.0.
1.8 Neutralize with 100 µL of 1M Tris, pH 9.0 per mL of eluate. It is recommended to elute with another 1-2 mL of elution buffer and collect 100 µL fractions (test for absorbance at 280 nm).
1.9 Alternatively, one could add 6 mL Elution Buffer and collect 1 mL fractions into 1.5 mL tubes containing 100 µL of 1M Tris, pH 9.0.
1.10 Combine fractions with highest absorbance (Remember to blank the spectrophotometer with a solution containing 100 µL 1M Tris, pH 9.0 per mL of Elution Buffer).
1.11 Concentration of IgG (mg/mL) = (A280/1.38).
1.12 Regenerate the column by:

1.12.1 Washing with ~ 5 volumes of Elution Buffer.
1.12.2 Equilibrate with 5 volumes of Binding Buffer containing 0.2% sodium azide.
1.12.3 Store upright at 4°C.

6. Notes

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