SANTA CRUZ BIOTECHNOLOGY, INC.

HB-EGF siRNA (m): sc-39421

BACKGROUND

Heparin-binding epidermal-like growth factor (HB-EGF), a member of the EGF family of mitogens, binds to the EGF receptor (EGFR) and to heparin sulfate proteoglycans on the cell surface. HB-EGF was originally isolated from medium conditioned by the growth of the human histocytic lymphoma cell U-937 on the basis of its heparin-binding ability and its mitogenic activity for Balb/3T3 fibroblasts. The HB-EGF gene encodes a 208 amino acid precursor containing a signal peptide and transmembrane domain. Mature HB-EGF is a soluble protein, 86 amino acids in length, and results from the enzymatic cleavage of the membrane-bound precursor. The membrane-bound form of HB-EGF has been identified as the diphtheria toxin receptor. Preincubation of Vero cells with phorbol 12-myristate 13-acetate (PMA) induces the proteolytic cleavage of HB-EGF outside the membrane anchor.

REFERENCES

1. Higashiyama, S., et al. 1991. A heparin-binding growth factor secreted by macrophage-like cells that is related to EGF. Science 251: 936-939.
2. Mitamura, T., et al. 1995. Diphtheria toxin binds to the epidermal growth factor (EGF)-like domain of human heparin-binding EGF-like growth factor/diphtheria toxin receptor and inhibits specifically its mitogenic activity. J. Biol. Chem. 270: 1015-1019.
3. Lee, Y.J., et al. 1995. Increased expression of heparin-binding epidermal growth factor-like growth factor mRNA in the kidney of streptozotocin-induced diabetic rats. Biochem. Biophys. Res. Commun. 207: 216-222.
4. Modjtahedi, H., et al. 1995. The binding of HB-EGF to tumour cells is blocked by mAbs which act as EGF and TGFα antagonists. Biochem. Biophys. Res. Commun. 207: 389-397.

CHROMOSOMAL LOCATION

Genetic locus: Hbegf (mouse) mapping to 18 B2.

PRODUCT

HB-EGF siRNA (m) is a pool of 3 target-specific 19-25 nt siRNAs designed to knock down gene expression. Each vial contains 3.3 nmol of lyophilized siRNA, sufficient for a 10 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see HB-EGF shRNA Plasmid (m): sc-39421-SH and HB-EGF shRNA (m) Lentiviral Particles: sc-39421-V as alternate gene silencing products.

For independent verification of HB-EGF (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-39421A, sc-39421B and sc-39421C.

STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at -20° C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at -20° C, avoid contact with RNAses and repeated freeze thaw cycles.

Resuspend lyophilized siRNA duplex in 330 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

HB-EGF siRNA (m) is recommended for the inhibition of HB-EGF expression in mouse cells.

SUPPORT REAGENTS

For optimal siRNA transfection efficiency, Santa Cruz Biotechnology's siRNA Transfection Reagent: sc-29528 (0.3 ml), siRNA Transfection Medium: sc-36868 (20 ml) and siRNA Dilution Buffer: sc-29527 (1.5 ml) are recommended. Control siRNAs or Fluorescein Conjugated Control siRNAs are available as 10 µM in 66 µl. Each contain a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA. Fluorescein Conjugated Control siRNAs include: sc-36869, sc-44239, sc-44240 and sc-44241. Control siRNAs include: sc-37007, sc-44230, sc-44231, sc-44232, sc-44233, sc-44234, sc-44235, sc-44236, sc-44237 and sc-44238.

GENE EXPRESSION MONITORING

HB-EGF (H-1): sc-365182 is recommended as a control antibody for monitoring of HB-EGF gene expression knockdown by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000) or immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500).

To ensure optimal results, the following support reagents are recommended: 1) Western Blotting: use m-IgGκ BP-HRP: sc-516102 or m-IgGκ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker™ Molecular Weight Standards: sc-2035, UltraCruz® Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgGκ BP-FITC: sc-516140 or m-IgGκ BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz® Mounting Medium: sc-24941 or UltraCruz® Hard-set Mounting Medium: sc-359850.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor HB-EGF gene expression knockdown using RT-PCR Primer: HB-EGF (m)-PR: sc-39421-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Yan, F., et al. 2013. A *Lactobacillus rhamnosus* GG-derived soluble protein, p40, stimulates ligand release from intestinal epithelial cells to transactivate epidermal growth factor receptor. J. Biol. Chem. 288: 30742-30751.
2. Zhao, G., et al. 2016. Activation of epidermal growth factor receptor in macrophages mediates feedback inhibition of M2 polarization and gastrointestinal tumor cell growth. J. Biol. Chem. 291: 20462-20472.
3. Wen, X., et al. 2022. DR1 activation promotes vascular smooth muscle cell apoptosis via up-regulation of CSE/H_2S pathway in diabetic mice. FASEB J. 36: e22070.

RESEARCH USE

For research use only, not for use in diagnostic procedures.