Alexa Fluor® 594 anti-mouse/human CD45R/B220 Antibody

Catalog# / Size	103254 / 100 µg
Clone	RA3-6B2
Regulatory Status	RUO
Other Names	B220
Isotype	Rat IgG2a, κ
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.

Product Details

Verified Reactivity	Mouse, Human
Reported Reactivity	Cat
Antibody Type	Monoclonal
Host Species	Rat
Immunogen	Abelson murine leukemia virus-induced pre-B tumor cells
Formulation	Phosphate-buffered solution, pH 7.2, containing 0.09% sodium azide.
Preparation	The antibody was purified by affinity chromatography and conjugated with Alexa Fluor® 594 under optimal conditions.
Concentration	0.5 mg/mL
Storage & Handling	The antibody solution should be stored undiluted between 2°C and 8°C, and protected from prolonged exposure to light. **Do not freeze.**
Application	IHC-F - *Quality tested* FC, 3D IHC - *Verified*
Recommended Usage	Each lot of this antibody is quality control tested by immunofluorescence staining. For immunohistochemistry, a concentration range of 2.5-5 µg/mL is recommended. For immunofluorescence microscopy, a concentration range of 1.25 - 10 µg/mL is recommended. For flow cytometric staining, the suggested use of this reagent is ≤0.25 µg per million cells in 100 µL volume. For 3D immunohistochemistry on formalin-fixed tissues, a concentration of 5.0 µg/mL is suggested. It is recommended that the reagent be titrated for optimal performance for each application. * Alexa Fluor® 594 has an excitation maximum of 590 nm, and a maximum emission of 617 nm. Alexa Fluor® and Pacific Blue™ are trademarks of Life Technologies Corporation. View full statement regarding label licenses
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: immunoprecipitation[1], *in vitro* and *in vivo* modulation of B cell responses[2-4], immunohistochemistry of acetone-fixed frozen sections and formalin-fixed paraffin-embedded sections[5,6], and spatial biology (IBEX)[14,15].
Application References	

(PubMed link indicates BioLegend citation)

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)
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6. Monteith CE, *et al.* 1996. *Can. J. Vet. Res.* 60:193. (IHC)
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13. Murakami R, *et al.* 2013. *PLoS One.* 8:73270. PubMed
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Product Citations

1. Baptista AP *et al.* 2019. Immunity. 50(5):1188-1201 . PubMed
2. Li S, *et al.* 2020. Cell Death Dis. 0.9875. PubMed
3. Pham THM, *et al.* 2020. Cell Host & Microbe. 27(1):54-67.e5.. PubMed
4. Counoupas C, *et al.* 2020. NPJ Vaccines. 0.28125. PubMed
5. Matryba P, *et al.* 2020. J Immunol. 1395:204. PubMed
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7. Kobia FM, *et al.* 2020. PLoS Biol. 18:e3000850. PubMed
8. Luo L, *et al.* 2021. J Neuroinflammation. 18:27. PubMed
9. Shi H *et al.* 2018. Immunity. 49(5):899-914 . PubMed
10. Song W, *et al.* 2022. Immunity. 55:290. PubMed
11. Goldberg MF *et al.* 2018. Immunity. 49(6):1090-1102 . PubMed
12. Kuhn NF *et al.* 2019. Cancer cell. 35(3):473-488 . PubMed
13. Bidet K, *et al.* 2019. NPJ Vaccines. 4:27. PubMed

RRID

AB_2563229 (BioLegend Cat. No. 103254)

Antigen Details

Structure

Protein tyrosine phosphatase (PTP) family, 180-240 kD

Distribution

B cells, T cell subset, NK cell subset

Function

Phosphatase, T and B cell activation

Ligand/Receptor

Galectin-1, CD2, CD3, CD4

Cell Type

B cells, NK cells, T cells

Biology Area

Cell Biology, Immunology, Inhibitory Molecules, Neuroscience, Neuroscience Cell Markers

Molecular Family

CD Molecules

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.

Gene ID

19264

5788

Related Protocols

Immunohistochemistry Protocol for Frozen Sections

Cell Surface Flow Cytometry Staining Protocol

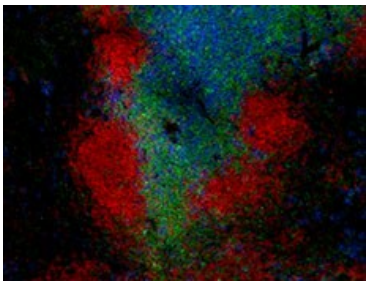
Immunocytochemistry Staining Protocol

Ce3D™ Tissue Clearing Kit

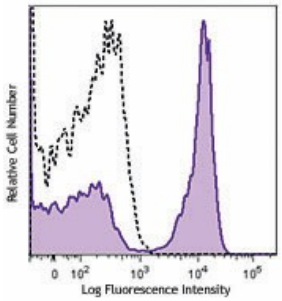
Other Formats

Alexa Fluor® 594 anti-mouse/human CD45R/B220, APC anti-mouse/human CD45R/B220, Biotin anti-mouse/human CD45R/B220, FITC anti-mouse/human CD45R/B220, PE anti-mouse/human CD45R/B220, PE/Cyanine5 anti-mouse/human CD45R/B220, Purified anti-mouse/human CD45R/B220, PE/Cyanine7 anti-mouse/human CD45R/B220, APC/Cyanine7 anti-mouse/human CD45R/B220, Alexa Fluor® 488 anti-mouse/human CD45R/B220, Alexa Fluor® 647 anti-mouse/human CD45R/B220, Pacific Blue™ anti-mouse/human CD45R/B220, Alexa Fluor® 700 anti-mouse/human CD45R/B220, PerCP anti-mouse/human CD45R/B220, PerCP/Cyanine5.5 anti-mouse/human CD45R/B220, Brilliant Violet 421™ anti-mouse/human CD45R/B220, Brilliant Violet 570™ anti-mouse/human CD45R/B220, Brilliant Violet 650™ anti-mouse/human CD45R/B220, Brilliant Violet 605™ anti-mouse/human CD45R/B220, Brilliant Violet 785™ anti-mouse/human CD45R/B220, Brilliant Violet 510™ anti-mouse/human CD45R/B220, Purified anti-mouse/human CD45R/B220 (Maxpar® Ready), Brilliant Violet 711™ anti-mouse/human CD45R/B220, PE/Dazzle™ 594 anti-mouse/human CD45R/B220, APC/Fire™ 750 anti-mouse/human CD45R/B220, Brilliant Violet 750™ anti-mouse/human CD45R/B220, TotalSeq™-A0103 anti-mouse/human CD45R/B220, Spark Blue™ 550 anti-mouse/human CD45R/B220, Spark NIR™ 685 anti-mouse/human CD45R/B220, TotalSeq™-B0103 anti-mouse/human CD45R/B220, Ultra-LEAF™ Purified anti-mouse/human CD45R/B220, TotalSeq™-C0103 anti-mouse/human CD45R/B220, PE/Fire™ 640 anti-mouse/human CD45R/B220, APC/Fire™ 810 anti-mouse/human CD45R/B220, PE/Fire™ 700 anti-mouse/human CD45R/B220, Spark Violet™ 538 anti-mouse/human CD45R/B220, Spark YG™ 581 anti-mouse/human CD45R/B220, Spark YG™ 570 anti-mouse/human CD45R/B220, PE/Fire™ 810 anti-mouse/human CD45R/B220, Spark Blue™ 574 anti-mouse/human CD45R/B220 Antibody, Spark Violet™ 423 anti-mouse/human CD45R/B220 Antibody, Spark Red™ 718 anti-mouse/human CD45R/B220

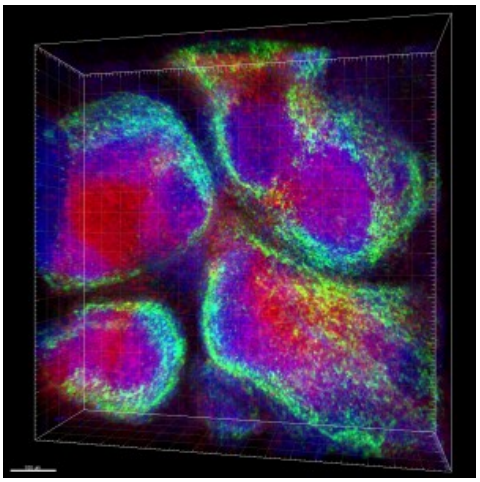
Product Data



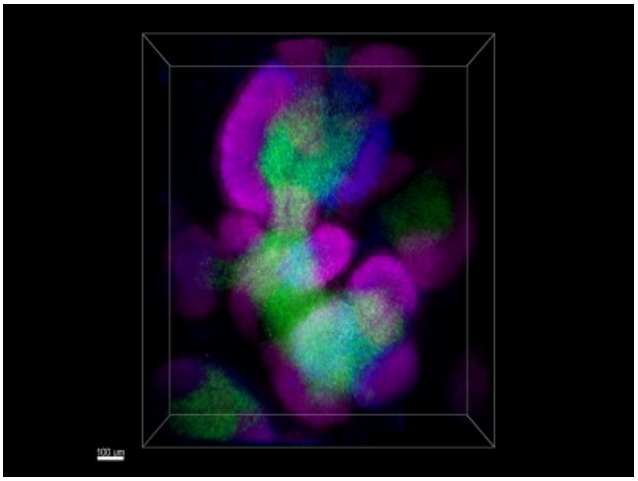
C57BL/6 mouse frozen lymph node section was fixed with 4% paraformaldehyde (PFA) for 10 minutes at room temperature and blocked with 5% FBS plus 5% rat serum for 1 hour at room temperature. Then the section was stained with 2.5 µg/mL of B220 (clone RA3-6B2) Alexa Fluor® 594 (red), 2.5 µg/mL of CD4 (clone RM4-5) Alexa Fluor® 488 (green), and 2.5 µg/mL of CD8 (clone 53-6.7) Alexa Fluor® 647 (blue) overnight at 4°C. The image was captured by 10X objective.



C57BL/6 mouse splenocytes were stained with CD45R/B220 (clone RA3-6B2) Alexa Fluor® 594 (filled histogram) or rat IgG2a, κ Alexa Fluor® 594 isotype control (open histogram). The data was acquired by BD LSRFortessa™ cell analyzer equipped with Yellow-Green Laser (561 nm).



Formalin-fixed, 300 micron-thick mouse spleen section was blocked, permeabilized and stained overnight with CD4 (clone RM4-5) Alexa Fluor® 647 (red), CD169 (Siglec-1)(clone 3D6.112) Alexa Fluor® 488 (green), and CD45R/B220 (clone RA3-6B2) Alexa Fluor® 594 (blue) all at 5 µg/mL, optically cleared, then analyzed at 220 µm imaging depth on a confocal microscope. Watch the video.



Paraformaldehyde-fixed (4%), 500 µm-thick mouse spleen section was processed according to the Ce3DTM Tissue Clearing Kit protocol (Cat. No. 427701). The section was costained with anti-mouse CD3ε Antibody (clone 500A2) Alexa Fluor® 488 at 5 µg/mL (green), and anti-mouse/human CD45R/B220 Antibody (clone RA3-6B2) Alexa Fluor® 594 at 5 µg/mL (magenta) and counterstained with DAPI (blue). The section was then optically cleared and mounted in a sample chamber. The image was captured with a 10X objective using Zeiss 780 confocal microscope and processed by Imaris image analysis software.
Watch the video.

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