

p22-phox siRNA (h): sc-36149

BACKGROUND

Mox1 and the glycoprotein gp91-phox are largely related proteins that are essential components of the NADPH oxidase. The superoxide-generating NADPH oxidase is present in phagocytes, neuroepithelial bodies, vascular smooth muscle cells and endothelial cells. It includes a membrane-bound flavocytochrome containing two subunits, gp91-phox and p22-phox, and the cytosolic proteins p47-phox and p67-phox. During activation of the NADPH oxidase, p47-phox and p67-phox migrate to the plasma membrane, where they associate with the flavocytochrome cytochrome b558 to form the active enzyme complex. The p22- and gp91-phox subunits also function as surface O₂ sensors that initiate cellular signaling in response to hypoxic conditions.

CHROMOSOMAL LOCATION

Genetic locus: CYBA (human) mapping to 16q24.3.

PRODUCT

p22-phox 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 p22-phox shRNA Plasmid (h): sc-36149-SH and p22-phox shRNA (h) Lentiviral Particles: sc-36149-V as alternate gene silencing products.

For independent verification of p22-phox (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-36149A, sc-36149B and sc-36149C.

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

p22-phox siRNA (h) is recommended for the inhibition of p22-phox 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

p22-phox (E-11): sc-271968 is recommended as a control antibody for monitoring of p22-phox 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).

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor p22-phox gene expression knockdown using RT-PCR Primer: p22-phox (h)-PR: sc-36149-PR (20 µl, 419 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Edirimanne, V.E., et al. 2007. Homocysteine stimulates NADPH oxidase-mediated superoxide production leading to endothelial dysfunction in rats. *Can. J. Physiol. Pharmacol.* 85: 1236-1247.
2. Zhao, R., et al. 2009. Involvements of NADPH oxidase in oxidized LDL-induced upregulation of heat shock factor-1 and plasminogen activator inhibitor-1 in vascular endothelial cells. *Am. J. Physiol. Endocrinol. Metab.* 297: E104-E111.
3. Jeon, S.M., et al. 2010. NADPH oxidase is involved in protein kinase CKII down-regulation-mediated senescence through elevation of the level of reactive oxygen species in human colon cancer cells. *FEBS Lett.* 584: 3137-3142.
4. Ravuri, C., et al. 2011. Endogenous production of reactive oxygen species by the NADPH oxidase complexes is a determinant of γ-glutamyltransferase expression. *Free Radic. Res.* 45: 600-610.
5. Nitsche, C., et al. 2012. The phosphatase PHLPP1 regulates Akt2, promotes pancreatic cancer cell death, and inhibits tumor formation. *Gastroenterology* 142: 377-387.
6. Hajas, G., et al. 2013. 8-oxoguanine DNA glycosylase-1 links DNA repair to cellular signaling via the activation of the small GTPase Rac1. *Free Radic. Biol. Med.* 61: 384-394.
7. Pandur, S., et al. 2014. Combined incubation of colon carcinoma cells with phorbol ester and mitochondrial uncoupling agents results in synergic elevated reactive oxygen species levels and increased γ-glutamyltransferase expression. *Mol. Cell. Biochem.* 388: 149-156.
8. Kim, Y.M., et al. 2017. ROS-induced ROS release orchestrated by Nox4, Nox2, and mitochondria in VEGF signaling and angiogenesis. *Am. J. Physiol., Cell Physiol.* 312: C749-C764.
9. Vara, D., et al. 2018. Direct activation of NADPH oxidase 2 by 2-deoxyribose-1-phosphate triggers nuclear factor κB-dependent angiogenesis. *Antioxid. Redox Signal.* 28: 110-130.

RESEARCH USE

For research use only, not for use in diagnostic procedures.