Technical Data Sheet

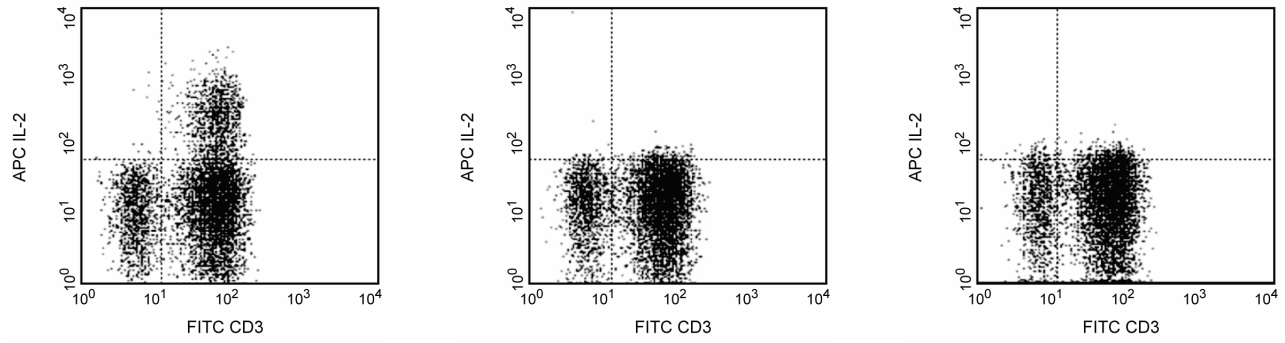
APC Rat Anti-Human IL-2

Product Information

Material Number:	**561054**
Size:	25 µg
Concentration:	0.2 mg/ml
Clone:	MQ1-17H12
Immunogen:	Human IL-2 Recombinant Protein
Isotype:	Rat IgG2a, κ
Reactivity:	QC Testing: Human
Storage Buffer:	Aqueous buffered solution containing ≤0.09% sodium azide.

Description

The MQ1-17H12 antibody reacts with human interleukin-2 (IL-2). The immunogen used to generate the MQ1-17H12 hybridoma was recombinant human IL-2. Unconjugated or purified forms of this antibody have been reported to be neutralizing for human IL-2 bioactivity.



Expression of IL-2 by stimulated CD3+ and CD3-human PBMC. *Human PBMC were stimulated for 6 h with PMA (Sigma, Cat. #P-8139) and calcium ionophore A23187 (Sigma, Cat. #C-9275), in the presence of GolgiStop™ (2 µM final concentration; (Cat. No. 554724). The PBMC were stained with FITC-anti-CD3 (FITC-UCHT1, Cat. No. 555332), fixed, permeabilized, and then stained with 0.25 µg of APC-rat anti-human IL-2 antibody (APC-MQ1-17H12, Cat. No. 554567) using The BD Pharmingen™ staining protocol (left panel). To demonstrate specificity of staining, the binding of APC-MQ1-17H12 was blocked by the preincubation of the conjugated antibody with excess recombinant human IL-2 (1.0 µg, Cat. No. 554603; middle panel), and by preincubation of the fixed/permeabilized cells with an excess of the unlabelled MQ1-17H12 antibody (10 µg, Cat. No. 554563; right panel) prior to staining with the APC-MQ1-17H12 antibody. The quadrant markers for the bivariate dot plots were set based on the autofluorescence control, and verified with the recombinant cytokine blocking (middle panel) and unlabelled antibody blocking specificity controls (right panel). This APC-conjugated reagent can be used in any flow cytometer equipped with a a dye, HeNE or red diode laser. These include the dual laser FACStarPLUS™, FACS Vantage™ or FACSCalibur™.*

Preparation and Storage

Store undiluted at 4°C and protected from prolonged exposure to light. Do not freeze.

The antibody was conjugated to APC under optimum conditions, and unconjugated antibody and free APC were removed.

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.

Application Notes

Application

Intracellular staining (flow cytometry)	Routinely Tested

Recommended Assay Procedure:

Immunofluorescent Staining and Flow Cytometric Analysis: The APC-conjugated MQ1-17H12 antibody (Cat. No. 554567) can be used for multicolor immunofluorescent staining and flow cytometric analysis to identify and enumerate IL-2-producing cells within mixed cell populations (see image). For optimal immunofluorescent staining with flow cytometric analysis, this anti-cytokine antibody should be titrated (≤ 0.5 µg mAb/1X10^6 cells). For specific methodology, please visit the protocols section on ELISA posted on our web site, www.bdbiosciences.com.

A useful control for demonstrating specificity of staining is either of the following: 1) pre-block the conjugated MQ1-17H12 antibody with a molar excess of ligand (e.g., recombinant human IL-2; Cat No. 554603) prior to staining, or 2) pre-block the fixed/permeabilized cells with unlabelled MQ117H12 antibody (Cat. No. 554563) prior to staining. A suitable rat IgG2a isotype control for assessing the level of background staining on paraformaldehyde-fixed/saponin-permeabilized human cells is APC-R35-95 (Cat. No. 554690); use at comparable concentrations to antibody of interest (e.g., ≤ 0.5 µg mAb/1X10^6 cells).

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Neutralization/Blocking: The NA/LE™ format of the MQ1-17H12 antibody (Cat. No. 554567) is useful for neutralization of human IL-2 bioactivity. A suitable NA/LE™ rat IgG2a isotype control to match the NA/LE™ MQ1-17H12 antibody is the R35-95 antibody, Cat. No. 554687.

Suggested Companion Products

Catalog Number	Name	Size	Clone
554603	Recombinant Human IL-2	10 µg	(none)
554563	Purified Rat Anti-Human IL-2	0.1 mg	MQ1-17H12
554690	APC Rat IgG2a κ Isotype Control	0.1 mg	R35-95
554724	Protein Transport Inhibitor (Containing Monensin)	0.7 ml	(none)

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
3. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
4. 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.
5. This product is manufactured and sold under license from Pestka Biomedical Laboratories, Inc. (d/b/a PBL InterferonSource) and may be used solely as indicated. This product may not be resold or incorporated in any manner into another product for resale. Any use for therapeutics is strictly prohibited. This product is covered by U.S. Patent No. 5,597,901 and Bulgarian Patent No. BG1895.
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References

Abrams J. Immunoenzymetric assay of mouse and human cytokines using NIP-labeled anti-cytokine antibodies. In: Coligan J, Kruisbeek A, Margulies D, Shevach E, Strober W, ed. *Current Protocols in Immunology.* New York: John Wiley and Sons; 1995:6.20-6.21. (Biology)

Abrams JS, Roncarolo MG, Yssel H, Andersson U, Gleich GJ, Silver JE. Strategies of anti-cytokine monoclonal antibody development: immunoassay of IL-10 and IL-5 in clinical samples. *Immunol Rev.* 1992; 127:5-24. (Biology)

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