

# EAR2 siRNA (h): sc-38822

## BACKGROUND

EAR2 (v-Erb-A avian erythroblastic leukemia viral oncogene homolog-like 2) protein binds a *cis* enhancer element (TGACCT motif) upstream of the Renin gene and represses Renin gene transcription. EAR2 also contributes to gonadotropin-dependent derepression of LHR promoter activity in granulosa cells. EAR2 mRNA is abundant in the liver and EAR2 protein localizes to the nucleus of As4.1 cells.

## REFERENCES

1. Ladias, J.A., et al. 1992. Transcriptional regulation of human apolipoprotein genes ApoB, ApoCIII, and ApoAII by members of the steroid hormone receptor superfamily HNF4, ARP1, EAR2, and EAR3. *J. Biol. Chem.* 267: 15849-15860.
2. Barnhart, K.M., et al. 1994. The sequence of a murine cDNA encoding EAR2, a nuclear orphan receptor. *Gene* 142: 313-314.
3. Islam, T.C., et al. 1994. Nuclear orphan receptor-binding retinoic acid response elements in keratinocytes. *Biochem. Biophys. Res. Commun.* 203: 545-552.
4. Chu, K., et al. 1997. The nuclear orphan receptors COUP-TFII and EAR2 act as silencers of the human oxytocin gene promoter. *J. Mol. Endocrinol.* 19: 163-172.
5. Chu, K., et al. 1998. Nuclear orphan receptors COUP-TFII and EAR2: presence in oxytocin-producing uterine cells and functional interaction with the oxytocin gene promoter. *Mol. Cell. Endocrinol.* 137: 145-154.
6. Zhang, Y., et al. 2000. Nuclear orphan receptors regulate transcription of the gene for the human luteinizing hormone receptor. *J. Biol. Chem.* 275: 2763-2770.
7. Zhu, X.G., et al. 2000. The orphan nuclear receptor EAR2 is a negative coregulator for thyroid hormone nuclear receptor function. *Mol. Cell. Biol.* 20: 2604-2618.
8. Zhang, Y. and Dufau, M.L. 2001. EAR2 and EAR3/COUP-TFI regulate transcription of the rat LH receptor. *Mol. Endocrinol.* 15: 1891-1905.
9. Liu, X., et al. 2003. Identification of a nuclear orphan receptor EAR2 as a negative regulator of renin gene transcription. *Circ. Res.* 92: 1033-1040.

## CHROMOSOMAL LOCATION

Genetic locus: NR2F6 (human) mapping to 19p13.11.

## PRODUCT

EAR2 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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see EAR2 shRNA Plasmid (h): sc-38822-SH and EAR2 shRNA (h) Lentiviral Particles: sc-38822-V as alternate gene silencing products.

For independent verification of EAR2 (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-38822A, sc-38822B and sc-38822C.

## 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  $\mu$ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330  $\mu$ l of RNase-free water makes a 10  $\mu$ M solution in a 10  $\mu$ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

## APPLICATIONS

EAR2 siRNA (h) is recommended for the inhibition of EAR2 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  $\mu$ M in 66  $\mu$ 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

COUP-TF/EAR2 (F-11): sc-166941 is recommended as a control antibody for monitoring of EAR2 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 $\kappa$  BP-HRP: sc-516102 or m-IgG $\kappa$  BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker<sup>™</sup> Molecular Weight Standards: sc-2035, UltraCruz<sup>®</sup> Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG $\kappa$  BP-FITC: sc-516140 or m-IgG $\kappa$  BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz<sup>®</sup> Mounting Medium: sc-24941 or UltraCruz<sup>®</sup> Hard-set Mounting Medium: sc-359850.

## RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor EAR2 gene expression knockdown using RT-PCR Primer: EAR2 (h)-PR: sc-38822-PR (20  $\mu$ 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](http://www.scbt.com) for detailed protocols and support products.