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ILT-2 siRNA (h): sc-42812

BACKGROUND

Leukocyte immunoglobulin-like receptors (LIRs) are members of the immunoglobulin superfamily of glycoproteins and are predominantly expressed by monocytes, B cells, dendritic cells, natural killer (NK) cells, peripheral blood leukocytes and tissues such as placenta, lung and liver. Immunoglobulin-like transcript 2 (ILT-2), also known as CD85 or MIR7, is a 650 amino acid glycoprotein that contains a 23 amino acid signal peptide, four extracellular C2-type IGSF domains and four intracellular ITIM motifs. ILT-2 can bind major histocompatibility (MHC) class I molecules and inhibit cell termination by natural killer (NK) and T cells, and inhibit Ca²⁺ mobilization in myeloid cells triggered through the B cell antigen receptor and histocompatibility leukocyte antigens (HLA)-DR. ILT-2 contains four putative cytoplasmic tyrosine-based inhibitory motifs and upon tyrosine phosphorylation, associates with the tyrosine phosphatase SHP-1.

REFERENCES

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- Colonna, M., et al. 1997. A common inhibitory receptor for major histocompatibility complex class I molecules on human lymphoid and myelomonocytic cells. *J. Exp. Med.* 186: 1809-1818.
- Cosman, D., et al. 1997. A novel immunoglobulin superfamily receptor for cellular and viral MHC class I molecules. *Immunity* 7: 273-282.
- Online Mendelian Inheritance in Man, OMIM™. 2000. Johns Hopkins University, Baltimore, MD. MIM Number: 604811. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
- Dietrich, J., et al. 2001. Ig-like transcript 2 (ILT-2)/leukocyte Ig-like receptor 1 (LIR1) inhibits TCR signaling and Actin cytoskeleton reorganization. *J. Immunol.* 166: 2514-2521.
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CHROMOSOMAL LOCATION

Genetic locus: LILRB1 (human) mapping to 19q13.42.

PRODUCT

ILT-2 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 ILT-2 shRNA Plasmid (h): sc-42812-SH and ILT-2 shRNA (h) Lentiviral Particles: sc-42812-V as alternate gene silencing products.

For independent verification of ILT-2 (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-42812A, sc-42812B and sc-42812C.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.

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

ILT-2 siRNA (h) is recommended for the inhibition of ILT-2 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

ILT-2 (VMP55): sc-20065 is recommended as a control antibody for monitoring of ILT-2 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_k BP-HRP: sc-516102 or m-IgG_k 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-IgG_k BP-FITC: sc-516140 or m-IgG_k 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 ILT-2 gene expression knockdown using RT-PCR Primer: ILT-2 (h)-PR: sc-42812-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

RESEARCH USE

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