

DGK- κ siRNA (h): sc-91229

BACKGROUND

Diacylglycerol kinase plays an important role in signal transduction through regulating the balance between two signaling lipids, diacylglycerol (DAG) and phosphatidic acid (PA). DGK- κ (diacylglycerol kinase κ), also known as DGKK, 142 kDa diacylglycerol kinase or diglyceride kinase kappa, is a 1,271 amino acid protein belonging to the eukaryotic diacylglycerol kinase family. DGK- κ contains one DAGKc domain, one PH domain and two phorbol-ester/DAG-type zinc fingers. DGK- κ generates PA by phosphorylating DAG and is inhibited in response to hydrogen peroxide. A peripheral membrane protein, DGK- κ does not form homooligomers and is expressed in testis, with lower levels in placenta.

REFERENCES

1. Itani, S.I., et al. 2002. Lipid-induced Insulin resistance in human muscle is associated with changes in diacylglycerol, protein kinase C, and I κ B- α . *Diabetes* 51: 2005-2011.
2. Imai, S., et al. 2005. Identification and characterization of a novel human type II diacylglycerol kinase, DGK κ . *J. Biol. Chem.* 280: 39870-39881.
3. Nair, S., et al. 2006. Pharmacogenomics of phenolic antioxidant butylated hydroxyanisole (BHA) in the small intestine and liver of Nrf2 knockout and C57BL/6J mice. *Pharm. Res.* 23: 2621-2637.
4. Merida, I., et al. 2008. Diacylglycerol kinases: at the hub of cell signalling. *Biochem. J.* 409: 1-18.
5. Hozumi, Y. 2009. Morphological study of the diacylglycerol kinase family. *Kaibogaku Zasshi* 84: 121-122.
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7. van der Zanden, L.F., et al. 2011. Common variants in DGKK are strongly associated with risk of hypospadias. *Nat. Genet.* 43: 48-50.

CHROMOSOMAL LOCATION

Genetic locus: DGKK (human) mapping to Xp11.22.

PRODUCT

DGK- κ 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 DGK- κ shRNA Plasmid (h): sc-91229-SH and DGK- κ shRNA (h) Lentiviral Particles: sc-91229-V as alternate gene silencing products.

For independent verification of DGK- κ (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-91229A, sc-91229B and sc-91229C.

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

DGK- κ siRNA (h) is recommended for the inhibition of DGK- κ 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

DGK- κ (4G12): sc-517147 is recommended as a control antibody for monitoring of DGK- κ 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 DGK- κ gene expression knockdown using RT-PCR Primer: DGK- κ (h)-PR: sc-91229-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.