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Mox1 siRNA (r): sc-156079

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. 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.

REFERENCES

- Henderson, L.M., et al. 1995. The arachidonate-activable, NADPH oxidase-associated H⁺ channel. Evidence that gp91-phox functions as an essential part of the channel. *J. Biol. Chem.* 270: 5909-5916.
- Ushio-Fukai, M., et al. 1996. p22^{phox} is a critical component of the superoxide-generating NADH/NADPH oxidase system and regulates Angiotensin II-induced hypertrophy in vascular smooth muscle cells. *J. Biol. Chem.* 271: 23317-23321.

CHROMOSOMAL LOCATION

Genetic locus: Nox1 (rat) mapping to Xq32.

PRODUCT

Mox1 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 μM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Mox1 shRNA Plasmid (r): sc-156079-SH and Mox1 shRNA (r) Lentiviral Particles: sc-156079-V as alternate gene silencing products.

For independent verification of Mox1 (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-156079A, sc-156079B and sc-156079C.

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

Mox1 siRNA (r) is recommended for the inhibition of Mox1 expression in rat 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

Mox1 (C-10): sc-518023 is recommended as a control antibody for monitoring of Mox1 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 Mox1 gene expression knockdown using RT-PCR Primer: Mox1 (r)-PR: sc-156079-PR (20 μl, 529 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

- Ajayi, A., et al. 2015. Altered p53 and NOX1 activity cause bioenergetic defects in a SCA7 polyglutamine disease model. *Biochim. Biophys. Acta* 1847: 418-428.
- 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.

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.