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Diagnostik & molekulare Diagnostik



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ErbB-3 siRNA (h2): sc-44298



The Power to Question

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. Full length ErbB-3 is overexpressed in human mammary tumors. The ErbB-3 gene also produces several alternative variants, including a secreted form which negatively regulates heregulin stimulated ErbB activation. ErbB-3 heterodimerizes with Neu and binds heregulin in order to activate phosphoinositide (PI) 3-kinase. The recruitment and activation of PI 3-kinase occurs via its interaction with phosphorylated YXXM motifs in the carboxy terminus of ErbB-3.

REFERENCES

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- Plowman, G.D., et al. 1990. Molecular cloning and expression of an additional epidermal growth factor receptor-related gene. Proc. Natl. Acad. Sci. USA 87: 4905-4909.
- Kraus, M.H., et al. 1993. Demonstration of ligand-dependent signaling by the ErbB-3 tyrosine kinase and its constitutive activation in human breast tumor cells. Proc. Natl. Acad. Sci. USA 90: 2900-2904.
- Rajkumar, T., et al. 1994. A monoclonal antibody to the human c-ErbB-3 protein stimulates the anchorage-independent growth of breast cancer cell lines. Br. J. Cancer 70: 459-465.
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CHROMOSOMAL LOCATION

Genetic locus: ERBB3 (human) mapping to 12q13.2.

PRODUCT

ErbB-3 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see ErbB-3 shRNA Plasmid (h2): sc-44298-SH and ErbB-3 shRNA (h2) Lentiviral Particles: sc-44298-V as alternate gene silencing products.

For independent verification of ErbB-3 (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-44298A, sc-44298B and sc-44298C.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.

STORAGE AND RESUSPENSION

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

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

APPLICATIONS

ErbB-3 siRNA (h2) is recommended for the inhibition of ErbB-3 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 μ M in 66 μ l. 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-44234, sc-44235, sc-44236, sc-44237 and sc-44238.

GENE EXPRESSION MONITORING

ErbB-3 (G-4): sc-7390 is recommended as a control antibody for monitoring of ErbB-3 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-lgG κ BP-HRP: sc-516102 or m-lgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz MarkerTM Molecular Weight Standards: sc-2035, UltraCruz[®] Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-lgG κ BP-FITC: sc-516140 or m-lgG κ 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 ErbB-3 gene expression knockdown using RT-PCR Primer: ErbB-3 (h2)-PR: sc-44298-PR (20 μ I, 563 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

RESEARCH USE

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

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