Chinese Hamster Ovary cell line, CHO-derived human TGF-beta 1 protein

Accession # P01137

N-terminal Sequence Analysis

Structure / Form

Disulfide-linked homodimer

Predicted Molecular Mass

12.8 kDa (monomer)

SDS-PAGE

12 kDa, reducing conditions

24 kDa, non-reducing conditions

Activity

Measured by its ability to inhibit the IL-4-dependent proliferation of HT-2 mouse T cells. Tsang, M. et al. (1995) Cytokine 7:389.

The ED₅₀ for this effect is 0.04-0.2 ng/mL.

The specific activity of Recombinant Human TGF-β1 is approximately 2.5 x 10⁴ U/μg, which is calibrated against human TGF-β1 Standard (NIBSC code: 89/514). Specific activity is for reference purposes only and is not routinely tested.

Endotoxin Level

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

Purity

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

Formulation

Lyophilized from a 0.2 μm filtered solution in Acetonitrile and TFA with BSA as a carrier protein. See Certificate of Analysis for details.

Reconstitute 2 μg vials at 20 μg/mL in sterile 4 mM HCl containing 1 mg/mL human or bovine serum albumin. Reconstitute 10 μg or larger vials at 100 μg/mL in sterile 4 mM HCl containing 1 mg/mL human or bovine serum albumin.

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

Use a manual defrost freezer and avoid repeated freeze-thaw cycles.

- 12 months from date of receipt, -20 to -70 °C as supplied.
- 1 month, 2 to 8 °C under sterile conditions after reconstitution.
- 3 months, -20 to -70 °C under sterile conditions after reconstitution.

Bioactivity

Recombinant Human TGF-beta 1 Protein

Bioactivity

Recombinant Human TGF-beta 1 (Catalog # 240-B) inhibits Recombinant Mouse IL-4 (Catalog # 404-ML) induced proliferation in the HT-2 mouse T cell line. The ED₅₀ for this effect is 0.04-0.2 ng/mL.

SDS-PAGE

1 μg/lane of Recombinant Human TGF-beta 1 Protein SDS-PAGE 1 μg/lane of Recombinant Human TGF-beta 1 was resolved with SDS-PAGE under reducing (R) and non-reducing (NR) conditions and visualized by silver staining, showing single bands at 12 kDa and 24 kDa, respectively.
Bioactivity

Recombinant Human TGF-beta 1 Protein Bioactivity
Epithelial to Mesenchymal Transition (EMT) was induced in the A549 human lung carcinoma cell line with cell culture media supplemented with Recombinant Human (rh) TGF-beta 1 (Catalog # 240-B). Control cells were cultured without rhTGF-beta 1. EMT induction was confirmed at 48 h by flow cytometric staining for E-Cadherin (filled; Catalog # FAB18381P), an epithelial cell marker, or an isotype control (Catalog # Catalog # IC0041P). TGF-beta 1 decreased the expression of E-Cadherin.

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 metalloproteases, 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: