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spectrin β III siRNA (h): sc-43432

BACKGROUND

Spectrin is an actin binding protein that is a major component of the plasma membrane skeleton. Spectrins function as membrane organizers and stabilizers by forming dimers, tetramers and higher polymers. Spectrin α and spectrin β are present in erythrocytes, whereas spectrin α II (also designated fodrin α) and spectrin β I (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. Spectrin β III is highly homologous to both spectrin β I and spectrin β II. Western blot analysis shows that spectrin β III migrates as an approximately 220 kDa polypeptide despite a calculated molecular mass of 271 kDa. Spectrin β III is highly expressed in brain, kidney, pancreas and liver, and at lower levels in lung and placenta. Specifically, spectrin β III constitutes a major component of the Golgi and vesicular membrane skeletons. The human SPTBN2 gene maps to chromosome 11q13.2.

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. Coelman, T.R., et al. J.S. 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, et al. 1994. A partial structural repeat forms the heterodimer self-association site of all β -spectrins. *J. Biol. Chem.* 269: 11400-11408.

CHROMOSOMAL LOCATION

Genetic locus: SPTBN2 (human) mapping to 11q13.2.

PRODUCT

spectrin β III 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 β III shRNA Plasmid (h): sc-43432-SH and spectrin β III shRNA (h) Lentiviral Particles: sc-43432-V as alternate gene silencing products.

For independent verification of spectrin β III (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-43432A, sc-43432B and sc-43432C.

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

spectrin β III siRNA (h) is recommended for the inhibition of spectrin β III 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 β III (A-8): sc-515737 is recommended as a control antibody for monitoring of spectrin β III 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 β III gene expression knockdown using RT-PCR Primer: spectrin β III (h)-PR: sc-43432-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.