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Mesothelin siRNA (h): sc-45386

BACKGROUND

Mesothelin is a glycosylphosphatidylinositol-linked cell-surface molecule expressed in the mesothelial lining of the body cavities and in many tumor cells. Mesothelin is a tumor antigen on the surface of human ovarian cancers and mesotheliomas. Mesothelin immunoreactivity is high in cancers of the ovary (serous papillary, endometrioid and undifferentiated) and pancreas, with less frequent staining seen in adenocarcinomas of the endometrium, lung and stomach/esophagus. In adult mouse tissues the Mesothelin transcript is present in lung, heart, spleen, liver, kidney and testis.

REFERENCES

1. Chang, K., et al. 1996. Molecular cloning of Mesothelin, a differentiation antigen present on mesothelium, mesotheliomas and ovarian cancers. *Proc. Natl. Acad. Sci. USA* 93: 136-140.
2. Chowdhury, P.S., et al. 1997. Isolation of anti-Mesothelin antibodies from a phage display library. *Mol. Immunol.* 34: 9-20.
3. Hassan, R., et al. 1999. 111Indium-labeled monoclonal antibody K1: bio-distribution study in nude mice bearing a human carcinoma xenograft expressing Mesothelin. *Int. J. Cancer* 80: 559-563.
4. Bera, T.K., et al. 2000. Mesothelin is not required for normal mouse development or reproduction. *Mol. Cell. Biol.* 20: 2902-2906.
5. Frierson, H.F., Jr., et al. 2003. Large-scale molecular and tissue microarray analysis of Mesothelin expression in common human carcinomas. *Hum. Pathol.* 34: 605-609.
6. Rump, A., et al. 2004. Binding of ovarian cancer antigen CA125/MUC16 to Mesothelin mediates cell adhesion. *J. Biol. Chem.* 279: 9190-9198.

CHROMOSOMAL LOCATION

Genetic locus: MSLN (human) mapping to 16p13.3.

PRODUCT

Mesothelin siRNA (h) 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 Mesothelin siRNA (h): sc-45386-SH and Mesothelin siRNA (h) Lentiviral Particles: sc-45386-V as alternate gene silencing products.

For independent verification of Mesothelin siRNA (h) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-45386A, sc-45386B and sc-45386C.

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.

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 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

Mesothelin siRNA (h) is recommended for the inhibition of Mesothelin 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-44233, sc-44234, sc-44235, sc-44236, sc-44237 and sc-44238.

GENE EXPRESSION MONITORING

Mesothelin (K1): sc-33672 is recommended as a control antibody for monitoring of Mesothelin 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 Mesothelin gene expression knockdown using RT-PCR Primer: Mesothelin siRNA (h)-PR: sc-45386-PR (20 μ l, 423 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.