

AKAP 95 siRNA (h): sc-29662

BACKGROUND

The type II cAMP-protein kinase (PKA) is a multifunctional kinase with a broad range of substrates. Specificity of PKA signaling is thought to be 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. The family of RII-anchoring proteins has been designated A-kinase anchoring proteins (AKAP). AKAP 95, also known as AKAP 8, is a nuclear matrix protein predominantly expressed in liver, heart, pancreas, kidney and skeletal muscle. During mitosis, AKAP 95 is recruited to the chromosomes and plays an essential role in mitotic progression. Characteristic of its family, AKAP 95 participates in PKA signaling through an interaction with the RII regulatory subunit. In addition, AKAP 95 forms a complex with HA95 and HDAC3 and is required for the deacetylation of Histone H3 in mitosis.

REFERENCES

1. Coghlan, V.M., et al. 1993. A-kinase anchoring proteins: a key to selective activation of cAMP-responsive events? *Mol. Cell. Biochem.* 127: 309-319.
2. Coghlan, V.M., et al. 1995. Association of protein kinase A and protein phosphatase 2B with a common anchoring protein. *Science* 267: 108-111.
3. Lester, L.B., et al. 1996. Cloning and characterization of a novel A-kinase anchoring protein. AKAP 220, association with testicular peroxisomes. *J. Biol. Chem.* 271: 9460-9465.
4. Collas, P., et al. 1999. The A-kinase-anchoring protein AKAP 95 is a multi-valent protein with a key role in chromatin condensation at mitosis. *J. Cell Biol.* 147: 1167-1180.
5. Arsenijevic, T., et al. 2004. A novel partner for D-type cyclins: protein kinase A-anchoring protein AKAP 95. *Biochem. J.* 378: 673-679.
6. Kamada, S., et al. 2005. A-kinase-anchoring protein 95 functions as a potential carrier for the nuclear translocation of active caspase 3 through an enzyme-substrate-like association. *Mol. Cell. Biol.* 25: 9469-9477.
7. Jungmann, R.A. and Kiryukhina, O. 2005. Cyclic AMP and AKAP-mediated targeting of protein kinase A regulates lactate dehydrogenase subunit A mRNA stability. *J. Biol. Chem.* 280: 25170-25177.

CHROMOSOMAL LOCATION

Genetic locus: AKAP8 (human) mapping to 1 19p13.12.

PRODUCT

AKAP 95 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 AKAP 95 shRNA Plasmid (h): sc-29662-SH and AKAP 95 shRNA (h) Lentiviral Particles: sc-29662-V as alternate gene silencing products.

For independent verification of AKAP 95 (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-29662A, sc-29662B and sc-29662C.

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

AKAP 95 siRNA (h) is recommended for the inhibition of AKAP 95 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

AKAP 95 (F-11): sc-390335 is recommended as a control antibody for monitoring of AKAP 95 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 AKAP 95 gene expression knockdown using RT-PCR Primer: AKAP 95 (h)-PR: sc-29662-PR (20 μ l, 586 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Zuo, H., et al. 2020. A-kinase anchoring proteins diminish TGF- β 1/cigarette smoke-induced epithelial-to-mesenchymal transition. *Cells* 9: 356.

RESEARCH USE

For research use only, not for use in diagnostic procedures.