

LDH-A siRNA (r): sc-270631

BACKGROUND

The lactate dehydrogenase family (LDH) catalyzes the final step of anaerobic glycolysis, the conversion of L-lactate and NAD to pyruvate and NADH. The LDH family consists of three members, LDH-A, LDH-B, and LDH-C, all of which form tetramers consisting four subunits. However, each family member displays a specific tissue distribution pattern with LDH-A and LDH-B predominant in several tissues, specifically LDH-A in muscle and LDH-B in heart, while LDH-C expression is confined to the testis and sperm. LDHs function as powerful markers for germ cell tumors. The genes encoding human LDH-A and LDH-C map to chromosome 11, while the human LDH-B gene maps to chromosome 12. Deficiency in the LDH-A gene is linked to exertional myoglobinuria.

REFERENCES

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- LeVan, K.M., et al. 1991. Properties of human testis-specific lactate dehydrogenase expressed from *Escherichia coli*. *Biochem. J.* 273: 587-592.
- Kanno, T., et al. 1995. Lactate dehydrogenase M-subunit deficiencies: clinical features, metabolic background, and genetic heterogeneities. *Muscle Nerve* 3: 54-60.
- Kopperschlager, G., et al. 1996. Methods for the separation of lactate dehydrogenases and clinical significance of the enzyme. *J. Chromatogr. B. Biomed. Appl.* 684: 25-49.
- Auerbach, G., et al. 1998. Lactate dehydrogenase from the hyperthermophilic bacterium *Thermotoga maritima*: the crystal structure at 2.1 Å resolution reveals strategies for intrinsic protein stabilization. *Structure* 6: 769-781.
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CHROMOSOMAL LOCATION

Genetic locus: *Ldha* (rat) mapping to 1q22.

PRODUCT

LDH-A siRNA (r) 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 LDH-A shRNA Plasmid (r): sc-270631-SH and LDH-A shRNA (r) Lentiviral Particles: sc-270631-V as alternate gene silencing products.

For independent verification of LDH-A (r) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-270631A, sc-270631B and sc-270631C.

PROTOCOLS

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

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

LDH-A siRNA (r) is recommended for the inhibition of LDH-A expression in rat 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

LDH-A (F-3): sc-137244 is recommended as a control antibody for monitoring of LDH-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 LDH-A gene expression knockdown using RT-PCR Primer: LDH-A (r)-PR: sc-270631-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.