

RAP55 siRNA (m): sc-152701

BACKGROUND

RAP55, also known as LSM14A (LSM14 homolog A), is a 463 amino acid member of the LSM14 family that contains a FFD-TFG box domain. The LSM14 domain and the RGG repeats of RAP55 are required for accumulation in P-bodies, and the region containing the FDF motif is responsible for cytoplasmic retention. RAP55, and all other Sm-like proteins, contain the Sm sequence motif, which consists of two regions separated by a linker of variable length that folds as a loop. The Sm-like proteins are thought to form a stable heteromer present in tri-snRNP particles, which are important for pre-mRNA splicing. RAP55 is phosphorylated upon DNA damage, probably by Atm or ATR. Existing as two alternatively spliced isoforms, the RAP55 gene is conserved in chimpanzee, canine, bovine, mouse, rat, chicken, zebrafish, mosquito, *Drosophila*, *C. elegans*, *S. pombe*, *S. cerevisiae*, *K. lactis*, *E. gossypii*, *M. grisea* and *N. crassa*, and maps to human chromosome 19q13.11.

REFERENCES

1. Lieb, B., et al. 1998. Identification of a novel mRNA-associated protein in oocytes of *Pleurodeles waltl* and *Xenopus laevis*. *Exp. Cell Res.* 245: 272-281.
2. Tanaka, K.J., et al. 2006. RAP55, a cytoplasmic mRNP component, represses translation in *Xenopus* oocytes. *J. Biol. Chem.* 281: 40096-40106.
3. Yang, W.H., et al. 2006. RNA-associated protein 55 (RAP55) localizes to mRNA processing bodies and stress granules. *RNA* 12: 547-554.
4. Matsuoka, S., et al. 2007. Atm and ATR substrate analysis reveals extensive protein networks responsive to DNA damage. *Science* 316: 1160-1166.
5. Online Mendelian Inheritance in Man, OMIM[™]. 2007. Johns Hopkins University, Baltimore, MD. MIM Number: 610677. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
6. Dephoure, N., et al. 2008. A quantitative atlas of mitotic phosphorylation. *Proc. Natl. Acad. Sci. USA* 105: 10762-10767.
7. Marnef, A., et al. 2009. RAP55: insights into an evolutionarily conserved protein family. *Int. J. Biochem. Cell Biol.* 41: 977-981.

CHROMOSOMAL LOCATION

Genetic locus: Lsm14a (mouse) mapping to 7 B1.

PRODUCT

RAP55 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 RAP55 shRNA Plasmid (m): sc-152701-SH and RAP55 shRNA (m) Lentiviral Particles: sc-152701-V as alternate gene silencing products.

For independent verification of RAP55 (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-152701A, sc-152701B and sc-152701C.

RESEARCH USE

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

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

RAP55 siRNA (m) is recommended for the inhibition of RAP55 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

RAP55 (C-5): sc-398552 is recommended as a control antibody for monitoring of RAP55 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 RAP55 gene expression knockdown using RT-PCR Primer: RAP55 (m)-PR: sc-152701-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.