

TLR9 siRNA (m): sc-40271

BACKGROUND

The Toll-like receptors (TLR) are a family of human receptors that share homology with the *Drosophila* Toll receptors, which are involved in mediating dorsoventral polarization in developing *Drosophila* embryos and participate in host immunity. The TLR family members are characterized by a highly conserved Toll homology (TH) domain, which is essential for Toll-induced signal transductions. TLRs are type I transmembrane receptors that contain an extracellular domain consisting of several leucine-rich regions and a single cytoplasmic Toll/IL-1R like domain. Three TLR family members, TLR7, TLR8 and TLR9, belong to a subfamily of TLRs which are differentially expressed. TLR7 is expressed in lung, placenta and spleen. TLR8 is expressed in lung and peripheral blood leukocytes, and TLR9 is predominantly expressed in spleen, lymph nodes, bone marrow and peripheral blood leukocytes. TLR7, TLR8 and TLR9 stimulate the NF κ B signaling pathway, suggesting that they play a role in the immune response.

REFERENCES

1. Gay, N.J. and Keith, F.J. 1991. *Drosophila* Toll and IL-1 receptor. Nature 351: 355-356.
2. Rock, F.L., et al. 1998. A family of human receptors structurally related to *Drosophila* Toll. Proc. Natl. Acad. Sci. USA 95: 588-593.
3. Brightbill, H.D., et al. 1999. Host defense mechanisms triggered by microbial lipoproteins through Toll-like receptors. Science 285: 732-736.

CHROMOSOMAL LOCATION

Genetic locus: Tlr9 (mouse) mapping to 9 F1.

PRODUCT

TLR9 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see TLR9 shRNA Plasmid (m): sc-40271-SH and TLR9 shRNA (m) Lentiviral Particles: sc-40271-V as alternate gene silencing products.

For independent verification of TLR9 (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-40271A, sc-40271B and sc-40271C.

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

TLR9 siRNA (m) is recommended for the inhibition of TLR9 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 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

TLR9 (5G5): sc-47723 is recommended as a control antibody for monitoring of TLR9 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 λ BP-HRP: sc-516132 or m-IgG λ BP-HRP (Cruz Marker): sc-516132-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 λ BP-FITC: sc-516185 or m-IgG λ BP-PE: sc-516186 (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 TLR9 gene expression knockdown using RT-PCR Primer: TLR9 (m)-PR: sc-40271-PR (20 μ l, 532 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Chen, M., et al. 2011. Blockade of TLR9 signaling in B cells impaired anti-dsDNA antibody production in mice induced by activated syngenic lymphocyte-derived DNA immunization. Mol. Immunol. 48: 1532-1539.
2. Levenson, E.A. and Kiick, K.L. 2014. DNA-polymer conjugates for immune stimulation through Toll-like receptor 9 mediated pathways. Acta Biomater. 10: 1134-1145.
3. Gao, Y., et al. 2020. Mitochondrial DNA from hepatocytes induces upregulation of interleukin-33 expression of macrophages in nonalcoholic steatohepatitis. Dig. Liver Dis. 52: 637-643.
4. Zhang, L. and Li, X. 2020. Toll-like receptor-9 (TLR-9) deficiency alleviates optic nerve injury (ONI) by inhibiting inflammatory response *in vivo* and *in vitro*. Exp. Cell Res. 396: 112159.

RESEARCH USE

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