RAP1 siRNA (m): sc-38555



The Power to Question

BACKGROUND

Telomeres are DNA-protein structures that protect the ends of linear chromosomes and help maintain genomic stability and cell phenotype. Mammalian telomeric proteins consist of TRF1 (telomeric repeat binding factor), TRF2, tankyrase, and TIN2, which have no recognized orthologs in the budding yeast, *Saccharomyces cerevisiae*, and RAP1, which is an ortholog to the yeast telomeric protein scRap1. Like scRap1, mammalian RAP1 regulates telomere elongation. RAP1 interacts with two proteins, Rif1 and Rif2, which contribute to telomere length homeostasis. Unlike scRap1, which binds telomeric DNA directly, RAP1 is recruited to telomeres by TRF2. The functional and structural similarities of scRap1 to mammalian RAP1 suggest that the budding yeast preserved RAP1 at telomeres, but lost the TRF component. The telomeric protein TRF1 requires TIN2 to control telomere length in human cells.

REFERENCES

- 1. Marcand, S., et al. 1997. A protein-counting mechanism for telomere length regulation in yeast. Science 275: 986-990.
- Wotten, D. et al. 1997. A novel RAP1p-interacting factor, Rif2p, cooperates with Rif1p to regulate telomere length in *Saccharomyces cerevisiae*. Genes Dev. 11: 748-760.
- 3. Kim, S.H., et al. 1999. TIN2, a new regulator of telomere length in human cells. Nat. Genet. 23: 405-412.
- Scherthan, H., et al. 2000. Mammalian meiotic telomeres: protein composition and redistribution in relation to nuclear pores. Mol. Cell. Biol. 11: 4189-203.
- Li, B., et al. 2000. Identification of human RAP1: implications for telomere evolution. Cell 101: 471-483.
- Arthur, W.T., et al. 2004. RAP1 promotes cell spreading by localizing Rac guanine nucleotide exchange factors. J. Cell. Biol. 167: 111-122.

CHROMOSOMAL LOCATION

Genetic locus: TERF2IP (human) mapping to 16q22.3; Terf2ip (mouse) mapping to 8 E1.

PRODUCT

RAP1 siRNA (m) is a pool of 3 target-specific 20-25 nt siRNAs designed to knock down gene expression. Each vial contains 3 nmol of lyophilized siRNA, sufficient for a 10 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections.

For independent verification of RAP1 (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3 nmol of lyophilized siRNA. These include: sc-38555A, sc-38555B and sc-38555C.

RESEARCH USE

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

PROTOCOLS

See our web site at www.scbt.com or our catalog 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

RAP1 siRNA (m) is recommended for the inhibition of RAP1 expression in mouse cells.

RAP1 (N-20): sc-13652 is recommended as a control antibody for Western Blotting (starting dilution 1:100, dilution range 1:100-1:1,000) or immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500) protein detection using the recommended secondary reagents listed below.

RECOMMENDED SECONDARY REAGENTS

To ensure optimal results, the following support (secondary) reagents are recommended: 1) Western Blotting: use donkey anti-goat IgG-HRP: sc-2020 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible donkey anti-goat IgG-HRP: sc-2033 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use donkey anti-goat IgG-FITC: sc-2024 (dilution range: 1:100-1:400) or donkey anti-goat IgG-TR: sc-2783 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

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 $60~\mu l$. 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. Semi-quantitative RT-PCR may be performed using RT-PCR Primer: RAP1 (m)-PR: sc-38555-PR (20 μl).

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