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Cytokeratin 20 shRNA (h) Lentiviral Particles: sc-43313-V



The Power to Question

BACKGROUND

Cytokeratins comprise a diverse group of intermediate filament proteins (IFPs) that are expressed as pairs in both keratinized and non-keratinized epithelial tissue, where they constitute up to 85% of mature keratinocytes in the vertebrate epidermis. Cytokeratins play a critical role in differentiation and tissue specialization and function to maintain the overall structural integrity of epithelial cells. The α -helical coiled-coil dimers associate laterally end-to-end to form 10 nm diameter filaments. Cytokeratins are useful markers of tissue differentiation, and in addition, they aid in the characterization of malignant tumors. Cytokeratin 20 is abundantly expressed in goblet cells and enterocytes of the gastrointestinal tract, and Cytokeratin 20 is a useful marker of pancreatic and colorectal cancer. Cytokeratin 20 is also helpful in distinguishing different types of highly related carcinomas, such as renal oncocytomas from renal cell carcinomas.

REFERENCES

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- Moll, R., et al. 1993. The human gene encoding cytokeratin 20 and its expression during fetal development and in gastrointestinal carcinomas. Differentiation 53: 75-93.
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- Marceau, N., et al. 1995. Cytokeratin expression, fibrillar organization and subtle function in liver cells. Biochem. Cell Biol. 73: 619-625.
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CHROMOSOMAL LOCATION

Genetic locus: KRT20 (human) mapping to 17q21.2.

PRODUCT

Cytokeratin 20 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 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 Cytokeratin 20 siRNA (h): sc-43313 and Cytokeratin 20 shRNA Plasmid (h): sc-43313-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

Cytokeratin 20 shRNA (h) Lentiviral Particles is recommended for the inhibition of Cytokeratin 20 expression in human cells.

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 µl frozen viral stock containing 1.0 x 10⁶ 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

Cytokeratin 20 (E-9): sc-271183 is recommended as a control antibody for monitoring of Cytokeratin 20 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) Immunofluo-rescence: 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 Cytokeratin 20 gene expression knockdown using RT-PCR Primer: Cytokeratin 20 (h)-PR: sc-43313-PR (20 μ I). 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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