

# IRAK-M siRNA (h): sc-39098

## BACKGROUND

Interleukin-1 receptor (IL1R)-associated kinases (IRAKs) are important mediators in the signal transduction of Toll-like receptor (TLR) and IL1R family members, collectively referred to as TIRs. Binding of IL-1 to its cognate receptor results in the activation of the NF $\kappa$ B signaling pathway. An IL-1-dependent kinase termed IRAK-1 (for IL-1 receptor-associated kinase 1) coimmunoprecipitates with activated IL-1RI and is implicated as an upstream mediator of NF $\kappa$ B activation. A related *Drosophila* protein, Pelle, is a known upstream activator of Dorsal, the *Drosophila* homolog of NF $\kappa$ B. IRAK-2 is a proximal mediator of IL1, a component of the IL1R signaling complex, and is required for IL1R-induced NF $\kappa$ B activation. IRAK-4, like IRAK-1 and Pelle, has auto- and cross-phosphorylation kinase activity. IRAK-4 is strongly expressed in kidney and is also found in lung, testis, small intestine, breast, liver, and placenta. In contrast to the other IRAKs that are expressed in most cell types, IRAK-M is restricted to monocytic cells. IRAK-M mRNA transcripts are found predominantly in PBL and the monocytic cell lines U937 and THP-1.

## REFERENCES

1. Croston, G.E., et al. 1995. NF $\kappa$ B activation by interleukin-1 (IL-1) requires an IL-1 receptor-associated protein kinase activity. *J. Biol. Chem.* 270: 16514-16517.
2. Cao, Z., et al. 1996. IRAK: a kinase associated with the interleukin-1 receptor. *Science* 271: 1128-1131.
3. Muzio, M., et al. 1997. IRAK (Pelle) family member IRAK-2 and MyD88 as proximal mediators of IL-1 signaling. *Science* 278: 1612-1615.
4. Scanlan, M.J., et al. 1999. Antigens recognized by autologous antibody in patients with renal-cell carcinoma. *Int. J. Cancer* 83: 456-464.

## CHROMOSOMAL LOCATION

Genetic locus: IRAK3 (human) mapping to 12q14.3.

## PRODUCT

IRAK-M 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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see IRAK-M shRNA Plasmid (h): sc-39098-SH and IRAK-M shRNA (h) Lentiviral Particles: sc-39098-V as alternate gene silencing products.

For independent verification of IRAK-M (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-39098A, sc-39098B and sc-39098C.

## 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  $\mu$ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330  $\mu$ l of RNase-free water makes a 10  $\mu$ M solution in a 10  $\mu$ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

## APPLICATIONS

IRAK-M siRNA (h) is recommended for the inhibition of IRAK-M 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  $\mu$ M in 66  $\mu$ 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-M (XX-6): sc-100389 is recommended as a control antibody for monitoring of IRAK-M 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 $\kappa$  BP-HRP: sc-516102 or m-IgG $\kappa$  BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker<sup>™</sup> Molecular Weight Standards: sc-2035, UltraCruz<sup>®</sup> Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG $\kappa$  BP-FITC: sc-516140 or m-IgG $\kappa$  BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz<sup>®</sup> Mounting Medium: sc-24941 or UltraCruz<sup>®</sup> Hard-set Mounting Medium: sc-359850.

## RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor IRAK-M gene expression knockdown using RT-PCR Primer: IRAK-M (h)-PR: sc-39098-PR (20  $\mu$ l, 479 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

## SELECT PRODUCT CITATIONS

1. Watanabe, T., et al. 2008. Muramyl dipeptide activation of nucleotide-binding oligomerization domain 2 protects mice from experimental colitis. *J. Clin. Invest.* 118: 545-559.
2. Chung, H., et al. 2010. Hepatitis C virus core protein induces homotolerance and cross-tolerance to Toll-like receptor ligands by activation of Toll-like receptor 2. *J. Infect. Dis.* 202: 853-861.
3. Al-Qahtani, A.A., et al. 2017. Middle east respiratory syndrome corona virus spike glycoprotein suppresses macrophage responses via DPP4-mediated induction of IRAK-M and PPAR $\gamma$ . *Oncotarget* 8: 9053-9066.

## RESEARCH USE

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