SANTA CRUZ BIOTECHNOLOGY, INC.

MARK3 siRNA (h): sc-43649

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

1. Ono, T., et al. 1997. Assignment of MARK3 alias KP78 to human chromosome band 14q32.3 by *in situ* hybridization. Cytogenet. Cell Genet. 79: 101-102.
2. 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.
3. Müller, J., et al. 2001. C-TAK1 regulates Ras signaling by phosphorylating the MAPK scaffold, KSR1. Mol. Cell 8: 983-993.
4. Sun, T.Q., et al. 2001. PAR-1 is a Dishevelled-associated kinase and a positive regulator of Wnt signalling. Nat. Cell Biol. 3: 628-636.
5. Hurov, J.B., et al. 2004. Atypical PKC phosphorylates PAR-1 kinases to regulate localization and activity. Curr. Biol. 14: 736-741.
6. Lizcano, J.M., et al. 2004. LKB1 is a master kinase that activates 13 kinases of the AMPK subfamily, including MARK/PAR-1. EMBO J. 23: 833-843.
7. Dequiedt, F., et al. 2006. New role for hPar-1 kinases EMK and C-TAK1 in regulating localization and activity of class IIa histone deacetylases. Mol. Cell. Biol. 26: 7086-7102.
8. Moravcevic, K., et al. 2010. Kinase associated-1 domains drive MARK/PAR1 kinases to membrane targets by binding acidic phospholipids. Cell 143: 966-977.

CHROMOSOMAL LOCATION

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

PRODUCT

MARK3 siRNA (h) is a pool of 3 target-specific 19-25 nt siRNAs designed to knock down gene expression. Each vial contains 3.3 nmol of lyophilized siRNA, sufficient for a 10 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see MARK3 shRNA Plasmid (h): sc-43649-SH and MARK3 shRNA (h) Lentiviral Particles: sc-43649-V as alternate gene silencing products.

For independent verification of MARK3 (h) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-43649A, sc-43649B and sc-43649C.

STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at -20° C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at -20° C, avoid contact with RNAses and repeated freeze thaw cycles.

Resuspend lyophilized siRNA duplex in 330 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

MARK3 siRNA (h) is recommended for the inhibition of MARK3 expression in human cells.

SUPPORT REAGENTS

For optimal siRNA transfection efficiency, Santa Cruz Biotechnology's siRNA Transfection Reagent: sc-29528 (0.3 ml), siRNA Transfection Medium: sc-36868 (20 ml) and siRNA Dilution Buffer: sc-29527 (1.5 ml) are recommended. Control siRNAs or Fluorescein Conjugated Control siRNAs are available as 10 µM in 66 µl. Each contain a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA. Fluorescein Conjugated Control siRNAs include: sc-36869, sc-44239, sc-44240 and sc-44241. Control siRNAs include: sc-37007, sc-44230, sc-44231, sc-44232, sc-44233, sc-44234, sc-44235, sc-44236, sc-44237 and sc-44238.

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 reagents are recommended: 1) Western Blotting: use m-IgGκ BP-HRP: sc-516102 or m-IgGκ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker™ Molecular Weight Standards: sc-2035, UltraCruz® Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgGκ BP-FITC: sc-516140 or m-IgGκ BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz® Mounting Medium: sc-24941 or UltraCruz® Hard-set Mounting Medium: sc-359850.

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.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

PROTOCOLS

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