

SNARK siRNA (m): sc-153652

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. SNARK (SNF1/AMP kinase-related kinase), also known as NUA2 (NUAK family, SNF1-like kinase, 2), is a 628 amino acid protein that contains one protein kinase domain and belongs to the Ser/Thr protein kinase family. Using magnesium as a cofactor, SNARK catalyzes the ATP-dependent phosphorylation of target proteins and is involved in regulating cell tolerance to stress-induced glucose starvation. Additionally, SNARK is thought to induce cell-cell detachment and may protect cells from Fap-1-mediated apoptosis, possibly playing a role in the motility and invasiveness of tumor cells.

REFERENCES

1. Lefebvre, D.L., et al. 2001. Identification and characterization of a novel sucrose-non-fermenting protein kinase/AMP-activated protein kinase-related protein kinase, SNARK. *Biochem. J.* 355: 297-305.
2. Online Mendelian Inheritance in Man, OMIM™. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 608131. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
3. Suzuki, A., et al. 2003. Induction of cell-cell detachment during glucose starvation through F-Actin conversion by SNARK, the fourth member of the AMP-activated protein kinase catalytic subunit family. *Biochem. Biophys. Res. Commun.* 311: 156-161.
4. Suzuki, A., et al. 2003. Identification of a novel protein kinase mediating Akt survival signaling to the ATM protein. *J. Biol. Chem.* 278: 48-53.
5. Lizcano, J.M., et al. 2004. LKB1 is a master kinase that activates 13 kinases of the AMPK subfamily, including MARK/PAR-1. *EMBO J.* 23: 833-843.
6. Legembre, P., et al. 2004. Identification of SNF1/AMP kinase-related kinase as an NFκB-regulated anti-apoptotic kinase involved in CD95-induced motility and invasiveness. *J. Biol. Chem.* 279: 46742-46747.
7. Kusakai, G., et al. 2004. Strong association of ARK5 with tumor invasion and metastasis. *J. Exp. Clin. Cancer Res.* 23: 263-268.
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CHROMOSOMAL LOCATION

Genetic locus: Nuak2 (mouse) mapping to 1 E4.

PRODUCT

SNARK siRNA (m) is a target-specific 19-25 nt siRNA designed to knock down gene expression. Each vial contains 3.3 nmol of lyophilized siRNA, sufficient for a 10 μM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see SNARK shRNA Plasmid (m): sc-153652-SH and SNARK shRNA (m) Lentiviral Particles: sc-153652-V as alternate gene silencing 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 μl of the RNase-free water provided. Resuspension of the siRNA duplex in 330 μl of RNase-free water makes a 10 μM solution in a 10 μM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

SNARK siRNA (m) is recommended for the inhibition of SNARK 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 μM in 66 μl. 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

SNARK (C-12): sc-374348 is recommended as a control antibody for monitoring of SNARK 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 SNARK gene expression knockdown using RT-PCR Primer: SNARK (m)-PR: sc-153652-PR (20 μl). 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 for detailed protocols and support products.