

**DESCRIPTION**

<b>Source</b>	<i>E. coli</i> -derived human IL-6 protein Pro29-Met212 Accession # Q75MH2 Produced using non-animal reagents in an animal-free laboratory. Manufactured and tested under cGMP guidelines.
<b>N-terminal Sequence Analysis</b>	Pro <sub>29</sub> -Val-Pro-Pro-Gly-Glu-Asp-Ser-Lys-Asp
<b>Predicted Molecular Mass</b>	20.9 kDa

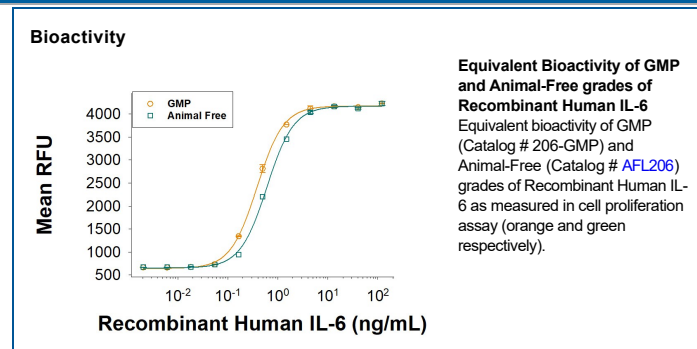
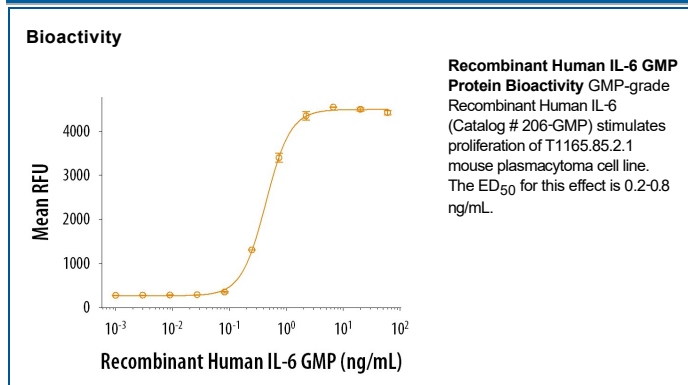
**SPECIFICATIONS**

<b>SDS-PAGE</b>	20-21 kDa, reducing conditions
<b>Activity</b>	Measured in a cell proliferation assay using T1165.85.2.1 mouse plasmacytoma cells. Nordan, R.P. <i>et al.</i> (1987) J. Immunol. <b>139</b> :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 >1.0 x 10 <sup>8</sup> IU/mg, which is calibrated against the human IL-6 WHO International Standard (NIBSC code: 89/548).
<b>Endotoxin Level</b>	<0.10 EU per 1 µg of the protein by the LAL method.
<b>Purity</b>	>97%, by SDS-PAGE under reducing conditions and visualized by silver stain.
<b>Host Cell Protein</b>	<0.5 ng per µg of protein when tested by ELISA.
<b>Mycoplasma</b>	Negative when tested in a ribosomal RNA hybridization assay.
<b>Host Cell DNA</b>	<0.0015 ng of DNA per µg of protein when tested by PCR.
<b>Formulation</b>	Lyophilized from a 0.2 µm filtered solution in PBS and NaCl. See Certificate of Analysis for details.

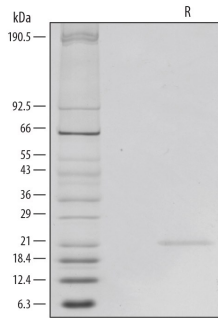
**PREPARATION AND STORAGE**

<b>Reconstitution</b>	Reconstitute at 100-200 µg/mL in sterile PBS.
<b>Shipping</b>	The product is shipped with polar packs. Upon receipt, store it immediately at the temperature recommended below.
<b>Stability &amp; Storage</b>	<b>Use a manual defrost freezer and avoid repeated freeze-thaw cycles.</b> <ul style="list-style-type: none"> <li>A minimum of 12 months when stored at ≤ -20 °C as supplied. Refer to lot specific COA for the Use by Date.</li> <li>1 month, 2 to 8 °C under sterile conditions after reconstitution.</li> <li>3 months, ≤ -20 °C under sterile conditions after reconstitution.</li> </ul>

**DATA**

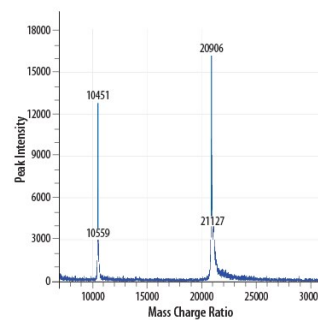


**SDS-PAGE**



**Recombinant Human IL-6 GMP Protein 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**



**Recombinant Human IL-6 GMP Protein 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:**

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2. Schuett, H. *et al.* (2009) Thromb. Haemost. **102**:215.
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4. Garbers, C. *et al.* (2012) Cytokine Growth Factor Rev. **23**:85.
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**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.

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- 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

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- 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 testing according to USP
- 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.

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- Purified proteins are stored in animal-free containers in a dedicated cold storage room.

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- High quality product obtained under stringent conditions.

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