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CILP-2 siRNA (m): sc-142343

BACKGROUND

Cartilage intermediate layer protein (CILP), an extracellular matrix protein, shows abundant expression in cartilaginous tissues. CILP is expressed as two isoforms, CILP and CILP-2, which are differentially expressed by chondrocytes and induced by TGF β 1. This induction is mediated by Smad3 through direct interactions with cis-elements in the CILP promoter region. TGF β also induces elevated chondrocyte extracellular inorganic pyrophosphate (PPi), which promotes the deposition of calcium pyrophosphate dihydrate crystals. The CILP isoforms have been implicated in common musculoskeletal disorders, including osteoarthritis, rheumatoid arthritis and lumbar disc disease.

REFERENCES

1. Johnson, K., et al. 2003. One of two chondrocyte-expressed isoforms of cartilage intermediate-layer protein functions as an Insulin-like growth factor 1 antagonist. *Arthritis Rheum.* 48: 1302-1314.
2. Yao, Z., et al. 2004. Characterization of cartilage intermediate layer protein (CILP)-induced arthropathy in mice. *Ann. Rheum. Dis.* 63: 252-258.
3. Valdes, A.M., et al. 2004. Association study of candidate genes for the prevalence and progression of knee osteoarthritis. *Arthritis Rheum.* 50: 2497-2507.
4. Lorenzo, P., et al. 2004. Altered patterns and synthesis of extracellular matrix macromolecules in early osteoarthritis. *Matrix Biol.* 23: 381-391.
5. Du, H., et al. 2005. The prevalence of autoantibodies against cartilage intermediate layer protein, YKL-39, osteopontin and cyclic citrullinated peptide in patients with early-stage knee osteoarthritis: evidence of a variety of autoimmune processes. *Rheumatol. Int.* 26: 35-41.
6. Seki, S., et al. 2005. A functional SNP in CILP, encoding cartilage intermediate layer protein, is associated with susceptibility to lumbar disc disease. *Nat. Genet.* 37: 607-612.
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CHROMOSOMAL LOCATION

Genetic locus: CILP2 (mouse) mapping to 8 B3.3.

PRODUCT

CILP-2 siRNA (m) is a pool of 2 target-specific 19-25 nt siRNAs designed to knock down gene expression. Each vial contains 3.3 nmol of lyophilized siRNA, sufficient for a 10 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see CILP-2 shRNA Plasmid (m): sc-142343-SH and CILP-2 shRNA (m) Lentiviral Particles: sc-142343-V as alternate gene silencing products.

For independent verification of CILP-2 (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-142343A and sc-142343B.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at -20° C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at -20° C, avoid contact with RNases and repeated freeze thaw cycles.

Resuspend lyophilized siRNA duplex in 330 μ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330 μ l of RNase-free water makes a 10 μ M solution in a 10 μ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

CILP-2 siRNA (m) is recommended for the inhibition of CILP-2 expression in mouse cells.

SUPPORT REAGENTS

For optimal siRNA transfection efficiency, Santa Cruz Biotechnology's siRNA Transfection Reagent: sc-29528 (0.3 ml), siRNA Transfection Medium: sc-36868 (20 ml) and siRNA Dilution Buffer: sc-29527 (1.5 ml) are recommended. Control siRNAs or Fluorescein Conjugated Control siRNAs are available as 10 μ M in 66 μ l. Each contain a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA. Fluorescein Conjugated Control siRNAs include: sc-36869, sc-44239, sc-44240 and sc-44241. Control siRNAs include: sc-37007, sc-44230, sc-44231, sc-44232, sc-44233, sc-44234, sc-44235, sc-44236, sc-44237 and sc-44238.

GENE EXPRESSION MONITORING

CILP-2 (B-1): sc-390297 is recommended as a control antibody for monitoring of CILP-2 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 reagents are recommended: 1) Western Blotting: use m-IgG κ BP-HRP: sc-516102 or m-IgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker™ Molecular Weight Standards: sc-2035, UltraCruz® Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG κ BP-FITC: sc-516140 or m-IgG κ BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz® Mounting Medium: sc-24941 or UltraCruz® Hard-set Mounting Medium: sc-359850.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor CILP-2 gene expression knockdown using RT-PCR Primer: CILP-2 (m)-PR: sc-142343-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

PROTOCOLS

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