

IRAK-1 siRNA (m): sc-35705

BACKGROUND

Three structurally related ligands for IL-1Rs have been described. These include two agonists, IL-1 α and IL-1 β , and a specific receptor antagonist, IL-1Ra. Two distinct receptors designated IL-1RI and IL-1RII have been identified, each of which belong to the Ig superfamily. The preponderance of evidence suggests IL-1RI to be the functional IL-1 receptor. Binding of IL-1 to its cognate receptor results in the activation of the NF κ B signaling pathway. The IL-1-dependent kinase termed IRAK (for IL-1 receptor-associated kinase) co-immunoprecipitates with activated IL-1RI and has been implicated as an upstream mediator of NF κ B activation. Additional support for this assertion comes from the fact that a related *Drosophila* protein, Pelle, is a known upstream activator of Dorsal, the *Drosophila* homolog of NF κ B.

CHROMOSOMAL LOCATION

Genetic locus: Irak1 (mouse) mapping to X A7.3.

PRODUCT

IRAK-1 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 IRAK-1 shRNA Plasmid (m): sc-35705-SH and IRAK-1 shRNA (m) Lentiviral Particles: sc-35705-V as alternate gene silencing products.

For independent verification of IRAK-1 (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-35705A, sc-35705B and sc-35705C.

STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at -20 $^{\circ}$ C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at -20 $^{\circ}$ 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

IRAK-1 siRNA (m) is recommended for the inhibition of IRAK-1 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

IRAK-1 (F-4): sc-5288 is recommended as a control antibody for monitoring of IRAK-1 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 κ BP-HRP: sc-516102 or m-IgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz MarkerTM Molecular Weight Standards: sc-2035, UltraCruz[®] Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG κ BP-FITC: sc-516140 or m-IgG κ 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 IRAK-1 gene expression knockdown using RT-PCR Primer: IRAK-1 (m)-PR: sc-35705-PR (20 μ l). Annealing temperature for the primers should be 55-60 $^{\circ}$ C and the extension temperature should be 68-72 $^{\circ}$ C.

SELECT PRODUCT CITATIONS

- Chadha, A., et al. 2015. Suppressive role of neddylation in dendritic cells during *Mycobacterium tuberculosis* infection. *Tuberculosis* 95: 599-607.
- Antony, C., et al. 2015. Regulation of L-type voltage gated calcium channel CACNA1S in macrophages upon *Mycobacterium tuberculosis* infection. *PLoS ONE* 10: e0124263.
- Vashishta, M., et al. 2015. Pneumococcal surface protein A (PspA) regulates programmed death ligand 1 expression on dendritic cells in a Toll-like receptor 2 and calcium dependent manner. *PLoS ONE* 10: e0133601.
- Guo, L., et al. 2015. IRAK1 mediates TLR4-induced ABCA1 downregulation and lipid accumulation in VSMCs. *Cell Death Dis.* 6: e1949.
- Lee, K.H., et al. 2018. Globular adiponectin exerts a pro-inflammatory effect via I κ B/NF κ B pathway activation and anti-inflammatory effect by IRAK-1 downregulation. *Mol. Cells* 41: 762-770.
- Liu, M., et al. 2020. Macrophage K63-linked ubiquitination of YAP promotes its nuclear localization and exacerbates atherosclerosis. *Cell Rep.* 32: 107990.

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.