Technical Data Sheet PE Mouse Anti-Human CD126

Product Information

Material Number:	551850	
Alternate Name:	IL-6 Receptor α-chain	
Size:	100 tests	
Vol. per Test:	20 µl	
Clone:	M5	
Immunogen:	Mixture of U266, XG-1, and BWD41 cells	
Isotype:	Mouse IgG1, ĸ	
Reactivity:	QC Testing: Human	
Storage Buffer:	Aqueous buffered solution containing BSA and $\leq 0.09\%$ sodium azide.	

Description

The M5 antibody reacts with human CD126 which is also known as the a subunit of the human IL-6 Receptor (IL-6Ra). The human IL-6 Rais an 80 kDa type I transmembrane glycoprotein, also known as B cell stimulatory factor-2 (BSF-2) receptor and IL-6 receptor. The IL-6Ra subunit associates with the 130-160 kDa gp130 subunit (IL-6 receptor ß chain, CD130), that is shared with the receptors for Leukemia Inhibitory Factor (LIF), Ciliary Neurotropic Factor (CNTF), Oncostatin M (OSM), IL-11, Cardiotropin 1 (CT-1) and possibly Neurotrophin-1/B Cell-Stimulating Factor 3 (NNT-1/BSF-3). The IL-6Ra chain binds IL-6 with low affinity, however the association with CD130 stabilizes the IL-6/IL-6Ra complex resulting in the formation of a high affinity complex. The IL-6R ßchain mediates signal transduction. IL-6Roi's are expressed at high levels by activated and EBV-transformed B cells, plasma cells and myeloma cells and at lower levels by most leucocytes, epithelial cells, fibroblasts, hepatocytes and neural cells. IL-6Ra exists in soluble form in human serum. The serum levels of soluble IL- $6R\alpha$ appear to elevate in pathological situations such as multiple myeloma, Grave's disease, juvenile chronic arthritis and HIV. The immunogen used to generate the M5 hybridoma was a mixture of U266, XG-1 (human myeloma cell line expressing membrane IL-6R) and BWD41 cells (murine thymoma cell line transfected with cDNA encoding the extracellular part of IL-6Rα).



Expression of cell surface IL-6R by human PBMC. Human PBMC isolated by density centrifugation (Ficoll-Paque™) were blocked with normal polyclonal human IgG and stained with R-PE-conjugated M5 (20 µg/106 cells, Cat No. 551850). Staining with the M5 antibody (filled histograms) is compared to staining obtained using the isotype control antibody (open histograms). Histograms in the figure were gated on the CD19-negative lymphocytes. Note: Certain human cell lines or cell types (e.g., neutrophils, monocytes) can first be treated with reagents that block receptors for the Fc regions of immunoglobulin to avoid nonspecific immunofluorescent staining mediated by Fc receptors.

Preparation and Storage

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

Application Notes

Application Flow cytometry

Routinely Tested

Recommended Assay Procedure:

Immunofluorescent Staining and Flow Cytometric Analysis: The R-PE conjugated M5 (Cat. No. 551850) antibody can be used for the **BD Biosciences**

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immunofluorescent staining (20 μ g/10e6 cells) and flow cytometric analysis of human nucleated cells to measure their expressed levels of surface IL-6Ra.15 An appropriate purified immunoglobulin isotype control is clone MOPC-21 (Cat. No. 555749).

ELISA: The purified M5 antibody (Cat. No. 551462) is useful as a capture for a sandwich ELISA that measures soluble human IL-6Ra protein levels. The M5 antibody can be paired with the M182 antibody and recombinant soluble human IL-6Ra as a standard.

Suggested Companion Products

Catalog Number	Name	Size	Clone
555749	PE Mouse IgG1, κ Isotype Control	100 tests	MOPC-21

Product Notices

- 1. This reagent has been pre-diluted for use at the recommended Volume per Test. We typically use 1 X 10e6 cells in a 100-µl experimental sample (a test).
- 2. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
- 3. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
- 4. Ficoll-Paque is a trademark of Amersham Biosciences Limited.
- 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.
- 6. Source of all serum proteins is from USDA inspected abattoirs located in the United States.

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