

B7-H3 siRNA (h): sc-45444

BACKGROUND

T cell activation and immune function are regulated by the innate immune system through positive and negative costimulatory molecules. One such molecule, B7-H3 (B7-homolog 3, also designated B7RP-2) belongs to the B7 immunoglobulin superfamily. Soluble B7-H3 binds a putative receptor on activated T cells that is distinct from CD28, CTLA-4, ICOS and PD-1. Widely expressed on nonlymphoid tissues, B7-H3 costimulates proliferation of both CD4⁺ and CD8⁺ T cells. The ability of B7-H3 to stimulate Th1 and cytotoxic-T cell responses suggest that it may have antitumor activity. B7-H3 interactions may play a role in regulating cell-mediated immune responses against cancer, implicating B7-H3 as a potential therapeutic tool.

REFERENCES

1. Chapoval, A.I., et al. 2001. B7-H3: a costimulatory molecule for T cell activation and IFN- γ production. *Nat. Immunol.* 2: 269-274.
2. Ferlazzo, G., et al. 2002. T lymphocytes express B7 family molecules following interaction with dendritic cells and acquire bystander costimulatory properties. *Eur. J. Immunol.* 32: 3092-3101.
3. Sun, M., et al. 2002. Characterization of mouse and human B7-H3 genes. *J. Immunol.* 168: 6294-6297.
4. Suh, W.K., et al. 2003. The B7 family member B7-H3 preferentially down-regulates T helper type 1-mediated immune responses. *Nat. Immunol.* 4: 899-906.
5. Sun, X., et al. 2003. Mouse B7-H3 induces antitumor immunity. *Gene Ther.* 10: 1728-1734.

CHROMOSOMAL LOCATION

Genetic locus: CD276 (human) mapping to 15q24.1.

PRODUCT

B7-H3 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 B7-H3 shRNA Plasmid (h): sc-45444-SH and B7-H3 shRNA (h) Lentiviral Particles: sc-45444-V as alternate gene silencing products.

For independent verification of B7-H3 (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-45444A, sc-45444B and sc-45444C.

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

B7-H3 siRNA (h) is recommended for the inhibition of B7-H3 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

B7-H3 (F-11): sc-376769 is recommended as a control antibody for monitoring of B7-H3 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 B7-H3 gene expression knockdown using RT-PCR Primer: B7-H3 (h)-PR: sc-45444-PR (20 μ l, 553 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Chen, Y.W., et al. 2008. The immunoregulatory protein human B7H3 is a tumor-associated antigen that regulates tumor cell migration and invasion. *Curr. Cancer Drug Targets* 8: 404-413.
2. Sugita, S., et al. 2020. Retinal pigment epithelial cells derived from induced pluripotent stem (iPS) cells suppress or activate T cells via costimulatory signals. *Int. J. Mol. Sci.* 21: E6507.

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.