SANTA CRUZ BIOTECHNOLOGY, INC.

CLIC5 siRNA (h): sc-62128

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The Power to Question

BACKGROUND

Chloride intracellular channel 5 (CLIC5), is a member of the highly conserved family of chloride ion channels that function in both soluble and integral membrane forms. Chloride channels regulate cellular traffic of chloride ions, a critical component of all living cells. They are involved in membrane potential stabilization, signal transduction, cell volume regulation and organic solute transport. CLIC5 associates with Actin-based cytoskeletal structures and may be involved in their assembly and maintenance. In addition, CLIC5 may also play an important role in inner ear function localizing to the stereocilia and possibly associating with Radixin. Two CLIC5 isoforms exist and they are referred to as CLIC5A and CLIC5B. The two isoforms share an identical C-terminus but have distinct N-terminal sequences. CLIC5B is the human ortholog of bovine p64 and avian p62. It binds AKAP350 and localizes to the Golgi apparatus.

REFERENCES

1. Berryman, M., et al. 2000. Identification of a novel member of the chloride intracellular channel gene family (CLIC5) that associates with the Actin cytoskeleton of placental microvilli. Mol. Biol. Cell 11: 1509-1521.
2. Rønnov-Jessen, L., et al. 2002. Differential expression of a chloride intracellular channel gene, CLIC4, in transforming growth factor-β1-mediated conversion of fibroblasts to myofibroblasts. Am. J. Pathol. 161: 471-480.
3. Shanks, R.A., et al. 2002. AKAP350 at the Golgi apparatus. II. Association of AKAP350 with a novel chloride intracellular channel (CLIC) family member. J. Biol. Chem. 277: 40973-40980.
4. Friedli, M., et al. 2003. Identification of a novel member of the CLIC family, CLIC6, mapping to 21q22.12. Gene 320: 31-40.
5. Berryman, M., et al. 2004. CLIC-5A functions as a chloride channel *in vitro* and associates with the cortical actin cytoskeleton *in vitro* and *in vivo*. J. Biol. Chem. 279: 34794-34801.
6. Gagnon, L.H., et al. 2006. The chloride intracellular channel protein CLIC5 is expressed at high levels in hair cell stereocilia and is essential for normal inner ear function. J. Neurosci. 26: 10188-10198.

CHROMOSOMAL LOCATION

Genetic locus: CLIC5 (human) mapping to 6p21.1.

PRODUCT

CLIC5 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 CLIC5 shRNA Plasmid (h): sc-62128-SH and CLIC5 shRNA (h) Lentiviral Particles: sc-62128-V as alternate gene silencing products.

For independent verification of CLIC5 (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-62128A, sc-62128B and sc-62128C.

RESEARCH USE

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

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

CLIC5 siRNA (h) is recommended for the inhibition of CLIC5 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

CLIC4/5/6 (A-11): sc-271863 is recommended as a control antibody for monitoring of CLIC5 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 CLIC5 gene expression knockdown using RT-PCR Primer: CLIC5 (h)-PR: sc-62128-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

PROTOCOLS

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