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GALE siRNA (m): sc-145310

BACKGROUND

GALE, also known as galactowaldenase, UDP-galactose-4-epimerase or SDR1E1, is a 348 amino acid protein that functions as the third enzyme in the Leloir pathway of galactose metabolism. A member of the sugar epimerase family, GALE exists as a homodimer, binds FAD as a cofactor and catalyzes the epimerization of UDP-N-acetylglucosamine to UDP-N-acetylgalactosamine and UDP-glucose to UDP-galactose. The gene encoding GALE maps to human chromosome 1p36.11 and mutations in this gene lead to the development of complex disorder known as epimerase-deficiency galactosemia (EDG) or galactosemia type 3, which is characterized by mental retardation, liver damage, cataracts and deafness.

REFERENCES

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2. Holton, J.B., et al. 1981. Galactosaemia: a new severe variant due to uridine diphosphate galactose-4-epimerase deficiency. *Arch. Dis. Child.* 56: 885-887.
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4. Kingsley, D.M., et al. 1986. Reversible defects in O-linked glycosylation and LDL receptor expression in a UDP-Gal/UDP-GalNAc 4-epimerase deficient mutant. *Cell* 44: 749-759.
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6. Maceratesi, P., et al. 1998. Human UDP-galactose 4' epimerase (GALE) gene and identification of five missense mutations in patients with epimerase-deficiency galactosemia. *Mol. Genet. Metab.* 63: 26-30.
7. Wohlers, T.M., et al. 1999. Identification and characterization of a mutation, in the human UDP-galactose-4-epimerase gene, associated with generalized epimerase-deficiency galactosemia. *Am. J. Hum. Genet.* 64: 462-470.

CHROMOSOMAL LOCATION

Genetic locus: Gale (mouse) mapping to 4 D3.

PRODUCT

GALE siRNA (m) is a pool of 2 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 GALE shRNA Plasmid (m): sc-145310-SH and GALE shRNA (m) Lentiviral Particles: sc-145310-V as alternate gene silencing products.

For independent verification of GALE (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-145310A and sc-145310B.

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

GALE siRNA (m) is recommended for the inhibition of GALE 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.

GENE EXPRESSION MONITORING

GALE (C-4): sc-390407 is recommended as a control antibody for monitoring of GALE 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 GALE gene expression knockdown using RT-PCR Primer: GALE (m)-PR: sc-145310-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.