

Human Interferon- α 1 (hIFN- α 1)

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rev. 03/10/20

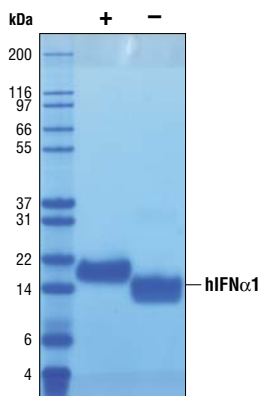
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Source: Recombinant human IFN- α 1 (hIFN- α 1) Cys24 - Asp189 (Accession # NP_076918) was produced in *E. coli* at Cell Signaling Technology.

Molecular Characterization: Recombinant hIFN- α 1 does not have a Met on the amino terminus and has a calculated MW of 19,386. DTT-reduced protein migrates as an 18 kDa polypeptide and non-reduced protein migrates as a 14 kDa polypeptide due to intramolecular cystines. The expected amino-terminal CDLPE of recombinant hIFN- α 1 was verified by amino acid sequencing.

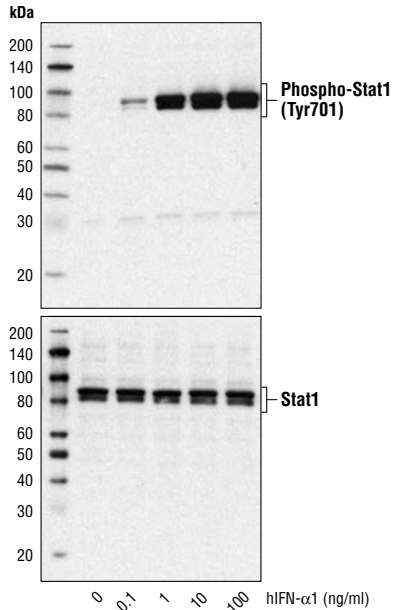
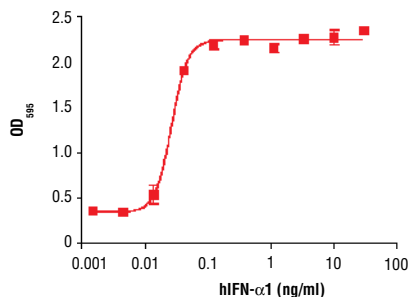
Endotoxin: Less than 0.01 ng endotoxin/1 μ g hIFN- α 1.

Purity: >98% as determined by SDS-PAGE of 6 μ g reduced (+) and non-reduced (-) recombinant hIFN- α 1. All lots are greater than 98% pure.



The purity of recombinant hIFN- α 1 was determined by SDS-PAGE of 6 μ g reduced (+) and non-reduced (-) recombinant hIFN- α 1 and staining overnight with Coomassie Blue.

Bioactivity: The bioactivity of hIFN- α 1 was determined in a virus protection assay. The ED₅₀ of each lot is between 0.01-1.0 ng/ml.



Western blot analysis of extracts from HeLa cells untreated or treated with hIFN- α 1 for 15 minutes, using Phospho-Stat1 (Tyr701) Antibody #9171 (upper) and Stat1 Antibody #9172 (lower).

◀ The bioactivity of recombinant hIFN- α 1 was determined in a virus protection assay. HeLa cells were pretreated with increasing concentrations of hIFN- α 1 for 24 hours. Cells were then inoculated 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 μ m filtered solution of PBS, pH 7.2 containing 20 μ g BSA per 1 μ g hIFN- α 1.

Carrier free: Lyophilized from a 0.22 μ m 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 hIFN- α 1 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 hIFN- α 1 to surfaces. Solubilize for 30 minutes at room temperature with occasional gentle vortexing. Stock hIFN- α 1 should be greater than 50 μ g/ml.

Storage: Stable in lyophilized state at -20°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: Interferon- α 1 is a member of the Type I IFN (1) family best known for their antiviral activity. Most nucleated cells produce one or more Type I IFNs in response to viral infection (2). Secreted Type I IFN then induces viral protective responses in neighboring non-infected cells. Type I IFNs also enhance virus-induced apoptosis (3). Other IFN- α 1 activities include enhancement of dendritic cell maturation and cytotoxic T cell activity (4). IFN- α 1 binds to the IFN- α R1 and IFN- α R2 heterodimer (1). Intracellular signaling through the Jak/Stat pathway is best characterized (3). However, the PI3K, ERK, and p38 kinase pathways are also involved (5). The antiviral activities of the IFNs have led to their use in treating viral infections (4). Type I IFNs also appear to have an integral role in several autoimmune diseases (6).

Background References:

- (1) Nagai, T. et al. (2003) *J Immunol* 171, 5233-43.
- (2) Stetson, D.B. and Medzhitov, R. (2006) *Immunity* 25, 373-81.
- (3) Vilcek, J. (2006) *Immunity* 25, 343-8.
- (4) Luft, T. et al. (2002) *Int Immunol* 14, 367-80.
- (5) van Boxel-Dezaire, A.H. et al. (2006) *Immunity* 25, 361-72.
- (6) Banchereau, J. and Pascual, V. (2006) *Immunity* 25, 383-92.