

PKD2 siRNA (m): sc-76156

BACKGROUND

PKD2 (protein kinase D2), also known as PRKD2 or HSPC187, is a widely expressed protein belonging to the protein kinase D (PKD) family of serine/threonine kinases. In mammals, there are three members of the PKD family, namely PKC μ , PKD2 and PKC ν , and each contain a homologous catalytic domain but differ in their tissue expression and subcellular localization. PKD family members are activated by G protein-coupled receptors (GPCRs) and are known to participate in biological processes such as proliferation, apoptosis, migration, signal transduction and vesicle shedding. Shuttling between the nucleus and the cytoplasm, PKD2 contains one PH domain, one protein kinase domain and two phorbol-ester/DAG-type zinc fingers, and functions as a calcium-independent, phospholipid-dependent protein kinase. Upon activation of CCK-BR, PKD2 is phosphorylated by casein kinase I isoforms and subsequently accumulates in the nucleus. The result of the nuclear accumulation of PKD2 is the transcriptional activation of Nur77 and the nuclear exclusion of HDAC7. This suggests that PKD2 mediates CCK-BR-induced transcriptional activation.

REFERENCES

1. Sturany, S., et al. 2001. Molecular cloning and characterization of the human protein kinase D2. A novel member of the protein kinase D family of serine threonine kinases. *J. Biol. Chem.* 276: 3310-3318.
2. Online Mendelian Inheritance in Man, OMIM[™]. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 607074. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
3. Kovalevska, L.M., et al. 2006. Immunohistochemical studies of protein kinase D (PKD) 2 expression in malignant human lymphomas. *Exp. Oncol.* 28: 225-230.
4. Irie, A., et al. 2006. Protein kinase D2 contributes to either IL-2 promoter regulation or induction of cell death upon TCR stimulation depending on its activity in Jurkat cells. *Int. Immunol.* 18: 1737-1747.
5. von Blume, J., et al. 2007. Phosphorylation at Ser 244 by CK1 determines nuclear localization and substrate targeting of PKD2. *EMBO J.* 26: 4619-4633.

CHROMOSOMAL LOCATION

Genetic locus: Prkd2 (mouse) mapping to 7 A2.

PRODUCT

PKD2 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 PKD2 shRNA Plasmid (m): sc-76156-SH and PKD2 shRNA (m) Lentiviral Particles: sc-76156-V as alternate gene silencing products.

For independent verification of PKD2 (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-76156A, sc-76156B and sc-76156C.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

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

PKD2 siRNA (m) is recommended for the inhibition of PKD2 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

PKD2 (F-2): sc-374344 is recommended as a control antibody for monitoring of PKD2 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 PKD2 gene expression knockdown using RT-PCR Primer: PKD2 (m)-PR: sc-76156-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.