SANTA CRUZ BIOTECHNOLOGY, INC.

CYP4X1 siRNA (m): sc-142738

BACKGROUND

Cytochrome P450 proteins are heme-thiolate monooxygenases that mediate NADPH-dependent electron transport and function to oxidize a variety of structurally unrelated compounds, including steroids, fatty acids and xenobiotics. Specifically, Cytochrome P450s are responsible for metabolizing arachidonic acid to hydroxyeicosatetraenoic acid (a regulator of blood pressure) and epoxyeicosatrienoic acid (a molecule involved in signaling events). CYP4X1 (cytochrome P450, family 4, subfamily X, polypeptide 1) is a 509 amino acid peripheral membrane protein of the microsome and endoplasmic reticulum. Expressed in brain, aorta and trachea, CYP4X1 may play a role in neurovascular function and is encoded by a gene that maps to human chromosome 1p33. Chromosome 1 spans 260 million base pairs, contains over 3,000 genes, and houses a large number of disease-associated genes, including those that are involved in familial adenomatous polyposis, Stickler syndrome, Parkinson's disease and Gaucher disease.

REFERENCES

1. Bowling, E.L., et al. 2000. The Stickler syndrome: case reports and literature review. Optometry 71: 177-182.
2. Ito, O., et al. 2001. Effects of converting enzyme inhibitors on renal P-450 metabolism of arachidonic acid. Am. J. Physiol. Regul. Integr. Comp. Physiol. 280: R822-R830.
3. Bylund, J., et al. 2002. Identification of a novel cytochrome P450, CYP4X1, with unique localization specific to the brain. Biochem. Biophys. Res. Commun. 296: 677-684.
4. Plasilova, M., et al. 2004. Exclusion of an extracolonic disease modifier locus on chromosome 1p33-36 in a large Swiss familial adenomatous polyposis kindred. Eur. J. Hum. Genet. 12: 365-371.
5. Nelson, D.R., et al. 2004. Comparison of cytochrome P450 (CYP) genes from the mouse and human genomes, including nomenclature recommendations for genes, pseudogenes and alternative-splice variants. Pharmacogenetics 14: 1-18.

CHROMOSOMAL LOCATION

Genetic locus: Cyp4x1 (mouse) mapping to 4 D1.

PRODUCT

CYP4X1 siRNA (m) is a pool of 2 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 CYP4X1 shRNA Plasmid (m): sc-142738-SH and CYP4X1 shRNA (m) Lentiviral Particles: sc-142738-V as alternate gene silencing products.

For independent verification of CYP4X1 (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-142738A and sc-142738B.

PROTOCOLS

See our web site at www.scbt.com or our catalog 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

CYP4X1 siRNA (m) is recommended for the inhibition of CYP4X1 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

CYP4X1 (Q-12): sc-162724 is recommended as a control antibody for monitoring of CYP4X1 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 (secondary) reagents are recommended: 1) Western Blotting: use donkey anti-goat IgG-HRP: sc-2020 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible donkey anti-goat IgG-HRP: sc-2033 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use donkey anti-goat IgG-FITC: sc-2024 (dilution range: 1:100-1:400) or donkey anti-goat IgG-TR: sc-2783 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor CYP4X1 gene expression knockdown using RT-PCR Primer: CYP4X1 (m)-PR: sc-142738-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.