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PGD2 synthase siRNA (h): sc-41640

BACKGROUND

Human PGD synthase is the key enzyme for production of the D and J series of prostanoids in the immune system and mast cells. This enzyme is the first member of the sigma class glutathione S-transferases (GST) from vertebrates and contains a prominent cleft as the active site, which is unique among members of the GST superfamily. The human PGD synthase gene, which maps to chromosome 4q22.3, is expressed in a species-specific manner. For instance, the human gene is widely distributed, whereas the mouse gene is only expressed in oviduct and skin. Human PGD synthase is expressed in the cytoplasm of human megakaryoblastic CMK cells prior to differentiation into platelets, which have no PGD synthase activity. Another member of the PGD synthase family, PGD2 synthase, catalyzes the conversion of PGH₂ to PGD₂ and is essential for the synthesis of PGD₂ in the brain. Unlike PGD synthase, PGD₂ synthase is not dependent on the presence of glutathione for its activity. The human PGD₂ synthase gene maps to chromosome 9q34.3.

REFERENCES

1. Nagata, A., et al. 1991. Human brain prostaglandin D synthase has been evolutionarily differentiated from lipophilic-ligand carrier proteins. *Proc. Natl. Acad. Sci. USA* 88: 4020-4024.
2. Mahmud, I., et al. 1997. Prostaglandin D synthase in human megakaryoblastic cells. *J. Biol. Chem.* 272: 28263-28266.
3. Kanaoka, Y., et al. 1997. Cloning and crystal structure of hematopoietic prostaglandin D synthase. *Cell* 90: 1085-1095.

CHROMSOMAL LOCATION

Genetic locus: PTGDS (human) mapping to 9q34.3.

PRODUCT

PGD₂ synthase 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 PGD₂ synthase shRNA Plasmid (h): sc-41640-SH and PGD₂ synthase shRNA (h) Lentiviral Particles: sc-41640-V as alternate gene silencing products.

For independent verification of PGD₂ synthase (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-41640A, sc-41640B and sc-41640C.

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

PGD₂ synthase siRNA (h) is recommended for the inhibition of PGD₂ synthase 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

PGD₂ synthase (F-7): sc-390717 is recommended as a control antibody for monitoring of PGD₂ synthase 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 PGD₂ synthase gene expression knockdown using RT-PCR Primer: PGD₂ synthase (h)-PR: sc-41640-PR (20 μl, 468 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Zhu, F., et al. 2010. Prolonged application of high fluid shear to chondrocytes recapitulates gene expression profiles associated with osteoarthritis. *PLoS ONE* 5: e15174.
2. Zhu, F., et al. 2010. Prostaglandin (PG)D₂ and 15-deoxy-Δ^{12,14}-PGJ₂, but not PGE₂, mediate shear-induced chondrocyte apoptosis via protein kinase A-dependent regulation of polo-like kinases. *Cell Death Differ.* 17: 1325-1334.

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.