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MTF-1 shRNA (m) Lentiviral Particles: sc-44354-V

BACKGROUND

The metal-responsive element (MRE)-binding transcription factor (MTF-1) stimulates the expression of metallothioneins in response to the exposure of cells to heavy metals. MTF-1 contains six zinc fingers in the DNA binding domain. The phosphorylation of MTF-1 in response to metal exposure appears to play a significant role in the ability of MTF-1 to activate metallothionein transcription. In addition to its role in metallothionein activation, MTF-1 is involved in a post-transcription regulatory complex for ribosomal protein S25. MTF-1, La and p53 inhibit the nuclear export of S25 mRNA in response to nutrient deprivation. Furthermore, MTF-1 acts as a chromatin insulator on integrated transgenes in cultured cells to insulate active loci against chromatin silencing.

REFERENCES

- Westin, G., et al. 1988. A zinc-responsive factor interacts with a metal-regulated enhancer element (MRE) of the mouse metallothionein-I gene. *EMBO J.* 7: 3763-3770.
- Radtke, F., et al. 1993. Cloned transcription factor MTF-1 activates the mouse metallothionein I promoter. *EMBO J.* 12: 1355-1362.
- Brugnera, E., et al. 1994. Cloning, chromosomal mapping and characterization of the human metal-regulatory transcription factor MTF-1. *Nucleic Acids Res.* 22: 3167-3173.
- Saydam, N., et al. 2002. Regulation of metallothionein transcription by the metal-responsive transcription factor MTF-1: identification of signal transduction cascades that control metal-inducible transcription. *J. Biol. Chem.* 277: 20438-20445.
- Adilakshmi, T., et al. 2002. Ribosomal protein S25 mRNA partners with MTF-1 and La to provide a p53-mediated mechanism for survival or death. *J. Biol. Chem.* 277: 4147-4151.

CHROMOSOMAL LOCATION

Genetic locus: Mtf1 (mouse) mapping to 4 D2.2.

PRODUCT

MTF-1 shRNA (m) Lentiviral Particles is a pool of concentrated, transduction-ready viral particles containing 3 target-specific constructs that encode 19-25 nt (plus hairpin) shRNA designed to knock down gene expression. Each vial contains 200 μ l frozen stock containing 1.0×10^6 infectious units of virus (IFU) in Dulbecco's Modified Eagle's Medium with 25 mM HEPES pH 7.3. Suitable for 10-20 transductions. Also see MTF-1 siRNA (m): sc-44354 and MTF-1 shRNA Plasmid (m): sc-44354-SH as alternate gene silencing products.

STORAGE

Store lentiviral particles at -80° C. Stable for at least one year from the date of shipment. Once thawed, particles can be stored at 4° C for up to one week. Avoid repeated freeze thaw cycles.

PROTOCOLS

See our web site at www.scbt.com or our catalog for detailed protocols and support products.

APPLICATIONS

MTF-1 shRNA (m) Lentiviral Particles is recommended for the inhibition of MTF-1 expression in mouse cells.

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 μ l frozen viral stock containing 1.0×10^6 infectious units of virus (IFU); contains an shRNA construct encoding a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA.

GENE EXPRESSION MONITORING

MTF-1 (H-300): sc-48775 is recommended as a control antibody for monitoring of MTF-1 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 (secondary) reagents are recommended: 1) Western Blotting: use goat anti-rabbit IgG-HRP: sc-2004 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible goat anti-rabbit IgG-HRP: sc-2030 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use goat anti-rabbit IgG-FITC: sc-2012 (dilution range: 1:100-1:400) or goat anti-rabbit IgG-TR: sc-2780 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor MTF-1 gene expression knockdown using RT-PCR Primer: MTF-1 (m)-PR: sc-44354-PR (20 μ l). Annealing temperature for the primers should be $55-60^\circ$ C and the extension temperature should be $68-72^\circ$ C.

BIOSAFETY

Lentiviral particles can be employed in standard Biosafety Level 2 tissue culture facilities (and should be treated with the same level of caution as with any other potentially infectious reagent). Lentiviral particles are replication-incompetent and are designed to self-inactivate after transduction and integration of shRNA constructs into genomic DNA of target cells.

RESEARCH USE

The purchase of this product conveys to the buyer the nontransferable right to use the purchased amount of the product and all replicates and derivatives for research purposes conducted by the buyer in his laboratory only (whether the buyer is an academic or for-profit entity). The buyer cannot sell or otherwise transfer (a) this product (b) its components or (c) materials made using this product or its components to a third party, or otherwise use this product or its components or materials made using this product or its components for Commercial Purposes.