

FMO3 siRNA (h): sc-72256

BACKGROUND

The Flavin containing monooxygenase family consists of five gene products, FMO1-5, that are major enzymatic oxidants involved in the metabolism of various therapeutics. Located in the liver, FMO3 is a hepatic microsomal enzyme that oxygenates soft nucleophiles such as secondary and tertiary amines. Through its N-oxygenase capabilities, FMO3 acts on a variety of xenobiotics to catalyze oxidative digestion. Defects in the FMO3 gene are the primary cause of trimethylaminuria (TMAuria), an inborn error of metabolism associated with a fishy body odor emitting from sweat, urine and breath. Genetic mutations in FMO3 lead to the N-oxidation of amino-trimethylamine derived from food products, thus producing the malodor associated with TMAuria.

REFERENCES

1. Burnett, V.L., et al. 1994. Cloning and sequencing of flavin-containing monooxygenases FMO3 and FMO4 from rabbit and characterization of FMO3. *J. Biol. Chem.* 269: 14314-14322.
2. Krause, R.J., et al. 1996. Characterization of the methionine S-oxidase activity of rat liver and kidney microsomes: immunochemical and kinetic evidence for FMO3 being the major catalyst. *Arch. Biochem. Biophys.* 333: 109-116.
3. Falls, J.G., et al. 1997. Molecular cloning, sequencing, and expression in *Escherichia coli* of mouse flavin-containing monooxygenase 3 (FMO3): comparison with the human isoform. *Arch. Biochem. Biophys.* 347: 9-18.
4. Petalcorin, M.I., et al. 2005. The fmo genes of *Caenorhabditis elegans* and *C. briggsae*: characterisation, gene expression and comparative genomic analysis. *Gene* 346: 83-96.
5. Koukouritaki, S.B., et al. 2005. Discovery of novel flavin-containing monooxygenase 3 (FMO3) single nucleotide polymorphisms and functional analysis of upstream haplotype variants. *Mol. Pharmacol.* 68: 383-392.
6. Honkatukia, M., et al. 2005. Fishy taint in chicken eggs is associated with a substitution within a conserved motif of the FMO3 gene. *Genomics* 86: 225-232.

CHROMOSOMAL LOCATION

Genetic locus: FMO3 (human) mapping to 1q24.3.

PRODUCT

FMO3 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 FMO3 shRNA Plasmid (h): sc-72256-SH and FMO3 shRNA (h) Lentiviral Particles: sc-72256-V as alternate gene silencing products.

For independent verification of FMO3 (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-72256A, sc-72256B and sc-72256C.

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

FMO3 siRNA (h) is recommended for the inhibition of FMO3 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

FMO3 (C-1): sc-515042 is recommended as a control antibody for monitoring of FMO3 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 FMO3 gene expression knockdown using RT-PCR Primer: FMO3 (h)-PR: sc-72256-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.