



# DAX-1 siRNA (h): sc-35175

## BACKGROUND

Adrenal hypoplasia congenita (AHC) is an X-linked disorder characterized by primary adrenal insufficiency. The disorder, which is lethal if untreated, results in adrenal insufficiency early in infancy and is characterized by low serum concentration of glucocorticoids, mineralocorticoids and androgens and failure to respond to ACTH. AHC has been mapped to chromosome Xp21 at the same or close to an X-linked locus involved in sex determination, DSS (for dosage-sensitive sex reversal). The gene corresponding to DSS and AHC (designated DAX-1 for DSS-AHC critical region on the X chromosome, gene 1) has been cloned and shown to be deleted in AHC deletion patients and mutated in AHC non-deletion patients. The carboxy terminal 250 amino acids of the DAX-1-encoded protein, DAX-1, exhibits approximately 50% continuous similarity to the ligand-binding domain of the members of the nuclear hormone receptor superfamily while the amino terminal domain contains a putative DNA-binding motif. DAX-1 binds to retinoic acid responsive elements and down regulates retinoic acid receptor-mediated transcriptional activation.

## REFERENCES

1. Walker, A.P., et al. 1993. Isolation of the human Xp21 glycerol kinase gene by positional cloning. *Hum. Mol. Genet.* 2: 107-114.
2. Worley, K.C., et al. 1993. Yeast artificial chromosome cloning in the glycerol kinase and adrenal hypoplasia congenita region of Xp21. *Genomics* 16: 407-416.
3. Zanaria, E., et al. 1994. An unusual member of the nuclear hormone receptor superfamily responsible for X-linked adrenal hypoplasia congenita. *Nature* 372: 635-641.
4. Muscatelli, F., et al. 1994. Mutations in the DAX-1 gene give rise to both X-linked adrenal hypoplasia congenita and hypogonadotropic hypogonadism. *Nature* 372: 672-676.

## CHROMOSOMAL LOCATION

Genetic locus: NROB1 (human) mapping to Xp21.2.

## PRODUCT

DAX-1 siRNA (h) is a target-specific 19-25 nt siRNA 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 DAX-1 shRNA Plasmid (h): sc-35175-SH and DAX-1 shRNA (h) Lentiviral Particles: sc-35175-V as alternate gene silencing products.

## 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

DAX-1 shRNA (h) Lentiviral Particles is recommended for the inhibition of DAX-1 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

DAX-1 (3G8): sc-293452 is recommended as a control antibody for monitoring of DAX-1 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™ Molecular Weight Standards: sc-2035, UltraCruz® 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® Mounting Medium: sc-24941 or UltraCruz® Hard-set Mounting Medium: sc-359850.

## RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor DAX-1 gene expression knockdown using RT-PCR Primer: DAX-1 (h)-PR: sc-35175-PR (20  $\mu$ l, 476 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

## SELECT PRODUCT CITATIONS

1. Li, M., et al. 2014. MicroRNA-561 promotes acetaminophen-induced hepatotoxicity in Hep G2 cells and primary human hepatocytes through downregulation of the nuclear receptor corepressor dosage-sensitive sex-reversal adrenal hypoplasia congenital critical region on the X chromosome, gene 1 (DAX-1). *Drug Metab. Dispos.* 42: 44-61.

## 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.