

# SKIP siRNA (h): sc-106937

## BACKGROUND

The inositol polyphosphate 5-phosphatases selectively remove the phosphate from the 5-position of various phosphatidylinositols, which generate second messengers in response to extracellular signals. SKIP (skeletal muscle and kidney enriched inositol phosphatase) is a Type II 5-phosphatase that contains two highly conserved catalytic motifs. It is predominantly expressed in skeletal muscle, heart, brain, and kidney, but can also be detected in some tissues as a shorter protein, which is produced by alternative splicing. SKIP has a high affinity for phosphatidylinositol 4,5-bisphosphate as a substrate as well as inositol 1,4,5-trisphosphate, inositol 1,3,4,5-tetrakisphosphate, and phosphatidylinositol 3,4,5-trisphosphate. SKIP is localized in the cytoplasm and at ruffling membranes. Cells expressing SKIP display a loss of actin stress fibers where the protein was localized, suggesting that SKIP plays a negative role in regulating the actin cytoskeletal structure.

## REFERENCES

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2. Zhang, X., et al. 1998. Phosphatidylinositol signalling reactions. *Semin. Cell Dev. Biol.* 9: 153-160.
3. Erneux, C., et al. 1998. The diversity and possible functions of the inositol polyphosphate 5-phosphatases. *Biochim. Biophys. Acta* 1436: 185-199.
4. Mochizuki, Y., et al. 1999. Novel inositol polyphosphate 5-phosphatase localizes at membrane ruffles. *J. Biol. Chem.* 274: 36790-36795.
5. Ijuin, T., et al. 2000. Identification and characterization of a novel inositol polyphosphate 5-phosphatase. *J. Biol. Chem.* 275: 10870-10875.
6. Gurung, R., et al. 2003. Identification of a novel domain in two mammalian inositol-polyphosphate 5-phosphatases that mediates membrane ruffle localization. The inositol 5-phosphatase skip localizes to the endoplasmic reticulum and translocates to membrane ruffles following epidermal growth factor stimulation. *J. Biol. Chem.* 278: 11376-11385.

## CHROMOSOMAL LOCATION

Genetic locus: INPP5K (human) mapping to 17p13.3.

## PRODUCT

SKIP 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 SKIP shRNA Plasmid (h): sc-106937-SH and SKIP shRNA (h) Lentiviral Particles: sc-106937-V as alternate gene silencing products.

For independent verification of SKIP (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-106937A, sc-106937B and sc-106937C.

## PROTOCOLS

See our web site at [www.scbt.com](http://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  $\mu$ 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

SKIP siRNA (h) is recommended for the inhibition of SKIP 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

SKIP (B-6): sc-365362 is recommended as a control antibody for monitoring of SKIP 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™ Molecular Weight Standards: sc-2035, UltraCruz® 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® Mounting Medium: sc-24941 or UltraCruz® Hard-set Mounting Medium: sc-359850.

## RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor SKIP gene expression knockdown using RT-PCR Primer: SKIP (h)-PR: sc-106937-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.