

L-type Ca⁺⁺ CP β 4 siRNA (m): sc-62049

BACKGROUND

Voltage-dependent calcium channels are important for the release of neurotransmitters in neurons. L-type (long lasting current) voltage-dependent calcium channels are composed of four subunits: an α 1 subunit, a β subunit, a γ subunit, and an α 2 δ subunit. The β subunit is encoded by four genes, β 1- β 4, differing by about 20%. The various β subunits contribute to the diversity of calcium currents and are also involved in membrane trafficking of the α 1 subunit. L-type Ca⁺⁺ CP β 4 (calcium channel voltage-dependent subunit β 4), also known as CACNB4, belongs to the calcium channel β subunit family. It is the most highly expressed subunit in the cerebellum. L-type Ca⁺⁺ CP β 4 localizes to the cytoplasm and functions by regulating G protein inhibition, current amplitude and voltage dependence of activation and inactivation. A splice variant exists for L-type Ca⁺⁺ CP β 4 which enhances cellular excitability. Mutations in the gene encoding L-type Ca⁺⁺ CP β 4 are associated with idiopathic generalized epilepsy (IGE) and juvenile myoclonic epilepsy (JME).

REFERENCES

1. Walker, D., et al. 1998. A β 4 isoform-specific interaction site in the carboxyl-terminal region of the voltage-dependent Ca²⁺ channel α 1A subunit. *J. Biol. Chem.* 273: 2361-2367.
2. Escayg, A., et al. 2000. Coding and noncoding variation of the human calcium-channel β 4-subunit gene CACNB4 in patients with idiopathic generalized epilepsy and episodic ataxia. *Am. J. Hum. Genet.* 66: 1531-1539.
3. Pagani, R., et al. 2003. Differential expression of α 1 and β subunits of voltage dependent Ca²⁺ channel at the neuromuscular junction of normal and P/Q Ca²⁺ channel knockout mouse. *Neuroscience* 123: 75-85.
4. Takahashi, E., et al. 2005. Expression pattern of voltage-dependent calcium channel α 1 and β subunits in adrenal gland of N-type Ca²⁺ channel α 1B subunit gene-deficient mice. *Mol. Cell. Biochem.* 271: 91-99.
5. Suzuki, T., et al. 2006. Mutation analyses of genes on 6p12-p11 in patients with juvenile myoclonic epilepsy. *Neurosci. Lett.* 405: 126-131.
6. Ma, S., et al. 2006. Mutations in the GABRA1 and EFHC1 genes are rare in familial juvenile myoclonic epilepsy. *Epilepsy Res.* 71: 129-134.

CHROMOSOMAL LOCATION

Genetic locus: Cacnb4 (mouse) mapping to 2 C1.1.

PRODUCT

L-type Ca⁺⁺ CP β 4 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 L-type Ca⁺⁺ CP β 4 shRNA Plasmid (m): sc-62049-SH and L-type Ca⁺⁺ CP β 4 shRNA (m) Lentiviral Particles: sc-62049-V as alternate gene silencing products.

For independent verification of L-type Ca⁺⁺ CP β 4 (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-62049A, sc-62049B and sc-62049C.

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

L-type Ca⁺⁺ CP β 4 siRNA (m) is recommended for the inhibition of L-type Ca⁺⁺ CP β 4 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

L-type Ca⁺⁺ CP β 4 (H-7): sc-376432 is recommended as a control antibody for monitoring of L-type Ca⁺⁺ CP β 4 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 L-type Ca⁺⁺ CP β 4 gene expression knockdown using RT-PCR Primer: L-type Ca⁺⁺ CP β 4 (m)-PR: sc-62049-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.

PROTOCOLS

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