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MIP-1 α shRNA (m) Lentiviral Particles: sc-44722-V

BACKGROUND

Chemokines are members of a superfamily of small inducible, secreted, pro-inflammatory cytokines. Members of the chemokine family exhibit 20 to 50% homology in their predicted amino acid sequences and are divided into four subfamilies. In C-C (or β) subfamily, the first two cysteines are adjacent. C-C chemokines are chemoattractants and activators for monocytes and T cells. C-C subfamily members include macrophage inflammatory protein (MIP)-1 α , MIP-1 β , MIP-2, MIP-3 α , MIP-3 β , MIP-4, HCC-1, MIP-5 (or HCC-2), RANTES, MCP-1/2/3 (and the murine homologs JE and MARC), I-309, murine C10 and TCA3. Research has shown that MIP-1 β is more selective than MIP-1 α , primarily attracting CD4⁺ T lymphocytes, with a preference for T cells of the naive phenotype. MIP-1 α is a more potent lymphocyte chemoattractant than MIP-1 β and exhibits a broader range of chemoattractant specificities. It has been suggested that CD8⁺ T lymphocytes are involved in the control of HIV infection *in vivo* by the release of HIV-suppressive factors (HIV-SF). MIP-1 α has been identified as one of the major HIV-SFs produced by CD8⁺ T cells, along with MIP-1 β and RANTES. Recombinant human MIP-1 α acts as an inhibitor of different strains of HIV-1, HIV-2 and SIV infection in a dose-dependent manner.

REFERENCES

1. Zipfel, P.F., et al. 1989. Mitogenic activation of human T cells induces two closely related genes which share structural similarities with a new family of secreted factors. *J. Immunol.* 142: 1582-1590.
2. Widmer, U., et al. 1993. Genomic cloning and promoter analysis of macrophage inflammatory protein (MIP)-2, MIP-1 α and MIP-1 β , members of the chemokine superfamily of proinflammatory cytokines. *J. Immunol.* 150: 4996-5012.
3. Schall, T.J., et al. 1993. Human macrophage inflammatory protein α (MIP-1 α) and MIP-1 β chemokines attract distinct populations of lymphocytes. *J. Exp. Med.* 177: 1821-1826.

CHROMOSOMAL LOCATION

Genetic locus: Ccl3 (mouse) mapping to 11 C.

PRODUCT

MIP-1 α 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 x 10⁶ 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 MIP-1 α siRNA (m): sc-44722 and MIP-1 α shRNA Plasmid (m): sc-44722-SH as alternate gene silencing products.

STORAGE

Store lentiviral particles at -80 $^{\circ}$ C. Stable for at least one year from the date of shipment. Once thawed, particles can be stored at 4 $^{\circ}$ 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

MIP-1 α shRNA (m) Lentiviral Particles is recommended for the inhibition of MIP-1 α expression in mouse cells.

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 μ l 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

MIP-1 α (C-5): sc-365691 is recommended as a control antibody for monitoring of MIP-1 α 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 MarkerTM compatible goat anti-mouse IgG-HRP: sc-2031 (dilution range: 1:2000-1:5000), Cruz MarkerTM 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 UltraCruzTM Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor MIP-1 α gene expression knockdown using RT-PCR Primer: MIP-1 α (m)-PR: sc-44722-PR (20 μ l, 497 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.