

ADAMTS-2 siRNA (h): sc-91785

BACKGROUND

ADAMTS (a disintegrin and metalloproteinase domain with Thrombospondin type-1 modules) is a family of zinc-dependent proteases that are implicated in a variety of normal and pathological conditions, including arthritis and cancer. ADAMTS protein family members contain an N-terminal propeptide domain, a metalloproteinase domain, a disintegrin-like domain and a C-terminus that contains a varying number of thrombospondin type-1 (TSP-1) motifs. ADAMTS genes are primarily expressed in fetal tissues, including the lung, kidney and liver. ADAMTS-2 cleaves the propeptides of Collagen Type I and II, but not Collagen Type III, prior to fibril assembly. It may also play a role in development aside from collagen biosynthesis. ADAMTS-2 is secreted and associated with the extracellular matrix, with the highest levels in skin, bone, tendon and aorta. Defects in ADAMTS2 are the cause of Ehlers-Danlos syndrome type VIIC (EDS VIIC), a recessively inherited connective-tissue disorder characterized clinically by severe skin fragility and joint hypermobility.

REFERENCES

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3. Li, S.W., et al. 2001. Transgenic mice with inactive alleles for procollagen N-proteinase (ADAMTS-2) develop fragile skin and male sterility. *Biochem. J.* 355: 271-278.
4. Cal, S., et al. 2002. Cloning, expression analysis, and structural characterization of seven novel human ADAMTSs, a family of metalloproteinases with disintegrin and Thrombospondin 1 domains. *Gene* 283: 49-62.
5. Wang, W.M., et al. 2003. Transforming growth factor β induces secretion of activated ADAMTS-2. A procollagen III N-proteinase. *J. Biol. Chem.* 278: 19549-19557.
6. Colige, A., et al. 2004. Novel types of mutation responsible for the dermatosparactic type of Ehlers-Danlos syndrome (Type VIIC) and common polymorphisms in the ADAMTS-2 gene. *J. Invest. Dermatol.* 123: 656-663.

CHROMOSOMAL LOCATION

Genetic locus: ADAMTS2 (human) mapping to 5q35.3.

PRODUCT

ADAMTS-2 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see ADAMTS-2 shRNA Plasmid (h): sc-91785-SH and ADAMTS-2 shRNA (h) Lentiviral Particles: sc-91785-V as alternate gene silencing products.

For independent verification of ADAMTS-2 (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-91785A, sc-91785B and sc-91785C.

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

ADAMTS-2 siRNA (h) is recommended for the inhibition of ADAMTS-2 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 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

ADAMTS-2 (F-4): sc-393562 is recommended as a control antibody for monitoring of ADAMTS-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 ADAMTS-2 gene expression knockdown using RT-PCR Primer: ADAMTS-2 (h)-PR: sc-91785-PR (20 μ l, 595 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

RESEARCH USE

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

PROTOCOLS

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