SANTA CRUZ BIOTECHNOLOGY, INC.

Cdt2 siRNA (h): sc-72852



BACKGROUND

WD-repeats are motifs that are found in a variety of proteins and are characterized by a conserved core of 40-60 amino acids that commonly form a tertiary propeller structure. While proteins that contain WD-repeats participate in a wide range of cellular functions, they are generally involved in regulatory mechanisms concerning chromatin assembly, cell cycle control, signal transduction, RNA processing, apoptosis and vesicular trafficking. Cdt2, also known as DTL (denticleless homolog), CDW1, DCAF2, L2DTL or RAMP, is a 730 amino acid protein that localizes to both the cytoplasm and the nuclear membrane and contains seven WD repeats. Expressed in testis, placenta, bone marrow, thymus and skeletal muscle, Cdt2 is required for DNA damage-induced Cdt1 proteolysis and is also thought to play an essential role in DNA replication and cell proliferation. Upon DNA damage, Cdt2 is subject to phosphorylation, probably by ATM or ATR. Two isoforms of Cdt2 exist due to alternative splicing events.

REFERENCES

- Cheung, W.M., et al. 2001. Cloning and expression of a novel nuclear matrix-associated protein that is regulated during the retinoic acidinduced neuronal differentiation. J. Biol. Chem. 276: 17083-17091.
- 2. Banks, D., et al. 2006. L2DTL/CDT2 and PCNA interact with p53 and regulate p53 polyubiquitination and protein stability through MDM2 and CUL4A/DDB1 complexes. Cell Cycle 5: 1719-1729.
- 3. Pan, H.W., et al. 2006. Role of L2DTL, cell cycle-regulated nuclear and centrosome protein, in aggressive hepatocellular carcinoma. Cell Cycle 5: 2676-2687.
- 4. Sansam, C.L., et al. 2006. DTL/CDT2 is essential for both CDT1 regulation and the early G_2/M checkpoint. Genes Dev. 20: 3117-3129.
- 5. Jin, J., et al. 2006. A family of diverse Cul4-Ddb1-interacting proteins includes Cdt2, which is required for S phase destruction of the replication factor Cdt1. Mol. Cell 23: 709-721.
- Higa, L.A., et al. 2006. CUL4-DDB1 ubiquitin ligase interacts with multiple WD40-repeat proteins and regulates histone methylation. Nat. Cell Biol. 8: 1277-1283.

CHROMOSOMAL LOCATION

Genetic locus: DTL (human) mapping to 1q32.3.

PRODUCT

Cdt2 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 Cdt2 shRNA Plasmid (h): sc-72852-SH and Cdt2 shRNA (h) Lentiviral Particles: sc-72852-V as alternate gene silencing products.

For independent verification of Cdt2 (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-72852A, sc-72852B and sc-72852C.

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

Cdt2 siRNA (h) is recommended for the inhibition of Cdt2 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

Cdt2 (B-8): sc-166735 is recommended as a control antibody for monitoring of Cdt2 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 Cdt2 gene expression knockdown using RT-PCR Primer: Cdt2 (h)-PR: sc-72852-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.