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GLDC siRNA (m): sc-145419

BACKGROUND

The glycine cleavage system is comprised of AMT (known as protein T), GCSH (known as protein H), DLD (known as protein L) and GLDC (known as protein P), all of which work together to catalyze the cleavage and degradation of glycine. GLDC (glycine dehydrogenase), also known as GCE, GCSP (glycine cleavage system P protein) or HYGN1, is a 1,020 amino acid protein that localizes to the mitochondria and belongs to the gcvP family. GLDC binds to glycine and enables the methylamine group from glycine to be transferred to the protein T. GLDC exists as a homodimer and utilizes pyridoxal phosphate as a cofactor. Mutations in the gene encoding GLDC leads to nonketotic hyperglycinemia (NKH), also known as glycine encephalopathy (GCE), an autosomal recessive disease characterized by accumulation of a large amount of glycine in body fluid and by severe neurological symptoms.

REFERENCES

1. Takayanagi, M., et al. 2000. Human glycine decarboxylase gene (GLDC) and its highly conserved processed pseudogene (psiGLDC): their structure and expression, and the identification of a large deletion in a family with non-ketotic hyperglycinemia. *Hum. Genet.* 106: 298-305.
2. Kure, S., et al. 2002. Heterozygous GLDC and GCSH gene mutations in transient neonatal hyperglycinemia. *Ann. Neurol.* 52: 643-646.
3. Toone, J.R., et al. 2002. Novel mutations in the P-protein (glycine decarboxylase) gene in patients with glycine encephalopathy (non-ketotic hyperglycinemia). *Mol. Genet. Metab.* 76: 243-249.
4. Flusser, H., et al. 2005. Mild glycine encephalopathy (NKH) in a large kindred due to a silent exonic GLDC splice mutation. *Neurology* 64: 1426-1430.
5. Conter, C., et al. 2006. Genetic heterogeneity of the GLDC gene in 28 unrelated patients with glycine encephalopathy. *J. Inher. Metab. Dis.* 29: 135-142.
6. Kanno, J., et al. 2007. Genomic deletion within GLDC is a major cause of non-ketotic hyperglycinaemia. *J. Med. Genet.* 44: e69.

CHROMOSOMAL LOCATION

Genetic locus: *Gldc* (mouse) mapping to 19 C1.

PRODUCT

GLDC siRNA (m) 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 GLDC shRNA Plasmid (m): sc-145419-SH and GLDC shRNA (m) Lentiviral Particles: sc-145419-V as alternate gene silencing products.

For independent verification of GLDC (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-145419A, sc-145419B and sc-145419C.

RESEARCH USE

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

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

GLDC siRNA (m) is recommended for the inhibition of GLDC 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

GLDC (H-9): sc-376196 is recommended as a control antibody for monitoring of GLDC 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 GLDC gene expression knockdown using RT-PCR Primer: GLDC (m)-PR: sc-145419-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

PROTOCOLS

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