SANTA CRUZ BIOTECHNOLOGY, INC.

NOD9 siRNA (h): sc-96800

BACKGROUND

The leucine-rich repeat (LRR) is a 20-30 amino acid motif that forms a hydrophobic α/β horseshoe fold, allowing it to accommodate several leucine residues within a tightly packed core. All LRRs contain a variable segment and a highly conserved segment, the latter of which accounts for 11 or 12 residues of the entire LRR motif. NOD9, also known as NLRX1, NOD26 or NOD5, is a 975 amino acid outer mitochondrial membrane protein that contains one NACHT domain and four LRRs. Expressed at high levels in heart, muscle and mammary gland, NOD9 plays a role in antiviral signaling, specifically via inhibition of virus-induced helicases, thereby acting as a negative regulator of antiviral responses. Two isoforms of NOD9 exist due to alternative splicing events.

REFERENCES

1. Inohara, N. and Nuñez, G. 2003. NODs: intracellular proteins involved in inflammation and apoptosis. Nat. Rev. Immunol. 3: 371-382.
2. Inohara, C. and Nuñez, G. 2005. NOD-LRR proteins: role in host-microbial interactions and inflammatory disease. Annu. Rev. Biochem. 74: 355-383.
3. O'Neill, L.A. 2008. Innate immunity: squelching anti-viral signalling with NLRX1. Curr. Biol. 18: R302-R304.
4. Komuro, A., et al. 2008. Negative regulation of cytoplasmic RNA-mediated antiviral signaling. Cytokine 43: 350-358.
5. Meylan, E. and Tschopp, J. 2008. NLRX1: friend or foe? EMBO Rep. 9: 243-245.
6. Moore, C.B., et al. 2008. NLRX1 is a regulator of mitochondrial antiviral immunity. Nature 451: 573-577.

CHROMOSOMAL LOCATION

Genetic locus: NLRX1 (human) mapping to 11q23.3.

PRODUCT

NOD9 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see NOD9 shRNA Plasmid (h): sc-96800-SH and NOD9 shRNA (h) Lentiviral Particles: sc-96800-V as alternate gene silencing products.

For independent verification of NOD9 (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-96800A, sc-96800B and sc-96800C.

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

NOD9 siRNA (h) is recommended for the inhibition of NOD9 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 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

NOD9 (F-2): sc-374514 is recommended as a control antibody for monitoring of NOD9 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 NOD9 gene expression knockdown using RT-PCR Primer: NOD9 (h)-PR: sc-96800-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Li, H., et al. 2016. NLRX1 attenuates apoptosis and inflammatory responses in myocardial ischemia by inhibiting MAVS-dependent NLRP3 inflammasome activation. Mol. Immunol. 76: 90-97.

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.