

NOSIP siRNA (h): sc-45708

BACKGROUND

Endothelial nitric oxide synthase (eNOS) interacting protein (NOSIP) is a modulator of eNOS activity. eNOS is an important nitric oxide (NO)-generating enzyme of the vasculature that is regulated by interactions with caveolin-1, Ca²⁺-calmodulin, HSP 90 and NOSIP. NOSIP modulates this activity by promoting the translocation of eNOS from the plasma membrane to intracellular sites, which in turn inhibits NO synthesis. NOSIP is involved in controlling airway and vascular diameter, synthesis of NO in ciliated epithelia and mucosal secretion, and is an important protein for mucociliary and bronchial function. NOSIP is highly expressed in endothelial cells and vascularized tissue.

REFERENCES

1. Dedio, J., et al. 2001. NOSIP, a novel modulator of endothelial nitric oxide synthase activity. *FASEB J.* 15: 79-89.
2. Konig, P., et al. 2002. Distribution of the novel eNOS-interacting protein NOSIP in the liver, pancreas and gastrointestinal tract of the rat. *Gastroenterology* 123: 314-324.
3. Dreyer, J., et al. 2003. Spinal upregulation of the nitric oxide synthase-interacting protein NOSIP in a rat model of inflammatory pain. *Neurosci. Lett.* 350: 13-16.
4. Dreyer, J., et al. 2004. Nitric oxide synthase (NOS)-interacting protein interacts with neuronal NOS and regulates its distribution and activity. *J. Neurosci.* 24: 10454-10465.
5. Konig, P., et al. 2005. NOSIP and its interacting protein, eNOS, in the rat trachea and lung. *J. Histochem. Cytochem.* 53: 155-164.
6. Schleicher, M., et al. 2005. Cell cycle-regulated inactivation of endothelial NO synthase through NOSIP-dependent targeting to the cytoskeleton. *Mol. Cell. Biol.* 25: 8251-8258.

CHROMOSOMAL LOCATION

Genetic locus: NOSIP (human) mapping to 19q13.33.

PRODUCT

NOSIP 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see NOSIP shRNA Plasmid (h): sc-45708-SH and NOSIP shRNA (h) Lentiviral Particles: sc-45708-V as alternate gene silencing products.

For independent verification of NOSIP (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-45708A, sc-45708B and sc-45708C.

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.

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

NOSIP siRNA (h) is recommended for the inhibition of NOSIP 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 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

NOSIP (C-2): sc-365363 is recommended as a control antibody for monitoring of NOSIP 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 NOSIP gene expression knockdown using RT-PCR Primer: NOSIP (h)-PR: sc-45708-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.