Technical Data Sheet

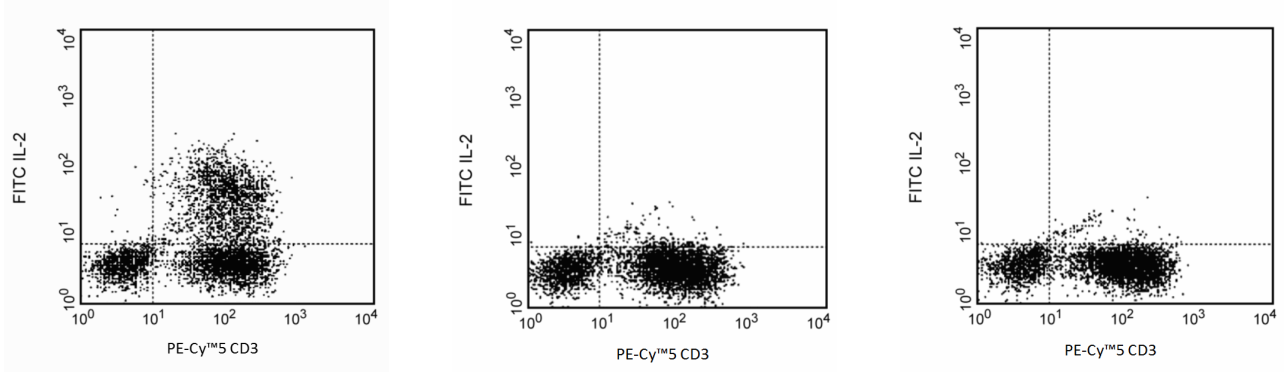
FITC Rat Anti-Human IL-2

Product Information

Material Number:	**561055**
Size:	25 µg
Concentration:	0.5 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+ human PBMC. *Human PBMC were stimulated for 6 hours with PMA (Sigma, Cat. #P-8139) and calcium ionophore A23187 (Sigma, Cat. #C-9275) in the presence of 2 µM GolgiStop™ (Cat. No. 554724). The PBMC's were stained with PE-Cy™5 anti-CD3 (Cat. No. 555334), fixed, permeabilized, and then stained with 0.25 µg of FITC-rat anti-human IL-2 antibody (Cat. No. 554565) (left panel). To demonstrate specificity of staining, the binding of FITC-MQ1-17H12 (Cat. No. 561055/554565) was blocked by the preincubation of the conjugated antibody with a molar excess of recombinant human IL-2 protein (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 FITC-MQ1-17H12 antibody. The quadrant markers for the bivariate dot plots were set based on the autofluorescence control, and verified with the blocking controls.*

Preparation and Storage

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

Application Notes

Application

Intracellular staining (flow cytometry)	Routinely Tested

Recommended Assay Procedure:

Immunofluorescent Staining and Flow Cytometry: The FITC-conjugated MQ1-17H12 antibody 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 for flow cytometric analysis, this anti-cytokine antibody should be titrated (≤ 0.5 µg mAb/1X10^6 cells) For specific methodology, please visit our web site, http://www.bdbiosciences.com/resources/index.jsp

A useful control for demonstrating specificity of staining is either of the following: 1) pre-block the conjugated MQ1-17H12 antibody with its ligand (e.g., recombinant human IL-2; Cat. No. 554603) prior to staining, or 2) pre-block the fixed/permeabilized cells with unlabelled MQ1-17H12 antibody (Cat. No. 554563) prior to staining. The intracellular cytokine staining technique and use of blocking controls are described in detail by C. Prussin and D. Metcalfe. A suitable rat IgG2a isotype control for assessing the level of background staining on paraformaldehyde-fixed/saponin-permeabilized mouse or human cells is FITC-R35-95 (Cat. No. 554688); use at comparable concentrations to the antibody of interest (e.g., ≤ 0.5 µg mAb/1X10^6 cells).

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Neutralization/Blocking: The NA/LE (Cat. No. 554562) format of the MQ1-17H12 antibody is useful for neutralization of human IL-2 bioactivity. A suitable NA/LE rat IgG2aisotype control is R35-95, Cat. No. 554687.

Suggested Companion Products

Catalog Number	Name	Size	Clone
554563	Purified Rat Anti-Human IL-2	0.1 mg	MQ1-17H12
554603	Recombinant Human IL-2	10 µg	(none)
554688	FITC Rat IgG2a, κ Isotype Control	0.1 mg	R35-95
554724	Protein Transport Inhibitor (Containing Monensin)	0.7 ml	(none)
555334	PE-Cy™5 Mouse Anti-Human CD3	100 tests	UCHT1
554562	Purified NA/LE Rat Anti-Human IL-2	0.5 mg	MQ1-17H12
554687	Purified NA/LE Rat IgG2a κ Isotype Control	0.5 mg	R35-95
554565	FITC Rat Anti-Human IL-2	0.1 mg	MQ1-17H12

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. 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.
3. 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.
4. All other brands are trademarks of their respective owners.
5. Cy is a trademark of Amersham Biosciences Limited.
6. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.

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)

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: Flow cytometry)

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