Equine IFN-γ Antibody

Antigen Affinity-purified Polyclonal Goat IgG
Catalog Number: AF1586

DESCRIPTION

Species Reactivity	Equine
Specificity	Detects equine IFN-γ in ELISAs and Western blots. In sandwich immunoassays, approximately 20% cross-reactivity with recombinant canine IFN-γ is observed, less than 6% cross-reactivity with recombinant bovine IFN-γ and recombinant feline IFN-γ is observed, and less than 0.2% cross-reactivity with recombinant human IFN-γ, recombinant mouse IFN-γ, recombinant rat IFN-γ, recombinant porcine IFN-γ, recombinant rhesus macaque IFN-γ, and recombinant cotton rat IFN-γ is observed.
Source	Polyclonal Goat IgG
Purification	Antigen Affinity-purified
Immunogen	*E. coli*-derived recombinant equine IFN-γ Ala25-Gln166 Accession # P42160
Endotoxin Level	<0.10 EU per 1 µg of the antibody by the LAL method.
Formulation	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied either lyophilized or as a 0.2 µm filtered solution in PBS.

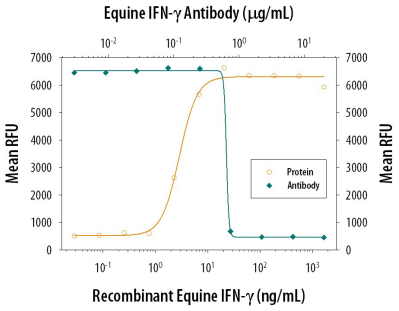
APPLICATIONS

Please Note: *Optimal dilutions should be determined by each laboratory for each application. General Protocols are available in the Technical Information section on our website.*

	Recommended Concentration	Sample
Western Blot	0.1 µg/mL	Recombinant Equine IFN-γ (Catalog # 1586-HG)
Immunocytochemistry	5-15 µg/mL	See Below
Equine IFN-γ Sandwich Immunoassay		**Reagent**
ELISA Capture	0.2-0.8 µg/mL	Equine IFN-γ Antibody (Catalog # AF1586)
ELISA Detection	0.1-0.4 µg/mL	Equine IFN-γ Biotinylated Antibody (Catalog # BAF1586)
Standard		Recombinant Equine IFN-γ (Catalog # 1586-HG)
Neutralization	Measured by its ability to neutralize IFN-γ inhibition of EMCV-induced cytopathy in the L-929 mouse fibroblast cell line. Vogel, S. and M. Hogan (1995) in Current Protocols in Immunology. Ciocio, R. (ed); John Wiley & Sons, Inc. p. 6. 9. 1. The Neutralization Dose (ND_{50}) is typically 0.25-1.25 µg/mL in the presence of 20 ng/mL Recombinant Equine IFN-γ.	

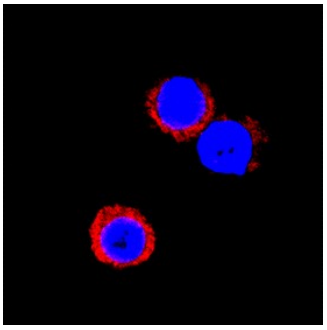
DATA

Neutralization



IFN-γ Inhibition of EMCV-induced Cytopathy and Neutralization by Equine IFN-γ Antibody. Recombinant Equine IFN-γ (Catalog # 1586-HG) reduces the Encephalomyocarditis Virus (EMCV)-induced cytopathy in the L-929 mouse fibroblast cell line in a dose-dependent manner (orange line), as measured by Resazurin (Catalog # AR002). Inhibition of EMCV activity elicited by Recombinant Equine IFN-γ (20 ng/mL) is neutralized (green line) by increasing concentrations of Goat Anti-Equine IFN-γ Antigen Affinity-purified Polyclonal Antibody (Catalog # AF1586). The ND_{50} is typically 0.25-1.25 µg/mL.

Immunocytochemistry



IFN-γ in Equine PBMCs. IFN-γ was detected in immersion fixed equine peripheral blood mononuclear cells (PBMCs) treated with Calcium Ionomycin and PMA using Goat Anti-Equine IFN-γ Antigen Affinity-purified Polyclonal Antibody (Catalog # AF1586) at 15 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Goat IgG Secondary Antibody (red; Catalog # NL001) and counterstained with DAPI (blue). Specific staining was localized to cytoplasm. View our protocol for Fluorescent ICC Staining of Non-adherent Cells.

PREPARATION AND STORAGE

Reconstitution	Reconstitute at 0.2 mg/mL in sterile PBS.
Shipping	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
Stability & Storage	**Use a manual defrost freezer and avoid repeated freeze-thaw cycles.** ● 12 months from date of receipt, -20 to -70 °C as supplied. ● 1 month, 2 to 8 °C under sterile conditions after reconstitution. ● 6 months, -20 to -70 °C under sterile conditions after reconstitution.

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BACKGROUND

Interferon-gamma (IFN-γ), also known as type II or immune interferon, exerts a wide range of immunoregulatory activities and is considered to be the prototype proinflammatory cytokine (1, 2). Mature equine IFN-γ exists as a noncovalently linked homodimer of 20-25 kDa variably glycosylated subunits (3, 4). It shares 73%-82% amino acid sequence identity with bovine, canine, feline, and porcine IFN-γ and 42%-64% with cotton rat, human, mouse, rat, and rhesus IFN-γ. IFN-γ dimers bind to IFN-γ RI (alpha subunits) which then interact with IFN-γ RII (beta subunits) to form the functional receptor complex of two α and two β subunits. Inclusion of IFN-γ RII increases the binding affinity for ligand and the efficiency of signal transduction (5, 6). IFN-γ is produced by a variety of immune cells under inflammatory conditions, notably by T cells and NK cells (7). It plays a key role in host defense by promoting the development and activation of Th1 cells, chemoattraction and activation of monocytes and macrophages, upregulation of antigen presentation molecules, and immunoglobulin class switching in B cells. It also exhibits antiviral, antiproliferative, and apoptotic effects (7, 8). In addition, IFN-γ functions as an anti-inflammatory mediator by promoting the development of regulatory T cells and inhibiting Th17 cell differentiation (9, 10). The pleiotropic effects of IFN-γ contribute to the development of multiple aspects of atherosclerosis (8).

References:

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