

MBD6 siRNA (h): sc-96201

BACKGROUND

Methylation of DNA contributes to the regulation of gene transcription in both mammalian and invertebrate systems. DNA methylation requires the enzymatic activity of DNA methyltransferase and predominates on cytosine residues that are present in dinucleotide motifs consisting of a 5' cytosine followed by guanosine (CpG), which results in transcriptional repression of the methylated gene. Several proteins have been identified that associate with the methyl-CpG sites, and they include methyl-CpG binding protein-1 (MBD1), MBD2, MBD3, MBD4 and MeCP2. Unlike other MBD proteins, MBD6 does not interact with methylated or unmethylated DNA, but does bind to heterochromatin. MBD6 contains one MBD domain and is encoded by a gene that maps to human chromosome 12q13.3.

REFERENCES

1. Wade, P.A. 2001. Methyl CpG-binding proteins and transcriptional repression. *Bioessays* 23: 1131-1137.
2. Nakao, M., et al. 2001. Regulation of transcription and chromatin by methyl-CpG binding protein MBD1. *Brain Dev.* 23 Suppl. 1: S174-S176.
3. Ballestar, E. and Wolffe, A.P. 2001. Methyl-CpG-binding proteins. Targeting specific gene repression. *Eur. J. Biochem.* 268: 1-6.
4. Roloff, T.C., et al. 2003. Comparative study of methyl-CpG-binding domain proteins. *BMC Genomics* 4: 1.
5. Laget, S., et al. 2010. The human proteins MBD5 and MBD6 associate with heterochromatin but they do not bind methylated DNA. *PLoS ONE* 5: e11982.
6. Defossez, P.A. and Stancheva, I. 2011. Biological functions of methyl-CpG-binding proteins. *Prog. Mol. Biol. Transl. Sci.* 101: 377-398.

CHROMOSOMAL LOCATION

Genetic locus: MBD6 (human) mapping to 12q13.3.

PRODUCT

MBD6 siRNA (h) is a pool of 2 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 MBD6 shRNA Plasmid (h): sc-96201-SH and MBD6 shRNA (h) Lentiviral Particles: sc-96201-V as alternate gene silencing products.

For independent verification of MBD6 (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-96201A and sc-96201B.

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 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

MBD6 siRNA (h) is recommended for the inhibition of MBD6 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor MBD6 gene expression knockdown using RT-PCR Primer: MBD6 (h)-PR: sc-96201-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.