SANTA CRUZ BIOTECHNOLOGY, INC.

Phd siRNA (h): sc-40839

BACKGROUND

Phosducin is a phototransducing protein that may participate in the feedback regulation of visual phototransduction or in the integration of photoreceptor metabolism. The human phosducin gene maps to chromosome 1q31.1 and encodes a 246 amino acid protein, also designated Phd. Phd is primarily expressed in the retina and the pineal gland, while lower levels are present in tissues such as liver, spleen, striated muscle and the brain. Retinal Phd is found exclusively in the outer and inner segments of photoreceptor cells, including the synaptic and nuclear layers. Phd modulates the phototransduction cascade through high affinity binding and sequestration of $G_{\beta/\gamma}$ subunits of heterotrimeric G proteins. Neutralization of $G_{\beta/\gamma}$ by phosducin inhibits G protein-mediated signaling, since G_{α} is unable to reassemble with $G_{\beta/\gamma}$ and provide a functional G protein trimer ($G_{\alpha/\beta/\gamma}$). In addition, phosducin can effectively slow down the mechanism of internalization of G protein-coupled opioid receptors.

REFERENCES

1. Ding, C., et al. 1993. The gene for human phosducin (PDC), a soluble protein that binds G-protein β γ dimers, maps to 1q25-q31.1. Genomics 18: 457-459.
2. Thulin, C.D., et al. 1999. The immunolocalization and divergent roles of phosducin and phosducin-like protein in the retina. Mol. Vis. 5: 40.
3. Savage, J.R., et al. 2000. Functional roles of the two domains of phosducin and phosducin-like protein. J. Biol. Chem. 275: 30399-30407.
4. Online Mendelian Inheritance in Man, OMIM™. 2000. Johns Hopkins University, Baltimore, MD. MIM Number: 171490. World Wide Web URL: http://www.ncbi.nlm.nih.gov/omim/
5. Schulz, R. 2001. The pharmacology of phosducin. Pharmacol. Res. 43: 1-10.
6. LocusLink Report (LocusID: 5132). http://www.ncbi.nlm.nih.gov/LocusLink/

CHROMOSOMAL LOCATION

Genetic locus: PDC (human) mapping to 1q31.1.

PRODUCT

Phd 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 Phd shRNA Plasmid (h): sc-40839-SH and Phd shRNA (h) Lentiviral Particles: sc-40839-V as alternate gene silencing products.

For independent verification of Phd (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-40839A, sc-40839B and sc-40839C.

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

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

Phd (E-1): sc-398752 is recommended as a control antibody for monitoring of Phd 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 Phd gene expression knockdown using RT-PCR Primer: Phd (h)-PR: sc-40839-PR (20 µl, 561 bp). 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.