

**DESCRIPTION**

<b>Source</b>	Chinese Hamster Ovary cell line, CHO-derived human TGF-beta 1 protein Ala279-Ser390 Accession # P01137 Manufactured and tested under cGMP guidelines.
<b>N-terminal Sequence Analysis</b>	Ala-Leu-Asp-Thr-Asn-Tyr-(Cys)-Phe-Ser-Ser
<b>Structure / Form</b>	Disulfide-linked homodimer
<b>Predicted Molecular Mass</b>	12.8 kDa (monomer)

**SPECIFICATIONS**

<b>SDS-PAGE</b>	12 kDa, reducing conditions 24 kDa, non-reducing conditions
<b>Activity</b>	Measured by its ability to inhibit the IL-4-dependent proliferation of HT-2 mouse T cells. Tsang, M. <i>et al.</i> (1995) Cytokine 7:389. The ED <sub>50</sub> for this effect is 0.04-0.2 ng/mL. The specific activity of recombinant human TGF-β1 GMP is approximately 2.5 x 10 <sup>4</sup> U/μg, which is calibrated against human TGF-β1 Standard (NIBSC code: 89/514).
<b>Endotoxin Level</b>	<0.10 EU per 1 μg of the protein by the LAL method.
<b>Purity</b>	>97%, by SDS-PAGE with silver staining.
<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>Adventitious Virus</b>	Master Cell Bank tested for adventitious viruses
<b>Formulation</b>	Lyophilized from a 0.2 μm filtered solution in Acetonitrile and TFA. See Certificate of Analysis for details.

**PREPARATION AND STORAGE**

<b>Reconstitution</b>	Reconstitute at 100 μg/mL in 4 mM HCl.
<b>Shipping</b>	The product is shipped at ambient temperature. 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**

<p><b>Bioactivity</b></p> <p>Mean RFU</p> <p>Recombinant Human TGF-β1 GMP (ng/mL)</p> <p>GMP-grade Recombinant Human TGF-β1 (Catalog # 240-GMP) inhibits Recombinant Mouse IL-4 (Catalog # 404-ML) induced proliferation in the HT-2 mouse T cell line. The ED<sub>50</sub> for this effect is 0.04-0.2 ng/mL.</p>	<p><b>SDS-PAGE</b></p> <p>1 μg/lane of GMP-grade Recombinant Human TGF-β1 (Catalog # 240-GMP) was resolved with SDS-PAGE under reducing (R) and nonreducing (NR) conditions and visualized by silver staining, showing single bands at 12 kDa and 24 kDa, respectively.</p>
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## BACKGROUND

TGF-β1 (transforming growth factor beta 1) is one of three closely related mammalian members of the large TGF-β superfamily that share a characteristic cystine knot structure (1-7). TGF-β1, -2 and -3 are highly pleiotropic cytokines that are proposed to act as cellular switches that regulate processes such as immune function, proliferation and epithelial-mesenchymal transition (1-4). Each TGF-β isoform has some non-redundant functions; for TGF-β1, mice with targeted deletion show defects in hematopoiesis and endothelial differentiation, and die of overwhelming inflammation (2). Human TGF-β1 cDNA encodes a 390 amino acid (aa) precursor that contains a 29 aa signal peptide and a 361 aa proprotein (8). A furin-like convertase processes the proprotein to generate an N-terminal 249 aa latency-associated peptide (LAP) and a C-terminal 112 aa mature TGF-β1 (8, 9). Disulfide-linked homodimers of LAP and TGF-β1 remain non-covalently associated after secretion, forming the small latent TGF-β1 complex (8-10). Covalent linkage of LAP to one of three latent TGF-β binding proteins (LTBPs) creates a large latent complex that may interact with the extracellular matrix (9, 10). TGF-β is activated from latency by pathways that include actions of the protease plasmin, matrix metalloproteinases, thrombospondin 1 and a subset of integrins (10). Mature human TGF-β1 shares 100% aa identity with pig, dog and cow TGF-β1, and 99% aa identity with mouse, rat and horse TGF-β1. It demonstrates cross-species activity (1). TGF-β1 signaling begins with high-affinity binding to a type II ser/thr kinase receptor termed TGF-β RII. This receptor then phosphorylates and activates a second ser/thr kinase receptor, TGF-β RI (also called activin receptor-like kinase (ALK) -5), or alternatively, ALK-1. This complex phosphorylates and activates Smad proteins that regulate transcription (3, 11, 12). Contributions of the accessory receptors betaglycan (also known as TGF-β RIII) and endoglin, or use of Smad-independent signaling pathways, allow for disparate actions observed in response to TGF-β in different contexts (11).

## References:

1. Derynck, R. and K. Miyazono (2008) Cold Spring Harbor Laboratory Press, 29.
2. Dunker, N. and K. Kriegstein (2000) Eur. J. Biochem. **267**:6982.
3. Wahl, S.M. (2006) Immunol. Rev. **213**:213.
4. Chang, H. et al. (2002) Endocr. Rev. **23**:787.
5. Lin, J.S. et al. (2006) Reproduction **132**:179.
6. Hinck, A.P. et al. (1996) Biochemistry **35**:8517.
7. Mittl, P.R.E. et al. (1996) Protein Sci. **5**:1261.
8. Derynck, R. et al. (1985) Nature **316**:701.
9. Miyazono, K. et al. (1988) J. Biol. Chem. **263**:6407.
10. Oklu, R. and R. Hesketh (2000) Biochem. J. **352**:601.
11. de Caestecker, M. et al. (2004) Cytokine Growth Factor Rev. **15**:1.
12. Zuniga, J.E. et al. (2005) J. Mol. Biol. **354**:1052.

## 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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- 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

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