SANTA CRUZ BIOTECHNOLOGY, INC.

C2 siRNA (h): sc-95541

BACKGROUND

The complement component proteins: C2, C3, C4, and C5 are potent anaphylatoxins that are released during complement activation. Binding of these proteins to their respective G protein-coupled receptors induces proinflammatory events such as cellular degranulation, smooth muscle contraction, arachidonic acid metabolism, cytokine release, leukocyte activation, and cellular chemotaxis. C2 deficiency (C2D) is the most common deficiency of the classical complement pathway and is mostly found in patients with autoimmune disease or susceptibility to bacterial infections. The N-terminal extracellular domain 1 of complement C2 receptor inhibitory trispanning, or CRIT, binds to C2 and specifically interacts with the C2a fragment. In doing so, CRIT blocks C2 cleavage and also prevents the classical pathway of C3 convertase formation.

REFERENCES

1. Manderson, A.P., et al. 2001. Continual low-level activation of the classical complement pathway. J. Exp. Med. 194: 747-756.
2. Online Mendelian Inheritance in Man, OMIM™. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 217000. World Wide Web URL: http://www.ncbi.nlm.nih.gov/omim/
3. Inal, J.M., et al. 2004. Complement C2 receptor inhibitor trispanning: a novel human complement inhibitory receptor. J. Immunol. 174: 356-366.
4. Skelly, P.J. 2004. Intravascular schistosomes and complement. Trends Parasitol. 20: 370-374.
5. Kitano, E., et al. 2005. Immunologic tests: C2. Nippon rinsho 7: 59-62.
6. Gold, B., et al. 2006. Variation in factor B (BF) and complement component 2 (C2) genes is associated with age-related macular degeneration. Nat. Genet. 38: 458-462.
7. Jönsson, G., et al. 2006. Homozygosity for the IgG_2 subclass allotype G2M(n) protects against severe infection in hereditary C2 deficiency. J. Immunol. 177: 722-728.
8. Selander, B., et al. 2006. Mannan-binding lectin activates C3 and the alternative complement pathway without involvement of C2. J. Clin. Invest. 116: 1425-1434.

CHROMOSOMAL LOCATION

Genetic locus: C2 (human) mapping to 6p21.33.

PRODUCT

C2 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 C2 shRNA Plasmid (h): sc-95541-SH and C2 shRNA (h) Lentiviral Particles: sc-95541-V as alternate gene silencing products.

For independent verification of C2 (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-95541A, sc-95541B and sc-95541C.

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

C2 siRNA (h) is recommended for the inhibition of C2 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

C2 (E-7): sc-373809 is recommended as a control antibody for monitoring of C2 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 C2 gene expression knockdown using RT-PCR Primer: C2 (h)-PR: sc-95541-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.