

Recombinant Human Granulocyte-Colony Stimulating Factor (CHO)

Certificate of Analysis and Data Sheet

➤ Source: CHO cells	➤ Catalog No. CTK-329
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➤ **Background :**

A glycoprotein of MW 20 kDa containing internal disulfide bonds. It induces the survival, proliferation, and differentiation of neutrophilic granulocyte precursor cells and functionally activates mature blood neutrophils. Among the family of colony-stimulating factors, G-CSF is the most potent inducer of terminal differentiation to granulocytes and macrophages of leukemic myeloid cell lines.

The synthesis of G-CSF can be induced by bacterial endotoxins , TNF , Interleukin-1 and GM-CSF . Prostaglandin E2 inhibits the synthesis of G-CSF. In epithelial, endothelial, and fibroblastic cells secretion of G-CSF is induced by Interleukin-17 .

➤ **Description :**

Recombinant Human Granulocyte Colony Stimulating Factor is a glycosylated, polypeptide chain containing 174 amino acids and having a molecular mass of 20 KD.

Human G-CSF is purified by proprietary chromatographic techniques.

➤ **Physical Appearance:**

Sterile Filtered White lyophilized (freeze-dried) powder.

➤ **Formulation:**

Human G-CSF was lyophilized from a concentrated (1mg/ml) solution containing 10mM Hydrochloric Acid pH=6.5, 0.4mg tween 20, 100mg mannitol, 160 mg L-arginine, 40 mg phenylalanin and 4mg methionin.

➤ **Solubility:**

It is recommended to reconstitute the lyophilized G-CSF in sterile 18MO-cm H2O not less than 100µg/ml, which can then be further diluted to other aqueous solutions.

➤ **Stability:**

Lyophilized G-CSF although stable at room temperature for 3 weeks, should be stored desiccated below -18 C. Upon reconstitution G-CSF should be stored at 4 C between 2-7 days and for future use below -18 C. For long term storage it is recommended to add a carrier protein (0.1% HSA or BSA).

Please avoid freeze-thaw cycles.

➤ **Purity:**

Greater than 98.0% as determined by:

- (a) Analysis by RP-HPLC.
- (b) Anion-exchange FPLC.
- (c) Analysis by reducing and non-reducing SDS-PAGE Silver Stained.

➤ **Amino-Acid Sequence :**

The sequence of the first five N-terminal amino acids was determined and was found to be

Thr-Pro-Leu-Gly-Pro.

➤ **Dimers and aggregates:**

Less than 1% as determined by silver-stained SDS-PAGE gel analysis.

➤ **Biological Activity:**

This G-CSF is fully biologically active when compared to standard. The ED₅₀, calculated by the dose-dependant proliferation of murine NFS-60 indicator cells (measured by ³H-thymidine uptake) is less than 0.1 ng/ml, corresponding to a Specific Activity of 1.27 x 10⁸ IU/mg.

➤ **Endotoxin:**

Less than 0.1 ng/μg (IEU/μg) of Recombinant Human Granulocyte Colony Stimulating Factor.

➤ **Protein content:**

G-CSF protein quantitation was carried out by two independent methods:

1. UV spectroscopy at 280 nm using the absorbency value of 0.815 as the extinction coefficient for a 0.1% (1mg/ml) solution. This value is calculated by the PC GENE computer analysis program of protein sequences (IntelliGenetics).
2. Analysis by RP-HPLC, using a calibrated solution of Recombinant Human Granulocyte Colony Stimulating Factor as a Reference Standard.

➤ **Usage:**

This material is offered for research, laboratory or further evaluation purposes.