

SMG1 siRNA (m): sc-76522

BACKGROUND

The phosphorylation and dephosphorylation of proteins on serine and threonine residues is an essential means of regulating a broad range of cellular functions in eukaryotes, including cell division, homeostasis and apoptosis. A group of proteins that are intimately involved in this process are the serine/threonine (Ser/Thr) protein kinases. SMG1, also known as ATX or LIP, is a 3,657 amino acid protein that localizes to both the nucleus and the cytoplasm and contains one FAT domain, one FATC domain, one HEAT repeat and one PI3K domain. Expressed in a variety of tissues, including heart and skeletal muscle, SMG1 functions as a Ser/Thr protein kinase that uses manganese as a cofactor to catalyze the phosphorylation of target proteins. Via its catalytic activity, SMG1 plays an important role in mRNA surveillance and genotoxic stress-induced response pathways. Multiple isoforms of SMG1 exist due to alternative splicing events.

REFERENCES

1. Yamashita, A., et al. 2001. Human SMG1, a novel phosphatidylinositol 3-kinase-related protein kinase, associates with components of the mRNA surveillance complex and is involved in the regulation of nonsense-mediated mRNA decay. *Genes Dev.* 15: 2215-2228.
2. Denning, G., et al. 2001. Cloning of a novel phosphatidylinositol kinase-related kinase: characterization of the human SMG1 RNA surveillance protein. *J. Biol. Chem.* 276: 22709-22714.
3. Online Mendelian Inheritance in Man, OMIM[™]. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 607032. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
4. Abraham, R.T. 2004. The ATM-related kinase, hSMG1, bridges genome and RNA surveillance pathways. *DNA Repair* 3: 919-925.

CHROMOSOMAL LOCATION

Genetic locus: Smg1 (mouse) mapping to 7 F2.

PRODUCT

SMG1 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 SMG1 shRNA Plasmid (m): sc-76522-SH and SMG1 shRNA (m) Lentiviral Particles: sc-76522-V as alternate gene silencing products.

For independent verification of SMG1 (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-76522A, sc-76522B and sc-76522C.

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.

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

SMG1 siRNA (m) is recommended for the inhibition of SMG1 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

SMG1 (E-4): sc-374557 is recommended as a control antibody for monitoring of SMG1 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 SMG1 gene expression knockdown using RT-PCR Primer: SMG1 (m)-PR: sc-76522-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.