

Rb siRNA (h): sc-29468

BACKGROUND

Pediatric cancer retinoblastoma and the formation of other human tumors can be attributed to mutations in the retinoblastoma tumor suppressor gene. The retinoblastoma tumor suppressor gene product, known as Rb or pRb, regulates differentiation, apoptosis and cell cycle control by coordinating the cell cycle, at G₁/S, with transcriptional machinery that includes the heterodimeric E2F family. During G₁, cyclin D (D1, D2, D3)-dependent kinase-mediated phosphorylation of Rb at Ser 795 marks the conversion of Rb from a transcriptionally repressive, hypophosphorylated state to an inactive, phosphorylated state, which may be sustained through mitosis by differential phosphorylation of up to 16 putative serine or threonine residues, including Ser 249/Thr 252, Thr 373, Thr 356, Ser 780, Ser 807/Ser 811 and Thr 821/Thr 826. Hypophosphorylated Rb represses the transcription of genes controlling cell cycle through direct protein-protein interactions, by binding and inactivating the promoters of transcription factors, and through recruitment of histone deacetylase, which deacetylates promoter regions and enhances nucleosome formation, thereby masking transcription enhancing *cis* elements.

CHROMOSOMAL LOCATION

Genetic locus: RB1 (human) mapping to 13q14.2.

PRODUCT

Rb siRNA (h) is a target-specific 19-25 nt siRNA 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 Rb shRNA Plasmid (h): sc-29468-SH and Rb shRNA (h) Lentiviral Particles: sc-29468-V as alternate gene silencing 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

Rb siRNA (h) is recommended for the inhibition of Rb 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.

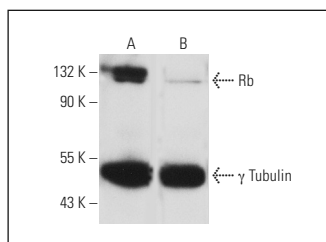
GENE EXPRESSION MONITORING

Rb (IF8): sc-102 is recommended as a control antibody for monitoring of Rb 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).

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Rb gene expression knockdown using RT-PCR Primer: Rb (h)-PR: sc-29468-PR (20 μ l, 526 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

DATA



Rb siRNA (h): sc-29468. Western blot analysis of Rb expression in non-transfected control (A) and Rb siRNA transfected (B) HeLa cells. Blot probed with Rb (IF8): sc-102. γ Tubulin (C-11): sc-17787 used as specificity and loading control.

SELECT PRODUCT CITATIONS

- Kinney, E.L., et al. 2008. Adenovirus E1A oncoprotein liberates c-Myc activity to promote cell proliferation through abating Bin1 expression via an Rb/E2F-1-dependent mechanism. *J. Cell. Physiol.* 216: 621-631.
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- Chatterjee, A., et al. 2015. Rapamycin-induced G₁ cell cycle arrest employs both TGF- β and Rb pathways. *Cancer Lett.* 360: 134-140.
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- Choi, C., et al. 2019. Cyclin D1 is associated with radiosensitivity of triple-negative breast cancer cells to proton beam irradiation. *Int. J. Mol. Sci.* 20: 4943.
- Das, A., et al. 2020. Evaluating anti-tumor activity of palbociclib plus radiation in anaplastic and radiation-induced meningiomas: pre-clinical investigations. *Clin. Transl. Oncol.* 22: 2017-2025.

RESEARCH USE

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