Monoclonal Anti-mouse/rat CD19-APC

Catalog Number: FAB7489A
Lot Number: ADAZ01
100 Tests

Reagents Provided

Allophycocyanin (APC)-conjugated mouse monoclonal anti-mouse/rat CD19: Supplied as 10 μg of antibody in 1 mL saline containing up to 0.5% BSA and 0.09% sodium azide.

Clone #: 771404

Isotype: mouse IgG_{2B}

Reagents Not Provided

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

Storage

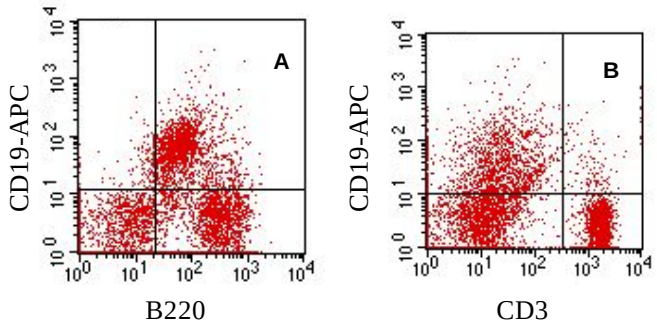
Reagents are stable for **twelve months** from the date of receipt when stored in the dark at 2-8 °C.

Intended Use

Designed to quantitatively determine the percentage of cells bearing CD19 within a population and qualitatively determine the density of CD19 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 CHO cell-derived recombinant rat CD19 (aa 1-287; Accession # NP_001013255). The IgG fraction of the tissue culture supernatant was purified by Protein A or G affinity chromatography. The purified antibody was then conjugated to APC fluorochrome. Cell surface expression of CD19 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.



(A) Mouse splenocytes were stained with APC-conjugated anti-mouse/rat CD19 (Catalog # FAB7489A) and CFS-conjugated anti-mouse B220 (Catalog # FAB1217F).

(B) Rat splenocytes were stained with APC-conjugated anti-mouse/rat CD19 (Catalog # FAB7489A) and PE-conjugated anti-rat CD3. Quadrant markers were set based on staining with an APC-conjugated isotype control (Catalog # IC0041A).

Background Information

CD19 (also surface antigen B4 and Leu12) is a 95-110 kDa member of the Immunoglobulin superfamily of molecules. It is expressed by B cells, and interacts with CD21 for the purpose of reducing the threshold of the antigen signal needed to activate the BCR. CD19 ligation also promotes B cell:follicular dendritic cell (FDC) interaction and B cell proliferation in the FDC zone of the spleen. Mature rat CD19 is a 529 amino acid (aa) type I transmembrane glycoprotein (aa 19-547). Based on mouse, it contains a 269 aa extracellular region (aa 19-287) plus a 236 aa cytoplasmic domain. The extracellular region possesses two C2-type Ig like domains (aa 20-113 and 171-271) and one utilized phosphorylation site at Ser225. The cytoplasmic domain contains five potential Tyr phosphorylation sites. There is one splice form that shows a two aa substitution after Gly489. Over aa 19-287, rat CD19 shares 88% and 57% aa sequence identity with mouse and human CD19, respectively.

Flow Cytometry Validation

This antibody has been tested for flow cytometry using mouse and rat splenocytes.

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 mouse 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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