



P2P-R siRNA (m): sc-40901

BACKGROUND

Rb protein and p53 are both cell cycle checkpoint components. Evidence suggests that p53 plays a role in regulating the phosphorylation of Rb by inducing p21 transcription, thus preventing Rb phosphorylation at the G₁ to S transition. Protein-protein interactions seem to be central in p53 cellular activities, as previously demonstrated with MDM2 and SV40 large T antigen. Two novel proteins have been identified by their abilities to bind to p53 and/or Rb. Human RBQ-1 (also designated RBBP6) has been cloned as a novel protein that binds to the retinoblastoma (Rb) gene product. A related mouse protein, P2P-R, also designated PACT (for p53 associated cellular protein-testis derived), has been shown to bind to both Rb and p53. Recombinant P2P-R binds to wildtype p53 but not to mutant p53, and it can interfere with p53 specific DNA binding. RBQ-1 may be a truncated human form of the P2P-R protein.

REFERENCES

1. Lane, D.P., et al. 1979. T antigen is bound to a host protein in SV4-transformed cells. *Nature* 278: 261-263.
2. DeCaprio, J.A., et al. 1988. SV40 large tumor antigen forms a specific complex with the product of the retinoblastoma susceptibility gene. *Cell* 54: 275-283.
3. Sturzbecher, H.W., et al. 1988. Mouse p53 blocks SV40 DNA replication *in vitro* and downregulates T antigen DNA helicase activity. *Oncogene* 3: 405-413.
4. Chellappan, S.P., et al. 1991. The E2F transcription factor is a cellular target for the Rb protein. *Cell* 65: 1053-1061.
5. Momand, J., et al. 1992. The MDM2 oncogene product forms a complex with the p53 protein and inhibits p53-mediated transactivation. *Cell* 69: 1237-1245.
6. El-Deiry, W.S., et al. 1993. WAF1, a potential mediator of p53 tumor suppression. *Cell* 75: 817-825.
7. Sakai, Y., et al. 1995. cDNA sequence and chromosomal localization of a novel human protein, RBQ-1 (RBBP6), that binds to the retinoblastoma gene product. *Genomics* 30: 98-101.
8. Simons, A., et al. 1997. PACT: cloning and characterization of a cellular p53 binding protein that interacts with Rb. *Oncogene* 14: 145-155.

CHROMOSOMAL LOCATION

Genetic locus: Rbbp6 (mouse) mapping to 7 F3.

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.

PRODUCT

P2P-R 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 P2P-R shRNA Plasmid (m): sc-40901-SH and P2P-R shRNA (m) Lentiviral Particles: sc-40901-V as alternate gene silencing products.

For independent verification of P2P-R (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-40901A, sc-40901B and sc-40901C.

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

P2P-R siRNA (m) is recommended for the inhibition of P2P-R 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

P2P-R (M56): sc-9962 is recommended as a control antibody for monitoring of P2P-R 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 P2P-R gene expression knockdown using RT-PCR Primer: P2P-R (m)-PR: sc-40901-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.