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Coronin 1B shRNA (h) Lentiviral Particles: sc-44695-V

BACKGROUND

Coronins are a family of WD repeat-containing, actin-binding proteins that localize to submembrane areas and regulate cell motility and cytoskeletal rearrangement. Coronin 1A (CORO1A, CLIPINA, CLABP, TACO, p57) can form coiled coil-mediated homotrimeric complexes that influence early phagosome formation. PKC-dependent phosphorylation of Coronin 1B (CORO1B) at Serine 2 regulates leading edge dynamics and cell motility in fibroblasts through interactions with Arp2/3 complex. Coronin 1C (CORO1C, Coronin 3, HCRN4) is abundant in differentiating Neuro-2a cells, PC-12 cells and primary oligodendrocytes, where it is thought to influence neuron morphogenesis and migration. Coronin 2A (CORO2A, CLIPINB, IR10, WDR2) is a component of the approximately 1.5-2 megadalton N-CoR (nuclear receptor corepressor) complex of 10-12 proteins, which recruits HDACs to generate repressive chromatin. Coronin 7 (CORO7, CRN7) localizes to the Golgi membrane and influences the organization of intracellular membrane compartments and vesicular trafficking. Coronin 2B (CORO2B, CLIPINC) and Coronin 6 (CORO6) are similar to other members of this family, since they possess a conserved basic N-terminal motif and 3-10 WD repeats clustered in one to two core domains.

REFERENCES

- Mishima, M., et al. 1999. Coronin localizes to leading edges and is involved in cell spreading and lamellipodium extension in vertebrate cells. *J. Cell Sci.* 112: 2833-2842.
- Spoerl, Z., et al. 2002. Oligomerization, F-actin interaction, and membrane association of the ubiquitous mammalian coronin 3 are mediated by its carboxyl terminus. *J. Biol. Chem.* 277: 48858-48867.
- Yoon, H.G., et al. 2003. Purification and functional characterization of the human N-CoR complex: the roles of HDAC3, TBL1 and TBLR1. *EMBO J.* 22: 1336-1346.

CHROMOSOMAL LOCATION

Genetic locus: CORO1B (human) mapping to 11q13.2.

PRODUCT

Coronin 1B 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 Coronin 1B siRNA (h): sc-44695 and Coronin 1B shRNA Plasmid (h): sc-44695-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

Coronin 1B shRNA (h) Lentiviral Particles is recommended for the inhibition of Coronin 1B 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

Coronin 1B (E-3): sc-271375 is recommended as a control antibody for monitoring of Coronin 1B 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 IgM-HRP: sc-2064 (dilution range: 1:500-1:5,000), TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use goat anti-mouse IgM-FITC: sc-2082 (dilution range: 1:100-1:400) or goat anti-mouse IgM-TR: sc-2983 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Coronin 1B gene expression knockdown using RT-PCR Primer: Coronin 1B (h)-PR: sc-44695-PR (20 μ l, 443 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.

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.