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Ox40L siRNA (h): sc-42824

BACKGROUND

The Ox40 ligand, Ox40L (also designated gp34), is a type II membrane protein, due to the absence of a signal peptide. Ox40L, a member of the tumor necrosis factor (TNF) superfamily, is a costimulatory molecule involved in dendritic cell:T cell interactions, T cell homing and B cell activation. Engagement of Ox40L with its receptor, Ox40, delivers a strong costimulatory signal to effector T cells. Ox40L is found preferentially on activated B cells and its receptor, Ox40, is a member of the tumor necrosis factor receptor (TNFR) family that is expressed on activated T cells. Ox40L plays a critical role in antigen-specific T cell responses *in vivo* and in both the priming and effector phases of T cell activation when expressed on antigen-presenting cells (APCs).

REFERENCES

- Hahne, M., et al. 1998. APRIL, a new ligand of the tumor necrosis factor family, stimulates tumor cell growth. *J. Exp. Med.* 188: 1185-1190.
- Chen, A.I., et al. 1999. Ox40-ligand has a critical costimulatory role in dendritic cell:T cell interactions. *Immunity* 11: 689-698.
- Matsumura, Y., et al. 1999. Intracellular signaling of gp34, the Ox40 ligand: induction of c-Jun and c-Fos mRNA expression through gp34 upon binding of its receptor, Ox40. *J. Immunol.* 163: 3007-3011.
- Weinberg, A.D., et al. 2000. Engagement of the Ox40 receptor *in vivo* enhances antitumor immunity. *J. Immunol.* 164: 2160-2169.
- Morimoto, S., et al. 2000. CD134L engagement enhances human B cell Ig production: CD154/CD40, CD70/CD27 and CD134/CD134L interactions coordinately regulate T cell-dependent B cell responses. *J. Immunol.* 164: 4097-4104.
- Murata, K., et al. 2000. Impairment of antigen-presenting cell function in mice lacking expression of Ox40 ligand. *J. Exp. Med.* 191: 365-374.
- Malmstrom, V., et al. 2001. CD134L expression on dendritic cells in the mesenteric lymph nodes drives colitis in T cell-restored SCID mice. *J. Immunol.* 166: 6972-6981.

CHROMOSOMAL LOCATION

Genetic locus: TNFSF4 (human) mapping to 1q25.1.

PRODUCT

Ox40L 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Ox40L shRNA Plasmid (h): sc-42824-SH and Ox40L shRNA (h) Lentiviral Particles: sc-42824-V as alternate gene silencing products.

For independent verification of Ox40L (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-42824A, sc-42824B and sc-42824C.

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 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

Ox40L siRNA (h) is recommended for the inhibition of Ox40L 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 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

Ox40L (RR19): sc-80271 is recommended as a control antibody for monitoring of Ox40L 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 Marker™ 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 Ox40L gene expression knockdown using RT-PCR Primer: Ox40L (h)-PR: sc-42824-PR (20 μ 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.