SANTA CRUZ BIOTECHNOLOGY, INC.

AOF1 siRNA (m): sc-105073

BACKGROUND

AOF1 (amine-oxidase flavin-containing domain 1), whose alternative names include KDM1B (lysine (K)-specific demethylase 1B) or LSD2 (lysine-specific histone demethylase 2), is an 823 amino acid nuclear protein belonging to the flavin monoamine oxidase family. As a histone demethylase, AOF1 specifically demethylates Lys4 of histone H3, a marker for epigenetic transcriptional activation, by interacting with a long stretch of the H3 N-terminal tail of mono- and dimethylated Lys4. Existing as three alternatively spliced isoforms, AOF1 is required for *de novo* DNA methylation of some imprinted genes in oocytes and contains a single SWIRM domain that is implicated in chromatin regulation. AOF1 contains one CW-type zinc finger and is encoded by a gene located on human chromosome 6p22.3.

REFERENCES

1. Shi, Y., et al. 2004. Histone demethylation mediated by the nuclear amine oxidase homolog LSD1. Cell 119: 941-953.
2. Culhane, J.C., et al. 2007. LSD1 and the chemistry of histone demethylation. Curr. Opin. Chem. Biol. 11: 561-568.
3. Forneris, F., et al. 2009. New roles of flavoproteins in molecular cell biology: histone demethylase LSD1 and chromatin. FEBS J. 276: 4304-4312.
4. Karytinos, A., et al. 2009. A novel mammalian flavin-dependent histone demethylase. J. Biol. Chem. 284: 17775-17782.
5. Ciccone, D.N., et al. 2009. KDM1B is a histone H3K4 demethylase required to establish maternal genomic imprints. Nature 461: 415-418.
6. Online Mendelian Inheritance in Man, OMIM™. 2009. Johns Hopkins University, Baltimore, MD. MIM Number: 613081. World Wide Web URL: http://www.ncbi.nlm.nih.gov/omim/

CHROMOSOMAL LOCATION

Genetic locus: Kdm1b (mouse) mapping to 13 A5.

PRODUCT

AOF1 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see AOF1 shRNA Plasmid (m): sc-105073-SH and AOF1 shRNA (m) Lentiviral Particles: sc-105073-V as alternate gene silencing products.

For independent verification of AOF1 (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-105073A, sc-105073B and sc-105073C.

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.

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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

AOF1 siRNA (m) is recommended for the inhibition of AOF1 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 µM in 66 µl. 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

AOF1 (E-6): sc-515565 is recommended as a control antibody for monitoring of AOF1 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 AOF1 gene expression knockdown using RT-PCR Primer: AOF1 (m)-PR: sc-105073-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.