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PDE4A siRNA (h): sc-41596

BACKGROUND

Phosphodiesterases (PDE) hydrolyze cAMP to 5'-AMP and thus play a critical role in the regulation of intracellular cAMP. Division of the PDE superfamily by sequence homology and enzymatic properties yields 11 PDE families. A unique upstream conserved region (UCR) characterizes the PDE4 family. Four separate genes (A-D) encode the PDE4 enzymes, and alternative splicing generates short or long isoforms of each gene. Long PDE4 isoforms contain both UCR1 and UCR2, while short PDE4 isoforms possess only UCR2. Both UCR domains are necessary for dimerization of PDE4 isoforms. The human PDE4A gene maps to chromosome 19p13.2 and spans 50 kilobases with 17 exons. The splice variants include isoforms PDE4A1-6.

REFERENCES

1. Bolger, G., et al. 1993. A family of human phosphodiesterases homologous to the dunce learning and memory gene product of *Drosophila melanogaster* are potential targets for antidepressant drugs. *Mol. Cell Biol.* 13: 6558-6571.
2. Horton, Y.M., et al. 1995. Molecular cloning of a novel splice variant of human type 4A (PDE4A) cyclic AMP phosphodiesterase and localization of the gene to the p13.2-q12 region of human chromosome 19. *Biochem. J.* 308: 683-691.
3. Francis, S.H., et al. 2001. Cyclic nucleotide phosphodiesterases: relating structure and function. *Prog. Nucleic Acid Res. Mol. Biol.* 65: 51-52.
4. Richter, W., et al. 2002. Dimerization of the type 4 cAMP-specific phosphodiesterases is mediated by the upstream conserved regions (UCRs). *J. Biol. Chem.* 277: 40212-40221.

CHROMOSOMAL LOCATION

Genetic locus: PDE4A (human) mapping to 19p13.2.

PRODUCT

PDE4A 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 PDE4A shRNA Plasmid (h): sc-41596-SH and PDE4A shRNA (h) Lentiviral Particles: sc-41596-V as alternate gene silencing products.

For independent verification of PDE4A (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-41596A, sc-41596B and sc-41596C.

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

PDE4A siRNA (h) is recommended for the inhibition of PDE4A 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

PDE4A (H-7): sc-74428 is recommended as a control antibody for monitoring of PDE4A 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 PDE4A gene expression knockdown using RT-PCR Primer: PDE4A (h)-PR: sc-41596-PR (20 μ l, 579 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Peng, Y., et al. 2018. PDE4A predicts poor prognosis and promotes metastasis by inducing epithelial-mesenchymal transition in hepatocellular carcinoma. *J. Cancer* 9: 2389-2396.

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.