

cubilin siRNA (h): sc-40099

BACKGROUND

Cubilin is an endocytic receptor that lacks a classical transmembrane region. It is a multidomain receptor that contains an amino terminal 110 residue segment followed by 8 epidermal growth factor (EGF)-like repeats and a contiguous stretch of 27 CUB domains. The gene encoding human cubilin maps to chromosome 10p13 and it is predominantly expressed in intestine, kidney and yolk sac. It also is expressed in intestinal membranes. Cubilin colocalizes with and binds to megalin, a member of the LDL receptor family that is required for the internalization of cubilin-bound ligands, such as vitamin B12, apolipoprotein A1 and albumin. Megalin specifically binds to cubilin in the amino terminal region that contains the EGF-like repeats and CUB domains 1 and 2. Mutations in the cubilin gene are thought to cause megaloblastic anemia 1 (MGA1), an autosomal recessive disorder also known as Imerslund-Grasbeck's disease, which causes intestinal malabsorption of vitamin B12.

REFERENCES

1. Kozyraki, R., et al. 1998. The human intrinsic factor-vitamin B12 receptor, cubilin: molecular characterization and chromosomal mapping of the gene to 10p within the autosomal recessive megaloblastic anemia (MGA1) region. *Blood* 91: 3593-3600.
2. Aminoff, M., et al. 1999. Mutations in CUBN, encoding the intrinsic factor-vitamin B12 receptor, cubilin, cause hereditary megaloblastic anaemia 1. *Nat. Genet.* 21: 309-313.
3. Kristiansen, M., et al. 2000. Cubilin P1297L mutation associated with hereditary megaloblastic anemia 1 causes impaired recognition of intrinsic factor-vitamin B12 by cubilin. *Blood* 96: 405-409.
4. Kozyraki, R., et al. 2001. Megalin-dependent cubilin-mediated endocytosis is a major pathway for the apical uptake of transferrin in polarized epithelia. *Proc. Natl. Acad. Sci. USA* 98: 12491-12496.
5. Yammani, R.R., et al. 2001. Cubilin and megalin expression and their interaction in the rat intestine: effect of thyroidectomy. *Am. J. Physiol. Endocrinol. Metab.* 281: E900-E907.
6. Kozyraki, R. 2001. Cubilin, a multifunctional epithelial receptor: an overview. *J. Mol. Med.* 79: 161-167.

CHROMOSOMAL LOCATION

Genetic locus: CUBN (human) mapping to 10p13.

PRODUCT

cubilin 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see cubilin shRNA Plasmid (h): sc-40099-SH and cubilin shRNA (h) Lentiviral Particles: sc-40099-V as alternate gene silencing products.

For independent verification of cubilin (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-40099A, sc-40099B and sc-40099C.

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

cubilin siRNA (h) is recommended for the inhibition of cubilin 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 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 cubilin gene expression knockdown using RT-PCR Primer: cubilin (h)-PR: sc-40099-PR (20 μ l, 494 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Basnayake, K., et al. 2010. Immunoglobulin light chains activate tubular epithelial cells through redox signaling. *J. Am. Soc. Nephrol.* 21: 1165-1173.

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.