SANTA CRUZ BIOTECHNOLOGY, INC.

PDI siRNA (h3): sc-270309

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), 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

1. Burgess, J.K., et al. 2000. Physical proximity and functional association of glycoprotein 1bα and protein-disulfide isomerase on the platelet plasma membrane. J. Biol. Chem. 275: 9758-9766.
2. Klappa, P., et al. 2000. Mutations that destabilize the a' domain of human protein-disulfide isomerase indirectly affect peptide binding. J. Biol. Chem. 275: 13213-13218.
3. Mayer, M., et al. 2000. BiP and PDI cooperate in the oxidative folding of antibodies *in vitro*. J. Biol. Chem. 275: 29421-29425.
4. Lahav, J., et al. 2000. Protein disulfide isomerase mediates integrin-dependent adhesion. FEBS Lett. 475: 89-92.
5. Maattanen, P., et al. 2006. ERp57 and PDI: multifunctional protein disulfide isomerases with similar domain architectures but differing substrate-partner associations. Biochem. Cell Biol. 84: 881-889.
6. Nuss, J.E., et al. 2008. Decreased enzyme activities of chaperones PDI and BiP in aged mouse livers. Biochem. Biophys. Res. Commun. 365: 355-361.
7. Appenzeller-Herzog, C., et al. 2008. The human PDI family: versatility packed into a single fold. Biochim. Biophys. Acta 1783: 535-548.
8. Shnyder, S.D., et al. 2008. Triplex profiling of functionally distinct chaperones (ERp29/PDI/BiP) reveals marked heterogeneity of the endoplasmic reticulum proteome in cancer. J. Proteome Res. 7: 3364-3372.

CHROMOSOMAL LOCATION

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

PRODUCT

PDI siRNA (h3) is a pool of 4 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 PDI shRNA Plasmid (h3): sc-270309-SH and PDI shRNA (h3) Lentiviral Particles: sc-270309-V as alternate gene silencing products.

For independent verification of PDI (h3) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-270309A, sc-270309B, sc-270309C and sc-270309D.

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

PDI siRNA (h3) 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 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

PDI (C-2): sc-74551 is recommended as a control antibody for monitoring of PDI 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 PDI gene expression knockdown using RT-PCR Primer: PDI (h3)-PR: sc-270309-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.

PROTOCOLS

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