

CAP1 siRNA (h): sc-88068

BACKGROUND

Cyclase-associated proteins (CAPs) are a family of evolutionary conserved proteins that participate in signal transduction and function to regulate events associated with the Actin cytoskeleton. CAP1 and CAP2 (adenylylate cyclase-associated protein 1 and 2, respectively) are two members of the CAP family, both of which localize to the cell membrane and contain one C-CAP/cofactor C-like domain. CAP1 is involved in the regulation of Actin filaments and is thought to mediate processes such as establishment of cell polarity and mRNA localization, while CAP2 has a bifunctional regulatory role and can interact directly with Actin. Although CAP1 is expressed throughout the body, CAP2 is predominately expressed in skin, brain, heart and skeletal muscle. Overexpression of CAP2 is associated with hepatocellular carcinoma, suggesting a possible role for CAP2 in carcinogenesis.

REFERENCES

1. Matviw, H., et al. 1992. Identification of a human cDNA encoding a protein that is structurally and functionally related to the yeast adenylyl cyclase-associated CAP proteins. *Mol. Cell. Biol.* 12: 5033-5040.
2. Yu, G., et al. 1994. Comparison of human CAP and CAP2, homologs of the yeast adenylyl cyclase-associated proteins. *J. Cell Sci.* 107: 1671-1678.
3. Hubberstey, A., et al. 1996. Mammalian CAP interacts with CAP, CAP2, and Actin. *J. Cell. Biochem.* 61: 459-466.
4. Moriyama, K., et al. 2002. Human CAP1 is a key factor in the recycling of Cofilin and Actin for rapid Actin turnover. *J. Cell Sci.* 115: 1591-1601.
5. Dodatko, T., et al. 2004. Crystal structure of the Actin binding domain of the cyclase-associated protein. *Biochemistry* 43: 10628-10641.

CHROMOSOMAL LOCATION

Genetic locus: CAP1 (human) mapping to 1p34.2.

PRODUCT

CAP1 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see CAP1 shRNA Plasmid (h): sc-88068-SH and CAP1 shRNA (h) Lentiviral Particles: sc-88068-V as alternate gene silencing products.

For independent verification of CAP1 (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-88068A, sc-88068B and sc-88068C.

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

CAP1 siRNA (h) is recommended for the inhibition of CAP1 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 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

CAP1 (E-4): sc-376191 is recommended as a control antibody for monitoring of CAP1 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 CAP1 gene expression knockdown using RT-PCR Primer: CAP1 (h)-PR: sc-88068-PR (20 μ l, 591 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Ramsey, A., et al. 2023. CAP1 (cyclase-associated protein 1) mediates the cyclic AMP signals that activate Rap1 in stimulating matrix adhesion of colon cancer cells. *Cell. Signal.* 104: 110589.

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.