SANTA CRUZ BIOTECHNOLOGY, INC.

# C9 siRNA (h): sc-62032

## BACKGROUND

C9 is a plasma protein synthesized in the liver and monocytes consisting of a single polypeptide chain. C9 is a part of the membrane attack complex (MAC), an important component of the immune system. The MAC forms upon complement system activation by invading pathogenic bacteria and consists of the four major complement proteins: C5b, C6, C7 and C8. These complement proteins bind to the outer surface of the plasma membrane of the invading cell. C9 binds to the membrane associated C5b-8 protein, which leads to the circular polymerization of 12-18 C9 molecules. These polymerized C9 molecules form a ring structure in the membrane. Molecules can then diffuse freely through this transmembrane channel, causing cell lysis and destruction of the invading bacterial cell.

## REFERENCES


1. Podack, E.R., et al. 1982. Membrane attach complex of complement (MAC): three-dimensional analysis of MAC-phospholipid vesicle recombinants. J. Immunol. 128: 2353-2357.
2. Tschopp, J., et al. 1983. Ultrastructure of the membrane attack complex of tetramolecular C9-polymerizing complex C5b-8. Proc. Natl. Acal. Sci. USA 79: 7474-7478.
3. Hatanaka, M., et al. 1992. Analysis of C5b-8 participation of two separate epitopes of C9 in C5b-8 binding. Mol. Immunol. 29: 911-916.
4. Wood A., et al. 1993. Specific induction of intracellular calcium oscillations by complement membrane attack on oligodendroglia. J. Neurosci. 13: 3319-3332.
5. Husler T., et al. 1996. Role of a disulfide-bonded peptide loop within human complement C9 in the species-selectivity of complement inhibitor CD59. Biochemistry 35: 3263-3269.
6. Farkas, I., et al. 2002. CD59 blocks not only the insertion of C9 into MAC but inhibits ion channel formation by homologous C5b-8 as well as C5b-9. J. Physiol. 539: 537-545.
7. Orren, A., et al. 2003. An abnormal but functionally active complement component C9 protein found in an Irish family with subtotal C9 deficiency. Immunology 108: 384-390.


## CHROMOSOMAL LOCATION

Genetic locus: C9 (human) mapping to 5p13.1.

## PRODUCT

C9 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 C9 shRNA Plasmid (h): sc-62032-SH and C9 shRNA (h) Lentiviral Particles: sc-62032-V as alternate gene silencing products.

For independent verification of C9 (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-62032A, sc-62032B and sc-62032C.

## 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

C9 siRNA (h) is recommended for the inhibition of C9 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

C9 (E-3): sc-390000 is recommended as a control antibody for monitoring of C9 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 C9 gene expression knockdown using RT-PCR Primer: C9 (h)-PR: sc-62032-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.