

CatSper δ siRNA (h): sc-97656

BACKGROUND

CatSper (cation channel, sperm associated proteins) are ion transport proteins located on the surface of sperm cells in the principal piece of the sperm tail. Vital to sperm motility, fertilization and cAMP-mediated calcium influx in sperm, CatSper contain a single, six-transmembrane-spanning segment and exhibit the voltage-dependent Ca²⁺ channel four-repeat structure. CatSper proteins are believed to assemble into a heterotetrameric complex, forming an alkalinization-activated Ca²⁺-selective channel. Mutations in any of the genes encoding CatSper family proteins can result in male infertility. CatSper δ (cation channel sperm-associated protein subunit δ) is a 798 amino acid single-pass type I membrane protein that is located in the principal piece of the sperm tail. CatSper δ exists as two alternatively spliced isoforms and is encoded by a gene that maps to human chromosome 19.

REFERENCES

- Jin, J., Jin, N., Zheng, H., Ro, S., Tafolla, D., Sanders, K.M. and Yan, W. 2007. Catsper3 and Catsper4 are essential for sperm hyperactivated motility and male fertility in the mouse. *Biol. Reprod.* 77: 37-44.
- Xia, J., Reigada, D., Mitchell, C.H. and Ren, D. 2007. CATSPER channel-mediated Ca²⁺ entry into mouse sperm triggers a tail-to-head propagation. *Biol. Reprod.* 77: 551-559.
- Li, H.G., Ding, X.F., Liao, A.H., Kong, X.B. and Xiong, C.L. 2007. Expression of CatSper family transcripts in the mouse testis during post-natal development and human ejaculated spermatozoa: relationship to sperm motility. *Mol. Hum. Reprod.* 13: 299-306.
- Qi, H., Moran, M.M., Navarro, B., Chong, J.A., Krapivinsky, G., Krapivinsky, L., Kirichok, Y., Ramsey, I.S., Quill, T.A. and Clapham, D.E. 2007. All four CatSper ion channel proteins are required for male fertility and sperm cell hyperactivated motility. *Proc. Natl. Acad. Sci. USA* 104: 1219-1223.
- Suarez, S.S., Marquez, B., Harris, T.P. and Schimenti, J.C. 2007. Different regulatory systems operate in the midpiece and principal piece of the mammalian sperm flagellum. *Soc. Reprod. Fertil. Suppl.* 65: 331-334.
- Wang, H., Liu, J., Cho, K.H. and Ren, D. 2009. A novel, single, transmembrane protein CATSPERG is associated with CATSPER1 channel protein. *Biol. Reprod.* 81: 539-544.

CHROMOSOMAL LOCATION

Genetic locus: CATSPERD (human) mapping to 19p13.3.

PRODUCT

CatSper δ 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 CatSper δ shRNA Plasmid (h): sc-97656-SH and CatSper δ shRNA (h) Lentiviral Particles: sc-97656-V as alternate gene silencing products.

For independent verification of CatSper δ (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-97656A, sc-97656B and sc-97656C.

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

CatSper δ siRNA (h) is recommended for the inhibition of CatSper δ 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

CatSper δ (A-7): sc-393749 is recommended as a control antibody for monitoring of CatSper δ 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 CatSper δ gene expression knockdown using RT-PCR Primer: CatSper δ (h)-PR: sc-97656-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.