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PIDD siRNA (h): sc-44656

BACKGROUND

PIDD (for p53 induced protein with a death domain) encodes a protein of 915 amino acids in mice (910 amino acids in humans) and contains seven tandem leucine rich repeats (LRR) in the amino terminus and a death domain in the carboxy terminus. PIDD mRNA is induced by γ -irradiation in a p53-dependent manner and the basal level of PIDD mRNA is dependent on p53 status. Over-expression of PIDD inhibits cell growth in a p53-like manner by inducing apoptosis. Antisense inhibition of PIDD expression has been shown to attenuate p53-mediated apoptosis, suggesting that PIDD expression is required for apoptosis. PIDD localizes to the cytosol.

REFERENCES

1. Lin, Y., et al. 2000. PIDD, a new death-domain-containing protein, is induced by p53 and promotes apoptosis. *Nat. Genet.* 26: 122-127.
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3. Benchimol, S., et al. 2001. p53-dependent pathways of apoptosis. *Cell Death Differ.* 8: 1049-1051.
4. Tinel, A., et al. 2004. The PIDDosome, a protein complex implicated in activation of caspase-2 in response to genotoxic stress. *Science* 304: 843-846.
5. Lai, M.D., et al. 2005. Phosphorylated and hypoacetylated mutant p53 enhances cisplatin-induced apoptosis through caspase-9 pathway in the absence of transcriptional activation or translation. *Int. J. Mol. Med.* 15: 725-734.
6. Nie, D.S., et al. 2005. Identification of a novel testis-specific gene mtLR1, which is expressed at specific stages of mouse spermatogenesis. *Biochem. Biophys. Res. Commun.* 328: 1010-1018.
7. Ren, J., et al. 2005. The Birc6 (Bruce) gene regulates p53 and the mitochondrial pathway of apoptosis and is essential for mouse embryonic development. *Proc. Natl. Acad. Sci. USA* 102: 565-570.

CHROMOSOMAL LOCATION

Genetic locus: PIDD1 (human) mapping to 11p15.5.

PRODUCT

PIDD 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see PIDD shRNA Plasmid (h): sc-44656-SH and PIDD shRNA (h) Lentiviral Particles: sc-44656-V as alternate gene silencing products.

For independent verification of PIDD (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-44656A, sc-44656B and sc-44656C.

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 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

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

PIDD (B-5): sc-514981 is recommended as a control antibody for monitoring of PIDD 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-IgG κ BP-HRP: sc-516102 or m-IgG κ 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-IgG κ BP-FITC: sc-516140 or m-IgG κ 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 PIDD gene expression knockdown using RT-PCR Primer: PIDD (h)-PR: sc-44656-PR (20 μ l, 542 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.