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Diagnostik & molekulare Diagnostik



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IFITM1 shRNA (h) Lentiviral Particles: sc-44549-V



The Power to Overtion

BACKGROUND

Interferons (IFNs) are potential antitumor agents, as they exhibit antiproliferative and differentiating properties, in addition to functioning in the defense against microbial infections. IFN exposure induces the regulation of expression levels of cellular proteins that mediate the pleiotropic effects of interferons. These effects may be mediated by soluble factors or by cell-cell interactions involving specific membrane proteins. The IFITM family of proteins are transmembrane proteins so named because their expression is IFN-inducible. IFITM proteins have been found upregulated in human colorectal carcinomas. Both mouse IFITM1 (also known as CD225) and IFITM3 demonstrate expression on the cell surfaces of primordial germ cells in a developmentally-regulated manner. They presumably modulate cell adhesion and influence cell differentiation. IFITM1 activity is required for primordial germ cell transit, and IFITM1 acts as a repulsive molecule by repelling non-IFITM1-expressing primordial germ cells from the mesoderm into the endoderm.

REFERENCES

- 1. Reid, L.E., et al. 1989. A single DNA response element can confer inducibility by both α and γ -interferons. Proc. Natl. Acad. Sci. USA 86: 840-844.
- Deblandre, G.A., et al. 1995. Expression cloning of an interferon-inducible 17 kDa membrane protein implicated in the control of cell growth. J. Biol. Chem. 270: 23860-23866.
- Perry, D.J., et al. 1999. Cloning of interferon-stimulated gene 17: the promoter and nuclear proteins that regulate transcription. Mol. Endocrinol. 13: 1197-1206.
- 4. Saitou, M., et al. 2002. A molecular programme for the specification of germ cell fate in mice. Nature 418: 293-300.

CHROMOSOMAL LOCATION

Genetic locus: IFITM1 (human) mapping to 11p15.5.

PRODUCT

IFITM1 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 μl frozen stock containing 1.0 x 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 IFITM1 siRNA (h): sc-44549 and IFITM1 shRNA Plasmid (h): sc-44549-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

IFITM1 shRNA (h) Lentiviral Particles is recommended for the inhibition of IFITM1 expression in human cells.

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 μ l frozen viral stock containing 1.0 x 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

IFITM1 (P-17): sc-34171 is recommended as a control antibody for monitoring of IFITM1 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 donkey anti-goat IgG-HRP: sc-2020 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible donkey anti-goat IgG-HRP: sc-2033 (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 donkey anti-goat IgG-FITC: sc-2024 (dilution range: 1:100-1:400) or donkey anti-goat IgG-TR: sc-2783 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor IFITM1 gene expression knockdown using RT-PCR Primer: IFITM1 (h)-PR: sc-44549-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° 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.

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