

ADAM5 siRNA (m): sc-61947

BACKGROUND

ADAMs (disintegrin and metalloproteinase domain), also known as MDCs (metalloproteinase, disintegrin and cysteine-rich domain) or cellular disintegrins, are a family of proteins that are ubiquitously expressed. They catalyze proteolysis, adhesion, fusion and intracellular signaling. ADAMs are membrane-anchored, glycosylated, Zn²⁺ dependent proteases, and there are over 30 different members in the family, with many diverse functions. ADAM1-6 localize to the testis, are developmentally regulated and are involved in spermatogenesis and sperm-egg binding and fusion. ADAM5 is expressed on the sperm surface. The ADAM5 precursor originates in the testis and is processed during epididymal maturation. Evidence suggests that ADAM5 plays a critical role in fertilization through a significant expressional relationship with ADAM2 and 3.

REFERENCES

1. Cho, C., et al. 1996. Chromosomal assignment of four testis-expressed mouse genes from a new family of transmembrane proteins (ADAMs) involved in cell-cell adhesion and fusion. *Genomics* 34: 413-417.
2. Yuan, R., et al. 1997. A role for the disintegrin domain of cyritestin, a sperm surface protein belonging to the ADAM family, in mouse sperm-egg plasma membrane adhesion and fusion. *J. Cell Biol.* 137: 105-112.
3. Sagane, K., et al. 1998. Metalloproteinase-like, disintegrin-like, cysteine-rich proteins MDC2 and MDC3: novel human cellular disintegrins highly expressed in the brain. *Biochem. J.* 334: 93-98.
4. Sagane, K., et al. 1999. Cloning and chromosomal mapping of mouse ADAM11, ADAM22 and ADAM23. *Gene* 236: 79-86.
5. Cal, S., et al. 2000. ADAM 23/MDC3, a human disintegrin that promotes cell adhesion via interaction with the $\alpha V\beta 3$ Integrin through an RGD-independent mechanism. *Mol. Biol. Cell* 11: 1457-1469.

CHROMOSOMAL LOCATION

Genetic locus: Adam5 (mouse) mapping to 8 A2.

PRODUCT

ADAM5 siRNA (m) 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 ADAM5 shRNA Plasmid (m): sc-61947-SH and ADAM5 shRNA (m) Lentiviral Particles: sc-61947-V as alternate gene silencing products.

For independent verification of ADAM5 (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-61947A, sc-61947B and sc-61947C.

PROTOCOLS

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

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

ADAM5 siRNA (m) is recommended for the inhibition of ADAM5 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

ADAM5 (C-6): sc-390150 is recommended as a control antibody for monitoring of ADAM5 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 ADAM5 gene expression knockdown using RT-PCR Primer: ADAM5 (m)-PR: sc-61947-PR (20 μ l). 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.