

Reagents Provided

Allophycocyanin (APC)-conjugated rat monoclonal anti-mouse CD45: Supplied as 10 µg of antibody in 1 mL saline containing up to 0.5% BSA and 0.1% sodium azide.

Clone #: 30-F11

Isotype: rat IgG_{2b}

Reagents Not Provided

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

Storage

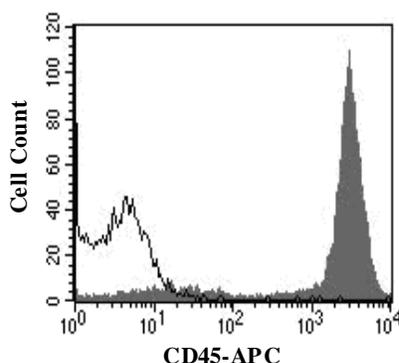
Reagents are stable for **twelve months** from the date of receipt when stored in the dark at 2° - 8° C.

Intended Use

Designed to quantitatively determine the percentage of cells bearing CD45 within a population and qualitatively determine the density of CD45 on cell surfaces by flow cytometry.

Product Description

This antibody was produced from a murine hybridoma elicited from a rat immunized with mouse thymus and spleen cells. 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 CD45 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.



Mouse splenocytes were stained with APC-conjugated anti-mouse CD45 (Catalog # FAB114A, filled histogram) or isotype control (Catalog # IC013A, open histogram).

Background Information

CD45, also known as leukocyte common antigen, T200 glycoprotein, B220, and Ly 5, is a transmembrane protein tyrosine phosphatase. It is expressed at high levels on the cell surface of all nucleated hematopoietic cells and their precursors. Alternative splicing of CD45 mRNA results in the expression of several distinct isoforms that are differentially expressed in cells of different lineages and activation states.

Flow Cytometry Validation

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

- Cells may be Fc-blocked with 1 µg of mouse IgG/10⁵ cells for 15 minutes at room temperature. Do not wash excess blocking IgG from this reaction.
- After blocking, 10 µL of conjugated antibody was added to up to 1 x 10⁶ cells and incubated for 30 minutes at room temperature.
- 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).
- 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.