

# Monoclonal Anti-mouse B220/CD45R-PerCP

Catalog Number: FAB1217C

Lot Number: ABAY01

100 Tests

## Reagents Provided

**Peridinin-Chlorophyll-Protein-Complex (PerCP)-conjugated rat monoclonal anti-mouse B220/CD45R:** Supplied as 25 µg of antibody in 1 mL saline containing up to 0.5% BSA and 0.1% sodium azide.

**Clone #:** RA3-6B2

**Isotype:** rat IgG<sub>2a</sub>

## Reagents Not Provided

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

## Storage

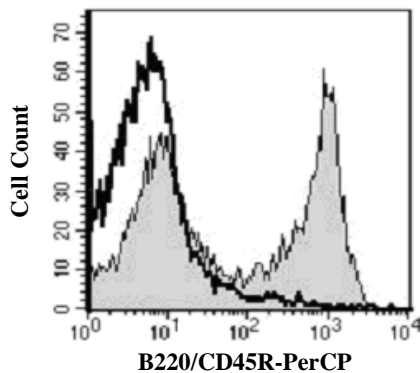
Reagents are stable for **twelve months** from the date of receipt when stored in the dark at 2° - 8° C.

## Intended Use

Designed to quantitatively determine the percentage of cells bearing B220/CD45R within a population and qualitatively determine the density of B220/CD45R 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 rat immunized with Abelson murine leukemia virus induced Pre-B tumor cells.<sup>1</sup> The IgG fraction of the tissue culture supernatant was purified by Protein G affinity chromatography. The purified antibody was then conjugated to PerCP fluorochrome. Cell surface expression of B220/CD45R is determined by flow cytometry. PerCP has a maximum absorption of 482 nm and 564 nm and a maximum emission of 675 nm.



Mouse splenocytes were stained with PerCP-conjugated anti-mouse B220/CD45R (Catalog # FAB1217C, filled histogram) or isotype control (Catalog # IC006C, open histogram).

## Background Information

B220 is a subset of mouse CD45 isoforms predominantly expressed on all B lymphocytes, including pro-, mature and activated B cells.<sup>1-3</sup> The level of B220 antigen expression on the B cell lineage is developmentally regulated<sup>2-4</sup> and this antibody is commonly used as a B cell marker. RA3-6B2 has been reported to inhibit *in vivo* B cell responses.<sup>5,6</sup>

## References

- Coffman, R.L. (1982) *Immuno. Rev.* **69**:5.
- Hardy, R.R. *et al.* (1991) *J. Exp. Med.* **173**:1213.
- Hathcock, K.S. *et al.* (1992) *J. Immunol.* **149**:2286.
- Allman, D.M. *et al.* (1992) *J. Immunol.* **149**:2533.
- Asensi, V.K. *et al.* (1989) *Immunology* **68**:204.
- Domati-Saad, R. *et al.* (1993) *J. Immunol.* **151**:5936.

## 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/10<sup>5</sup> 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 1 - 2.5 x 10<sup>5</sup> 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 analysis by flow cytometry. As a control for this analysis, cells in a separate tube should be treated with PerCP-labeled rat IgG<sub>2a</sub> antibody. This procedure may need to be modified, depending upon cell type and final utilization. Individual users may need to titrate to determine 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.