

# Annexin IV siRNA (h): sc-29684

## BACKGROUND

The annexin family of calcium-binding proteins is composed of at least ten mammalian genes. It is characterized by a conserved core domain which binds to phospholipids in a  $Ca^{2+}$ -dependent manner and a unique amino terminal region which may confer binding specificity. Annexin family members have been implicated as regulators of such diverse processes as ion flux, endocytosis and exocytosis, and cellular adhesion. For example, the crystal structure of Annexin III has suggested a hydrophilic amino terminus with possible  $Ca^{2+}$  channel activity. Similarly, Annexin V has ion channel properties. Annexin IV, also referred to as endonexin, functions to regulate  $Cl^-$  flux by mediating calmodulin kinase II (CaMKII) activity and Annexin V has been shown to regulate PKC activity.

## REFERENCES

1. Smith, P.D., et al. 1994. Structural evolution of the annexin supergene family. *Trends Genet.* 10: 241-246.
2. Chan, H.C., et al. 1994. Annexin IV inhibits calmodulin-dependent protein kinase II-activated chloride conductance. A novel mechanism for ion channel regulation. *J. Biol. Chem.* 269: 32464-32468.
3. Rothhut, B., et al. 1995. Inhibitory effect of annexin V on protein kinase C activity in mesangial cell lysates. *Eur. J. Biochem.* 232: 865-872.

## CHROMOSOMAL LOCATION

Genetic locus: ANXA4 (human) mapping to 2p13.3.

## PRODUCT

Annexin IV 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 Annexin IV shRNA Plasmid (h): sc-29684-SH and Annexin IV shRNA (h) Lentiviral Particles: sc-29684-V as alternate gene silencing products.

For independent verification of Annexin IV (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-29684A, sc-29684B and sc-29684C.

## STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at  $-20^{\circ}$  C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at  $-20^{\circ}$  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

Annexin IV siRNA (h) is recommended for the inhibition of Annexin IV 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

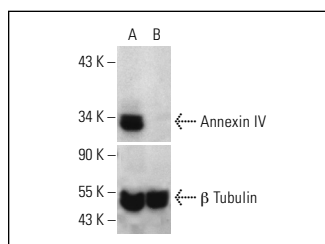
Annexin IV (D-2): sc-46693 is recommended as a control antibody for monitoring of Annexin IV gene expression knockdown by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000) or immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500).

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>TM</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 Annexin IV gene expression knockdown using RT-PCR Primer: Annexin IV (h)-PR: sc-29684-PR (20  $\mu$ l, 459 bp). Annealing temperature for the primers should be 55-60 $^{\circ}$  C and the extension temperature should be 68-72 $^{\circ}$  C.

## DATA



Annexin IV siRNA (h): sc-29684. Western blot analysis of Annexin IV expression in non-transfected control (A) and Annexin IV siRNA transfected (B) HeLa cells. Blot probed with Annexin IV (N-19): sc-1930.  $\beta$  Tubulin (D-10): sc-5274 used as specificity and loading control.

## RESEARCH USE

For research use only, not for use in diagnostic procedures.