IMMBIOMED

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matrix bound monoclonal antibody against γ-carboxyglutamyl (Gla) residues

Product No. ADG3570Mx

Description

REF ADG3570Mx is a monoclonal antibody (REF 3570) directed against γ-carboxyglutamyl (Gla) residues found in various proteins of human and other origin, and snake venoms, bound to SEPHAROSE™ CL-4B. It is intended for use in the affinity purification of Gla-containing proteins such as prothrombin, factor VII/VIIa, factor IX/IXa, factor X/Xa, Protein C, Protein S and growth arrest-specific protein-6 (Gas6).

Properties

The antibody is bound to the sepharose at a concentration of 5 mg of antibody/mL of settled gel.

Purification Protocol

Using the following buffers, a suggested purification protocol is:

Loading/Washing Buffer: 20 mM Tris-HCl, pH 7.4
150 mM NaCl, 10 mM EDTA,

Elution Buffer: 20 mM Tris-HCl, pH 7.4
150 mM NaCl, 50 mM $CaCl_2$,

1. Pack the gel in a suitable column, approx. 16 mm in diameter
2. Equilibrate the column with Loading Buffer at a flow rate of 2 mL/min.
3. Load the sample to be purified using a flow rate of 0.5 mL/min.
4. Wash the column with Washing Buffer using a flow rate of 2 mL/min.
5. Once the A280 of the effluent has decreased to the baseline, elute the bound protein with Elution Buffer using a flow rate of 2 mL/min.
6. Dialyze the elution fraction against a suitable buffer (e.g. TBS, Tris Buffered Saline, pH 7.4) and store at –20°C.

Note: As Ca^{2+} ions in the sample may inhibit binding, the presence of a chelator such as EDTA (2-10 mM) is recommended during the binding step. In some cases, Ca^{2+} concentrations >50 mM may enhance the elution step.

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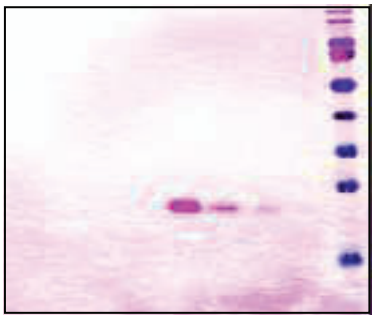
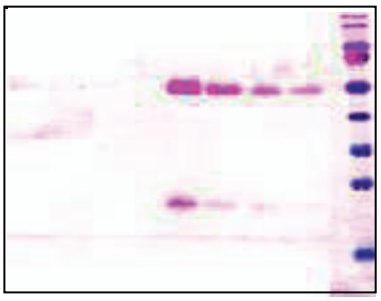
Example

Factor X (5 µg) was purified using ADG3570Mx sepharose (500 µl, 1:1 suspension) in a spin column. The fractions were analysed by Western blot analysis using (A) the monoclonal anti-Gla antibody M3B or (B) a polyclonal anti-Factor X antibody. (1= flow through, 2-4 Wash, 5-8 Eluate, M= Protein marker)

A 1 2 3 4 5 6 7 8 M



B 1 2 3 4 5 6 7 8 M



Presentation

Approximately 1 mL of settled gel immobilized with 5 mg of purified antibody with approximately 1.0 mL of PBS, pH 7.4 with 0.02% NaN_3 added as a preservative.

Storage

Store the immobilized gel at 2° - 8°C. The gel should be washed thoroughly with PBS, pH 7.4 pH prior to use to remove traces of the sodium azide preservative.

References

1. Brown, M. A., et al. Journal of Biological Chemistry 2000, 275: 19795-19802.

2. Stenberg, L. M. et al. Biochemical and Biophysical Research Communications 2001, 280: 1036–1041.

3. Stenberg, L. M. et al. Biochemical and Biophysical Research Communications 2001, 283: 454–459.

Hinweis/Note:
Der Packungsbeileger dient nur als erste Information. Der relevante Packungsbeileger liegt der Ware bei.

The datasheet is for information purposes only. The current datasheet will be enclosed with product shipment.

For research use only!

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