



Monoclonal Anti-mouse Podocalyxin Antibody

ORDERING INFORMATION

Catalog Number: MAB1556

Clone: 192703

Lot Number: IPF02

Size: 100 µg

Formulation: 0.2 µm filtered solution in PBS with 5% trehalose

Storage: -20° C

Reconstitution: sterile PBS

Specificity: mouse Podocalyxin

Immunogen: NS0-derived rmPodocalyxin

Ig class: rat IgG_{2B}

Recommended Applications:

Flow cytometry
Immunocytochemistry
Western blot

Background

Podocalyxin, also known as Podocalyxin-like protein-1 (PCLP1 or PODXL), is a type I transmembrane glycoprotein. It belongs to the CD34/Podocalyxin family of sialomucins that share structural similarity and sequence homology. Podocalyxin is a major sialoprotein in the podocytes of the kidney glomerulus and is also expressed by both endothelium and multipotent hematopoietic progenitors. It has been identified as a novel cell surface marker for hemangioblasts, the common precursors of hematopoietic and endothelial cells.¹

Preparation

This antibody was produced from a hybridoma resulting from the fusion of a mouse myeloma with B cells obtained from a rat immunized with NS0-derived, recombinant mouse Podocalyxin extracellular domain (rmPodocalyxin; aa 21 - 402; Accession # Q9R0M4). The IgG fraction of the tissue culture supernatant was purified by Protein G affinity chromatography.

Formulation

Lyophilized from a 0.2 µm filtered solution in phosphate-buffered saline (PBS) with 5% trehalose.

Reconstitution

Reconstitute with sterile PBS. If 1 mL of PBS is used, the antibody concentration will be 100 µg/mL.

Storage

Lyophilized samples are stable for twelve months from date of receipt when stored at -20° C to -70° C. Upon reconstitution, the antibody can be stored at 2° - 8° C for 1 month without detectable loss of activity. Reconstituted antibody can also be aliquotted and stored frozen at -20° C to -70° C in a manual defrost freezer for six months without detectable loss of activity. **Avoid repeated freeze-thaw cycles.**

Specificity

This antibody detects rmPodocalyxin in direct ELISAs and Western blots. It also detects Podocalyxin on the mouse embryonic stem cell line D3 by flow cytometry as well as immunocytochemistry.

Applications

Flow cytometry - Dilute this antibody to 0.1 mg/mL and add 10 µL of the diluted solution to 1 - 2.5 x 10⁵ cells in a total reaction volume not exceeding 200 µL. The binding of unlabeled monoclonal antibodies may be visualized by adding 10 µL of a 25 µg/mL stock solution of a secondary developing reagent such as goat anti-rat IgG conjugated to a fluorochrome.

Immunocytochemistry - This antibody can be used with the appropriate secondary reagents at a concentration of 10 µg/mL in fixed cells. For chromogenic detection of labeling, the use of R&D Systems Cell and Tissue Staining Kits (CTS Series) is recommended.

Western blot - This antibody can be used at 1 - 2 µg/mL with the appropriate secondary reagents to detect mouse Podocalyxin. Using a colorimetric detection system, the detection limit for rmPodocalyxin is approximately 5 ng/lane and 100 ng/lane under non-reducing and reducing conditions, respectively. Chemiluminescent detection will increase sensitivity by 5 to 50 fold.

Optimal dilutions should be determined by each laboratory for each application.

Reference:

1. Hara, T. *et al.*, 1999, *Immunity* 11(5):567 - 578.

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