SANTA CRUZ BIOTECHNOLOGY, INC.

cGKIα/β siRNA (r): sc-270330

BACKGROUND

cGKII (cGMP-dependent protein kinase type II) is a major receptor of intracellular cGMP, and mediates a plethora of physiological responses. cGKII contains a conserved leucine zipper motif at the amino terminus. It is expressed in small intestine, colon, prostate, and human brain tissues, and the cGKII gene maps to chromosome 4q21.21. cGKII has been shown to regulate the ion transport system in the intestine. Myristoylation of the penultimate glycine in cGKII appears to be essential for directing cGKII to the membrane, since cGKII is devoid of any hydrophobic transmembrane domains. The translocation of cGKII from the cytosol to the membrane allows it to function properly in regulating intestinal ion transport. cGMP-dependent protein kinase 1 (cGKI) lowers the intracellular level of calcium and is therefore considered important for the relaxation of vascular smooth muscle. There are two isoforms of cGKI, α and β, which differ only in their N-terminal sequence.

REFERENCES

1. Gamm, D.M., et al. 1995. The type II isoform of cGMP-dependent protein kinase is dimeric and possesses regulatory and catalytic properties distinct from the type I isoforms. J. Biol. Chem. 270: 27380-27388.
2. Tamura, N., et al. 1996. cDNA cloning and gene expression of human type I α cGMP-dependent protein kinase. Hypertension 27: 552-5577.
3. Vaandrager, A.B. and de Jonge, H.R. 1996. Signalling by cGMP-dependent protein kinases. Mol. Cell. Biochem. 157: 23-30.
4. Orstavik, S., et al. 1997. Characterization of the human gene encoding the type I α and type I β cGMP-dependent protein kinase (PRKG1). Genomics 42: 311-318.

CHROMOSOMAL LOCATION

Genetic locus: Prkg1 (rat) mapping to 1q43.

PRODUCT

cGKIα/β siRNA (r) is a pool of 2 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see cGKIα/β shRNA Plasmid (r): sc-270330-SH and cGKIα/β shRNA (r) Lentiviral Particles: sc-270330-V as alternate gene silencing products.

For independent verification of cGKIα/β (r) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-270330A and sc-270330B.

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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

cGKIα/β siRNA (r) is recommended for the inhibition of cGKIα/β expression in rat 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 µM in 66 µl. 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

cGKIα/β (G-3): sc-271766 is recommended as a control antibody for monitoring of cGKIα/β 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 cGKIα/β gene expression knockdown using RT-PCR Primer: cGKIα/β (r)-PR: sc-270330-PR (20 µl, 402 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Piwkowska, A., et al. 2014. Insulin stimulates glucose transport via protein kinase G type I α-dependent pathway in podocytes. Biochem. Biophys. Res. Commun. 446: 328-334.
2. Rachubik, P., et al. 2020. The PKGIα/VASP pathway is involved in Insulin- and high glucose-dependent regulation of albumin permeability in cultured rat podocytes. J. Biochem. 168: 575-588.

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.