SANTA CRUZ BIOTECHNOLOGY, INC.

H3-K9-HMTase siRNA (h): sc-62429

BACKGROUND

H3-K9-HMTase (histone-lysine N-methyltransferase SETDB2) is a 719 amino acid protein encoded by the human gene SETDB2. H3-K9-HMTase, which belongs to the histone-lysine methyltransferase family, contains one MBD (methyl-CpG-binding) domain, one pre-SET domain and one SET domain. H3-K9-HMTase is believed to be a probable histone methyltransferase with catalytic activity. Epigenetic gene silencing in eukaryotes is regulated in part by lysine methylation of the core histone proteins. While histone lysine methylation is known to control gene expression through the recruitment of modification-specific effector proteins, it remains unknown whether non-histone chromatin proteins are targets for similar modification-recognition systems. Located in the nucleus, H3-K9-HMTase is ubiquitously expressed with highest expression found in heart, testis and ovary.

REFERENCES

1. Ebbs, M.L., et al. 2005. H3 lysine 9 methylation is maintained on a transcribed inverted repeat by combined action of SUVH6 and SUVH4 methyltransferases. Mol. Cell. Biol. 25: 10507-10515.
2. Estève, P.O., et al. 2005. Functional analysis of the N- and C-terminus of mammalian G9a Histone H3 methyltransferase. Nucleic Acids Res. 33: 3211-3223.
3. Tachibana, M., et al. 2005. Histone methyltransferases G9a and GLP form heteromeric complexes and are both crucial for methylation of euchromatin at H3-K9. Genes Dev. 19: 815-826.
4. Nishijima, H., et al. 2006. Nuclear RanGAP is required for the heterochromatin assembly and is reciprocally regulated by Histone H3 and Clr4 histone methyltransferase in *Schizosaccharomyces pombe*. Mol. Biol. Cell 17: 2524-2536.
5. Henkels, C.H. and Khorasanizadeh, S. 2007. Implications of a histone code mimic in epigenetic signaling. Mol. Cell 27: 521-522.
6. Tzeng, T.Y., et al. 2007. Epigenetic regulation of the *Drosophila* chromosome 4 by the histone H3K9 methyltransferase dSETDB1. Proc. Natl. Acad. Sci. USA 104: 12691-12696.

CHROMOSOMAL LOCATION

Genetic locus: SETDB2 (human) mapping to 13q14.2.

PRODUCT

H3-K9-HMTase siRNA (h) 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 H3-K9-HMTase shRNA Plasmid (h): sc-62429-SH and H3-K9-HMTase shRNA (h) Lentiviral Particles: sc-62429-V as alternate gene silencing products.

For independent verification of H3-K9-HMTase (h) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-62429A, sc-62429B and sc-62429C.

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

H3-K9-HMTase siRNA (h) is recommended for the inhibition of H3-K9-HMTase expression in human 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

H3-K9-HMTase (LD87): sc-130475 is recommended as a control antibody for monitoring of H3-K9-HMTase 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 H3-K9-HMTase gene expression knockdown using RT-PCR Primer: H3-K9-HMTase (h)-PR: sc-62429-PR (20 µl, 550 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Ea, C.K., et al. 2012. EHMT1 protein binds to nuclear factor-κB p50 and represses gene expression. J. Biol. Chem. 287: 31207-31217.

RESEARCH USE

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