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# Zonadhesin siRNA (m): sc-155824

## BACKGROUND

Zonadhesin, also known as ZAN, is a sperm-specific, transmembrane protein that binds the spermatozoa to the zona pellucida of the oocyte, thereby initiating acrosomal fusion. Expressed primarily in the testis and localized to the apical region of the sperm head, Zonadhesin is a 2,812 amino acid protein that mediates species-specific cell adhesion in the sperm head and is essential for proper fertilization. Zonadhesin contains three MAM (Meprin/A5 antigen/m receptor tyrosine phosphatase) domains and four VWF (von willebrand factor) domains which mediate sperm adhesion and covalent oligomerization, respectively. Seven isoforms of Zonadhesin exist due to alternative splicing events. Four of the seven splice variants are expressed at low levels due to the presence of a premature mRNA stop codon that results in non-sense-mediated mRNA decay.

## REFERENCES

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- Gao, Z. and Garbers, D.L. 1998. Species diversity in the structure of Zonadhesin, a sperm-specific membrane protein containing multiple cell adhesion molecule-like domains. *J. Biol. Chem.* 273: 3415-3421.
- Lea, I.A., et al. 2001. Zonadhesin: characterization, localization, and zona pellucida binding. *Biol. Reprod.* 65: 1691-1700.
- Bi, M., et al. 2003. Processing, localization and binding activity of Zonadhesin suggest a function in sperm adhesion to the zona pellucida during exocytosis of the acrosome. *Biochem. J.* 375: 477-488.
- Olson, G.E., et al. 2004. Zonadhesin assembly into the hamster sperm acrosomal matrix occurs by distinct targeting strategies during spermiogenesis and maturation in the epididymis. *Biol. Reprod.* 71: 1128-1134.
- Herlyn, H. and Zischler, H. 2005. Sequence evolution, processing, and posttranslational modification of Zonadhesin D domains in primates, as inferred from cDNA data. *Gene* 362: 85-97.
- Hunt, P.N., et al. 2005. Expression and genomic organization of Zonadhesin-like genes in three species of fish give insight into the evolutionary history of a mosaic protein. *BMC Genomics* 6: 165.

## CHROMOSOMAL LOCATION

Genetic locus: Zan (mouse) mapping to 5 G2.

## PRODUCT

Zonadhesin 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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Zonadhesin shRNA Plasmid (m): sc-155824-SH and Zonadhesin shRNA (m) Lentiviral Particles: sc-155824-V as alternate gene silencing products.

For independent verification of Zonadhesin (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-155824A, sc-155824B and sc-155824C.

## 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

Zonadhesin siRNA (m) is recommended for the inhibition of Zonadhesin 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  $\mu$ 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

Zonadhesin (H-2): sc-514467 is recommended as a control antibody for monitoring of Zonadhesin 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™ Molecular Weight Standards: sc-2035, UltraCruz® 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® Mounting Medium: sc-24941 or UltraCruz® Hard-set Mounting Medium: sc-359850.

## RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Zonadhesin gene expression knockdown using RT-PCR Primer: Zonadhesin (m)-PR: sc-155824-PR (20  $\mu$ 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.

## PROTOCOLS

See our web site at [www.scbt.com](http://www.scbt.com) for detailed protocols and support products.