Recombinant Human Epo (Tissue Culture Grade)

Catalog Number: 287-TC

Specifications and Use

Source

- A DNA sequence encoding the human EPO precursor protein (Jacobs, K. *et al*., 1985, Nature **313**:806 - 810) was expressed in a Chinese hamster ovary cell line.
- **Note:** *This is one of multiple forms available for this protein. Check R&D Systems' website,* ***www.RnDSystems.com****, for a complete listing of the variants.*

Molecular Mass

- Mature human EPO, containing 165 amino acid residues, has a predicted molecular mass of approximately 21 kDa. As a result of glycosylation, the recombinant protein migrates with an apparent molecular mass of 37 kDa in SDS-PAGE.

Endotoxin Level

- < 0.1 EU per 100 units of the cytokine as determined by the LAL method.

Activity

- Calibrated against the second international reference preparation of erythropoietin (Annable, L. *et al*., 1972. Bull. Hlth. Org. **47**:99).
- The *in vitro* biological activity of this preparation was measured in a cell proliferation assay using a factor-dependent human erythroleukemic cell line TF-1 (Kitamura, T. *et al*., 1989, J. Cell. Physiol. **140**:323).
- The ED_{50} for this effect is typically 0.015 - 0.075 units/mL.

Formulation

- Lyophilized from a 0.2 μm filtered solution of 0.025% BSA in PBS.

Reconstitution

- It is recommended that sterile phosphate-buffered saline containing at least 0.1% human serum albumin or bovine serum albumin be added to the vial to prepare a stock solution of no less than 500 U/mL of the cytokine.

Storage

- Lyophilized samples are stable for up to twelve months from date of receipt at -20° C to -70° C.
- Upon reconstitution, this cytokine can be stored under sterile conditions at 2° - 8° C for one month or at -20° C to -70° C **in a manual defrost freezer** for three months without detectable loss of activity.
- **Avoid repeated freeze-thaw cycles.**

Human Erythropoietin

Erythropoietin (Epo) is a 34 kDa glycoprotein hormone in the type I cytokine family and is related to thrombopoietin (1). Its three N-glycosylation sites, four alpha helices, and N- to C-terminal disulfide bond are conserved across species (2, 3). Glycosylation of Epo is required for biological activities *in vivo* (4). Mature human Epo shares 75% - 84% amino acid sequence identity with bovine, canine, equine, feline, mouse, ovine, porcine, and rat EPO. Epo is primarily produced in the kidney by a population of fibroblast-like cortical interstitial cells adjacent to the proximal tubules (5). It is also produced in much lower, but functionally significant amounts by fetal hepatocytes and in adult liver and brain (6 - 8). Epo promotes erythrocyte formation by preventing the apoptosis of early erythroid precursors which express the Epo receptor (Epo R) (8, 9). Epo R has also been described in brain, retina, heart, skeletal muscle, kidney, endothelial cells, and a variety of tumor cells (7, 8, 10, 11). Ligand induced dimerization of Epo R triggers JAK2-mediated signaling pathways followed by receptor/ligand endocytosis and degradation (1, 12). Rapid regulation of circulating Epo allows tight control of erythrocyte production and hemoglobin concentrations. Anemia or other causes of low tissue oxygen tension induce Epo production by stabilizing the hypoxia-inducible transcription factors HIF-1α and HIF-2α (1, 6). Epo additionally plays a tissue-protective role in ischemia by blocking apoptosis and inducing angiogenesis (7, 8, 13).

References:

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R&D Systems, Inc.
1-800-343-7475