

SPAG6 siRNA (h): sc-90404

BACKGROUND

SPAG6 (sperm-associated antigen 6), also known as PF16 or Repro-SA-1, is a cytoplasmic protein that contains eight armadillo repeats and belongs to the armadillo repeat family. Proteins with armadillo repeats typically participate in protein-protein interactions, suggesting that SPAG6 associates with other proteins and may function as a structural or regulatory protein. Sharing 64% amino acid identity, SPAG6 is the mammalian homolog of the *Chlamydomonas reinhardtii* protein pf16, a component of the central apparatus in flagella. Predominantly expressed in testis, SPAG6 is a central apparatus protein that plays an important role in the sperm tail and is essential for flagellar motility and male fertility. SPAG6 interacts with SPAG16 and, together, these proteins may cooperate in the regulation of sperm motility. Four isoforms exist for SPAG6 due to alternative splicing events.

REFERENCES

1. Neilson, L.I., et al. 1999. cDNA cloning and characterization of a human sperm antigen (SPAG6) with homology to the product of the *Chlamydomonas* PF16 locus. *Genomics* 60: 272-280.
2. Sapiro, R., et al. 2000. Sperm antigen 6 is the murine homologue of the *Chlamydomonas reinhardtii* central apparatus protein encoded by the PF16 locus. *Biol. Reprod.* 62: 511-518.
3. Sapiro, R., et al. 2002. Male infertility, impaired sperm motility, and hydrocephalus in mice deficient in sperm-associated antigen 6. *Mol. Cell. Biol.* 22: 6298-6305.
4. Online Mendelian Inheritance in Man, OMIM[™]. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 605730. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
5. Zhang, Z., et al. 2005. Dissecting the axoneme interactome: the mammalian orthologue of *Chlamydomonas* PF6 interacts with sperm-associated antigen 6, the mammalian orthologue of *Chlamydomonas* PF16. *Mol. Cell. Proteomics* 4: 914-923.

CHROMOSOMAL LOCATION

Genetic locus: SPAG6 (human) mapping to 10p12.2.

PRODUCT

SPAG6 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 SPAG6 shRNA Plasmid (h): sc-90404-SH and SPAG6 shRNA (h) Lentiviral Particles: sc-90404-V as alternate gene silencing products.

For independent verification of SPAG6 (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-90404A, sc-90404B and sc-90404C.

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

SPAG6 siRNA (h) is recommended for the inhibition of SPAG6 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

SPAG6 (H-4): sc-518235 is recommended as a control antibody for monitoring of SPAG6 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 SPAG6 gene expression knockdown using RT-PCR Primer: SPAG6 (h)-PR: sc-90404-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.