SANTA CRUZ BIOTECHNOLOGY, INC.

C6 siRNA (m): sc-72770

BACKGROUND

The complement cascade is a multi-protein system that functions to clear pathogens from an infected host. Part of the innate (unchanging) immune system, the complement cascade consists of proteins and inactive zymogens that are present in blood and are stimulated by one of several triggers. Once stimulated, the cascade relays amplified responses throughout the body, ultimately activating the cell-killing membrane attack complex which can insert itself into the cell membrane and cause the cell to lyse. C6 (Complement component C6) is a 934 amino acid secreted protein that plays a role in the complement cascade, specifically functioning as part of the membrane attack complex. Expressed as two transcript variants, C6 contains one EGF-like domain, one LDL-receptor class A domain, one MACPF domain, two Sushi domains and three TSP type-1 domains. C6 deficiency is correlated with a higher risk of bacterial infection, further supporting the importance of C6 in the innate immune system.

REFERENCES

1. DiScipio, R.G., et al. 1989. The molecular architecture of human complement component C6. J. Biol. Chem. 264: 16197-16206.
2. Haefliger, J.A., et al. 1989. Complete primary structure and functional characterization of the sixth component of the human complement system. Identification of the C5b-binding domain in complement C6. J. Biol. Chem. 264: 18041-18051.
3. Hobart, M.J., et al. 1993. Structure of the human C6 gene. Biochemistry 32: 6198-6205.
4. González, S., et al. 1996. Characterization of the human C6 promoter: requirement of the CCAAT enhancer binding protein binding site for C6 gene promoter activity. J. Immunol. 157: 2282-2290.
5. Chamberlain-Banoub, J., et al. 2006. Complement membrane attack is required for endplate damage and clinical disease in passive experimental myasthenia gravis in Lewis rats. Clin. Exp. Immunol. 146: 278-286.
6. Qian, Z., et al. 2006. Antibody and complement mediated injury in transplants following sensitization by allogeneic blood transfusion. Transplantation 82: 857-864.

CHROMOSOMAL LOCATION

Genetic locus: C6 (mouse) mapping to 15 A1.

PRODUCT

C6 siRNA (m) 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 C6 shRNA Plasmid (m): sc-72770-SH and C6 shRNA (m) Lentiviral Particles: sc-72770-V as alternate gene silencing products.

For independent verification of C6 (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-72770A, sc-72770B and sc-72770C.

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

C6 siRNA (m) is recommended for the inhibition of C6 expression in mouse 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

C6 (3G11): sc-66191 is recommended as a control antibody for monitoring of C6 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 C6 gene expression knockdown using RT-PCR Primer: C6 (m)-PR: sc-72770-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.