Miltenyi Biotec

Human TGF-β1
premium grade

Contents

1. Description

Products Human TGF-β1, premium grade.

Recombinant human transforming growth factor β1.

Content in µg	Order no.
5	130-095-067
25	130-095-066
100	130-108-969
1000	130-108-971

Biological activity The ED_{50} is ≤0.2 ng/mL corresponding to an activity of ≥5×10^6 U/mg. Lot-specific activities are stated in the Certificate of Analysis (www.miltenyibiotec.com/certificates).

▲ **Note:** The ED50 is determined by inhibition assay using IL-5 induced TF-1 cells according to Randall *et al.* The proliferation assay was calibrated with the standard for human TGF-β1 (NIBSC code 89/514) provided by the WHO/National Institute for Biological Standards and Control.

Primary structure Two identical, non-glycosylated disulfide-linked polypeptide chains (112 amino acid residues without LAP).

Molecular mass 25.6 kDa (dimer).

Source Produced in HEK293 cells.

Product format 5 µg, 25 µg, 100 µg: Lyophilized from a filtered (0.2 µm) buffer solution.

1000 µg: Liquid, filtered (0.2 µm) acetic acid solution (40 mM) without stabilizers.

Protein concentration 1000 µg: 0.5 mg/mL

Stabilizer Mannitol and trehalose.

Purity >97% as determined by SDS-PAGE analysis.

Endotoxin level Low endotoxin (<0.1 EU/µg cytokine) as determined by Limulus Amebocyte Lysate (LAL) assay.

Storage Human TGF-β1, premium grade should be stored at –20 °C. The expiration date is indicated on the vial label. Aliquots should be stored at –20 °C or below. Avoid repeated freeze-thaw cycles. Further dilutions should be prepared with 0.1 % bovine serum albumin (BSA) or human serum albumin (HSA) in phosphate-buffered saline.

Reconstitution It is recommended to reconstitute lyophilized Human TGF-β1, premium grade (5 µg, 25 µg, 100 µg) with deionized sterile-filtered water in a minimal volume of 250 µL.

1.1 Background information

Transforming growth factor β1 (TGF-β1) belongs to a family of homologous, disulfide-linked, homodimeric proteins. These highly pleiotropic cytokines inhibit proliferation of most cells, but can promote the growth of mesenchymal cells and enhance extracellular matrix formation. The pivotal function of TGF-β1 in the immune system is to mediate immunosuppression and maintain tolerance by regulating lymphocyte proliferation, differentiation, and survival. In addition, TGF-β1 controls inflammatory responses through chemotactic attraction and activation of inflammatory cells and fibroblasts. TGF-β1 is produced by many cell types, but is reported to be most abundant in mammalian platelets and bone. All three TGF-β members are synthesized as an homodimeric precursor of 390 residues, which is intracellularly processed by proteolysis into a 112 aa form. The resulting N-terminal latency-associated peptide (LAP) remains non-covalently associated with the TGF-β dimer, and the complex binds to another protein called Latent TGF-β-Binding Protein (LTBP), forming a larger complex called Large Latent Complex (LLC). The LLC is secreted into the extracellular matrix, and prevents the binding of TGF-β to its specific cell surface receptor. Several extracellular factors such as matrix metalloproteases, low pH, reactive oxigen species and thrombospondin-1 can induce release of the active mature TGF-β dimer from the inactive complex. This sophisticated mechanism of activation is important for a fine-tuning of TGF-β signaling. Human TGF-β1 is a recombinant homodimer corresponding to the fully mature form of TGF-β1 without LAP. The amino acid sequence of human TGF-β1 shares 99% identity with TGF-β1 from mouse and rat, therefore human TGF-β1 is commonly used also for mouse cell culture.

1.2 Applications

Human TGF-β1 can be used for a variety of applications, including:

- *In vitro* differentiation of naive $CD4^+$ T cells towards Th17 cells.
- *In vitro* generation of $FoxP3^+$ inducible regulatory T cells (iTregs).

140-002-831.05

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- Embryonic stem cell differentiation models, for example, for vasculogenesis and angiogenesis.
- *In vitro* chondrogenesis of mesenchymal progenitor cells and redifferentiation of expanded chondrocytes.

Optimal concentration for a specific application should be determined by a dose-response experiment.

2. References

1. Randall, L. A. *et al.* (1993) A novel, sensitive bioassay for transforming growth factor β. J. Immunol. Methods 164: 61–67.
2. Souza-Fonseca-Guimaraes, F. *et al.* (2012) NK cell tolerance to TLR agonists mediated by regulatory T cells after polymicrobial sepsis. J. Immunol. 188 (12): 5850–5858.
3. Nguyen, T. L. *et al.* (2011) Antigen-specific TGF-β-induced regulatory T cells secrete chemokines, regulate T cell trafficking, and suppress ongoing autoimmunity. J. Immunol. 187 (4): 1745–1753.

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