SANTA CRUZ BIOTECHNOLOGY, INC.

NOSTRIN siRNA (m): sc-72296

BACKGROUND

NOSTRIN (nitric oxide synthase trafficker isoform 1), also known as endothelial nitric oxide synthase traffic inducer, is a member of the pombe Cdc15 homology (PCH) family of proteins. NOSTRIN is expressed in the vascular endothelial cells of highly vascularized tissues such as placenta, lung, kidney and heart. It consists of an N-terminal Cdc15 domain with an FCH (Fes/CIP homology) region, two coiled coil domains and a C-terminal SH3 domain. NOSTRIN typically exists as a trimer. It functions as an adaptor protein binding to caveolin-1 via an internal domain and NOS3 via its SH3 domain, forming a ternary complex which facilitates caveolar transport of NOS3. The NOS3 protein is responsible for the production of nitric oxide (NO), a potent mediator in various biological processes. The translocation of NOS3 from the plasma membrane to intracellular vesicle-like structures diminishes NO production. NOSTRIN also interacts with Dynamin and N-WASP via its SH3 domain.

REFERENCES

1. Zimmermann, K., et al. 2002. NOSTRIN: a protein modulating nitric oxide release and subcellular distribution of endothelial nitric oxide synthase. Proc. Natl. Acad. Sci. USA 99: 17167-17172.
2. Choi, Y.J., et al. 2005. Cloning and characterization of mouse disabled 2 interacting protein 2, a mouse orthologue of human NOSTRIN. Biochem. Biophys. Res. Commun. 326: 594-599.
3. Icking, A., et al. 2005. NOSTRIN functions as a homotrimeric adaptor protein facilitating internalization of eNOS. J. Cell Sci. 118: 5059-5069.
4. Kim, H.W., et al. 2005. Mouse disabled 2 interacting protein 2 functions as a transcriptional repressor through direct binding onto its own promoter. Biochem. Biophys. Res. Commun. 337: 75-81.
5. Xiang, W., et al. 2005. Expression of endothelial nitric oxide synthase traffic inducer in the placentas of women with pre-eclampsia. Int. J. Gynaecol. Obstet. 89: 103-107.
6. Xiang, W., et al. 2006. Expression of endothelial nitric oxide synthase traffic inducer in the placenta of pregnancy induced hypertension. J. Huazhong Univ. Sci. Technol. Med. Sci. 26: 356-358.
7. Icking, A., et al. 2006. FCH/Cdc.domain determines distinct subcellular localization of NOSTRIN. FEBS Lett. 580: 223-228.

CHROMOSOMAL LOCATION

Genetic locus: Nostrin (mouse) mapping to 2 C2.

PRODUCT

NOSTRIN 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 NOSTRIN shRNA Plasmid (m): sc-72296-SH and NOSTRIN shRNA (m) Lentiviral Particles: sc-72296-V as alternate gene silencing products.

For independent verification of NOSTRIN (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-72296A, sc-72296B and sc-72296C.

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

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

NOSTRIN (B-9): sc-373954 is recommended as a control antibody for monitoring of NOSTRIN 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 NOSTRIN gene expression knockdown using RT-PCR Primer: NOSTRIN (m)-PR: sc-72296-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.

PROTOCOLS

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