Product Information

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Anti-Keratinocyte Growth Factor
produced in goat, affinity isolated antibody

Catalog Number **K4760**

Synonyms: Anti-KGF; Anti-FGF-7

Product Description
Anti-Keratinocyte Growth Factor is produced in goat using as immunogen a purified recombinant human KGF/fibroblast growth factor 7 expressed in *E. coli*. FGF-7 specific IgG was purified by human FGF-7 affinity chromatography.

Anti-Keratinocyte Growth Factor recognizes recombinant human FGF-7 by various immunochemical techniques including neutralization, immunoblotting, immunohistochemistry, and ELISA. Based on ELISA and immunoblotting, this antibody shows less than 20% cross-reactivity with recombinant human FGF-9 and less than 10% cross-reactivity with recombinant human FGF-4.

Fibroblast growth factors (FGFs) are members of a large family of structurally related polypeptides (17-38 kDa) that exert biological activities toward cells of mesenchymal, neuronal, and epithelial origin.[1, 2] All members of the FGF superfamily have two conserved cysteine residues and a conserved 120 amino acid core region that contains six identical, interspersed amino acids.[3-5] All FGFs share 30-50 % amino acid sequence identity. FGFs are involved in normal development, wound healing and repair, angiogenesis, and a variety of neurotrophic activities. They are also involved in hematopoiesis as well as in tissue remodeling and maintenance. FGFs are potent physiological regulators of growth and differentiation for a variety of cells of mesodermal, ectodermal, and endodermal origin. They have been implicated in pathological conditions such as tumorigenesis and metastasis. To date, the FGF family consists of 23 members designated FGF-1 through FGF-23.[5]

Four distinct tyrosine kinase FGF receptors (FGFRs) from four separate genes have been identified: FGFR-1 (flg, cek-1), FGFR-2 (bek, cek-3), FGFR-3 (cek-2), and FGFR-4.[6-8] The high affinity cell surface FGF receptors have an extracellular region containing three immunoglobulin-like domains, a transmembrane region, and a cytosolic tyrosine kinase domain activated by ligand binding. Multiple additional variants (isoforms) arising from alternative splicing have also been reported.[7] Ligand binding specificity, signal transduction, and membrane attachment may be modified by alternative splicings.

Keratinocyte Growth Factor (KGF, FGF-7) was originally isolated from the conditioned medium of a human embryonic lung fibroblast cell line as a mitogen specific for epithelial cells. The transcript for KGF is detected in stromal but not epithelial cells. It has been suggested that KGF is a mesenchymal cell-derived paracrine growth factor that stimulates epithelial cell growth. The N-terminal residues (31 amino acid residues) of KGF are cleaved from the KGF precursor protein (194 amino acid residues) to generate the mature form of KGF (163 amino acid residues).[9] Human KGF shows species cross-reactivity and is active on mouse, monkey, and porcine cells. Mature human and mouse FGF-7 have 96 % amino acid sequence identity, while human and rat FGF-7 have 92 % amino acid sequence identity.[9]

KGF is expressed in a number of stromal fibroblast cell lines but is absent from normal glial cells and epithelial cell lines. KGF released from stromal and dermal fibroblasts stimulates proliferation and differentiation of keratinocytes and other cells in the epithelium. Adult cells known to express FGF-7 include fibroblasts, γδ T cells, smooth muscle cells, and ovarian theca cells. In the embryo, KGF is expressed at many stages of development throughout the mesenchyme.

Reagent
Supplied lyophilized from a 0.2 μm filtered solution of phosphate buffered saline with 5% trehalose.

Preparation Instructions
To one vial of lyophilized powder, add 1 mL of sterile phosphate buffered saline, pH 7.4, to produce a 0.25 mg/mL stock solution of antibody.

Storage/Stability
Prior to reconstitution, store at –20 °C. Reconstituted product may be stored at 2-8 °C for up to one month without detectable loss of activity. For prolonged storage, freeze in working aliquots at –20 °C. Avoid repeated freezing and thawing. Do not store in a frost-free freezer.

Product Profile
Anti-Keratinocyte Growth Factor has the ability to neutralize the biological activity of recombinant human FGF-7 (KGF) in monkey epithelial 4MBr-5 cells. Recombinant human FGF-7 is added to various concentrations of the antibody for 1 hour at 22 °C in a 96 well microplate. Following this pre-incubation, 4MBr-5 cells are added to the mixture. The assay mixture in a total volume of 100 µL, containing antibody at concentrations of 0.01 to 100 µg/mL, recombinant human FGF-7 (KGF) at 125 ng/mL, and cells at 1.5×10^5 cells/mL, is incubated at 37 °C for 48 hours in a humidified CO_2 incubator. The mixture is pulsed with ^{3}H-thymidine during the final 24 hours. The cells are detached and harvested onto glass fiber filters, and the ^{3}H-thymidine incorporated into the DNA is measured.

The Neutralization $Dose_{50}$ (ND_{50}) for Anti-FGF-7 is ~ 6-12 µg/mL in the presence of 125 ng/mL of recombinant human FGF-7 using the 4MBr-5 cell line.

The ND_{50} is the concentration of antibody required to yield one-half maximal inhibition of the cytokine activity on a responsive cell line, when the cytokine is present at a concentration just high enough to elicit a maximum response.

The exact concentration of antibody required to neutralize human FGF-7 activity is dependent on the cytokine concentration, cell type, growth conditions, and the type of activity studied.

Immunoblotting: a working concentration of 0.1-0.2 µg/mL is recommended. The detection limit for recombinant human FGF-7 is ~1 ng/lane and 0.5 ng/lane under non-reducing and reducing conditions, respectively.

ELISA: a working concentration of 0.5-1.0 µg/mL is recommended. The detection limit for recombinant human FGF-7 is ~0.08 ng/well.

Note: In order to obtain the best results in various techniques and preparations, we recommend determining optimal working dilutions by titration.

Endotoxin level is < 0.1 EU (endotoxin units) per 1 µg antibody as determined by the LAL (Limulus amebocyte lysate) method.

References

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