SANTA CRUZ BIOTECHNOLOGY, INC.

CD6 siRNA (h): sc-35015

BACKGROUND

CD6 is a type I transmembrane glycoprotein that is present on mature thymocytes, peripheral T cells and a subset of B cells. The CD6 glycoprotein is tyrosine phosphorylated during TCR-mediated T-cell activation and the size difference between the CD6 forms is due in part to differences in phosphorylation state. CD6 protein contains a 24-amino acid signal sequence, three extracellular "scavenger receptor cysteine-rich" (SRCR) domains, a membrane-spanning domain, and a 44-amino acid cytoplasmic domain. CD6 shows significant homology to CD5. CD6, which is also found in brain and B cell chronic lymphocytic leukemias, plays an important role in interactions of thymocytes with thymic epithelial cells. CD6 molecules can physically associate with the TCR/CD3 complex.

REFERENCES

1. Bazil, V., et al. 1989. Monoclonal antibodies against human leucocyte antigens. III. Antibodies against CD45R, CD6, CD44 and two newly described broadly expressed glycoproteins MEM-53 and MEM-102. Folia Biol. 35: 289-297.
2. Swack, J.A., et al. 1991. Biosynthesis and post-translational modification of CD6, a T cell signal-transducing molecule. J. Biol. Chem. 266: 7137-7143.
3. Aruffo, A., et al. 1991. The lymphocyte glycoprotein CD6 contains a repeated domain structure characteristic of a new family of cell surface and secreted proteins. J. Exp. Med. 174: 949-952.
4. Singer, N.G., et al. 1996. Role of the CD6 glycoprotein in antigen-specific and autoreactive responses of cloned human T lymphocytes. Immunology 88: 537-543.
5. Gimferrer, I., et al. 2003. The accessory molecules CD5 and CD6 associate on the membrane of lymphoid T cells. J. Biol. Chem. 278: 8564-8571.
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7. Hassan, N.J., et al. 2004. Frontline: optimal T cell activation requires the engagement of CD6 and CD166. Eur. J. Immunol. 34: 930-940.
8. Gimferrer, I., et al. 2004. Relevance of CD6-mediated interactions in T cell activation and proliferation. J. Immunol. 173: 2262-2270.

CHROMOSOMAL LOCATION

Genetic locus: CD6 (human) mapping to 11q12.2.

PRODUCT

CD6 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 CD6 shRNA Plasmid (h): sc-35015-SH and CD6 shRNA (h) Lentiviral Particles: sc-35015-V as alternate gene silencing products.

For independent verification of CD6 (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-35015A, sc-35015B and sc-35015C.

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

CD6 shRNA (h) Lentiviral Particles is recommended for the inhibition of CD6 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

CD6 (SPV-L14): sc-7320 is recommended as a control antibody for monitoring of CD6 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 CD6 gene expression knockdown using RT-PCR Primer: CD6 (h)-PR: sc-35015-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.

PROTOCOLS

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