

IP3KB siRNA (h): sc-39066

BACKGROUND

Inositol 1,4,5-trisphosphate (Ins(1,4,5)P₃) regulates the level of calcium within the cell by releasing calcium from intracellular stores. Ins(1,4,5)P₃ is phosphorylated by inositol 1,4,5-trisphosphate 3-kinase (IP3K) to form inositol 1,3,4,5-tetrakisphosphate (Ins(1,4,5)P₄), which is thought to regulate the influx of calcium across the plasma membrane. IP3K exists as three isoforms, IP3KA, B, and C. IP3KA, the most highly characterized isoform, is expressed in rat brain and testis. IP3KB is expressed in various rat tissues such as lung, thymus, testis, brain, and heart. IP3K activity is stimulated in the presence of calmodulin via phosphorylation by cAMP-dependent protein kinase, protein kinase C, or calcium/calmodulin dependent protein kinase II and, subsequently, mediates the inositol phosphate signaling pathways.

REFERENCES

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2. Berridge, M.J. and Irvine, R.F. 1989. Inositol phosphates and cell signaling. *Nature* 341: 197-205.
3. Sim, S.S., et al. 1990. Regulation of D-myo-inositol 1,4,5-trisphosphate 3-kinase by cAMP-dependent protein kinase and protein kinase C. *J. Biol. Chem.* 265: 10367-10372.
4. Takazawa, K., et al. 1990. Cloning and expression in *Escherichia coli* of a rat brain cDNA encoding a Ca²⁺/calmodulin-sensitive inositol 1,4,5-trisphosphate 3-kinase. *Biochem. J.* 272: 107-112.
5. Irvine, R.F. 1991. Inositol tetrakisphosphate as a second messenger: confusions, contradictions, and a potential resolution. *Bioessays* 13: 419-427.
6. Vanweyenberg, V., et al. 1995. Tissue and cell-specific expression of Ins(1,4,5)P₃ 3-kinase isoenzymes. *Biochem. J.* 306: 429-435.
7. Woodring, P.J., et al. 1997. Expression, purification, and regulation of two isoforms of the inositol 1,4,5-trisphosphate 3-kinase. *J. Biol. Chem.* 272: 30447-30454.

CHROMOSOMAL LOCATION

Genetic locus: ITPKB (human) mapping to 1q42.12.

PRODUCT

IP3KB 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 IP3KB shRNA Plasmid (h): sc-39066-SH and IP3KB shRNA (h) Lentiviral Particles: sc-39066-V as alternate gene silencing products.

For independent verification of IP3KB (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-39066A, sc-39066B and sc-39066C.

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

IP3KB siRNA (h) is recommended for the inhibition of IP3KB 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

IP3KB (J-15): sc-100385 is recommended as a control antibody for monitoring of IP3KB 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 IP3KB gene expression knockdown using RT-PCR Primer: IP3KB (h)-PR: sc-39066-PR (20 μ 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.