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Cacna2d2 shRNA (h) Lentiviral Particles: sc-45522-V

BACKGROUND

CACNA2D2 is a gene coding for the protein calcium channel, voltage-dependent $\alpha_2\delta$ -2 (Cacna2d2), a regulatory subunit of the voltage dependent calcium channels. The protein interacts with α -1, β and γ subunits in a 1:1:1:1 ratio to form a channel mediating calcium influx. Protein expression occurs in the brain, heart, and other tissues, and is involved in central nervous system function. Disruptions of the CACNA2D2 gene may be involved in cerebellar ataxias and epileptic episodes in humans. The gene is localized to the tumor suppressor region of chromosome 3p21.3 in humans. Expression deficiency occurs in lung, breast and other cancers in humans. Part of a family of $\alpha_2\delta$ subunits involved in voltage-dependent calcium influx, Cacna2d2 shares 56% amino acid homology with the $\alpha_2\delta$ -1 subunit, although they have different patterns of tissue expression.

REFERENCES

1. Alden, K.J., et al. 2001. Differential effect of gabapentin on neuronal and muscle calcium currents. *J. Pharmacol. Exp. Ther.* 297: 727-735.
2. Barclay, J., et al. 2001. Ducky mouse phenotype of epilepsy and ataxia is associated with mutations in the Cacna2d2 gene and decreased calcium channel current in cerebellar Purkinje cells. *J. Neurosci.* 21: 6095-6104.
3. Brodbeck, J., et al. 2002. The ducky mutation in Cacna2d2 results in altered Purkinje cell morphology and is associated with the expression of a truncated $\alpha_2\delta$ -2 protein with abnormal function. *J. Biol. Chem.* 277: 7684-7693.
4. Ji, L., et al. 2002. Expression of several genes in the human chromosome 3p21.3 homozygous deletion region by an adenovirus vector results in tumor suppressor activities *in vitro* and *in vivo*. *Cancer Res.* 62: 2715-2720.
5. Braga, E.A., et al. 2003. New tumor suppressor genes in hot spots of human chromosome 3: new methods of identification. *Mol. Biol.* 37: 194-211.

CHROMOSOMAL LOCATION

Genetic locus: CACNA2D2 (human) mapping to 3p21.31.

PRODUCT

Cacna2d2 shRNA (h) Lentiviral Particles is a pool of concentrated, transduction-ready viral particles containing 3 target-specific constructs that encode 19-25 nt (plus hairpin) shRNA designed to knock down gene expression. Each vial contains 200 μ l frozen stock containing 1.0×10^6 infectious units of virus (IFU) in Dulbecco's Modified Eagle's Medium with 25 mM HEPES pH 7.3. Suitable for 10-20 transductions. Also see Cacna2d2 siRNA (h): sc-45522 and Cacna2d2 shRNA Plasmid (h): sc-45522-SH as alternate gene silencing products.

STORAGE

Store lentiviral particles at -80° C. Stable for at least one year from the date of shipment. Once thawed, particles can be stored at 4° C for up to one week. Avoid repeated freeze thaw cycles.

PROTOCOLS

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

APPLICATIONS

Cacna2d2 shRNA (h) Lentiviral Particles is recommended for the inhibition of Cacna2d2 expression in human cells.

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 μ l frozen viral stock containing 1.0×10^6 infectious units of virus (IFU); contains an shRNA construct encoding a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA.

GENE EXPRESSION MONITORING

Cacna2d2 (G-5): sc-365911 is recommended as a control antibody for monitoring of Cacna2d2 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 (secondary) reagents are recommended: 1) Western Blotting: use goat anti-mouse IgG-HRP: sc-2005 (dilution range: 1:2000-1:32,000) or Cruz Marker™ compatible goat anti-mouse IgG-HRP: sc-2031 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use goat anti-mouse IgG-FITC: sc-2010 (dilution range: 1:100-1:400) or goat anti-mouse IgG-TR: sc-2781 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Cacna2d2 gene expression knockdown using RT-PCR Primer: Cacna2d2 (h)-PR: sc-45522-PR (20 μ l). Annealing temperature for the primers should be 55 - 60° C and the extension temperature should be 68 - 72° C.

BIOSAFETY

Lentiviral particles can be employed in standard Biosafety Level 2 tissue culture facilities (and should be treated with the same level of caution as with any other potentially infectious reagent). Lentiviral particles are replication-incompetent and are designed to self-inactivate after transduction and integration of shRNA constructs into genomic DNA of target cells.

RESEARCH USE

The purchase of this product conveys to the buyer the nontransferable right to use the purchased amount of the product and all replicates and derivatives for research purposes conducted by the buyer in his laboratory only (whether the buyer is an academic or for-profit entity). The buyer cannot sell or otherwise transfer (a) this product (b) its components or (c) materials made using this product or its components to a third party, or otherwise use this product or its components or materials made using this product or its components for Commercial Purposes.