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NMRAL1 siRNA (m): sc-150010

BACKGROUND

HSCARG, also known as NMRAL1 (NmrA-like family domain-containing protein 1), is a 299 amino acid redox sensor protein that belongs to the NmrA-type oxidoreductase family. Localizing primarily to the cytoplasm and perinuclear region, HSCARG localization to the nucleus may occur with increased intracellular nitric oxide and reduced NADPH/NADP⁺ ratios. Existing as a homodimer, HSCARG interacts with ASS1, inhibiting ASS1 activity in the presence of low NADPH/NADP⁺ ratios. HSCARG gets induced by nitric oxide, cGMP and proinflammatory cytokines. The gene encoding HSCARG maps to human chromosome 16p13.3 and mouse chromosome 16 A1. Overexpression of the gene encoding HSCARG results in increased cell viability.

REFERENCES

1. Martin, J., et al. 2004. The sequence and analysis of duplication-rich human chromosome 16. *Nature* 432: 988-994.
2. Dai, X., et al. 2006. Protein expression, crystallization and preliminary X-ray crystallographic studies on HSCARG from *Homo sapiens*. *Protein Pept. Lett.* 13: 955-957.
3. Zheng, X., et al. 2007. Restructuring of the dinucleotide-binding fold in an NAD(P)H sensor protein. *Proc. Natl. Acad. Sci. USA* 104: 8809-8814.
4. Zhao, Y., et al. 2008. An NADPH sensor protein (HSCARG) down-regulates nitric oxide synthesis by association with argininosuccinate synthetase and is essential for epithelial cell viability. *J. Biol. Chem.* 283: 11004-11013.
5. Lian, M. and Zheng, X. 2009. HSCARG regulates NFκB activation by promoting the ubiquitination of RelA or COMMD1. *J. Biol. Chem.* 284: 17998-18006.
6. Gan, Q., et al. 2009. HSCARG inhibits activation of NFκB by interacting with IκB kinase-β. *J. Cell Sci.* 122: 4081-4088.
7. Dai, X., et al. 2009. NADPH is an allosteric regulator of HSCARG. *J. Mol. Biol.* 387: 1277-1285.

CHROMOSOMAL LOCATION

Genetic locus: Nmr1 (mouse) mapping to 16 A1.

PRODUCT

NMRAL1 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 NMRAL1 shRNA Plasmid (m): sc-150010-SH and NMRAL1 shRNA (m) Lentiviral Particles: sc-150010-V as alternate gene silencing products.

For independent verification of NMRAL1 (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-150010A, sc-150010B and sc-150010C.

PROTOCOLS

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

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

NMRAL1 siRNA (m) is recommended for the inhibition of NMRAL1 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

HSCARG (D-1): sc-514369 is recommended as a control antibody for monitoring of NMRAL1 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 NMRAL1 gene expression knockdown using RT-PCR Primer: NMRAL1 (m)-PR: sc-150010-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.