

nephrocystin-2 siRNA (m): sc-149912

BACKGROUND

Left-right asymmetry in vertebrates is essential for the development of lateral unpaired organs, including the heart, stomach and spleen, and is dependent on the differential expression of specific genes, which include nodal, lefty and nephrocystin-2. Nephrocystin-2, also known as inversin (INV), inversion of embryo turning homolog or NPHP2, is a 1,065 amino acid protein that exists as three alternatively spliced isoforms and is essential for establishment of the left-right axis and normal renal development. Localizing to the cytoplasm, cytoskeleton, membrane and nucleus, nephrocystin-2 is expressed during presomite-stage embryos and persists in adulthood, with high levels of expression in liver and kidney. Mice expressing nephrocystin-2 mutations are primarily generated by random insertional mutagenesis and result in the reversal of left/right polarity and cyst formation in the kidneys. Furthermore, altered nephrocystin-2 function reverses nodal and lefty expression, indicating that nephrocystin-2 signaling occurs upstream of these proteins involved in the development of asymmetry.

REFERENCES

1. Lux, S.E., et al. 1990. Analysis of cDNA for human erythrocyte ankyrin indicates a repeated structure with homology to tissue-differentiation and cell-cycle control proteins. *Nature* 344: 36-42.
2. Yokoyama, T., et al. 1990. Conserved cysteine to serine mutation in tyrosinase is responsible for the classical albino mutation in laboratory mice. *Nucleic Acids Res* 18: 7293-7298.
3. Yokoyama, T., et al. 1993. Reversal of left-right asymmetry: a situs inversus mutation. *Science* 260: 679-682.
4. Lowe, L.A., et al. 1996. Conserved left-right asymmetry of nodal expression and alterations in murine situs inversus. *Nature* 381: 158-161.
5. Mochizuki, T., et al. 1998. Cloning of inv, a gene that controls left/right asymmetry and kidney development. *Nature* 395: 177-181.

CHROMOSOMAL LOCATION

Genetic locus: Invs (mouse) mapping to 4 B1.

PRODUCT

nephrocystin-2 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see nephrocystin-2 shRNA Plasmid (m): sc-149912-SH and nephrocystin-2 shRNA (m) Lentiviral Particles: sc-149912-V as alternate gene silencing products.

For independent verification of nephrocystin-2 (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-149912A, sc-149912B and sc-149912C.

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 μ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330 μ l of RNase-free water makes a 10 μ M solution in a 10 μ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

nephrocystin-2 siRNA (m) is recommended for the inhibition of nephrocystin-2 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 μ M in 66 μ l. 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-2 gene expression knockdown using RT-PCR Primer: nephrocystin-2 (m)-PR: sc-149912-PR (20 μ l). 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.