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Mucin 16 shRNA (h) Lentiviral Particles: sc-44971-V

BACKGROUND

The mucins are a family of highly glycosylated, secreted proteins with a basic structure consisting of a variable number of tandem repeats (VNTRs). Membrane-associated and secretory Mucins are high molecular weight glycoproteins of the glycocalyx (polysaccharide biofilm) that protects mucosal epithelium from particulate matter and microorganisms. Epithelial mucins are large, secreted and cell surface glycoproteins crucial for adhesion modulation, signaling and epithelial cell protection. The number of repeats is highly polymorphic and varies among different alleles. The mucin family consists of Mucins 1-4, Mucin 5 (AC and B), Mucins 6-8, Mucins 11-13 and Mucins 15-17. The Mucin 16 protein (also commonly referred to as CA125), encoded by the gene MUC16, is a very high molecular weight tumor antigen consisting of three domains: a carboxy-terminal domain, an extracellular domain and an amino-terminal domain. Mucin 16, an ovarian cancer-associated antigen, is used as a marker to monitor the progress of epithelial ovarian cancer. It is a hydrophilic membrane-associated protein that may be involved in vitamin A functions.

REFERENCES

1. Yin, B.W., et al. 2001. Molecular cloning of the CA125 ovarian cancer antigen: identification as a new mucin, MUC16. *J. Biol. Chem.* 276: 27371-27375.
2. Maeda, T., et al. 2004. Solution structure of the SEA domain from the murine homologue of ovarian cancer antigen CA125 (MUC16). *J. Biol. Chem.* 279: 13174-13182.
3. Hori, Y., et al. 2005. Effect of retinoic acid on gene expression in human conjunctival epithelium: secretory phospholipase A2 mediates retinoic acid induction of Mucin 16. *Invest. Ophthalmol. Vis. Sci.* 46: 4050-4061.

CHROMOSOMAL LOCATION

Genetic locus: MUC16 (human) mapping to 19p13.2.

PRODUCT

Mucin 16 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 Mucin 16 siRNA (h): sc-44971 and Mucin 16 shRNA Plasmid (h): sc-44971-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

Mucin 16 shRNA (h) Lentiviral Particles is recommended for the inhibition of Mucin 16 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

Mucin 16 (C-6): sc-365002 is recommended as a control antibody for monitoring of Mucin 16 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 Mucin 16 gene expression knockdown using RT-PCR Primer: Mucin 16 (h)-PR: sc-44971-PR (20 μ l, 586 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.