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SANTA CRUZ BIOTECHNOLOGY, INC.

Odf2 siRNA (h): sc-43410

BACKGROUND

The major cytoskeletal structures in the mammalian sperm tail are the outer dense fibers (ODF) and the fibrous sheath (FS). The ODFs are located on the outside of the axoneme, and they help maintain the passive elastic structures and elastic recoil of the sperm tail. Human ODFs consist of approximately 10 major and at least 15 minor proteins. The major proteins of the ODF include Odf1, Odf2, and Odf3, which compose a family of proteins that are preferentially expressed during mammalian spermiogenesis. The human Odf1 gene maps to chromosome 8q22. The human Odf2 gene maps to chromosome 9q34.11. Both Odf1 and Odf2 are exclusively expressed in testis. Odf2 interacts with Odf1 during assembly of the outer dense fibers by means of leucine zippers in both proteins. Odf1 can also self interact. The Odf proteins may be involved in male infertility as a result of flagellar dysfunction.

REFERENCES

1. Gastmann, O., et al. 1993. Sequence, expression, and chromosomal assignment of a human sperm outer dense fiber gene. Mol. Reprod. Dev. 36: 407-418.
2. Shao, X., et al. 1996. Self-interaction of the major 27 kilodalton outer dense fiber protein is in part mediated by a leucine zipper domain in the rat. Biol. Reprod. 55: 1343-1350.
3. Shao, X., et al. 1998. Human outer dense fiber gene, ODF2, localizes to chromosome 9q34. Cytogenet. Cell Genet. 83: 221-223.
4. Schalles, U., et al. 1998. Developmental expression of the 84 kDa ODF sperm protein: localization to both the cortex and medulla of outer dense fibers and to the connecting piece. Dev. Biol. 199: 250-260.
5. Petersen, C., et al. 1999. Outer dense fibre proteins from human sperm tail: molecular cloning and expression analyses of two cDNA transcripts encoding proteins of approximately 70 kDa. Mol. Hum. Reprod. 5: 627-635.
6. Shao, X., et al. 2001. Testicular protein Spag5 has similarity to mitotic spindle protein Deepest and binds outer dense fiber protein Odf1. Mol. Reprod. Dev. 59: 410-416.
7. Kierszenbaum, A.L. 2002. Keratins: unraveling the coordinated construction of scaffolds in spermatogenic cells. Mol. Reprod. Dev. 61: 1-2.

CHROMOSOMAL LOCATION

Genetic locus: ODF2 (human) mapping to 9q34.11.

PRODUCT

Odf2 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 Odf2 shRNA Plasmid (h): sc-43410-SH and Odf2 shRNA (h) Lentiviral Particles: sc-43410-V as alternate gene silencing products.

For independent verification of Odf2 (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-43410A, sc-43410B and sc-43410C.

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

Odf2 siRNA (h) is recommended for the inhibition of Odf2 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

Odf2 (G-8): sc-365874 is recommended as a control antibody for monitoring of Odf2 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 Odf2 gene expression knockdown using RT-PCR Primer: Odf2 (h)-PR: sc-43410-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.

PROTOCOLS

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