Technical Data Sheet

Biotin Mouse Anti-Human IFN-γ

Product Information

Material Number:	**554550**
Size:	0.5 mg
Concentration:	0.5 mg/ml
Clone:	4S.B3
Immunogen:	Partially purified human IFN-γ from supernatants of human PBMC stimulated with Staphylococcus aureus
Isotype:	Mouse IgG1, κ
Reactivity:	QC Testing: Human
Storage Buffer:	Aqueous buffered solution containing ≤0.09% sodium azide.

Description

The 4S.B3 antibody reacts with human interferon-γ (IFN-γ). The immunogen used to generate this hybridoma was partially purified human IFN-γ obtained from supernatants of human PBMC stimulated with *Staphylococcus aureus*. This is a neutralizing antibody.

This antibody is routinely tested by ELISA Detection. Other applications were tested at BD Biosciences Pharmingen during antibody development only or reported in the literature.

Preparation and Storage

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

Application Notes

Application

ELISA Detection	Routinely Tested

Recommended Assay Procedure:

ELISA Detection: The biotinylated 4S.B3 antibody (Cat. No. 554550) is useful as a detection antibody in a sandwich ELISA for measuring human IFN-γ protein levels. Biotinylated 4S.B3 antibody can be paired with the purified NIB42 antibody (Cat. No. 551221) as the capture antibody, with recombinant human IFN-γ protein (Cat. No. 554616 or 554617) as the standard. Biotinylated 4S.B3 antibody should be titrated (0.5 -2.0 µg/ml) to determine optimal concentration for ELISA detection. To obtain linear standard curves, doubling dilutions of human IFN-γ ranging from ~15 to 2,000 pg/ml are recommended for inclusion in each ELISA plate. For specific methodology, please visit our web site, www.bdbiosciences.com, and go to the protocols section or the chapter on ELISA in the Immune Function Handbook.

Note 1: This ELISA pair shows no cross-reactivity with any of the cytokines or chemokines tested (*e.g.,* mouse IL-1β, IL-2, IL-3, IL-4, IL-5, IL6, IL-7, IL-9, IL-10, IL-12 p70, IL-15, GM-CSF, IFN-γ, MCP-1, TCA-3, TNF; human IL-1α, IL-1β, IL-2, IL-3, IL-4, IL-5, IL-6, IL-7, IL-8, IL-9, IL-10, IL-11, IL-12 p70, IL-12 p40, IL-13, IL-15, G-CSF, GM-CSF, lymphotactin, MCP-1, MCP-2, MIP-1α, MIP-1β, NT-3, PDGF-AA, sCD23 , SCF, TNF, LT-α, VEGF; rat IL-2, IL-4, IL-6, IL-10, GM-CSF, IFN-γ, TNF).

Note 2: This ELISA pair is recommended primarily for measuring cytokine from experimental cell culture systems. These ELISA reagents are not recommended for assaying serum or plasma samples. For measuring human IFN-γ in serum or plasma our human IFN-γ OptEIA Set (Cat. No. 555142) or BD OptEIA Kit II (Cat. No. 550612) are specially formulated and recommended.

Immunofluorescent Staining and Flow Cytometric Analysis: The 4S.B3 antibody is useful for immunofluorescent staining and flow cytometric analysis to identify and enumerate IFN-γ producing cells within mixed cell populations. FITC-and PE-conjugated 4S.B3 antibodies (Cat. No. 554551; No. 554552) are especially suitable for these studies. The use of a specificity control, such as one of the following, is suggested: 1) recombinant human IFN-γ (Cat. No. 554616), 2) unlabeled 4S.B3 antibody (554549), or 3) mouse IgG 1 isotype controls, FITC-MOPC-21 (Cat. No. 554679) and PE-MOPC-21 (Cat. No. 554680).

Western Blot: The 4S.B3 antibody has been reported to be useful for Western blotting. Please note that this application is not routinely tested at BD Biosciences Pharmingen.

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Suggested Companion Products

Catalog Number	Name	Size	Clone
550612	Human IFN-γ OptEIA Kit II	2 plates	(none)
555142	Human IFN-γ OptEIA Set	20 tests	(none)
554616	Recombinant Human IFN-γ	25 µg	(none)
551221	Purified Mouse Anti-Human IFN-γ	1.0 mg	NIB42

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.

References

Meager A. Characterization of interferons and immunoassays. In: Clemens MJ, Morris AG, Gearing AJH, ed. *Lymphockies and Interferons. A Practical Approach.* Oxford: IRL Press Ltd; 1987:105-127.(Biology)

Meager A, Parti S, Barwick S, Spragg J, O'Hagan K. Detection of hybridomas secreting monoclonal antibodies to human gamma interferon using a rapid screening technique and specificity of certain monoclonal antibodies to gamma interferon. *J Interferon Res.* 1984; 4(4):619-625.(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)