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# MARK3 shRNA (h) Lentiviral Particles: sc-43649-V

## BACKGROUND

MARK3 (MAP/microtubule affinity-regulating kinase 3), also known as C-TAK1 (Cdc25C-associated protein kinase 1), EMK-2 (ELKL motif kinase 2), protein kinase STK10 or serine/threonine-protein kinase p78, is a 753 amino acid peripheral membrane protein that phosphorylates Cdc25C on Serine 216 and is ubiquitously expressed in various human tissue and cell lines. Existing as six alternatively spliced isoforms, MARK3 belongs to the Ser/Thr protein kinase family and the protein kinase superfamily. MARK3 has been suggested to mediate the binding of the 14-3-3 proteins through its kinase activity and acts as a negative regulator of mitosis. The gene encoding MARK3 maps to human chromosome 14q32.32 and mouse chromosome 12 F1.

## REFERENCES

- Ogg, S., et al. 1994. Purification of a serine kinase that associates with and phosphorylates human Cdc25C on serine 216. *J. Biol. Chem.* 269: 30461-30469.
- Sanchez, Y., et al. 1997. Conservation of the Chk1 checkpoint pathway in mammals: linkage of DNA damage to Cdk regulation through Cdc25. *Science* 277: 1497-1501.
- Peng, C.Y., et al. 1997. Mitotic and G<sub>2</sub> checkpoint control: regulation of 14-3-3 protein binding by phosphorylation of Cdc25C on serine-216. *Science* 277: 1501-1505.
- Peng, C.Y., et al. 1998. C-TAK1 protein kinase phosphorylates human Cdc25C on serine 216 and promotes 14-3-3 protein binding. *Cell Growth Differ.* 9: 197-208.
- Dalal, S.N., et al. 1999. Cytoplasmic localization of human Cdc25C during interphase requires an intact 14-3-3 binding site. *Mol. Cell. Biol.* 19: 4465-4479.

## CHROMOSOMAL LOCATION

Genetic locus: MARK3 (human) mapping to 14q32.32.

## PRODUCT

MARK3 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 µl frozen stock containing 1.0 x 10<sup>6</sup> 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 MARK3 siRNA (h): sc-43649 and MARK3 shRNA Plasmid (h): sc-43649-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](http://www.scbt.com) or our catalog for detailed protocols and support products.

## APPLICATIONS

MARK3 shRNA (h) Lentiviral Particles is recommended for the inhibition of MARK3 expression in human cells.

## SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 µl frozen viral stock containing 1.0 x 10<sup>6</sup> 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

MARK3 (G-11): sc-365461 is recommended as a control antibody for monitoring of MARK3 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 MARK3 gene expression knockdown using RT-PCR Primer: MARK3 (h)-PR: sc-43649-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.