Monoclonal Anti-mouse JAM-C-APC

Catalog Number: FAB7050A
Lot Number: ACHX01
100 Tests

Reagents Provided

Allophycocyanin (APC)-conjugated rat monoclonal anti-mouse JAM-C: Supplied as 10 μg of antibody in 1 mL saline containing up to 0.5% BSA and 0.1% sodium azide.

Clone #: 209628

Isotype: rat IgG_{2B}

Reagents Not Provided

- Flow Cytometry Staining Buffer (Catalog # FC001) or other BSA-supplemented saline buffer.

Storage

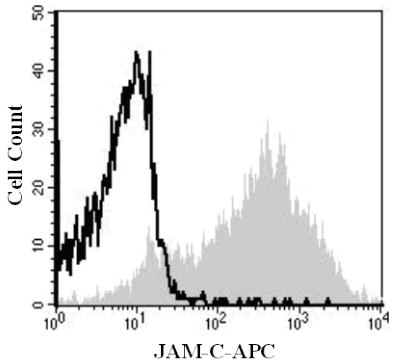
Reagents are stable for **twelve months** from the date of receipt when stored in the dark at 2° - 8° C.

Intended Use

Designed to quantitatively determine the percentage of cells bearing JAM-C within a population and qualitatively determine the density of JAM-C on cell surfaces by flow cytometry.

Product Description

This antibody was produced from a hybridoma resulting from the fusion of a mouse myeloma with B cells obtained from a mouse immunized with purified, NS0-derived, recombinant mouse junctional adhesion molecule C (JAM-C) extracellular domain. The IgG fraction of the tissue culture supernatant was purified by Protein G affinity chromatography. The purified antibody was then conjugated to APC fluorochrome. Cell surface expression of JAM-C is determined by flow cytometry using 620 - 650 nm wavelength excitation and monitoring emitted fluorescence with a detector optimized to collect peak emissions at 660 - 670 nm.



B16-F1 cells were stained with APC-conjugated anti-mouse JAM-C (Catalog # FAB7050A, filled histogram) or APC-conjugated isotype control (Catalog # IC013A, open histogram).

Background Information

The family of junctional adhesion molecules (JAM), which is comprised of at least three members, contains type I transmembrane receptors belonging to the immunoglobulin (Ig) superfamily. These proteins are localized in the tight junctions between endothelial cells or epithelial cells. Some family members are also found on blood leukocytes and platelets. Mouse JAM-C is highly expressed during embryogenesis. In adult tissues, mouse JAM-C is restricted to endothelial cells, lymph endothelial cells in the kidney, lymph node, and Peyer's patches, where the protein can be localized to the high endothelial venules. Although human JAM-C is expressed on human platelets and a subset of leukocytes, mouse JAM-C expression was not detected on any mouse lymphocytes. Mouse JAM-C shares 86% amino acid (aa) sequence identity with its human homologue. It also shares approximately 31% and 35% aa sequence homology with mouse JAM-A and JAM-B, respectively.

Flow Cytometry Validation

This antibody has been tested for flow cytometry using B16-F1 cells.

1. Cells may be Fc-blocked with 1 μg of mouse IgG/10^5 cells for 15 minutes at room temperature. Do not wash excess blocking IgG from this reaction.
2. After blocking, 10 μL of conjugated antibody was added to up to 1 x 10^6 cells and incubated for 30 minutes at room temperature.
3. Unbound antibody was removed by washing the cells twice in Flow Cytometry Staining Buffer (Catalog # FC001). Note that whole blood requires a RBC lysis step at this point using Flow Cytometry Mouse Lyse Buffer (Catalog # FC003).
4. The cells were resuspended in Flow Cytometry Staining Buffer for final flow cytometric analysis. As a control for this analysis, cells in a separate tube should be treated with APC-labeled rat IgG_{2B} antibody. This procedure may need to be modified, depending upon the cell type and final utilization. Individual users may need to titrate to determine the optimal reagent amount for their specific use.

Warning: Contains sodium azide as a preservative - sodium azide may react with lead and copper plumbing to form explosive metal azides. Flush with large volumes of water during disposal.

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