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Biglycan shRNA (m) Lentiviral Particles: sc-45735-V

BACKGROUND

Biglycan, a class I small leucine rich proteoglycan (SLRP) present in the extracellular matrix, influences bone cell differentiation and proliferation. Biglycan contains two chondroitin sulfate glucosaminoglycan (GAG) chains attached near its amino terminus, whereas a closely related SLRP, Decorin, contains only one. Biglycan deficient specimens possess diminished capacity to produce bone cells precursors, a lessened response to TGF β , reduced collagen synthesis, and increased apoptosis. Patients with rheumatoid arthritis express increased immunity to Biglycan whereas osteoarthritis patients do not, suggesting that higher immunity to SLRPs may play a role in the pathogenesis of inflammatory rheumatic diseases.

REFERENCES

1. Dodge, G.R., et al. 1998. Effects of IFN- γ and TNF α on the expression of the genes encoding Aggrecan, Biglycan, and Decorin core proteins in cultured human chondrocytes. *Arthritis Rheum.* 41: 274-283.
2. Klezovitch, O. and Scanu A.M. 2001. Domains of apolipoprotein E involved in the binding to the protein core of Biglycan of the vascular extracellular matrix: potential relationship between retention and anti-atherogenic properties of this apolipoprotein. *Trends Cardiovasc. Med.* 11: 263-268.
3. Goldberg, M., et al. 2002. Biglycan is a repressor of amelogenin expression and enamel formation: an emerging hypothesis. *J. Dent. Res.* 81: 520-524.

CHROMOSOMAL LOCATION

Genetic locus: Bgn (mouse) mapping to X A7.3.

PRODUCT

Biglycan 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 Biglycan siRNA (m): sc-45735 and Biglycan shRNA Plasmid (m): sc-45735-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.

PROTOCOLS

See our web site at www.scbt.com or our catalog for detailed protocols and support products.

APPLICATIONS

Biglycan shRNA (m) Lentiviral Particles is recommended for the inhibition of Biglycan 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

Biglycan (L-15): sc-27936 is recommended as a control antibody for monitoring of Biglycan 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 Biglycan gene expression knockdown using RT-PCR Primer: Biglycan (m)-PR: sc-45735-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.