Technical Data Sheet Purified Rat Anti-Mouse CD45R/B220

Product Information		
Material Number:	553084	
Size:	0.5 mg	
Concentration:	0.5 mg/ml	
Clone:	RA3-6B2	
Immunogen:	Mouse Pre-B Tumor	
Isotype:	Rat IgG2a, ĸ	
Reactivity:	QC Testing: Mouse	
	Reported: Human	
Storage Buffer:	Aqueous buffered solution containing $\leq 0.09\%$ sodium azide.	

Description

The RA3-6B2 antibody reacts with an epitope on the extracellular domain of CD45 glycoprotein which is dependent upon the expression of exon A and specific carbohydrate residues. It is expressed on B lyphocytes 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. Levels of expression of CD45R/B220 on the B-cell lineage appear to be developmentally regulated. It is also 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/B220 antigen is not 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/B220 is commonly used as a pan B-cell marker; however, CD19 expression, detected by mAb 1D3, is reported to be more restricted to the B-cell lineage. mAb 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 RA3-6B2 mAb with activated human T lymphocytes has been observed.

This antibody is routinely tested by flow cytometric analysis. Other applications were tested at BD Biosciences Pharmingen during antibody development only or reported in the literature.

Preparation and Storage

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography. Store undiluted at 4° C.

Application Notes

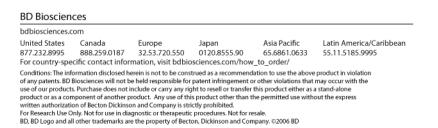
Application

Flow cytometry	Routinely Tested
Immunoprecipitation	Reported
Immunohistochemistry-frozen	Reported
Immunohistochemistry-zinc-fixed	Reported

Recommended Assay Procedure:

Sodium azide is a reversible inhibitor of oxidative metabolism; therefore, antibody preparations containing this preservative agent must not be used in cell cultures nor injected into animals. Sodium azide may be removed by washing stained cells or plate-bound antibody or dialyzing soluble antibody in sodium azide-free buffer. Since endotoxin may also affect the results of functional studies, we recommend the NA/LETM antibody format for in vitro and in vivo use.

For IHC, we recommend the use of purified RA3-6B2 mAb in our special formulation for immunohistochemistry, Cat. No. 550286.





Suggested Companion Products

Catalog Number	Name	Size	Clone
550286	Purified Rat Anti-Mouse CD45R/B220	1.0 ml	RA3-6B2

Product Notices

- 1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
- 2. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
- 3. 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.

References

Allman DM, Ferguson SE, Cancro MP. Peripheral B cell maturation. I. Immature peripheral B cells in adults are heat-stable antigenhi and exhibit unique signaling characteristics. *J Immunol.* 1992; 149(8):2533-2540.(Biology)

Asensi V, Kimeno K, Kawamura I, Sakumoto M, Nomoto K. Treatment of autoimmune MRL/lpr mice with anti-B220 monoclonal antibody reduces the level of anti-DNA antibodies and lymphadenopathies. *Immunology*. 1989; 68(2):204-208.(Biology)

Ballas ZK, Rasmussen W. Lymphokine-activated killer cells. VII. IL-4 induces an NK1.1+CD8 alpha+beta- TCR-alpha beta B220+ lymphokine-activated killer subset. J Immunol. 1993; 150(1):17-30.(Biology)

Bleesing JJ, Morrow MR, Uzel G, Fleisher TA. Human T cell activation induces the expression of a novel CD45 isoform that is analogous to murine B220 and is associated with altered O-glycan synthesis and onset of apoptosis. *Cell Immunol.* 2001; 213(1):72-81.(Biology)

Coffman RL. Surface antigen expression and immunoglobulin gene rearrangement during mouse pre-B cell development. *Immunol Rev.* 1982; 69:5-23.(Biology) Domiati-Saad R, Ogle EW, Justement LB. Administration of anti-CD45 mAb specific for a B cell-restricted epitope abrogates the B cell response to a T-dependent antigen in vivo. *J Immunol.* 1993; 151(11):5936-5947.(Biology)

Driver DJ, McHeyzer-Williams LJ, Cool M, Stetson DB, McHeyzer-Williams MG. Development and maintenance of a B220- memory B cell compartment. J Immunol. 2001; 167(3):1393-1405.(Biology)

George A, Rath S, Shroff KE, Wang M, Durdik JM. Ligation of CD45 on B cells can facilitate production of secondary Ig isotypes. J Immunol. 1994; 152(3):1014-1021. (Biology)

Hardy RR, Carmack CE, Shinton SA, Kemp JD, Hayakawa K. Resolution and characterization of pro-B and pre-pro-B cell stages in normal mouse bone marrow. J Exp Med. 1991; 173(5):1213-1225. (Biology)

Hathcock KS, Hirano H, Murakami S, Hodes RJ. CD45 expression by B cells. Expression of different CD45 isoforms by subpopulations of activated B cells. J Immunol. 1992; 149(7):2286-2294. (Biology)

Johnson P, Maiti A. CD45: A family of leukocyte-specific cell surface glycoproteins. In: Herzenberg LA, Weir DM, Blackwell C, ed. Weir's Handbook of Experimental Immunology, Vol 2. Cambridge: Blackwell Science; 1997:62.1-62.16.(Biology)

Kobata T, Takasaki K, Asahara H, et al. Apoptosis with FasL+ cell infiltration in the periphery and thymus of corrected autoimmune mice. *Immunology*. 1997; 92(2):206-213.(Biology)

Krop I, de Fougerolles AR, Hardy RR, Allison M, Schlissel MS, Fearon DT. Self-renewal of B-1 lymphocytes is dependent on CD19. Eur J Immunol. 1996; 26(1):238-242. (Biology)

Laouar Y, Ezine S. In vivo CD4+ lymph node T cells from lpr mice generate CD4-CD8-B220+TCR-beta low cells. *J Immunol.* 1994; 153(9):3948-3955.(Biology) Puzanov IJ, Bennett M, Kumar V. IL-15 can substitute for the marrow microenvironment in the differentiation of natural killer cells. *J Immunol.* 1996; 157(10):4282-4285.(Biology)

Renno T, Hahne M, Tschopp J, MacDonald HR. Peripheral T cells undergoing superantigen-induced apoptosis in vivo express B220 and upregulate Fas and Fas ligand. J Exp Med. 1996; 183(2):431-437. (Biology)

Rolink A, ten Boekel E, Melchers F, Fearon DT, Krop I, Andersson J. A subpopulation of B220+ cells in murine bone marrow does not express CD19 and contains natural killer cell progenitors. J Exp Med. 1996; 183(1):187-194. (Biology)

Sagara S, Sugaya K, Tokoro Y, et al. B220 expression by T lymphoid progenitor cells in mouse fetal liver. J Immunol. 1997; 158(2):666-676.(Biology)