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SANTA CRUZ BIOTECHNOLOGY, INC.

Neu siRNA (h2): sc-156048

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. Neu, a glycoprotein, undergoes transactivation upon heterodimerization with other EGF receptor family members. Neu heterodimerization with ErbB-3 recruits heregulin, which induces phosphoinositide (PI) 3-kinase activation. Activation of Neu potentiates tumor cell motility and protease secretion and invasion, and also modulates cell cycle checkpoint function, DNA repair and apoptotic responses. Amplification and/or overexpression of Neu occurs in 20-30% of breast carcinomas. Measurement of increased Neu expression can be a predictor of disease prognosis. Neu may also prove to be a promising target for therapeutic agents.

REFERENCES

1. Rubin, I. and Yarden, Y. 2001. The basic biology of HER2. Ann. Oncol. 12: S3-S8.
2. Eccles, S.A. 2001. The role of c-ErbB-2/HER2/Neu in breast cancer progression and metastasis. J. Mammary Gland Biol. Neoplasia 6: 393-406.
3. Hellyer, N.J., et al. 2001. Heregulin-dependent activation of phosphoinositide 3-kinase and Akt via the ErbB-2/ErbB-3 co-receptor. J. Biol. Chem. 276: 42153-42161.
4. Ukita, Y., et al. 2002. Gene amplification and mRNA and protein overexpression of c-ErbB-2 (HER-2/Neu) in human intrahepatic cholangiocarcinoma as detected by fluorescence *in situ* hybridization, *in situ* hybridization, and immunohistochemistry. J. Hepatol. 36: 780-785.
5. Hayes, D.F. and Thor, A.D. 2002. c-ErbB-2 in breast cancer: development of a clinically useful marker. Semin. Oncol. 29: 231-245.
6. Baxevanis, C.N., et al. 2002. HER-2/Neu-derived peptide epitopes are also recognized by cytotoxic $CD3^+CD56^+$ (natural killer T) lymphocytes. Int. J. Cancer 98: 864-872.
7. Cho, H.S., et al. 2003. Structure of the extracellular region of HER-2 alone and in complex with the Herceptin Fab. Nature 421: 756-760.

CHROMOSOMAL LOCATION

Genetic locus: ERBB2 (human) mapping to 17q12.

PRODUCT

Neu 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 Neu shRNA Plasmid (h2): sc-156048-SH and Neu shRNA (h2) Lentiviral Particles: sc-156048-V as alternate gene silencing products.

For independent verification of Neu (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-156048A, sc-156048B and sc-156048C.

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

Neu siRNA (h2) is recommended for the inhibition of Neu 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

Neu (3B5): sc-33684 is recommended as a control antibody for monitoring of Neu 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 Neu gene expression knockdown using RT-PCR Primer: Neu (h2)-PR: sc-156048-PR (20 µl, 548 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.

PROTOCOLS

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