

E-cadherin siRNA (h2): sc-44222

BACKGROUND

Cadherins comprise a family of Ca^{2+} -dependent adhesion molecules that function to mediate cell-cell binding critical to the maintenance of tissue structure and morphogenesis. Members of this family of adhesion proteins include rat cadherin K (and its human homolog, cadherin-6), R-cadherin, B-cadherin, E/ P-cadherin and cadherin-5. The classical cadherins, E-, N- and P-cadherin, consist of large extracellular domains characterized by a series of five homologous NH_2 terminal repeats. The most distal of these cadherins is thought to be responsible for binding specificity, transmembrane domains and carboxy-terminal intracellular domains. The relatively short intracellular domains interact with a variety of cytoplasmic proteins, such as β -catenin, to regulate cadherin function.

REFERENCES

1. Hirsch, H.A., et al. 1978. Surgical therapy of breast cancer. *Gynakol. Rundsch.* 18: 132-141.
2. Takeichi, M. 1988. The cadherins: cell-cell adhesion molecules controlling animal morphogenesis. *Development* 102: 639-655.
3. Hatta, M., et al. 1991. Genomic organization and chromosomal mapping of the mouse P-cadherin gene. *Nucleic Acids Res.* 19: 4437-4441.
4. Umbas, R., et al. 1992. Expression of the cellular adhesion molecule E-cadherin is reduced or absent in high-grade prostate cancer. *Cancer Res.* 52: 5104-5109.
5. Koch, P.J., et al. 1994. Desmosomal cadherins: another growing multigene family of adhesion molecules. *Curr. Opin. Cell Biol.* 6: 682-687.

CHROMOSOMAL LOCATION

Genetic locus: CDH1 (human) mapping to 16q22.1.

PRODUCT

E-cadherin 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 E-cadherin shRNA Plasmid (h2): sc-44222-SH and E-cadherin shRNA (h2) Lentiviral Particles: sc-44222-V as alternate gene silencing products.

For independent verification of E-cadherin (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-44222A, sc-44222B and sc-44222C.

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

E-cadherin siRNA (h2) is recommended for the inhibition of E-cadherin 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

E-cadherin (G-10): sc-8426 is recommended as a control antibody for monitoring of E-cadherin 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-IgG κ BP-HRP: sc-516102 or m-IgG κ 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-IgG κ BP-FITC: sc-516140 or m-IgG 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 E-cadherin gene expression knockdown using RT-PCR Primer: E-cadherin (h2)-PR: sc-44222-PR (20 μl , 429 bp). Annealing temperature for the primers should be $55-60^\circ\text{C}$ and the extension temperature should be $68-72^\circ\text{C}$.

SELECT PRODUCT CITATIONS

1. Saydam, O., et al. 2009. Downregulated microRNA-200a in meningiomas promotes tumor growth by reducing E-cadherin and activating the Wnt/ β -catenin signaling pathway. *Mol. Cell. Biol.* 29: 5923-5940.

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.