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KRIT1 siRNA (m): sc-146571

BACKGROUND

The Krev interaction-trapped 1 (KRIT1) gene encodes a 529 amino acid microtubule-associated protein. Specifically, during interphase, KRIT1 localizes along the length of microtubules, whereas during metaphase it localizes on spindle pole bodies and on the mitotic spindle. During later phases of mitosis, KRIT1 localizes to the midbody where plus ends from dividing cells overlap. KRIT1 interacts with both Krev1 and integrin cytoplasmic domain-associated protein-1 α (ICAP1 α), suggesting that KRIT1 may help determine endothelial cell shape and function in response to cell-cell and cell-matrix interactions by guiding cytoskeletal structure. In addition, KRIT1 mutations are implicated in individuals with Cerebral Cavous Malformations (CCM). CCMs are capillary-venous abnormalities located mostly within the central nervous system, and occasionally within the skin and/or retina. CCMs may occur either sporadically or as an autosomal dominant condition and can result in cerebral hemorrhages, strokes and seizures.

REFERENCES

1. Serebrii, I., et al. 1997. Association of Krev-1/rap1a with Krit1, a novel ankyrin repeat-containing protein encoded by a gene mapping to 7q21-22. *Oncogene* 15: 1043-1049.
2. Craig, H.D., et al. 1998. Multilocus linkage identifies two new loci for a mendelian form of stroke, cerebral cavernous malformation, at 7p15-13 and 3q25.2-27. *Hum. Mol. Genet.* 7: 1851-1858.
3. Gunel, M., et al. 2002. KRIT1, a gene mutated in cerebral cavernous malformation, encodes a microtubule-associated protein. *Proc. Natl. Acad. Sci. USA* 99: 10677-10682.
4. Denier, C., et al. 2002. Krit1/cerebral cavernous malformation 1 mRNA is preferentially expressed in neurons and epithelial cells in embryo and adult. *Mech Dev.* 117: 363-367.
5. Kehrer-Sawatzki, H., et al. 2002. Mutation and expression analysis of the KRIT1 gene associated with cerebral cavernous malformations (CCM1). *Acta Neuropathol.* 104: 231-240.

CHROMOSOMAL LOCATION

Genetic locus: Krit1 (mouse) mapping to 5 A1.

PRODUCT

KRIT1 siRNA (m) 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see KRIT1 shRNA Plasmid (m): sc-146571-SH and KRIT1 shRNA (m) Lentiviral Particles: sc-146571-V as alternate gene silencing products.

For independent verification of KRIT1 (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-146571A, sc-146571B and sc-146571C.

PROTOCOLS

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

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 μ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330 μ l of RNase-free water makes a 10 μ M solution in a 10 μ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

KRIT1 siRNA (m) is recommended for the inhibition of KRIT1 expression in mouse 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 μ M in 66 μ l. 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

KRIT1 (E-8): sc-514371 is recommended as a control antibody for monitoring of KRIT1 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-IgG κ BP-HRP: sc-516102 or m-IgG κ 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-IgG κ BP-FITC: sc-516140 or m-IgG κ 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 KRIT1 gene expression knockdown using RT-PCR Primer: KRIT1 (m)-PR: sc-146571-PR (20 μ l). 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.