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FOXM1 shRNA (h) Lentiviral Particles: sc-43769-V

BACKGROUND

The Fox family of transcription factors is a large group of proteins that share a common DNA binding domain termed a winged-helix or forkhead domain. FOXM1, also known as FKHL16, MPP2 or TRIDENT, is primarily expressed in proliferating cells. The gene encoding human FOXM1 maps to chromosome 12p13.33. The transcription element that restricts FOXM1 expression to proliferating cells is located 300 bp upstream of the start codon. FOXM1 is most abundant in thymus, testis, small intestine and colon. Alternative splicing generates FOXM1A and FOXM1B isoforms that contain PEST regions involved in rapid protein degradation. A decrease in FOXM1 expression is associated with age-related defects in cellular proliferation. Conversely, an increase in FOXM1B expression in the livers of older transgenic mice restore hepatocyte DNA replication rates to the higher rate present in young livers. FOXM1B activates the transcription of cyclin B1, cyclin D1 and Cdc25B.

REFERENCES

1. Ye, H., et al. 1997. Hepatocyte nuclear factor 3/forkhead homolog 11 is expressed in proliferating epithelial and mesenchymal cells of embryonic and adult tissues. *Mol. Cell. Biol.* 17: 1626-1641.
2. Korver, W., et al. 1997. The human TRIDENT/HFH-11/FKHL16 gene: structure, localization and promoter characterization. *Genomics* 46: 435-442.
3. Yao, K.M., et al. 1997. Molecular analysis of a novel winged-helix protein, WIN. Expression pattern, DNA binding property and alternative splicing within the DNA binding domain. *J. Biol. Chem.* 272: 19827-19836.
4. Ly, D.H., et al. 2000. Mitotic misregulation and human aging. *Science* 287: 2486-2492.
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CHROMOSOMAL LOCATION

Genetic locus: FOXM1 (human) mapping to 12p13.33.

PRODUCT

FOXM1 shRNA (h) 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 FOXM1 siRNA (h): sc-43769 and FOXM1 shRNA Plasmid (h): sc-43769-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

FOXM1 shRNA (h) Lentiviral Particles is recommended for the inhibition of FOXM1 expression in human 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

FOXM1 (G-5): sc-376471 is recommended as a control antibody for monitoring of FOXM1 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-mouse IgG-HRP: sc-2005 (dilution range: 1:2000-1:32,000) or Cruz Marker™ compatible goat anti-mouse IgG-HRP: sc-2031 (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-mouse IgG-FITC: sc-2010 (dilution range: 1:100-1:400) or goat anti-mouse IgG-TR: sc-2781 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor FOXM1 gene expression knockdown using RT-PCR Primer: FOXM1 (h)-PR: sc-43769-PR (20 μ l, 491 bp). 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.