

CD45R (B220) Monoclonal Antibody (RA3-6B2), Alexa Fluor™ 561, eBioscience™

Product Details	
Size	100 µg
Species Reactivity	Human, Mouse
Host/Isotype	Rat / IgG2a, kappa
Recommended Isotype Control	Rat IgG2a kappa Isotype Control (eBR2a), Alexa Fluor™ 561, eBioscience™
Class	Monoclonal
Type	Antibody
Clone	RA3-6B2
Conjugate	Alexa Fluor™ 561
Excitation/Emission Max	556/569 nm
Form	Liquid
Concentration	0.2 mg/mL
Purification	Affinity chromatography
Storage buffer	PBS, pH 7.2
Contains	0.09% sodium azide
Storage conditions	4° C, store in dark, DO NOT FREEZE!
RRID	AB_2896201

Applications	Tested Dilution	Publications
Immunohistochemistry (Paraffin) (IHC (P))	1:100	-
Flow Cytometry (Flow)	0.5 µg/test	-

Product Specific Information

Description: The RA3-6B2 monoclonal antibody reacts with exon A-restricted isoform of mouse CD45, a 220 kDa surface molecule. CD45R/B220 epitope is mainly expressed by the B cell lineage from early Pro-B to mature B cells. However, some activated T cells, lymphokine activated killer cells (LAK), NK cell progenitors in the bone marrow, and T cells of the lpr/lpr mutant mouse also express this antigen.

Applications Reported: This RA3-6B2 antibody has been reported for use in flow cytometric analysis.

Our internal testing shows that Alexa Fluor 561 non-specifically stains B cells in Swiss Webster and SJL mice. Non-specific staining has not been observed in BALB/c or C57BL/6 mice. Other strains have not been tested. See the Antibody Testing Data for an example of this strain-dependent difference.

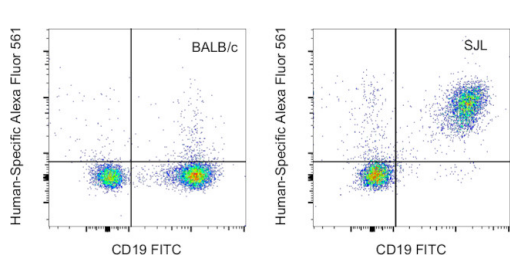
Applications Tested: This RA3-6B2 antibody has been tested by flow cytometric analysis of mouse splenocytes. This may be used at less than or equal to 0.5 µg per test. A test is defined as the amount (µg) of antibody that will stain a cell sample in a final volume of 100 µL. Cell number should be determined empirically but can range from 10⁵ to 10⁸ cells/test. It is recommended that the antibody be carefully titrated for optimal performance in the assay of interest.

Alexa Fluor 561 emits at 575 nm and is intended for use on spectral cytometers where it may be multiplexed with both PE and

PE-eFluor 610.

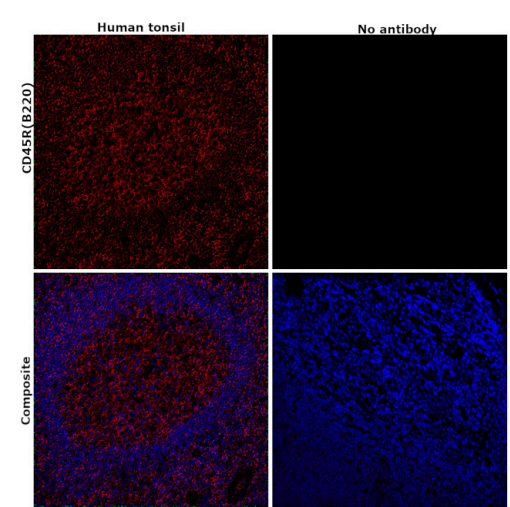
Excitation: 558 nm; Emission: 575 nm; Laser: Yellow-Green Laser

Product Images For CD45R (B220) Monoclonal Antibody (RA3-6B2), Alexa Fluor™ 561, eBioscience™



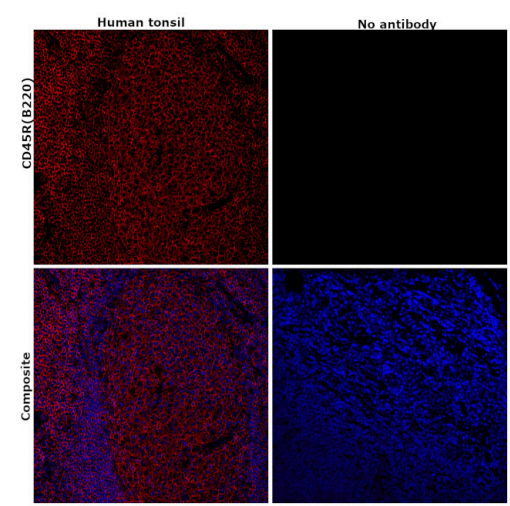
CD45R (B220) Antibody (505-0452-82) in Flow

Alexa Fluor 561 non-specific staining of B cells in the SJL strain of mice. Splenocytes from BALB/c (left) and SJL (right) strains of mice were stained with Anti-Mouse CD19 Monoclonal Antibody conjugated to FITC and a non-cross-reactive, human-specific monoclonal antibody conjugated to Alexa Fluor 561. These data show that Alexa Fluor 561-conjugated antibodies non-specifically stain B cells in SJL mice (right) and outbred, Swiss Webster mice (data not shown). Non-specific staining has not been observed in BALB/c mice (left) and C57BL/6 mice (data not shown).



CD45R (B220) Antibody (505-0452-82) in IHC (P)

Immunohistochemical analysis of CD45R (B220) was performed using formalin-fixed paraffin-embedded human tonsil tissue sections. To expose the target protein, heat-induced epitope retrieval was performed on de-paraffinized sections using eBioscience™ IHC Antigen Retrieval Solution - High pH (10X) (Product # 00-4956-58) diluted to 1X solution in water in a decloaking chamber at 110 degree Celsius for 15 minutes. Following antigen retrieval, the sections were blocked with 2% normal goat serum in 1X PBS for 45 minutes at room temperature and then probed with or without CD45R (B220) Monoclonal Antibody (RA3-6B2), Alexa Fluor™ 561, eBioscience™ (Product # 505-0452-82) at 1:100 dilution in 0.1% normal goat serum overnight at 4 degree Celsius in a humidified chamber. ReadyProbes™ Tissue Autofluorescence Quenching Kit (Product # R37630) was used to quench autofluorescence from the tissues. Nuclei were stained with DAPI (Product # D1306) and the sections were mounted using ProLong™ Glass Antifade Mountant (Product # P36984). The images were captured on EVOS™ M7000 Imaging System (Product # AMF7000) at 20X magnification and externally deconvoluted.



CD45R (B220) Antibody (505-0452-82) in IHC (P)

Immunohistochemical analysis of CD45R (B220) was performed using formalin-fixed paraffin-embedded human tonsil tissue sections. To expose the target protein, heat-induced epitope retrieval was performed on de-paraffinized sections using eBioscience™ IHC Antigen Retrieval Solution - Low pH (10X) (Product # 00-4955-58) diluted to 1X solution in water in a decloaking chamber at 110 degree Celsius for 15 minutes. Following antigen retrieval, the sections were blocked with 2% normal goat serum in 1X PBS for 45 minutes at room temperature and then probed with or without CD45R (B220) Monoclonal Antibody (RA3-6B2), Alexa Fluor™ 561, eBioscience™ (Product # 505-0452-82) at 1:100 dilution in 0.1% normal goat serum overnight at 4 degree Celsius in a humidified chamber. ReadyProbes™ Tissue Autofluorescence Quenching Kit (Product # R37630) was used to quench autofluorescence from the tissues. Nuclei were stained with DAPI (Product # D1306) and the sections were mounted using ProLong™ Glass Antifade Mountant (Product # P36984). The images were captured on EVOS™ M7000 Imaging System (Product # AMF7000) at 20X magnification and externally deconvoluted.

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