

CBS siRNA (r): sc-270531

BACKGROUND

Strongly expressed in human liver and pancreas, with weaker expression in heart and brain, the cytoplasmic protein cystathionine β -synthase (CBS) operates in the first step of homocysteine transsulfuration. CBS, which belongs to the cysteine synthase/cystathionine β -synthase family of proteins, catalyzes the formation of cystathionine from the thrombogenic amino acid homocysteine using pyridoxal phosphate cofactor. Allosteric activation by adenosyl-methionine regulates CBS activity. Deficiencies in CBS are associated with homocystinuria, a recessively inherited error in sulfur amino acid metabolism that affects many organs and tissues. Symptoms of homocystinuria include arteriosclerosis, thrombosis, dislocated optic lenses, mental retardation and skeletal abnormalities.

REFERENCES

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CHROMOSOMAL LOCATION

Genetic locus: Cbs (rat) mapping to 20p12.

PRODUCT

CBS 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 CBS shRNA Plasmid (r): sc-270531-SH and CBS shRNA (r) Lentiviral Particles: sc-270531-V as alternate gene silencing products.

For independent verification of CBS (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-270531A, sc-270531B and sc-270531C.

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

CBS siRNA (r) is recommended for the inhibition of CBS 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

CBS (B-4): sc-133154 is recommended as a control antibody for monitoring of CBS 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 CBS gene expression knockdown using RT-PCR Primer: CBS (r)-PR: sc-270531-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.