SANTA CRUZ BIOTECHNOLOGY, INC.

DDX36 siRNA (h): sc-78053

BACKGROUND

DEAD box proteins, characterized by the conserved motif Asp-Glu-Ala-Asp, are putative RNA helicases implicated in several cellular processes involving modifications of RNA secondary structure. Specifically, DEAD box proteins are involved in translation initiation, nuclear and mitochondrial splicing, and ribosome and spliceosome assembly. Based on their distribution patterns, members of this family may be involved in embryogenesis, spermatogenesis, and cellular growth and division. DDX30, DDX35 and DDX36 each contain one helicase ATP-binding domain and one helicase C-terminal domain. DDX36 (DEAH box protein 36), also known as MLE-like protein 1 and RNA helicase associated with AU-rich element ARE, is a 1,008 amino acid protein that is expressed in testis and may function in sex development and spermatogenesis. DDX36 plays a role in degradation and deadenylation of mRNAs that contain the consensus ARE sequence element in their 3'-UTR. There are three isoforms of DDX36 that exist as a result of alternative splicing events.

REFERENCES

1. Fu, J.J., Li, L.Y. and Lu, G.X. 2002. Molecular cloning and characterization of human DDX36 and mouse Ddx36 genes, new members of the DEAD/H box superfamily. Sheng Wu Hua Xue Yu Sheng Wu Wu Li Xue Bao 34: 655-661.
2. Fu, J.J., Li, L.Y., Liu, S.F., Xing, X.W., Liu, G. and Lu, G.X. 2003. Expression research for human DDX36 and mouse Ddx36 gene in the adult testis. Yi Chuan Xue Bao 30: 201-208.
3. Abdelhaleem, M. 2005. RNA helicases: regulators of differentiation. Clin. Biochem. 38: 499-503.
4. Cordin, O., Banroques, J., Tanner, N.K. and Linder, P. 2006. The DEAD-box protein family of RNA helicases. Gene 367: 17-37.
5. Wang, Y. and Bogenhagen, D.F. 2006. Human mitochondrial DNA nucleoids are linked to protein folding machinery and metabolic enzymes at the mitochondrial inner membrane. J. Biol. Chem. 281: 25791-25802.

CHROMOSOMAL LOCATION

Genetic locus: DHX36 (human) mapping to 3q25.2.

PRODUCT

DDX36 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see DDX36 shRNA Plasmid (h): sc-78053-SH and DDX36 shRNA (h) Lentiviral Particles: sc-78053-V as alternate gene silencing products.

For independent verification of DDX36 (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-78053A, sc-78053B and sc-78053C.

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

DDX36 siRNA (h) is recommended for the inhibition of DDX36 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 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

DDX36 (B-6): sc-377485 is recommended as a control antibody for monitoring of DDX36 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 DDX36 gene expression knockdown using RT-PCR Primer: DDX36 (h)-PR: sc-78053-PR (20 µl, 594 bp). 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.