SANTA CRUZ BIOTECHNOLOGY, INC.

CKAP4 siRNA (m): sc-142354

BACKGROUND

CKAP4 (cytoskeleton-associated protein 4), also known as p63, CLIMP-63 or ERGIC-63, is a 602 amino acid single-pass type II transmembrane protein that links the endoplasmic reticulum (ER) to the cytoskeleton. Considered a novel protein in maintaining ER morphology, CKAP4 anchors the ER to microtubles which is required for maintaining ER spatial distribution during interphase of the cell cycle. CKAP4 can be reversibly palmitoylated and phosphorylated and is a major substrate of the palmitoyl acyltransferase, ZDHHC2. It is suggested that CKAP4 binds with high affinity to an inhibitor of cell proliferation, antiproliferative factor (APF), and blocks its activity on bladder epithelial cells. Two isoforms of CKAP4 exist due to alternative splicing.

REFERENCES

1. Klopfenstein, D.R., et al. 2001. Subdomain-specific localization of CLIMP-63 (p63) in the endoplasmic reticulum is mediated by its luminal α-helical segment. J. Cell Biol. 153: 1287-1300.
2. Vedrenne, C., et al. 2005. Phosphorylation controls CLIMP-63-mediated anchoring of the endoplasmic reticulum to microtubules. Mol. Biol. Cell 16: 1928-1937.
3. Gupta, N., et al. 2006. Identification and characterization of p63 (CKAP4/ERGIC-63/CLIMP-63), a surfactant protein A binding protein, on type II pneumocytes. Am. J. Physiol. Lung Cell. Mol. Physiol. 291: L436-L446.
4. Conrads, T.P., et al. 2006. CKAP4/p63 is a receptor for the frizzled-8 protein-related antiproliferative factor from interstitial cystitis patients. J. Biol. Chem. 281: 37836-37843.
5. Wesierska-Gadek, J., et al. 2007. A new, unexpected action of olomoucine, a CDK inhibitor, on normal human cells: up-regulation of CLIMP-63, a cytoskeleton-linking membrane protein. J. Cell. Biochem. 102: 1405-1419.
6. Nikonov, A.V., et al. 2007. Climp-63-mediated binding of microtubules to the ER affects the lateral mobility of translocon complexes. J. Cell Sci. 120: 2248-2258.

CHROMOSOMAL LOCATION

Genetic locus: Ckap4 (mouse) mapping to 10 C1.

PRODUCT

CKAP4 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see CKAP4 shRNA Plasmid (m): sc-142354-SH and CKAP4 shRNA (m) Lentiviral Particles: sc-142354-V as alternate gene silencing products.

For independent verification of CKAP4 (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-142354A, sc-142354B and sc-142354C.

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 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

CKAP4 siRNA (m) is recommended for the inhibition of CKAP4 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 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

CKAP4 (A-3): sc-393544 is recommended as a control antibody for monitoring of CKAP4 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 CKAP4 gene expression knockdown using RT-PCR Primer: CKAP4 (m)-PR: sc-142354-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.