

Gelsolin siRNA (r): sc-45925

BACKGROUND

Gelsolin (also known as brevin, Actin-depolymerizing factor or ADF), a protein of leukocytes, platelets and other cells, severs Actin filaments in the presence of submicromolar calcium, thereby isolating cytoplasmic Actin gels. A calcium-independent mechanism reverses the process. A Gelsolin variant with 23 more amino-terminal amino acids is a plasma component probably involved in the clearance of Actin, the most abundant human protein, from the circulation. It has been suggested that a single gene encodes both cell and plasma gelsolins. Gelsolin may be unique in that it is made for both secretion and intracytoplasmic location. Amino acid homology was identified between Gelsolin and the amyloid of the Finnish variety of amyloidosis. The amyloid in this disorder is antigenically and structurally related to Gelsolin. Gelsolin is the principal intracellular and extracellular Actin-severing protein. Gelsolin and Gc protein together constitute the extracellular Actin-scavenger system which prevents the toxic effects of Actin release into the extracellular space under circumstances of cell necrosis.

REFERENCES

1. Lind, S.E., et al. 1984. Human plasma Gelsolin binds to Fibronectin. *J. Biol. Chem.* 259: 13262-13266.
2. Fernandes-Alnemri, T., et al. 1995. Mch3, a novel human apoptotic cysteine protease highly related to CPP32. *Cancer Res.* 55: 6045-6052.
3. Takahashi, A., et al. 1996. Cleavage of Lamin A by Mch2 a but not CPP32: multiple interleukin-1 β -converting enzyme-related proteases with distinct substrate recognition properties are active in apoptosis. *Proc. Natl. Acad. Sci. USA* 93: 8395-8400.
4. Rao, L., et al. 1996. Lamin proteolysis facilitates nuclear events during apoptosis. *J. Cell Biol.* 135: 1441-1455.
5. Liu, X., et al. 1997. DFF, a heterodimeric protein that functions downstream of caspase-3 to trigger DNA fragmentation during apoptosis. *Cell* 89: 175-184.
6. Salvesen, G.S., et al. 1997. Caspases: intracellular signaling by proteolysis. *Cell* 91: 443-446.
7. Kothakota, S., et al. 1997. Caspase-3-generated fragment of Gelsolin: effector of morphological change in apoptosis. *Science* 278: 294-298.

CHROMOSOMAL LOCATION

Genetic locus: Gsn (rat) mapping to 3p11.

PRODUCT

Gelsolin siRNA (r) 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 Gelsolin shRNA Plasmid (r): sc-45925-SH and Gelsolin shRNA (r) Lentiviral Particles: sc-45925-V as alternate gene silencing products.

For independent verification of Gelsolin (r) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-45925A, sc-45925B and sc-45925C.

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

Gelsolin siRNA (r) is recommended for the inhibition of Gelsolin expression in rat 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

Gelsolin (F-5): sc-514502 is recommended as a control antibody for monitoring of Gelsolin 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 MarkerTM 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 Gelsolin gene expression knockdown using RT-PCR Primer: Gelsolin (r)-PR: sc-45925-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.

PROTOCOLS

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