

DESCRIPTION

Source *E. coli*-derived human IL-18/IL-1F4 protein
Tyr37-Asp193
Accession # Q14116.1
Produced using non-animal reagents in an animal-free laboratory.
Manufactured and tested under cGMP guidelines.

N-terminal Sequence Analysis Tyr37-Phe-Gly-Lys-Leu-Glu-Ser-Lys-Leu-Ser

Predicted Molecular Mass 18 kDa

SPECIFICATIONS

SDS-PAGE 18 kDa, under reducing conditions.

Activity Measured by its ability to induce IFN- γ secretion by KG-1 human acute myelogenous leukemia cells in the presence of TNF- α .
The ED₅₀ for this effect is 1.25-15.0 ng/mL.
The specific activity of GMP Recombinant Human IL-18/IL-1F4 is > 5.00 x 10⁶ units/mg, which is calibrated against the human IL-18 WHO Standard (NIBSC code: 03/200).

Endotoxin Level <0.10 EU per 1 μ g of the protein by the LAL method.

Purity >97%, by SDS-PAGE with quantitative densitometry by Coomassie® Blue Staining.

Mass Spectrometry The molecular weight by mass spectrometry is 18205 Da \pm 5 Da.

Host Cell Protein <0.200 ng per μ g of protein when tested by ELISA.

Mycoplasma Negative for mycoplasma.

Host Cell DNA <0.00150 ng per μ g of protein when tested by PCR.

Formulation Lyophilized from a 0.2 μ m filtered solution in PBS and TCEP with Trehalose. See Certificate of Analysis for details.

PREPARATION AND STORAGE

Reconstitution Reconstitute at 500 μ g/mL in sterile water.

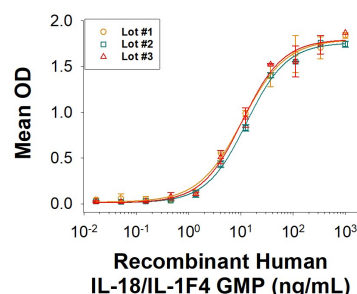
Shipping The product is shipped with polar packs. Upon receipt, store it immediately at the temperature recommended below.

Stability & Storage **Use a manual defrost freezer and avoid repeated freeze-thaw cycles.**

- A minimum of 12 months when stored at ≤ -20 °C as supplied. Refer to lot specific COA for the Use by Date.
- 1 month, 2 to 8 °C under sterile conditions after reconstitution.
- 3 months, ≤ -20 °C under sterile conditions after reconstitution.

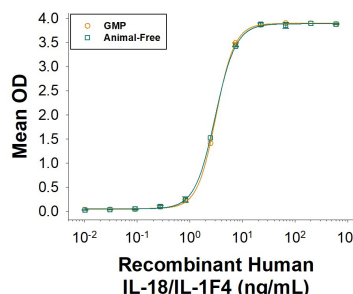
DATA

Bioactivity



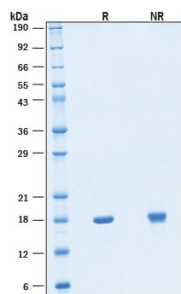
Recombinant Human IL-18/IL-1F4 GMP Protein Bioactivity. The bioactivity of Recombinant Human IL-18/IL-1F4 GMP Protein (Catalog # BT-018-GMP) was measured by its ability to induce IFN- γ secretion by KG-1 human acute myelogenous leukemia cells in the presence of TNF- α . Three independent lots were tested for bioactivity and plotted on the same graph to show lot-to-lot consistency of GMP IL-18/IL-1F4 protein.

Bioactivity



Equivalent Bioactivity of GMP and Animal-Free grades of Recombinant Human IL-18/IL-1F4. Equivalent bioactivity of GMP (Catalog # BT-018-GMP) and Animal-Free (Catalog # BT-018-AFL) grades of Recombinant Human IL-18/IL-1F4 as measured by its ability to induce IFN- γ secretion by KG-1 human acute myelogenous leukemia cells in the presence of TNF- α (orange and green, respectively).

SDS-PAGE



Recombinant Human IL-18/IL-1F4 GMP Protein SDS-PAGE. 2 μ g/lane of Recombinant Human IL-18/IL-1F4 GMP Protein (Catalog # BT-018-GMP) was resolved with SDS-PAGE under reducing (R) and non-reducing (NR) conditions and visualized by Coomassie® Blue staining, showing a band at 18 kDa under reducing conditions.

BACKGROUND

Interleukin-18 (IL-18) is a proinflammatory cytokine in the IL-1 family that exerts distinct immune effects depending on the local cytokine environment. It is expressed as a 24 kDa precursor by endothelial and epithelial cells, keratinocytes, $\gamma\delta$ T cells, and phagocytes. The precursor is activated intracellularly by Caspase-1 mediated proteolysis to release the 17 kDa mature cytokine. The precursor can also be released by necrotic cells for extracellular cleavage by multiple proteases. IL-18 activation is induced by infection or tissue damage and contributes to disease pathology in chronic inflammation (1-3). IL-18 binds to the widely expressed IL-18 R α which recruits IL-18 R β to form the signaling receptor complex (4, 5). Its bioactivity is negatively regulated by interactions with IL-18 binding proteins and virally encoded IL-18BP homologs (6). In the presence of IL-12 or IL-15, IL-18 enhances anti-viral Th1 immune responses by inducing IFN- γ production and the cytolytic activity of CD8 $^{+}$ T cells and NK cells (7, 8). In the absence of IL-12 or IL-15, however, IL-18 promotes production of the Th2 cytokines IL-4 and IL-13 by CD4 $^{+}$ T cells and basophils (9, 10). In the presence of IL-1 β or IL-23, IL-18 induces the antigen-independent production of IL-17 by $\gamma\delta$ T cells and CD4 $^{+}$ T cells (11). IL-18 also promotes myeloid dendritic cell maturation and triggers neutrophil respiratory burst (12, 13). In cancer, IL-18 exhibits diverse activities including enhancing anti-tumor immunity, inhibiting or promoting angiogenesis, and promoting tumor cell metastasis (14). Mature human IL-18 shares approximately 63% amino acid sequence identity with mouse and rat IL-18 (15). Alternative splicing in human ovarian cancer generates an isoform that is resistant to Caspase-1 activation (16). A cell surface form can be expressed on M-CSF induced macrophages and released in response to bacterial endotoxin (17).

References:

- Dinareello, C.A. *et al.* (2013) *Front. Immunol.* **4**:289.
- Smith, D.E. (2011) *J. Leukoc. Biol.* **89**:383.
- Gu, Y. *et al.* (1997) *Science* **275**:206.
- Torigoe, K. *et al.* (1997) *J. Biol. Chem.* **272**:25737.
- Cheung, H. *et al.* (2005) *J. Immunol.* **174**:5351.
- Novick, D. *et al.* (1999) *Immunity* **10**:127.
- Fehniger, T.A. *et al.* (1999) *J. Immunol.* **162**:4511.
- Yoshimoto, T. *et al.* (1998) *J. Immunol.* **161**:3400.
- Yoshimoto, T. *et al.* (2000) *Nat. Immunol.* **1**:132.
- Kroeger, K.M. *et al.* (2009) *J. Leukoc. Biol.* **86**:769.
- Lalor, S.J. *et al.* (2011) *J. Immunol.* **186**:5738.
- Li, J. *et al.* (2004) *Cell. Immunol.* **227**:103.
- Elbim, C. *et al.* (2005) *Clin. Diagn. Lab. Immunol.* **12**:436.
- Fabbi, M. *et al.* (2015) *J. Leukoc. Biol.* **97**:665.
- Ushio, S. *et al.* (1996) *J. Immunol.* **156**:4274.
- Gaggero, A. *et al.* (2004) *Oncogene* **23**:7552.
- Bellora, F. *et al.* (2012) *Eur. J. Immunol.* **42**:1618.

MANUFACTURING SPECIFICATIONS

GMP Proteins

R&D Systems, a Bio-Techne Brand's GMP proteins are produced according to relevant sections of the following documents: USP Chapter 1043, Ancillary Materials for Cell, Gene and Tissue-Engineered Products and Eu. Ph. 5.2.12, Raw Materials of Biological Origin for the Production of Cell-based and Gene Therapy Medicinal Products.

R&D Systems' quality focus includes:

- Manufactured and tested under an ISO 9001:2015 and ISO 13485:2016 certified quality system
- Documented processes and QA control of documentation and process changes
- Personnel training programs
- Raw material testing and vendor qualification/monitoring
- Fully validated equipment, processes and test methods
- Equipment calibration schedules using a computerized calibration program
- Facility maintenance, safety programs and pest control
- Material review process for variances
- Monitoring of stability over product shelf-life

R&D Systems strives to provide our customers with the analytical characteristics of each product so that customers may determine whether our products are appropriate for their research. The Certificate of Analysis provided contains the following lot specific information:

- N-terminal amino acid analysis, SDS-PAGE analysis, mass spectrometry, and endotoxin level (as determined by LAL assay) performed on each bulk QC lot, not on individual bottlings of each QC lot
- Post-bottling lot-specific bioassay results (compliance with an established range) and results of microbial testing according to USP <71>
- Host Cell Protein testing performed by ELISA
- Mycoplasma testing by ribosomal RNA hybridization assay

Additional testing and documentation requested by the customer can be arranged at an additional cost.

Production records and facilities are available for examination by appropriate personnel on-site at R&D Systems in Minneapolis, Minnesota USA.

R&D Systems sells GMP grade products for preclinical or clinical *ex vivo* use. They are not for *in vivo* use. Please read the following End User Terms prior to using this product.

Animal-Free Manufacturing Conditions

Our dedicated controlled-access animal-free laboratories ensure that at no point in production are the products exposed to potential contamination by animal components or byproducts. Every stage of manufacturing is conducted in compliance with R&D Systems' stringent Standard Operating Procedures (SOPs). Production and purification procedures use equipment and media that are confirmed animal-free.

Production

- All molecular biology procedures use animal-free media and dedicated labware.
- Dedicated fermentors are utilized in committed animal-free areas.

Purification

- Protein purification columns are animal-free.
- Bulk proteins are filtered using animal-free filters.
- Purified proteins are stored in animal-free containers in a dedicated cold storage room.

Quality Assurance

- Low Endotoxin Level.
- No impairment of biological activity.
- High quality product obtained under stringent conditions.

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