

DDX33 siRNA (h): sc-93804

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 and ribosome/spliceosome assembly. Based on their distribution patterns, some members of this family may be involved in embryogenesis, spermatogenesis and cellular growth and division. DDX33 (DEAD (Asp-Glu-Ala-His) box polypeptide 33), also known as DHX33, is a 707 amino acid nucleolar protein belonging to the DEAD box helicase family. Containing a helicase ATP-binding domain and a helicase C-terminal domain, DDX33 is encoded by a gene located on human chromosome 17. Chromosome 17 comprises over 2.5% of the human genome and encodes over 1,200 genes. Two isoforms of DDX33 exist due to alternative splicing events.

REFERENCES

- Schmid, S.R. and Linder, P. 1992. D-E-A-D protein family of putative RNA helicases. *Mol. Microbiol.* 6: 283-291.
- Will, C.L., Urlaub, H., Achsel, T., Gentzel, M., Wilm, M. and Lührmann, R. 2002. Characterization of novel SF3b and 17S U2 snRNP proteins, including a human Prp5p homologue and an SF3b DEAD-box protein. *EMBO J.* 21: 4978-4988.
- Abdelhaleem, M., Maltais, L. and Wain, H. 2003. The human DDX and DHX gene families of putative RNA helicases. *Genomics* 81: 618-622.
- Cordin, O., Tanner, N.K., Doère, M., Linder, P. and Banroques, J. 2004. The newly discovered Q motif of DEAD-box RNA helicases regulates RNA-binding and helicase activity. *EMBO J.* 23: 2478-2487.
- Xu, Y.Z., Newnham, C.M., Kameoka, S., Huang, T., Konarska, M.M. and Query, C.C. 2004. Prp5 bridges U1 and U2 snRNPs and enables stable U2 snRNP association with intron RNA. *EMBO J.* 23: 376-385.

CHROMOSOMAL LOCATION

Genetic locus: DHX33 (human) mapping to 17p13.2.

PRODUCT

DDX33 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 DDX33 shRNA Plasmid (h): sc-93804-SH and DDX33 shRNA (h) Lentiviral Particles: sc-93804-V as alternate gene silencing products.

For independent verification of DDX33 (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-93804A, sc-93804B and sc-93804C.

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

DDX33 siRNA (h) is recommended for the inhibition of DDX33 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

DDX33 (B-4): sc-390573 is recommended as a control antibody for monitoring of DDX33 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 DDX33 gene expression knockdown using RT-PCR Primer: DDX33 (h)-PR: sc-93804-PR (20 μ l, 530 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.