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LRF shRNA (h) Lentiviral Particles: sc-44574-V

BACKGROUND

LRF, formerly identified as Pokemon, is a poxvirus zinc finger (POZ) domain-containing transcription factor that influences cell differentiation. LRF (for leukemia/lymphoma related factor) is also known as zinc finger and BTB domain containing 7A, ZBTB7, TIP21, FBI1 and FBI-1. POZ-domain transcription factors contain a POZ or BTB type protein-protein interaction domain at their N-terminus and Krüppel-type zinc fingers at their C-terminus. LRF is inducible during both murine and human preadipocyte differentiation and may contribute to adipogenesis through influencing the switch from cellular proliferation to terminal differentiation. LRF can associate with active chromatin and stimulate TAT-activated HIV-1 transcription through interactions with the HIV-1 long terminal repeat. 3T3L1 cells stably overexpressing LRF show a reduction in DNA synthesis and in expression of cyclin A, cyclin-dependent kinase 2 and p107.

REFERENCES

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2. Kukita, A., et al. 1999. Osteoclast-derived zinc finger (OCZF) protein with POZ domain, a possible transcriptional repressor, is involved in osteoclastogenesis. *Blood* 94: 1987-1997.
3. Pendergrast, P.S., et al. 2002. FBI-1 can stimulate HIV-1 TAT activity and is targeted to a novel subnuclear domain that includes the TAT-P-TEFb-containing nuclear speckles. *Mol. Biol. Cell* 13: 915-929.
4. Lee, D.K., et al. 2002. POZ domain transcription factor, FBI-1, represses transcription of ADH5/FDH by interacting with the zinc finger and interfering with DNA binding activity of Sp1. *J. Biol. Chem.* 277: 26761-26768.
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6. Laudes, M., et al. 2004. Role of the POZ zinc finger transcription factor FBI-1 in human and murine adipogenesis. *J. Biol. Chem.* 279: 11711-11718.
7. Maeda, T., et al. 2005. Role of the proto-oncogene Pokemon in cellular transformation and ARF repression. *Nature* 433: 278-285.

CHROMOSOMAL LOCATION

Genetic locus: ZBTB7A (human) mapping to 19p13.3.

PRODUCT

LRF 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 LRF siRNA (h): sc-44574 and LRF shRNA Plasmid (h): sc-44574-SH as alternate gene silencing products.

APPLICATIONS

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

LRF (H-6): sc-393012 is recommended as a control antibody for monitoring of LRF 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 LRF gene expression knockdown using RT-PCR Primer: LRF (h)-PR: sc-44574-PR (20 μ l, 449 bp). 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.

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.