## **Technical Data Sheet**

# BV786 Rat Anti-Mouse IFN-y

#### **Product Information**

Material Number: 563773

Alternate Name: IFN-γ; IFN-g; IFN-gamma; Interferon gamma; Type II Interferon

 Size:
 50 μg

 Concentration:
 0.2 mg/ml

 Clone:
 XMG1.2

**Immunogen:** Mouse IFN-γ Recombinant Protein

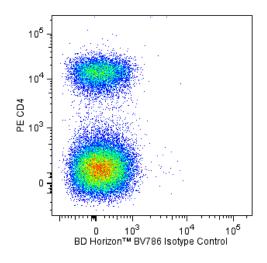
Isotype:Rat IgG1,  $\kappa$ Reactivity:QC Testing: Mouse

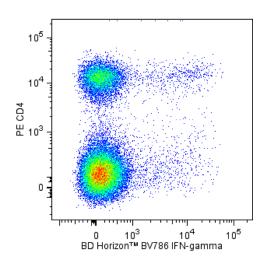
**Storage Buffer:** Aqueous buffered solution containing BSA and ≤0.09% sodium azide.

## Description

The XMG1.2 monoclonal antibody specifically binds to mouse interferon- $\gamma$  (IFN- $\gamma$ ) protein. IFN- $\gamma$  is a pleiotropic cytokine, of approximately 15-17 kDa, involved in the regulation of inflammatory and immune responses. It plays an important role in activation, growth, and differentiation of T and B lymphocytes, macrophages, NK cells and other non-hematopoietic cell types. IFN- $\gamma$  production is associated with the Th1 cell differentiation. The purified form of this antibody has been reported to be a neutralizing antibody.

The antibody was conjugated to BD Horizon<sup>TM</sup> BV786 which is part of the BD Horizon<sup>TM</sup> Brilliant Violet<sup>TM</sup> family of dyes. This dye is a tandem fluorochrome of BD Horizon<sup>TM</sup> BV421 with an Ex Max of 405-nm and an acceptor dye with an Em Max at 786-nm. BD Horizon<sup>TM</sup> BV786 can be excited by the violet laser and detected in a filter used to detect Cy7<sup>TM</sup>-like dyes (eg, 780/60-nm filter).





Two-color flow cytometric analysis of IFN-y expression by stimulated mouse splenocytes. Mouse splenic leucocytes were stimulated for 5 hours with Phorbol 12-Myristate 13-Acetate (PMA; Sigma P-8139; 50 ng/ml) and lonomycin (Sigma I-0634; 1 µg/ml) in the presence of BD GolgiStop™ Protein Transport Inhibitor (containing Monensin) (Cat. No. 554724). The cells were harvested, washed with BD Pharmingen™ Stain Buffer (FBS) (Cat. No. 554656), and fixed with BD Cytofix™ Fixation Buffer (Cat. No. 554655). The cells were then washed and stained in BD Perm/Wash™ Buffer (Cat. No. 554723) with PE Rat Anti-Mouse CD4 antibody (Cat. No. 553048/553049/561837) and either BD Horizon™ BV786 Rat IgG1, κ Isotype Control (Cat. No. 563847; Left Panel) or BD Horizon™ BV786 Rat Anti-Mouse IFN-γ antibody (Cat. No. 563773; Right Panel) by using the BD Biosciences Intracellular Cytokine Staining protocol. Two-color flow cytometric dot plots show the correlated expression of IFN-γ (or Ig Isotype control staining) versus CD4 were derived from gated events with the forward and side light-scatter characteristics of intact stimulated leucocytes. Flow cytometric analysis was performed using a BD™ LSR II Flow Cytometer System.

#### **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™ BV786 under optimum conditions, and unconjugated antibody and free BD Horizon™ BV786 were removed.

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### **Application Notes**

#### Application

Intracellular staining (flow cytometry) Routinely Tested	
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#### **Suggested Companion Products**

Catalog Number	Name	Size	Clone	
554656	Stain Buffer (FBS)	500 ml	(none)	
554724	Protein Transport Inhibitor (Containing Monensin)	0.7 ml	(none)	
554655	Fixation Buffer	100 ml	(none)	
554723	Perm/Wash Buffer	100 ml	(none)	
563847	BV786 Rat IgG1, κ Isotype Control	50 μg	R3-34	
553048	PE Rat Anti-Mouse CD4	0.1 mg	RM4-5	
561837	PE Rat Anti-Mouse CD4	25 μg	RM4-5	
553049	PE Rat Anti-Mouse CD4	0.2 mg	RM4-5	
554715	BD Cytofix/Cytoperm Plus Kit (with BD GolgiStop)	250 tests	(none)	

#### **Product Notices**

- 1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
- 2. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
- 3. An isotype control should be used at the same concentration as the antibody of interest.
- 4. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
- 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. Cy is a trademark of Amersham Biosciences Limited.
- Brilliant Violet<sup>TM</sup> 421 is a trademark of Sirigen.
- 9. Brilliant Violet<sup>TM</sup> 786 is a trademark of Sirigen.

#### 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: ELISA, Neutralization)

Cherwinski HM, Schumacher JH, Brown KD, Mosmann TR. Two types of mouse helper T cell clone. III. Further differences in lymphokine synthesis between Th1 and Th2 clones revealed by RNA hybridization, functionally monospecific bioassays, and monoclonal antibodies. *J Exp Med.* 1987; 166(5):1229-1244. (Clone-specific: ELISA)

Klinman D and Nutman T. ELISPOT assay to detect cytokine-secreting murine and human cells. In: Coligan J, Kruisbeek A, Margulies D, Shevach E, Strober W, ed. Current Protocols in Immunology. 1994:6-19. (Clone-specific: ELISPOT)

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)

Sander B, Hoiden I, Andersson U, Moller E, Abrams JS. Similar frequencies and kinetics of cytokine producing cells in murine peripheral blood and spleen. Cytokine detection by immunoassay and intracellular immunostaining. *J Immunol Methods*. 1993; 166(2):201-214. (Clone-specific)

Suzuki Y, Yang Q, Conley FK, Abrams JS, Remington JS. Antibody against interleukin-6 reduces inflammation and numbers of cysts in brains of mice with toxoplasmic encephalitis. *Infect Immun.* 1994; 62(7):2773-2778. (Clone-specific: In vivo exacerbation, Neutralization)

Yang X, HayGlass KT. A simple, sensitive, dual mAb based ELISA for murine gamma interferon determination: comparison with two common bioassays. *J Immunoassay*. 1993; 14(3):129-148. (Clone-specific: ELISA)

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