

## RP1 siRNA (h): sc-37609

### BACKGROUND

APC (adenomatous polyposis coli) is a tumor suppressor gene that is frequently mutated in colorectal cancers and is one of the earliest indicators of colorectal carcinogenesis. APC is widely expressed and is largely similar to the related brain-specific homolog APCL. These proteins both associate with  $\beta$ -catenin and functionally regulate the levels of intracellular  $\beta$ -catenin. Additionally, these homologs interact with the microtubule cytoskeletal protein EB1 to regulate cell cycle progression or growth control. EB1 family proteins (EB1, RP1 (EB2) and EB3) interact with cytoplasmic microtubules in interphase cells, with mitotic spindles, and with the APC tumor suppressor gene. The functional inactivation of the APC gene product is a key event in colorectal tumorigenesis. RP1 is localized in the plus ends of microtubule networks in the presence or absence of APC. The gene which encodes RP1 maps to human chromosome 18q12.1.

### REFERENCES

1. Cottrell, S., et al. 1992. Molecular analysis of APC mutations in familial adenomatous polyposis and sporadic colon carcinomas. *Lancet* 340: 626-630.
2. Su, L.K., et al. 1993. Association of the APC tumor suppressor protein with catenins. *Science* 262: 1734-1737.
3. Nakagawa, H., et al. 1998. Identification of a brain-specific APC homologue, APCL, and its interaction with  $\beta$ -catenin. *Cancer Res.* 58: 5176-5181.
4. Morrison, E.E., et al. 1998. EB1, a protein which interacts with the APC tumour suppressor, is associated with the microtubule cytoskeleton throughout the cell cycle. *Oncogene* 17: 3471-3477.
5. Juwana, J.P., et al. 1999. EB/RP gene family encodes tubulin binding proteins. *Int. J. Cancer* 81: 275-284.
6. Nakagawa, H., et al. 2000. APCL, a central nervous system-specific homologue of adenomatous polyposis coli tumor suppressor, binds to p53-binding protein 2 and translocates it to the perinucleus. *Cancer Res.* 60: 101-105.
7. Wadle, A., et al. 2001. Chromosomal localization and promoter analysis of the adenomatous polyposis coli binding protein RP1. *Oncogene* 20: 5920-5929.

### CHROMOSOMAL LOCATION

Genetic locus: MAPRE2 (human) mapping to 18q12.1.

### PRODUCT

RP1 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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see RP1 shRNA Plasmid (h): sc-37609-SH and RP1 shRNA (h) Lentiviral Particles: sc-37609-V as alternate gene silencing products.

For independent verification of RP1 (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-37609A, sc-37609B and sc-37609C.

### 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  $\mu$ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330  $\mu$ l of RNase-free water makes a 10  $\mu$ M solution in a 10  $\mu$ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

### APPLICATIONS

RP1 siRNA (h) is recommended for the inhibition of RP1 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  $\mu$ M in 66  $\mu$ 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

RP1 (F-11): sc-390375 is recommended as a control antibody for monitoring of RP1 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 $\kappa$  BP-HRP: sc-516102 or m-IgG $\kappa$  BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker<sup>TM</sup> Molecular Weight Standards: sc-2035, UltraCruz<sup>®</sup> Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG $\kappa$  BP-FITC: sc-516140 or m-IgG $\kappa$  BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz<sup>®</sup> Mounting Medium: sc-24941 or UltraCruz<sup>®</sup> Hard-set Mounting Medium: sc-359850.

### RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor RP1 gene expression knockdown using RT-PCR Primer: RP1 (h)-PR: sc-37609-PR (20  $\mu$ 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](http://www.scbt.com) for detailed protocols and support products.