

MCM10 siRNA (h): sc-61000

BACKGROUND

The mini-chromosome maintenance (MCM) family of proteins include MCM2, MCM3, MCM4 (Cdc21), MCM5 (Cdc46), MCM6 (Mis5), MCM7 (Cdc47), MCM8, MCM9 and MCM10 (Dna43). The proteins in this family are regulators of DNA replication that act to ensure replication occurs only once in the cell cycle. Expression of MCM proteins increases during cell growth, peaking at G₁ to S phase. MCM proteins are crucial components of the pre-replication complex (pre-RC) and are involved in replication fork formation and the recruitment of other DNA replication proteins. The MCM proteins each contain an ATP-binding motif, which is predicted to mediate ATP-dependent opening of double-stranded DNA. MCM proteins are regulated by E2F transcription factors, which induce MCM expression, and by protein kinases, which interact with MCM proteins to maintain the postreplicative state of the cell. MCM10 interacts with replication factors and can interact with MCM2 and MCM6.

REFERENCES

1. Koonin, E.V. 1993. A common set of conserved motifs in a vast variety of putative nucleic acid-dependent ATPases including MCM proteins involved in the initiation of eukaryotic DNA replication. *Nucleic Acids Res.* 21: 2541-2547.
2. Fujita, M., et al. 1998. Cell cycle- and chromatin binding state-dependent phosphorylation of human MCM heterohexameric complexes. A role for Cdc2 kinase. *J. Biol. Chem.* 273: 17095-17101.
3. Leone, G., et al. 1998. E2F3 activity is regulated during the cell cycle and is required for the induction of S phase. *Genes Dev.* 12: 2120-2130.
4. Coverley, D., et al. 1998. Protein kinase inhibition in G₂ causes mammalian MCM proteins to reassociate with chromatin and restores ability to replicate. *Exp. Cell Res.* 238: 63-69.
5. Ricke, R.M. and Bielinsky, A.K. 2004. MCM10 regulates the stability and chromatin association of DNA polymerase- α . *Mol. Cell* 16: 173-185.
6. Yoshida, K. and Inoue, I. 2004. Expression of MCM10 and TopBP1 is regulated by cell proliferation and UV irradiation via the E2F transcription factor. *Oncogene* 23: 6250-6260.
7. Izumi, M., et al. 2004. Localization of human MCM10 is spatially and temporally regulated during the S phase. *J. Biol. Chem.* 279: 32569-32577.

CHROMOSOMAL LOCATION

Genetic locus: MCM10 (human) mapping to 10p13.

PRODUCT

MCM10 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see MCM10 shRNA Plasmid (h): sc-61000-SH and MCM10 shRNA (h) Lentiviral Particles: sc-61000-V as alternate gene silencing products.

For independent verification of MCM10 (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-61000A, sc-61000B and sc-61000C.

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

MCM10 siRNA (h) is recommended for the inhibition of MCM10 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 MCM10 gene expression knockdown using RT-PCR Primer: MCM10 (h)-PR: sc-61000-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. Romani, B., et al. 2015. HIV-1 Vpr protein enhances proteasomal degradation of MCM10 DNA replication factor through the Cul4-DDB1[VprBP] E3 ubiquitin ligase to induce G₂/M cell cycle arrest. *J. Biol. Chem.* 290: 17380-17389.

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.