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SZABO-SCANDIC HandelsgmbH

Quellenstraße 110, A-1100 Wien

T. +43(0)1 489 3961-0

F. +43(0)1 489 3961-7

mail@szabo-scandic.com

www.szabo-scandic.com

[linkedin.com/company/szaboscandic](https://www.linkedin.com/company/szaboscandic) 



DNAM-1 shRNA (m) Lentiviral Particles: sc-45776-V

BACKGROUND

The T cell antigen receptor (TCR) recognizes foreign antigens and translates such recognition events into intracellular signals that elicit a change in the cell from a dormant to an activated state. Additional proteins termed "accessory molecules" are also required for activation and for cytotoxic T lymphocyte (CTL)-mediated cytotoxicity. For instance, CD2, CD4, CD8, LFA-1, and CD28 are examples of well characterized accessory molecules. An accessory molecule designated DNAX accessory molecule-1 or DNAM-1 has been described. DNAM-1 is a transmembrane glycoprotein that is 318 amino acids in length and contains two immunoglobulin-like domains. DNAM-1 is expressed on both T cells and natural killer (NK) cells and participates in primary adhesion during CTL-mediated cytotoxicity.

REFERENCES

- Arthos, J., et al. 1989. Identification of the residues in human CD4 critical for the binding of HIV. *Cell* 57: 469-481.
- Weiss, A., et al. 1991. Signal transduction by the T cell antigen receptor. *Semin. Immunol.* 3: 313-324.
- Allison, J.P., et al. 1991. The immunobiology of T cells with invariant gamma delta antigen receptors. *Annu. Rev. Immunol.* 9: 679-705.
- Ehrlich, E.W., et al. 1993. T cell receptor interaction with peptide/major histocompatibility complex (MHC) and superantigen/MHC ligands is dominated by antigen. *J. Exp. Med.* 178: 713-722.
- Julius, M., et al. 1993. Distinct roles for CD4 and CD8 as co-receptors in antigen receptor signalling. *Immunol. Today* 14: 177-183.
- Vignali, D.A. 1994. The interaction between CD4 and MHC class II molecules and its effect on T cell function. *Behring Inst. Mitt.* 94: 133-147.

CHROMOSOMAL LOCATION

Genetic locus: Cd226 (mouse) mapping to 18 E4.

PRODUCT

DNAM-1 shRNA (m) 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 DNAM-1 siRNA (m): sc-45776 and DNAM-1 shRNA Plasmid (m): sc-45776-SH as alternate gene silencing products.

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.

APPLICATIONS

DNAM-1 shRNA (m) Lentiviral Particles is recommended for the inhibition of DNAM-1 expression in mouse 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

DNAM-1 (M-13): sc-31040 is recommended as a control antibody for monitoring of DNAM-1 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 DNAM-1 gene expression knockdown using RT-PCR Primer: DNAM-1 (m)-PR: sc-45776-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.

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.