

# ULBP4 siRNA (h): sc-63183

## BACKGROUND

The immune system contains genetically encoded subsystems, which monitor the extracellular environment in order to eliminate pathogens and resolve abnormal or transformed tissues. Cytomegalovirus UL16 binding proteins, known as ULBPs, are GPI-linked glycoproteins that belong to the extended MHC class I family and are distantly related to MHC class I polypeptide-related sequence B, known as MICB. ULBP and MICB proteins are ligands for the activating receptor NKG2D/DAP10, which causes lymphocyte activation resulting in the secretion of cytokines, such as interferon- $\gamma$ , and tumor cell lysis. The interaction of ULBP or MICB with NKG2D/DAP10 can be blocked by the soluble form of UL16. ULBPs stimulate cytokine and chemokine production from NK cells, CD8  $\alpha/\beta$  T cells and  $\gamma/\delta$  T cells. Soluble forms of ULBPs induce protein tyrosine phosphorylation and activation of the Janus kinase 2, Stat5, extracellular signal-regulated kinase, mitogen-activated protein kinase and phosphatidylinositol 3-kinase (PI 3-kinase)/Akt signal transduction pathways.

## REFERENCES

1. Kubin, M., Cassiano, L., Chalupny, J., Chin, W., Cosman, D., Fanslow, W., Mullberg, J., Rousseau, A.M., Ulrich, D. and Armitage, R. 2001. ULBP1, 2, 3: novel MHC class I-related molecules that bind to human cytomegalovirus glycoprotein UL16, activate NK cells. *Eur. J. Immunol.* 31: 1428-1437.
2. Cosman, D., Mullberg, J., Sutherland, C.L., Chin, W., Armitage, R., Fanslow, W., Kubin, M. and Chalupny, N.J. 2001. ULBPs, novel MHC class I-related molecules, bind to CMV glycoprotein UL16 and stimulate NK cytotoxicity through the NKG2D receptor. *Immunity* 14: 123-133.
3. Steinle, A., Li, P., Morris, D.L., Groh, V., Lanier, L.L., Strong, R.K. and Spies, T. 2001. Interactions of human NKG2D with its ligands MICA, MICB, and homologs of the mouse RAE-1 protein family. *Immunogenetics* 53: 279-287.
4. Sutherland, C.L., Chalupny, N.J., Schooley, K., VandenBos, T., Kubin, M. and Cosman, D. 2002. UL16-binding proteins, novel MHC class I-related proteins, bind to NKG2D and activate multiple signaling pathways in primary NK cells. *J. Immunol.* 168: 671-679.
5. LocusLink Report (LocusID: 4277). <http://www.ncbi.nlm.nih.gov/LocusLink/>

## CHROMOSOMAL LOCATION

Genetic locus: RAET1E (human) mapping to 6q25.1.

## PRODUCT

ULBP4 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 ULBP4 shRNA Plasmid (h): sc-63183-SH and ULBP4 shRNA (h) Lentiviral Particles: sc-63183-V as alternate gene silencing products.

For independent verification of ULBP4 (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-63183A, sc-63183B and sc-63183C.

## 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

ULBP4 siRNA (h) is recommended for the inhibition of ULBP4 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

ULBP4 (6E6): sc-53133 is recommended as a control antibody for monitoring of ULBP4 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 ULBP4 gene expression knockdown using RT-PCR Primer: ULBP4 (h)-PR: sc-63183-PR (20  $\mu$ l). 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.

## PROTOCOLS

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