SANTA CRUZ BIOTECHNOLOGY, INC.

CH25H siRNA (m): sc-142309

BACKGROUND

CH25H (Cholesterol 25-hydroxylase), also known as h25OH, is a 272 amino acid endoplasmic membrane protein that belongs to the sterol desaturase family. CH25H contains clusters of histidine residues essential for catalytic activity and is involved in cholesterol and lipid metabolism. CH25H catalyzes the formation of 25-hydroxycholesterol from cholesterol leading to the repression of cholesterol biosynthetic enzymes. CH25H regulates lipid metabolism by synthesizing a corepressor that blocks sterol regulatory element binding protein (SREBP) processing. CH25H utilizes diiron cofactors to catalyze the hydroxylation of hydrophobic substrates.

REFERENCES

1. Riemenschneider, M., et al. 2004. Association analysis of genes involved in cholesterol metabolism located within the linkage region on chromosome 10 and Alzheimer's disease. Neurobiol. Aging 25: 1305-1308.
2. Papassotiropoulos, A., et al. 2005. Cholesterol 25-hydroxylase on chromosome 10q is a susceptibility gene for sporadic Alzheimer's disease. Neurodegener. Dis. 2: 233-241.
3. Wang, J.H., et al. 2006. Regulation of cholesterol 25-hydroxylase expression by vitamin D3 metabolites in human prostate stromal cells. Biochem. Biophys. Res. Commun. 345: 720-725.
4. Shibata, N., et al. 2006. Association studies of cholesterol metabolism genes (CH25H, ABCA1 and CH24H) in Alzheimer's disease. Neurosci. Lett. 391: 142-146.
5. Morgan, A.R., et al. 2007. Association studies of 23 positional/functional candidate genes on chromosome 10 in late-onset Alzheimer's disease. Am. J. Med. Genet. B Neuropsychiatr. Genet. 144B: 762-770.
6. Carter, C.J. 2007. Convergence of genes implicated in Alzheimer's disease on the cerebral cholesterol shuttle: APP, cholesterol, lipoproteins, and atherosclerosis. Neurochem. Int. 50: 12-38.

CHROMOSOMAL LOCATION

Genetic locus: Ch25h (mouse) mapping to 19 C1.

PRODUCT

CH25H siRNA (m) 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see CH25H shRNA Plasmid (m): sc-142309-SH and CH25H shRNA (m) Lentiviral Particles: sc-142309-V as alternate gene silencing products.

For independent verification of CH25H (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-142309A, sc-142309B and sc-142309C.

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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

CH25H siRNA (m) is recommended for the inhibition of CH25H expression in mouse 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 µM in 66 µl. 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

CH25H (1G8): sc-293256 is recommended as a control antibody for monitoring of CH25H 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 CH25H gene expression knockdown using RT-PCR Primer: CH25H (m)-PR: sc-142309-PR (20 µl). 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.