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ERR γ shRNA (h) Lentiviral Particles: sc-44704-V

BACKGROUND

Estrogen and progesterone receptors are members of a family of transcription factors that are regulated by the binding of their cognate ligands. The interaction of hormone-bound estrogen receptors with estrogen responsive elements (EREs) alters transcription of ERE-containing genes. Estrogen receptor-related proteins, (ERR α , β and γ), are orphan nuclear receptors. Like estrogen receptors, ERRs bind specifically to EREs to activate reporter genes. EREs are constitutively active without binding to estrogen. The biological response to progesterone is mediated by two distinct forms of the human progesterone receptor (PR-A and PR-B), which arise from alternative splicing. In most cells, PR-B functions as a transcriptional activator of progesterone-responsive genes, whereas PR-A functions as a transcriptional inhibitor of all steroid hormone receptors. mPR is a membrane progesterin receptor. The predicted 436-amino acid ERR γ protein which presumably localizes to the nucleus, is expressed in the heart, kidney, brain, lung, bone marrow, adrenal gland, trachea, spinal cord and thyroid gland tissues.

REFERENCES

1. Eudy, J.D., et al. 1998. Isolation of a gene encoding a novel member of from the critical region of Usher syndrome type IIa at 1q41. *Genomics* 50: 382-384.
2. Hong, H., et al. 1999. Hormone-independent transcriptional activation and coactivator binding by novel orphan nuclear receptor ERR3. *J. Biol. Chem.* 274: 22618-22626.
3. Heard, D.J., et al. 2000. Human ERR γ , a third member of the estrogen receptor-related receptor (ERR) subfamily of orphan nuclear receptors: tissue-specific isoforms are expressed during development and in the adult. *Mol. Endocrinol.* 14: 382-392.

CHROMOSOMAL LOCATION

Genetic locus: ESRRG (human) mapping to 1q41.

PRODUCT

ERR γ shRNA (h) 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 ERR γ siRNA (h): sc-44704 and ERR γ shRNA Plasmid (h): sc-44704-SH as alternate gene silencing products.

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.

APPLICATIONS

ERR γ shRNA (h) Lentiviral Particles is recommended for the inhibition of ERR γ 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

ERR γ (D-1): sc-393969 is recommended as a control antibody for monitoring of ERR γ 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 ERR γ gene expression knockdown using RT-PCR Primer: ERR γ (h)-PR: sc-44704-PR (20 μ l, 489 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.

STORAGE

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

PROTOCOLS

See our web site at www.scbt.com or our catalog for detailed protocols and support products.