SANTA CRUZ BIOTECHNOLOGY, INC.

NOS2 siRNA (m): sc-36092

BACKGROUND

Nitric oxide (NO) has a broad range of biological activities and has been implicated in signaling pathways in phylogenetically diverse species. Nitric oxide synthases (NOSs), the enzymes responsible for synthesis of NO, contain an N-terminal oxygenase domain and a C-terminal reductase domain. NOS activity requires homodimerization as well as three cosubstrates (L-arginine, NADPH and O_2) and five cofactors or prosthetic groups (FAD, FMN, calmodulin, tetrahydrobiopterin and heme). Several distinct NOS isoforms have been described and been shown to represent the products of three distinct genes. These include two constitutive Ca^{2+}/CaM-dependent forms of NOS, including NOS1 (also designated ncNOS) whose activity was first identified in neurons, and NOS3 (also designated ecNOS), first identified in endothelial cells. The inducible form of NOS, NOS2 (also designated iNOS), is Ca^{2+}-independent and is expressed in a broad range of cell types.

REFERENCES

1. Nathan, C., et al. 1994. Nitric oxide synthases: roles, tolls, and controls. Cell 78: 915-918.
2. Heiss, L.N., et al. 1994. Epithelial autotoxicity of nitric oxide: role in the respiratory cytopathology of pertussis. Proc. Natl. Acad. Sci. USA 91: 267-270.
3. Farias-Eisner, R., et al. 1994. Nitric oxide is an important mediator for tumoricidal activity *in vivo*. Proc. Natl. Acad. Sci. USA 91: 9407-9411.

CHROMOSOMAL LOCATION

Genetic locus: Nos2 (mouse) mapping to 11 B5.

PRODUCT

NOS2 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 NOS2 shRNA Plasmid (m): sc-36092-SH and NOS2 shRNA (m) Lentiviral Particles: sc-36092-V as alternate gene silencing products.

For independent verification of NOS2 (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-36092A, sc-36092B and sc-36092C.

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

NOS2 siRNA (m) is recommended for the inhibition of NOS2 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

NOS2 (C-11): sc-7271 is recommended as a control antibody for monitoring of NOS2 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).

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor NOS2 gene expression knockdown using RT-PCR Primer: NOS2 (m)-PR: sc-36092-PR (20 µl, 460 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Huang, Z., et al. 2012. Stimulation of unprimed macrophages with immune complexes triggers a low output of nitric oxide by calcium-dependent neuronal nitric-oxide synthase. J. Biol. Chem. 287: 4492-4502.
2. Kumar, A., et al. 2014. Inducible nitric oxide synthase is key to peroxynitrite-mediated, LPS-induced protein radical formation in murine microglial BV2 cells. Free Radic. Biol. Med. 73: 51-59.
3. Hwang, H.J., et al. 2015. Improved islet transplantation outcome by the co-delivery of siRNAs for iNOS and 17β-estradiol using an R3V6 peptide carrier. Biomaterials 38: 36-42.
4. Li, J.T., et al. 2016. Subanesthetic isoflurane relieves zymosan-induced neutrophil inflammatory response by targeting NMDA glutamate receptor and Toll-like receptor 2 signaling. Oncotarget 7: 31772-31789.
5. Lee, H.J., et al. 2017. Hydrogen sulfide inhibits high glucose-induced NADPH oxidase 4 expression and matrix increase by recruiting inducible nitric oxide synthase in kidney proximal tubular epithelial cells. J. Biol. Chem. 292: 5665-5675.
6. Kapralov, A.A., et al. 2020. Redox lipid reprogramming commands susceptibility of macrophages and microglia to ferroptotic death. Nat. Chem. Biol. 16: 278-290.
7. Maher, P. 2021. Investigations into the role of metabolism in the inflammatory response of BV2 microglial cells. Antioxidants 10: 109.

RESEARCH USE

For research use only, not for use in diagnostic procedures.