rBAT siRNA (h): sc-106486



The Power to Question

BACKGROUND

Heterodimeric amino acid transporters mediate the transfer of amino acids between organs and between different cell types. The heavy chain subunit is a type II membrane protein with an intracellular amino terminus, a single transmembrane helix, and a large intracellular domain. The SLC3A1 gene encodes one of these heavy chains, rBAT, which dimerize with a light chain subunit (seven types have been identified) to facilitate reabsorption of dibasic amino acids and cystine in renal and intenstinal epithelial cells. Defects in this transport system causes cystinuria, a disease that manifests as the development of kidney stones. Mutations in SLC3A1 or the gene encoding the light chain subunit, SLC7A9, both cause cystinuria, the former classified as "type I" and the latter as "non-type I", however, the clinical presentation of the two is indistinguishable, expounding the importance of the functional complex, and not just one subunit, for normal amino acid transport.

REFERENCES

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- Botzenhart, E., et al. 2002. Cystinuria in children: distribution and frequencies of mutations in the SLC3A1 and SLC7A9 genes. Kidney Int. 62: 1136-1142.
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- Moschen, I., et al. 2002. Influence of rBAT-mediated amino acid transport on cytosolic pH. Nephron 91: 631-636.
- Peters, T., et al. 2003. A mouse model for cystinuria type I. Hum. Mol. Genet. 12: 2109-2120.
- 6. Orts Costa, J.A., et al. 2003. Cystinuria update: clinical, biochemical and genetic aspects. An. Med. Interna 20: 317-326.
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CHROMOSOMAL LOCATION

Genetic locus: SLC3A1 (human) mapping to 2p21.

PRODUCT

rBAT 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 rBAT shRNA Plasmid (h): sc-106486-SH and rBAT shRNA (h) Lentiviral Particles: sc-106486-V as alternate gene silencing products.

For independent verification of rBAT (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-106486A, sc-106486B and sc-106486C.

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

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

rBAT (G-5): sc-518240 is recommended as a control antibody for monitoring of rBAT 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).

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor rBAT gene expression knockdown using RT-PCR Primer: rBAT (h)-PR: sc-106486-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.

PROTOCOLS

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

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