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CRMP-2 shRNA (h) Lentiviral Particles: sc-44485-V

BACKGROUND

Collapsin response mediator proteins (CRMPs), including CRMP-1 (DRP-1), CRMP-2 (DRP-2 or TOAD64), CRMP-3 (DRP-4), CRMP-4 (DRP-3) and CRMP-5 (DRP-5), mediate signal transduction after exposure of neural cells to the axon guidance molecule Semaphorin 3A/collapsin. CRMPs are present in the developing cerebral cortex and neocortical neurons and are responsive to Semaphorin 3A. In the adult brain, the expression of CRMPs is dramatically downregulated. However, they remain expressed in structures that retain their capacity for differentiation and plasticity. CRMP-2 is involved in axonal growth and guidance. The human CRMP-2 gene is located on 8p21.2, a chromosomal region that has been previously shown to have a significant linkage to schizophrenia and to several deficit symptoms of schizophrenia.

REFERENCES

1. Kitamura, K., et al. 1999. Characterization of the human dihydropyrimidinase-related protein 2 (DRP-2) gene. *DNA Res.* 6: 291-297.
2. Gu, Y., et al. 2000. Neurofibrillary tangle-associated collapsin response mediator protein-2 (CRMP-2) is highly phosphorylated on Thr 509, Ser 518 and Ser 522. *Biochemistry* 6: 4267-4275.
3. Nakata, K., 2003. The human dihydropyrimidinase-related protein 2 gene on chromosome 8p21 is associated with paranoid-type schizophrenia. *Biol. Psychiatry* 53: 571-576.
4. Rosslenbroich, V., et al. 2003. Subcellular localization of collapsin response mediator proteins to lipid rafts. *Biochem. Biophys. Res. Commun.* 305: 392-399.
5. Quach, TT., et al. 2004. Involvement of collapsin response mediator proteins in the neurite extension induced by neurotrophins in dorsal root ganglion neurons. *Mol. Cell. Neurosci.* 25: 433-443.

CHROMOSOMAL LOCATION

Genetic locus: DPYSL2 (human) mapping to 8p21.2.

PRODUCT

CRMP-2 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 CRMP-2 siRNA (h): sc-44485 and CRMP-2 shRNA Plasmid (h): sc-44485-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.

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.

APPLICATIONS

CRMP-2 shRNA (h) Lentiviral Particles is recommended for the inhibition of CRMP-2 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

CRMP-2 (E-9): sc-376739 is recommended as a control antibody for monitoring of CRMP-2 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 CRMP-2 gene expression knockdown using RT-PCR Primer: CRMP-2 (h)-PR: sc-44485-PR (20 μ l, 579 bp). Annealing temperature for the primers should be $55-60^\circ$ C and the extension temperature should be $68-72^\circ$ 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.