SANTA CRUZ BIOTECHNOLOGY, INC.

Hint1 siRNA (m): sc-145966

BACKGROUND

Hint1 (histidine triad nucleotide binding protein 1), also known as PRKCNH1 or PKCI1, is a 126 amino acid protein that localizes to both the cytoplasm and the nucleus and contains one HIT (histidine triad) domain. Widely expressed and existing as a homodimer, Hint1 functions to hydrolyze adenosine 5'-monophosphoramidate substrates, such as AMP-N-alanine methyl ester, AMP-morpholidate and AMP-α-acetyl lysine methyl ester, and may also inhibit PKC function and influence apoptosis. Knockdown of Hint1 mRNA results in downregulation of p53 and Bax, suggesting that Hint1 is a haploinsufficient tumor suppressor. Also, it is suspected that promoter hypermethylation of the gene encoding Hint1 may play a role in hepatocarcinogenesis.

REFERENCES

1. Pearson, J.D., et al. 1990. Amino acid sequence and characterization of a protein inhibitor of protein kinase C. J. Biol. Chem. 265: 4583-4591.
2. Robinson, K. and Aitken, A. 1994. Identification of a new protein family which includes bovine protein kinase C inhibitor-1. Biochem. J. 304: 662-664.
3. Brzoska, P.M., et al. 1996. Cloning, mapping, and *in vivo* localization of a human member of the PKCI-1 protein family (PRKCNH1). Genomics 36: 151-156.
4. Lima, C.D., et al. 1996. Three-dimensional structure of human protein kinase C interacting protein 1, a member of the HIT family of proteins. Proc. Natl. Acad. Sci. USA 93: 5357-5362.
5. Brenner, C. 2002. Hint, Fhit, and GalT: function, structure, evolution, and mechanism of three branches of the histidine triad superfamily of nucleotide hydrolases and transferases. Biochemistry 41: 9003-9014.
6. Online Mendelian Inheritance in Man, OMIM™. 2006. Johns Hopkins University, Baltimore, MD. MIM Number: 601314. World Wide Web URL: http://www.ncbi.nlm.nih.gov/omim/
7. Zhang, Y.J., et al. 2008. Silencing of Hint1, a novel tumor suppressor gene, by promoter hypermethylation in hepatocellular carcinoma. Cancer Lett. 275: 277-284.

CHROMOSOMAL LOCATION

Genetic locus: Hint1 (mouse) mapping to 11 B1.3.

PRODUCT

Hint1 siRNA (m) is a pool of 2 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 Hint1 shRNA Plasmid (m): sc-145966-SH and Hint1 shRNA (m) Lentiviral Particles: sc-145966-V as alternate gene silencing products.

For independent verification of Hint1 (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-145966A and sc-145966B.

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

Hint1 siRNA (m) is recommended for the inhibition of Hint1 expression in mouse 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

Hint1 (B-5): sc-271790 is recommended as a control antibody for monitoring of Hint1 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 Hint1 gene expression knockdown using RT-PCR Primer: Hint1 (m)-PR: sc-145966-PR (20 µl). 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.