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Certificate of Analysis

IGF-I

(recombinant protein expressed in *E. coli*)

Catalog # 01-208

Lot # 23329

Source: Recombinant human IGF-I expressed in *E. coli*. Purity >95% as determined by HPLC. Mr ~ 7649Da.

Purity: Single protein band on SDS-PAGE gel.

Sterility: Sterilized through a 0.2 μ membrane filter and packaged aseptically.

Storage and Stability: Lyophilized: 2 years at 4°C. Rehydrated: 6 months at -20°C. Rehydrated solutions can be stored for 6 months at -20°C without detectable loss of activity. More dilute solutions stored at -20°C are less stable. Aliquot to avoid repeated freezing and thawing.

Formulation: 25 μ g/vial. Lyophilized from 100 μ l 0.1M acetic acid. Lyophilized powder.

Rehydration: Rehydrate in 0.1M acetic acid or 10mM HCl.

**FOR IN VITRO RESEARCH USE ONLY
NOT FOR USE IN HUMANS OR ANIMALS**

Quality Control Testing and Research Applications

Biological Activity: 5ng/ml of this lot supported a 1.4-fold increase in chicken embryo fibroblasts over control cultures without IGF-I. Cell viability was assessed using an ATP endpoint assay (ATPLite™-M, Packard Instruments). The optimal IGF-I concentration in cell culture can vary depending on cell type.

IGF Growth Assay Protocol

1. 2 days prior to the assay, seed 100 μ l of chicken embryo fibroblasts (CEF) in 2% FBS to a density of 5x10⁴ cells/ml in a 96 well tissue culture plate.
2. 1 day prior to the assay, serum starve by adding 100 μ l serum free media to the CEF.
3. On the day of the assay, aspirate off the media and add 100 μ l **IGF-I** (0.1-20ng/ml) in serum free media. Incubate for 24 hours at 37°C, 5% CO₂. Include appropriate positive and negative controls.
4. Assay for cell number using ATP endpoint assay per vendor's instructions. Read ATP levels and compare readings to control wells containing cells not incubated with IGF-I.