SANTA CRUZ BIOTECHNOLOGY, INC.

NSDHL siRNA (m): sc-150073

BACKGROUND

NSDHL (NAD(P) dependent steroid dehydrogenase-like), also known as H105E3, XAP104 or SDR31E1, is a 373 amino acid single-pass membrane protein expressed in brain, heart, liver, lung, kidney, skin and placenta. Belonging to the 3-β-HSD family, NSDHL localizes to the endoplasmic reticulum and is involved in cholesterol biosynthesis. NSDHL is thought to be involved in the demethylation of sterol precursors in one of the later steps of cholesterol biosynthesis. Defects in the gene encoding NSDHL causes congenital hemidysplasia with ichthyosiform erythroderma and limb defects (CHILD), which is an X-linked dominant disorder of lipid metabolism with defective cholesterol biosynthesis that usually results in male lethality. CHILD is characterized by congenital hemidysplasia with ichthyosiform erythroderma and ipsilateral hypoplasia of limbs and other parts of the skeleton.

REFERENCES

1. Hummel, M., et al. 2003. Left-sided CHILD syndrome caused by a nonsense mutation in the NSDHL gene. Am. J. Med. Genet. A 122A: 246-251.
2. Caldas, H. and Herman, G.E. 2003. NSDHL, an enzyme involved in cholesterol biosynthesis, traffics through the Golgi and accumulates on ER membranes and on the surface of lipid droplets. Hum. Mol. Genet. 12: 2981-2991.
3. Ohashi, M., et al. 2003. Localization of mammalian NAD(P)H steroid dehydrogenase-like protein on lipid droplets. J. Biol. Chem. 278: 36819-36829.
4. Murata, K., et al. 2003. A unique point mutation in the NSDHL gene in a Japanese patient with CHILD syndrome. J. Dermatol. Sci. 33: 67-69.
5. Mehra, S., et al. 2005. A novel somatic mutation of the 3β-hydroxysteroid dehydrogenase gene in sporadic cutaneous verruciform xanthoma. Arch. Dermatol. 141: 1263-1267.
6. Bittar, M., et al. 2006. CHILD syndrome in 3 generations: the importance of mild or minimal skin lesions. Arch. Dermatol. 142: 348-351.

CHROMOSOMAL LOCATION

Genetic locus: Nsdhl (mouse) mapping to X A7.3.

PRODUCT

NSDHL 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see NSDHL shRNA Plasmid (m): sc-150073-SH and NSDHL shRNA (m) Lentiviral Particles: sc-150073-V as alternate gene silencing products.

For independent verification of NSDHL (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-150073A, sc-150073B and sc-150073C.

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 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

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

NSDHL (D-11): sc-390871 is recommended as a control antibody for monitoring of NSDHL 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 NSDHL gene expression knockdown using RT-PCR Primer: NSDHL (m)-PR: sc-150073-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.