

Technical Data Sheet

BUV615 Rat Anti-Mouse CD45R/B220

Product Information

Material Number:	751580
Size:	50 µg
Clone:	RA3-6B2
Alternative Name:	B220; Ly-5; CD45R; LCA; Ptpcr; Protein tyrosine phosphatase receptor type C
Reactivity:	Mouse (Tested in Development)
Isotype:	Rat IgG2a, κ
Immunogen:	Mouse Abelson Leukemia Virus-Induced pre-B tumor cells
Application:	Flow cytometry (Qualified)
Concentration:	0.2 mg/ml
Storage Buffer:	Aqueous buffered solution containing ≤0.09% sodium azide.
Regulatory Status:	RUO

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.

The antibody was conjugated to BD Horizon BUV615 which is part of the BD Horizon Brilliant™ Ultraviolet family of dyes. This dye is a tandem fluorochrome with an Ex Max near 350 nm and an Em Max near 615 nm. BD Horizon Brilliant BUV615 can be excited by the ultraviolet laser (355 nm) and detected with a 610/20 filter and a 595 nm LP. Due to the excitation of the acceptor dye by the blue/yellow-green laser line, there may be significant spillover into channels detecting PE-CF594 like emissions (eg, 610/20-nm filter).

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 the dye under optimum conditions that minimize unconjugated dye and antibody.

Recommended Assay Procedure

BD™ CompBeads can be used as surrogates to assess fluorescence spillover (Compensation). When fluorochrome conjugated antibodies are bound to BD CompBeads, they have spectral properties very similar to cells. However, for some fluorochromes there can be small differences in spectral emissions compared to cells, resulting in spillover values that differ when compared to biological controls. It is strongly recommended that when using a reagent for the first time, users compare the spillover on cells and BD CompBead to ensure that BD CompBeads are appropriate for your specific cellular application.

For optimal and reproducible results, BD Horizon Brilliant Stain Buffer should be used anytime two or more BD Horizon Brilliant dyes are used in the same experiment. Fluorescent dye interactions may cause staining artifacts which may affect

data interpretation. The BD Horizon Brilliant Stain Buffer was designed to minimize these interactions. More information can be found in the Technical Data Sheet of the BD Horizon Brilliant Stain Buffer (Cat. No. 563794/566349) or the BD Horizon Brilliant Stain Buffer Plus (Cat. No. 566385).

Note: When using high concentrations of antibody, background binding of this dye to erythroid cell subsets (mature erythrocytes and precursors) has been observed. For researchers studying these cell populations, or in cases where light scatter gating does not adequately exclude these cells from the analysis, this background may be an important factor to consider when selecting reagents for panel(s).

Suggested Companion Products

Catalog Number	Name	Size	Clone
566385	Brilliant Stain Buffer Plus	1000 Tests	
566349	Brilliant Stain Buffer	1000 Tests	
553141	Purified Rat Anti-Mouse CD16/CD32 (Mouse BD Fc Block™)	0.1 mg	2.4G2
565804	Red Nucleic Acid Stain	0.5 mL	
613005	BUV615 Rat IgG2a, κ Isotype Control	50 μ g	R35-95
554656	Stain Buffer (FBS)	500 mL	
554657	Stain Buffer (BSA)	500 mL	
563794	Brilliant Stain Buffer	100 Tests	
555899	Lysing Buffer	100 mL	

Product Notices

1. The production process underwent stringent testing and validation to assure that it generates a high-quality conjugate with consistent performance and specific binding activity. However, verification testing has not been performed on all conjugate lots.
2. Researchers should determine the optimal concentration of this reagent for their individual applications.
3. An isotype control should be used at the same concentration as the antibody of interest.
4. 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.
5. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
6. Please refer to www.bdbiosciences.com/us/s/resources for technical protocols.
7. BD Horizon Brilliant Stain Buffer is covered by one or more of the following US patents: 8,110,673; 8,158,444; 8,575,303; 8,354,239.
8. Please refer to <http://regdocs.bd.com> to access safety data sheets (SDS).
9. CF™ is a trademark of Biotium, Inc.
10. BD Horizon Brilliant Ultraviolet 615 is covered by one or more of the following US patents: 8,110,673; 8,158,444; 8,227,187; 8,575,303; 8,354,239.

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