SANTA CRUZ BIOTECHNOLOGY, INC.

POPX2 siRNA (m): sc-62845

BACKGROUND

The phosphorylation and dephosphorylation of proteins on serine and threonine residues is an essential means of regulating a broad range of cellular functions in eukaryotes, including division, homeostasis and apoptosis. A group of proteins that are intimately involved in this process are the serine/threonine protein phosphatases. POPX1, also designated partner of PIX 1, PPM1E (protein phosphatase 1E) or PP2CH, and POPX2, also designated partner of PIX 2, PPM1F (protein phosphatase 1F), CaMKPase (CaM-kinase phosphatase), or FEM-2, belong to the PP2C family of serine/threonine phosphatases. Members of the PP2C family are negative regulators of cell stress response pathways. POPX2 is a ubiquitously expressed protein and POPX1 is predominantly expressed in brain and testis. POPX1 and POPX2 specifically interact with PIX (PAK interacting exchange factor) proteins and negatively regulate the activity of αPAK, a protein kinase that can lead to the breakdown of Actin stress fibers and other morphological changes. POPX2 can also interact with and regulate CaMKII activity. Overexpression of POPX2 can result in caspase-dependent apoptosis.

REFERENCES

1. Nomura, N., et al. 1994. Prediction of the coding sequences of unidentified human genes. I. The coding sequences of 40 new genes (KIAA0001-KIAA0040) deduced by analysis of randomly sampled cDNA clones from human immature myeloid cell line KG-1. DNA Res. 1: 27-35.
2. Kikuno, R., et al. 1999. Prediction of the coding sequences of unidentified human genes. XIV. The complete sequences of 100 new cDNA clones from brain which code for large proteins *in vitro*. DNA Res. 6: 197-205.
3. Tan, K.M., et al. 2001. The *Caenorhabditis elegans* sex-determining protein FEM-2 and its human homologue, hFEM-2, are Ca^{2+}/calmodulin-dependent protein kinase phosphatases that promote apoptosis. J. Biol. Chem. 276: 44193-44202.
4. Koh, C.G., et al. 2002. The p21-activated kinase PAK is negatively regulated by POPX1 and POPX2, a pair of serine/threonine phosphatases of the PP2C family. Curr. Biol. 12: 317-321.
5. Harvey, B.P., et al. 2004. Regulation of the multifunctional Ca^{2+}/calmodulin-dependent protein kinase II by the PP2C phosphatase PPM1F in fibroblasts. J. Biol. Chem. 279: 24889-24898.

CHROMOSOMAL LOCATION

Genetic locus: Ppm1f (mouse) mapping to 16 A3.

PRODUCT

POPX2 siRNA (m) 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 POPX2 shRNA Plasmid (m): sc-62845-SH and POPX2 shRNA (m) Lentiviral Particles: sc-62845-V as alternate gene silencing products.

For independent verification of POPX2 (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-62845A, sc-62845B and sc-62845C.

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

POPX2 siRNA (m) is recommended for the inhibition of POPX2 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 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

POPX2 (E-2): sc-514894 is recommended as a control antibody for monitoring of POPX2 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 POPX2 gene expression knockdown using RT-PCR Primer: POPX2 (m)-PR: sc-62845-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.