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ARAP1 siRNA (h): sc-44444

BACKGROUND

ARAP1 (ARFGAP, Rho GAP, Ankyrin repeat and pleckstrin homology domains-containing protein 1), also designated Centaurin $\delta 2$ (CENTD2), contains ARF GAP, Rho GAP, Ankyrin repeat, RAS-associating and pleckstrin homology domains. ARAP1 displays Rho GAP and phosphatidylinositol (3,4,5) trisphosphate (PIP3)-dependent ARFGAP activity. It associates with the Golgi, and the ARF GAP activity mediates changes in the Golgi and the formation of filopodia. The Rho GAP activity may mediate cell rounding and loss of stress fibers. At least three transcript variants encoding different isoforms have been found for this gene. ARAP1 can associate with G Protein receptor Angiotensin 1 (AT1) and influences recycling of the AT1 receptor to the plasma membrane. ARAP1 transcript levels are abundant in ovary, lung, liver and kidney. Northern blots indicate an ubiquitous 5.5 kb ARAP1 transcript and an additional 7 kb transcript present in heart and skeletal muscle.

REFERENCES

1. Nagase, T., et al. 1998. Prediction of the coding sequences of unidentified human genes. XI. The complete sequences of 100 new cDNA clones from brain which code for large proteins *in vitro*. DNA Res. 5: 277-286.
2. Jacques, K.M., et al. 2002. ARF1 dissociates from the clathrin adaptor GGA prior to being inactivated by ARF GTPase-activating proteins. J. Biol. Chem. 277: 47235-47241.
3. Miura, K., et al. 2002. ARAP1: a point of convergence for ARF and Rho signaling. Mol. Cell 9: 109-119.
4. Santy, L.C., et al. 2002. GTPase signaling: bridging the GAP between ARF and Rho. Curr. Biol. 12: R360-R362.
5. Guo, D.F., et al. 2003. Type 1 angiotensin II receptor-associated protein ARAP1 binds and recycles the receptor to the plasma membrane. Biochem. Biophys. Res. Commun. 310: 1254-1265.

CHROMOSOMAL LOCATION

Genetic locus: ARAP1 (human) mapping to 11q13.4.

PRODUCT

ARAP1 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see ARAP1 shRNA Plasmid (h): sc-44444-SH and ARAP1 shRNA (h) Lentiviral Particles: sc-44444-V as alternate gene silencing products.

For independent verification of ARAP1 (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-44444A, sc-44444B and sc-44444C.

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

APPLICATIONS

ARAP1 siRNA (h) is recommended for the inhibition of ARAP1 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 μ M in 66 μ 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

ARAP1 (A-3): sc-393138 is recommended as a control antibody for monitoring of ARAP1 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 κ BP-HRP: sc-516102 or m-IgG κ 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 κ BP-FITC: sc-516140 or m-IgG κ 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 ARAP1 gene expression knockdown using RT-PCR Primer: ARAP1 (h)-PR: sc-44444-PR (20 μ 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.