

CPOX siRNA (m): sc-142545

BACKGROUND

CPOX (coproporphyrinogen III oxidase) is a 454 amino acid mitochondrial enzyme that is localized to the inner membrane space of erythrocytes. It participates in the sixth step of heme biosynthesis by catalyzing the formation of protoporphyrinogen IX from coproporphyrinogen III. Mutations in the gene encoding CPOX are the cause of coproporphyrinemia, an autosomal dominant disease characterized by skin photosensitivity and neurological disturbances. Symptoms are often experienced as attacks, which include severe abdominal and nerve pain. People affected by coproporphyrinemia overexcrete coproporphyrinogen III in feces and urine and the enzymatic activity of CPOX is found to be approximately half that of normal, leading to a decrease in overall heme synthesis. There is no cure for coproporphyrinemia, but preventative treatment to relieve symptoms usually involves dietary changes and avoidance of drugs and alcohol.

REFERENCES

1. Taketani, S., et al. 1994. Molecular cloning, sequencing and expression of cDNA encoding human coproporphyrinogen oxidase. *Biochim. Biophys. Acta* 1183: 547-549.
2. Martasek, P., et al. 1994. Homozygous hereditary coproporphyrinemia caused by an arginine to tryptophane substitution in coproporphyrinogen oxidase and common intragenic polymorphisms. *Hum. Mol. Genet.* 3: 477-480.
3. Delfau-Larue, M.H., et al. 1994. Coproporphyrinogen oxidase: gene organization and description of a mutation leading to exon 6 skipping. *Hum. Mol. Genet.* 3: 1325-1330.
4. Lamoril, J., et al. 1995. A molecular defect in coproporphyrinogen oxidase gene causing harderoporphyria, a variant form of hereditary coproporphyrinemia. *Hum. Mol. Genet.* 4: 275-278.
5. Daimon, M., et al. 1997. A novel missense mutation in exon 4 of the human coproporphyrinogen oxidase gene in two patients with hereditary coproporphyrinemia. *Hum. Genet.* 99: 199-201.
6. Lamoril, J., et al. 1997. Three novel mutations in the coproporphyrinogen oxidase gene. *Hum. Mutat.* 9: 78-80.

CHROMOSOMAL LOCATION

Genetic locus: Cpx (mouse) mapping to 16 C1.2.

PRODUCT

CPOX 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see CPOX shRNA Plasmid (m): sc-142545-SH and CPOX shRNA (m) Lentiviral Particles: sc-142545-V as alternate gene silencing products.

For independent verification of CPOX (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-142545A, sc-142545B and sc-142545C.

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

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

CPOX (B-9): sc-393388 is recommended as a control antibody for monitoring of CPOX 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 MarkerTM 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 CPOX gene expression knockdown using RT-PCR Primer: CPOX (m)-PR: sc-142545-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.