SANTA CRUZ BIOTECHNOLOGY, INC.

Calmegin siRNA (m): sc-60317

BACKGROUND

Calmegin, belonging to the calreticulin family, is expressed in testis as an endoplasmic reticulum membrane protein, where it acts as a chaperone protein and plays a role in spermatogenesis. Calmegin is a single-pass type I membrane protein that is transcriptionally regulated in coordination by CpG methyltransferase and histone deacetylase (HDAC). First expressed in meiotic prophase of spermatocytes, Calmegin facilitates sperm-egg zona pellucida binding through association with sperm membrane proteins (fertilin α and β). A loss in Calmegin results in male sterility. However, if the zona pellucida is partially dissected and fertilized *in vitro,* the egg will develop normally.

REFERENCES

1. Siep, M., Sleddens-Linkels, E., Mulders, S., van Eenennaam, H., Wassenaar, E., Van Cappellen, W.A., Hoogerbrugge, J., Grootegoed, J.A. and Baarends, W.M. 2004. Basic helix-loop-helix transcription factor TCFL5 interacts with the Calmegin gene promoter in mouse spermatogenesis. Nucleic Acids Res. 32: 6425-6436.
2. Nakanishi, T., Isotani, A., Yamaguchi, R., Ikawa, M., Baba, T., Suarez, S.S. and Okabe, M. 2004. Selective passage through the uterotubal junction of sperm from a mixed population produced by chimeras of Calmegin-knockout and wildtype male mice. Biol. Reprod. 71: 959-965.
3. Kim, D.H., Shim, J.S. and Kwon, H.J. 2005. Coordinated transcriptional regulation of Calmegin, a testis-specific molecular chaperon, by histone deacetylase and CpG methyltransferase. Exp. Mol. Med. 37: 492-496.

CHROMOSOMAL LOCATION

Genetic locus: Clgn (mouse) mapping to 8 C2.

PRODUCT

Calmegin 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Calmegin shRNA Plasmid (m): sc-60317-SH and Calmegin shRNA (m) Lentiviral Particles: sc-60317-V as alternate gene silencing products.

For independent verification of Calmegin (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-60317A, sc-60317B and sc-60317C.

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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

Calmegin siRNA (m) is recommended for the inhibition of Calmegin 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 µM in 66 µl. 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Calmegin gene expression knockdown using RT-PCR Primer: Calmegin (m)-PR: sc-60317-PR (20 µl). 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.