Technical Data Sheet

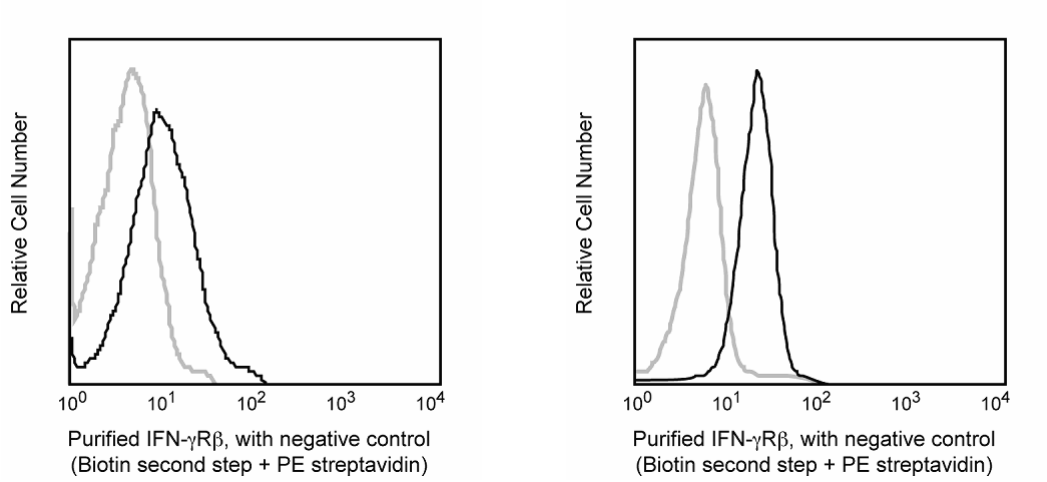
Purified Hamster Anti-Mouse IFN-γ Receptor β Chain

Product Information

Material Number:	**559917**
Size:	0.5 mg
Concentration:	0.5 mg/ml
Clone:	MOB-47
Immunogen:	Mouse IFN-γRβ chain protein
Isotype:	Armenian Hamster IgG
Reactivity:	QC Testing: Mouse
Storage Buffer:	Aqueous buffered solution containing ≤0.09% sodium azide.

Description

The MOB-47 antibody recognizes the 60-65 kD beta chain subunit of the mouse interferon-γ receptor (IFN-γRβ). The IFN-γ receptor β chain is expressed by a variety of normal mouse cell types and cell lines that are responsive to IFN-γ including T and B lymphocytes, NK cells, monocytes, macrophages, granulocytes and fibroblasts. The IFN-γRβ receptor epitope recognized by the MOB-47 antibody is blocked by bound IFN-γ. The immunogen used to generate this hybridoma was the purified soluble extracellular domain of the mouse IFN-γRβ chain protein.



Expression of cell surface IFN-γRβ chains by BALB/c splenic lymphocytes (Left panel) and mouse A20 cells (Right Panel). *BALB/c spleen cells and A20 cells (mouse B cell lymphoma) were preincubated (~15 min., 4°C) with purified 2.4G2 antibody [rat anti-mouse CD16 (FcγIII)/CD32 (FcγII); Cat. No. 553142; 1 µg antibody/10e6 cells] to block nonspecific staining due to immunoglobulin Fc receptors. The cells were stained (45 min., 4°C) with purified MOB-47 antibody (either at 0.5 µg mAb/10e6 BALB/c cells or at 0.125 µg mAb/10e6 A20 cells; Cat. No. 559917). After washing, the cells were incubated (30 min., 4°C) with a biotin-conjugated cocktail of mouse anti-hamster antibodies (Clones G70-204 + G94-56; Cat. No. 554010; 0.5 µg mAb cocktail/10e6 cells). Finally, the cells were washed and incubated with R-PE-conjugated streptavidin (Cat. No. 554061; 0.015 µg PE-SA/10e6 cells). After washing, the cells were analyzed with a FACScan™ Flow Cytometer. The immunofluorescent staining patterns for splenic lymphocytes (left panel) and A20 cells (right panel) that were either not stained with MOB-47 (grey line) or were stained with purified MOB-47 (black line) followed by the 2nd and 3rd layer reagents. The overlapping histograms were generated from reanalyzed flow cytometric data files that were gated for cells that had the light-scattering characteristics of lymphocytes (left panel) or viable tumor cells (right panel).*

Preparation and Storage

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

BD Biosciences

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Application Notes

Application

Flow cytometry	Routinely Tested
Immunoprecipitation	Reported
Western blot	Reported

Recommended Assay Procedure:

Immunofluorescent Staining and Flow Cytometric Analysis: The purified form of MOB-47 (Cat. No. 559917) can be used for the immunofluorescent staining (≤ 1 µg antibody/10e6 cells) and flow cytometric analysis of normal mouse cells or cell lines to measure their expressed levels of IFN-γRβ. An appropriate purified immunoglobulin isotype control is clone A19-3 (Cat. No. 553969). A three-layer staining protocol is recommended for maximizing the detection IFN-γRβ chains expressed by cells as detailed in the figure legend.

Note: The IFN-γRβ receptor epitope recognized by the MOB-47 antibody is blocked by bound IFN-γ. Thus, it has been reported that cells which have bound IFN-γ can be pretreated with dilute acid to remove the bound ligand before they are stained.

IP/WB: The MOB-47 antibody has been reported to be useful for the immunoprecipitation and Western blotting of IFN-γRβ chains from lysates of cloned mouse T cells.

Suggested Companion Products

Catalog Number	Name	Size	Clone
553969	Purified Hamster IgG1, κ Isotype Control	0.5 mg	A19-3
553142	Purified Rat Anti-Mouse CD16/CD32 (Mouse BD Fc Block™)	0.5 mg	2.4G2
554061	PE Streptavidin	0.5 mg	(none)
554010	Biotin Mouse Anti-Armenian and Syrian Hamster IgG Cocktail	0.5 mg	(none)

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.
3. 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.
4. Although hamster immunoglobulin isotypes have not been well defined, BD Biosciences Pharmingen has grouped Armenian and Syrian hamster IgG monoclonal antibodies according to their reactivity with a panel of mouse anti-hamster IgG mAbs. A table of the hamster IgG groups, Reactivity of Mouse Anti-Hamster Ig mAbs, may be viewed at http://www.bdbiosciences.com/pharmingen/hamster_chart_11x17.pdf.

References

Bach EA, Szabo SJ, Dighe AS, et al. Ligand-induced autoregulation of IFN-gamma receptor beta chain expression in T helper cell subsets. *Science*. 1995; 270(5239):1215-1218.(Clone-specific: Flow cytometry, Immunoprecipitation, Western blot)