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Fgl2 shRNA (h) Lentiviral Particles: sc-44691-V

BACKGROUND

Fibrinogen-like protein 2 (Fgl2), also known as fibroleukin, is secreted by T cells and is involved in diseases in which thrombosis plays a pivotal role, such as virus-induced fulminant hepatitis, fetal loss syndrome, and xenograft rejection. Constitutively expressed in cytotoxic T-cells, Fgl2 exerts immunosuppressive effects on both T cell proliferation and dendritic cell maturation. Fgl2 is a serine protease and directly cleaves prothrombin to thrombin. Fgl2 functions in the pathogenesis of diseases including viral-induced hepatitis and Th1 cytokine-induced fetal loss syndrome.

REFERENCES

1. Ning, Q., et al. 1999. The nucleocapsid protein of murine hepatitis virus type 3 induces transcription of the novel fgl2 prothrombinase gene. *J. Biol. Chem.* 274: 9930-9936.
2. Yuwaraj S., et al. 2001. Genomic characterization, localization, and functional expression of FGL2, the human gene encoding fibroleukin: a novel human procoagulant. *Genomics* 71: 330-338.
3. Chan C.W., et al. 2003. Soluble fibrinogen-like protein 2/fibroleukin exhibits immunosuppressive properties: suppressing T cell proliferation and inhibiting maturation of bone marrow-derived dendritic cells. *J. Immunol.* 170: 4036-4044.
4. Ning, Q., et al. 2003. Induction of prothrombinase fgl2 by the nucleocapsid protein of virulent mouse hepatitis virus is dependent on host hepatic nuclear factor-4 α . *J. Biol. Chem.* 278: 15541-15549.
5. Olson, G.E., et al. 2004. Region-specific expression and secretion of the fibrinogen-related protein, fgl2, by epithelial cells of the hamster epididymis and its role in disposal of defective spermatozoa. *J. Biol. Chem.* 279: 51266-51274.
6. Ghanekar, A., et al. 2004. Endothelial induction of fgl2 contributes to thrombosis during acute vascular xenograft rejection. *J. Immunol.* 172: 5693-5701.

CHROMOSOMAL LOCATION

Genetic locus: FGL2 (human) mapping to 7q11.23.

PRODUCT

Fgl2 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 Fgl2 siRNA (h): sc-44691 and Fgl2 shRNA Plasmid (h): sc-44691-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.

APPLICATIONS

Fgl2 shRNA (h) Lentiviral Particles is recommended for the inhibition of Fgl2 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

Fgl2 (K-17): sc-30870 is recommended as a control antibody for monitoring of Fgl2 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 Fgl2 gene expression knockdown using RT-PCR Primer: Fgl2 (h)-PR: sc-44691-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.

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