Monoclonal Anti-mouse CD161/KLRB-1C-Alexa Fluor® 488

Catalog Number: FAB7614G
Lot Number: ACYW01
100 Tests

Reagents Provided

Alexa Fluor® 488-conjugated rat monoclonal anti-mouse CD161/KLRB-1C: Supplied as 25 μg of antibody in 0.5 mL saline containing up to 0.5% BSA and 0.1% sodium azide.

Clone #: 694370

Isotype: rat IgG_{2A}

Reagents Not Provided

- Flow Cytometry Staining Buffer (Catalog # FC001) or other BSA-supplemented saline buffer.

Storage

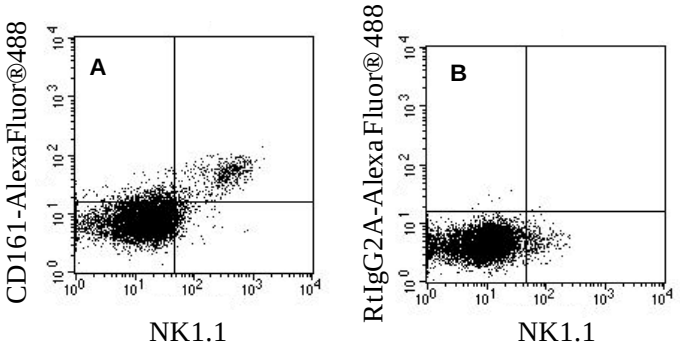
Reagents are stable for **twelve months** from the date of receipt when stored in the dark at 2-8 °C.

Intended Use

Designed to quantitatively determine the percentage of cells bearing CD161/KLRB-1C within a population and qualitatively determine the density of CD161/KLRB-1C on cell surfaces by flow cytometry.

Product Description

This antibody was produced from a hybridoma resulting from the fusion of a mouse myeloma with B cells obtained from a rat immunized with purified NS0-derived recombinant mouse CD161/KLRB-1C (rmKLRB-1C, aa 66-220). The IgG fraction of the tissue culture supernatant was purified by Protein A or G affinity chromatography. The purified antibody was then conjugated to Alexa Fluor® 488 fluorochrome. Cell surface expression of CD161/KLRB-1C is determined by flow cytometry using 488 nm wavelength excitation and monitoring emitted fluorescence with a detector optimized to collect peak emissions at 515-545 nm.



C57/B6 mouse splenocytes were stained with APC-conjugated anti-mouse NK1.1 (Clone PK136) and A) Alexa Fluor® 488-conjugated anti-mouse CD161/KLRB-1C (Catalog # FAB7614G) or Alexa Fluor® 488-conjugated isotype control (Catalog # IC006G).

Background Information

CD161, also known as NK1.1, NKR-P1A, and KLRB1 is a 40 kDa type II transmembrane glycoprotein that contains one C-type lectin domain in its extracellular region. CD161 is expressed as a disulfide-linked dimer on the surface of Th17 cells and natural killer (NK) cells, as well as on subsets of CD1-restricted T cells, intestinal NT cells, peripheral memory T cells, monocytes, and dendritic cells. It binds to OCIL/CLEC2d, leading to an inhibition of NK cell-mediated cytotoxicity and IFN-γ secretion. Alternatively, CD161 can enhance TCR activation by CD1d ligation. $CD161^+$ cell populations are depleted in ulcerative colitis, Grave's disease, and AIDS, although $CD161^+$ T cells are activated during asthmatic attacks. Additional related proteins are expressed in mouse but not human: the inhibitory NKR-P1B and D, and the stimulatory NKR-P1A, C, and F. In competition studies, clone 694370 inhibited binding of the NK1.1 clone PK136, indicating an overlap of antibody epitopes between the two clones.

Flow Cytometry Validation

This antibody has been tested for flow cytometry using C57/B6 mouse splenocytes.

1. Cells may be Fc-blocked with 1 μg of mouse IgG/10^5 cells for 15 minutes at room temperature. Do not wash excess blocking IgG from this reaction.
2. After blocking, 5 μL of conjugated antibody was added to up to 1 x 10^6 cells and incubated for 30 minutes at room temperature.
3. Unbound antibody was removed by washing the cells twice in Flow Cytometry Staining Buffer (Catalog # FC001). Note that whole blood requires a RBC lysis step at this point using Flow Cytometry Mouse Lyse Buffer (Catalog # FC003).
4. The cells were resuspended in Flow Cytometry Staining Buffer for final flow cytometric analysis. As a control for this analysis, cells in a separate tube should be treated with Alexa Fluor® 488-labeled rat IgG_{2A} antibody. This procedure may need to be modified, depending upon the cell type and final utilization. Individual users may need to titrate to determine the optimal reagent amount for their specific use.

Warning: Contains sodium azide as a preservative - sodium azide may react with lead and copper plumbing to form explosive metal azides. Flush with large volumes of water during disposal.

Legal

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