SANTA CRUZ BIOTECHNOLOGY, INC.

PGD siRNA (h): sc-78779

BACKGROUND

PGD (phosphogluconate dehydrogenase), also referred to as 6PGD, is a 483 amino acid enzyme that is involved in the pentose phosphate shunt. Pentose is required for nucleic acid biosynthesis and the pentose phosphate cycle is a major source of NADPH. As the second dehydrogenase in the pentose phosphate cycle, PGD catalyzes the oxidative decarboxylation of PGD to ribulose 5-phosphate, following the release of CO_2 and the reduction of NADP. PGD deficiency increases the level of erythrocyte pyruvate kinase (PK) activity and reduces glutathione synthetase (GSH), resulting in hemolysis. Defects in PGD are generally asymptomatic and are inherited in an autosomal dominant fashion. Catalytic active regions of PGD, such as those forming the substrate and coenzyme binding sites, are highly conserved in most species.

REFERENCES

1. Weitkamp, L.R., et al. 1970. Genetic linkage relations of the loci for 6-phosphogluconate dehydrogenase and adenosine deaminase in man. Am. J. Hum. Genet. 22: 216-220.
2. Niehaus, W.G., et al. 1996. Slow-binding inhibition of 6-phosphogluconate dehydrogenase by zinc ion. Arch. Biochem. Biophys. 333: 333-337.
3. Rippa, M., et al. 1998. 6-phosphogluconate dehydrogenase: the mechanism of action investigated by a comparison of the enzyme from different species. Biochim. Biophys. Acta 1429: 83-92.
4. Caprari, P., et al. 2001. 6-phosphogluconate dehydrogenase deficiency in an Italian family. Ann. Hematol. 80: 41-44.
5. Hanau, S., et al. 2004. 6-phosphogluconate dehydrogenase: a target for drugs in African trypanosomes. Curr. Med. Chem. 11: 2639-2650.
6. Goulielmos, G.N., et al. 2004. Functional constraints of 6-phosphogluconate dehydrogenase (6-PGD) based on sequence and structural information. J. Mol. Evol. 59: 358-371.
7. Ceyhan, D., et al. 2005. Purification and kinetic properties of 6-phosphogluconate dehydrogenase from rat small intestine. Protein J. 24: 293-301.

CHROMOSOMAL LOCATION

Genetic locus: PGD (human) mapping to 1p36.22.

PRODUCT

PGD 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 PGD shRNA Plasmid (h): sc-78779-SH and PGD shRNA (h) Lentiviral Particles: sc-78779-V as alternate gene silencing products.

For independent verification of PGD (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-78779A, sc-78779B and sc-78779C.

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

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

PGD (G-2): sc-398977 is recommended as a control antibody for monitoring of PGD 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 PGD gene expression knockdown using RT-PCR Primer: PGD (h)-PR: sc-78779-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.