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KIF13A shRNA (h) Lentiviral Particles: sc-43380-V

BACKGROUND

The kinesins constitute a large family of microtubule-dependent motor proteins, which are responsible for the distribution of numerous organelles, vesicles and macromolecular complexes throughout the cell. Kinesins also play crucial roles in cell division, intracellular transport and membrane trafficking events including endocytosis and transcytosis. KIF13A, a novel plus end-directed microtubule-dependent motor protein, belongs to the unc-104/KIF1A kinesin subfamily and represents the orthologue of *Drosophila* kinesin-73. KIF13A has several alternative transcripts, which are differentially expressed in human tissues. KIF13A associates with β 1-Adaptin, a subunit of the AP-1 adaptor complex. Transmembrane receptors and some membrane-bound proteins are postulated to bind KIFs to cargo vesicles. KIF13A associates with cargo vesicles that contain AP-1 and mannose-6-phosphate receptor (M6PR). KIF13A transports M6PR-containing vesicles and targets M6PR from the *trans*-Golgi network to the plasma membrane via a direct interaction with the AP-1 adaptor complex. Overexpression of KIF13A results in mislocalization of AP-1 and M6PR, and functional blocking of KIF13A reduces M6PR cell surface expression. KIF13A is also found to have significant linkage to schizophrenia.

REFERENCES

1. Hamm-Alvarez, S.F. 1998. Molecular motors and their role in membrane traffic. *Adv. Drug Deliv. Rev.* 29: 229-242.
2. Cole, D.G. 1999. Kinesin-II, the heteromeric kinesin. *Cell. Mol. Life Sci.* 56: 217-226.
3. Nakagawa, T., et al. 2000. A novel motor, KIF13A, transports mannose-6-phosphate receptor to plasma membrane through direct interaction with AP-1 complex. *Cell* 103: 569-581.
4. Yang, Z., et al. 2001. Molecular cloning and functional analysis of mouse C-terminal kinesin motor KIFC3. *Mol. Cell. Biol.* 21: 765-770.

CHROMOSOMAL LOCATION

Genetic locus: KIF13A (human) mapping to 6p22.3.

PRODUCT

KIF13A 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 KIF13A siRNA (h): sc-43380 and KIF13A shRNA Plasmid (h): sc-43380-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

KIF13A shRNA (h) Lentiviral Particles is recommended for the inhibition of KIF13A 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

KIF13A (C-20): sc-16789 is recommended as a control antibody for monitoring of KIF13A 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 KIF13A gene expression knockdown using RT-PCR Primer: KIF13A (h)-PR: sc-43380-PR (20 μ l, 587 bp). Annealing temperature for the primers should be $55-60^\circ\text{C}$ and the extension temperature should be $68-72^\circ\text{C}$.

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.