SANTA CRUZ BIOTECHNOLOGY, INC.

nephrocystin siRNA (m): sc-40770

BACKGROUND

Clinical features of familial juvenile nephronophthisis include anemia, polyuria, polydipsia, isosthenuria, and death in uremia. Juvenile nephronophthisis type 1 is caused by mutations of NPHP1, the gene encoding for nephrocystin. Nephrocystin interacts with $p130^{Cas}$ (BCAR1), proline-rich tyrosine kinase-2 (PTK2B or Pyk2), and tensin in embryonic kidney and testis, indicating that these proteins participate in a common signaling pathway. Nephrocystin and $p130^{Cas}$ interact in mammalian cells and both proteins prominently localize at or near sites of cell-cell contact in polarized Madin-Darby canine kidney epithelial cells. Expression of nephrocystin results in phosphorylation of Pyk2 on Tyrosine 402 as well as activation of downstream mitogen-activated protein kinases, such as ERK1 and ERK2. Nephrocystin contains a src-homology 3 (SH_3) domain, which is highly conserved throughout evolution. The gene which encodes nephrocystin maps to human chromosome 2q13.

REFERENCES

1. Medhioub, M., et al. 1994. Refined mapping of agene (NPH1) causing familial juvenile nephronophthisis and evidence for genetic heterogeneity. Genomics 22: 296-301.
2. Donaldson, J.C., et al. 2000. Crk-associated substrate $p130^{Cas}$ interacts with nephrocystin and both proteins localize to cell-cell contacts of polarized epithelial cells. Exp. Cell Res. 256: 168-178.
3. Benzing, T., et al. 2001. Nephrocystin interacts with Pyk2, $p130^{Cas}$, and tensin and triggers phosphorylation of Pyk2. Proc. Natl. Acad. Sci. USA 98: 9784-9789.
4. Hildebrandt, F., et al. 2001. New insights: nephronophthisis-medullary cystic kidney disease. Pediatr. Nephrol. 16: 168-176.
5. LocusLink Report (LocusID: 256100). http://www.ncbi.nlm.nih.gov/LocusLink

CHROMOSOMAL LOCATION

Genetic locus: Nphp1 (mouse) mapping to 2 F1.

PRODUCT

nephrocystin 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 nephrocystin shRNA Plasmid (m): sc-40770-SH and nephrocystin shRNA (m) Lentiviral Particles: sc-40770-V as alternate gene silencing products.

For independent verification of nephrocystin (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-40770A, sc-40770B and sc-40770C.

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

nephrocystin siRNA (m) is recommended for the inhibition of nephrocystin 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 nephrocystin gene expression knockdown using RT-PCR Primer: nephrocystin (m)-PR: sc-40770-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.