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PRPK siRNA (m): sc-152499

BACKGROUND

p53, a DNA-binding, oligomerization domain- and transcription activation domain-containing tumor suppressor, upregulates growth arrest and apoptosis-related genes in response to stress signals, thereby influencing programmed cell death, cell differentiation and cell cycle control mechanisms. PRPK (p53-related protein kinase), also known as TP53RK, is a 253 amino acid protein kinase that phosphorylates Ser15 of p53. PRPK phosphorylation of p53 causes increased stabilization and activity of p53. CGI-121 may act as an inhibitor of the PRPK-p53 interaction, thus preventing the phosphorylation of p53. Unphosphorylated p53 is degraded by the ubiquitin-proteasome pathway, which may ultimately lead to cell proliferation. PRPK contains a protein kinase domain with a conserved catalytic core. PRPK is localized to the nucleus of the cell and is highly expressed in testis, with lower expression in heart, kidney and spleen.

REFERENCES

1. Abe, Y., et al. 2001. Cloning and characterization of a p53-related protein kinase expressed in interleukin-2-activated cytotoxic T cells, epithelial tumor cell lines, and the testes. *J. Biol. Chem.* 276: 44003-44011.
2. Online Mendelian Inheritance in Man, OMIM™. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 608679. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
3. Facchin, S., et al. 2003. Functional homology between yeast piD261/BUD32 and human PRPK: both phosphorylate p53 and PRPK partially complements piD261/Bud32 deficiency. *FEBS Lett.* 549: 63-66.
4. Miyoshi, A., et al. 2003. Identification of CGI-121, a novel PRPK (p53-related protein kinase)-binding protein. *Biochem. Biophys. Res. Commun.* 303: 399-405.
5. Abe, Y., et al. 2006. A Small Ras-like protein Ray/Rab1c modulates the p53-regulating activity of PRPK. *Biochem. Biophys. Res. Commun.* 344: 377-385.
6. Facchin, S., et al. 2007. Phosphorylation and activation of the atypical kinase p53-related protein kinase (PRPK) by Akt/PKB. *Cell. Mol. Life Sci.* 64: 2680-2689.

CHROMOSOMAL LOCATION

Genetic locus: Trp53rk (mouse) mapping to 2 H3.

PRODUCT

PRPK siRNA (m) 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 PRPK shRNA Plasmid (m): sc-152499-SH and PRPK shRNA (m) Lentiviral Particles: sc-152499-V as alternate gene silencing products.

For independent verification of PRPK (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-152499A, sc-152499B and sc-152499C.

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

PRPK siRNA (m) is recommended for the inhibition of PRPK expression in mouse 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

PRPK (C-1): sc-514703 is recommended as a control antibody for monitoring of PRPK 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 PRPK gene expression knockdown using RT-PCR Primer: PRPK (m)-PR: sc-152499-PR (20 μ l). 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.