

# IRAK-1 siRNA (h): sc-35704

## BACKGROUND

Three structurally related ligands for IL-1Rs have been described. These include two agonists, IL-1 $\alpha$  and IL-1 $\beta$ , 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 $\kappa$ 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 $\kappa$ 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 $\kappa$ B.

## CHROMOSOMAL LOCATION

Genetic locus: IRAK1 (human) mapping to Xq28.

## PRODUCT

IRAK-1 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-1 shRNA Plasmid (h): sc-35704-SH and IRAK-1 shRNA (h) Lentiviral Particles: sc-35704-V as alternate gene silencing products.

For independent verification of IRAK-1 (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-35704A, sc-35704B and sc-35704C.

## 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-1 siRNA (h) is recommended for the inhibition of IRAK-1 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-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 $\kappa$  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-1 gene expression knockdown using RT-PCR Primer: IRAK-1 (h)-PR: sc-35704-PR (20  $\mu$ l, 431 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

## SELECT PRODUCT CITATIONS

1. Park, H., et al. 2015. MicroRNA-146a and microRNA-146b regulate human dendritic cell apoptosis and cytokine production by targeting TRAF6 and IRAK-1 proteins. *J. Biol. Chem.* 290: 2831-2841.
2. Kanda, T., et al. 2015. Interleukin(IL)-36 $\alpha$  and IL-36 $\gamma$  induce proinflammatory mediators from human colonic subepithelial myofibroblasts. *Front. Med.* 2: 69.
3. Mehto, S., et al. 2015. *Mycobacterium tuberculosis* and human immunodeficiency virus type 1 cooperatively modulate macrophage apoptosis via Toll like receptor 2 and calcium homeostasis. *PLoS ONE* 10: e0131767.
4. Rana, M., et al. 2016. IRAK regulates macrophage foam cell formation by modulating genes involved in cholesterol uptake and efflux. *Bioessays* 38: 591-604.
5. Nishida, A., et al. 2017. Interleukin-36 $\alpha$  induces inflammatory mediators from human pancreatic myofibroblasts via a MyD88 dependent pathway. *Pancreas* 46: 539-548.
6. Li, H.N., et al. 2019. miR-146a-5p suppresses ATP-binding cassette subfamily G member 1 dysregulation in patients with refractory *Mycoplasma pneumoniae* via interleukin 1 receptor-associated kinase 1 downregulation. *Int. J. Mol. Med.* 44: 2003-2014.

## RESEARCH USE

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

## PROTOCOLS

See our web site at [www.scbt.com](http://www.scbt.com) for detailed protocols and support products.