

SPAG1 siRNA (m): sc-153701

BACKGROUND

Mammalian sperm flagellum contain two cytoskeletal structures associated with the the axoneme: the outer dense fibers and the fibrous sheath. The outer dense fibers surround the axoneme in the midpiece and principal piece, whereas the fibrous sheath surrounds outer dense fibers of the tail of the principal piece. SPAG1 (sperm associated antigen 1), also known as SP75 or TPIS, is a 926 amino acid cytoplasmic protein detected in pachytene primary spermatocytes where it predominantly localizes to the neck and midpiece. Involved in fertilization, SPAG1 expression is also high in the cytoplasm of malignant pancreatic ductal adenocarcinomas (PDAC) and is therefore considered a novel marker of PDAC progression. Containing an ATP/GTP-binding site, multiple putative phosphorylation sites and three tetratricopeptide (TPR) motifs, SPAG1 possesses GTPase activity and binds GTP.

REFERENCES

1. Zhang, M.L., et al. 1992. Isolation and sequencing of the cDNA encoding the 75-kD human sperm protein related to infertility. *Chin. Med. J.* 105: 998-1003.
2. Schuler, G.D., et al. 1996. A gene map of the human genome. *Science* 274: 540-546.
3. Takaishi, M., et al. 1999. A tetratricopeptide repeat-containing protein gene, tpis, whose expression is induced with differentiation of spermatogenic cells. *Biochem. Biophys. Res. Commun.* 264: 81-85.
4. Kanazawa, R., et al. 2003. Isolation and characterization of a human sperm antigen gene h-Sp-1. *Int. J. Androl.* 26: 226-235.
5. Ficarro, S., et al. 2003. Phosphoproteome analysis of capacitated human sperm. Evidence of tyrosine phosphorylation of a kinase-anchoring protein 3 and valosin-containing protein/p97 during capacitation. *J. Biol. Chem.* 278: 11579-11589.
6. Liu, N., et al. 2006. A sperm component, HSD-3.8 (SPAG1), interacts with G-protein β 1 subunit and activates extracellular signal-regulated kinases (ERK). *Front. Biosci.* 11: 1679-1689.
7. Neesse, A., et al. 2007. Sperm-associated antigen 1 is expressed early in pancreatic tumorigenesis and promotes motility of cancer cells. *Oncogene* 26: 1533-1545.

CHROMOSOMAL LOCATION

Genetic locus: Spag1 (mouse) mapping to 15 B3.1.

PRODUCT

SPAG1 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see SPAG1 shRNA Plasmid (m): sc-153701-SH and SPAG1 shRNA (m) Lentiviral Particles: sc-153701-V as alternate gene silencing products.

For independent verification of SPAG1 (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-153701A, sc-153701B and sc-153701C.

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

SPAG1 siRNA (m) is recommended for the inhibition of SPAG1 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 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor SPAG1 gene expression knockdown using RT-PCR Primer: SPAG1 (m)-PR: sc-153701-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Huang, C., et al. 2016. The GTPase SPAG1 orchestrates meiotic program by dictating meiotic resumption and cytoskeleton architecture in mouse oocytes. *Mol. Biol. Cell* 27: 1776-1785.

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.