

# Thrombospondin 5 siRNA (h): sc-43195

## BACKGROUND

Thrombospondin 5 (also designated TSP 5, cartilage oligomeric matrix protein or COMP) is the fifth member of the Thrombospondin family of extracellular matrix proteins. The Thrombospondin family share overall homology, with significant homology in their carboxy-terminal globular domains. They all contain type 2 (epidermal growth factor-like) and type 3 (calmodulin-like) repeats in their central domains. The human COMP/TSP 5 gene maps to chromosome 19p13.1. Thrombospondin 5 is expressed in all types of cartilage, tendon and vascular smooth muscle. Its localization in cartilage is developmentally regulated to the chondrocyte territorial and interterritorial matrix. Thrombospondin 5 also binds to Collagen Type I, II and IX in a zinc-dependent manner. Mutations in the COMP/TSP 5 gene are associated with the human genetic disorders pseudoachondroplasia (PSACH) and some types of multiple epiphyseal dysplasia (MED). PSACH and MED are autosomal dominant chondrodysplasias, which cause mild to severe short-limb dwarfism and early-onset osteoarthritis.

## REFERENCES

- Hedbom, E., et al. 1992. Cartilage matrix proteins. An acidic oligomeric protein (COMP) detected only in cartilage. *J. Biol. Chem.* 267: 6132-6136.
- Newton, G., et al. 1994. Characterization of human and mouse cartilage oligomeric matrix protein. *Genomics* 24: 435-439.
- Shen, Z., et al. 1995. Distribution and expression of cartilage oligomeric matrix protein and bone sialoprotein show marked changes during rat femoral head development. *Matrix Biol.* 14: 773-781.
- Briggs, M.D., et al. 1995. Pseudoachondroplasia and multiple epiphyseal dysplasia due to mutations in the cartilage oligomeric matrix protein gene. *Nat. Genet.* 10: 330-336.
- Riessen, R., et al. 2001. Cartilage oligomeric matrix protein (Thrombospondin 5) is expressed by human vascular smooth muscle cells. *Arterioscler. Thromb. Vasc. Biol.* 21: 47-54.
- Svensson, L., et al. 2002. Cartilage oligomeric matrix protein-deficient mice have normal skeletal development. *Mol. Cell. Biol.* 22: 4366-4371.

## CHROMOSOMAL LOCATION

Genetic locus: COMP (human) mapping to 19p13.11.

## PRODUCT

Thrombospondin 5 siRNA (h) is a pool of 3 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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Thrombospondin 5 shRNA Plasmid (h): sc-43195-SH and Thrombospondin 5 shRNA (h) Lentiviral Particles: sc-43195-V as alternate gene silencing products.

For independent verification of Thrombospondin 5 (h) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-43195A, sc-43195B and sc-43195C.

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

## APPLICATIONS

Thrombospondin 5 siRNA (h) is recommended for the inhibition of Thrombospondin 5 expression in human 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  $\mu$ M in 66  $\mu$ 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

Thrombospondin 5 (F-7): sc-374660 is recommended as a control antibody for monitoring of Thrombospondin 5 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 $\kappa$  BP-HRP: sc-516102 or m-IgG $\kappa$  BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker<sup>™</sup> Molecular Weight Standards: sc-2035, UltraCruz<sup>®</sup> Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG $\kappa$  BP-FITC: sc-516140 or m-IgG $\kappa$  BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz<sup>®</sup> Mounting Medium: sc-24941 or UltraCruz<sup>®</sup> Hard-set Mounting Medium: sc-359850.

## RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Thrombospondin 5 gene expression knockdown using RT-PCR Primer: Thrombospondin 5 (h)-PR: sc-43195-PR (20  $\mu$ l, 489 bp). 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](http://www.scbt.com) for detailed protocols and support products.