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PDI siRNA (h2): sc-44319

BACKGROUND

Oxidoreductase-protein disulfide isomerase (PDI) is a homodimer consisting of subunits that catalyzes thiol-disulfide exchange, mediates folding of newly synthesized proteins and functions as a molecular chaperone. PDI localizes to the lumen of the endoplasmic reticulum (ER), where in conjunction with folding-helper proteins, such as immunoglobulin heavy chain binding protein (BiP), it mediates tertiary and quaternary protein processing. Cell surface PDI induces sulfhydryl-mediated conformational changes in integrin-mediated adhesion receptor-ligand interactions, thereby regulating integrin responses and cell adhesion. Additionally, PDI functions as a subunit of two more complex enzyme systems: the prolyl-4-hydroxylase and the triacylglycerol transfer proteins.

REFERENCES

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- Mayer, M., Kies, U., Kammermeier, R. and Buchner, J. 2000. BiP and PDI cooperate in the oxidative folding of antibodies *in vitro*. *J. Biol. Chem.* 275: 29421-29425.
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CHROMOSOMAL LOCATION

Genetic locus: P4HB (human) mapping to 17q25.3.

PRODUCT

PDI siRNA (h2) 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see PDI shRNA Plasmid (h2): sc-44319-SH and PDI shRNA (h2) Lentiviral Particles: sc-44319-V as alternate gene silencing products.

For independent verification of PDI (h2) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-44319A, sc-44319B and sc-44319C.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

PROTOCOLS

See our web site at www.scbt.com or our catalog 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 μ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330 μ l of RNase-free water makes a 10 μ M solution in a 10 μ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

PDI siRNA (h2) is recommended for the inhibition of PDI 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 μ M in 66 μ l. 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

PDI (A-1): sc-376370 is recommended as a control antibody for monitoring of 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 (secondary) reagents are recommended: 1) Western Blotting: use goat anti-mouse IgG-HRP: sc-2005 (dilution range: 1:2000-1:32,000) or Cruz Marker™ compatible goat anti-mouse IgG-HRP: sc-2031 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use goat anti-mouse IgG-FITC: sc-2010 (dilution range: 1:100-1:400) or goat anti-mouse IgG-TR: sc-2781 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor PDI gene expression knockdown using RT-PCR Primer: PDI (h2)-PR: sc-44319-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.