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MARK2 shRNA (m) Lentiviral Particles: sc-45794-V

BACKGROUND

Microtubule affinity-regulating kinase 2 (MARK2), also known as EMK1 (ELKL motif kinase 1) or Par1b, is a 788 amino acid protein that is a member of the protein kinase superfamily, MARK subfamily. Highly expressed in heart, brain, skeletal muscle and pancreas, MARK2 is essential for the asymmetric development of membrane domains around polarized epithelial cells. Activation of MARK2 by phosphorylation on Thr 208 allows the protein to modulate the building of a columnar versus a hepatic epithelial cell. MARK2 contains one KA1 (kinase-associated) domain, one protein kinase domain and one UBA domain. MARK2 is expressed as 14 isoforms produced by alternative splicing events. Some of these isoforms may function in graft rejection.

REFERENCES

- Marx, A., et al. 2006. Structural variations in the catalytic and ubiquitin-associated domains of microtubule-associated protein/microtubule affinity regulating kinase (MARK) 1 and MARK2. *J. Biol. Chem.* 281: 27586-27599.
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- Wang, J.W., et al. 2007. Activation of PAR-1 kinase and stimulation of tau phosphorylation by diverse signals require the tumor suppressor protein LKB1. *J. Neurosci.* 27: 574-581.
- Terabayashi, T., et al. 2007. Polarity-regulating kinase partitioning-defective 1/microtubule affinity-regulating kinase 2 negatively regulates development of dendrites on hippocampal neurons. *J. Neurosci.* 27: 13098-13107.
- Zeaiter, Z., et al. 2008. Analysis of detergent-resistant membranes of *Helicobacter pylori* infected gastric adenocarcinoma cells reveals a role for MARK2/Par1b in CagA-mediated disruption of cellular polarity. *Cell. Microbiol.* 10: 781-794.

CHROMOSOMAL LOCATION

Genetic locus: Mark2 (mouse) mapping to 19 A.

PRODUCT

MARK2 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 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 MARK2 siRNA (m): sc-45794 and MARK2 shRNA Plasmid (m): sc-45794-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

MARK2 shRNA (m) Lentiviral Particles is recommended for the inhibition of MARK2 expression in mouse 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

MARK2 (H-86): sc-98800 is recommended as a control antibody for monitoring of MARK2 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 MARK2 gene expression knockdown using RT-PCR Primer: MARK2 (m)-PR: sc-45794-PR (20 μ l). 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.

PROTOCOLS

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