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spectrin α I siRNA (h): sc-43430

BACKGROUND

Spectrin, an Actin binding protein that is a major component of the cytoskeletal superstructure of the erythrocyte plasma membrane, is essential in determining the properties of the membrane including its shape and deformability. Spectrins function as membrane organizers and stabilizers, composed of non-homologous α and β chains, which aggregate side-to-side in an antiparallel fashion to form dimers, tetramers and higher polymers. Spectrin α I and spectrin β I are present in erythrocytes, whereas spectrin α II (also designated fodrin α) and spectrin β II (also designated fodrin β) are present in other somatic cells. The spectrin tetramers in erythrocytes act as barriers to lateral diffusion, but spectrin dimers seem to lack this function. Defects of spectrin α I are present in the erythrocytes of many patients with abnormalities of red blood cell shape including hereditary spherocytosis and elliptocytosis.

REFERENCES

1. Speicher, D.W. 1986. The present status of erythrocyte spectrin structure: the 106-residue repetitive structure is a basic feature of an entire class of proteins. *J. Cell. Biochem.* 30: 245-258.
2. Gardner, K., et al. 1987. Modulation of spectrin-actin assembly by erythrocyte adducin. *Nature* 328: 359-362.
3. Coleman, T.R., et al. 1989. Functional diversity among spectrin isoforms. *Cell Motil. Cytoskeleton* 12: 225-247.
4. Saxton, M.J. 1989. The spectrin network as a barrier to lateral diffusion in erythrocytes. A percolation analysis. *Biophys. J.* 55: 21-28.
5. Kennedy, S.P., et al. 1994. A partial structural repeat forms the heterodimer self-association site of all β -spectrins. *J. Biol. Chem.* 269: 11400-11408.
6. Online Mendelian Inheritance in Man, OMIM[™]. 1999. Johns Hopkins University, Baltimore, MD. MIM Number: 182860. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
7. Boulanger, L., et al. 2002. Erythroid expression of the human α -spectrin gene promoter is mediated by GATA-1- and NF-E2-binding proteins. *J. Biol. Chem.* 277: 41563-41570.

CHROMOSOMAL LOCATION

Genetic locus: SPTA1 (human) mapping to 1q23.1.

PRODUCT

spectrin α I 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 spectrin α I shRNA Plasmid (h): sc-43430-SH and spectrin α I shRNA (h) Lentiviral Particles: sc-43430-V as alternate gene silencing products.

For independent verification of spectrin α I (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-43430A, sc-43430B and sc-43430C.

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

spectrin α I siRNA (h) is recommended for the inhibition of spectrin α I 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

spectrin α I (B-12): sc-271130 is recommended as a control antibody for monitoring of spectrin α I 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 spectrin α I gene expression knockdown using RT-PCR Primer: spectrin α I (h)-PR: sc-43430-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.