

DTWD1 siRNA (h): sc-89974

BACKGROUND

DTWD1 (DTW domain-containing protein 1), also known as MDS009, is a 304 amino acid protein that contains one DXTW motif and belongs to the DTW family. Existing as three alternatively spliced isoforms, DTWD1 is encoded by a gene that maps to human chromosome 15q21.2. Chromosome 15 is made up of approximately 106 million base pairs and is about 3% of the human genome. Angelman and Prader-Willi syndromes are associated with loss of function or deletion of genes in the 15q11-q13 region. In the case of Angelman syndrome, this loss is due to inactivity of the maternal 15q11-q13 encoded UBE3A gene in the brain by either chromosomal deletion or mutation. In cases of Prader-Willi syndrome, there is a partial or complete deletion of this region from the paternal copy of chromosome 15. Tay-Sachs disease is a lethal disorder associated with mutations of the HEXA gene, which is encoded by chromosome 15. Marfan syndrome is associated with chromosome 15 through the FBN1 gene.

REFERENCES

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2. Zody, M.C., et al. 2006. Analysis of the DNA sequence and duplication history of human chromosome 15. *Nature* 440: 671-675.
3. Diene, G., et al. 2007. The Prader-Willi syndrome. *Ann. Endocrinol.* 68: 129-137.
4. Lalande, M. and Calciano, M.A. 2007. Molecular epigenetics of Angelman syndrome. *Cell. Mol. Life Sci.* 64: 947-960.
5. Maegawa, G.H., et al. 2007. Pyrimethamine as a potential pharmacological chaperone for late-onset forms of GM2 gangliosidosis. *J. Biol. Chem.* 282: 9150-9161.
6. Makoff, A.J. and Flomen, R.H. 2007. Detailed analysis of 15q11-q14 sequence corrects errors and gaps in the public access sequence to fully reveal large segmental duplications at breakpoints for Prader-Willi, Angelman, and inv dup(15) syndromes. *Genome Biol.* 8: R114.

CHROMOSOMAL LOCATION

Genetic locus: DTWD1 (human) mapping to 15q21.2.

PRODUCT

DTWD1 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 DTWD1 shRNA Plasmid (h): sc-89974-SH and DTWD1 shRNA (h) Lentiviral Particles: sc-89974-V as alternate gene silencing products.

For independent verification of DTWD1 (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-89974A, sc-89974B and sc-89974C.

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

DTWD1 siRNA (h) is recommended for the inhibition of DTWD1 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

DTWD1 (E-5): sc-398408 is recommended as a control antibody for monitoring of DTWD1 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 DTWD1 gene expression knockdown using RT-PCR Primer: DTWD1 (h)-PR: sc-89974-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.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.