SANTA CRUZ BIOTECHNOLOGY, INC.

EGFR siRNA (h2): sc-44340

BACKGROUND

The EGF receptor family comprises several related receptor tyrosine kinases that are frequently overexpressed in a variety of carcinomas. Members of this receptor family include EGFR (HER1), Neu (ErbB-2, HER2), ErbB-3 (HER3) and ErbB-4 (HER4), which form either homodimers or heterodimers upon ligand binding. Exons in the EGFR gene product are frequently either deleted or duplicated to produce deletion mutants (DM) or tandem duplication mutants (TDM), respectively, which are detected at various molecular weights. EGFR binds several ligands, including epidermal growth factor (EGF), transforming growth factor α (TGFα), Amphiregulin and heparin binding-EGF (HB-EGF). Ligand binding promotes the internalization of EGFR via Clathrin-coated pits and its subsequent degradation in response to its intrinsic tyrosine kinase. EGFR is involved in organ morphogenesis and maintenance and repair of tissues, but upregulation of EGFR is associated with tumor progression. The oncogenic effects of EGFR include initiation of DNA synthesis, enhanced cell growth, invasion and metastasis. Abrogation of EGFR results in cell cycle arrest, apoptosis or dedifferentiation of cancer cells, suggesting that EGFR may be an effective therapeutic target.

REFERENCES

1. Downward, J., et al. 1984. Autophosphorylation sites on the epidermal growth factor receptor. Nature 311: 483-485.
2. Gullick, W.J., et al. 1985. Antibodies to the autophosphorylation sites of the epidermal growth factor receptor protein-tyrosine kinase as probes of structure and function. EMBO J. 4: 2869-2877.
3. Gullick, W.J., et al. 1986. Expression of epidermal growth factor receptors on human cervical, ovarian, and vulval carcinomas. Cancer Res. 46: 285-292.
4. Berger, M.S., et al. 1987. Epidermal growth factor receptors in lung tumours. J. Pathol. 152: 297-307.
5. Gamou, S., et al. 1988. Biosynthesis of the epidermal growth factor receptor in human squamous cell carcinoma lines: secretion of the truncated receptor is not common to epidermal growth factor receptor-hyperproducing cells. Cell Struct. Funct. 13: 25-38.
6. Fenstermaker, R.A. and Ciesielski, M.J. 2000. Deletion and tandem duplication of exons 2-7 in the epidermal growth factor receptor gene of a human malignant glioma. Oncogene 19: 4542-4548.

CHROMOSOMAL LOCATION

Genetic locus: EGFR (human) mapping to 7p11.2.

PRODUCT

EGFR siRNA (h2) 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 EGFR shRNA Plasmid (h2): sc-44340-SH and EGFR shRNA (h2) Lentiviral Particles: sc-44340-V as alternate gene silencing products.

For independent verification of EGFR (h2) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-44340A, sc-44340B and sc-44340C.

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

EGFR siRNA (h2) is recommended for the inhibition of EGFR expression in human 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

EGFR (A-10): sc-373746 is recommended as a control antibody for monitoring of EGFR 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 EGFR gene expression knockdown using RT-PCR Primer: EGFR (h2)-PR: sc-44340-PR (20 µl, 429 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Ma, S., et al. 2016. Ferroptosis is induced following siramesine and lapatinib treatment of breast cancer cells. Cell Death Dis. 7: e2307.

RESEARCH USE

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