

L-type Ca⁺⁺ CP β3 siRNA (h): sc-95841

BACKGROUND

Voltage-dependent calcium channels are essential for the release of neurotransmitters. L-type (long lasting current) voltage-dependent calcium channels are composed of four subunits: an α1 subunit, a β subunit, a γ subunit and an α2δ subunit. The β subunit is encoded by four genes, designated β1-β4, all of which contribute to the diversity of calcium currents and are involved in membrane trafficking of the α1 subunit. L-type Ca⁺⁺ CP β3, also known as CACNB3 (calcium channel voltage-dependent subunit β 3), CACNLB3 or CAB3, is a 484 amino acid protein that contains one SH3 domain and is expressed in ovary, brain and smooth muscle. Functioning as one of the four components of the β subunit, L-type Ca⁺⁺ CP β3 increases the peak calcium current in voltage-dependent calcium channels, thereby shifting the voltage dependencies of activation and inactivation and controlling G protein inhibition and α1 membrane targeting. Two isoforms of L-type Ca⁺⁺ CP β3 exist due to alternative splicing events.

REFERENCES

1. Collin, T., et al. 1994. Cloning, chromosomal location and functional expression of the human voltage-dependent calcium-channel β3 subunit. *Eur. J. Biochem.* 220: 257-262.
2. Yamada, Y., et al. 1995. The structures of the human calcium channel α1 subunit (CACNL1A2) and β subunit (CACNLB3) genes. *Genomics* 27: 312-319.
3. Murakami, M., et al. 1996. Gene structure of the murine calcium channel β3 subunit, cDNA and characterization of alternative splicing and transcription products. *Eur. J. Biochem.* 236: 138-143.
4. Murakami, M., et al. 2002. Pain perception in mice lacking the β3 subunit of voltage-activated calcium channels. *J. Biol. Chem.* 277: 40342-40351.
5. Colecraft, H.M., et al. 2002. Novel functional properties of Ca²⁺ channel β subunits revealed by their expression in adult rat heart cells. *J. Physiol.* 541: 435-452.
6. Qin, N., et al. 2002. Molecular cloning and characterization of the human voltage-gated calcium channel α(2)δ-4 subunit. *Mol. Pharmacol.* 62: 485-496.

CHROMOSOMAL LOCATION

Genetic locus: CACNB3 (human) mapping to 12q13.12.

PRODUCT

L-type Ca⁺⁺ CP β3 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 μM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see L-type Ca⁺⁺ CP β3 shRNA Plasmid (h): sc-95841-SH and L-type Ca⁺⁺ CP β3 shRNA (h) Lentiviral Particles: sc-95841-V as alternate gene silencing products.

For independent verification of L-type Ca⁺⁺ CP β3 (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-95841A, sc-95841B and sc-95841C.

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

L-type Ca⁺⁺ CP β3 siRNA (h) is recommended for the inhibition of L-type Ca⁺⁺ CP β3 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 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

L-type Ca⁺⁺ CP β3 (7D1): sc-130560 is recommended as a control antibody for monitoring of L-type Ca⁺⁺ CP β3 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 L-type Ca⁺⁺ CP β3 gene expression knockdown using RT-PCR Primer: L-type Ca⁺⁺ CP β3 (h)-PR: sc-95841-PR (20 μl, 557 bp). 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.