

▶ BTLA siRNA (h): sc-45458

BACKGROUND

B and T lymphocyte attenuator (BTLA) is an immunoglobulin domain-containing glycoprotein whose expression is induced during T cell activation. BTLA is similar to CTLA-4 and PD-1, all of which are co-inhibitory receptors belonging to the CD28 family. However, unlike CTLA-4 and PD-1, BTLA does not interact with B7-Ig family counter receptors. Rather, the herpesvirus entry mediator (HVEM), a TNF receptor, acts as a molecular switch that modulates T cell activation by propagating inhibitory signals through BTLA. The BTLA-HVEM interaction is conserved between mouse and human, suggesting that this system is an important pathway regulating lymphocyte activation.

REFERENCES

1. Watanabe, N., et al. 2003. BTLA is a lymphocyte inhibitory receptor with similarities to CTLA-4 and PD-1. *Nat. Immunol.* 4: 670-679.
2. Gonzalez, L.C., et al. 2005. A coreceptor interaction between the CD28 and TNF receptor family members B and T lymphocyte attenuator and herpesvirus entry mediator. *Proc. Natl. Acad. Sci. USA* 102: 1116-1121.
3. Cheung, T.C., et al. 2005. Evolutionarily divergent herpesviruses modulate T cell activation by targeting the herpesvirus entry mediator cosignaling pathway. *Proc. Natl. Acad. Sci. USA* 102: 13218-13223.
4. Tao, R., et al. 2005. Differential effects of B and T lymphocyte attenuator and programmed death-1 on acceptance of partially versus fully MHC-mismatched cardiac allografts. *J. Immunol.* 175: 5774-5782.
5. Krieg, C., et al. 2005. Functional analysis of B and T lymphocyte attenuator engagement on CD4⁺ and CD8⁺ T cells. *J. Immunol.* 175: 6420-6427.
6. Sedy, J.R., et al. 2005. B and T lymphocyte attenuator regulates T cell activation through interaction with herpesvirus entry mediator. *Nat. Immunol.* 6: 90-98.

CHROMOSOMAL LOCATION

Genetic locus: BTLA (human) mapping to 3q13.2.

PRODUCT

BTLA 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 BTLA shRNA Plasmid (h): sc-45458-SH and BTLA shRNA (h) Lentiviral Particles: sc-45458-V as alternate gene silencing products.

For independent verification of BTLA (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-45458A, sc-45458B and sc-45458C.

RESEARCH USE

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

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

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

BTLA (4B8): sc-517004 is recommended as a control antibody for monitoring of BTLA 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 BTLA gene expression knockdown using RT-PCR Primer: BTLA (h)-PR: sc-45458-PR (20 μ l, 159 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.