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ARMS shRNA (h) Lentiviral Particles: sc-44511-V

BACKGROUND

Ankyrin repeat-rich membrane-spanning protein (ARMS), also designated kinase D-interacting substance 220 or Kidins220, is a highly conserved, 1,715 amino acid protein containing multiple domains, including 4 putative transmembrane domains and several ankyrin repeats. ARMS is expressed in regions rich in neurotrophin (Trk) and ephrin (Eph) receptors, such as the brain and neuroendocrine cells (where it concentrates at the tip of neurites) and in plastic areas of the adult brain. It is also detected in peripheral blood immature dendritic cells and PC12 cells. ARMS functions as a substrate for protein kinase D and is a downstream target for both Trk and Eph receptors. It is a highly conserved protein, which suggests it has an evolutionary conserved role. The gene encoding for the protein maps to chromosome 2p25.1.

REFERENCES

- Iglesias, T., et al. 2000. Identification and cloning of Kidins220, a novel neuronal substrate of protein kinase D. *J. Biol. Chem.* 275: 40048-40056.
- Kong, H., et al. 2001. An evolutionarily conserved transmembrane protein that is a novel downstream target of neurotrophin and ephrin receptors. *J. Neurosci.* 21: 176-85.
- Arevalo, J.C., et al. 2004. A unique pathway for sustained neurotrophin signaling through an ankyrin-rich membrane-spanning protein. *EMBO. J.* 23: 2358-2368.
- Riol-Blanco, L., et al. 2004. The neuronal protein Kidins220 localizes in a raft compartment at the leading edge of motile immature dendritic cells. *Eur. J. Immunol.* 34: 108-118.
- Luo, S., et al. 2005. α -Syntrophin regulates ARMS localization at the neuromuscular junction and enhances EphA4 signaling in an ARMS-dependent manner. *J. Cell Biol.* 169: 813-824.

CHROMOSOMAL LOCATION

Genetic locus: KIDINS220 (human) mapping to 2p25.1.

PRODUCT

ARMS 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 ARMS siRNA (h): sc-44511 and ARMS shRNA Plasmid (h): sc-44511-SH as alternate gene silencing products.

APPLICATIONS

ARMS shRNA (h) Lentiviral Particles is recommended for the inhibition of ARMS 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

ARMS (P-20): sc-324828 is recommended as a control antibody for monitoring of ARMS 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 donkey anti-goat IgG-HRP: sc-2020 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible donkey anti-goat IgG-HRP: sc-2033 (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 donkey anti-goat IgG-FITC: sc-2024 (dilution range: 1:100-1:400) or donkey anti-goat IgG-TR: sc-2783 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor ARMS gene expression knockdown using RT-PCR Primer: ARMS (h)-PR: sc-44511-PR (20 μ l, 595 bp). 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.

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.

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.

PROTOCOLS

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