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SANTA CRUZ BIOTECHNOLOGY, INC.

PDE9A siRNA (r): sc-270131

BACKGROUND

Phosphodiesterases (PDEs) also designated cyclic nucleotide phosphodiesterases, are important for the downregulation of the intracellular level of the second messenger cyclic adenosine monophosphate (cAMP) by hydrolyzing cAMP to 5'AMP. Phosphodiesterase 9A (PDE9A) is a 593 amino acid protein that plays a role in signal transduction via regulation of the intracellular concentration of cyclic nucleotides and has a high affinity for cGMP. There are 15 known isoforms of PDE9A. It is expressed in various tissues including testis, brain, small intestine, skeletal muscle, heart, lung, thymus, spleen, placenta, kidney, liver, pancreas, ovary and prostate. Highest levels of PDE9A expression occur in brain, kidney, spleen, colon, heart and colon, while there is no detection of PDE9A in blood. PDE9A is composed of an N-terminal regulatory domain and a C-terminal catalytic domain containing two possible divalent metal sites. It may be implicated in affective bipolar disorder.

REFERENCES

1. Shimosato, Y., et al. 1978. Squamous cell carcinoma of the thymus. An analysis of eight cases. Am. J. Surg. Pathol. 1: 109-121.
2. Soderling, S.H., et al. 1998. Identification and characterization of a novel family of cyclic nucleotide phosphodiesterases. J. Biol. Chem. 273: 15553-15558.
3. Fisher, D.A., et al. 1998. Isolation and characterization of PDE9A, a novel human cGMP-specific phosphodiesterase. J. Biol. Chem. 273: 15559-155564.
4. Guipponi, M., et al. 1998. Identification and characterization of a novel cyclic nucleotide phosphodiesterase gene (PDE9A) that maps to 21q22.3: alternative splicing of mRNA transcripts, genomic structure and sequence. Hum. Genet. 103: 386-392.
5. Rentero, C., et al. 2003. Identification and distribution of different mRNA variants produced by differential splicing in the human phosphodiesterase 9A gene. Biochem. Biophys. Res. Commun. 301: 686-692.
6. Wang, P., et al. 2003. Identification and characterization of a new human type 9 cGMP-specific phosphodiesterase splice variant (PDE9A5). Differential tissue distribution and subcellular localization of PDE9A variants. Gene 314: 15-27.

CHROMOSOMAL LOCATION

Genetic locus: Pde9a (rat) mapping to 20p12.

PRODUCT

PDE9A siRNA (r) 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 PDE9A shRNA Plasmid (r): sc-270131-SH and PDE9A shRNA (r) Lentiviral Particles: sc-270131-V as alternate gene silencing products.

For independent verification of PDE9A (r) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-270131A, sc-270131B and sc-270131C.

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

PDE9A siRNA (r) is recommended for the inhibition of PDE9A expression in rat 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.

GENE EXPRESSION MONITORING

PDE9A (D-7): sc-376271 is recommended as a control antibody for monitoring of PDE9A 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 PDE9A gene expression knockdown using RT-PCR Primer: PDE9A (r)-PR: sc-270131-PR (20 µl). 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.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.