

# GALT siRNA (h): sc-92612

## BACKGROUND

GALT (galactose-1-phosphate uridylyltransferase) is a 379 amino acid member of the galactose-1-phosphate uridylyltransferase type 1 family of proteins. GALT exists as a homodimer and is believed to play a role in galactose metabolism. More specifically, GALT is responsible for catalyzing the reaction of UDP-glucose with  $\alpha$ -D-galactose 1-phosphate to produce  $\alpha$ -D-glucose 1-phosphate and UDP-galactose. This is the second step of the Leloir pathway of galactose metabolism. The products of this reaction will either enter the glycolytic pathway to yield energy ( $\alpha$ -D-glucose 1-phosphate) or be used as a galactosyl donor in the synthesis of glycoproteins and glycolipids (UDP-galactose). Mutations in the gene encoding GALT can lead to galactosemia, a disorder (occurring from the inability to metabolize galactose) that is characterized by cataracts, mental retardation and jaundice. In newborns, galactosemia can be fatal if lactose is not removed from the diet.

## REFERENCES

1. Reichardt, J.K., et al. 1988. Cloning and characterization of a cDNA encoding human galactose-1-phosphate uridyl transferase. *Mol. Biol. Med.* 5: 107-122.
2. Reichardt, J.K., et al. 1992. Molecular characterization of two galactosemia mutations and one polymorphism: implications for structure-function analysis of human galactose-1-phosphate uridylyltransferase. *Biochemistry* 31: 5430-5433.
3. Ninfali, P., et al. 1996. Molecular basis of galactose-1-phosphate uridylyltransferase deficiency involving skeletal muscle. *J. Neurol.* 243: 102-103.

## CHROMOSOMAL LOCATION

Genetic locus: GALT (human) mapping to 9p13.3.

## PRODUCT

GALT 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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see GALT shRNA Plasmid (h): sc-92612-SH and GALT shRNA (h) Lentiviral Particles: sc-92612-V as alternate gene silencing products.

For independent verification of GALT (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-92612A, sc-92612B and sc-92612C.

## STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at  $-20^{\circ}$  C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at  $-20^{\circ}$  C, avoid contact with RNAses and repeated freeze thaw cycles.

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

## APPLICATIONS

GALT siRNA (h) is recommended for the inhibition of GALT 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  $\mu$ M in 66  $\mu$ 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

GALT (G-1): sc-365577 is recommended as a control antibody for monitoring of GALT 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 $\kappa$  BP-HRP: sc-516102 or m-IgG $\kappa$  BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker<sup>™</sup> Molecular Weight Standards: sc-2035, UltraCruz<sup>®</sup> Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG $\kappa$  BP-FITC: sc-516140 or m-IgG $\kappa$  BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz<sup>®</sup> Mounting Medium: sc-24941 or UltraCruz<sup>®</sup> Hard-set Mounting Medium: sc-359850.

## RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor GALT gene expression knockdown using RT-PCR Primer: GALT (h)-PR: sc-92612-PR (20  $\mu$ l). Annealing temperature for the primers should be 55-60 $^{\circ}$  C and the extension temperature should be 68-72 $^{\circ}$  C.

## SELECT PRODUCT CITATIONS

1. Villegas-Comonfort, S., et al. 2012. Arachidonic acid induces an increase of  $\beta$ -1,4-galactosyltransferase I expression in MDA-MB-231 breast cancer cells. *J. Cell. Biochem.* 113: 3330-3341.

## RESEARCH USE

For research use only, not for use in diagnostic procedures.

## PROTOCOLS

See our web site at [www.scbt.com](http://www.scbt.com) for detailed protocols and support products.