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# NOSIP shRNA (h) Lentiviral Particles: sc-45708-V

## 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, Ca<sup>2+</sup>-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. König, 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. König, 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 shRNA (h) Lentiviral Particles is a pool of concentrated, transduction-ready viral particles containing 3 target-specific constructs that encode 19-25 nt (plus hairpin) shRNA designed to knock down gene expression. Each vial contains 200 µl frozen stock containing 1.0 x 10<sup>6</sup> infectious units of virus (IFU) in Dulbecco's Modified Eagle's Medium with 25 mM HEPES pH 7.3. Suitable for 10-20 transductions. Also see NOSIP siRNA (h): sc-45708 and NOSIP shRNA Plasmid (h): sc-45708-SH as alternate gene silencing products.

## RESEARCH USE

The purchase of this product conveys to the buyer the nontransferable right to use the purchased amount of the product and all replicates and derivatives for research purposes conducted by the buyer in his laboratory only (whether the buyer is an academic or for-profit entity). The buyer cannot sell or otherwise transfer (a) this product (b) its components or (c) materials made using this product or its components to a third party, or otherwise use this product or its components or materials made using this product or its components for Commercial Purposes.

## APPLICATIONS

NOSIP shRNA (h) Lentiviral Particles is recommended for the inhibition of NOSIP expression in human cells.

## SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 µl frozen viral stock containing 1.0 x 10<sup>6</sup> infectious units of virus (IFU); contains an shRNA construct encoding a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA.

## 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 (secondary) reagents are recommended: 1) Western Blotting: use goat anti-mouse IgG-HRP: sc-2005 (dilution range: 1:2000-1:32,000) or Cruz Marker™ compatible goat anti-mouse IgG-HRP: sc-2031 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use goat anti-mouse IgG-FITC: sc-2010 (dilution range: 1:100-1:400) or goat anti-mouse IgG-TR: sc-2781 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

## 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 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

## BIOSAFETY

Lentiviral particles can be employed in standard Biosafety Level 2 tissue culture facilities (and should be treated with the same level of caution as with any other potentially infectious reagent). Lentiviral particles are replication-incompetent and are designed to self-inactivate after transduction and integration of shRNA constructs into genomic DNA of target cells.

## STORAGE

Store lentiviral particles at -80° C. Stable for at least one year from the date of shipment. Once thawed, particles can be stored at 4° C for up to one week. Avoid repeated freeze thaw cycles.

## PROTOCOLS

See our web site at [www.scbt.com](http://www.scbt.com) or our catalog for detailed protocols and support products.