

**DESCRIPTION**

**Source** *E. coli*-derived  
Pro29-Met212  
Accession # Q75MH2  
Produced using non-animal reagents in an animal-free laboratory.  
Manufactured and tested under cGMP guidelines.

**N-terminal Sequence Analysis** Pro<sub>29</sub>-Val-Pro-Pro-Gly-Glu-Asp-Ser-Lys-Asp

**Predicted Molecular Mass** 20.9 kDa

**SPECIFICATIONS**

**SDS-PAGE** 20-21 kDa, reducing conditions

**Activity** Measured in a cell proliferation assay using T1165.85.2.1 mouse plasmacytoma cells. Nordan, R.P. *et al.* (1987) J. Immunol. **139**:813. The ED<sub>50</sub> for this effect is 0.2-0.8 ng/mL.  
The specific activity of Recombinant Human IL-6 is approximately 1.1 x 10<sup>5</sup> IU/μg, which is calibrated against human IL-6 WHO International Standard (NIBSC code: 89/548).

**Endotoxin Level** <0.10 EU per 1 μg of the protein by the LAL method.

**Purity** >97%, by SDS-PAGE under reducing conditions and visualized by silver stain.

**Host Cell Protein** <0.5 ng per μg of protein when tested by ELISA.

**Mycoplasma** Negative when tested in a ribosomal RNA hybridization assay.

**Formulation** Lyophilized from a 0.2 μm filtered solution in PBS and NaCl. See Certificate of Analysis for details.

**PREPARATION AND STORAGE**

**Reconstitution** Reconstitute at 100-200 μg/mL in sterile PBS.

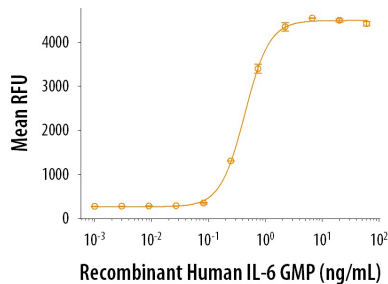
**Shipping** The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below.

**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.

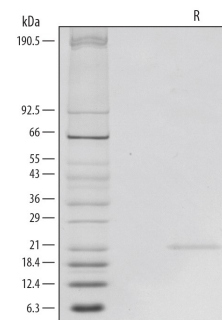
**DATA**

**Bioactivity**



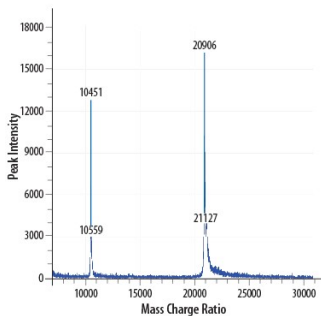
GMP-grade Recombinant Human IL-6 (Catalog # 206-GMP) stimulates proliferation of T1165.85.2.1 mouse plasmacytoma cell line. The ED<sub>50</sub> for this effect is 0.2-0.8 ng/mL.

**SDS-PAGE**



1 μg/lane of GMP-grade Recombinant Human IL-6 (Catalog # 206-GMP) was resolved with SDS-PAGE under reducing conditions (R) and visualized by silver staining, showing a single band at 21 kDa.

**Mass Spectrometry**



MALDI-TOF analysis of GMP-grade Recombinant Human IL-6 (Catalog # 206-GMP). The major peak corresponds to the calculated molecular mass, 20910 Da. The minor peak at 21127 Da is a matrix-associated artifact of the MALDI-TOF.

**BACKGROUND**

Interleukin-6 (IL-6) is a pleiotropic,  $\alpha$ -helical, 22-28 kDa phosphorylated and variably glycosylated cytokine that plays important roles in the acute phase reaction, inflammation, hematopoiesis, bone metabolism, and cancer progression (1-5). Mature human IL-6 is 183 amino acids (aa) in length and shares 39% aa sequence identity with mouse and rat IL-6 (6). Alternative splicing generates several isoforms with internal deletions, some of which exhibit antagonistic properties (7-10). IL-6 induces signaling through a cell surface heterodimeric receptor complex composed of a ligand binding subunit (IL-6 R  $\alpha$ ) and a signal transducing subunit (gp130). IL-6 binds to IL-6 R $\alpha$ , triggering IL-6 R $\alpha$  association with gp130 and gp130 dimerization (11). gp130 is also a component of the receptors for CLC, CNTF, CT-1, IL-11, IL-27, LIF, and OSM (12). Soluble forms of IL-6 R $\alpha$  are generated by both alternative splicing and proteolytic cleavage (5). In a mechanism known as trans-signaling, complexes of soluble IL-6 and IL-6 R $\alpha$  elicit responses from gp130-expressing cells that lack cell surface IL-6 R $\alpha$  (5). Trans-signaling enables a wider range of cell types to respond to IL-6, as the expression of gp130 is ubiquitous, while that of IL-6 R $\alpha$  is predominantly restricted to hepatocytes, monocytes, and resting lymphocytes (2, 5). Soluble splice forms of gp130 block trans-signaling from IL-6/IL-6 R $\alpha$  but not from other cytokines that use gp130 as a co-receptor (5, 13). IL-6, along with TNF- $\alpha$  and IL-1, drives the acute inflammatory response and the transition from acute inflammation to either acquired immunity or chronic inflammatory disease (1-5). When dysregulated, it contributes to chronic inflammation in obesity, insulin resistance, inflammatory bowel disease, arthritis, sepsis, and atherosclerosis (1, 2, 5). IL-6 can also function as an anti-inflammatory molecule, as in skeletal muscle where it is secreted in response to exercise (2). In addition, it enhances hematopoietic stem cell proliferation and the differentiation of Th17 cells, memory B cells, and plasma cells (1, 14).

**References:**

1. Mansell, A. and B.J. Jenkins (2013) Cytokine Growth Factor Rev. **24**:249.
2. Schuett, H. *et al.* (2009) Thromb. Haemost. **102**:215.
3. Erta, M. *et al.* (2012) Int. J. Biol. Sci. **8**:1254.
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6. Hirano, T. *et al.* (1986) Nature **324**:73.
7. Kestler, D.P. *et al.* (1995) Blood **86**:4559.
8. Kestler, D.P. *et al.* (1999) Am. J. Hematol. **61**:169.
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11. Murakami, M. *et al.* (1993) Science **260**:1808.
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13. Mitsuyama, K. *et al.* (2006) Clin. Exp. Immunol. **143**:125.
14. Cerutti, A. *et al.* (1998) J. Immunol. **160**:2145.

## MANUFACTURING SPECIFICATIONS

### GMP Proteins

R&D Systems, a Bio-Techne Brand's GMP proteins are produced according to relevant sections of the following documents: WHO TRS, No. 822, 1992 Annex 1, Good Manufacturing Practices for Biological Products; USP Chapter 1043, Ancillary Materials for Cell, Gene and Tissue-Engineered Products and USP Chapter 92, Growth Factors and Cytokines Used in Cell Therapy Manufacturing.

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, 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 bioburden testing (using broth culture, Sabourand's dextrose and blood agar plates with results reported at 3 days and at 7 days)
- 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. Testing may include, but is not limited to, USP< 61> bioburden testing, positive identity testing, testing for adventitious agents and testing for residual host cell content.

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 its GMP grade recombinant protein products for research use or further manufacturing use in *ex vivo* cell therapy applications. They are not for *in vivo* use or for use as therapeutic or other drugs, biologic products or devices. 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.
- For *ex vivo* research or bioproduction, [additional documentation](#) can be provided.

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