# **Technical Data Sheet**

# **Purified Rat Anti-Mouse CD124**

### **Product Information**

551853 **Material Number:** 

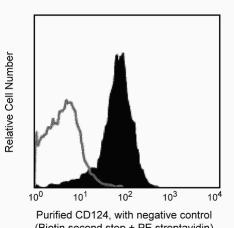
Alternate Name: IL-4 Receptor α chain

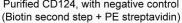
0.1 mg **Concentration:** 0.5 mg/mlClone: mIL4R-M1 Rat IgG2a, ĸ Isotype: QC Testing: Mouse Reactivity:

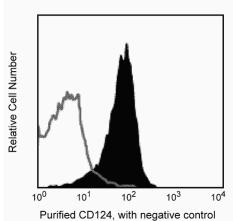
Storage Buffer: Aqueous buffered solution containing ≤0.09% sodium azide.

## Description

The mIL4R-M1 monoclonal antibody specifically binds to CD124 which is also known as the α subunit of the mouse Interleukin-4 Receptor (IL-4R $\alpha$ ). The mouse IL-4R $\alpha$  is a 140 kDa transmembrane glycoprotein that is expressed by B and T lymphocytes and a variety of other hematopoietic and nonhematopoietic cells and cell lines. The cell surface IL-4Ra chain binds IL-4 with high affinity and associates with either the common  $\gamma$  chain (IL-4R $\alpha/\gamma c$ ; aka, type I IL-4R) or the IL-13 receptor alpha subunit (IL-4R $\alpha$ /IL-13R $\alpha$ ; aka, type II IL-4R complex) to form two distinct types of signal-transducing IL-4R complexes. The type I IL-4 receptor complex specifically binds IL-4 whereas the type II IL-4R binds and transduces signals from either IL-4 or IL-13. The mIL4R-M1 antibody blocks IL-4 binding to cells and is reported to be a potent inhibitor of IL-4's biological activities. The mIL4R-M1 antibody also recognizes naturally-occurring, soluble truncated forms of IL-4Rα (sIL-4R) that result either from enzymatic cleavage of the cell surface extracellular IL-4Rα domain or from differential mRNAsplicing and secretion by cells. These sIL-4R retain their high-affinity ligand binding domain and appear to either enhance or inhibit IL-4-mediated functions depending on the relative local levels of IL-4 and sIL-4R.







(Biotin second step + PE streptavidin)

Expression of cell surface CD124 by normal splenocyres from BALB/c and C57BL/6 mice. Spleen cells from BALB/c (left panel) and C57BL/6 (right panel) mice were treated with ACK lysis buffer, were washed, and were labeled with purified Fc Block (Cat. No. 553142; Rat IgG2b anti-mouse CD16/CD32) to block mouse Fc receptors. The cells were then stained with mlL4R-M1 (1 μg) followed by biotinylated RG7/1.30 (mouse anti-rat IgG2α; 0.25 μg, Cat. No. 553894) and streptavidin phycoerythrin (0.015 μg, Cat. No. 554061) in a three-layer staining protocol to amplify immunofluorescent signals. Staining with the mlL4R-M1 antibody (filled histograms) is compared to staining with the secondary reagents alone (open histograms)

## **Preparation and Storage**

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography. Store undiluted at 4° C.

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# **Application Notes**

### Application

Flow cytometry	Routinely Tested
ELISA	Tested During Development
Immunoprecipitation	Reported
Neutralization	Reported

## **Recommended Assay Procedure:**

Immunofluorescent Staining and Flow Cytometric Analysis: The purified form of the mIL4R-M1 antibody can be used for the immunofluorescent staining ( $\leq 1 \mu g$  antibody/10e6 cells) and flow cytometric analysis of nucleated mouse cells to measure their expressed levels of surface CD124. An appropriate purified Ig isotype control is clone R35-95 (Cat. No. 553927). A three-layer staining protocol is recommended for maximizing the detection of IL-4R $\alpha$  expressed by cells as detailed in the figure legend.

**ELISA:** The purified mIL4R-M1 antibody is useful for a sandwich ELISA that measures soluble mouse CD124 protein levels. The biotinylated mIL-4R-M1 antibody (Cat. No. 552508) can be paired with the purified mIL4R-M2 (rat anti-mouse sIL-4R) antibody, Cat. No. 552952, and recombinant soluble mIL-4R $\alpha$  as the standard.

**Neutralization:** The mIL4R-M1 antibody reportedly blocks IL-4's binding to and biological effects on IL-4R-positive cells. Note: for use in in vivo and in vitro systems, our NA/LE (no azide, low endotoxin) format, Cat. No. 552288 is recommended.

**Immunoprecipitation:** The mIL4R-M1 antibody is reported to immunoprecipitate mouse IL-4R proteins. Please note that this application is not routinely tested at BD Biosciences Pharmingen.

### **Suggested Companion Products**

Catalog Number	Name	Size	Clone
553142	Purified Rat Anti-Mouse CD16/CD32 (Mouse BD Fc Block™)	0.5 mg	2.4G2
553894	Biotin Mouse Anti-Rat IgG2a	0.5 mg	RG7/1.30
554061	PE Streptavidin	0.5 mg	(none)
553927	Purified Rat IgG2a, κ Isotype Control	0.5 mg	R35-95
552288	Purified NA/LE Anti-mouse CD124	0.5 mg	mIL4R-M1

#### **Product Notices**

- 1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
- 2. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
- 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

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Feldman GM, Ruhl S, Bickel M, Finbloom DS, Pluznik DH. Regulation of interleukin-4 receptors on murine myeloid progenitor cells by interleukin-6. *Blood.* 1991; 78(7):1678-1684.(Clone-specific: Flow cytometry)

Gessner A, Rollinghoff M. Biologic functions and signaling of the interleukin-4 receptor complexes. *Immunobiology*. 2000; 201(3-4):285-307. (Biology) Hassuneh MR, Nagarkatti PS, Nagarkatti M. Evidence for the participation of interleukin-2 (IL-2) and IL-4 in the regulation of autonomous growth and tumorigenesis of transformed cells of lymphoid origin. *Blood*. 1997; 89(2):610-620. (Clone-specific: Flow cytometry)

Kubo M, Yamashita M, Abe R, et al. CD28 costimulation accelerates IL-4 receptor sensitivity and IL-4-mediated Th2 differentiation. *J Immunol.* 1999; 63(5):2432-2442.(Clone-specific: Flow cytometry)

Lowenthal JW, Castle BE, Christiansen J, et al. Expression of high affinity receptors for murine interleukin 4 (BSF-1) on hemopoietic and nonhemopoietic cells. *J Immunol.* 1988; 140(2):456-464.(Biology)

Mosley B, Beckmann MP, March CJ, et al. The murine interleukin-4 receptor: molecular cloning and characterization of secreted and membrane bound forms. *Cell.* 1989; 59(2):335-348.(Biology)

Sempowski GD, Beckmann MP, Derdak S, Phipps RP. Subsets of murine lung fibroblasts express membrane-bound and soluble IL-4 receptors. Role of IL-4 in enhancing fibroblast proliferation and collagen synthesis. *J Immunol.* 1994; 152(7):3606-3614.(Clone-specific: Flow cytometry)

551853 Rev. 1 Page 2 of 2