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# KIF3B shRNA (m) Lentiviral Particles: sc-43377-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. Individual kinesin members play crucial roles in cell division, intracellular transport, and membrane trafficking events including endocytosis and transcytosis. Members of the heterotrimeric kinesin II family of microtubule associated motors generally contain two different motor subunits from the KIF3 family, which includes KIF3A, B and C. KIF3 isoforms mediate anterograde transport of membrane bound organelles in neurons and melanosomes, transport between the endoplasmic reticulum and the Golgi, and transport of protein complexes within cilia and flagella required for their morphogenesis. The human KIF3B gene maps to chromosome 20 and encodes a 747-amino acid protein that is ubiquitously expressed. The KIF3B protein is essential for the left-right determination through a mechanism that produces a putative morphogen gradient along the left-right axis in the node.

## REFERENCES

1. Yamazaki, H., et al. 1995. KIF3A/B: a heterodimeric kinesin superfamily protein that works as a microtubule plus end-directed motor for membrane organelle transport. *J. Cell Biol.* 130: 1387-1399.
2. Hamm-Alvarez, S.F. 1998 Molecular motors and their role in membrane traffic. *Adv. Drug Deliv. Rev.* 29: 229-242.
3. Nonaka, S., et al. 1998. Randomization of left-right asymmetry due to loss of nodal cilia generating leftward flow of extraembryonic fluid in mice lacking KIF3B motor protein. *Cell* 95: 829-837.
4. Cole, D.G. 1999 Kinesin-II, the heteromeric kinesin. *Cell. Mol. Life Sci.* 56: 217-226.
5. Hirokawa, N. 2000. Stirring up development with the heterotrimeric kinesin KIF3. *Traffic* 1: 29-34.
6. Yang, Z., et al. 2001 Molecular cloning and functional analysis of mouse C-terminal kinesin motor KifC3. *Mol. Cell. Biol.* 21: 75-70.

## CHROMOSOMAL LOCATION

Genetic locus: Kif3b (mouse) mapping to 2 H1.

## PRODUCT

KIF3B shRNA (m) 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 µl frozen stock containing  $1.0 \times 10^6$  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 KIF3B siRNA (m): sc-43377 and KIF3B shRNA Plasmid (m): sc-43377-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.

## APPLICATIONS

KIF3B shRNA (m) Lentiviral Particles is recommended for the inhibition of KIF3B expression in mouse cells.

## SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 µl frozen viral stock containing  $1.0 \times 10^6$  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

KIF3B (H-60): sc-50456 is recommended as a control antibody for monitoring of KIF3B 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-rabbit IgG-HRP: sc-2004 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible goat anti-rabbit IgG-HRP: sc-2030 (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-rabbit IgG-FITC: sc-2012 (dilution range: 1:100-1:400) or goat anti-rabbit IgG-TR: sc-2780 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

## RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor KIF3B gene expression knockdown using RT-PCR Primer: KIF3B (m)-PR: sc-43377-PR (20 µl). 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.

## 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

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