

PAP-2a siRNA (m): sc-72263

BACKGROUND

PAP-2 (phosphatidic acid phosphatase 2), also known as lipid phosphate phosphohydrolase (LPP), is a family of integral membrane glycoproteins that dephosphorylate a variety of lipid phosphates and play a role in signal transduction via the phospholipase D pathway. PAP-2 proteins function independently of Mg^{2+} and are insensitive to NEM (N-ethylmaleimide) inhibition. The lipid phosphates degraded by this family include ceramide 1-phosphate (C1P), sphingosine 1-phosphate (S1P), phosphatidic acid (PA) and lysophosphatidic acid (LPA). There are three PAP-2 isozymes: PAP-2a, PAP-2b and PAP-2c (also known as LPP1, LPP3 and LPP2 respectively). PAP-2a and PAP-2b are ubiquitously expressed and most effectively hydrolyze PA and LPA. PAP-2c is predominantly expressed in human brain, placenta and pancreas, and in mouse liver, lung and kidney. PAP-2c most effectively hydrolyzes LPA and S1P and may function as a cell cycle regulator.

REFERENCES

1. Roberts, R., et al. 1998. Human type 2 phosphatidic acid phosphohydrolases. Substrate specificity of the type 2a, 2b, and 2c enzymes and cell surface activity of the 2a isoform. *J. Biol. Chem.* 273: 22059-22067.
2. Nanjundan, M. and Possmayer, F. 2000. Characterization of the pulmonary N-ethylmaleimide-insensitive phosphatidate phosphohydrolase. *Exp. Lung Res.* 26: 361-381.
3. Pasquaré, S.J., et al. 2001. Aging promotes a different phosphatidic acid utilization in cytosolic and microsomal fractions from brain and liver. *Exp. Gerontol.* 36: 1387-1401.
4. Simon, M.F., et al. 2002. Expression of ectolipid phosphate phosphohydrolases in 3T3F442A preadipocytes and adipocytes. Involvement in the control of lysophosphatidic acid production. *J. Biol. Chem.* 277: 23131-23136.
5. Jia, Y.J., et al. 2003. Differential localization of lipid phosphate phosphatases 1 and 3 to cell surface subdomains in polarized MDCK cells. *FEBS Lett.* 552: 240-246.
6. Escalante-Alcalde, D., et al. 2003. The lipid phosphatase LPP3 regulates extra-embryonic vasculogenesis and axis patterning. *Development* 130: 4623-4637.

CHROMOSOMAL LOCATION

Genetic locus: Ppap2a (mouse) mapping to 13 D2.2.

PRODUCT

PAP-2a 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see PAP-2a shRNA Plasmid (m): sc-72263-SH and PAP-2a shRNA (m) Lentiviral Particles: sc-72263-V as alternate gene silencing products.

For independent verification of PAP-2a (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-72263A, sc-72263B and sc-72263C.

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

PAP-2a siRNA (m) is recommended for the inhibition of PAP-2a 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

PAP-2a (A-4): sc-515517 is recommended as a control antibody for monitoring of PAP-2a 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 PAP-2a gene expression knockdown using RT-PCR Primer: PAP-2a (m)-PR: sc-72263-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.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.