

OATP-A siRNA (h): sc-42548

BACKGROUND

The organic anion transporting polypeptides, OATP-A (also designated OATP1, OATP1A2 and SLC21A3) and OATP-C (also designated OATP2, SLC21A6 and LST1), mediate hepatic uptake of cardiac glycosides. The expression of OATP-C, but not OATP-A, is inducible by phenobarbital and pregnenolone-16 α -carbonitrile, resulting in the increased capacity of the liver to extract cardiac glycosides from the plasma. OATP-A is expressed in liver and kidney and helps mediate sodium-independent uptake of the anionic steroid conjugates dehydroepiandrosterone sulfate, estradiol-17-glucuronide and prostaglandin. OATP-C is exclusively expressed in liver and is localized to the basolateral hepatocyte membrane. Although OATP-C mRNA levels decrease during pregnancy and increase postpartum, OATP-C protein levels remain relatively constant. OATP-C transports taurocholic acid, the adrenal androgen dehydroepiandrosterone sulfate, thyroid hormone, hydroxymethylglutaryl-CoA reductase inhibitor and pravastatin. OATP-C is therefore a novel organic anion transport protein that has overlapping but not identical substrate specificities with other subtypes of OATP. OATP-A and OATP-C are both pravastatin transporters, suggesting that they are responsible for the hepatic uptake of the liver-specific hydroxymethylglutaryl-CoA reductase inhibitor in mouse, rat and human.

REFERENCES

1. Hsiang, B., et al. 1999. A novel human hepatic organic anion transporting polypeptide (OATP2). *J. Biol. Chem.* 274: 37161-37168.
2. Konig, J., et al. 2000. Localization and genomic organization of a new hepatocellular organic anion transporting polypeptide. *J. Biol. Chem.* 275: 23161-23168.
3. Cao, J., et al. 2001. Differential regulation of hepatic bile salt and organic anion transporters in pregnant and postpartum rats and the role of prolactin. *Hepatology* 33: 140-147.
4. Rausch-Derra, L.C., et al. 2001. Differential effects of microsomal enzyme-inducing chemicals on the hepatic expression of rat organic anion transporters, OATP1 and OATP2. *Hepatology* 33: 1469-1478.
5. Isern, J., et al. 2001. Functional analysis and androgen-regulated expression of mouse organic anion transporting polypeptide 1 (Oatp1) in the kidney. *Biochem. Biophys. Acta* 1518: 73-78.

CHROMOSOMAL LOCATION

Genetic locus: SLC01A2 (human) mapping to 12p12.1.

PRODUCT

OATP-A 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 OATP-A shRNA Plasmid (h): sc-42548-SH and OATP-A shRNA (h) Lentiviral Particles: sc-42548-V as alternate gene silencing products.

For independent verification of OATP-A (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-42548A, sc-42548B and sc-42548C.

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

OATP-A siRNA (h) is recommended for the inhibition of OATP-A 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.

GENE EXPRESSION MONITORING

OATP-A (E-7): sc-365007 is recommended as a control antibody for monitoring of OATP-A 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).

To ensure optimal results, the following support reagents are recommended: 1) Western Blotting: use m-IgG κ BP-HRP: sc-516102 or m-IgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker[™] Molecular Weight Standards: sc-2035, UltraCruz[®] Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG κ BP-FITC: sc-516140 or m-IgG κ BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz[®] Mounting Medium: sc-24941 or UltraCruz[®] Hard-set Mounting Medium: sc-359850.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor OATP-A gene expression knockdown using RT-PCR Primer: OATP-A (h)-PR: sc-42548-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.