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HMG-I/HMG-Y siRNA (h2): sc-44333

BACKGROUND

High mobility group (HMG) I (Y) chromatin proteins bind to the minor groove of AT-rich DNA sequences with high affinity. Evidence suggests that the binding of HMG proteins to DNA induces alterations in the DNA architecture including DNA bending and unwinding of the helix. HMG proteins synergize with Oct-2, members of the NF κ B family, ATF-2 and c-Jun to activate transcription. Other studies indicate that phosphorylation of HMG protein is required to stimulate the transcriptional activity of the protein. Human HMG I (Y) contains two DNA-binding domains, termed HMG boxes. HMG proteins bind single-stranded DNA but induce conformational changes in double-stranded DNA alone.

REFERENCES

1. Wen, L., et al. 1989. A human placental cDNA clone that encodes nonhistone chromosomal protein HMG-1. *Nucleic Acids Res.* 17: 1197-1214.
2. Bustin, M., et al. 1990. Structural features of the HMG chromosomal proteins and their genes. *Biochim. Biophys. Acta* 1049: 231-243.
3. Shirakawa, H., et al. 1992. Structure of a gene coding for human HMG2 protein. *J. Biol. Chem.* 267: 6641-6635.
4. Nissen, M.S., et al. 1995. Changes in superhelicity are introduced into closed circular DNA by binding of high mobility group protein I/Y. *J. Biol. Chem.* 270: 4355-4360.
5. Wang, D.Z., et al. 1995. Interleukin 4-inducible phosphorylation of HMG-I(Y) is inhibited by rapamycin. *J. Biol. Chem.* 270: 22924-22932.

CHROMOSOMAL LOCATION

Genetic locus: HMGA1 (human) mapping to 6p21.31.

PRODUCT

HMG-I/HMG-Y siRNA (h2) 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 HMG-I/HMG-Y shRNA Plasmid (h2): sc-44333-SH and HMG-I/HMG-Y shRNA (h2) Lentiviral Particles: sc-44333-V as alternate gene silencing products.

For independent verification of HMG-I/HMG-Y (h2) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-44333A, sc-44333B and sc-44333C.

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

HMG-I/HMG-Y siRNA (h2) is recommended for the inhibition of HMG-I/HMG-Y 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 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

HMG-I/HMG-Y (D-12): sc-393213 is recommended as a control antibody for monitoring of HMG-I/HMG-Y 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 HMG-I/HMG-Y gene expression knockdown using RT-PCR Primer: HMG-I/HMG-Y (h2)-PR: sc-44333-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Saayman, S.M., et al. 2016. Long non-coding RNA BGas regulates the cystic fibrosis transmembrane conductance regulator. *Mol. Ther.* 24: 1351-1357.

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.