

Technical Data Sheet

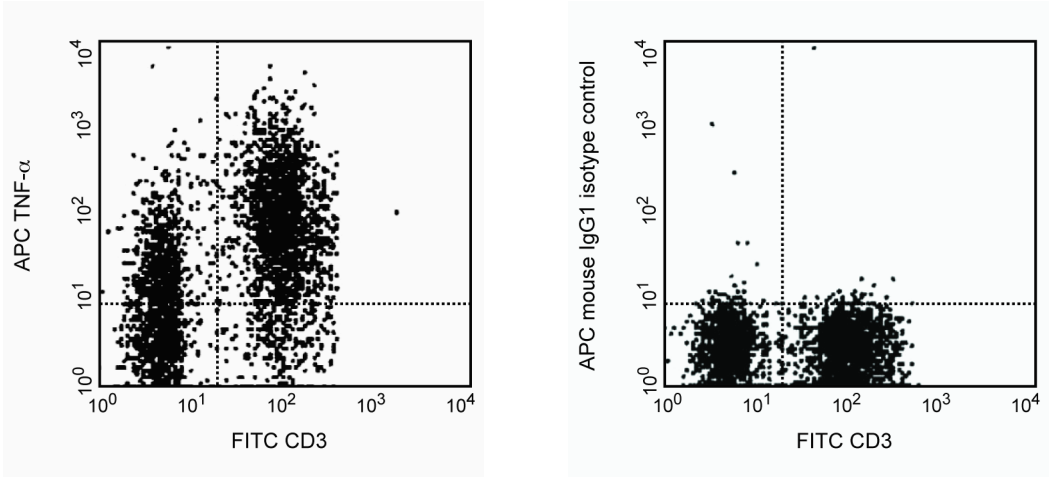
APC Mouse IgG1 κ Isotype Control

Product Information

Material Number:	554681
Size:	0.1 mg
Concentration:	0.2 mg/ml
Clone:	MOPC-21
Isotype:	Mouse IgG1, κ
Storage Buffer:	Aqueous buffered solution containing ≤0.09% sodium azide.

Description

The MOPC-21 immunoglobulin is a mouse myeloma protein. The MOPC-21 immunoglobulin was selected as an isotype control following screening for low background on a variety of mouse and human tissues.



Expression of TNF by stimulated human peripheral blood mononuclear cells (PBMC). Human PBMC were stimulated for 6 hours with PMA (50 ng/ml final concentration; Sigma) and calcium ionophore A23187 (1 µg/ml final concentration; Sigma), in the presence of GolgiStop™ (2 µM final concentration; Cat. No. 554724). The PMBC were stained with FITC-anti-CD3 (FITC-UCHT1, Cat. No. 555332), fixed, permeabilized, and subsequently stained with 0.125 µg of APC-mouse anti-human TNF (APC-MAb11, Cat. No. 554514, left panel) or with 0.125 µg APC-mouse IgG1 (Cat. No. 554681, right panel) using BD Pharmingen™ staining protocol. To demonstrate specificity, the binding of APC-MAb11 antibody was blocked by perincubation of fixed/permeabilized cells with excess unlabelled MAb11 antibody (5 µg; Cat. No. 554510). The quadrant markers for the bivariate dot plot were set based on isotype controls and verified using the unlabelled MAb11 antibody blocking control. The APC-conjugated reagent can be used in any flow cytometer equipped with a dye, HeNe or red diode laser. These include the dual laser FACStarPLUS™, FACSVantage™, or FACSCalibur™.

Preparation and Storage

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography. The antibody was conjugated to APC under optimum conditions, and unconjugated antibody and free APC were removed. Store undiluted at 4°C and protected from prolonged exposure to light. Do not freeze.

Application Notes

Application

Intracellular staining (flow cytometry)	Routinely Tested
Isotype control	Routinely Tested

Recommended Assay Procedure:

Immunofluorescent Staining and Flow Cytometric Analysis: The APC-MOPC-21 immunoglobulin (Cat. No. 554681) is a suitable mouse IgG1 isotype control for assessing the level of background staining on paraformaldehyde fixed/saponin-permeabilized mouse or human cells for flow cytometric analysis. Use at comparable concentrations to antibody of interest (e.g., ≤ 0.5 µg mAb/1 million cells) (see figure). For specific methodology, visit the protocols section of our website, or refer to the Immune Function Handbook, which is posted on our web site at www.bdbiosciences.com

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Product Notices

1. Please refer to www.bdbiosciences.com/pharming/en/protocols for technical protocols.
2. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
3. For fluorochrome spectra and suitable instrument settings, please refer to our Fluorochrome Web Page at www.bdbiosciences.com/colors.
4. This APC-conjugated reagent can be used in any flow cytometer equipped with a dye, HeNe, or red diode laser.
5. Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before discarding to avoid accumulation of potentially explosive deposits in plumbing.

References

Prussin C, Metcalfe DD. Detection of intracytoplasmic cytokine using flow cytometry and directly conjugated anti-cytokine antibodies. *J Immunol Methods*. 1995; 188(1):117-128. (Methodology)