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CYPOR shRNA (h2) Lentiviral Particles: sc-43715-V

BACKGROUND

P450 enzymes constitute a family of monooxygenase enzymes that are involved in the metabolism of a wide array of endogenous and xenobiotic compounds. Several P450 enzymes have been classified by sequence similarities as members of the CYP1A and CYP2A subfamilies. CYPOR, also known as cytochrome P450 reductase and NADPH cytochrome P450 reductase, is a microsomal enzyme responsible for the transfer of electrons from NADPH to cytochrome P450 enzymes during the P450 catalytic cycle. CYPOR is localized to the endoplasmic reticulum, where it is also able to transfer electrons to heme oxygenase and cytochrome b5. CYPOR is structurally related to two separate flavo-protein families, ferredoxin nucleotide reductase (FNR) and flavodoxin. Electron transfer of CYPOR requires the binding of two flavin cofactors, FAD and FMN, to the FNR and flavodoxin domains, respectively.

REFERENCES

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- Ohgjiya, S., et al. 1994. Mouse NADPH cytochrome P450 oxidoreductase: molecular cloning and functional expression in yeast. *Biochem. Biophys. Acta* 1186: 137-141.
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CHROMOSOMAL LOCATION

Genetic locus: POR (human) mapping to 7q11.23.

PRODUCT

CYPOR shRNA (h2) Lentiviral Particles is a pool of concentrated, transduction-ready viral particles containing 3 target-specific constructs that encode 19-25 nt (plus hairpin) shRNA designed to knock down gene expression. Each vial contains 200 μ l frozen stock containing 1.0×10^6 infectious units of virus (IFU) in Dulbecco's Modified Eagle's Medium with 25 mM HEPES pH 7.3. Suitable for 10-20 transductions. Also see CYPOR siRNA (h2): sc-43715 and CYPOR shRNA Plasmid (h2): sc-43715-SH as alternate gene silencing products.

STORAGE

Store lentiviral particles at -80° C. Stable for at least one year from the date of shipment. Once thawed, particles can be stored at 4° C for up to one week. Avoid repeated freeze thaw cycles.

APPLICATIONS

CYPOR shRNA (h2) Lentiviral Particles is recommended for the inhibition of CYPOR expression in human cells.

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 μ l frozen viral stock containing 1.0×10^6 infectious units of virus (IFU); contains an shRNA construct encoding a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA.

GENE EXPRESSION MONITORING

CYPOR (F-10): sc-25270 is recommended as a control antibody for monitoring of CYPOR 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 goat anti-mouse IgG-HRP: sc-2005 (dilution range: 1:2000-1:32,000) or Cruz Marker™ compatible goat anti-mouse IgG-HRP: sc-2031 (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 goat anti-mouse IgG-FITC: sc-2010 (dilution range: 1:100-1:400) or goat anti-mouse IgG-TR: sc-2781 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor CYPOR gene expression knockdown using RT-PCR Primer: CYPOR (h2)-PR: sc-43715-PR (20 μ l, 476 bp). Annealing temperature for the primers should be $55-60^{\circ}$ C and the extension temperature should be $68-72^{\circ}$ C.

BIOSAFETY

Lentiviral particles can be employed in standard Biosafety Level 2 tissue culture facilities (and should be treated with the same level of caution as with any other potentially infectious reagent). Lentiviral particles are replication-incompetent and are designed to self-inactivate after transduction and integration of shRNA constructs into genomic DNA of target cells.

RESEARCH USE

The purchase of this product conveys to the buyer the nontransferable right to use the purchased amount of the product and all replicates and derivatives for research purposes conducted by the buyer in his laboratory only (whether the buyer is an academic or for-profit entity). The buyer cannot sell or otherwise transfer (a) this product (b) its components or (c) materials made using this product or its components to a third party, or otherwise use this product or its components or materials made using this product or its components for Commercial Purposes.

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