Sterile

#8923

Mouse Interleukin-3 (mIL-3)

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rev. 04/04/17

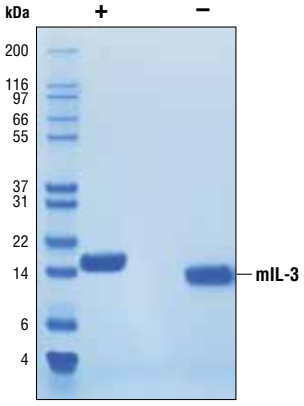
For Research Use Only. Not For Use In Diagnostic Procedures.

Source: Recombinant mouse Interleukin-3 (mIL-3) Ala27-Cys166 (Accession # NM_010556) was produced in *E. coli* at Cell Signaling Technology.

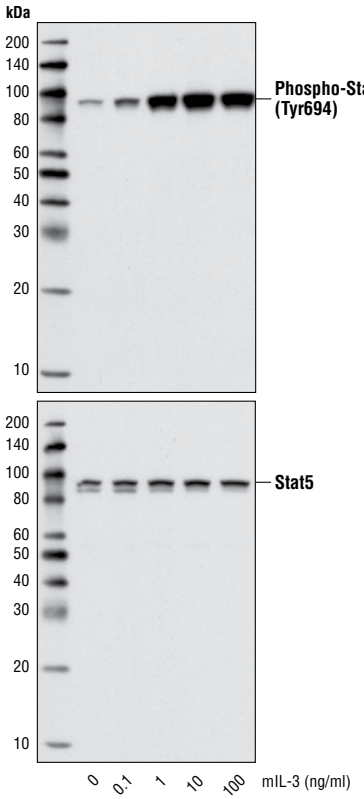
Molecular Characterization: Recombinant mIL-3 does not have a Met on the amino terminus and has a calculated MW of 15,674. DTT-reduced protein migrates as a 16 kDa polypeptide and non-reduced protein migrates as a 14 kDa polypeptide due to intramolecular cystines. The expected amino-terminal ASISG of recombinant mIL-3 was verified by amino acid sequencing.

Endotoxin: Less than 0.01 ng endotoxin/1 μg mIL-3.

Purity: >98% as determined by SDS-PAGE of 6 μg reduced (+) and non-reduced (-) recombinant mIL-3. All lots are greater than 98% pure.

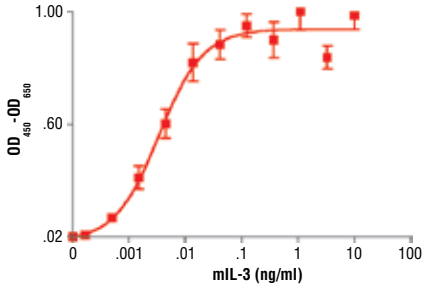


The purity of recombinant mIL-3 was determined by SDS-PAGE of 6 μg reduced (+) and non-reduced (-) recombinant mIL-3 and staining overnight with Coomassie Blue.

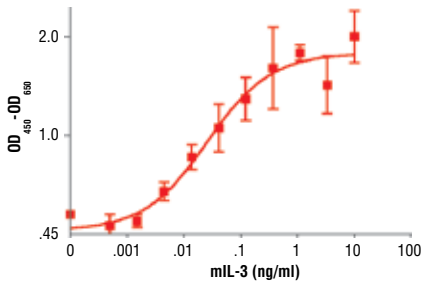


Western blot analysis of extracts from BaF3 cells, untreated or treated with mIL-3 for 10 minutes, using Phospho-Stat5 (Tyr694) (C11C5) Rabbit mAb #9359 (upper) and Stat5 (3H7) Rabbit mAb #9358 (lower).

Bioactivity: The bioactivity of recombinant mIL-3 was determined in MC/9 and BaF3 cell proliferation assays. The ED50 of each lot is between 2-50 pg/ml in MC/9 cells and 20-90 pg/mL in BaF3 cells.



The proliferation of MC/9 cells treated with increasing concentrations of mIL-3 was assessed. After 72 hour treatment with mIL-3, cells were incubated with a tetrazolium salt and the OD_{450} - OD_{650} was determined.



The proliferation of BaF3 cells treated with increasing concentrations of mIL-3 was assessed. After 48 hour treatment with mIL-3, cells were incubated with a tetrazolium salt and the OD_{450} - OD_{650} 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 mIL-3.

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 mIL-3 concentration of greater than 50 μg/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 mIL-3 to surfaces. Solubilize for 30 minutes at room temperature with occasional gentle vortexing. Stock mIL-3 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: IL-3 is produced by T cells, mast cells and eosinophils (1). Target cells include hematopoietic progenitors, neutrophils, macrophages, mast cells, eosinophils, lymphoid and erythroid cells (1). IL-3 supports growth and differentiation and is used as a media additive to support culture of many cell types (1). The IL-3 receptor is a heterodimer of the IL-3 specific α-chain and the common β-chain, βc, which is also used by GM-CSF and IL-5. (1). Binding of IL-3 can also involve substitution of the βc by a βIL-3-chain that appears to be specific for IL-3 (1,2). Binding of IL-3 to its cognate receptor(s) induces activation of Jak2, phosphorylation of multiple Stats (1,3,5,6), and the PI3K/Akt pathway (1). IL-3 may play an important role in the development of airway inflammation associated with asthma (3,4,5).

Background References:

(1) Reddy, E.P. et al. (2000) *Oncogene* 19, 2532–47.

(2) Hara, T. and Miyajima, A. (1992) *EMBO J* 11, 1875–84.

(3) Asquith, K.L. et al. (2008) *J Immunol* 180, 1199–206.

(4) Schroeder, J.T. et al. (2009) *J Immunol* 182, 2432–8.

(5) Munitz, A. et al. (2006) *J Immunol* 177, 77–83.