

NTH1 siRNA (m): sc-38134

BACKGROUND

The human endonuclease III (hNTH1), a homolog of the *E. coli* enzyme (Nth), is a DNA glycosylase with abasic (apurinic/aprimidinic (AP)) lyase activity that specifically cleaves oxidatively damaged pyrimidines in DNA. The enzyme carries out β -elimination and forms a Schiff base between the active site at Lysine-212 and the deoxyribose generated after base removal. Full-length human NTH1 sorts exclusively into the nuclei, whereas most mouse NTH1 protein sorts into the mitochondria, with a relatively small amount localized in the nuclei. This difference is due to the presence of a nuclear localization sequence in the human NTH1 that is absent in the mouse form of the protein. The mammalian NTH1 gene lies immediately adjacent to one of the tuberous sclerosis disease-determining genes, TSC2, in a head-to-head orientation. The two genes share a promoter with bidirectional activity essential for the transcription of both genes. DNA glycosylases such as NTH1 play an important role in the excision of damaged bases in the genome.

REFERENCES

- Ikeda, S., et al. 1998. Purification and characterization of human NTH1, a homolog of *Escherichia coli* endonuclease III. Direct identification of Lys-212 as the active nucleophilic residue. *J. Biol. Chem.* 273: 21585-21593.
- Sarker, A.H., et al. 1998. Cloning and characterization of a mouse homologue (mNth1) of *Escherichia coli* endonuclease III. *J. Mol. Biol.* 282: 761-764.
- Ikeda, S., et al. 2000. Identification of functional elements in the bidirectional promoter of the mouse Nth1 and Tsc2 genes. *Biochem. Biophys. Res. Commun.* 273: 1063-1068.
- Ikeda, S., et al. 2002. Differential intracellular localization of the human and mouse endonuclease III homologs and analysis of the sorting signals. *DNA Repair* 1: 847-854.
- Hazra, T.K., et al. 2004. The discovery of a new family of mammalian enzymes for repair of oxidatively damaged DNA, and its physiological implications. *Carcinogenesis* 24: 155-157.

CHROMOSOMAL LOCATION

Genetic locus: Nth1 (mouse) mapping to 17 A3.3.

PRODUCT

NTH1 siRNA (m) 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 NTH1 shRNA Plasmid (m): sc-38134-SH and NTH1 shRNA (m) Lentiviral Particles: sc-38134-V as alternate gene silencing products.

For independent verification of NTH1 (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-38134A, sc-38134B and sc-38134C.

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

NTH1 siRNA (m) is recommended for the inhibition of NTH1 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 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

NTH1 (2660C1a): sc-130644 is recommended as a control antibody for monitoring of NTH1 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).

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor NTH1 gene expression knockdown using RT-PCR Primer: NTH1 (m)-PR: sc-38134-PR (20 μ l). 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.