Monoclonal Anti-human CD21-Phycoerythrin

Catalog Number: FAB4909P
Lot Number: ABCU01
100 Tests

Reagents Provided

Phycoerythrin (PE)-conjugated mouse monoclonal anti-human CD21: Supplied as 25 μg of antibody in 1 mL saline containing up to 0.5% BSA and 0.09% sodium azide.

Clone #: 544408

Isotype: mouse IgG_1

Reagents Not Provided

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

Storage

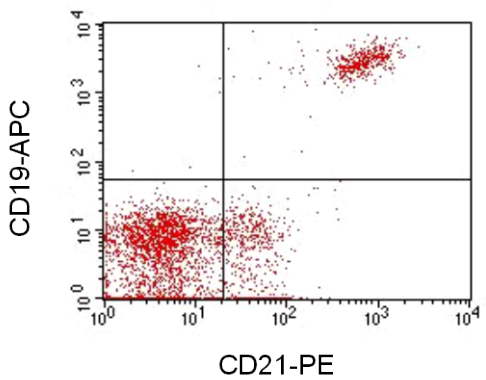
Reagents are stable for **twelve months** from date of receipt when stored in the dark at 2-8 °C.

Intended Use

Designed to quantitatively determine the percentage of cells bearing CD21 within a population and qualitatively determine the density of CD21 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 mouse immunized with purified, CHO-derived, recombinant human CD21 extracellular domain (aa 21-971). The IgG fraction of the tissue culture supernatant was purified by Protein G affinity chromatography. The purified antibody was then conjugated to PE fluorochrome. Cell surface expression of CD21 is determined by flow cytometry using 488 nm wavelength excitation and monitoring emitted fluorescence with a detector optimized to collect peak emissions at 565-605 nm.



PBMC lymphocytes were stained with PE-conjugated anti-human CD21 (Catalog # FAB4909P) and APC-conjugated anti-human CD19 (Catalog # FAB4867A). Quadrant markers were set based on isotype control staining (Catalog # IC002P and IC002A).

Background Information

CD21 (also EBV receptor and CR2) is a 145 kDa member of the RCA (receptors of complement activation) family of proteins. It is expressed on T cells, B cells, and follicular dendritic cells. On the B cell surface, it combines with the BCR and CD19 to form a B cell-activating complex. Mature human CD21 is 1013 amino acids (aa) in length. It is a type I transmembrane (TM) protein that contains a 951 aa extracellular domain (ECD; aa 21-971) and a short 34 aa cytoplasmic tail. The ECD exhibits fifteen 60 aa SUSHI repeats. Soluble CD21 is apparently generated by cleavage near the TM domain. One potential splice variant shows a deletion of aa 847-908, while another shows an insertion of 59 aa after Lys659. The ECD of human CD21 is 71% aa identical to the ECD of mouse CD21.

Flow Cytometry Validation

This antibody has been tested for flow cytometry using PBMC lymphocytes.

1. Cells may be Fc-blocked with 1 μg of human IgG/10^5 cells for 15 minutes at room temperature. Do not wash excess blocking IgG from this reaction.
2. After blocking, 10 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 Human Lyse Buffer (Catalog # FC002).
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 PE-labeled mouse IgG_1 antibody. This procedure may need to be modified, depending upon 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.

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