Technical Data Sheet

PE-Cy[™]7 Rat Anti-Mouse CD45R/B220

Product Information

Material Number: 561881

Alternate Name: B220; Ly-5; CD45R; LCA; Ptprc; Protein tyrosine phosphatase receptor type C

Size 0.2 mg/ml Concentration: RA3-6B2 Clone:

Immunogen: Mouse Abelson Leukemia Virus-Induced pre-B tumor cells

Isotype: Rat IgG2a, κ Reactivity: QC Testing: Mouse

Tested in Development: Human

Storage Buffer: Aqueous buffered solution containing ≤0.09% sodium azide.

Description

The rat anti-mouse CD45R antibody (clone RA3-6B2) has been reported to react with an epitope on the extracellular domain of the transmembrane CD45 glycoprotein which is dependent upon the expression of exon A and specific carbohydrate residues. It is expressed on B lymphocytes at all stages from pro-B through mature and activated B cell, but it is decreased on plasma cells and a subset of memory B cells. The levels of CD45R expression on the B-cell lineage appear to be developmentally regulated. It is also reportedly found on the abnormal T cells involved in the lymphadenopathy of lpr/lpr and gld/gld mutant mice, on lytically active subsets of lymphokine-activated killer cells (NK cells and non-MHC-restricted CTL), on apoptotic T lymphocytes of mice injected with bacterial superantigen, on a population of NK-cell precursors in the bone marrow, and on B-lymphocyte, T-lymphocyte, and macrophage progenitors in fetal liver. The CD45R antigen has been reported not to be on hematopoietic stem cells, naive T lymphocytes, or MHC-restricted CTL. CD45 is a member of the Protein Tyrosine Phosphatase (PTP) family. Its intracellular (COOH-terminal) region contains two PTP catalytic domains, and the extracellular region is highly variable due to alternative splicing of exons 4, 5, and 6 (designated A, B, and C, respectively), plus differing levels of glycosylation. The CD45 isoforms detected in the mouse are cell type-, maturation, and activation state-specific. The CD45 isoforms play complex roles in T-cell and B-cell antigen receptor signal transduction. CD45R is commonly used as a pan B-cell marker; however, CD19 expression, detectable by the rat anti-mouse CD19 antibody (clone 1D3), is reported to be more restricted to the B-cell lineage. The rat anti-mouse CD45R antibody (clone RA3-6B2) has been reported to enhance isotype switching during in vitro B-cell responses and to inhibit in vivo B-cell responses. Cross-reaction of the RA3-6B2 clone with activated human T lymphocytes has also been reportedly observed.

Preparation and Storage

Store undiluted at 4°C and protected from prolonged exposure to light. Do not freeze.

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.

The antibody was conjugated with PE-Cy7 under optimum conditions, and unconjugated antibody and free PE-Cy7 were removed.

Application Notes

Application

Flow cytometry	Routinely Tested	

Suggested Companion Products

Catalog Number	Name	Size	Clone
552784	PE-Cy TM 7 Rat IgG2a, κ Isotype Control	0.1 mg	R35-95
553061	FITC Hamster Anti-Mouse CD3e	0.1 mg	145-2C11
554656	Stain Buffer (FBS)	500 ml	(none)

Product Notices

- Since applications vary, each investigator should titrate the reagent to obtain optimal results.
- Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
- For fluorochrome spectra and suitable instrument settings, please refer to our Fluorochrome Web Page at www.bdbiosciences.com/colors.
- Please observe the following precautions: Absorption of visible light can significantly alter the energy transfer occurring in any tandem fluorochrome conjugate; therefore, we recommend that special precautions be taken (such as wrapping vials, tubes, or racks in aluminum foil) to prevent exposure of conjugated reagents, including cells stained with those reagents, to room illumination.

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- 5. PE-Cy7 is a tandem fluorochrome composed of R-phycoerythrin (PE), which is excited by 488-nm light and serves as an energy donor, coupled to the cyanine dye Cy7, which acts as an energy acceptor and fluoresces maximally at 780 nm. PE-Cy7 tandem fluorochrome emission is collected in a detector for fluorescence wavelengths of 750 nm and higher. Although every effort is made to minimize the lot-to-lot variation in the efficiency of the fluorochrome energy transfer, differences in the residual emission from PE may be observed. Therefore, we recommend that individual compensation controls be performed for every PE-Cy7 conjugate. PE-Cy7 is optimized for use with a single argon ion laser emitting 488-nm light, and there is no significant overlap between PE-Cy7 and FITC emission spectra. When using dual-laser cytometers, which may directly excite both PE and Cy7, we recommend the use of cross-beam compensation during data acquisition or software compensation during data analysis.
- 6. Cy is a trademark of Amersham Biosciences Limited. This conjugated product is sold under license to the following patents: US Patent Nos. 5,486,616; 5,569,587; 5,569,766; 5,627,027.
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- Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before
 discarding to avoid accumulation of potentially explosive deposits in plumbing.
- 9. Warning: Some APC-Cy7 and PE-Cy7 conjugates show changes in their emission spectrum with prolonged exposure to formaldehyde. If you are unable to analyze fixed samples within four hours, we recommend that you use BDTM Stabilizing Fixative (Cat. No. 338036).
- 10. An isotype control should be used at the same concentration as the antibody of interest.

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