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DC-SIGN shRNA (h) Lentiviral Particles: sc-43719-V

BACKGROUND

Dendritic cells (DCs) are antigen-presenting immune system cells that are present on peripheral mucosal tissues and migrate to lymphoid tissues. DC-SIGN (DC-specific ICAM-3 grabbing nonintegrin) is a type II membrane protein that is exclusively expressed by DCs. DC-SIGN, also designated CD209, binds to ICAM-3 to mediate the initial interaction between DCs and resting T cells through the immunological synapse. The DCs that are present in the initial sites of HIV-1 infection capture HIV-1 through DC-SIGN, which then facilitates the migration of DCs to areas of T cell-rich secondary lymphoid organs, where it promotes efficient *trans* HIV-1 infection of those T cells. DC-SIGN functions to transport HIV-1 from exposed mucosal surfaces to a lymphoid compartment.

REFERENCES

1. Curtis, B.M., et al. 1992. Sequence and expression of a membrane-associated C-type lectin that exhibits its CD2-independent binding of human immunodeficiency virus envelope glycoprotein gp120. *Proc. Natl. Acad. Sci. USA* 89: 8356-8360.
2. Steinman, R.M. 2000. DC-SIGN: a guide to some mysteries of dendritic cells. *Cell* 100: 491-494.
3. Geijtenbeek, T.B.H., et al. 2000. Identification of DC-SIGN, a novel dendritic cell-specific ICAM-3 receptor that supports primary immune responses. *Cell* 100: 575-585.
4. Geijtenbeek, T.B.H., et al. 2000. DC-SIGN, a dendritic cell-specific HIV-1-binding protein that enhances *trans* infection of T cells. *Cell* 100: 587-597.
5. Cohen, J. 2000. AIDS research. Novel protein delivers HIV to target cells. *Science* 287: 1567.
6. Steinberg, D. 2000. Receptor boosts HIV infection. *The Scientist* 14: 12

CHROMOSOMAL LOCATION

Genetic locus: CD209 (human) mapping to 19p13.2.

PRODUCT

DC-SIGN 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 DC-SIGN siRNA (h): sc-43719 and DC-SIGN shRNA Plasmid (h): sc-43719-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

DC-SIGN shRNA (h) Lentiviral Particles is recommended for the inhibition of DC-SIGN 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

DC-SIGN (C-20): sc-11038 is recommended as a control antibody for monitoring of DC-SIGN 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 DC-SIGN gene expression knockdown using RT-PCR Primer: DC-SIGN (h)-PR: sc-43719-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.