Animal-Free™ Recombinant Human EGF

Catalog Number: BT-EGF-AFL

DESCRIPTION

Source	*E. coli*-derived human EGF protein Asn971-Arg1023, with an N-terminal Met Accession # P01133.2 Produced using non-animal reagents in an animal-free laboratory.
N-terminal Sequence Analysis	Met
Predicted Molecular Mass	6 kDa

SPECIFICATIONS

SDS-PAGE	7 kDa, under reducing conditions.
Activity	Measured in a cell proliferation assay using Balb/3T3 mouse embryonic fibroblast cells. Rubin, J.S. *et al.* (1991) Proc. Natl. Acad. Sci. USA **88**:415. The ED_{50} for this effect is 20.0-100 pg/mL. The specific activity of Recombinant Human EGF is >8.0 x 10^5 IU/mg, which is calibrated against the human EGF WHO International Standard (NIBSC code: 91/530).
Endotoxin Level	<0.10 EU per 1 µg of the protein by the LAL method.
Purity	>97%, by SDS-PAGE with quantitative densitometry by Coomassie® Blue Staining.
Formulation	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details.

PREPARATION AND STORAGE

Reconstitution	Reconstitute at 500 µg/mL in sterile water.
Shipping	The product is shipped with polar packs. Upon receipt, store it immediately at the temperature recommended below.
Stability & Storage	**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.

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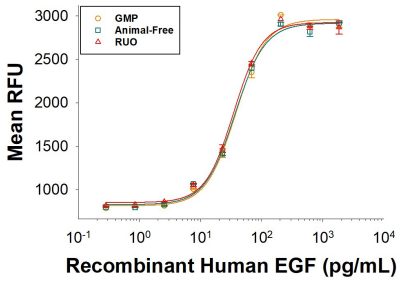
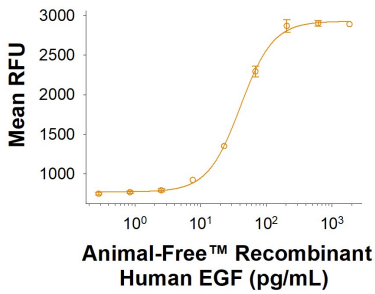
DATA

Bioactivity



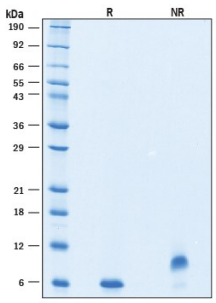
Equivalent Bioactivity of GMP, Animal-Free and RUO grades of Recombinant Human EGF. Equivalent bioactivity of GMP (Catalog # BT-EGF-GMP), Animal-Free (Catalog # BT-EGF-AFL) and RUO (Catalog # 236-EG) grades of Recombinant Human EGF as measured in a cell proliferation assay using the cell proliferation of Balb/3T3 mouse embryonic fibroblast cell line (orange, green, red, respectively).

Bioactivity



Animal-Free™ Recombinant Human EGF Protein Bioactivity. Animal-Free™ Recombinant Human EGF Protein (Catalog # BT-EGF-AFL) induces cell proliferation of Balb/3T3 mouse embryonic fibroblast cells.

SDS-PAGE



Animal-Free™ Recombinant Human EGF Protein SDS-PAGE. 2 µg/lane of Animal-Free™ Recombinant Human EGF Protein (Catalog # BT-EGF-AFL) was resolved with SDS-PAGE under reducing (R) and non-reducing (NR) conditions and visualized by Coomassie® Blue staining, showing a band at 7 kDa under reducing conditions.

BACKGROUND

Epidermal growth factor (EGF) is a small, potent growth factor capable of inducing cell proliferation, differentiation, and survival. EGF is the founding member of the EGF family that also includes TGF-alpha, amphiregulin (AR), betacellulin (BTC), epiregulin (EPR), heparin-binding EGF-like growth factor (HB-EGF), epigen, and the neuregulins (NRG)-1 through -6 (1). Members of The EGF family are characterized by a shared structural motif, the EGF-like domain, which contains three intramolecular disulfide bonds that are formed by six similarly spaced, conserved cysteine residues (2). These disulfide bonds are essential for proper protein conformation and receptor binding. All EGF family members are synthesized as type I transmembrane precursor proteins that may contain several EGF domains in the extracellular region. The mature proteins are released from the cell surface by regulated proteolysis (1). The full length EGF protein is 1207 amino acids (aa) (EGF precursor) containing nine EGF domains and nine LDLR class B repeats. However, the mature protein is much smaller, only 53 aa, and is generated by proteolytic cleavage of the EGF domain proximal to the transmembrane region (3). EGF is well conserved across mammals with mature human EGF 70% identical to mature mouse and rat EGF. Physiologically, EGF is found in various body fluids, including blood, milk, urine, saliva, seminal fluid, pancreatic juice, cerebrospinal fluid, and amniotic fluid (4). EGF is a high affinity ligand of the EGF receptor (ErbB). Four ErbB (HER) family receptor tyrosine kinases including EGFR/ErbB1, ErbB2, ErbB3 and ErbB4, mediate responses to EGF family members (5). EGF binding induces dimerization of the EGF receptor resulting in activation of the protein tyrosine kinase signaling pathway. These receptors undergo a complex pattern of ligand-induced homo- or hetero-dimerization to transduce EGF family signals (6, 7). EGF binds ErbB1 and depending on the context, induces the formation of homodimers or heterodimers containing ErbB2. Dimerization results in autophosphorylation of the receptor at specific tyrosine residues to create docking sites for a variety of signaling molecules (5, 8). Biological activities ascribed to EGF include epithelial development, angiogenesis, inhibition of gastric acid secretion, fibroblast proliferation, and colony formation of epidermal cells in culture.

References:

1. Harris, R.C. *et al.* (2003) Exp. Cell Res. **284**:2.
2. Carpenter, G. and Cohen, S. (1990) J. Biol. Chem. **265**:7709.
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5. Jorissen, R.N. *et al.* (2003) Exp. Cell Res. **284**:31.
6. Gamett, D.C. *et al.* (1997) J. Biol. Chem. **272**:12052.
7. Qian, X. *et al.* (1994) Proc. Natl. Acad. Sci. **91**:1500.
8. Qian, X. *et al.* (1999) J. Biol. Chem. **274**:574.

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MANUFACTURING SPECIFICATIONS

Animal-Free Manufacturing Conditions

Our dedicated controlled-access animal-free laboratories ensure that at no point in production are the products exposed to potential contamination by animal components or byproducts. Every stage of manufacturing is conducted in compliance with R&D Systems' stringent Standard Operating Procedures (SOPs). Production and purification procedures use equipment and media that are confirmed animal-free.

Production

- All molecular biology procedures use animal-free media and dedicated labware.
- Dedicated fermentors are utilized in committed animal-free areas.

Purification

- Protein purification columns are animal-free.
- Bulk proteins are filtered using animal-free filters.
- Purified proteins are stored in animal-free containers in a dedicated cold storage room.

Quality Assurance

- Low Endotoxin Level.
- No impairment of biological activity.
- High quality product obtained under stringent conditions.

Please read our complete Animal-Free Statement

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