SANTA CRUZ BIOTECHNOLOGY, INC.

N-type Ca^{++} CP α1B siRNA (h): sc-42698

BACKGROUND

N-type calcium channels are localized in high density presynaptic nerve terminals and are crucial elements in neuronal excitation-secretion coupling. Peripherally distributed N-type Ca^{++} channel plays a key role in cardiovascular regulation through autonomic nervous system. The high-voltage activated Ca^{2+} channels that have been characterized biochemically are complexes of a pore-forming α-1 subunit; a transmembrane, disulfide-linked complex of α-2 and δ subunits; an intracellular β subunit; and in some cases, a transmembrane γ subunit. The α-1 subunit conducts N-type Ca^{2+} currents, which initiate rapid synaptic transmission. In addition to mediating Ca^{2+} entry to initiate transmitter release, N-type Ca^{2+} channels are thought to interact directly with proteins of the synaptic vesicle docking and fusion machinery. The synaptic protein interaction sites in the intracellular loop II-III of subunit α-1B of N-type Ca^{2+} channels bind to syntaxin, SNAP-25 and synaptotagmin.

REFERENCES

1. Catterall, W.A. 1999. Interactions of presynaptic Ca^{2+} channels and snare proteins in neurotransmitter release. Ann. N.Y. Acad. Sci. 868: 144-159.
2. Fossier, P., et al. 1999. Calcium transients and neurotransmitter release at an identified synapse. Trends Neurosci. 4: 161-166.
3. Uneyama, H., et al. 1999. Pharmacology of N-type Ca^{2+} channels distributed in cardiovascular system. Int. J. Mol. Med. 5: 455-466.
4. Catterall, W.A. 2000. Structure and regulation of voltage-gated Ca^{2+} channels. Annu. Rev. Cell Dev. Biol. 16: 521-555.

CHROMOSOMAL LOCATION

Genetic locus: CACNA1B (human) mapping to 9q34.3.

PRODUCT

N-type Ca^{++} CP α1B 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 N-type Ca^{++} CP α1B shRNA Plasmid (h): sc-42698-SH and N-type Ca^{++} CP α1B shRNA (h) Lentiviral Particles: sc-42698-V as alternate gene silencing products.

For independent verification of N-type Ca^{++} CP α1B (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-42698A, sc-42698B and sc-42698C.

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

N-type Ca^{++} CP α1B siRNA (h) is recommended for the inhibition of N-type Ca^{++} CP α1B 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

N-type Ca^{++} CP α1B (A-2): sc-377489 is recommended as a control antibody for monitoring of N-type Ca^{++} CP α1B 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 N-type Ca^{++} CP α1B gene expression knockdown using RT-PCR Primer: N-type Ca^{++} CP α1B (h)-PR: sc-42698-PR (20 µl). 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.