Technical Data Sheet

PE-CF594 Mouse Anti-Human IFN-γ

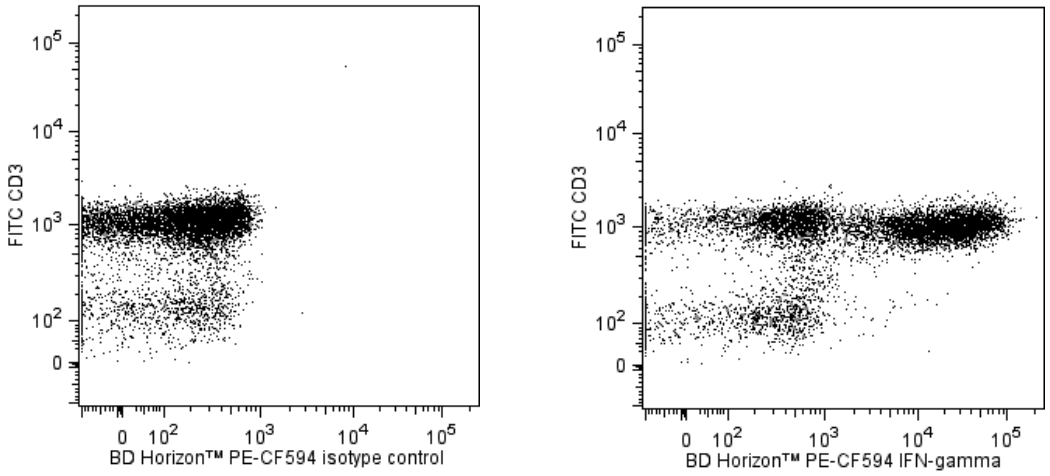
Product Information

Material Number:	**562392**
Alternate Name:	IFNG; Interferon-gamma; Interferon-γ; Type II interferon; MAF
Size:	50 Tests
Vol. per Test:	5 µl
Clone:	B27
Immunogen:	Human IFN-γ Recombinant Protein
Isotype:	Mouse IgG1, κ
Reactivity:	QC Testing: Human Tested in Development: Rhesus, Cynomolgus, Baboon
Storage Buffer:	Aqueous buffered solution containing BSA and ≤0.09% sodium azide.

Description

The B27 monoclonal antibody specifically binds to human interferon-γ (IFN-γ), a 14-18 kDa glycoprotein containing 143 amino acid residues. IFN-γ is a potent multifunctional cytokine produced by several activated cell types including NK, NKT, CD4+TCRαβ+, CD8+TCRαβ+, and TCRγδ+ T cells. IFN-γ exerts its biological effects through specific binding to the high-affinity IFN-γ receptor complex comprised of IFN-γRα (CD119) and IFN-γRβ subunits. In addition to its antiviral effects, IFN-γ upregulates a number of lymphoid cell functions including the antimicrobial and anti-tumor responses of macrophages, NK cells, and neutrophils. In addition, IFN-γ influences the regulation of proliferation, differentiation, and effector responses of B cell and T cell subsets. These influences can involve IFN-γ's capacity to boost MHC class I and II expression by antigen-presenting cells as well as direct effects on B cells and T cells themselves. B27 is a neutralizing antibody. The use of B27 antibody for epitope mapping of human IFN-γ has been described. The B27 antibody has been reported not to bind to denatured IFN-γ.

This antibody is conjugated to BD Horizon™ PE-CF594, which has been developed exclusively by BD Biosciences as a better alternative to PE-Texas Red®. PE-CF594 excites and emits at similar wavelengths to PE-Texas Red® yet exhibits improved brightness and spectral characteristics. Due to PE having maximal absorption peaks at 496 nm and 564 nm, PE-CF594 can be excited by the blue (488-nm), green (532-nm) and yellow-green (561-nm) lasers and can be detected with the same filter set as PE-Texas Red® (eg 610/20-nm filter).



Multicolor flow cytometric analysis of IFN-γ expressed in stimulated human peripheral blood mononuclear cells.
HiCK-1 Human Cytokine Positive Control Cells (Cat. No. 555061) were permeabilized with BD Perm/Wash™ Buffer (Cat. No. 554723). The cells were then stained with either a BD Horizon™ PE-CF594 Mouse IgG1, κ Isotype Control (Cat No. 562292, Left Panel) or with the BD Horizon™ PE-CF594 Mouse Anti-Human IFN-γ antibody (Cat No. 562392, Right Panel) in conjunction with a FITC Mouse Anti-Human CD3 antibody (Cat. No. 555332/561806/561807). Two-color flow cytometric dot plots showing the expression of IFN-γ (or Ig Isotype Control staining) versus CD3 were derived from gated events with the forward and side light-scatter characteristics of intact lymphocytes. Flow cytometry was performed using a BD™ LSR II Flow Cytometer System.

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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 BD Horizon™ PE-CF594 under optimum conditions, and unconjugated antibody and free PE-CF594 were removed.

Application Notes

Application

Intracellular staining (flow cytometry)	Routinely Tested

Suggested Companion Products

Catalog Number	Name	Size	Clone
562292	PE-CF594 Mouse IgG1, κ Isotype Control	0.1 mg	X40
554723	Perm/Wash Buffer	100 mL	(none)
555061	HiCK-1 Human Cytokine Positive Control Cells	1 mL	(none)
555332	FITC Mouse Anti-Human CD3	100 Tests	UCHT1
561806	FITC Mouse Anti-Human CD3	25 Tests	UCHT1
561807	FITC Mouse Anti-Human CD3	500 Tests	UCHT1
554656	Stain Buffer (FBS)	500 mL	(none)
554657	Stain Buffer (BSA)	500 mL	(none)

Product Notices

1. This reagent has been pre-diluted for use at the recommended Volume per Test. We typically use 1 × 10^6 cells in a 100-µl experimental sample (a test).
2. An isotype control should be used at the same concentration as the antibody of interest.
3. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
4. Please observe the following precautions: Absorption of visible light can significantly alter the energy transfer occurring in any tandem fluorochrome conjugate; therefore, we recommend that special precautions be taken (such as wrapping vials, tubes, or racks in aluminum foil) to prevent exposure of conjugated reagents, including cells stained with those reagents, to room illumination.
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. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
7. Texas Red is a registered trademark of Molecular Probes, Inc., Eugene, OR.
8. CF™ is a trademark of Biotium, Inc.
9. When excited by the yellow-green (561-nm) laser, the fluorescence may be brighter than when excited by the blue (488-nm) laser.
10. This product is provided under an Agreement between BIOTIUM and BD Biosciences. The manufacture, use, sale, offer for sale, or import of this product is subject to one or more patents or pending applications owned or licensed by Biotium, Inc. This product, and only in the amount purchased by buyer, may be used solely for buyer's own internal research, in a manner consistent with the accompanying product literature. No other right to use, sell or otherwise transfer (a) this product, or (b) its components is hereby granted expressly, by implication or by estoppel. This product is for research use only. Diagnostic uses require a separate license from Biotium, Inc. For information on purchasing a license to this product including for purposes other than research, contact Biotium, Inc., 3159 Corporate Place, Hayward, CA 94545, Tel: (510) 265-1027. Fax: (510) 265-1352. Email: btinfo@biotium.com.
11. Because of the broad absorption spectrum of the tandem fluorochrome, extra care must be taken when using multi-laser cytometers, which may directly excite both PE and CF™594.
12. Species testing during development may have been performed with a different format of the same clone. Selected applications have been tested for cross-reactivity.
13. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.

References

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. (Clone-specific)

Favre C, Wijdenes J, Cabrillat H, Djossou O, Banchereau J, de Vries JE. Epitope mapping of recombinant human gamma interferon using monoclonal antibodies. *Mol Immunol.* 1989; 26(1):17-25. (Clone-specific: Immunoprecipitation, Neutralization)

Fonteneau JF, Le Drean E, Le Guiner S, Gervois N, Diez E, Jotereau F. Heterogeneity of biologic responses of melanoma-specific CTL. *J Immunol.* 1997; 159(6):2831-2839. (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)

Rotteveel FT, Kokkelink I, van Lier RA, et al. Clonal analysis of functionally distinct human CD4+ T cell subsets. *J Exp Med.* 1988; 168(5):1659-1673. (Biology)