SANTA CRUZ BIOTECHNOLOGY, INC.

CHERP siRNA (h): sc-97408

BACKGROUND

The regulation of the intracellular concentration of calcium is important for proper maintenance of voltage-gated ion channels which control muscle and nerve function. Calcium homeostasis is regulated by a variety of proteins. CHERP (calcium homeostasis endoplasmic reticulum protein), also known as SRA1, DAN16 or SCAF6, is a 916 amino acid protein that localizes to the cytoplasm and the endoplasmic reticulum (ER). Expressed in pancreas, brain, lung, placenta, liver, kidney, heart and skeletal muscle, CHERP is involved in maintaining calcium homeostasis and plays a role in cell growth and proliferation. CHERP contains one G-patch domain, one RPR domain and one SURP motif and is expressed as two isoforms due to alternative splicing events.

REFERENCES

1. O'Rourke, F., et al. 1994. Ca^{2+} release by inositol 1,4,5-trisphosphate is blocked by the K^{+}-channel blockers apamin and tetrapentylammonium ion, and a monoclonal antibody to a 63 kDa membrane protein: reversal of blockade by K^{+} ionophores nigericin and valinomycin and purification of the 63 kDa antibody-binding protein. Biochem. J. 300: 673-683.
2. Laplante, J.M., et al. 2000. Cloning of human Ca^{2+} homoeostasis endoplasmic reticulum protein (CHERP): regulated expression of antisense cDNA depletes CHERP, inhibits intracellular Ca^{2+} mobilization and decreases cell proliferation. Biochem. J. 348: 189-199.
3. Ding, W., et al. 2002. Human T-cell lymphotropic virus type 1 p12(I) expression increases cytoplasmic calcium to enhance the activation of nuclear factor of activated T cells. J. Virol. 76: 10374-10382.
4. O'Rourke, F.A., et al. 2003. Antisense-mediated loss of calcium homoeostasis endoplasmic reticulum protein (CHERP; ERPROT213-21) impairs Ca^{2+} mobilization, nuclear factor of activated T-cells (NFAT) activation and cell proliferation in Jurkat T-lymphocytes. Biochem. J. 373: 133-143.
5. Brandenberger, R., et al. 2004. Transcriptome characterization elucidates signaling networks that control human ES cell growth and differentiation. Nat. Biotechnol. 22: 707-716.

CHROMOSOMAL LOCATION

Genetic locus: CHERP (human) mapping to 19p13.1.

PRODUCT

CHERP 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 CHERP shRNA Plasmid (h): sc-97408-SH and CHERP shRNA (h) Lentiviral Particles: sc-97408-V as alternate gene silencing products.

For independent verification of CHERP (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-97408A, sc-97408B and sc-97408C.

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

CHERP siRNA (h) is recommended for the inhibition of CHERP 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 60 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

CHERP (SS5): sc-100650 is recommended as a control antibody for monitoring of CHERP 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 CHERP gene expression knockdown using RT-PCR Primer: CHERP (h)-PR: sc-97408-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.