

CD20 siRNA (h): sc-29972

BACKGROUND

CD20 is a leukocyte surface antigen consisting of four transmembrane regions and cytoplasmic N- and C-termini. The cytoplasmic domain of CD20 contains multiple phosphorylation sites, leading to additional isoforms. CD20 is expressed primarily on B cells but has also been detected on both normal and neoplastic T cells. CD20 functions as a calcium-permeable cation channel, and it is known to accelerate the G₀ to G₁ progression induced by IGF-1. CD20 is activated by the IGF-1 receptor via the α subunits of the heterotrimeric G proteins. Activation of CD20 significantly increases DNA synthesis and is thought to involve basic helix-loop-helix leucine zipper transcription factors.

REFERENCES

1. Tedder, T.F., et al. 1994. CD20: a regulator of cell-cycle progression of B lymphocytes. *Immunol. Today* 15: 450-454.
2. Schlossman, S., et al., eds. 1995. *Leucocyte Typing V*. New York: Oxford University Press.
3. Szollosi, J., et al. 1996. Supramolecular complexes of MHC class I, MHC class II, CD20, and tetraspan molecules (CD53, CD81, and CD82) at the surface of a B cell line JY. *J. Immunol.* 157: 2939-2946.
4. Algino, K.M., et al. 1996. CD20 (pan-B cell antigen) expression on bone marrow-derived T cells. *Am. J. Clin. Pathol.* 106: 78-81.
5. Smiers, F.J., et al. 1996. CD20 and CD40 mediated mitogenic responses in B-lineage acute lymphoblastic leukaemia. *Br. J. Haematol* 93: 125-130.
6. Himmelmann, A., et al. 1997. PU.1/Pip and basic helix loop helix zipper transcription factors interact with binding sites in the CD20 promoter to help confer lineage- and stage-specific expression of CD20 in B lymphocytes. *Blood* 90: 3984-3995.

CHROMOSOMAL LOCATION

Genetic locus: MS4A1 (human) mapping to 11q12.2.

PRODUCT

CD20 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 CD20 shRNA Plasmid (h): sc-29972-SH and CD20 shRNA (h) Lentiviral Particles: sc-29972-V as alternate gene silencing products.

For independent verification of CD20 (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-29972A, sc-29972B and sc-29972C.

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

CD20 shRNA (h) Lentiviral Particles is recommended for the inhibition of CD20 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

CD20 (D-10): sc-393894 is recommended as a control antibody for monitoring of CD20 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 CD20 gene expression knockdown using RT-PCR Primer: CD20 (h)-PR: sc-29972-PR (20 μ l, 581 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.