SANTA CRUZ BIOTECHNOLOGY, INC.

AKAP 3 siRNA (h): sc-60143

BACKGROUND

The type II cAMP-dependent protein kinase (PKA) is a multifunctional kinase with a broad range of substrates. Specificity of PKA signaling is mediated by the compartmentalization of the kinase to specific sites within the cell. To maintain this specific localization, the R subunit (RII) of PKA interacts with specific RII-anchoring proteins, designated A-kinase anchoring proteins (AKAP). AKAP 3, also known as AKAP 110, FSP95, PRKA3 and SOB1, binds both PKA and PDE4A and functions as a scaffolding protein in spermatozoa to regulate local cAMP concentrations and modulate sperm functions. Expression of AKAP 3 in normal tissues is restricted to the testis, where bicarbonate stimulates tyrosine phosphorylation of AKAP 3, thereby increasing its recruitment of PKA. AKAP-3 also exhibits high expression in patients with epithelial ovarian cancer (EOC). It demonstrates tumor-restricted expression and appears to be associated with worse overall survival, which make AKAP 3 a potential target for antigen-specific immunotherapy in EOC.

REFERENCES

1. Vijayaraghavan, S., et al. 1999. Isolation and molecular characterization of AKAP 110, a novel, sperm-specific protein kinase A-anchoring protein. Mol. Endocrinol. 13: 705-717.
2. Niu, J., et al. 2001. Interaction of heterotrimeric G13 protein with an A-kinase-anchoring protein 110 (AKAP 110) mediates cAMP-independent PKA activation. Curr. Biol. 11: 1686-1690.
3. Turner, R.M., et al. 2001. Molecular genetic analysis of two human sperm fibrous sheath proteins, AKAP 4 and AKAP 3, in men with dysplasia of the fibrous sheath. J. Androl. 22: 302-315.
4. Hasegawa, K., et al. 2003. A-kinase anchoring protein 3 messenger RNA expression in ovarian cancer and its implication on prognosis. Int. J. Cancer 108: 86-90.
5. Lea, I.A., et al. 2004. Association of sperm protein 17 with A-kinase anchoring protein 3 in flagella. Reprod. Biol. Endocrinol. 2: 57.
6. Luconi, M., et al. 2005. Tyrosine phosphorylation of the a kinase anchoring protein 3 (AKAP3) and soluble adenylate cyclase are involved in the increase of human sperm motility by bicarbonate. Biol. Reprod. 72: 22-32.
7. Bajpai, M., et al. 2005. AKAP3 selectively binds PDE4A isoforms in bovine spermatozoa. Biol. Reprod. 74: 109-118.
8. Sharma, S., et al. 2005. A-kinase anchoring protein 3 messenger RNA expression correlates with poor prognosis in epithelial ovarian cancer. Gynecol. Oncol. 99: 183-188.

CHROMOSOMAL LOCATION

Genetic locus: AKAP3 (human) mapping to 12p13.32.

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.

PRODUCT

AKAP 3 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 AKAP 3 shRNA Plasmid (h): sc-60143-SH and AKAP 3 shRNA (h) Lentiviral Particles: sc-60143-V as alternate gene silencing products.

For independent verification of AKAP 3 (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-60143A, sc-60143B and sc-60143C.

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

AKAP 3 siRNA (h) is recommended for the inhibition of AKAP 3 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor AKAP 3 gene expression knockdown using RT-PCR Primer: AKAP 3 (h)-PR: sc-60143-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.