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ErbB-3 shRNA (h2) Lentiviral Particles: sc-44298-V

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

1. Kraus, M.H., et al. 1989. Isolation and characterization of ErbB-3, a third member of the ErbB/epidermal growth factor receptor family: evidence for overexpression in a subset of human mammary tumors. *Proc. Natl. Acad. Sci. USA* 86: 9193-9197.
2. 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.
3. 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.
4. 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.
5. Rajkumar, T. and Gullick, W.J. 1994. The type I growth factor receptors in human breast cancer. *Breast Cancer Res. Treat.* 29: 3-9.

CHROMOSOMAL LOCATION

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

PRODUCT

ErbB-3 shRNA (h2) Lentiviral Particles is a pool of concentrated, transduction-ready viral particles containing 3 target-specific constructs that encode 19-25 nt (plus hairpin) shRNA designed to knock down gene expression. Each vial contains 200 μ l frozen stock containing 1.0×10^6 infectious units of virus (IFU) in Dulbecco's Modified Eagle's Medium with 25 mM HEPES pH 7.3. Suitable for 10-20 transductions. Also see ErbB-3 siRNA (h2): sc-44298 and ErbB-3 shRNA Plasmid (h2): sc-44298-SH as alternate gene silencing products.

STORAGE

Store lentiviral particles at -80° C. Stable for at least one year from the date of shipment. Once thawed, particles can be stored at 4° C for up to one week. Avoid repeated freeze thaw cycles.

PROTOCOLS

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

APPLICATIONS

ErbB-3 shRNA (h2) Lentiviral Particles is recommended for the inhibition of ErbB-3 expression in human cells.

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 μ l frozen viral stock containing 1.0×10^6 infectious units of virus (IFU); contains an shRNA construct encoding a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA.

GENE EXPRESSION MONITORING

ErbB-3 (C-17): sc-285 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 (secondary) reagents are recommended: 1) Western Blotting: use goat anti-rabbit IgG-HRP: sc-2004 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible goat anti-rabbit IgG-HRP: sc-2030 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use goat anti-rabbit IgG-FITC: sc-2012 (dilution range: 1:100-1:400) or goat anti-rabbit IgG-TR: sc-2780 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

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 l, 563 bp). Annealing temperature for the primers should be $55-60^{\circ}$ C and the extension temperature should be $68-72^{\circ}$ C.

BIOSAFETY

Lentiviral particles can be employed in standard Biosafety Level 2 tissue culture facilities (and should be treated with the same level of caution as with any other potentially infectious reagent). Lentiviral particles are replication-incompetent and are designed to self-inactivate after transduction and integration of shRNA constructs into genomic DNA of target cells.

RESEARCH USE

The purchase of this product conveys to the buyer the nontransferable right to use the purchased amount of the product and all replicates and derivatives for research purposes conducted by the buyer in his laboratory only (whether the buyer is an academic or for-profit entity). The buyer cannot sell or otherwise transfer (a) this product (b) its components or (c) materials made using this product or its components to a third party, or otherwise use this product or its components or materials made using this product or its components for Commercial Purposes.