

Produktinformation



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Diagnostik & molekulare Diagnostik



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fractalkine shRNA (h) Lentiviral Particles: sc-43771-V



The Power to Question

BACKGROUND

Chemokines are members of a superfamily of 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 the subfamily designated C-C or β , the first two cysteines are adjacent. In the C-X-C or α subfamily, the first two of four cysteine residues are separated by a single amino acid. C subfamily members, also designated γ chemokines, lack the first and third cysteine residues of the conserved motif. Chemokines in these three subfamilies are small, secreted proteins. Fractalkine, also designated neurotactin, is the first characterized member of a fourth chemokine subfamily. Fractalkine contains a novel C-X3-C motif in which the first two cysteines are separated by three amino acid residues. Fractalkine mRNA has been detected in brain and heart and is upregulated in microglia and endothelial cells by inflammatory signals. The protein exists both as a membrane-bound form and as a chemotactic soluble form.

REFERENCES

- 1. Oppenheim, J.J., et al. 1991. Properties of the novel proinflammatory supergene "intercrine" cytokine family. Ann. Rev. Immunol. 9: 617-648.
- Miller, M.D., et al. 1992. Biology and biochemistry of the chemokines: a family of chemotactic and inflammatory cytokines. Crit. Rev. Immunol. 12: 17-46.
- 3. Taub, D.D., et al. 1993. Review of the chemokine meeting of the Third International Symposium of Chemotactic Cytokines. Cytokine 5: 175-179.
- Schall, T.J., et al. 1994. Chemokines, leukocyte trafficking, and inflammation. Curr. Opin. Immunol. 6: 865-873.
- 5. Taub, D.D., et al. 1996. β chemokines costimulate lymphocyte cytolysis, proliferation, and lymphokine production. J. Leuk. Biol. 59: 53-60.

CHROMOSOMAL LOCATION

Genetic locus: CX3CL1 (human) mapping to 16q21.

PRODUCT

fractalkine 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 μl 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 fractalkine siRNA (h): sc-43771 and fractalkine shRNA Plasmid (h): sc-43771-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

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

SUPPORT REAGENTS

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

fractalkine (A-9): sc-166200 is recommended as a control antibody for monitoring of fractalkine 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) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor fractalkine gene expression knockdown using RT-PCR Primer: fractalkine (h)-PR: sc-43771-PR (20 μ l, 575 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.

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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