SANTA CRUZ BIOTECHNOLOGY, INC.

nm23-H2 siRNA (h): sc-40774

BACKGROUND

The nm23 gene, a potential suppressor of metastasis, was originally identified by differential hybridization between two murine melanoma sublines, one with a high and the second with a low metastatic capacity. Highly metastatic sub-lines exhibit much lower levels of nm23 than less metastatic cells. Based on sequence analysis, nm23 appears highly related to nucleotide diphosphate kinases (NDP). In humans, NDP kinases A and B are identical to two isotypes of human nm23 homologs, namely nm23-H1 and H2, respectively. nm23-H2 is identical in sequence to PuF, a transcription factor that binds to nuclease-hypersensitive elements at positions 142 to 115 of the human c-Myc promotor.

REFERENCES

1. Steeg, P.S., et al. 1988. Evidence for a novel gene associated with low tumor metastatic potential. J. Natl. Cancer Inst. 80: 200-209.
2. Lacombe, M., et al. 1990. Functional cloning of a nucleoside diphosphate kinase from *Dictyostelium discoideum*. J. Biol. Chem. 265: 10012-10018.
3. Kimura, N., et al. 1990. Isolation and characterization of a cDNA clone encoding rat nucleoside diphosphate kinase. J. Biol. Chem. 265: 15744-15749.
4. Stahl, J.A., et al. 1991. Identification of a second human nm23 gene, nm23-H2. Cancer Res. 51: 445-449.
5. Urano, T., et al. 1992. Molecular cloning and functional expression of the second mouse nm23/NDP kinase gene, nm23-M2. FEBS Lett. 309: 358-362.
6. Urano, T., et al. 1993. Expression of nm23/NDP kinase proteins on the cell surface. Oncogene 8: 1371-1376.

CHROMOSOMAL LOCATION

Genetic locus: NME1-NME2 (human) mapping to 17q21.33.

PRODUCT

nm23-H2 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see nm23-H2 shRNA Plasmid (h): sc-40774-SH and nm23-H2 shRNA (h) Lentiviral Particles: sc-40774-V as alternate gene silencing products.

For independent verification of nm23-H2 (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-40774A, sc-40774B and sc-40774C.

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

nm23-H2 siRNA (h) is recommended for the inhibition of nm23-H2 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

nm23-H2 (X-42): sc-100400 is recommended as a control antibody for monitoring of nm23-H2 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 nm23-H2 gene expression knockdown using RT-PCR Primer: nm23-H2 (h)-PR: sc-40774-PR (20 µl, 422 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Thakur, R.K., et al. 2009. Metastases suppressor nm23-H2 interaction with G-quadruplex DNA within c-Myc promoter nuclease hypersensitive element induces c-Myc expression. Nucleic Acids Res. 37: 172-183.
2. Shan, C., et al. 2015. Chemical intervention of the nm23-H2 transcriptional programme on c-Myc via a novel small molecule. Nucleic Acids Res. 43: 6677-6691.
3. Sengupta, P. and Chatterjee, S. 2020. Inosine 5'-diphosphate, a molecular decoy rescues nucleoside diphosphate kinase from c-MYC G-quadruplex unfolding. Biochim. Biophys. Acta Gen. Subj. 1864: 129649.

RESEARCH USE

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