SONY

Product Data Sheet

Alexa Fluor® 488 anti-mouse/human CD45R/B220

Catalog # / Size: 1116140 / 25 µg
1116125 / 100 µg

Clone: RA3-6B2

Isotype: Rat IgG2a, κ

Immunogen: Abelson murine leukemia virus-induced pre-B tumor cells

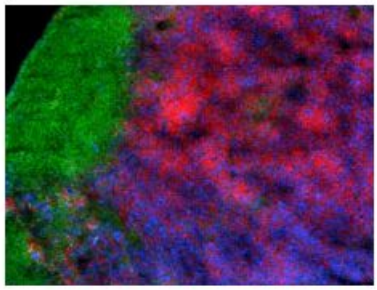
Reactivity: Human, Mouse

Preparation: The antibody was purified by affinity chromatography and conjugated with Alexa Fluor® 488 under optimal conditions.

Formulation: Phosphate-buffered solution, pH 7.2, containing 0.09% sodium azide.

Workshop Number: 810 under optimal conditions.

Concentration: 0.5



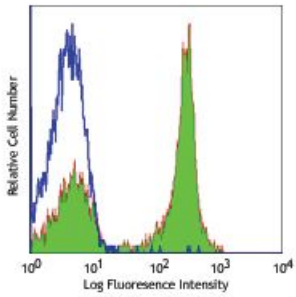
C57BL/6 mouse frozen lymph node section was fixed with 4% paraformaldehyde (PFA) for 10 minutes at room temperature and blocked with 5% FBS plus 5% rat serum for 1 hour at room temperature. Then the section was stained with 5 µg/ml of CD8 (clone 53-6.7) Alexa Fluor® 594 (red), 5 µg/ml of CD4 (clone GK1.5) Alexa Fluor® 647 (blue), and 5 µg/ml of B220 (clone RA3-6B2) Alexa Fluor® 488 (green) overnight at 4°C. The image was captured by 10X objective.

Applications:

Applications: Flow Cytometry, Immunohistochemistry, Other

Recommended Usage: Each lot of this antibody is quality control tested by immunofluorescent staining with flow cytometric analysis. For flow cytometric staining, the suggested use of this reagent is ≤2.0 µg per million cells in 100 µl volume. For immunohistochemisty, a concentration range of 2.5-5 µg/ml is suggested. For immunofluorescence microscopy, a concentration range of 1.25-10 µg/ml is recommended. It is recommended that the reagent be titrated for optimal performance for each application.

* Alexa Fluor® 488 has a maximum emission of 519 nm when it is excited at 488 nm.



C57BL/6 mouse splenocytes stained with RA3-6B2 Alexa Fluor® 488

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Application Notes: Clone RA3-6B2 has been described 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. Additional reported applications (for the relevant formats) include: immunoprecipitation1, *in vitro* and *in vivo* modulation of B cell responses[2-4], and immunohistochemistry of acetone-fixed frozen sections and formalin-fixed paraffin-embedded sections[5,6]. The LEAF™ purified antibody (Endotoxin <0.1 EU/µg, Azide-Free, 0.2 µm filtered) is recommended for functional assays (Cat. No. 103216).

Application References:

1. Coffman RL. 1982. *Immunol. Rev.* 69:5. (IP)
2. George A, *et al.* 1994. *J. Immunol.* 152:1014. (Activ)
3. Asensi V, *et al.* 1989. *Immunology* 68:204. (Activ)
4. Domiati-Saad R, *et al.* 1993. *J. Immunol.* 151:5936. (Activ)
5. Hata H, *et al.* 2004. *J. Clin. Invest.* 114:582. (IHC)
6. Monteith CE, *et al.* 1996. *Can. J. Vet. Res.* 60:193. (IHC)
7. Shih FF, *et al.* 2006. *J. Immunol.* 176:3438. (FC)
8. Chang C L-T, *et al.* 2007. *J. Immunol.* 178:6984.
9. Fazilleau N, *et al.* 2007. *Nature Immunol.* 8:753.
10. Lang GL, *et al.* 2008. *Blood* 111:2158. PubMed
11. Charles N, *et al.* 2010. *Nat. Med.* 16:701. (FC) PubMed
12. del Rio ML, *et al.* 2011. *Transpl. Int.* 24:501. (FC) PubMed
13. Murakami R, *et al.* 2013. *PLoS One.* 8:73270. PubMed

Description: CD45R, also known as B220, is an isoform of CD45. It is a member of the protein tyrosine phosphatase (PTP) family with a molecular weight of approximately 180-240 kD. CD45R is expressed on B cells (at all developmental stages from pro-B cells through mature B cells), activated B cells, and subsets of T and NK cells. CD45R (B220) is also expressed on a subset of abnormal T cells involved in the pathogenesis of systemic autoimmunity in MRL-*Fas*^{lpr} and MRL-*Fas*^{gld} mice. It plays a critical role in TCR and BCR signaling. The primary ligands for CD45 are galectin-1, CD2, CD3, and CD4. CD45R is commonly used as a pan-B cell marker; however, CD19 may be more appropriate for B cell specificity.

Antigen References:

1. Barclay A, *et al.* 1997. The Leukocyte Antigen FactsBook Academic Press.
2. Trowbridge IS, *et al.* 1993. *Annu. Rev. Immunol.* 12:85.
3. Kishihara K, *et al.* 1993. *Cell* 74:143.
4. Pulido R, *et al.* 1988. *J. Immunol.* 140:3851.

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