SANTA CRUZ BIOTECHNOLOGY, INC.

DAZ siRNA (h): sc-270452

BACKGROUND

Spermatogenesis is the process by which male spermatogonia develop into mature spermatozoa. DAZ (deleted in azoospermia) are RNA-binding proteins that play an essential role in spermatogenesis. DAZ proteins influence the first stages of spermatogenesis and the maintenance of germ cell populations. DAZ proteins (DAZ1, DAZ2, DAZ3, DAZ4 and DAZ5) are encoded by separate genes on chromosome Y, each of which contain an AZFc domain in their codinga region. DAZ proteins are localized to the nucleus of spermatogonia, but relocate to the cytoplasm during meiosis. DAZ proteins contain an RRM (RNA recognition motif) domain that may regulate mRNA translation by binding to the 3'UTR. Deletions in the genes encoding DAZ proteins may cause azoospermia or oligospermia which can lead to male infertility.

REFERENCES

1. Reijo, R., et al. 1995. Diverse spermatogenic defects in humans caused by Y chromosome deletions encompassing a novel RNA-binding protein gene. Nat. Genet. 10: 383-393.
2. Tsui, S., et al. 2000. Identification of two novel proteins that interact with germ-cell-specific RNA-binding proteins DAZ and DAZL1. Genomics 65: 266-273.
3. Saxena, R., et al. 2000. Four DAZ genes in two clusters found in the AZFc region of the human Y chromosome. Genomics 67: 256-267.
4. Foresta, C., et al. 2002. Inhibin B plasma concentrations in infertile patients with DAZ gene deletions treated with FSH. Eur. J. Endocrinol. 6: 801-806.
5. Online Mendelian Inheritance in Man, OMIM™. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 400003. World Wide Web URL: http://www.ncbi.nlm.nih.gov/omim/
6. Skaletsky, H., et al. 2003. The male-specific region of the human Y chromosome is a mosaic of discrete sequence classes. Nature 423: 825-837.

CHROMOSOMAL LOCATION

Genetic locus: DAZ1/DAZ2 (human) mapping to Yq11.223, DAZ3/DAZ4 (human) mapping to Yq11.23.

PRODUCT

DAZ 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 DAZ shRNA Plasmid (h): sc-270452-SH and DAZ shRNA (h) Lentiviral Particles: sc-270452-V as alternate gene silencing products.

For independent verification of DAZ (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-270452A, sc-270452B and sc-270452C.

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

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

DAZ (Z6Q): sc-100705 is recommended as a control antibody for monitoring of DAZ 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 DAZ gene expression knockdown using RT-PCR Primer: DAZ (h)-PR: sc-270452-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.