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KLC1 shRNA (h) Lentiviral Particles: sc-43880-V

BACKGROUND

The kinesin family of motor proteins comprise at least two forms of conventional kinesin. They are encoded by different genes and designated as ubiquitous kinesin, which is expressed in all cells and tissues, and neuronal kinesin, which is expressed exclusively in neuronal cells. Conventional kinesin, kinesin-I, is a heterotetramer of two kinesin heavy chain subunits and two kinesin light chain subunits. While the kinesin heavy chain contains motor activity, evidence suggests that the kinesin light chain is involved in either modulation of kinesin heavy chain activity or in cargo binding. The motor protein kinesin is a heterotetramer composed of two heavy chains and two light chains. Kinesin motor activity is dependent on the presence of ATP and microtubules.

REFERENCES

1. Vignali, G., et al. 1997. Expression of neuronal kinesin heavy chain is developmentally regulated in the central nervous system of the rat. *J. Neurochem.* 69: 1840-1849.
2. Diefenbach, R.J., et al. 1998. The C-terminal region of the stalk domain of ubiquitous human kinesin heavy chain contains the binding site for kinesin light chain. *Biochemistry* 37: 16663-16670.
3. Rahman, A., et al. 1998. Two kinesin light chain genes in mice. Identification and characterization of the encoded proteins. *J. Biol. Chem.* 273: 15395-15403.
4. Rahman, A., et al. 1999. Defective kinesin heavy chain behavior in mouse kinesin light chain mutants. *J. Cell Biol.* 146: 1277-1288.
5. Vancoillie, G., et al. 2000. Kinesin and kinectin can associate with the melanosomal surface and form a link with microtubules in normal human melanocytes. *J. Invest. Dermatol.* 114: 421-429.

CHROMOSOMAL LOCATION

Genetic locus: KLC1 (human) mapping to 14q32.33.

PRODUCT

KLC1 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 KLC1 siRNA (h): sc-43880 and KLC1 shRNA Plasmid (h): sc-43880-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

KLC1 shRNA (h) Lentiviral Particles is recommended for the inhibition of KLC1 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

KLC1 (L-15): sc-13361 is recommended as a control antibody for monitoring of KLC1 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 KLC1 gene expression knockdown using RT-PCR Primer: KLC1 (h)-PR: sc-43880-PR (20 μ l, 414 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.