SANTA CRUZ BIOTECHNOLOGY, INC.

Fibrocystin siRNA (m): sc-60638

BACKGROUND

Fibrocystin is a type I membrane protein that undergoes regulated proteolysis. Many proteolytic cleavages occur on the ectodomain whereas at least one cleavage occurs on the cytoplasmic portion of Fibrocystin. The latter generates a C-terminal intracellular fragment that localizes to the nucleus. This proteolysis requires activation of protein kinase C (PKC) and release of intracellular calcium. Fibrocystin is expressed in the cilia of the bile duct epithelium and leads to abnormalities in the rubric of the ductal plate malformation. The intracellular C-terminus of Fibrocystin interacts with calcium modulating cyclophilin ligand (CAML), a protein implicated in calcium signaling. Fibrocystin may participate in the mediation of intracellular calcium in the distal nephron in a manner similar to PKD1 and PKD2. Mutations in the PKHD1 gene, which encodes Fibrocystin, result in autosomal recessive polycystic kidney disease (ARPKD), a severe form of polycystic kidney disease characterized by enlarged kidneys and congenital hepatic fibrosis.

REFERENCES

1. Masyuk, T.V., et al. 2003. Defects in cholangiocyte Fibrocystin expression and ciliary structure in the PCK rat. Gastroenterology 125: 1303-1310.
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3. Bergmann, C., et al. 2005. Algorithm for efficient PKHD1 mutation screening in autosomal recessive polycystic kidney disease (ARPKD). Hum. Mutat. 25: 225-231.
4. Bergmann, C., et al. 2005. Clinical consequences of PKHD1 mutations in 164 patients with autosomal-recessive polycystic kidney disease (ARPKD). Kidney Int. 67: 829-848.
5. Losekoot, M., et al. 2005. Analysis of missense variants in the PKHD1 gene in patients with autosomal recessive polycystic kidney disease (ARPKD). Hum. Genet. 118: 185-206.
6. Mai, W., et al. 2005. Inhibition of Pkhd1 impairs tubulomorphogenesis of cultured IMCD cells. Mol. Biol. Cell 16: 4398-4409.
7. Nagano, J., et al. 2005. Fibrocystin interacts with CAML, a protein involved in Ca^{2+} signaling. Biochem. Biophys. Res. Commun. 338: 880-889.

CHROMOSOMAL LOCATION

Genetic locus: Pkhd1 (mouse) mapping to 1 A3.

PRODUCT

Fibrocystin 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 Fibrocystin shRNA Plasmid (m): sc-60638-SH and Fibrocystin shRNA (m) Lentiviral Particles: sc-60638-V as alternate gene silencing products.

For independent verification of Fibrocystin (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-60638A, sc-60638B and sc-60638C.

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

Fibrocystin siRNA (m) is recommended for the inhibition of Fibrocystin 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Fibrocystin gene expression knockdown using RT-PCR Primer: Fibrocystin (m)-PR: sc-60638-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.

PROTOCOLS

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