



gp91-phox siRNA (r): sc-61838

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 O_2 sensors that initiate cellular signaling in response to hypoxic conditions. Mox1 and gp91 contain identical C-terminal sequence identity, yet they have distinct expression patterns. gp91-phox is expressed in eosinophils, neutrophils, monocytes and B-lymphocytes, whereas Mox1 is predominantly detected in the colon, and low expression is also detected in the uterus and prostate. Mox1 is also upregulated in vascular smooth-muscle cells in response to PDGF stimulation, which collectively indicates that Mox1 may function analogously to gp91-phox, yet regulate the NADPH superoxide production in non-phagocytic cells.

CHROMOSOMAL LOCATION

Genetic locus: Cybb (rat) mapping to Xq13.

PRODUCT

gp91-phox siRNA (r) 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 gp91-phox shRNA Plasmid (r): sc-61838-SH and gp91-phox shRNA (r) Lentiviral Particles: sc-61838-V as alternate gene silencing products.

For independent verification of gp91-phox (r) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-61838A, sc-61838B and sc-61838C.

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

gp91-phox siRNA (r) is recommended for the inhibition of gp91-phox expression in rat cells.

PROTOCOLS

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

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

gp91-phox (54.1): sc-130543 is recommended as a control antibody for monitoring of gp91-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 gp91-phox gene expression knockdown using RT-PCR Primer: gp91-phox (r)-PR: sc-61838-PR (20 μ l, 539 bp). Annealing temperature for the primers should be $55-60^\circ\text{C}$ and the extension temperature should be $68-72^\circ\text{C}$.

SELECT PRODUCT CITATIONS

- Shen, E., et al. 2009. Rac1 is required for cardiomyocyte apoptosis during hyperglycemia. *Diabetes* 58: 2386-2395.
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- Liu, X.H., et al. 2016. NADPH oxidase 4 contributes to connective tissue growth factor expression through Smad3-dependent signaling pathway. *Free Radic. Biol. Med.* 94: 174-184.
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RESEARCH USE

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