

PIPK I α siRNA (h): sc-36232

BACKGROUND

Phosphatidylinositol-4-phosphate-5-kinase (PIPK) synthesizes phosphatidylinositol-4,5-bisphosphate, which regulates various processes including cell proliferation, survival, membrane trafficking and cytoskeletal organization. The PIPK family is divided into type I, type II and type III. Each type of the PIPK family phosphorylate distinct substrates and they contain an activation loop, which determines their enzymatic specificity and subcellular targeting. The phosphatidylinositol-4-phosphate-5-kinase type I consists of three members, PIPK I α , β and γ , which are characterized by phosphorylating PI4P on the 5-hydroxyl. PIPK I α (designated PIPK I β in mouse) is expressed in brain tissue. PIPK I β , designated PIPK I α in mouse, is also called STM7. PIPK I γ has two variants produced by alternative splicing which are expressed in lung, brain and kidneys.

REFERENCES

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2. Loijens, J.C., et al. 1996. Type I phosphatidylinositol-4-phosphate 5-kinases are distinct members of this novel lipid kinase family. *J. Biol. Chem.* 271: 32937-32943.
3. Tolia, K.F., et al. 1998. Type I phosphatidylinositol-4-phosphate 5-kinases synthesize the novel lipids phosphatidylinositol 3,5-bisphosphate and phosphatidylinositol 5-phosphate. *J. Biol. Chem.* 273: 18040-18046.
4. Rao, V.D., et al. 1998. Structure of Type II β phosphatidylinositol phosphate kinase: a protein kinase fold flattened for interfacial phosphorylation. *Cell* 94: 829-839.
5. Ishihara, H., et al. 1998. Type I phosphatidylinositol-4-phosphate 5-kinases. Cloning of the third isoform and deletion/substitution analysis of members of this novel lipid kinase family. *J. Biol. Chem.* 273: 8741-8748.
6. Kunz, J., et al. 2000. The activation loop of phosphatidylinositol phosphate kinases determines signaling specificity. *Mol. Cell* 5: 1-11.
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CHROMOSOMAL LOCATION

Genetic locus: PIP5K1A (human) mapping to 1q21.3.

PRODUCT

PIPK I α 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see PIPK I α shRNA Plasmid (h): sc-36232-SH and PIPK I α shRNA (h) Lentiviral Particles: sc-36232-V as alternate gene silencing products.

For independent verification of PIPK I α (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-36232A, sc-36232B and sc-36232C.

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

PIPK I α siRNA (h) is recommended for the inhibition of PIPK I α 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 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

PIPK I α (D-12): sc-377021 is recommended as a control antibody for monitoring of PIPK I α 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 PIPK I α gene expression knockdown using RT-PCR Primer: PIPK I α (h)-PR: sc-36232-PR (20 μ l, 600 bp). 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.