SANTA CRUZ BIOTECHNOLOGY, INC.

DDX25 siRNA (m): sc-72230

BACKGROUND

DDX25, also designated GRTH for gonadotropin-regulated testicular RNA helicase, is a testis-specific member of the DEAD-box protein family found in Leydig and germ cells. DDX25 has both ATPase and RNA helicase activity, regulating such translational-associated events as mRNA nuclear export during spermatid development. DDX25 can be phosphorylated on threonine residues, with the phosphorylated form found only in the cytoplasm and not in the nucleus. Essential for spermatogenesis, DDX25 participates in germ cell development and is up-regulated by gonadotropin at the transcriptional level. Genetic variations in DDX25 may contribute to male infertility due to spermatogenic impairment.

REFERENCES

1. Sheng, Y., et al. 2003. Cell-specific and hormone-regulated expression of gonadotropin-regulated testicular RNA helicase gene (GRTH/Ddx25) resulting from alternative utilization of translation initiation codons in the rat testis. J. Biol. Chem. 278: 27796-27803.
2. Tsai-Morris, C.H., et al. 2004. Gonadotropin-regulated testicular RNA helicase (GRTH/Ddx25) is essential for spermatid development and completion of spermatogenesis. Proc. Natl. Acad. Sci. USA 101: 6373-6378.
3. Tsai-Morris, C.H., et al. 2004. Genomic organization and transcriptional analysis of gonadotropin-regulated testicular RNA helicase—GRTH/DDX25 gene. Gene 331: 83-94.
4. Mee, L., et al. 2005. Hydrolethalus syndrome is caused by a missense mutation in a novel gene HYLS1. Hum. Mol. Genet. 14: 1475-1488.
5. Abdelhaleem, M. 2005. RNA helicases: regulators of differentiation. Clin. Biochem. 38: 499-503.
6. A, Z., et al. 2006. Single nucleotide polymorphisms of the gonadotrophin-regulated testicular helicase (GRTH) gene may be associated with the human spermatogenesis impairment. Hum. Reprod. 21: 755-759.

CHROMOSOMAL LOCATION

Genetic locus: Ddx25 (mouse) mapping to 9 A4.

PRODUCT

DDX25 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 DDX25 shRNA Plasmid (m): sc-72230-SH and DDX25 shRNA (m) Lentiviral Particles: sc-72230-V as alternate gene silencing products.

For independent verification of DDX25 (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-72230A, sc-72230B and sc-72230C.

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

DDX25 siRNA (m) is recommended for the inhibition of DDX25 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

DDX25 (F-10): sc-271730 is recommended as a control antibody for monitoring of DDX25 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 DDX25 gene expression knockdown using RT-PCR Primer: DDX25 (m)-PR: sc-72230-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.