SANTA CRUZ BIOTECHNOLOGY, INC.

JMJD2A siRNA (m): sc-62516

BACKGROUND

JMJD2A (jumonji domain containing 2A), also designated jumonji C domain-containing histone demethylation protein 3A, is a 1,064 amino acid protein encoded by the human gene JMJD2A. JMJD2A belongs to the JHDM3 histone demethylase family and contains one JmjC domain, one JmjN domain, two PHD-type zinc fingers and two Tudor domains. JMJD2A is histone demethylase that specifically demethylates Lys 9 and Lys 36 residues of Histone H3, thereby playing a central role in histone code. It does not demethylate histone H3 Lys 4, H3 Lys 27 nor H4 Lys 20, however, it will demethylate trimethylated H3 Lys 9 and H3 Lys 36 residue, while it has no activity on mono- and dimethylated residues. JMJD2A demethylation of lysine residues will generate formaldehyde and succinate. It also participates in transcriptional repression of ASCL2 and E2F-responsive promoters via the recruitment of histone deacetylases and NCOR1, respectively. JMJD2A is a ubiquitously expressed nuclear protein.

REFERENCES

1. Katoh, M., et al. 2004. Identification and characterization of JMJD2 family genes in silico. Int. J. Oncol. 24: 1623-1628.
2. Gray, S.G., et al. 2005. Functional characterization of JMJD2A, a histone deacetylase- and retinoblastoma-binding protein. J. Biol. Chem. 280: 28507-28518.
3. Zhang, D., et al. 2005. JMJD2A is a novel N-CoR-interacting protein and is involved in repression of the human transcription factor achaete scute-like homologue 2 (ASCL2/Hash2). Mol. Cell. Biol. 25: 6404-6414.
4. Huang, Y., et al. 2006. Recognition of Histone H3 Lysine 4 methylation by the double tudor domain of JMJD2A. Science 312: 748-751.
5. Whetstine, J.R., et al. 2006. Reversal of histone lysine trimethylation by the JMJD2 family of histone demethylases. Cell 125: 467-481.
6. Lee, J., et al. 2007. Distinct binding modes specify the recognition of methylated histones H3K4 and H4K20 by JMJD2A-Tudor. Nat. Struct. Mol. Biol. 15: 109-111.
7. Ng, S.S., et al. 2007. Crystal structures of histone demethylase JMJD2A reveal basis for substrate specificity. Nature 448: 87-91.

CHROMOSOMAL LOCATION

Genetic locus: Kdm4a (mouse) mapping to 4 D2.1.

PRODUCT

JMJD2A 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 JMJD2A shRNA Plasmid (m): sc-62516-SH and JMJD2A shRNA (m) Lentiviral Particles: sc-62516-V as alternate gene silencing products.

For independent verification of JMJD2A (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-62516A, sc-62516B and sc-62516C.

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

JMJD2A siRNA (m) is recommended for the inhibition of JMJD2A 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

JMJD2A (D-9): sc-271210 is recommended as a control antibody for monitoring of JMJD2A 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 JMJD2A gene expression knockdown using RT-PCR Primer: JMJD2A (m)-PR: sc-62516-PR (20 µl, 591 bp). 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.