SANTA CRUZ BIOTECHNOLOGY, INC.

Annexin V siRNA (h2): sc-44282

BACKGROUND

The Annexin family of calcium-binding proteins is composed of at least ten mammalian genes and is characterized by a conserved core domain, which binds phospholipids in a Ca^{2+}-dependent manner, and a unique amino-term-inal 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. Annexin V is ubiquitously expressed at high levels in tissues and cells grown in tissue culture, while Annexin VIII exhibits a more limited distribution. Where co-expressed in the same tissues, Annexin VIII is often expressed at a 100-fold lower level than Annexin V. However, Annexin VIII is preferentially expressed in acute promyelocytic leukemia (APL) cells, which may relate to its role in hematopoietic cell differentiation.

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. Reutelingsperger, C.P., et al. 1994. Differential tissue expression of Annexin VIII in human. FEBS Lett. 349: 120-124.
4. Liu, J.H., et al. 1994. Expression of the Annexin VIII gene in acute promyelocytic leukemia. Leuk. Lymphoma 13: 381-386.
5. 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.
6. Mailliard, W.S., et al. 1996. Calcium-dependent binding of S100C to the N-terminal domain of Annexin I. J. Biol. Chem. 271: 719-725.

CHROMOSOMAL LOCATION

Genetic locus: ANXA5 (human) mapping to 4q27.

PRODUCT

Annexin V siRNA (h2) 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Annexin V shRNA Plasmid (h2): sc-44282-SH and Annexin V shRNA (h2) Lentiviral Particles: sc-44282-V as alternate gene silencing products.

For independent verification of Annexin V (h2) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-44282A, sc-44282B and sc-44282C.

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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

Annexin V siRNA (h2) is recommended for the inhibition of Annexin V 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 µM in 66 µl. 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 V (H-3): sc-74438 is recommended as a control antibody for monitoring of Annexin V 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 Annexin V gene expression knockdown using RT-PCR Primer: Annexin V (h2)-PR: sc-44282-PR (20 µl). 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 for detailed protocols and support products.