

APIP siRNA (m): sc-61977

BACKGROUND

APIP (Apaf-1-interacting protein), also known as AIP2, is a member of the aldolase class II family and has a highly conserved C-terminal from *C. elegans* to humans. It is ubiquitously expressed, with high expression levels in heart, kidney and skeletal muscle. Alternative splicing produces two isoforms of APIP. Isoform 1 is the full length, 242 amino acid protein; isoform 2 is missing residues 1-38 and contains a distinct sequence for amino acids 39-53. APIP plays an important role in preventing muscle ischemic damage. It suppresses hypoxia-induced cell death by inducing the activation of Akt and ERK 1/2, which are responsible for inhibition of caspase-9 via phosphorylation, and competing with caspase-9 to bind the caspase recruitment domain (CARD) of Apaf-1. Through these mechanisms, APIP negatively regulates the activation of caspase-9 and Apaf-1-mediated cell death.

REFERENCES

1. Lai, C.H., et al. 2000. Identification of novel human genes evolutionarily conserved in *Caenorhabditis elegans* by comparative proteomics. *Genome Res.* 10: 703-713.
2. Cao, G., et al. 2004. Cloning of a novel Apaf-1-interacting protein: a potent suppressor of apoptosis and ischemic neuronal cell death. *J. Neurosci.* 24: 6189-6201.
3. Cho, D.H., et al. 2004. Induced inhibition of ischemic/hypoxic injury by APIP, a novel Apaf-1-interacting protein. *J. Biol. Chem.* 279: 39942-39950.
4. Anichini, A., et al. 2006. APAF-1 signaling in human melanoma. *Cancer Lett.* 238: 168-179.
5. Schafer, Z.T., et al. 2006. The apoptosome: physiological, developmental, and pathological modes of regulation. *Dev. Cell* 10: 549-561.
6. Cho, D.H., et al. 2007. Suppression of hypoxic cell death by APIP-induced sustained activation of AKT and ERK1/2. *Oncogene* 26: 2809-2814.
7. LocusLink Report (LocusID: 51074). <http://www.ncbi.nlm.nih.gov/LocusLink/>

CHROMOSOMAL LOCATION

Genetic locus: Apip (mouse) mapping to 2 E2.

PRODUCT

APIP siRNA (m) is a pool of 2 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 APIP shRNA Plasmid (m): sc-61977-SH and APIP shRNA (m) Lentiviral Particles: sc-61977-V as alternate gene silencing products.

For independent verification of APIP (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-61977A and sc-61977B.

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

APIP siRNA (m) is recommended for the inhibition of APIP expression in mouse 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

APIP (G-2): sc-393194 is recommended as a control antibody for monitoring of APIP 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 APIP gene expression knockdown using RT-PCR Primer: APIP (m)-PR: sc-61977-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.