

ORDERING INFORMATION

Catalog Number: AF3250

Lot Number: WOQ01

Size: 100 µg

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

Storage: -20° C

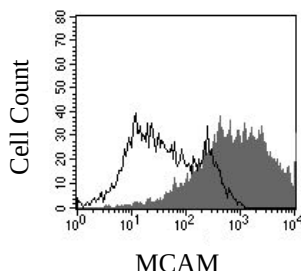
Reconstitution: sterile PBS

Specificity: rat MCAM extracellular domain

Immunogen: NS0-derived rrMCAM extracellular domain

Ig Type: goat IgG

Applications: Western blot
Flow cytometry
Direct ELISA



Rat PC-12 cells were stained with anti-MCAM (R&D Systems, Cat. # AF3250, filled histogram), or control antibody (R&D Systems, Cat. # AB-108-C, open histogram), followed by PE-conjugated anti-goat antibody (R&D Systems, Cat. # F0107).

Preparation

Produced in goats immunized with purified, NS0-derived, recombinant rat Melanoma Cell Adhesion Molecule (rrMCAM; aa 19 - 560; Accession # Q9EPF2) extracellular domain. Rat MCAM specific IgG was purified by rat MCAM affinity chromatography. MCAM, also known as CD146 and MUC18, is a type I transmembrane adhesion molecule belonging to the Ig superfamily. It is expressed on all endothelial cells, different epithelial cells, multiple lymphocyte subsets and various tumor cells. Two splice isoforms that differ in their cytoplasmic domains and vasolateral localization have been described. A soluble MCAM is shed and can be detected in circulation. The amino acid sequence of rat MCAM is 72% and 90% identical to that of human and mouse MCAM, respectively.

Formulation

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

Reconstitution

Reconstitute with sterile PBS. If 0.5 mL of PBS is used, the antibody concentration will be 0.2 mg/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 has been selected for its ability to recognize rat MCAM in the applications listed below.

Applications

Western blot - This antibody can be used at 0.1 - 0.2 µg/mL with the appropriate secondary reagents to detect rat MCAM. The detection limit for rrMCAM is approximately 5 ng/lane under non-reducing and reducing conditions. In this format, this antibody shows approximately 5% cross-reactivity with rhMCAM.

Flow Cytometry - This antibody has been tested on PC-12 cells for use in flow cytometry. Dilute this antibody to 50 µg/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 antibodies may be visualized by adding 10 µL of a 25 µg/mL stock solution of a secondary developing reagent such as anti-goat IgG conjugated to a fluorochrome.

Direct ELISA - This antibody can be used at 0.5 - 1.0 µg/mL with the appropriate secondary reagents to detect rat MCAM. The detection limit for rrMCAM is approximately 0.3 ng/well.

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