



GCL siRNA (h): sc-72182

BACKGROUND

Germ cell-less (GCL) represses transcription during the later stages of spermatogenesis both in *Drosophila* and mammalian species. GCL localizes to the nuclear envelope, where it competes with BAF to interact with emerlin, a complex that is required for appropriate gene expression. Defective sperm motility as well as impaired nuclear envelope integrity in liver, pancreas and testis cells result from a lack of GCL gene expression. Thus it appears that the essential function of GCL is to facilitate normal nuclear-lamina organization, which results in normal sperm morphogenesis and chromatin remodeling.

REFERENCES

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2. Holaska, J.M., et al. 2003. Transcriptional repressor Germ cell-less (GCL) and barrier to autointegration factor (BAF) compete for binding to emerlin *in vitro*. *J. Biol. Chem.* 278: 6969-6975.
3. Kimura, T., et al. 2003. Mouse germ cell-less as an essential component for nuclear integrity. *Mol. Cell. Biol.* 23: 1304-1315.
4. Masuhara, M., et al. 2003. Enhanced degradation of MDM2 by a nuclear envelope component, mouse germ cell-less. *Biochem. Biophys. Res. Commun.* 308: 927-932.
5. Wilkinson, F.L., et al. 2003. Emerin interacts *in vitro* with the splicing-associated factor, YT521-B. *Eur. J. Biochem.* 270: 2459-2466.
6. Haraguchi, T., et al. 2004. Emerin binding to Btf, a death-promoting transcriptional repressor, is disrupted by a missense mutation that causes Emery-Dreifuss muscular dystrophy. *Eur. J. Biochem.* 271: 1035-1045.

CHROMOSOMAL LOCATION

Genetic locus: GMCL1 (human) mapping to 2p13.3.

PRODUCT

GCL 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 GCL shRNA Plasmid (h): sc-72182-SH and GCL shRNA (h) Lentiviral Particles: sc-72182-V as alternate gene silencing products.

For independent verification of GCL (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-72182A, sc-72182B and sc-72182C.

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.

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

GCL siRNA (h) is recommended for the inhibition of GCL 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

GCL (B-4): sc-271330 is recommended as a control antibody for monitoring of GCL 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 GCL gene expression knockdown using RT-PCR Primer: GCL (h)-PR: sc-72182-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.