SANTA CRUZ BIOTECHNOLOGY, INC.

ARC siRNA (h): sc-29722

BACKGROUND

ARC (apoptosis repressor with CARD domain), also designated nucleolar protein 3 (NOL3, NOP, NOP30) is a caspase-inhibiting protein that requires phosphorylation in order to prevent apoptosis. 5.5- and 1.0-kb ARC human transcripts are present in skeletal muscle and heart. Expression of the 1.0-kb transcript inhibits apoptosis in a dose-dependent manner when coexpressed with caspase-8. ARC interacts with caspase-2 and caspase-8 through its N-terminal death effector domain and is able to bind to caspase-8 in the mitochondria. ARC inhibits apoptosis induced by stimulation of CD95/FAS, tumor necrosis factor receptor-1 and TRAMP/death receptor-3. It is phosphorylated at Threonine 149 by CK2, and this phosphorylation targets ARC to mitochondria.

REFERENCES

1. Koseki, T., et al. 1998. ARC, an inhibitor of apoptosis expressed in skeletal muscle and heart that interacts selectively with caspases. Proc. Natl. Acad. Sci. USA 95: 5156-5160.
2. Stoss, O., et al. 1999. Alternative splicing determines the intracellular localization of the novel nuclear protein NOP30 and its interaction with the splicing factor SRp30c. J. Biol. Chem. 274: 10951-10962.
3. Li, P.F., et al. 2002. Phosphorylation by protein kinase CK2: a signaling switch for the caspase-inhibiting protein ARC. Mol. Cell 10: 247-258.
4. Ekhterae, D., et al. 2003. Apoptosis repressor with caspase domain inhibits cardiomyocyte apoptosis by reducing K^+ currents. Am. J. Physiol., Cell Physiol. 284: C1405-C1410.
5. Jo, D.G., et al. 2004. Calcium binding of ARC mediates regulation of caspase-8 and cell death. Mol. Cell. Biol. 24: 9763-9770.

CHROMOSOMAL LOCATION

Genetic locus: NOL3 (human) mapping to 16q22.1.

PRODUCT

ARC 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see ARC shRNA Plasmid (h): sc-29722-SH and ARC shRNA (h) Lentiviral Particles: sc-29722-V as alternate gene silencing products.

For independent verification of ARC (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-29722A, sc-29722B and sc-29722C.

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.

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

ARC siRNA (h) is recommended for the inhibition of ARC 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

ARC (F-11): sc-374177 is recommended as a control antibody for monitoring of ARC 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 ARC gene expression knockdown using RT-PCR Primer: ARC (h)-PR: sc-29722-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.