SANTA CRUZ BIOTECHNOLOGY, INC.

PDE6G siRNA (h): sc-61311

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), as they are responsible for hydrolyzing cAMP to 5'AMP. PDE6G, also designated phosphodiesterase 6G cGMP-specific rod γ, is an oligomer composed of two catalytic chains (α and β), an inhibitory chain (γ) and an δ chain. PDEG6G functions in the processes of transmission and amplification of the visual signal. A mutation in the rod PDE-γ gene desensitizes and delays murine rod photoreceptors. PDE6H, also designated phosphodiesterase 6H cGMP-specific cone γ, is a tetramer composed of two catalytic chains (α and β), and two inhibitory chains (γ). It functions similarly to PDE6H in vision processes. Defects of the PDE6H gene cause retinal cone dystrophy 3 (rcd3), also designated cone dystrophy with night blindness and supernormal rod responses.

REFERENCES

1. Hamilton, S.E. and Hurley, J.B. 1990. A phosphodiesterase inhibitor specific to a subset of bovine retinal cones. J. Biol. Chem. 265: 11259-11264.
2. Tsang, S.H., Gouras, P., Yamashita, C.K., Kjeldbye, H., Fisher, J., Farber, D.B. and Goff, S.P. 1996. Retinal degeneration in mice lacking the γ subunit of the rod cGMP phosphodiesterase. Science 272: 1026-1029.
3. Shimizu-Matsumoto, A., Itoh, K., Inazawa, J., Nishida, K., Matsumoto, Y., Kinoshita, S., Matsubara, K. and Okubo, K. 1996. Isolation and chromosomal localization of the human cone cGMP phosphodiesterase γ cDNA (PDE6H). Genomics 32: 121-124.
4. Salchow, D.J., Gouras, P., Doi, K., Goff, S.P., Schwinger, E. and Tsang, S.H. 1999. A point mutation (W70A) in the rod PDE-γ gene desensitizing and delaying murine rod photoreceptors. Invest. Ophthalmol. Vis. Sci. 40: 3262-3267.

CHROMOSOMAL LOCATION

Genetic locus: PDE6G (human) mapping to 17q25.3.

PRODUCT

PDE6G 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 PDE6G shRNA Plasmid (h): sc-61311-SH and PDE6G shRNA (h) Lentiviral Particles: sc-61311-V as alternate gene silencing products.

For independent verification of PDE6G (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-61311A, sc-61311B and sc-61311C.

PROTOCOLS

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

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

PDE6G siRNA (h) is recommended for the inhibition of PDE6G 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.

GENE EXPRESSION MONITORING

PDE6G/H (A-2): sc-166350 is recommended as a control antibody for monitoring of PDE6G 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 PDE6G gene expression knockdown using RT-PCR Primer: PDE6G (h)-PR: sc-61311-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.