Sterile	Mouse Interferon-y (mIFN-y)		Cel	l Signaling
222	 SC 100 μg (With Carrier) SF 100 μg (Carrier Free) 		Orders Support	877-616-CELL (2355) orders@cellsignal.com 877-678-TECH (8324) info@cellsignal.com
\$ #	Multi-milligram quantities available	New 03/11	Web	www.cellsignal.com
This product is intended for research purposes only. This product is not intended to be used for				

therapeutic or diagnostic purposes in humans or animals.

Source: Recombinant mouse IFN- γ (mIFN- γ) His23-Cys155 (Accession # NP_032363) was produced in *E. coli* at Cell Signaling Technology.

Molecular Characterization: Recombinant mIFN- γ has a Met on the amino terminus and has a calculated MW of 15,652. DTT-reduced and non-reduced protein migrate as 14 kDa polypeptides. The expected amino-terminus MHGTV of recombinant mIFN- γ was verified by amino acid sequencing.

Endotoxin: Less than 0.01 ng endotoxin/1 µg mIFN-y.

Purity: 98% as determined by SDS-PAGE of 6 μ g reduced (+) and non-reduced (-) recombinant mIFN- γ . All lots are greater than 98% pure.





Western blot analysis of extracts from L-929 cells, untreated or treated with mIFN- γ for 20 minutes, using Phospho-Stat1 (Tyr701) Antibody #9171 (upper) and Stat1 Antibody #9172 (lower).

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The purity of recombinant mIFN- γ was determined by SDS-PAGE of 6 μ g reduced (+) and non-reduced (-) recombinant mIFN- γ and staining overnight with Coomassie Blue.

Bioactivity: The bioactivity of mIFN- γ was determined in a virus protection assay. The ED₅₀ of each lot is between 30-150 pg/ml.



The bioactivity of recombinant mIFN- γ was determined in a virus protection assay. L-929 cells were pretreated with increasing concentrations of mIFN- γ for 24 hours. Cells were then innoculated with encephalomyocarditis virus (EMCV) and incubated for an additional 48 hours. Surviving cells were then fixed and stained with crystal violet and the OD₅₉₅ was determined.

Formulation: With carrier: Lyophilized from a 0.22 μm filtered solution of PBS, pH 7.2 containing 20 μg BSA per 1 μg mIFN- $\gamma.$

Carrier free: Lyophilized from a 0.22 μm filtered solution of PBS, pH 7.2.

Reconstitution:

With carrier: Add sterile PBS or PBS containing 1% bovine or human serum albumin or 5-10% FBS to a final mIFN- γ concentration of greater than 50 μ g/ml. Solubilize for 30 minutes at room temperature with occasional gentle vortexing.

Carrier free: Add sterile PBS or PBS containing protein to minimize absorption of mIFN- γ to surfaces. Solubilize for 30 minutes at room temperature with occasional gentle vortexing. Stock mIFN- γ should be greater than 50 µg/ml.

Storage: Stable in lyophilized state at 4°C for 1 year after receipt. Sterile stock solutions reconstituted with carrier protein are stable at 4°C for 2 months and at -20°C for 6 months. Avoid repeated freeze-thaw cycles.

Maintain sterility. Storage at -20°C should be in a manual defrost freezer.

Applications: Optimal concentration for the desired application should be determined by the user.

Background: IFN-y plays key roles in both the innate and adaptive immune response. IFN- γ activates the cytotoxic activity of innate immune cells such as macrophages and NK cells (1,2). IFN- γ production by NK cells and antigen-presenting cells (APCs) promotes cell-mediated adaptive immunity by inducing IFN-y production by T lymphocytes, increased class I and class II MHC expression, and enhancing peptide antigen presentation (1). The anti-viral activity of IFN- γ is due to its induction of PKR and other regulatory proteins. Binding of IFN-y to the IFNGR1/IF-NGR2 complex promotes dimerization of the receptor complexes. Binding induces a conformational change in receptor intracellular domains and signaling involves Jak1, Jak2, and Stat1 (3). The critical role of IFN-y in amplification of immune surveillance and function is supported by increased susceptibility to pathogen infection by IFN-y or IFNGR knockout mice, and in humans with inactivating mutations in IFNGR1 or IFNGR2. IFN-y also appears to have a role in atherosclerosis (4).

Background References:

(1) Schroder, K. et al. (2004) *J Leukoc Biol* 75, 163-89.

(2) Martinez, F.O. et al. (2009) Annu Rev Immunol 27, 451-83.

- (3) Kotenko, S.V. et al. (1995) J Biol Chem 270, 20915-21.
- (4) McLaren, J.E. and Ramji, D.P. (2009) *Cytokine Growth Factor Rev* 20, 125-35.