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CLASP1 shRNA (h) Lentiviral Particles: sc-44348-V

BACKGROUND

Members of the CLASP family, including CLASP1, are mammalian microtubule plus-end binding proteins that interact with CLIPs in order to stabilize the microtubule structures in transfected cells. CLASP1 localizes near the distal end of growing spindle microtubules during mitosis and is a component of the outer corona region of kinetochores. CLASP proteins stabilize microtubules by promoting pauses and restricting MT growth. Defects in CLASP1 cause collapse of the spindle, attachment of kinetochores to short microtubules, and other abnormal mitotic behaviors.

REFERENCES

1. Lemos, C.L., et al. 2000. Mast, a conserved microtubule-associated protein required for bipolar mitotic spindle organization. *EMBO J.* 19: 3668-3682.
2. Akhmanova, A., et al. 2001. Clasps are CLIP-115 and -170 associating proteins involved in the regional regulation of microtubule dynamics in motile fibroblasts. *Cell* 104: 923-935.
3. Maiato, H., et al. 2003. Human CLASP1 is an outer kinetochore component that regulates spindle microtubule dynamics. *Cell* 113: 891-904.
4. Maiato, H., et al. 2003. How do kinetochores CLASP dynamic microtubules? *Cell Cycle* 2: 511-514
5. Mimori-Kiyosue, Y., et al. 2005. CLASP1 and CLASP2 bind to EB1 and regulate microtubule plus-end dynamics at the cell cortex. *J. Cell Biol.* 168: 141-153.
6. Venables, J.P., et al. 2005. Up-regulation of the ubiquitous alternative splicing factor Tra2 β causes inclusion of a germ cell-specific exon. *Hum. Mol. Genet.* 14: 2289-2303.

CHROMOSOMAL LOCATION

Genetic locus: CLASP1 (human) mapping to 2q14.2.

PRODUCT

CLASP1 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 CLASP1 siRNA (h): sc-44348 and CLASP1 shRNA Plasmid (h): sc-44348-SH as alternate gene silencing products.

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.

APPLICATIONS

CLASP1 shRNA (h) Lentiviral Particles is recommended for the inhibition of CLASP1 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

CLASP1 (D-8): sc-390159 is recommended as a control antibody for monitoring of CLASP1 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 IgG-HRP: sc-2005 (dilution range: 1:2000-1:32,000) or Cruz Marker™ compatible goat anti-mouse IgG-HRP: sc-2031 (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-mouse IgG-FITC: sc-2010 (dilution range: 1:100-1:400) or goat anti-mouse IgG-TR: sc-2781 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor CLASP1 gene expression knockdown using RT-PCR Primer: CLASP1 (h)-PR: sc-44348-PR (20 μ l). 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.

PROTOCOLS

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