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LSD1 siRNA (r): sc-156072

BACKGROUND

Histone methylation regulates chromatin structure and transcription and maintains an epigenetic state of the cell. Histone methylation is dynamically regulated by histone methylases and demethylases. Lysine-specific histone demethylase 1 (LSD1), also designated BHC110, is a flavin-dependent amine oxidase which catalyzes the removal of one or two methyl groups from the methyl-lysine-4 side chain of Histone H3. The LSD1 protein contains a SWIRM domain, a FAD-binding motif and an amine oxidase domain. Association with CoREST, a SANT domain-containing corepressor, positively regulates LSD1. CoREST mediates the demethylation ability of LSD1 and protects it from proteasomal degradation *in vivo*. PHF21A (also designated BCH80), a PHD domain-containing protein, inhibits activity of LSD1/CoREST mediated demethylation. The LSD1 protein also co-localizes with the androgen receptor in human prostate tumor cells and in unaffected prostate cells, stimulating androgen-receptor-dependent transcription.

REFERENCES

1. Shi, Y., et al. 2004. Histone demethylation mediated by the nuclear amine oxidase homolog LSD1. *Cell* 119: 941-953.
2. Forneris, F., et al. 2005. Histone demethylation catalysed by LSD1 is a flavin-dependent oxidative process. *FEBS Lett.* 579: 2203-2207.
3. Lee, M.G., et al. 2005. An essential role for CoREST in nucleosomal Histone 3 lysine 4 demethylation. *Nature* 437: 432-435.
4. Metzger, E., et al. 2005. LSD1 demethylates repressive histone marks to promote androgen-receptor-dependent transcription. *Nature* 437: 436-439.
5. Shi, Y.J., et al. 2005. Regulation of LSD1 histone demethylase activity by its associated factors. *Mol. Cell* 19: 857-864.
6. Treweek, S.C., et al. 2005. Methylation: lost in hydroxylation? *EMBO Rep.* 6: 315-320.
7. Wysocka, J., et al. 2005. Taking LSD 1 to a new high. *Cell* 122: 654-658.

CHROMOSOMAL LOCATION

Genetic locus: Kdm1a (rat) mapping to 5q36.

PRODUCT

LSD1 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 LSD1 shRNA Plasmid (r): sc-156072-SH and LSD1 shRNA (r) Lentiviral Particles: sc-156072-V as alternate gene silencing products.

For independent verification of LSD1 (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-156072A, sc-156072B and sc-156072C.

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

LSD1 siRNA (r) is recommended for the inhibition of LSD1 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

LSD1 (B-9): sc-271720 is recommended as a control antibody for monitoring of LSD1 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 LSD1 gene expression knockdown using RT-PCR Primer: LSD1 (r)-PR: sc-156072-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.