## **Technical Data Sheet**

# Alexa Fluor® 647 Rat Anti-Mouse CD45R

#### **Product Information**

Material Number: 557683

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

 Size:
 0.1 mg

 Concentration:
 0.2 mg/ml

 Clone:
 RA3-6B2

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

 Isotype:
 Rat IgG2a, κ

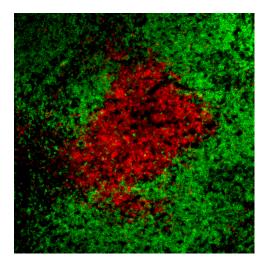
 Reactivity:
 QC Testing: Mouse

Reported Reactivity: Human

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

### Description

The RA3-6B2 monoclonal antibody specifically binds to 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.



Immunohistofluorescent analysis of CD45R/B220 expression by cells within C57BL/6 mouse spleen. A mouse spleen cryosection (5 µm) was fixed with BD Cytofix™ Fixation Buffer (Cat. No. 554655), blocked with 5% goat serum and 1% BSA diluted in 1x PBS, and stained with Alexa Fluor® 647 Rat Anti-Mouse CD45R/B220 antibody (Cat. No. 557683, pseudo-colored green) and BD Horizon™ BV421 Rat Anti-Mouse CD3 Molecular Complex antibody (Cat. No. 564008, pseudo-colored red). Images were captured on a standard epifluorescence microscope. Original magnification, 20x.

### **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 to Alexa Fluor® 647 under optimum conditions, and unreacted Alexa Fluor® 647 was removed.

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### **Application Notes**

#### Application

Flow cytometry	Routinely Tested
Immunofluorescence	Tested During Development

#### **Suggested Companion Products**

Catalog Number	Name	Size	Clone	
557690	Alexa Fluor® 647 Rat IgG2a, κ Isotype Control	0.1 mg	R35-95	
564907	DAPI Solution	1 mg	(none)	
554656	Stain Buffer (FBS)	500 mL	(none)	
554657	Stain Buffer (BSA)	500 mL	(none)	
554655	Fixation Buffer	100 mL	(none)	
564008	BV421 Rat Anti-Mouse CD3 Molecular Complex	50 μg	17A2	

#### **Product Notices**

- Since applications vary, each investigator should titrate the reagent to obtain optimal results.
- 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.
- For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
- Alexa Fluor® 647 fluorochrome emission is collected at the same instrument settings as for allophycocyanin (APC). 4.
- The Alexa Fluor®, Pacific Blue<sup>TM</sup>, and Cascade Blue® dye antibody conjugates in this product are sold under license from Molecular Probes, Inc. for research use only, excluding use in combination with microarrays, or as analyte specific reagents. The Alexa Fluor® dyes (except for Alexa Fluor® 430), Pacific Blue<sup>TM</sup> dye, and Cascade Blue® dye are covered by pending and issued patents.
- Alexa Fluor® is a registered trademark of Molecular Probes, Inc., Eugene, OR.
- 7. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.

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