

Anti-hIFN- γ -IgA2

Neutralizing IgA2 monoclonal antibody to human interferon gamma

Catalog code: hifng-mab7-2

<https://www.invivogen.com/anti-hifng-iga>

For research use only

Version 24F24-NJ

PRODUCT INFORMATION

Contents: 200 μ g purified anti-hIFN- γ -IgA2 monoclonal antibody (mAb), provided azide-free and lyophilized.

Target: Human interferon gamma (hIFN- γ)

Clone: H7WM120

Source: Chinese hamster ovary (CHO) cells

Isotype: Human IgA2, kappa

Purification: Affinity chromatography with protein M

Formulation: 0.2 μ m filtered solution in Tris HCl buffer with glycine, saccharose, and stabilizing agents

Tested applications: Neutralization & blocking *in vitro*

Antibody resuspension (0.1 mg/ml)

Note: Ensure you see the lyophilized pellet before resuspension.

Resuspend anti-hIFN- γ -hIgA2 with sterile water:

Add 2 ml of sterile water per 200 μ g vial.

Storage and stability

- Product is shipped at room temperature. Upon receipt, store lyophilized antibody at -20°C.

- Reconstituted antibody is stable for 1 month when stored at 4°C and for 1 year when stored at -20°C. Avoid repeated freeze-thaw cycles.

Quality control

- The product has been validated for neutralization using cellular assays.

- The complete sequence of this antibody has been verified.

- The absence of bacterial contamination (e.g. lipoproteins and endotoxins) has been confirmed using HEK-Blue™ TLR2 and HEK-Blue™ TLR4 cells.

BACKGROUND

Interferon gamma (IFN- γ) is a Type II interferon, secreted by CD4+ T-helper 1 (Th1) cells and activated natural killer (NK) cells. It plays an important role in activating lymphocytes to enhance anti-microbial and anti-tumor effects^{1,2}. In addition, IFN- γ regulates the proliferation, differentiation, and response of lymphocyte subsets. Signaling takes place through a IFN Receptor complex consisting of two alpha chains (Type I receptor) and two beta chains (Type 2 receptor)^{3,4}. Upon phosphorylation by JAK1, STAT1-homodimers translocate to the nucleus, where they bind interferon-gamma-activated sites (GAS) in the promoter of IFN- γ inducible genes⁴.

1. Shtrichman R. & Samuel CE., 2001. The role of gamma interferon in antimicrobial immunity. *Curr Opin Microbiol.* 4(3):251-9. 2. Sato A. et al., 2006. Antitumor activity of IFN-lambda in murine tumor models. *J Immunol.* 176(12):7686-94. 3. Platanias LC., 2005. Mechanisms of type-I- and type-II-interferon-mediated signalling. *Nat Rev Immunol.* 5(5):375-86. 4. Schroder K. et al., 2004. Interferon-gamma: an overview of signals, mechanisms and functions. *J Leukoc Biol.* 75(2):163-89.

TECHNICAL SUPPORT

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DESCRIPTION

Anti-hIFN- γ -IgA2 is a recombinant mAb specific for human interferon γ (hIFN- γ). It was generated by combining the constant domain of the human IgA2 molecule with the murine variable region of the clone H7WM120. Anti-hIFN- γ -IgA2 has been selected for its ability to efficiently neutralize the biological activity of hIFN- γ . It is produced in CHO cells and purified by affinity chromatography.

APPLICATIONS

Anti-hIFN- γ -IgA2 is a neutralizing antibody. It can be used to block hIFN- γ -induced cellular activation *in vitro*, as described below.

NEUTRALIZATION PROTOCOL

The exact concentration of antibody required to neutralize hIFN- γ activity is dependent on the cytokine concentration, cell type and growth conditions. Below is a protocol using recombinant human IFN- γ as well as HEK-Blue™ IFN- γ cells. These cells are HEK293 cells stably expressing the human STAT1 and an IFN- γ -inducible SEAP (secreted embryonic alkaline phosphatase) reporter gene. Changes in SEAP activity in the supernatant due to inhibition of IFN- γ receptor binding can be assessed using QUANTI-Blue™ Solution, a SEAP detection reagent.

In a 96-well plate:

1. Prepare a serial dilution of the Anti-hIFN- γ -IgA2 (1 μ g/ml - 10 ng/ml final concentration).

Note: We recommend using Anti- β -Gal-hlgA2 (which targets E. coli β -galactosidase) as a negative control antibody.

2. Add 1 ng/ml of recombinant hIFN- γ to a final volume of 40 μ l.

3. Incubate for 30 minutes at 37°C, 5% CO₂.

4. Prepare a suspension of HEK-Blue™ IFN- γ cells (~3.0 x 10⁵ cells/ml) in culture medium.

5. Add 160 μ l (5 x 10⁴ cells/well) of the cell suspension to each well

6. Incubate the plate at 37°C in a 5% CO₂ incubator for 18-24 h.

7. Monitor SEAP production using QUANTI-Blue™ Solution following the instructions on the data sheet.

RELATED PRODUCTS

Product	Cat. Code
HEK-Blue™ IFN- γ Cells	hkb-ifng
Anti- β -Gal-hlgA2	bgal-mab7
Recombinant human IFN- γ	rcyec-hifng
QUANTI-Blue™ Solution	rep-qb-1