

CaMKI γ siRNA (h): sc-88278

BACKGROUND

The Ca²⁺/calmodulin-dependent protein kinases (CaMKs) comprise a structurally related subfamily of serine/threonine kinases. CaMKI γ (calcium/calmodulin-dependent protein kinase I γ), also known as VWS1 or CLICKIII, is a 476 amino acid protein that localizes to both the cytoplasm and to the membrane of the Golgi apparatus and contains one protein kinase domain. Expressed predominately in brain and present at lower levels in spleen, liver, kidney and skeletal muscle, CaMKI γ functions as a Ca²⁺/calmodulin-dependent protein kinase that uses ATP to catalyze the phosphorylation of target proteins, such as the transcription factor CREB-1. CaMKI γ exists as multiple alternatively spliced isoforms and is encoded by a gene which maps to human chromosome 1.

REFERENCES

1. Soderling, T.R. 1999. The Ca-calmodulin-dependent protein kinase cascade. *Trends Biochem. Sci.* 24: 232-236.
2. Schutte, B.C., et al. 2000. A preliminary gene map for the Van der Woude syndrome critical region derived from 900 kb of genomic sequence at 1q32-q41. *Genome Res.* 10: 81-94.
3. Hook, S.S., et al. 2001. Ca²⁺/CaM-dependent kinases: from activation to function. *Annu. Rev. Pharmacol. Toxicol.* 41: 471-505.
4. Takemoto-Kimura, S., et al. 2003. Molecular cloning and characterization of CLICK-III/CaMKI γ , a novel membrane-anchored neuronal Ca²⁺/calmodulin-dependent protein kinase (CaMK). *J. Biol. Chem.* 278: 18597-18605.
5. Nishimura, H., et al. 2003. Cloning, characterization and expression of two alternatively splicing isoforms of Ca²⁺/calmodulin-dependent protein kinase I γ in the rat brain. *J. Neurochem.* 85: 1216-1227.
6. Takemoto-Kimura, S., et al. 2007. Regulation of dendritogenesis via a lipid-raft-associated Ca²⁺/calmodulin-dependent protein kinase CLICKIII/CaMKI γ . *Neuron* 54: 755-770.

CHROMOSOMAL LOCATION

Genetic locus: CAMK1G (human) mapping to 1q32.2.

PRODUCT

CaMKI γ 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 CaMKI γ shRNA Plasmid (h): sc-88278-SH and CaMKI γ shRNA (h) Lentiviral Particles: sc-88278-V as alternate gene silencing products.

For independent verification of CaMKI γ (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-88278A, sc-88278B and sc-88278C.

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

CaMKI γ siRNA (h) is recommended for the inhibition of CaMKI γ 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

CaMKI γ (10G8): sc-134296 is recommended as a control antibody for monitoring of CaMKI γ 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 CaMKI γ gene expression knockdown using RT-PCR Primer: CaMKI γ (h)-PR: sc-88278-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.